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P. Angelini, E. Bricchi, N. Zeppilli, L. Dimitriu, M. Rondolini, G. Angeles, S. Covino & R. Venanzoni

Screening of the antifungal activity of essential oils against human and plant pathogenic filamentous fungi

Abstract

Angelini, P., Bricchi, E., Zeppilli, N., Dimitriu, L., Rondolini, M., Angeles, G., Covino, S. & Venanzoni, R.: Screening of the antifungal activity of essential oils against human and plant pathogenic filamentous fungi. — Fl. Medit. 29: 5-12. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

In this study the antifungal activity of five essential oils (*Canarium luzonicum*, *Cymbopogon martinii*, *Ledum palustre* subsp. *groenlandicum*, *Matricaria chamomilla*, and *Ocimum tenuiflorum*) against both clinical (*Penicillium chrysogenum*, *Aspergillus tubingensis*, *A. minutus*) and plant pathogenic filamentous fungi (*Verticillium* spp., *Fusarium oxysporum*, *Sclerotina sclerotiorum*) were evaluated. Minimum Inhibitory Concentrations (MIC) were determined following CLSI M38-A2 recommendations.
All tested essential oils showed antifungal activity. *C. martini* and *O. tenuiflorum* essential oils were very effective with MIC range values similar or lower than those of terbinafine. *Sclerotinia sclerotiorum* (PeruMicA 26) and *Verticillium* spp. (PeruMicA 24) were the most sensitive strains to essential oils, while *A. tubingensis* (PeruMicA 21) showed the lowest sensitivity to the essential oils.

Key words: broth microdilution assay, drug resistance, MIC, natural products.

Introduction

There is an increasing consumer demand for natural preservatives to reduce the use of chemical compounds as antimicrobial agents in the field of nutrition and to combat various infections due to increasingly aggressive microorganisms that are resistant to the use of synthetic drugs (Melyssa & al. 2014).

In this context, the use of plant derived substances, such as hydro-alcoholic extracts or essential oils, can certainly play a fundamental role. These substances have an enormous versatility, nowadays the same plant still represents an important pool for the identification of novel drug leads, with a very broad spectrum of action due to their different chemical structure (Atanasov & al. 2015).

One of the major problems of conventional medicine is the current, excessive use of synthetic antimicrobials, resulting in the hypersensitivity and toxicity of the drugs (Etebu & Arikekpar 2016).

Pathogenic fungi can be the cause of fungal infections to which both humans and plants are susceptible, with some synthetic fungicides known to effectively control them. However, the emergence of resistant fungus strains limits the use of synthetic fungicides, with some of the latter possessing considerable toxicity. Moreover, the increased health and environmental hazard linked with synthetic molecules is of growing public concern. One of the most promising natural products for fungal inhibition can be found in essential oils (EOs) (Angelini & al. 2018a). Intense antifungal properties have, in fact, been exhibited in many types of EOs obtained from different plants, bryophytes and fungi (Properzi & al. 2013; Nazzaro & al. 2017; Angelini & al. 2014, 2016, 2017, 2018b; Dey & Mukherjee 2015; Al-Fatimi & al. 2018).

The composition, structure, and functional group of EOs have an important role in determining their biological activities. Their versatile character of antioxidant, antibacterial, antifungal, antidiabetic, antimutagenic and anticancer properties is well documented by many scientists (Angelini & al. 2006; Tirillini & al. 2009; Pagiotti & al. 2012; Properzi & al. 2013; Angelini & al. 2018a).

In particular, the antimicrobial activity of plant oils and extracts has formed the basis of many applications, including raw and processed food preservation, pharmaceuticals, alternative medicine and natural therapies (Hammer & al. 1999). The antimicrobial activity of essential oil might be caused by the properties of terpenes/terpenoids, that due to their highly lipophilic nature and low molecular weight are capable of disrupting the cell membrane, causing cell death or inhibiting the sporulation and germination of fungi.

While some of the EOs used on the basis of their reputed antimicrobial properties have well documented *in vitro* activity, there are few published data for many others (Tolouee & al. 2010; Mishra & Mishra 2011; Soni & Soni 2014; Zhang & al. 2017). Therefore, the aims of the current investigation were: i) to study the antifungal activity of *Canarium luzonicum* (Blume) A. Gray (Elemi, *Burseraceae*), *Cymbopogon martini* (Roxb.) W.Watson (Palmarosa, *Poaceae*), *Ledum palustre* subsp. *groenlandicum* (Oeder) Hultén (Ledum, *Ericaceae*), *Matricaria chamomilla* L. (Italian Camomilla, *Lamiaceae*) and *Ocimum tenuiflorum* L. (Tulsi, *Lamiaceae*) EOs against human and pathogenic filamentous fungi; ii) to verify the activity of the above-mentioned EOs against some fungal species resistant to common synthetic drugs.

Materials and Methods

Essential oils

The EOs used in this study were obtained from *C. luzonicum, C. martinii*, *L. palustre* subsp. *groenlandicum*, *M. chamomilla* and *O. tenuiflorum*. These oils were selected based on a literature survey due to being little known for their antifungal activity and were all bought from local markets.

Fungal strains

In vitro antifungal activity of essential oils was assessed against six clinical [*Aspergillus tubingensis* (PeruMicA 21), *A. minutus* (PeruMicA 22), *Penicillium chrysogenum* (PeruMicA 23)] and plant pathogenic filamentous fungal strains [*Verticillium* spp. (PeruMicA 24), *Fusarium oxysporum* (PeruMicA 25), *Sclerotina sclerotiorum* (PeruMicA 26)].

Two yeast strains, *Candida parapsilosis* (ATCC 22019) and *C. krusei* (ATCC 6258), were used as quality controls (CLSI 2008; CLSI 2012; CLSI 2017) for antifungal assays.

Voucher microbial cultures are maintained in the PeruMycA culture collection of the Department of Chemistry, Biology and Biotechnology (DCBB) (University of Perugia, Italy) and are available upon request.

Determination of Minimum Inhibitory Concentration (MIC)

MIC was evaluated using the broth microdilution assay, according to the CLSI M38-A2 protocol (2008). The antifungal drug tested was terbinafine (Sigma-Aldrich, Milan, Italy). The medium used was RPMI (Roswell Park Memorial Institute) 1640 medium (Sigma) with L-glutamine and without sodium bicarbonate, supplemented with 2% glucose (w/v), buffered at pH 7.0 with 0.165 M morpholine propane sulfonic acid (MOPS buffer).

The fungal inoculum suspension was prepared using a spectrophotometer to match the optical density with that of 70% transmittance at a 530 nm wavelength. The final inoculum concentration was from 0.2 to $0.4 \times 10^{4\text{-}5}$ CFU/ml.

Essential oils had a MIC range of 0.016-16 μg ml^{-1} and terbinafine (Novartis, Basel, Switzerland) had a MIC range of 0.03–16 μg ml^{-1}. MIC end-points (μg ml^{-1}) were determined after 48 hours (for *P. chrysogenum*, *A. tubingensis*, *A. minutus*, *Verticillium* spp.) and 72 hours (for *F. oxysporum*, *S. sclerotiorum*) of incubation in ambient air at 28-30 °C (CLSI 2008; CLSI 2017).

For all tested EOs, the MIC end-points were defined as the lowest concentration that showed total growth inhibition (Pagiotti & al. 2011). The MIC end-points for terbinafine were defined as the lowest concentration that inhibited 80% of the growth when compared with the growth control (Mota & al. 2009).

Geometric means and MIC ranges were determined from the three biological replicates to allow comparisons between the activities of EOs.

Results and discussion

Recently, the scientific interest into antimicrobial properties of EOs has been increasing. In particular, a large number of papers have been published on the EOs antimicrobial activity, but many of them focused on the activity against bacteria and yeasts while little investigation on the effect of EOs against filamentous fungi has been performed (Angelini & al. 2008, 2009; Khan & al. 2010; Prasad & al. 2010; Jamalian & al. 2012; Nikolić & al. 2016; Zhang & al. 2017). EOs from *C. luzonicum, C. martinii*, *L. palustre* subsp. *groenlandicum*, *M. chamomilla* and, *O. tenuiflorum* have been used for their medicinal properties for centuries; they

Table 1. Minimum inhibitory concentrations (MICs) of essential oils and terbinafine against clinical and plant pathogenic filamentous fungi.

	MIC[1] (µg ml^{-1})					
Essential oils	*C. luzonicum*	*C. martini*	*L. palustre* subsp. *groenlandicum*	*M. chamomilla*	*O. tenuiflorum*	Terbinafine
Fungal strains						
Aspergillus tubingensis (PeruMicA 21)	12.7 (8.0-16.0)	2.52 (2.0-4.0)	10.07 (8.0-16.0)	12.7 (8.0-16.0)	5.04 (4.0-8.0)	10.07 (8 - 16)
Aspergillus minutus (PeruMicA 22)	3.17 (2.0-8.0)	0.79 (0.5-1.0)	1.6 (1.0-4.0)	5.04 (4.0-8.0)	2 (1.0-4.0)	1.25 (0.5 - 2)
Penicillium chrysogenum (PeruMicA 23)	2.52 (2.0-4.0)	0.25 (0.25-1.0)	0.79 (0.5-1.0)	4 (2.0-8.0)	0.4 (0.25-0.5)	0.62 (0.5 - 1)
Verticillium spp. (PeruMicA 24)	0.79 (0.5-1.0)	0.4 (0.25-0.5)	0.63 (0.5-1.0)	1.59 (1.0-2.0)	0.25 (0.125-0.5)	0.2 (0.12 - 0.25)
Fusarium oxysporum (PeruMicA 25)	3.17 (2.0-4.0)	0.63 (0.5-1)	1.0 (0.5-2.0)	3.17 (2.0-4.0)	0.4 (0.25-0.5)	0.63 (0.5 - 1)
Sclerotinia sclerotiorum (PeruMicA 26)	0.5 (0.25-1.0)	0.2 (0.125-0.25)	0.63 (0.5-1.0)	1.59 (1.0-2.0)	0.25 (0.125-0.5)	0.39 (0.25 - 0.5)

[1]MIC values are reported as geometric means of three independent replicates (n=3); MIC range concentrations are reported within brachets.

possess antibacterial, antifungal, antioxidant, and anti-inflammatory properties (Harris 2010; Mogana & al. 2011; Singh & al. 2011; Raut & Karuppayil 2014; Yamani & al. 2016).

In the present study, the antifungal activity of these five EOs against both clinical and plant pathogenic filamentous fungi were evaluated by CLSI document M38-A2 (2008).

Antifungal activity of essential oils

Table 1 shows the MIC ranges and geometric means of EOs and terbinafine against the fungal species which has been tested. The MIC values of quality control strains that were reproducible, fell within the established ranges published by CLSI protocol (2008). All tested EOs with concentrations ranging from 16-0.16 showed antifungal activity (Table 1). *C. martini* and *O. tenuiflorum* essential oils, were extremely effective on all tested fungi, with MIC range values similar or lower than those of terbinafine (0.2-2.52 µl ml^{-1} and 0.25-5.04 µl ml^{-1} respectively), while *C. luzonicum* and *M. chamomilla* EOs exhibited a lower level of inhibition. The *S. sclerotiorum* and *V.* spp. strains tested demonstrated the most sensitive fungal species to essential oils, with MIC ranges of 0.125-2 µg ml^{-1}, while *A. tubingensis* strain revealed the least sensitivity to the essential oils (MIC ranges: 2.52-12.7 µg ml^{-1}). Plant pathogens fungal strains were much more sensitive to terbinafine and essential oils than clinical strains (Table 1).

Terbinafine showed good activity *in vitro* against *Verticillium* spp. (MIC: 0.2 µg ml^{-1}), *S. sclerotiorum* (MIC: 0.39 µg ml^{-1}), *P. chrysogenum* (MIC: 0.62 µg ml^{-1}), *F. oxysporum* (MIC range: 0.63 µg ml^{-1}) while, *A. minutus and A. tubingensis* strains were less sensitive to this allylamine-antifungal drug, with a geometric MIC of 1.25- and 10 µg ml^{-1}, respectively. Currently, clinically relevant breakpoints for terbinafine are not available. However, since for Ryder & al. (1998) breakpoint of> 8 µg / ml was taken to indicate *in vitro* terbinafine resistance, *A. tubingensis* may be considered a fungal isolate terbinafine resistance.

A. tubingensis and *A. minutus* strains are also reported as amphotericin- and itraconazole-resistant (Pagiotti & al. 2018).

Conclusions

This study provides a range of information about the antifungal activity of EOs of *C. luzonicum, C. martini*, *L. palustre* subsp. *groenlandicum*, *M. chamomilla* and, *O. tenuiflorum*. It is difficult to compare antifungal activity results from different studies because of the different extraction methods, test organisms and test systems used. Of the five EOs studied, *C. luzonicum* and *C. martini* were highly effective against all fungal strains tested, showing two promising sources in the search for new antifungal drugs. Moreover, most EOs are safe and free of adverse side effects when used properly.

The most important safety factor for EOs is their dosage (Edris 2007). Specific pharmacological approaches will be needed in future to validate its use as a phytotherapeutic product.

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Article

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E. Kozuharova, I. Ionkova & F. M. Raimondo

Invasive alien species: potential cheap resources of plant substances for medicinal use*

Abstract

Kozuharova, E., Ionkova, I. & Raimondo, F. M.: Invasive alien species: potential cheap resources of plant substances for medicinal use. — Fl. Medit. 29: 13-25. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

On the basis of the literature examined, the scientific acquisitions concerning the pharmacological properties and medicinal uses of *Ambrosia artemisiifolia* and *Erigeron canadensis* – two American vascular plants of Asteraceae family that have become invasive in Europe and others continents – are collected and discussed. The data reveal the potential of the invasive as cheap sources of compounds with valuable pharmacological activities. In addition to the two plants presented as a case study, there are hundreds of plant species at hand as potential assets to explore and make money.

Key words: *Ambrosia artemisiifolia*, *Erigeron canadensis*, *Asteraceae*, biologically active compounds, pharmacological activity, management, ecosystem services.

Introduction

Some alien plant species have high tolerance of various habitat conditions and elevated propagation ability. This promotes their aggressive invasive behaviour. Often they over compete the local vegetation. Additionally many of them suppress the seed germination and seedling development of local plants. In the newly invaded habitats they might not have suitable herbivores to control their populations (DAISIE 2009). The only effective enemy might be *Homo sapiens*. Humans are known with their destructive power once an object has become significant for industrial utilization. Due to the fact that these are aggressive invasive species, they can provide abundant and cheap resources of plant bioactive compounds which can be used for medicinal purposes. Additionally, excessive harvesting for medicinal use might contribute to decrease their populations and reduce the destructive impact of these species on natural habitats.

*Extended and enriched version of the oral presentation given at the International Symposium "Botany at the intersection of Nature, Culture, Art and Science", Selinunte, 28-30 June 2018.

The good practice in the traditional use of plants for remedial purposes is a precondition for successful phyto-therapeutic application. On these basis a modern scientific approach such as phytochemical and pharmacological investigations affirms the therapeutic effect.

Echinacea purpurea (L.) Moench. is an example of a plant with growing modern popularity based on good practices in Native Americans' traditional medicine (Austin 2004, Wilkes 2012).

The aim of this contribution is to review research data and reveal the potential of two invasive plants of *Asteraceae* family as cheap sources of compounds with valuable pharmacological activities.

Material and metods

On the basis of the literature examined, the scientific acquisitions concerning the pharmacological properties and uses of *Ambrosia artemisiifolia* L. and *Erigeron canadensis* L., two American species that have become invasive on different continents, are collected and discussed. Some brief botanical, distributive and ecological indications on these two species are given below.

Ambrosia artemisiifolia is an annual plant native to North America across Canada, the eastern and central United States, the Great Plains, and in Alaska; the Caribbean on Cuba, Hispaniola, and Jamaica; and South America in the southern bioregion (Argentina, Chile, Paraguay, Uruguay), the western bioregion (Bolivia, Peru), and Brazil. This species has been introduced in Europe at the end of 19th century in seed crops at various independent geographical points and at various times since its introduction in natural habitats. Recently, the number of naturalized populations increased considerably fast and it is considered to be the one of the most dangerous invasive alien species of Europe (Chauvel & al. 2006; Kazinczi & al 2008; Essl & al. 2009; DAISIE 2009; Hodgins & al. 2013).

Erigeron canadensis [= *Conyza canadensis* (L.) Cronquist] is an erect, annual plant. It is native throughout most of North America and Central America and invasive in Europe. It was introduced into Europe in the mid 17th century, likely along with Canadian furs shipped to France (Tilley 2012). This plant is associated with disturbed, open and unshaded habitats, such as cultivated land, abandoned fields, roadsides, ruderal places, and other open habitats. Canadian horseweed can reduce crop yields through direct competition for resources. (Tilley 2012). It also contains allelopathic chemicals which can inhibit germination and reduce seedling growth in several species (Shaukat & al. 2003).

On the two plants chosen as a case study (*Ambrosia artemisiifolia* and *Erigeron canadensis*), here we present: 1) traditional ethnobotanical data from their native habitats; 2) modern investigations of pharmacological activity and essential secondary compounds.

The case studies

Ambrosia artemisiifolia (Fig.1).

This species contains phytotoxins and shows significant inhibitory effects on the seed germination and primary growth of crops (Ritter & Coble 1981; Wang & Zhu 1995;

Fig. 1. Drawing of *Ambrosia artemisiifolia* from Bulgarian population (by E. Kozuharova).

Brückner & al. 2003; Vidotto & al. 2013). High levels of naturally occurring variation in the ALS gene sequence have been found in *A. artemisiifolia* allowing rapid and widespread selection for resistance when ALS-inhibiting herbicides are used (Tranel & al. 2004). The main problem is that its pollen is well known as noxious allergen (Mirone & al. 2004; D'Amato & al. 2007; Léonard & al. 2010).

There are different therapeutic effects known to the Native Americans, which are well documented (Gilmore 1913; Speck 1941; Tantaquidgeon 1972; Hamel & al. 1975; Herrick 1977; Shemluck 1982; Anderson & al 1988; Austin 2004, Table 1).

Table 1. Ethnobotanical data and therapeutic effects of *Ambrosia artemisiifolia* known to the Native Americans.

Tribe	Therapeuti effect	Method of application	References
Cherokee	Dermatological Aid	Crushed leaves rubbed on insect sting and infusion of leaf rubbed on hives.	Hamel & al. 1975, Moerman 1998
	Disinfectant	Juice of wilted leaves applied to infected toes.	Hamel & al. 1975 Moerman 1998
	Febrifuge	Infusion of leaf taken for fever.	Hamel & al. 1975 Moerman 1998
	Pulmonary Aid	Infusion taken for pneumonia.	Hamel & al. 1975 Moerman 1998
Dakota	Antidiarrheal	Infusion of leaves and plant tops taken for bloody flux.	Gilmore 1913 Moerman 1998
	Antiemetic	Infusion of leaves and plant tops taken for vomiting.	Gilmore 1913 Moerman 1998
Delaware	Blood Medicine	Poultice of plant used to prevent blood poisoning.	Tantaquidgeon 1972, Moerman 1998
Houma	Gynecological Aid	Decoction of root taken for menstrual troubles.	Speck 1941, Moerman 1998
Iroquois	Antidiarrheal	Compound decoction of plants taken for diarrhea with bleeding.	Herrick 1977, Moerman 1998
	Heart Medicine	Infusion of roots taken for stroke.	Herrick 1977, Moerman 1998
	Orthopedic Aid	Decoction of plants taken for cramps from picking berries.	Herrick 1977, Moerman 1998
Lakota	Antirheumatic (External)	Infusion of leaves applied to swellings.	Rogers 1980, Moerman 1998
		Plant used for toilet paper.	Rogers 1980, Austin 2004, Moerman 1998
Luiseno	Emetic	Plant used as an emetic.	Sparkman 1908, Moerman 1998
Mahuna	Dermatological Aid	Infusion of plant used as a wash for minor skin eruptions and scalp diseases.	Romero1954, Moerman 1998
Oto	Antiemetic	Bruised leaves laid on scarified abdomen for nausea.	Gilmore 1919, Moerman 1998

It contains sesquiterpenoids-sesquiterpene lactones, etc. (Bianchi & al. 1968; David & al. 1999; Sturgeon & al. 2005; Taglialatela-Scafati & al. 2012; Huang & al. 2014; Ding & al. 2015; Kiss & al. 2017) as well as polyphenols and flavonoids (Parkhomenko & al. 2005, 2006; Maksimovic 2008) or other constituents (Georgiev & al. 2007).

Pharmacological tests revealed that *Ambrosia artemisiifolia* possess numerous activities such as cytotoxic, antimalarial, antimicrobial, anti-inflammatory, pronounced hepatoprotective and hypolipemic-lowers the concentration of fats in the blood (Table 2).

Principal constituents of the essential oil obtained by steam destilation are germacrene D (24.1%), limonene (16.83%), α-pinene (8.0%) and myrcene (7.4%) with significant bactericidal and fungicidal activity (Chalchat & al. 2004, Table 2).

Table 2. Pharmacological activities of *Ambrosia artemisiifolia*.

Activity of the plant extracts	Reference
Antioxidant	Maksimovic 2008
Hypolipemic	Parkhomenko & al. 2005, 2006
Hepatoprotective	Parkhomenko & al. 2005, 2006
Cytotoxic	Bianchi & al. 1968; David & al. 1999; Sturgeon & al. 2005; Huang & al. 2014; Kiss & al. 2017
Antimalarial	David & al. 1999
Antimicrobial: antifungal, antibacterial, antiviral	Georgiev 2007; Solujić & al. 2008
Anti-inflammatory	Pérez 1996
Activity of the essential oil	
Significant bactericidal and fungicidal activity	Chalchat & al. 2004

Erigeron canadensis (Fig. 2).

Ethnobotanical investigations reveal that there are number of therapeutic effects known to the Native Americans (Moerman 1998, 2009; Pennacio & al. 2010; Austin 2010, Table 3). It is claimed that in folk medicine this plant is used in diarrhea, dysentery uterine hemorrhages, dropsy, gravel, cystitis, calculus, bronchial catarrh, and hemoptysis (Yan & al. 2010, Shakirullah & al. 2011) but there is no indication for original native American tradition Phytochemical studies revealed that *Erigeron canadensis* contained saponins, diterpenoids, terpenoids, glycosides, tannin, anthraquinone, steroids and flavonoids (quercetin-7-O-beta-D-galacto pyranoside,quercetin, luteolin, apigenin, 5,7,4'- trihydroxy-3'-methoxy flavone, quercetin-3-alpharhamnopyranoside, quercetin-3-O-beta-Dglucopyranoside, apigenin-7-O-beta-D-gluco pyranoside, luteolin-7-O-beta-D-glucuronide methyl ester,4'-hydroxy baicalein-7-O-beta-Dglucopyranoside, baicalein and rutin). Also conyzolide; conyzoflavone; conyzapyranone A; conyzapyranone B; 4 Z,8 Z-matricaria- γ-lactone; 4 E,8 Z-matricaria- γ-lactone; 9,12,13-trihydroxy10(E)-octadecenoic acid; epifriedelanol; friedeline; taraxerol; simiarenol; spinasterol; stigmasterol; β- sitosterol; C10 acetylenes; sesqui-

Table 3. Ethnobotanical data and therapeutic effects of *Erigeron canadensis* known to the Native Americans.

Tribe	Method of application	Therapeutic effect	References
Native Americans	smoke the flowers and leaves	for pleasure or to relive head colds.	Pennacio & al. 2010
Native Americans		gastointestinal aid	Moerman 2009
Chumash	grind the plant tea	relive pain kidney problems	Austin 2010
Navajo	crush leaves to	treat skin problems	Austin 2010

Table 4. Pharmacological activities of *Erigeron canadensis*.

Activity of the plant extracts	Reference
Antioxidant (potent radical scavenging activity)	Olas & al. 2006; Saluk-Juszczak & al. 2010; Shah & al. 2012; Park & al. 2013
Anti-platelet and anticoagulant	Olas & al. 2006; Saluk-Juszczak & al. 2010; Pawlaczyk & al. 2011
Antiinflammatory	Lenfeld & al. 1986, Sung & al. 2014
Antiproliferative	Réthy 2007; Réthy & al. 2007; Csupor-Löffler & al. 2009, 2011
Anti-gastric ulcer	Park & al. 2013
De-pigmentation	Hong & al. 2008
Antibacterial	Biswas & Sinha 2014
Reduce blood glucose level in vitro	Aslam & al. 2018
Activity of the essential oil	
Anti-inflammatory	Guenter 1976
Haemostatic	Guenter 1976
Stimulant	Guenter 1976
Carminative	Guenter 1976
Antiproliferative	Choi 2008,
Antifungal - evaluated to weak	Curini & al. 2003
Antifungal - evaluated to moderate or strong activity against *Candida, Cryptococcus, Trichophyton, Rhodotorula* but no antibacterial activity	Veres & al. 2012
Anti-melanoma B16 activity	Yan & al. 2010

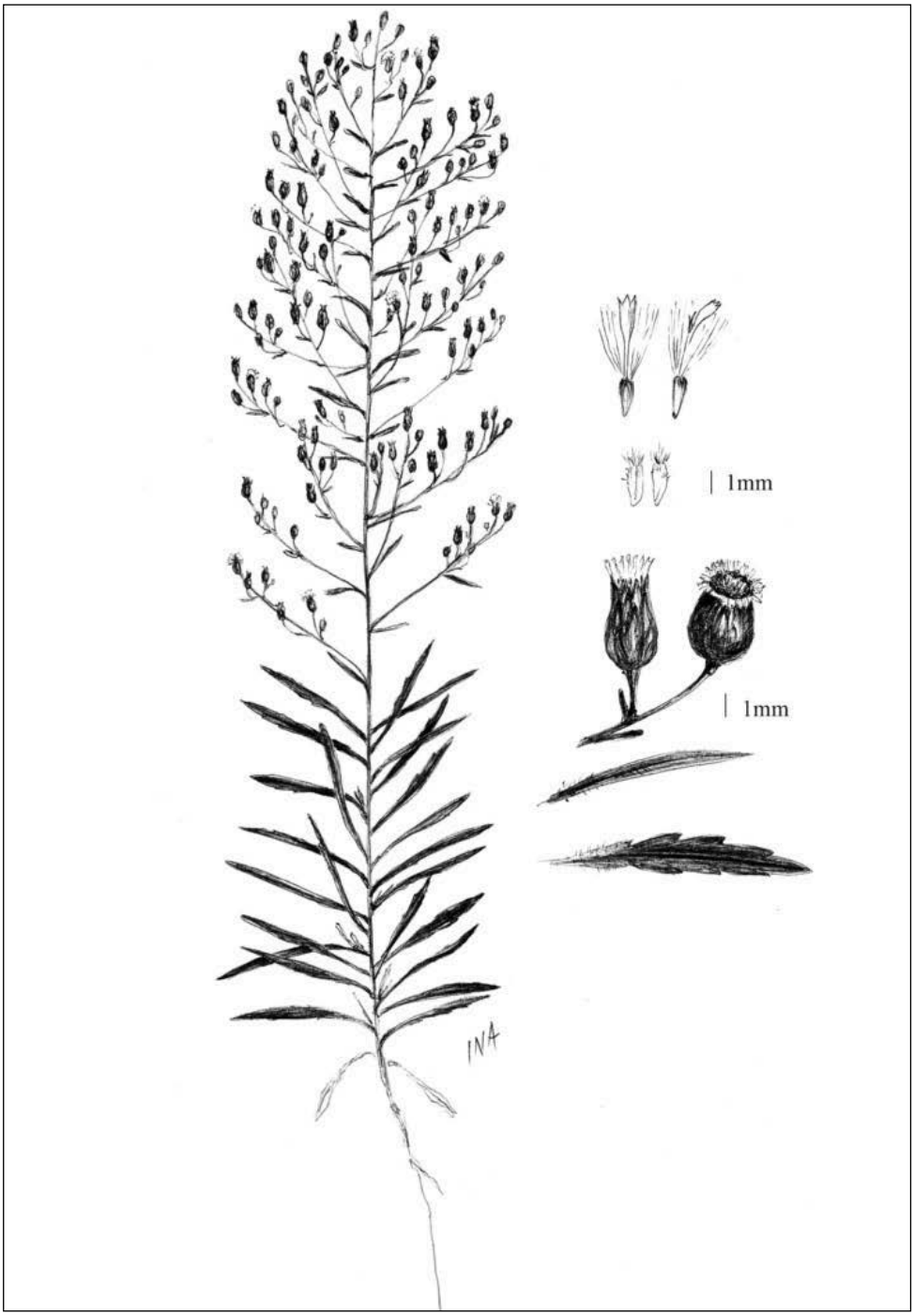

Fig. 2. Drawing of *Erigeron canadensis* from Bulgarian population (by E. Kozuharova).

terpene hydrocarbons, beta-santalene, beta-himachalene, cuparene, alpha-curcumene, gamma-cadinene, sphingolipids 1,3,5-trihydroxy-2-hexadecanoyl amino-(6E,9E)-heptacosdiene; 1,3,5-trihydroxy-2- hexadecanoylamino-(6E,9E)-heptacosdiene-1-Oglucopyranoside; 1,3-dihydroxy-2-hexanoylamino- (4E)-heptadecene; p-hydroxybenzoic acid, 3,5- dihydroxybenzoic acid, 3,5-dimethoxybenzoic acid; 3beta-hydroxyolean-12-en-28-oic acid; 3beta-erythrodiol; beta-sitosterol; stigmasterol; beta-sitosterol 3-O-beta-D-glucoside and harmine were isolated from different parts of the plant (Bohlmann & Jakupovic 1979; Lenfeld & al. 1986; Czeczot & al. 1990; Mukhtar & al. 2002a, 2002b; Wei & al. 2007; Shakirullah & al. 2011; Csupor-Löffler & al. 2011; Shao 2012; Shah & al. 2012; Veres & al. 2012; Biswas & Sinha 2014).

Extracts of *Erigeron canadensis* were reported to have anti-inflammatory and anti-proliferative activity as well as gastric ulcer protective effect. Also anti-inflammatory, de-pigmentation, anti-coagulant, anti-platelet and anti-oxidant effects are reported (Table 4).

Health risks or side effects following the proper administration of designated therapeutic dosages were not recorded (Anonymous 2000).

The essential oil contains more than 30 constituents but is mainly composed of monoterpenoids-limomene, camphene, α and β-pinenes etc., and sesquiterpenoids – caryophyllene, germacrene D and α-curcumene etc. (Miyazawa & al. 1992; Lis & Góra 2000; Rustaiyan & al. 2004; Tzakou & al. 2005; Lis & al. 2005; Unnithan & al. 2014, Ayaz & al. 2017). A few non-terpenoid acetylenic compounds were also detected (Unnithan & al. 2014). The compounds isolated from essential oils differ among different locations which may be attributed to the different environmental and climatic conditions (Unnithan & al. 2014).

The essential oil of *Erigeron canadensis* possess anti-inflammatory, haemostatic, stimulant, carminative and antifungal activity (Table 4).

Discussion and conclusion

Ethnobotanical data from their habitats reveal promising medicinal potential. A growing body of scientific literature points to their therapeutic properties. Valuable chemical constituents of these alien invasive species are sesquiterpene lactones, essential oils etc. They posses different activities such as anticancer activity, as well as antitussive, antifungal, antiinflammatory, antinociceptive, hypoglycaemic, antimitotic, antioxidant, antitrypanosomal, CNS depressant activity, diuretic effects, contact dermatitis, insecticidal and herbicidal activities, hepatoprotective and hypolipemic activities etc.

Due to the fact that these are aggressive invasive species, they can provide abundant and cheap resources reach of plant chemical constituents which can be utilized for therapeutic purposes. Additionally, exploitation of the biomass for medicinal use might contribute to relieving the destructive impact of these species on natural habitats.

The invasive plant species considered in this contribution deserve further investigations as they have valuable pharmacological activities. Harvesting of the plants for their medicinal value may reduce the populations due to decreased seed production and propagation. This way additionally the risk of allergy caused by *Ambrosia artemisiifolia* pollen will decrease.

There are many studies conducted on the two species here examined and there is good reason to believe that there are prospects of study that can still provide other products that can

contribute to give further value to negatively evaluated biological resources, since they are seen as harmful and competitive agents and not as a resource. Therefore, unexplored fields remain to which new research can be directed. In addition to the plants presented as a case study, there are hundreds of species at hand as potential assets to explore and make money.

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Article

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"Re-flowering flowers": the hope of an eternal blooming since Roman times*

Abstract

Caneva, G., Monaco, A., Virgili, P. & Bartoli, F.: "Re-flowering flowers": the hope of an eternal blooming since Roman times. — Fl. Medit. 29: 27-44. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

"Re-flowering flowers", i.e., the metamorphic artistic representations of plants in which one flower gives birth to other ones, are often detectable in the phytoiconography of the Greek-Roman art. Through an extensive analysis of archaeological artworks in the Euro-Mediterranean and West-Asian area, we found the diffusion of this motif starting from the Hellenistic period (IV century B.C.). The metamorphic flowers motif became a dominant element in triumphal arches, and later also in coffered ceilings, forming the so-called "rosettes". The identification of the single plant elements of these compositions can be carried out both on pottery (among which the best examples come from the Apulian and Greek vases) and on carved structures, where colours are no longer present. We analyzed in detail the botanical compositions of the scrolls of the *Ara Pacis* and in the triumphal Arches of *Titus* and *Septimius Severus* in Rome (Italy). The results enhanced the representation of a relevant floristic richness with some recurrent flowers, such as those of *Lilium*, *Anemone*, *Silene*, *Stellaria*, *Anthemis*, *Calendula*, *Scabiosa*, *Asphodelus*, *Nuphar*, *Carlina* and *Laurus*, but also fruits, shoots, bulbs and floral buds. This motif seems linked to the leading thread of the metamorphosis in the Hellenistic culture and the revived Pythagorean theories of the Augustan age. The continuous transformation of an element into another suggests a spatial translation of temporal concepts: the absence of an end; death as a prelude to a new life. We should better understand the meanings of natural elements in the ancient artistic representations since they were not used only as a mere decorative motif but were part of a widely shared symbolic language.

Key words: flowers representation; plant iconography; phytoiconology; plant symbolism; Roman archaeology.

Introduction

Analyzing art representations in the Greek-Roman culture, spread in western cultures for hundreds of years, we can highlight the presence of "Re-flowering Flowers" (RF), i.e., metamorphic plant images in which one flower gives birth to another one, in sequence

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(Fig. 1). Such scheme is widely recurrent in scrolls and the so-called “rosettes” (Ramage & Ramage 2008), and these iconographic elements had a great fortune from ancient times until nowadays, as they were used in the coffered ceiling of aristocratic palaces, in churches and “classic” architectonic elements (Fig. 2). We can even assume that this type of decoration, highly diffused from Augustan times in the Euro-Mediterranean area, seems to be even more ancient (Vandi 2002).

The archaeological literature neglects these representations in their constitutive polymorphic and metamorphic structure, even in the case of Pompeii, where some attention was given to plant representations in paintings (Comes 1879; Jashemski & al. 2002; Ciarallo 2004, 2006; Caneva 2014). In fact, a description of the composite elements of floristic representations was carried out rarely, such as in the case of the *Ara Pacis* freezes (Caneva 2010), and in other archaeological remains in Rome (Caneva & al. 2014; Kumbaric & Caneva 2014). Despite the absence of specific studies, the ceilings of triumphal arches of Roman Emperors seem to represent a relevant source of examples for understanding the structure and composition of the RF motif.

Moreover, often the floristic parts have been considered only as a decorative motif. It seems contradictory with the knowledge that- in ancient cultures- the representation of an image was the bearer of a message, which needed to be read in relation to its provenience and context to be fully understood (Zanker 1989; Pensabene 2011). For the ancient Roman culture, as for other ancient civilizations, the importance of giving a symbolic meaning to the subject of representations, is clearly underlined by *Vitruvius* in his famous treaty on Architecture (*De Architectura*, Book I, 5th paragraph): *“the architect must possess a good knowledge of history, which permits him to explain to the eventual interlocutors the symbolical meaning with which he often embellishes his buildings”*. The primary role of nature in such a means of communication was evident from the literary texts of *Horatius (Odes), Gaius Plinius Secundum (Naturalis Historia), Virgilius (Bucolicae),* and *Ovidius (Metamorphosis),* as well as from an in-depth iconographic analysis of archaeological remains (Caneva 2010; Caneva & al. 2014; Kumbaric & al. 2014; Kumbaric & Caneva 2014). In fact, all the ancient civilizations were in close contact with the natural world, and

Fig. 1. Examples of “Re-flowering flowers”, i.e., images in which one flower sprouts from another one, in a sequence of two, three or four elements (from the *Ara Pacis* scrolls, Caneva 2010).

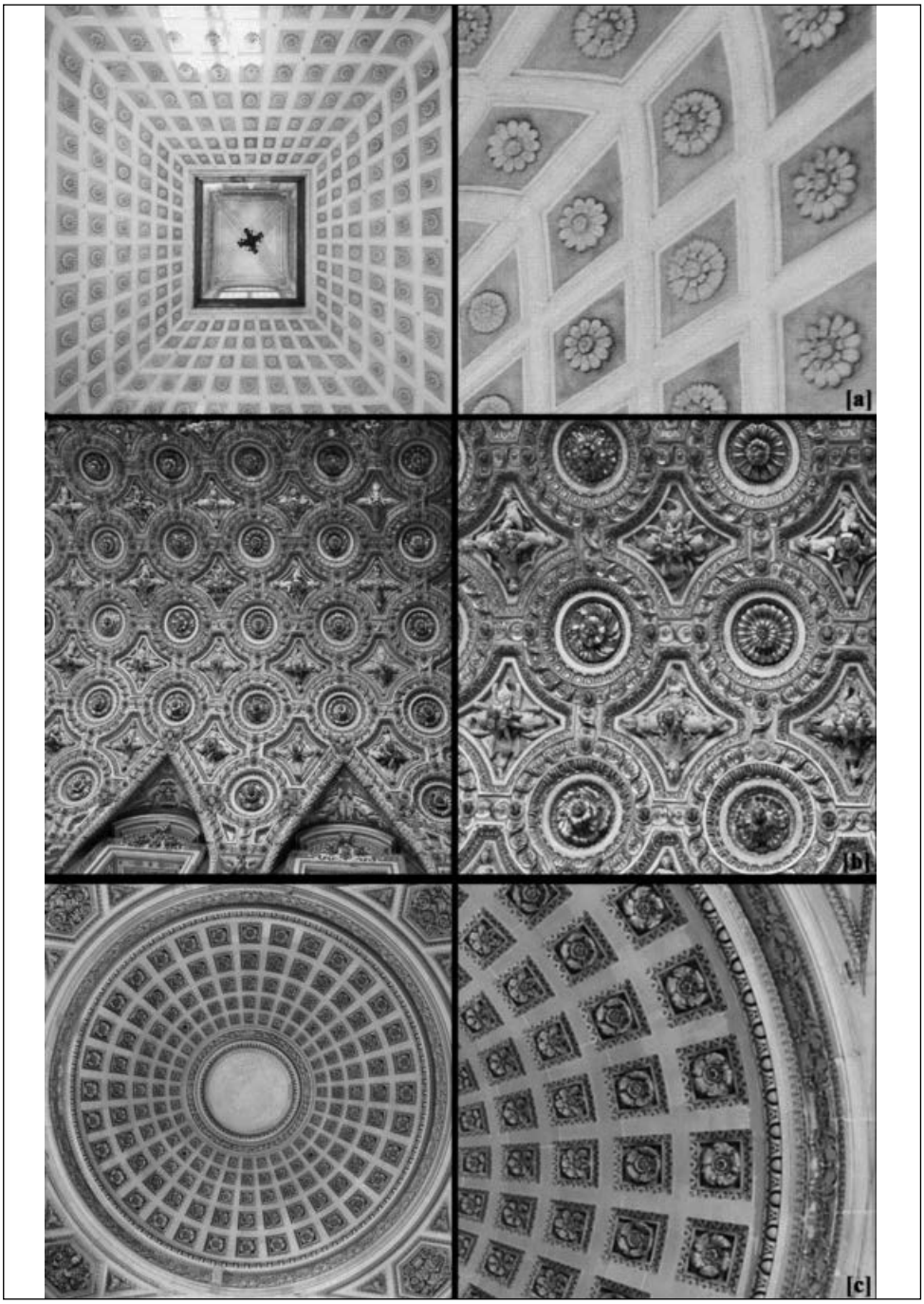

Fig. 2. The RF motif in the coffered ceiling of aristocratic palaces, in churches and in the "classic" architectonic elements: (a) Grimani Palace, Venice; (b) Quirinale Palace, Rome; (c) Pantheon, Paris.

recurrently explained natural phenomena as deity expressions affecting humans (Vandi 2002), and we should better understand the meanings of natural elements in the ancient artistic representations, to better comprehend their culture.

Considering the importance of explaining this recurrent motif, which was insufficiently analyzed, this study aims to define: i) its origin, ii) its symbolical meaning and iii) the variants of the RF motif, taking into consideration a wide range of examples, including some examples of ceilings of triumphal Roman arches. So, in this work, we will discuss its origin and diffusion, the scheme of composition in some relevant monuments and its symbolic and cultural meanings.

Methods

The analysis of the RF motif started from an extensive bibliographic research, looking at different type of architectural, ceramic and jewellery artifacts, mainly from the Hellenistic period in the Euro-Mediterranean area, but also from the Assyrian and Babylonian area (Kleiner 2012). It was useful to analyse both the origin of the motif and its diffusion over time and in different regions. The bibliographic analysis was associated with field research carried out in Rome (Roman *Forum*, Colosseum valley, Imperial *Fori*, Trajan Markets and *Ara Pacis*), to point out how widely this kind of motif was employed and how differently it was realized. We used the collected images together with the results of the bibliographical analysis, to produce a database, which was used to select three outstanding examples of RF representations, which are described here with their historical background and iconographic structure. Following the idea of carrying out a visual “break down” of elements that are in successive transformation, which was suggested for the *Ara Pacis* scrolls (Caneva 2010), we carried out a botanical analysis identifying the “various elementary components”. Specifically, we identified various plant parts (calyx, corolla, stamens, pistils, fruits, bulbs, leaves and derived structures), through the comparison with flora and botanic atlases of reference (Pignatti 1982; http://dryades.units.it/floritaly). The meaning of this motif was finally hypothesized considering the previously cited ancient textbooks of *Plinius, Virgilius, Vitruvius* and *Ovidius*.

The analyzed monuments

The *Ara Pacis* was conceived as an altar dedicated to the peace, which was built (from 13 B.C. to 9 B.C.) to celebrate *Augustus*’s return from his victorious expedition in Gaul and Spain (Fig. 3a). Marking the end of the external rebellions and internal struggles that started with *Julius Caesar’s* murder in 44 B.C., it was an augural tribute for a new peaceful age of Roman dominance coincident with the Empire’s founding (La Rocca 1983; Zanker 1990; Castriota 1995; Cohon 2002). The botanical scrolls are here widely carved in the six panels of the lower parts of the external faces, below the representations of the scenes of the Roman origin and of the finally reached prosperity (four panels), as well as below the two panels representing the parade that was welcoming *Augustus* at his return. This monument can be considered the most relevant example of an extensive use of plant representations for iconographic and symbolic purposes (Caneva 2010).

The arches of *Titus* and *Septimius Severus* in Rome are other earlier and highly representative momuments that include such iconographic motif. The origin of such architectur-

al element (the arch) was possibly related to the *propylaea* of the Hellenistic cities, to the *tetrapylia* of the columnnade streets and elements of *Alexandria* and *Pergamum*. However, the arch seems also to be related to the Etruscan-Italian door of the city, but the first time the word *arcus* appears in a document (of the Decurion assembly in Pisa) was during the Augustan age, when it was referred to the building of the arch of *Caius* and *Lucius Caesar* (4 A.D.) (La Rocca & al. 2008).

The arch of *Titus* (Fig. 3b) was realized in 81 A.D., as an honorific monument built by the Domitian emperor on the *via Sacra*, to reinforce the community consensus on the Flavian dynasty, commemorating the victories of his brother *Titus*, as a deed of pietas (De Maria 1988). It celebrates the *bellum judaicum* and the conquest of Jerusalem in the 71 A.D., and it represents *Titus*'s triumphal return, with the booty of the temple of Jerusalem, a four-horse chariot, crowned by a personification of Victory, and *Titus*'s deification being carried to heaven by an eagle. Other representations in the arch celebrate *Honos* and *Virtus*, Rome and the *Genius* of the Roman people (Torelli & al. 2008).

The arch of *Septimius Severus* (Fig. 3c) was built in the Roman *Forum*, and it was dedicated by the Senate in 203 A.D. to celebrate the *Princeps Virtutis* and his sons *Caracalla* and *Geta,* winning the wars against *Parthia*. The historic relieves, located on the smaller fornix, imitates the triumphal pictures and the friezes of the columns as well. The narration starts from the bottom to the top, showing the salient moments of the war of Mesopotamia (Torelli & al. 2008). The decoration of the arch exhibits a polysemy of meanings. Looking at the archivolt, we can see the Victories carrying trophies, while underneath there are the *Putti* of seasons representing the *aeternitas* of the imperial power. Moreover, the fluvial deities in the smaller fornix symbolize the territorial extension of the Empire, and the arch is rich of images carrying messages connected to peace, prosperity, power and the Emperor (De Maria 1988).

Results

The origin and diffusion of the RF motif

Our bibliographical analysis and field surveys showed as, in ancient times, the *RF* motif was adopted in different architectural elements in ancient Greece and Macedonia, and in the wide *Magna Grecia* area (Tab. 1). Moreover, this motif was not confined only to a fixed context, but it was diffused in a wide artistic production. Examples are present on marble reliefs, in gold necklaces, on mosaic pavements and pottery vases. On pottery, significant examples come from the Apulian and Greek vases where the use of still-visible colour makes easier the analysis of the single elements of composition. In this case, such as on marble friezes or in mosaics, the RF motif includes compositions that start from a central *Acanthus* and end in a flower. In the vault of the arch and ceilings the representation is not connected with other plant elements, and this type of execution is close to examples of the motif in jewellery (e.g., Diadem of Verghina, Taranto necklace) (Becatti 1965).

Considering that the most ancient monuments where the RF motif is detectable, such as the Temples of *Saturnus, Castor* and *Pollux,* and *Apollo Sosianus,* were widely modified during the centuries with the addition of new elements, the most likely origin of such motif seems to be the "red-figure vases" from ancient *Apulia*. The chronology of

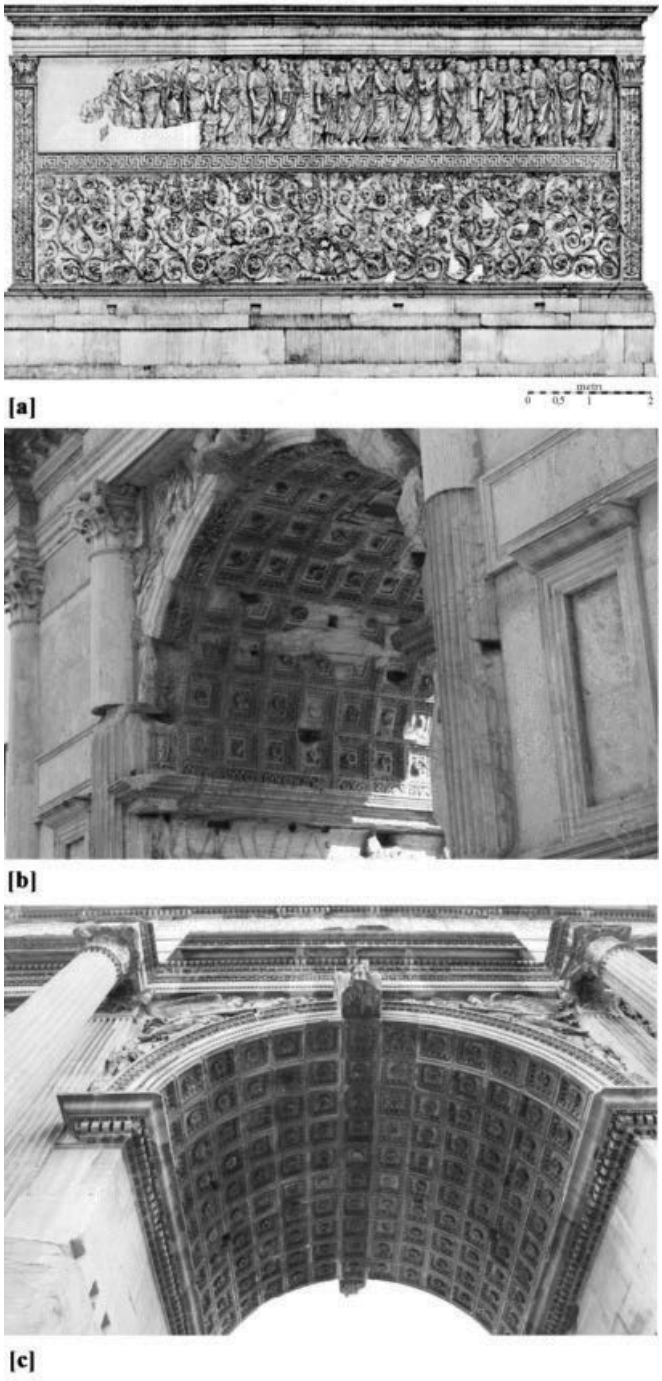

Fig. 3. The analyzed monuments: a) *Ara Pacis*; b) Arch of *Titus*; c) Arch of *Septimius Severus.*

Table 1. A Chronological bibliographic database of RF motifs in the earlier stages of their use.

Object	Date	Originated from	Located in	Part of	Place/ City
Temple of *Saturnus*	V cent. B.C. (restored in 42 B.C. and 283 A.D.)	Roman *Forum*	Roman *Forum*	coffered ceiling	Rome, Italy
Temple of *Castor* and *Pollux*	V cent. B.C. (restored in 117 and 73 B.C. and 6 A.D.)	Roman *Forum*	Roman *Forum*	coffered ceiling	Rome, Italy
Temple of *Apollo Sosianus*	V cent. B.C. (restored in 353 and 117 and 34 B.C.)	*Campus Martius*	*Campus Martius*	coffered ceiling	Rome, Italy
Gold Necklace	IV cent. B.C.	*Apulia*	British Museum	whole	London, England
The golden larnax	366 B.C.	Tomb of Philip II (Macedonia)	Museum of the Royal Tombs of Aigai	whole	Verghina, Greece
Drape with Acantino decoration	366 B.C.	Tomb of Philip II (Macedonia)	Salonika Museum	whole	Thessaloniki, Greece
Diadem	366 B.C.	Tomb of Philip II (Macedonia)	Salonika Museum	whole	Thessaloniki, Greece
Krater with a mythological scene	350 B.C. (Bari painter)	*Apulia*	British Museum	krater neck	London, England
Krater with the funeral of *Patroclus*	340 B.C. (*Darius* painter)	*Apulia*	British Museum	krater neck	London, England
Krater with *a naiskos* scene	325 B.C. (Baltimora painter)	*Apulia*	British Museum	krater neck	London, England
Mosaic with a hunting scene	316 B.C.	*Pella*	Pella Museum	edge	Pella, Greece
Tomb of *Lucius Scipio Barbatus*	280 B.C.	Sepulchre of the *Scipio* family	Vatican Museum	metope	Rome, Italy
Two marble reliefs	270 B.C.	*Pergamum*, Sanctuary of *Demeter*	Istanbul Museum	whole	Istanbul, Turkey
Mosaic	Eumen II age (197-160 B.C.)	*Pergamum*	Berlin Museum	edge	Berlin, Germany
Tombs of *Larthia*	150-130 B.C.	Chiusi	Archaeological Museum	metope	Florence, Italy

Table 1. continued.

Tombs of *Thanunia Seianti*	150-130 B.C.	Chiusi	British Museum, London	metope	London, England
Basilica Julia	I cent. B.C.	Roman *Forum*	Roman *Forum*	Frieze and coffered ceiling	Rome, Italy
Forum Julium	46 B.C.	Imperial *Forum*	Museum of Trajan markets	coffered ceiling	Rome, Italy
Venus genitrix temple	46 B.C.	Imperial *Forum*	Museum of Trajan markets	coffered ceiling	Rome, Italy
Marble *plutei* relief	First Augustan age (31 B.C.-10 B.C.)	*Horti Sallustiani*	Museum of Montemartini central	whole	Rome, Italy
Ara Pacis	9 B.C.	*Campus Martius*	*Campus Martius*	frieze	Rome, Italy
Forum of *Augustus*	2 B.C.	Imperial *Forum*	Museum of Trajan markets	coffered ceiling	Rome, Italy
Temple of *Mars Ultur*	2 B.C.	Imperial *Forum*	Museum of Trajan markets	capitel	Rome, Italy
Arch of *Titus*	81 A.D.	Roman *Forum*	Roman *Forum*	coffered ceiling of the vault	Rome, Italy
Forum of *Nerva*	97 A.D.	Imperial *Forum*	Museum of Trajan markets	coffered ceiling	Rome, Italy
Trajan’s Market	100-110 A.D.	Imperial *Forum*	Museum of Trajan markets	coffered ceiling	Rome, Italy
Trajan’s *Forum*	112 A.D.	Imperial *Forum*	Museum of Trajan markets	frieze with cupids	Rome, Italy
Temple of *Antoninus* and *Faustina*	141 A.D.	Roman *Forum*	Roman *Forum*	external frieze	Rome, Italy
Arch of *Septimus Severus*	203 A.D.	Roman *Forum*	Roman *Forum*	coffered ceiling of the vault	Rome, Italy

Apulian vases is connected to the period in which Taranto (*Taras*) reached its heights, being a forced waypoint for the commercial routes that connected Greece to Italy, through the Adriatic Sea (Cerchiai & al. 2002).

The most used shapes for the Apulian vases were the “volute-krater” and the *pseudo-panathenaic amphorae*, due to their monumental dimension. In the Apulian vases, the

recurrent motifs of the Greek culture were re-elaborated, as the Greek influence bloomed in the Apulian region (Becatti 1965). The subjects of these vases were taken from drama, tragedy and mythology (Trendal 1989). Here, all the floristic elements show the image of double or triple flowers connected with the previous one from the central part (Boardman 1966). Flowers and plants moved from the centre to the side and motifs of RF are usually placed in the lower register (Fig. 4). In the upper register, there are floristic elements, even though they are depicted in a static way (Todisco 2012).

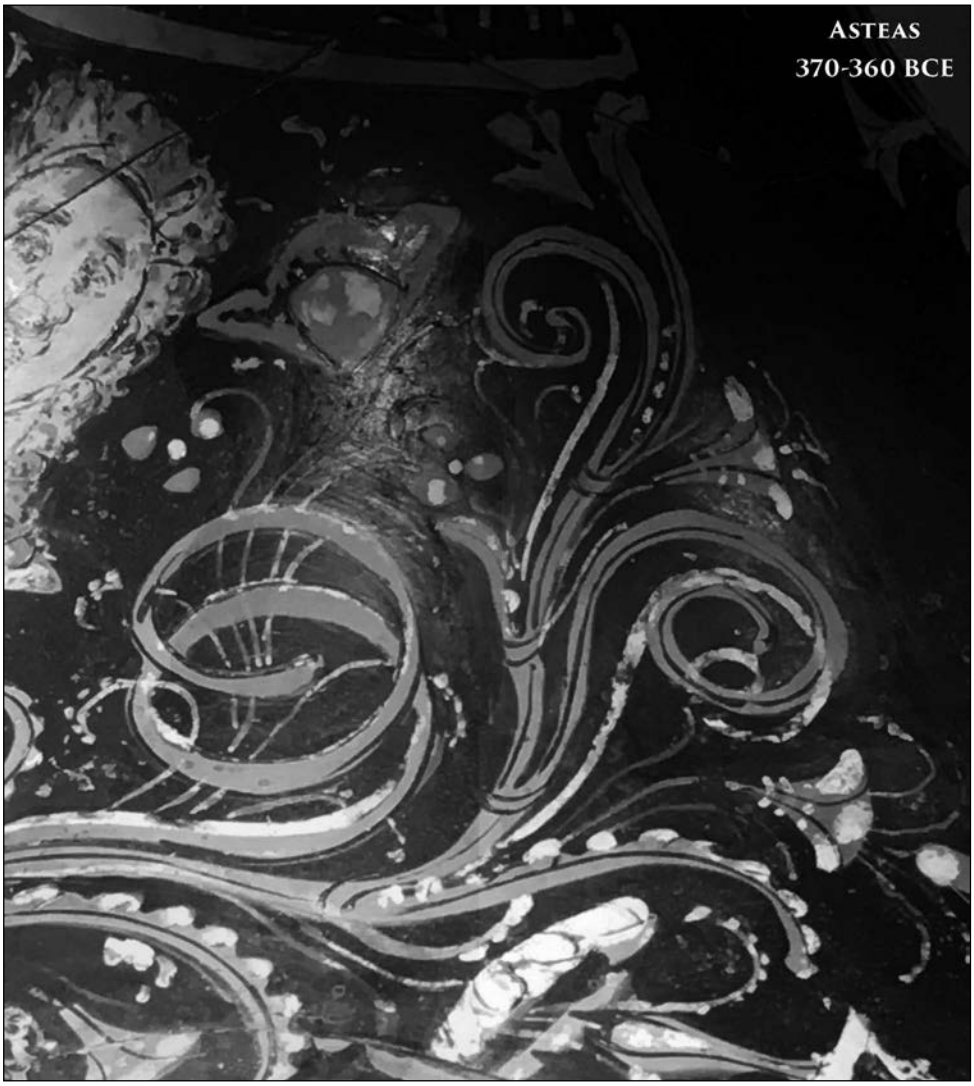

Fig. 4. An Apulian vase (Montesarchio, Archaeological National Museum of Sannio Caudino): Flowers and plants develop from the centre to the side.

A further example of the use of floristic elements is the gold necklace of the second half of IV century B.C., from the grave goods of a tomb from Taranto. The jewel shows a decoration of a triple flower that recalls the floristic elements in the ceramic production. This is a clear demonstration of the motif that switches from different materials (Masiello & Indellicati 2011).

The RF motif was also used in the funeral context like the case of so-called diadem of the princess Meda of Verghina (Macedonia) in one of the three tombs of the Royal *Tumulus* (350 B.C.). There, it was found a gold diadem with a Herculean knot, and a lace embellished with leaves, flowers and petals. It was also discovered a drape with an *acanthus* decoration enriched with flowers and tendrils. In this context, we have a clear archetype of the motif of the re-flowering flowers in both metal and textile materials.

In the earlier Roman context, we also find this motif in the Republican *sarcophagus* of *Lucius Cornelius Scipio Barbatus*, a member of the *Scipio* family that built the *sarcophagus* that was found in the tomb on the *via Appia*. The coffin, carved out in volcanic stone, is modelled on the type of the Greek altar, typical of southern Italy. Here we have a clear example of a mixture of architectural elements of an Ionic temple, provided by the use of volutes on the lid and a Doric temple by the presence of metopes filled with the re-flowering flower elements (Ramage & Ramage 2008). This shows how fast the circulation of art was in the Republican age, where a motif taken far away was re-elaborated and suddenly used in the local art.

A relevant example of the RF motif comes from the stylistic decoration of “*acanthus motif*”, which had a big fortune during the III century B.C. Their first examples are found in the tomb gravestones of Thebe, realized as an imitation of the temple trabeation in which the *acanthus* shoot was placed to fill the space in the architrave, *tympanum* and connecting lines of the *acroterion* (Castriota 1995). In the sanctuary of Demeter (270 B.C.) an elaborate vegetal frieze, carved in marble reliefs, arises from a central *acanthus* that develops axial stems and racemes, generating different plants and RF as deities in miniature (Centanni 2007). This composition, such as that on the *Pergamon* altar, had an important role in the study of the Roman art, also considering their symbolic functions (Castriota 1995). Remarkably the representation of the central *acanthus* seems to have a formal affinity with the *Ara Pacis,* built in honour of *Augustus.*

The botanical analysis of RF in the composition in Roman archaeological monuments

The recognized plants in the different examples of RF sum up to about 35 species in the three selected monuments, but this number represents only a part of the original floristic richness of such representations. The botanical composition and their approximate recurrence (Fig. 5) are shown in Tab. 2.

In general, the six external panels of *Ara Pacis* present a very rich flora in their composition, with about 100 species, as previously documented (Caneva 2010). The flora of the scrolls consists of 16 species, with dominance, for the external flowers, of species belonging to *Cardueae,* or the genera *Carlina, Lilium, Nymphaea, Anemone*, *Hedera, Anthemis* and *Scabiosa.* In the case of the central elements (the terminals), the most recurrent elements are fruits of *Asphodelus* and *Malva* or buds and shoots of various species (Tab. 2).

Both *Titus* and *Septimius Severus* arches showed, indeed, a rich coffered ceiling, which was built up respectively by 82 and 140 tiles containing the RF motif. Despite the large use

Fig. 5. Botanical details: External elements (a, b, c) from the *Ara Pacis* (scrolls, Caneva 2010) and (d, e) from the *Septimius Severus* Arch; Central elements (Terminals) (f, g, h) from the *Ara Pacis* scrolls (Caneva 2010) and (i, l) from the Arch of *Titus*.

Table 2. Re-flowering Flowers: Botanical elements in the Ara Pacis and Roman trium-phal arches.

Scientific names	**Features**	**External elements**			**Central elements**		
		Ara Pacis	**Titus Arch**	**SS Arch**	**Ara Pacis**	**Titus Arch**	**SS Arch**
1) *Carduea*	Capoline leaves, often thorny	***					
2) *Carlina* cfr. *utzka*	Capoline leaves with the typical shape	**	*	*			
3) *Lilium candidum*	Flowers (symmetry and number of elements)	**	*				
4) *Nymphaea* sp.	Flowers (morphology and number of elements)	**					
5) *Hedera helix*	Fruits (morphology of single elements)	**				*	*
6) *Lilium* sp.	Flowers morphology (or typical bulb structure)	*	*	*			
7) *Anemone* cfr. *apenina*	Flowers (symmetry and number of elements)	*	*	*			
8) *Anemone* cfr. *coronaria*	Flowers (symmetry and number of elements)	*	*	*			
9) *Scabiosa* sp.	Capoline (morphology of ligulate flowers)	*		**			
10) *Narcissus* sp.	Flowers (symmetry and shape)	*		*			
11) *Helianthemum* cfr. *nummularium*	Flowers (symmetry, margin shape and number of elements)	*					
12) *Alchemilla* cfr. *vulgaris*	Flowers (symmetry and number of elements)	*					
13) *Sedum* sp.	Flowers (symmetry and number of elements)	r					
14) *Ecballium elaterium*	Flowers (symmetry and number of elements)	r					
15) *Rosa* sp.	Flowers (symmetry and number of elements)	r					
16) *Anthemis* sp.	Capoline morphology with a combination of tubulate and ligutae flowers	*	*	*			
17) *Calendula arvensis*	Capoline morphology with many ligulate flowers		*	**			
18) *Silene* sp.	Flowers (symmetry, number of elements, typical petal shape)		**	**			
19) *Stellaria* cf. *media*	Flowers (symmetry and number of elements)		*	*			
20) *Chaerophyllum* cfr. *temulum*	Flowers (symmetry number of elements, typical petal shape)		*	*			

Table 2. continued.

21) *Cerastium* sp.	Flowers (symmetry and number of elements)			*			
22) *Punica granatum*	Calyx (typical shape)			**r**			
23) *Asphodelus* sp.	flowers and fruits (symmetry, number of elements, typical trimerous shape of the capsule)			**	**	*	*
24) *Malva* sp.	Fruits (shape of the mericarps)				**		
25) *Arum italicum*	ripening fruits (transformation of the inflorescence)				**		
26) *Nuphar luteum*	flower buds (typical shape)				*	*	*
27) *Cydonia oblonga*	Fruit (pommes with typical shape)				*		
28) *Panicum miliaceum*	Growing spikelet				*		
29) *Asparagus* sp.	Shoot (elongate growing stem)				*		
30) Orchids	Pollinodia, with elongate structure				**r**		
31) *Biarum tenuifolium*	Acute shoot				*		
32) *Tragopogon* sp.	Flower buds (typical elongate shape)				r		
33) *Petasites* sp.	Flower buds showing composite structure				**r**		
34) *Allium sativum*	Bulb composed by bulbils				*		
35) *Laurus nobilis*	Flower buds					**	**

Legend: ** = frequent, * = 2-3 representations; r = rare

of plant elements, especially in the cases of the tiles of the arches, the identification of most flowers to a specific taxonomic detail is now impossible, due to the bad state of conservation or to the insufficient carved details, and thus we can only suppose the genus or the family of the represented plants. As in the *Ara Pacis* case, the flowers of *Carlina, Lilium, Anemone*, and *Scabiosa* were recurrent in the external elements, but the most frequent were those of *Silene, Stellaria*, and *Calendula,* and also of *Chaerophyllum* and *Asphodelus,* which were rare or absent in the Augustan altar. In the case of central elements (the terminals), the most recurrent still resulted the fruits of *Asphodelus* and *Malva*, but also the flower buds of *Laurus nobilis*, which were absent in the *Ara Pacis* scrolls (Tab. 2).

The general structure is built up by external flowers with actinomorphic symmetry and the terminal elements with flower buds or shoots, and its build-up follows three schemes, respectively constituted by two, three, or four levels. In the *Ara Pacis*, a higher floristic richness is represented with a higher number of selected species. In both arches, the external flowers have from 6 to 14 petals, and the most common representation is that with six petals on the external level and a trimer bud on the internal level. The arch of *Septimius Severus* is in a better state of conservation and appears to have a high number of elements. They all have at least three levels

of composition: one external and two internal. Differing from the arch of *Titus,* the composition shows a more elaborate way of execution, and each flower presents a great variety of elements.

The symbolic meaning

In the case of the Apulian vases, the “plant volutes” and the spiralling branches have been associated with the goddess of fecundity (Zanker 1989). In fact, the ceramic production is related to the period in which *Taras* reached its height. The floral spring realized on the neck of the vases is the symbolic representation of this period. The most representative motifs of this production had shoots formed by undulate stems enriched with tendrils encircled in spirals, animated by flowers, corollas, palmettes, and dentate leaves in arabesque volutes; the symbolic representation of the female head in the middle of the floristic elements sets the human representation no longer in a mythological context, but indicating a close relation and fusion with nature (Becatti 1965). The RF motif is presented at the end of the path of the vegetal composition, reflecting the dynamic sense of a process which has no end. The plant decoration is the representation of an eternal blooming that starts with a flower, carrying on in another flower, with the idea of eternal prosperity of the city.

This meaning is evident in the case of monument representations. In fact, the monuments constituted the most relevant elements of a period, and the monuments are the most representative carriers of ideas, and messages of whom decided to build them. A building or its elements are strictly connected with whom committed its construction and with the message that the client wanted to convey. In this way, the single elements that are usually designed as a decorative motif, need to be considered in the same way as the inscriptions or the reliefs.

For the *Ara Pacis* and the monumental arches, the double or triple flower representation is an augural motif that evokes the idea of eternal blooming, in relation to time and its cyclic conception. The use of plant elements is connected with the new *Augustus* political program. In the plant representation, there are also deities’ attributes. Specifically, *Dionysus* and *Apollo* specific attributes are represented to celebrate their connection with the Attalid family. In this way, the metonymic presence of the deities in the vegetal frieze of the *Ara Pacis* recalls the *Pergamum*’s reliefs and the will of *Augustus* to be accompanied during its new political program and approved in lineage as *Divus filius* (Centanni 2007).

The architecture of the arches changed its purpose giving more emphasis to both political and propagandistic ideas of power, which was carried through inscription (Mazzilli 2016) and symbolic representations. The interior vaults, accompanied by the deification of the emperors, were made by a coffered ceiling, where the metamorphic flowers in the tiles stand as an augural signal reinforcing the idea of prosperity (Torelli & al. 2008). The recurrence of laurel flower buds in the final elements of the arches has a clear augural meaning of victory, in coherence with the aims of the arches theirselves.

Discussion

The origin and diffusion of the RF motif

Assyrian and Babylon cultures used the representation of metamorphic elements, which gave rise to chimeric animals and “tree of life” structures. The tree of life has been described in the book of the Genesis and in the Assyrian Babylonian culture as a symbolic representation

of the garden of Eden, the biblical paradise (Murphy & Murphy 2002). However, we cannot find examples in the Western Asian ancient representations, where flowers were used as single elements for their symbolic meanings (e.g., Ishtar Gate, Babylon) (Kleiner 2012).

The origin of the RF motif from the Apulian area can be explained considering the influence of Spartans colonization, occurred during the VII Century B.C., on the autochthonous culture, which created a new culture. As other southern Italian regions (Campania, Calabria and Sicily) of the so-called *Magna Graecia*, *Apulia* was inhabited by both indigenous and Greek people, and the local artistic production was an elaboration of previous elements merged together with the new ones (Giuliano 1989). The ceramic wares had a key role in this context, and they are considered a means through which the locals assimilated new messages as well as thoughts from Greece (Becatti 1965). First, the ruling class wanted to have the Attic ceramic for auto-representation needs, and then they started to create their ceramics, with a specific structure. Moreover, the ancient city was first ruled by a severe aristocracy that switched to democratic systems after military defeats (Masiello & Indellicati 2011). Moreover, during the time of the Pythagorean *Archytas* (mathematician and philosopher, which assumed a primary role in the city from 367 to 361 B.C.), *Taras* was open to sciences, arts and intellectuals, as testified by the great production of vases, sculptures, terracottas and jewellery (Trendal 1989).

In ancient civilizations, different aspects of other cultures have merged together and influenced each other, and artworks reflect something of the past that can be repeated or simply adopted with a new message. Considering these aspects, it is necessary to look at the phytomorphic images cautiously, considering all the little elements that compose the representation. Thus, there is a connection between the images presented and their meaning. What is shown through artistic representations is the result of many factors that connect various cultural elements within a community.

Then, from the III and II century B.C., when Romans conquered the Greek world, they started to be in contact with the Hellenic culture, which influenced religion, lifestyle and morality (Zanker 1989). Especially the artistic representations of the Republican period seem to have a relationship with the Greek-Hellenistic tradition, especially during the years of the conquest of Macedonia (146 A.D.) (Hölscher 1987). The members of the senator class were fascinated by the new culture, also renamed with the Latin term *asiatica luxuria* (the Latin noun *luxuria* is related to the abundance and wealth that Romans had seen at the Hellenic court). Moreover, the new noble families started to show their cosmopolitism as well as their political ambitions. The Republican years acquired a key role in the development of the Roman art, becoming a prelude of the *Augustus* age. In fact, later *Octavianus* kept many aspects of the Republican structure in his political program. This led the emperor to be in contact with the people, an aspect also reflected in art, where we can underline a relationship of offer and return. This event has to be considered as a key moment in the evolution of art since Rome opened the doors to an entirely new world that slowly disclosed its secrets (Hölscher 1987). In the spoils of the war after *Actium's* battle, around 31 B.C., there were not only riches and artworks but also, and most importantly, Egyptian's scientists, whom, heir of the Assyrian and Babylonian at first, then of the Greek culture, transmitted to the Romans their ancient knowledge.

The recurrence of the RF motif in many celebrative monuments, such as temples, basilicas and in the triumphal arches, can be explained considering the origin of the arch itself, and to their symbolic meaning, which will later be discussed.

The botanical analysis of the RF motif in the Roman archaeological monuments

The botanical identification seems useful to deepen and to contribute to the understanding of the role and use of the RF motif in the architectural decoration of the past. In fact, several flowers and different plant elements were used to create metamorphic structures, and each one had a specific aim, even if the relevance could be different (for more details, see Caneva 2010).

Among the most significant plant elements, we need to consider especially those located in the central area of the motif, i.e., the terminal part of the blooming process. Then, the carving of *Malva* and *Asphodelus* fruits can then be connected to their use in the ritual offering to the Apollonian ceremonies, giving them the meaning of hope of a return of the golden age (Detienne, 1975). Great value should be given to *Laurus nobilis* flowering buds in the central part of the RF motif in the triumphal arches since they express an Apollonian symbol of victory. The bulb of *Allium sativum* is a propitiatory element, known since antiquity for its therapeutic and magical properties, and Romans, such as other cultures, considered it as one of the most important medicinal plants. *Cydonia oblonga* fruits were dedicated to both *Hera/Juno* and *Aphrodite/Venus*, and they bore the significance of driving away the negative/bad influences. *Arum* and *Biarum* species seem elements linked to the idea of fertility and female divinities of maternity. Analogously, *Panicum miliaceum* was a cereal associated to the idea of fertility and *Ceres/Demeter*.

Dealing with the external flowers, as starting points of the metamorphic process, we have observed a high variety of elements. For example, *Cardueae* and related plants had the iconographic significance of reference to the bitterness of the earth, as emerged from biblical texts, by referring to their thorniness and their ability to thus protect themselves. Lilies constituted an emblem of beauty and fertility; ivy, as an example of the noblest blooming, was a Dionysian reference. Anemones were associated to the myth of *Adonis'* death and the idea of the ephemeral, and Narcissus to the idea of beauty, but also death, being a flower associated to the myth of *Narcissus.* Nymphs in general and lotus in particular represented, according to ancient eastern beliefs, the "flower of the sun, of creation and rebirth". *Silene* flowers were associated with the moon, but also with *Silenus,* who in mythology appears as the clumsy tutor of *Bacchus*. Flowers of *Asphodelus* had a different symbolic meaning with respects to their fruits, and according to Greek mythology grew in the Underworld, where "the shadows of heroes" walked, and that thus they were "flowers of the deceased" planted close to tombs. Their presences in the arches probably honour the deaths in the military campaigns, which concurred to the victory.

The overall symbolic meaning of the RF motif

In this context, the origin of the RF motif is coherent with one of the fundamental ideas of the Hellenic philosophy of the continuous metamorphosis, in particular in the pre-Socratic naturalism. As expressed by the speculative and deeply metaphysical genius of *Heraclitus*, *"to be is to flow"* and *"everything flows, nothing stays"; "the world would consist of a perennial transformation":* each one of us lives in that one develops, in that one continuously renovates. At the same time the becoming of the being is linked with the non-being, however *"this death is not a total dying, but a transmutation into something else".* The myth of transformation and metamorphosis was already mentioned in the Alexandrine literature, in *Callimachus'*, *Eratosthenes*'s, earlier than in *Virgilius*, *Catullus* and *Ovidius*. Starting from the Pythagoras's theory stating the never-ending transformation

in the universe, in *Ovidius*'s *Metamorphosis* is shown a lively nature where everything is changing in one eternal mutation, revealing the deceptiveness of the appearance.

The idea of a continuous metamorphosis of one element into another creates both a physical and ideological interconnection between the elements of nature and such a process of generation creates a sort of *unitary link between all the elements of the creation*, which is quite an important leading thread in the Hellenistic culture and the revived Pythagorean ideas of the Augustan age. The idea that the individual is only an element, a means but not an end, emerges from the observation of this continuous ending and transformation into something else contributes to giving the *idea of a starting point of further growths.* The richness of different plant elements contributes to the idea that nature follows such rules.

RF elements found a very suitable place in the ceiling of the arches, which express the purpose of emphasizing both political and propagandistic ideas of power (Mazzilli 2016). They become symbols of victory, and they find in the symbols of a never-ending flowering a perfect symbiosis.

Conclusions

The diffusion of the RF motif started during the Hellenistic period (IV Century B.C.). It became a dominant element in triumphal arches, and later also in coffered ceilings, forming the so-called "rosettes". Through the analysis of specific artworks and monuments, such vases, jewellery, marble reliefs and the Roman arches, we were able to confirm the symbolic value of nature, and its great diversity of plant elements. The floristic elements were often considered only as a decorative motif, neglecting the symbolic language linked to the primary role nature had in the ancient culture. It symbolically expresses the hope on an "eternal blooming" even if today its original meaning is lost.

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Article

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New records, distribution and taxonomic notes for non-native vascular flora of Tunisia – I. *Poaceae*

Abstract

El Mokni, R. & Verloove, F.: New records, distribution and taxonomic notes for non-native vascular flora of Tunisia – I. *Poaceae*. — Fl. Medit. 29: 45-53. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Six new taxa of the *Poaceae* family are added to the non-native flora of Tunisia: *Cenchrus longisetus* M. C. Johnst., *Digitaria ciliaris* (Retz.) Koeler, *Eleusine indica* (L.) Gaertn., *Eragrostis cilianensis* subsp. *thyrsiflora* (Willk.) H. Scholz & Valdés, *Eragrostis pilosa* (L.) P. Beauv. and *Paspalum dilatatum* Poir. Five of them are considered naturalized and horticultural trade is considered the most likely pathway for their introduction, whereas *Eragrostis pilosa* (L.) P. Beauv., might have been introduced as impurity in cereals for field crops. Populations of Tunisian *E. pilosa* are characteristic in possessing glandular pits, especially on inflorescence branches, a character not observed in *E. pilosa* s. str. in Europe and the Mediterranean area. Actual distribution and taxonomic notes are also given for all newly reported reported taxa.

Key words: chorology, *Cenchrus*, *Digitaria*, *Eleusine*, *Eragrostis*, *Paspalum,* North Africa.

Introduction

Interest on alien terrestrial flora in Tunisia has grown rapidly in the last two decades, with many new alien species having been reported (see e.g., El Mokni & El Aouni 2011a, 2011b, 2012, 2013, El Mokni & al. 2012, 2013, 2016; Sayari & al. 2016). Still, information on alien plants is incomplete and frequently updated with new records (see e.g., Iamonico & El Mokni 2016, 2017a, 2017b; El Mokni & Verloove 2017; El Mokni & Domina 2017, 2018, El Mokni & Véla 2017). According to recent research, more than 10 alien species were added to Tunisian terrestrial flora (see e.g., El Mokni 2018; El Mokni & Iamonico 2018). Despite the rather small size of the country (163,610 km²), the flora of Tunisia belongs to two major floristic subdivisions of the World (Takhtajan, 1986): Mediterranean Region (South Mediterranean province) encompassing the major part of the country, and Saharo-Arabian Region (Saharan province) that includes the southernmost part.

In the framework of the extensive field surveys aiming at updating and improving the knowledge on the Tunisian vascular flora, mainly focused on the coastal regions, many small populations of naturalized plants within the *Poaceae* were found within

coastal plant communities. *Cenchrus longisetus* M. C. Johnst., *Digitaria ciliaris* (Retz.) Koeler, *Eleusine indica* (L.) Gaertn., *Eragrostis cilianensis* subsp. *thyrsiflora* (Willk.) H. Scholz & Valdés, *Eragrostis pilosa* (L.) P. Beauv. and *Paspalum dilatatum* Poir.) are here reported for the first time from the country.

Materials and Methods

The work is based on extensive field surveys, analysis of literature, and examination of the specimens kept in the Herbarium of the Botanic Garden Meise, Belgium (BR) (acronym according to Thiers 2019+), and in the personal collection of one of the authors (R. El Mokni) which is deposited in the Herbarium of the Faculty of Pharmacy of Monastir and of the personal Herbarium of El Mokni. Reported taxa are presented alphabetically.

Results and Discussion

Cenchrus longisetus M. C. Johnst.
= *Pennisetum villosum* R. Br. ex Fresen.

Distribution and habitat

Originating from East Africa (Eritrea, Ethiopia, and Somalia), *C. longisetus* was introduced for ornament, widely used in public landscaping and became naturalized in the Mediterranean region (see i.e., Sommier & Caruana Gatto 1915; Domina & al. 2018; Galasso & al. 2018). It is increasing as a result of global warming (Fried 2012). Outside Europe, its invasiveness is widely recognized and it has naturalized in many warm parts of the world including America, Australia, and South Africa. In North Africa, *C. longisetus* was known so far in Morocco and Algeria as naturalized alien, whereas in Egypt it is noted as widely cultivated taxon (Valdés & Scholz 2009). It is here reported for the first time from Tunisia where it occurs since many years as naturalized weed in open grasslands and at some points alongside the railway leading to the Manzil Bourguiba city from Bizerta town.

Notes

This species is best recognized by its very showy inflorescences. These are whitish, plume-like, and the spikelets have tufts of silky hairs.

Examined specimens (new records)

TUNISIA: Bizerte (Center-Ville, N-E of Tunisia), 37°16’16” N, 09°52’41” E, 3 m a.s.l. 22 November 2010, *R. El Mokni* (Herb. Univ. Bizerta); Bizerte (Bab-Mateur, N-E of Tunisia), 37°15’55” N, 09°51’28” E, 4 m a.s.l. 23 September 2011, *R. El Mokni* (Herb. Univ. Bizerta); Bizerte (Sekma, N-E of Tunisia), 37°15’54” N, 09°51’13” E, 13 m a.s.l. 26 March 2013, *R. El Mokni* (BR, Herb. Univ. Bizerta).

Digitaria ciliaris (Retz.) Koeler

Distribution and habitat

Digitaria ciliaris is a weedy species, found in open, disturbed areas in most warm temperate and tropical regions of the world. Although still often overlooked, it is now known as a naturalized alien in many parts of southern Europe (cfr. Wilhalm 2009 for an extensive overview). In some areas, for instance in the Canary Islands, it is much more frequent than the related and very similar *Digitaria sanguinalis* (L.) Scop. Surprisingly, it is still hardly known in northwestern Africa where it may have been overlooked (cfr. Verloove 2016). In North Africa, it was reported for the first time from Algeria in 2007 (Zeddam & Scholz 2007), although it was shown subsequently to have been present in Morocco since at least 1947 (Verloove 2016). It is here reported for the first time from Tunisia where it occurs as widely naturalized weed in many plantation lots, annual cultivated areas, ornamental pots and plant nurseries.

Notes

Digitaria ciliaris has inflorescences on a long culm, usually much taller than the foliage, consisting of 2-9 racemes that are 5-10(-15) cm long. It has smooth marginal nerves of lower lemmas, without minute spines or with very few spinules in the upper one-third, a lower glume usually longer than 0.2 mm and upper surface of leaves usually glabrous or with some long-scattered hairs near base. *D. sanguinalis*, in contrast, has marginal nerves of lower lemmas with minute spinules, more or less throughout, lower glume usually shorter than 0.2 mm and upper side of leaves usually hairy throughout (rarely glabrous) (cfr. Verloove 2016).

Examined specimens (new records)

TUNISIA: Bizerte (Center-Ville, N-E of Tunisia), 37°16’16” N, 09°52’41” E, 3 m a.s.l. 22 November 2015, *R. El Mokni* (Herb. Univ. Bizerta); Bizerte (Route Panoramique, N-E of Tunisia), 37°18’48” N, 09°51’47” E, 17 m a.s.l. 24 December 2016, *R. El Mokni* (BR, Herb. Univ. Monastir); Bizerte (Av. Habib-Bourguiba, N-E of Tunisia), 37°16’12” N, 09°52’01” E, 10 m a.s.l. 24 December 2016, *R. El Mokni* (BR, Herb. Univ. Monastir).

Eleusine indica (L.) Gaertn.

Distribution and habitat

Eleusine comprises about 10 species, distributed in the tropical and subtropical parts of the world (De Wet 2006). This genus is represented by two neophyte taxa in Europe – *E. indica* and *E. tristachya* (Lam.) Lam. (Hansen 1980; Nikoli 2009). *E. indica* (Crow’s foot grass) is widespread throughout tropical and sub-tropical regions of the world. It has an African origin (Phillips 1972), although an alternative view states it was originally native to India (Holm & al. 1977; Holm & al. 1979; Waterhouse 1993). It is widely naturalized throughout the tropics, sub-tropics and temperate regions of the world, including Africa, Asia, SE Asia, Australia, the Pacific and the Americas (Waterhouse 1994).

In North Africa, *E. indica* was known so far as alien from Morocco, Algeria, Lybia and Egypt (Valdés & Scholz 2009). It is here reported for the first time from Tunisia where it occurs as a weed in irrigated public gardens, sidewalks, lawns, etc.

Notes

Eleusine indica is a variable species that is treated here in its traditional sense. Recent studies have suggested that *E. indica* subsp. *africana* is closer to *E. coracana* (L.) Gaertn. and is best subsumed under that species (as *E. coracana* subsp. *africana* (Kenn.-O'Byrne) Hilu & de Wet) (Hilu & Johnson 1997) or even considered a distinct species (*E. africana* Kenn.-O'Byrne; Peterson & al. 2015). Both are separated on spikelet size, glume nervation and shape and ornamentation of the seed. However, although typical plants of both taxa are easily distinguished, intermediate plants are often encountered and their separation therefore is rarely straightforward. This also applies to at least part of the material seen from Tunisia.

Examined specimens (new records)

TUNISIA: Ile de Djerba, Houmt Souk., within city, neglected area, 28 August 2005*, J. Lambinon* 05/Tu/628 (BR, LG); Nabeul (Hammamet-Sud, Cap-Bon, N-E of Tunisia), 36°24'34" N, 10°34'35" E, 10 m a.s.l., 21 December 2008, *R. El Mokni* (Herb. Univ. Bizerta); Ariana (Sokra, N-E of Tunisia), 36°50'06" N, 10°09'59" E, 33 m a.s.l., 07 August 2011, *R. El Mokni* (Herb. Univ. Bizerta); Bizerte (Route Panoramique, N-E of Tunisia), 37°19'24" N, 09°51'31" E, 38 m a.s.l., 03 September 2013, *R. El Mokni* (Herb. Univ. Bizerta); Bizerte (Bab-Mateur, N-E of Tunisia), 37°16'07" N, 09°52'05" E, 6 m a.s.l., 18 October 2013, *R. El Mokni* (Herb. Univ. Bizerta); Medenine (Jerba, S-E of Tunisia), 33°52'28" N, 10°51'39" E, 8 m a.s.l., 17 December 2014, *R. El Mokni* (Herb. Univ. Bizerta); Monastir (Centre-Ville, C-E of Tunisia), 35°46'16" N, 10°49'52" E, 15 m a.s.l., 23 December 2015, *R. El Mokni* (BR, Herb. Univ. Monastir); Nabeul (Hammamet-Sud, Cap-Bon, N-E of Tunisia), 36°26'26" N, 10°42'25" E, 10 m a.s.l., 17 January 2017, *R. El Mokni* (Herb. Univ. Monastir); Bizerte (Route Corniche-Kheyam, N-E of Tunisia), 37°19'31" N, 09°51'52" E, 6 m a.s.l., 22 January 2017, *R. El Mokni* (BR, Herb. Univ. Monastir).

Eragrostis cilianensis subsp. ***thyrsiflora*** (Willk.) H. Scholz & Valdés

Distribution and habitat

Originating from southern Europe, *E. cilianensis* was introduced in southern Canada, U.S.A, Mexico, Central America, Caribbean, Argentina, Bolivia, Brazil, Colombia, Ecuador, Paraguay, Peru, Uruguay, and Venezuela (Peterson 2001; Peterson & Boechat 2001). It has widely spread to other tropical and temperate areas around the world: South, East and Central Europe, North Africa, tropical and southern Africa, South-west islands of the Indian Ocean, West Asia and Arabian Peninsula, India and Pakistan, China, South-east Asia, Indonesia and Australia (Holm & al. 1977; Holm & al. 1997). *E. cilianensis* subsp. *thyrsiflora*, in contrast, was known so far only in France and Spain as native taxon with no reports for North Africa (Valdés & Scholz 2009). It is here reported for the first time from

Northern Africa where it occurs as a weed in cultivated lands of the Bizerta region in Northeastern Tunisia.

Notes

E. cilianensis is best recognized by its ovate panicle, 4-30 cm long, fairly dense, contracted, stiffly branched, usually with glands on pedicels and branchlets; crateriform glands on the leaf-margins and the rather stiff panicles of yellowish green or leaden grey spikelets. It closely resembles *E. minor* Host and both may intergrade. The latter usually has a less dense inflorescence, single-veined upper glumes (vs. 3-veined), narrower spikelets with less numerous florets and a more or less elliptical to ovoid caryopsis (vs. nearly orbicular). Subsp. *thyrsiflora* is distinguished from the nominal taxon by spikelets that are much elongated with more numerous florets (spikelets more than 16 mm long with 24-60 florets, vs. less than 16 mm long with 22-30 florets; Portal 2002).

Examined specimen (new record)

TUNISIA: Bizerta (Ghar El-Melh, N-E of Tunisia), 37°09'50" N, 10°08'26" E, 10 m a.s.l., 22 March 2015, *R. El Mokni* (BR, Herb. Univ. Monastir).

Eragrostis pilosa (L.) P. Beauv.

Distribution and habitat

E. pilosa is native to Europe, naturalized in North, Central, and South America (excluding Surinam). It occurs in disturbed habitats, often in wet sandy soils, along forest margins in sandy or gravelly sites and city sidewalks (Giraldo-Cañas & al. 2012). It is mostly found in coastal countries worldwide, but not in cooler northern temperate areas. It is widespread in Africa and temperate Asia, and naturalized in Australia and the Americas. In tropical Asia, it occurs in India, Indochina, Indonesia, Malaysia, Myanmar, Nepal, Pakistan and Sri Lanka. In Europe, it has been reported as a weed in, for instance, Albania, Austria, Bulgaria, Czechoslovakia, France, Germany, Greece, Hungary, Italy, Portugal, Romania, Russian Federation (European part), Spain, Switzerland, Ukraine and former Yugoslavia (Holm & al. 1997). In North Africa, *E. pilosa* was known so far in Morocco, Algeria, Lybia and Egypt (Valdés & Scholz 2009). It is here reported for the first time from Tunisia where it occurs as a weed in open grasslands subjected to excessive grazing of the Monastir region.

Notes

E. pilosa belongs to the *E. pectinacea-pilosa* group (Scholz 1996), a complex assemblage of variable, closely similar species. All are annuals with non-disarticulating rachillas, rounded or dorsally slightly compressed caryopses and micro-hairs in which the basal cell is only 0.8-1.6 times the length of the apical cell (Koch 1974). The plants recently found in Tunisia are very characteristic in possessing few to many glandular pits, especially on inflorescence branches, a character not observed in *E. pilosa* s. str. in Europe and the Mediterranean area. Within the *E. pectinacea-pilosa* complex this feature is encountered in three taxa: the North American *E. perplexa* Harvey, the Asian *E. amurensis* Probatova (syn.: *E. voronensis* Scholz) and the predominantly African *E. pilosa* subsp. *neglecta* Scholz (Scholz 1988, Seregin 2012). Tunisian plants correspond rather well with the latter,

a taxon known from Niger, Sudan and Pakistan but probably present elsewhere in the Sahel in Africa. The taxonomic value of the presence of glandular pits in *E. pilosa* and related species is unknown. According to Scholz (1988) they are never present in *E. pilosa* s. str., while other authors accept that they can be present in small number (e.g. Giraldo-Cañas & al. 2012). Further study is required, preferably using molecular techniques. For this reason, and at least for the time being, the Tunisian populations are assigned to *E. pilosa* s. l.

Examined specimens (new records)
TUNISIA: Monastir (Zeramdine-Ridène, C-E of Tunisia), 35°36'28" N, 10°41'52" E, 33 m a.s.l., 06 November 2016, *R. El Mokni* (BR, Herb. Univ. Monastir); Monastir (Jemmel-Birettayeb, C-E of Tunisia), 35°37'44" N, 10°44'37" E, 20 m a.s.l., 28 November 2016, *R. El Mokni* (Herb. Univ. Monastir).

Paspalum dilatatum Poir.

Distribution and habitat
P. dilatatum is a tufted perennial with short rhizomes and is of South American origin. It is introduced and naturalized in many Mediterranean countries (Clayton in Tutin & al. 1980). In North Africa, *P. dilatatum* was known so far in Morocco, Algeria and Egypt (Zeddam & Scholz 2007; Valdés & Scholz 2009). It is here reported for the first time from Tunisia where it occurs as a weed on the edges of certain non-permanent streams in the north-west and on coastal dunes immersed from time to time by waste water in the region of Bizerte.

Notes
Characteristic features of *P. dilatatum* are racemes 3-7(10), alternate, 6-8 cm long, axils of racemes pilose. Spikelets are obtuse, 3-4 mm long. Upper glume and lower lemma have a marginal fringe of relatively long white hairs (1.5-2 mm long).

Examined specimens (new records)
TUNISIA: Jendouba (Tabarka-Houamdia, N-W of Tunisia), 36°56'15" N, 08°47'31" E, 6 m a.s.l. 03 February 2012, R. El Mokni (Herb. Univ. Bizerta); Bizerte (Route Corniche-Sidi Salem, North-East of Tunisia), 37°17'21" N, 09°52'27" E, 1 m a.s.l. 19 August 2014, *R. El Mokni* (Herb. Univ. Bizerta).

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Article

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Découverte de *Christella dentata* (*Thelypteridaceae*) en Algérie

Abstract

Rebbas, K., Vela, E., Bougaham, A. F., Belharrat, A., De Belair, G. & Prelli, R.: Découverte de *Christella dentata* (*Thelypteridaceae*) en Algérie. — Fl. Medit. 29: 55-66. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Christella dentata (= *Cylosorus dentatus*, *Thelypteridaceae*, *Pteridophyta*), native to the tropical and subtropical regions of Africa and Asia, has been observed between Jijel and Bejaia in northeastern Algeria. Distribution, ecological notes and comparison with related taxa as *Cyclosorus interruptus* (*Dryopteris gongylodes* p.p.) are also provided. This is the first report of this species in North Africa.

Key words: Pteridophytes, biodiversity survey, flora of Algeria, North Africa, Important Plant Area.

Introduction

Le littoral montagneux du tell littoral algéro-tunisien constitue un point chaud de biodiversité dénommé « Kabylies-Numidie-Kroumirie » (Vela & Benhouhou 2007) et abrite de nombreuses zones importantes pour les plantes (Radford & al. 2011; Yahi & al. 2012). Cependant elles n'ont plus fait l'objet d'explorations botaniques depuis le milieu du 20^{e} siècle, jusqu'à une période plus récente marquée par un renouveau des découvertes taxonomiques ou chorologiques d'importance nationale voire nord-africaine (cf. Rebbas & Vela 2008; Martin 2008; Daoud-Bouattour & al. 2009; Laribi & al. 2009; Laribi & al. 2011; De Belair & Vela 2011; Haou & al. 2011; Vela & al. 2012a-2012b; Ouarmim & al. 2013; Hadji & Rebbas 2014; El Mokni & al. 2015a, 2015b; Vela & al. 2015) sans parler des nombreux xénophytes d'apparition et/ou de naturalisation récente (à titre d'exemple cf. Vela & al. 2013).

Malgré une reprise des recherches botaniques sur le terrain, en Kroumirie tunisienne, en Numidie littorale algérienne, et en quelques points de Kabylie (Béjaïa, Akfadou) de nombreux secteurs demeurent sous-étudiés dont la Kabylie des Babors, région montagneuse difficile d'accès (voir néanmoins Gharzouli & Djellouli 2005). Malgré une reprise des recherches botaniques sur le terrain, en Kroumirie tunisienne, en Numidie littorale algérienne, et en quelques points de Kabylie (Béjaïa, Akfadou) de nombreux secteurs demeurent sous-étudiés dont la Kabylie des Babors, région montagneuse difficile d'accès (voir néanmoins Gharzouli & Djellouli 2005).

A l'occasion des recherches sur la végétation rupicole, sources et suintement des rochers du littoral de petite Kabylie effectués dans le cadre des travaux d'une thèse de doctorat (cfr. Rebbas 2014), nous avons visité plusieurs reprises l'oued Ziama, situé à la limite administrative entre la wilaya de Béjaïa et celle de Jijel. Une première station a été découverte par l'un de nous (K.R.) en juillet 2007 sur un petit talus rocheux de la berge de l'oued au niveau d'une petite source. L'observation de jeunes pieds immatures de *Pteris cretica* et d'une fougère indéterminée nous encouragea à renouveler les prospections. Une nouvelle visite en août 2009, nous a permis de retrouver la première station, malheureusement les individus de la fougère inconnue, bien que mieux développés étaient toujours immatures, et l'individu de *Pteris* avait disparu suite à un petit éboulement. Quelques jours plus tard, en prospectant plus en amont, que nous avons découvert plusieurs individus bien développées directement dans la ripisylve de l'oued, et avons ainsi pu démarrer le travail d'identification. Ce n'est que plus récemment, pendant la préparation de cette publication, que deux autres stations, plus éloignées mais bien plus abondantes et mieux conservées, ont pu être découvertes en septembre 2018 et en mars 2019 (A.F.B. et A.B.), confirmant l'indigénat et la pérennité de l'espèce dans ce secteur.

Les flores classiques en usage en Algérie pour les Ptéridophytes (Maire 1952; Quézel & Santa 1962) nous conduisaient systématiquement vers *Dryopteris gongylodes* subsp. *propinqua* (R. Br.) Christensen, à défaut d'autre candidat potentiel. En revanche, les flores d'Europe et notamment l'ouvrage spécialisé le plus usité par les botanistes francophones (Prelli 2001) nous conduisaient vers *Christella dentata* (Forssk.) Brownsey & Jermy, là aussi à défaut d'autre candidat. D'après la synthèse récente de Dobignard & Chatelain (2010) pour le nord de l'Afrique (incluant Madère et Canaries), le premier nom est désormais retenu sous le nom synonyme de *Cyclosorus interruptus* (Willd.) H. Itô, tandis que le second est un nom retenu mais a aussi pour synonyme le nom désuet *Cyclosorus dentatus* (Forssk.) Ching, ce qui montre bien, sinon leur parenté réelle, du moins leur parenté longtemps supposée et la délicatesse de leur distinction. Le plus remarquable dans cet état de fait était que les flores du Maghreb ignoraient le second (seul le premier étant présent), tandis que les flores d'Europe (Madère et canaries incluses) ignoraient le premier (seul le second étant présent). Notre découverte du second en Algérie avait aussitôt été signalée à Cyrille Chatelain et Alain Dobignard qui ont ainsi pu l'ajouter dans leur « Suppléments et corrigenda aux volumes 1 à 4 » (in Dobignard & Chatelain 2013 : 380).

C'est suite à cela que nous détaillons ici l'identification de notre nouvelle fougère et que nous révisons la situation taxonomique et chorologique du genre *Cyclosorus* sensu lato (*Thelypteridaceae*) en Algérie, et par extension en Afrique du Nord puis en Méditerranée occidentale.

Description des sites et du matériel étudié

Les coordonnées sont fournies par GPS de localisation au sol. Les altitudes approximatives données par le GPS sont vérifiées et corrigées si besoin d'après un logiciel de cartographie avec modèle numérique de terrain. Les stations de *Christella dentata* sont les suivantes (Fig. 1) :
Algérie, Jijel, Ziama, Oued Ziama, 36.6533°N, 05.4541°E, 30 m, 20.08.2009, K. Rebbas;
Algérie, Jijel, Ziama, Oued Ziama, 36.6536°N, 05.4530°E, 30 m, 20.08.2009, K. Rebbas;

Fig. 1. Localisation géographique des stations de *Christella dentata* à l'est de la wilaya de Béjaia et à l'ouest de la wilaya de Jijel (carte réalisée avec GeoCAT, http://geocat.kew.org).

Algérie, Jijel, Ziama, Oued Ziama, 36.6536°N, 05.4511°E, 30 m, 20.08.2009, K. Rebbas;
Algérie, Jijel, Ziama, Oued Akherkhor, 36.6531°N, 05.4448°E, 20 m, 18.12.2018, K. Rebbas;
Algérie, Jijel, Ziama, Oued Akherkhor, 36.6541°N, 05.4453°E, 20 m, 18.12.2018, K. Rebbas;
Algérie, Jijel, Ziama, Oued Akherkhor, 36.6504°N, 05.4397°E, 40m, 23.12.2018, K. Rebbas;
Algérie, Jijel, Ziama, Oued Akherkhor, 36.6468°N, 05.4379°E, 50m, 23.12.2018, K. Rebbas;
Algérie, Jijel, Ziama, Oued Akherkhor, 36.6462°N, 05.4392°E, 60m, 23.12.2018, K. Rebbas;
Algérie, Jijel, Parc national de Taza, Oued Taza, T'bola, 36.70417°N, 5.60015°E, 170m, 11.09.2018, A.F. Bougaham et A. Bouchareb;
Algérie, Bejaia, Souk El Tenine, Oued Boulazazene, Taramanet, 36.6119°N, 5.3727°E, 50m, 19.03.2019, A.F. Bougaham et A. Belharrat;
Algérie, Bejaia, Souk El Tenine, Oued Boulazazene, Gar Dourar, 36.6069°N, 5.3777°E, 60m, 19.03.2019, A.F. Bougaham et A. Belharrat.

L'oued Ziama en petite Kabylie représente un bassin versant incluant la face nord de l'Adrar El-Alem (1167 m) et du Djebel El-Kouf (1369 m), l'ensemble du Kef Es-Safsaf et la face ouest du Djebel Mrada (617 m). Son embouchure rejoint la Méditerranée entre les villes de Ziama Mansouriah et de Melbou (ex-Les Falaises). Les stations sont situées entre un et deux kilomètres de l'embouchure, sur l'oued Ziama et son affluent rive gauche l'oued Akherkhor, un peu avant la confluence de ces deux derniers. Il s'agit d'un fleuve côtier à débit permanent constitué d'eaux claires à courant modérée. Le lit est principalement recouvert de galets et par endroit de limons fins. Les berges sont couvertes par une ripisylve d'Aulne glutineux (*Alnus glutinosa* (L.) Gaertn.) formant une galerie dense et en très

bon état de conservation. Nous avons dénombré environ deux centaines de touffes de *Christella dentata*, dont 110 sur la seule station la plus en amont.

La localité de T'bola se trouve le long de l'oued Taza dans la forêt domaniale de Guerrouche située au sein de la zone centrale du Parc national de Taza. La station est couverte par une forêt de Chêne zéen (*Quercus canariensis* Willd.) en bon état de conservation. Le sous-bois est assez diversifié avec principalement *Viburnum tinus* L., *Laurus nobilis* L., *Alnus glutinosa* (L.) Gaertn., *Rubus incanescens* (DC.) Bertol., *Hedera algeriensis* Hibberd, *Asplenium onopteris* L. Nous avons dénombré 130 touffes de *Christella dentata* sur une surface de 400 m^2 (Fig. 2).

La troisième station de *Christella dentata* a été découverte dans une ripisylve s'étendant le long de l'oued Boulezazene (affluent de l'oued Agarioune) qui prend source de Djebel Tababort (1969 m.), située dans le village de Tizi War dans la commune de Melbou de la Daira de Souk El Tenine. Les deux localités à *Christella dentata* sont caractérisées par une formation végétale dominée par *Alnus glutinosa* (L.) Gaertn., *Populus alba* L. et *Salix alba* L. Cette espèce pousse le long des anciens canaux d'irrigation destinés à arroser les vergers abandonnés. Le sous-bois est formé principalement par *Nerium oleander* L., *Rubus ulmifolius* Schott., *Dittrichia viscosa* (L.) Greuter, *Phragmites communis* Trin, *Hedera algeriensis* Hibberd et *Vinca major* L. Nous avons dénombré 80 touffes de *Christella dentata* dans la localité Taramanet et 28 touffes dans les gorges Gar-Dourar.

Dans ces sites le climat est pluvieux avec des précipitations annuelles moyennes comprises entre 1000 et 1200 mm (A.N.R.H. 1993) mais méditerranéen car l'été reste sec, avec moins de 40 mm en moyenne cumulés sur les trois mois (juin, juillet et août). Il se situe dans l'étage bioclimatique humide à hiver chaud au sens de Daget (1977).

Le long de l'oued Ziama, cinq espèces de ptéridophytes sont présentes en mélange sur au moins huit stations distantes chacune de 100 à 200 m. Il s'agit de: *Adiantum capillus-veneris* L., *Christella dentata* (Forssk.) Brownsey & Jermy, *Polystichum setiferum* (Forssk.) T. Moore ex Woyn., *Pteris cretica* L., *P. longifolia* L. (= *P. vittata* L.). La première (*Adiantum*) et la troisième (*Polystichum*) sont assez communes en Algérie, tandis que la dernière (*Pteris longifolia*) est signalée comme rare et présente, entre autres, en Petite Kabylie (Quézel & Santa 1962) où nous la connaissons de plusieurs sites le long de l'oued Agrioun (cascade de Kefrida, sources de Chaabet El Akra…). Récemment, une nouvelle station de cette ptéridophyte a été découverte à Ath Yanni dans la région du Djurdjura septentrional et une autre dans la région de Toudja (Béjaia) par un des auteurs (K.R.). *Pteris cretica* est en revanche très rare en Algérie et présente pour toute l'Afrique du nord seulement en petite Kabylie, dans les montagnes de la wilaya de Jijel (Battandier & Trabut 1905; Maire 1952; Quezel & Santa 1962). Enfin si *Cyclosorus interruptus* (= *Dryopteris gongyloides* subsp. *propinqua* auct.; *Nephrodium unitum* subsp. *callense* Trab.) est très rare dans tout le Maghreb (connu jusqu'à présent en Algérie seulement en Numidie littorale au bord des lacs de la région d'El Kala, (Desfontaines 1799; Battandier & Trabut 1905; Maire 1952; Quézel & Santa 1962) et au Maroc seulement dans les marais du littoral atlantique près de Larache (Jahandiez & Maire 1931; Maire 1952), *Christella dentata* (= *Cyclosorus dentatus*), quant à elle n'avait jamais été signalée au Maghreb, mais seulement des îles macaronésiennes (Azores, Madère, Canaries, Cap Vert), d'Espagne continentale (Andalousie, Galice), d'Italie (Campanie, Sicile) et de Crète (cf. Prelli 2001).

Fig. 2. *Christella dentata.* En haut : la plante et son biotope ; en bas : face supérieure et inférieure des pinnules (Photos: K. Rebbas, Oued Ziama, 20.08.2009).

Le matériel récolté est déposé à MPU (Montpellier, France) et ENSA (El Harrach, Alger), et dans ce dernier pour majeure partie sous l'acronyme GDB intégré à ENSA. Deux copies (iso-récoltes) sont déposées à PAL (Herbarium Mediterraneum, Palerme, Italie). De plus, des photos (ph) complétant le matériel GDB sont également consultables à l'adresse http://gdebelair.com/tax/zzpz.html#Thelypteridaceae [consulté le 23/12/2018]:

Polystichum setiferum (Forssk.) Woyn., Algérie, Jijel, Oued Ziama x O. Akherkhour, 2009-08-20, K. Rebbas, hEV n°0025 (MPU);

Christella dentata (Forssk.) Brownsey & Jermy, Algérie, Jijel, Oued Ziama, 2009-08-20, K. Rebbas, hEV n°0026 (MPU);

Christella dentata (Forssk.) Brownsey & Jermy, Algérie, Jijel, Oued Ziama, 2009-08-20, K. Rebbas, hEV n°0026bis (PAL);

Pteris cretica L., Algérie, Jijel, Oued Ziama x O. Akherkhour, 2009-08-20, K. Rebbas, hEV n°0027 (MPU);

Pteris vittata L., Algérie, Jijel, Oued Ziama x O. Akherkhour, 2009-08-20, K. Rebbas, hEV n°0028 (MPU);

Christella dentata (Forssk.) Brownsey & Jermy, Algérie, Jijel, Oued Ziama x O. Akherkhour, 2009-08-20, K. Rebbas, hEV n°0029 (MPU);

Christella dentata (Forssk.) Brownsey & Jermy, Algérie, Jijel, Oued Ziama x O. Akherkhour, 2009-08-20, K. Rebbas, hEV n°0029bis (PAL);

Christella dentata (Forssk.) Brownsey & Jermy, Algérie, Jijel, Oued Ziama, 36.6541°N, 05.4453°E, 20 m, 2018-12-18, K. Rebbas, RK n°0001 (ENSA);

Cyclosorus interruptus (Willd.) H. Itô, Algérie, El Tarf, lac Bleu, 36.91069°N, 8.34081°E, 16 m, G. de Bélair, 1996-10-06, ENSA/GDB n°029_14; 1990-10-01, ENSA/GDB n°033_06; 1993-09-15, ENSA/GDB n°033_08; 1997-11-03, ENSA/GDB n°075_19;

Cyclosorus interruptus (Willd.) H. Itô, Algérie, El Tarf, aunaie Oum el Agareb, 36.82189°N, 8.20965°E, 15 m, 1990-10-26, G. de Bélair, ENSA/GDB n°033_07;

Cyclosorus interruptus (Willd.) H. Itô, Algérie, El Tarf, aunaie Bou Glès, 36.82679°N, 8.21664°E, 22 m, G. de Bélair, 1992-05-08, ENSA/GDB n°033_09; 1991-06-23, ENSA/GDB n°033_12; 2005-12-23, GDB ph013_10; 2005-12-23, GDB ph013_11;

Cyclosorus interruptus (Willd.) H. Itô, Algérie, El Tarf, aunaie Righia 2, 36.85339°N, 8.18883°E, 22 m, 1989-06-19, G. de Bélair, ENSA/GDB n°033_10;

Cyclosorus interruptus (Willd.) H. Itô, Algérie, Skikda, aunaie Demnat Ataoua, 36.88987°N, 7.25918°E, 38 m, 1990-05-01, G. de Bélair, ENSA/GDB n°033_11;

Thelypteris palustris Schott, Algérie, El Tarf, garaet Esstah, 36.84253°N, 7.98214°E, 5 m, G. de Bélair, 1996-12-02, ENSA/GDB n°029_15; 1993-09-05, ENSA/GDB n°033_13; 1991-09-05, ENSA/GDB n°033_16; 1992-06-15, ENSA/GDB n°033_18 ; 1992-09-25, ENSA/GDB n°033_19; 1995-12-15, ENSA/GDB n°033_20; 1996-12-02, ENSA/GDB n°033_21;

Thelypteris palustris Schott, Algérie, Skikda, aunaie Demnat Ataoua, 36.88987°N, 7.25918°E, 38 m, G. de Bélair, 1991-09-21, ENSA/GDB n°033_15; 1993-06-07, ENSA/GDB n°033_14; 1991-11-24, ENSA/GDB n°033_17; 2014-06-13, ENSA/GDB n°108_25; 1993-06-07, GDB ph013_12; 2011-05-14, GDB ph013_13; 2012-12-21, GDB ph013_14; 2012-12-21, GDB ph013_15; 2014-06-13, GDB ph015_47;

Thelypteris palustris Schott, Algérie, El Tarf, Goureiate, 36.85388°N, 8.17392°E, 25 m, 1998-08-30, G. de Bélair, ENSA/GDB n°075_17;

Description taxonomique

Christella dentata a des feuilles en touffes, de 30-70 cm de longueur, à pétiole assez court; limbe fortement pubescent (sur les faces ainsi que sur les axes) nettement réduit à la base, à texture rêche; pennes incisées jusqu'à la moitié de la longueur des pinnules, celles-ci entières et à sommet arrondi. Nervation très particulière, la nervure basale de chaque pinnule s'unissant à celle de la pinnule voisine et formant une nervure verticale qui rejoint le sinus. Un très léger dimorphisme entre les feuilles stériles et fertiles, ces dernières plus longues et dressées, à pennes plus étroites. Sore ronds, à indusie réniforme et pubescent, les pédicelles des sporanges portant des glandes microscopiques oranges. Risque de confusion avec *Oreopteris limbosperma* et avec les *Dryopteris* du groupe de la Fougère-mâle; la pilosité du limbe et la nervation de *C. dentata* permettent de trancher. Plante vivace à développement printanier et fructification estivale; les feuilles persistent tout l'hiver et se dessèchent au cours du printemps suivant (Prelli 2001).

Cette espèce est facilement confondue avec *C. parasitica* (= *Cyclosorus parasiticus*) (Wagner 1950). Les plantes vivantes de *C. parasitica* sont plus pâles et d'un vert plus terne, et ont des pennes proximales plus étroites. Plus important encore, ses frondes ne sont pas dimorphes et n'ont pas la rupture nette entre les frondes stériles et fertiles comme *C. dentata*. Les feuilles stériles de *C. dentata* sont plus larges, plus courtes et plus étalées que les autres, et généralement entièrement dépourvues de sores. Une clé des espèces chinoises du sous-genre *Cyclosoriopsis* (incluant à la fois *Cyclosorus parasiticus* et *Christella dentata*) est fournie par Li & al. (2013).

La séparation des deux genres *Christella* et *Cyclosorus* quant à elle, se justifie dans la mesure où l'on suit la récente synthèse du Pteridophyte Phylogeny Group (2016), dans laquelle le genre *Christella* est maintenu indépendant des *Cyclosorus*.

Répartition géographique

Chirstella dentata est originaire des régions tropicales de l'Ancien Monde (répandues en Asie et en Afrique) et des régions tempérées d'Australie et de Nouvelle-Zélande, mais est adventive en Amérique du Nord et en Amérique centrale. Il a été décrit comme ayant une distribution cosmopolite tropicale, mais il se rencontre aussi dans des sites subtropicaux qui ne gèlent pas ou presque, survivant peut-être à quelques gelées par an dans le nord de la Floride (Iwatsuki 1963 ; McCarthy 1998).

C. dentata a été observée pour la première fois au Japon dans les années 1950 et s'est depuis propagée vers le nord (probablement en tant que contaminant de serre) à raison de 3 km par an, persistant pendant les hivers (Murakami & al. 2007). À Hawaii, l'espèce a été récoltée beaucoup plus tôt et a probablement été introduite et établie à la fin des années 1800 (MacCaughey 1918; Wagner 1950).

C. dentata est parfois considéré comme indigène dans l'île Lord Howe, de l'île Norfolk, des Tonga et de Tahiti, bien qu'une publication suggère qu'elle a été introduite à Tahiti (Iwatsuki 1963, Rodd & Pickard 1983 ; Murdock & Smith 2003 ; Meyer & al. 2009). Il a également été observé en Nouvelle-Calédonie, au Vanuatu, au Samoa et aux Îles Cook (CABI 2018).

En Europe, *C. dentata* est présente et indigène dans les régions suivantes : en Grèce sur l'île de Crète, au Portugal aux Azores et à Madère, en Espagne aux Canaries et sur la péninsule (Brownsey & Jermy, 1973 ; Prelli 2001). Elle est considérée comme douteuse au Maroc (Dobignard & Chatelain 2010) tandis qu'en Sicile et Italie péninsulaire elle est tantôt considérée comme douteuse (Pignatti 2017) tantôt comme xénophyte occasionnelle (Galasso & al. 2018), mais sans justification spécifique.

Sa présence en Afrique du Nord dans un site sauvage, en bon état de conservation, riche en espèces subtropicales (*Pteris vittata*, *Pteris cretica*, etc.), en plein cœur d'une région historiquement inexplorée sur le plan botanique, nous incite à la considérer comme parfaitement indigène et en position de relique.

Clé d'identification des *Thelypteridaceae* d'Afrique-du-Nord

Etant donné qu'aucun pays méditerranéen ni aucune rive continentale de la méditerranée ne présentait jusqu'à ce jour les deux espèces *Cyclosorus interruptus* et *Christella dentata*, aucune clé complète ne permet de les distinguer dans les flores usuelles en Europe et en Afrique du Nord. C'est en revanche le cas dans la flore de Chine, très riche en espèces du genre *Cyclosorus* sensu lato (Youxing & al. 2013).

Nous fournissons ici une clé des trois espèces de *Thelypteridaceae* connues en Afrique du nord continentale (Macaronésie exclue), et basée sur le modèle de la flore d'Algérie de Quézel & Santa (1962 : 24).

1. Feuilles adultes glabres (les jeunes feuilles sont légèrement poilues et glanduleuses). Pinnules minces, à nervures bien visibles par transparence, divisées jusqu'à la nervure. – Marais et bois marécageux – RR: K3: plaine des Guerbès-Senhadja (Ben Azzouz), mares de la Mafragh (Berrihane) – Subcosm. – (= *Polystichum thelypteris* (L.) Roth ; *Nephrodium thelypteris* (L.) Desv. ; *Dryopteris thelypteris* (L.) A. Gray): .... ***Thelypteris palustris*** Schott

1'. Feuilles densément poilues-glanduleuses en-dessous. Pinnules coriaces, à nervures peu visibles par transparence, à lobes plus ou moins profond mais n'atteignant jamais la nervure. Indusie couverte de poils denses : **2**

2. Feuilles adultes à limbe démarrant nettement à la base. Rachis des pennes avec grandes écailles isolées à la face inférieure. Pinnules divisées jusqu'à la moitié ou moins, avec 6 à 10 paries de nervures [Fig. 3a] – Marais et bois marécageux – R: K3: lacs et mares de La Calle à la Mafragh (El Kala, Bouteldja, Berrihane), Plaine des Guerbès-Senhadja (Ben Azzouz) – Trop. – « Maas » – (= *Nephrodium unitum* subsp. *callense* Trab.; *Polypodium unitum* Desf., non L. ; *Dryopteris gongyloides* (Schkuhr) Kuntze subsp. *propinqua* (R. Br.) Christ.) : ..... ***Cyclosorus interruptus*** (Willd.) H. Itô

2'. Feuilles adultes à limbe progressivement rétréci à la base. Rachis des pennes sans écaille. Pinnules divisées jusqu'à la moitié ou plus, avec 6 à 8 paries de nervures [Fig. 3b] – Bords de ruisseaux ombragés – RR: K2: Oued Ziama, Oued Taza – Subtrop. – (= *Thelypteris dentata* (Forssk.) E.P. St. John ; *Cyclosorus dentatus* (Forssk.) Ching) ..... ***Christella dentata*** (Forssk.) Brownsey & Jermy

Fig. 3a, 3b. Comparaison entre *Cyclosurus interruptus* (à gauche) avec écailles brunâtres éparpillées le long du rachis (photo E. Vela, legit G. de Bélair, Lac Bleu, 15.09.1993, GDB n°033_08) et *Christella dentata* (à droite) avec absence d'écaille brunâtre le long du rachis (photo E. Vela, legit K. Rebbas, Oued Ziama, 20.08.2009).

Conclusion

Tandis que *Cyclosorus interruptus* est confirmée pour l'Algérie dans la Numidie littorale (K3) où elle côtoie *Thelypteris palustris*, *Christella dentata* est une nouveauté pour la flore d'Afrique du Nord. Cette découverte enrichit davantage la ptéridoflore algérienne et vient relancer l'intérêt de la rechercher également au Maroc, notamment dans la péninsule tingitane et le Rif, tandis que *Cyclosorus interruptus* est signalée dans le Maroc atlantique nord (secteur « Man ») où elle côtoie là aussi *Thelypteris palustris* (Fennane & al. 1999).

Ce secteurs méconnus de l'oued Ziama apparaît en continuité entre deux zones importantes pour les plantes (ZIP), celle du parc national de Taza, dont fait partie l'oued Taza, et la ZIP mitoyenne de Babors-Kefrida (Yahi & al. 2012; Bouchibane & al. 2017 ; Benhouhou & al. 2018). La poursuite de ce travail visera l'exploration botanique d'autres secteurs de la Kabylie. Une généralisation des recherches sur les ptéridophytes de toute l'Algérie sera nécessaire à leur clarification taxonomique et nomenclaturale.

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Short note

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Emilio Badalamenti

Notes about the naturalization in Sicily of *Paulownia tomentosa* (*Paulowniaceae*) and remarks about its global spread

Abstract

Badalamenti, E.: Notes about the naturalization in Sicily of *Paulownia tomentosa* (*Paulowniaceae*) and remarks about its global spread. — Fl. Medit. 29: 67-70. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Paulownia tomentosa is native to different regions of East Asia. Here, the first case of natural regeneration in Sicily is reported. Although *Paulownia* is not still invading typically Mediterranean areas, mainly due to ecological constraints, its recognized invasiveness at a global level impose the need to monitor the ongoing process.

Key words: invasive alien species, woody plants, Mediterranean, Princess Tree.

Introduction

Biological invasions occupy a remarkable place among the causes of alteration of ecosystems and reduction of biodiversity on a global scale. The spread and impacts by invasive alien species (IAS) seem to be bound to exacerbating in the next decades, due to the increases in global trade and tourism pressure (Lambdon & al. 2008), as well as due to the promoting effect played by the climate change and the modification of disturbance regimes. IAS are particularly suited to face rapid changes in environmental conditions and disturbance is regarded as one of the most influential factors for invasive success. This helps explain why urban habitats, including Mediterranean ones, are particularly prone to plant invasions. Mediterranean coastal cities are very exposed to invasive species also due to the high rates of plant introduction and the many introduction pathways (Domina & Mazzola 2008; Badalamenti & al. 2014; Pasta & al. 2017), which considerably increased the likelihood of naturalization of alien plants there (Lambdon & al. 2008). Since urban areas have been sometimes the first sites where the naturalization of plant species occurred, even by those taxa which turned out to be invasive, the observation of the early signs of natural regeneration could be important even there. This could allow to follow the invasion process from the beginning, better understanding the possible evolution of invasive tendencies. In the last decades, the number of casual or naturalized alien trees has steadily increased in Sicily (e.g. Scafidi & al. 2016; Badalamenti & La Mantia 2018). As

part of specific surveys on alien flora of Sicily, the first records of natural regeneration of *Paulownia tomentosa* (Thunb.) Steud. (hereinafter *Paulownia*) are here reported.

Material and methods

In the last 4 years, periodic observations of mature trees were randomly carried out in green areas of Palermo (parks and gardens) where *Paulownia* is cultivated. Only individuals clearly originating from seed were considered. Systematics and nomenclature follow APG IV (2016).

Results and Conclusions

During periodic surveys ca. 10 self-sown individuals of *Paulownia* were found. Natural regeneration exclusively occurred at the base of walls or sidewalks, where some water and nutrients may accumulate. Seedling establishment was confined to ecological conditions characterized by thermal and water stress, in full light and low plant competition. The height of detected individuals ranged from 15-20 cm up to 1.5 m, and they have been found up to 120 m of distance from mother plants. *Paulownia* is the only genus within the *Paulowniaceae* family, encompassing six woody species coming from East Asia, including *Paulownia tomentosa* from East China, Korea and Japan (Zhao-Hua & al. 1986). *Paulownia* thrives in areas characterized by mean annual temperatures of 11-17 °C, mean annual rainfall of 500-1,500 mm and length of the dry period from 3 to 9 months. In the native habitats, the species colonizes mesophilous sites, such as forests, ravines and open valleys, and it represents a minor component of hardwood forests (Zhao-Hua & al. 1986). *Paulownia* is a pioneer species characterized by juvenile fast growth and sprouting ability, relatively short lifespan (60-70 years), early achievement of sexual maturity (8-10 years), and abundant production of easily wind-dispersed seeds (Essl 2007).

Paulownia has been widely introduced outside Asia for its impressive flowering, as well as for its hardiness to abiotic stresses and fast-growing. It was firstly introduced to Europe in 1834 (Ostinelli 1910), and from there to United States (US). Although these regions have shared the introduction period and its main purpose, they have been affected by notably different invasive patterns. In the US, the species underwent to a rapid naturalization process and has invaded man-made sites, but also semi-natural and natural habitats, such as xerophilous rocky cliffs, forest edges and gaps, and stream banks. *Paulownia* is extremely able to exploit disturbance (especially wildfires), giving rise to massive spread and large-scale invasion processes, threatening native species and ecosystems (Kuppinger & al. 2010). Other areas in the world where *Paulownia* has significantly escaped from cultivation include Hawaii and NE Australia (Randall 2017). In Europe, until recently *Paulownia* had been only found in disturbed and anthropic habitats such as railways, wall crevices, and ruderal vegetation, whereas rarely invading river banks or afforestations (Essl 2007). However, the notable increase in the records of natural regeneration in the last few decades suggests that the full naturalization in temperate central Europe, as well as the entry within semi-natural and natural habitats are highly expected (Essl 2007).

Paulownia tomentosa (sub *Paulownia imperialis* Siebold.) is present in Sicily at least since 1858, when it was cultivated in the Palermo Botanical Garden (Todaro 1858). Subsequently,

the species has been rarely used for ornamental purposes in green areas (parks and villas) and urban spaces (squares and streets). It is currently cultivated in Alimena, Isnello, and Palermo (province of Palermo), Trapani (province of Trapani), Mistretta and Vulcano (province of Messina) (Rossini-Oliva & al. 2002; Bazan & al. 2005; Domina & Mazzola 2008). *Paulownia* has been introduced to Italy in 1843 (Ridolfi 1843 in Maniero 2000). Nowadays, it is found in the floras of 15 out of 20 Italian regions (Galasso & al. 2018), where it has mostly invaded ruderal sites, dry and exposed areas and urban contexts (e.g. Olivieri 2017). Notably, the species has been included in the management list of invasive alien plants in Piedmont (DGR 33-5174, 12/06/2017), for its adverse impacts on biodiversity and buildings.

There is large uncertainty about the future behavior of *Paulownia* in Mediterranean-climate areas (Crosti & al. 2010). One of the possible reasons is its prevalent use as an ornamental species, which makes most natural environments physically unreachable. According to Essl (2007), both the origin and the naturalization process of *Paulownia* resemble the history of *Ailanthus altissima* (Mill.) Swingle; they were also found together invading the synanthropic vegetation of mesic habitats, as well as mixed broad-leaved forests. However, *Ailanthus* has proven to be particularly suited to establish within Mediterranean ecosystems (e.g. Badalamenti & al. 2015). Conversely, *Paulownia* does not seem to be equipped to cope with Mediterranean drought period. Indeed, the expected rise of average temperatures due to climate change, which likely will favor its spread northward, may represent one limiting factor in Mediterranean ecosystems as *Paulownia* would require water during the warm season (Zhao-Hua & al. 1986). However, introduced plants may experience considerable shifts in the ecological behavior even many years after the introduction.

In conclusion, the observation of some self-sown individuals suggests that *Paulownia* should be considered as a casual alien species in Sicily. Its recognized invasiveness at a global level impose the need to monitor the ongoing process (Randall 2017).

Specimina visa:

ITALY (SIC): Palermo, Campus of University (WGS84 38°06’16.10”N, 13°20’57.13”E), 44 m a.s.l., wall base, 10 May 2016, *E. Badalamenti s.n.* (SAF!).

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Short note

Fl. Medit. 29: 71-74
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H. M. A. Eltajoury, T. A. Mukassabi, F. M. Altera & A. M. Elmgasapi

Scolymus maculatus (*Asteraceae*) new for the Flora of Libya

Abstract

Eltajoury, H. M. A., Mukassabi, T. A., Altera, F. M. & Elmgasapi, A. M.: *Scolymus maculatus* (*Asteraceae*) new for the Flora of Libya. — Fl. Medit. 29: 71-74. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Scolymus maculatus L. (*Asteraceae*) is a species native to the Mediterranean basin found for the first time in Libya. It was discovered growing in a typical silty habitat with other mesophyte plants, very close to the shoreline, about 45 km east of the city of Benghazi. Records date back to July 2017 and June 2018.

Key words: flora, *Compositae*, Cyrenaica, North Africa.

Introduction

The *Asteraceae* is one of the largest plant families in the globe, and consists of more than 30 000 species, and 1900 genera (Jafri & El-Gadi 1983). *Scolymus* belongs to tribe *Cichorieae* Lam. & DC. and includes only three species: *S. maculatus* L., *S. hispanicus* L., and *S. grandiflorus* Desf. (Vazquez 2000). Globally, *Scolymus* occurs native in South Europe, North Africa and the Mediterranean basin (Vazquez 2000). Up to now only *S. hispanicus* and *S. grandiflorus* are reported from Libya (Greuter 2006). *S. hispanicus* is considered introduced and was collected from near Al Beyda located at 800 m above the sea level.

Indeed, the flora of Libya is not yet completely known and a number of new plant records are published each year (e.g. Mahklouf 2016).

Materials and Methods

Plant specimens were first collected from Elmabni area (32° 26’ 07.26” N, 20° 26’ 51.39” E, 2 m a.s.l.); forty-five km east of Benghazi, 600 m away from the left side of the main road towards east (Fig. 1a). This species was found during a vegetation analysis project in May 2017 and June 2018. The collected samples were first checked according to the Flora of Libya (Jafri & El-Gadi 1983). Moreover, further taxonomic investigations were done in the Cyrenaica Herbarium (CHUG) based on Flora of Egypt (Tackholm 1974; Boulos 2002) and Flora of Turkey (Davis & al. 1975). Plant

specimens were deposited in the Cyrenaica Herbarium (CHUG), Department of Botany, Faculty of Sciences, University of Benghazi.

Results

Collection and Taxonomy

The main differences between the three species of *Scolymus* are described by Vázquez (2000), since *S. maculatus* is an annual herb with more than five involucral leaves per capitula, capitula terminal and achenes without pappus (Fig. 1b). Whereas, the two other species *S. hispanicus* and *S. grandifloras* that are annual, biennial or perennial herbs, have three or less involuclar leaves per capitulum, capitula terminal or axillary and achenes with pappus. The population sampled belongs without any doubt to *S. maculatus*.

S. maculatus was found in relatively a large population, which covers an area of about eight hectares (Fig. 1c) and was obvious that the occurrence of this species dates back to at least 3-4 years or more. We belive that this spceis in Libya can be considered as an alien naturalized of recent introduction. Ten random quadrats of four m^2 were made within the concerned area to assess the density and frequency of *S. maculatus*. The individuals were found in 70% of quadrats made, with a density of 1.2 individual/m^2. This species dominates the whole area in association with: *Calendula* arvensis (Vaill.) L., *Lamarckia aurea* (L.) Moench, *Matricaria aurea* (Loefl.) Sch. Bip., *Malva parviflora* L., *Nicotiana glauca* Graham, *Paronychia arabica* (L.) DC. and *Suaeda aegyptiaca* (Hasselq.) Zohary. Some other species were found scattered within the site such as: *Diplotaxis muralis* (L.) DC., *Emex spinosa* (L.) Campd., *Plantago coronopus* L., *Silybum marianum* (L.) Gaertn. and *Vulpia bromoides* (L.) Gray.

General distribution

Scolymus maculatus is a species native to all countries of the Mediterranean including N. Africa, S. Europe, Crimea, S. Russia and Transcaucasia (Greuter 2006; African Plant Database 2012). The occurrence in Libya was expected since this species was known from all the neighbouring territories (Boulos 2002 ; Le-Floc'h & al. 2010; Bartolucci & al. 2018).

Site and climate description

It was discovered growing in a typical silty habitat in the field among other mesophyte plants and about 130 m south of halophyte communities, 2500 m south of the shoreline and 45 km east of the city of Benghazi. The climate in this area is primarily Mediterranean, very dry summers (June-August) and relatively wet winters (November-April). The highest mean monthly rainfall do not exceed 65 mm and usually in December and January. The mean annual rainfall within the last two decades is around 300 mm although very spatially erratic. The mean maximum monthly temperature reaches 41 ºC in June and decreases to 21 ºC in January. The lowest mean minimum monthly temperature is recorded in January and December at 6 ºC and 7 ºC, respectively (Mukassabi & al. 2017).

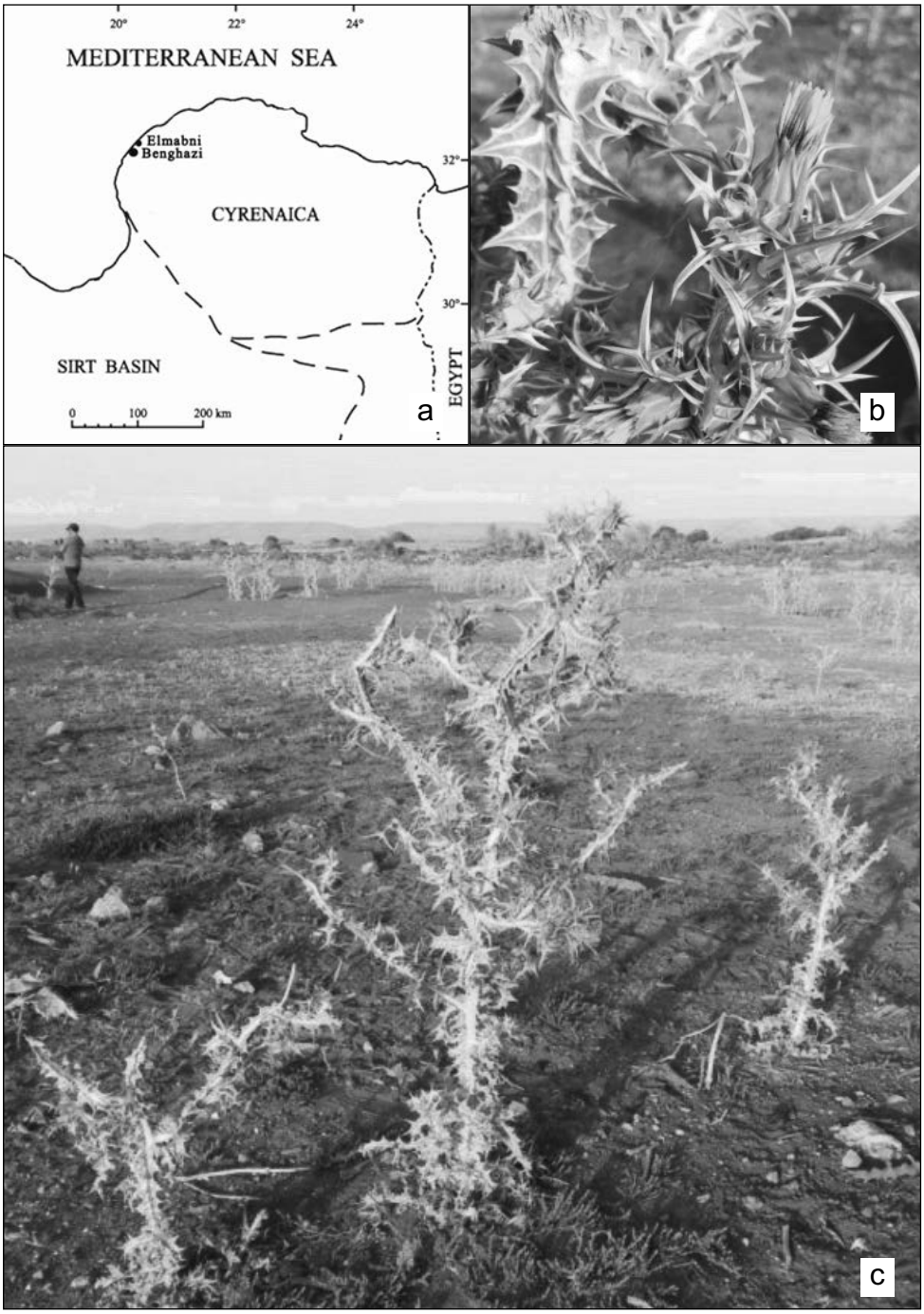

Fig. 1. a) The location of the new record of *Scolymus maculatus*; b) detail of stem and inflorescence; c) the population covers an area of about eight hectares (Photos by T. Mukassabi).

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Article

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Species diversity, chorology and conservation of the lichen flora in Tessala Mountains forest (North-West Algeria)

Abstract

Hamralaine, A. S., Benhassaini, H., Miara, M. D., Ait Hammou, M. & Hamralaine, O.: Species diversity, chorology and conservation of the lichen flora in Tessala Mountains forest (North-West Algeria). — Fl. Medit. 29: 75-91. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

This study is a first investigation of the lichen flora of the Tessala Mountains green oak forest in northwestern Algeria. Field research using stratified sampling method allowed us to propose a first checklist of the lichen flora of this region containing 53 taxa with taxonomic, chorological and biogeographical data in North-West Africa (Algeria, Morocco and Tunisia) of each of them. Among these lichens, 3 taxa are cited for the first time for the NW Africa. Comments on some interesting species are added, especially those with wide distribution reported currently only in Algeria, which are to be found in neighboring countries (Morocco, Tunisia). Finally, 17 taxa present in our list are protected by the Algerian law. The forest of Tessala Mountains must urgently benefit from an official protection status in order to preserve this lichen biodiversity of proven interest.

Key words: checklist, inventory, North-West Africa, *Quercus ilex* subsp. *ballota*, Sidi Bel Abbès.

Introduction

In North Africa, the lichen flora is mainly Mediterranean with a very high endemism (Werner 1955). It is essentially the region of folded mountains at the northwestern of the continent dominated by the Atlas Mountains with a Mediterranean climate (White 1996).

The study of lichen of Algeria is a relatively old (Amrani & al. 2015) dating back to the written first contribution in 1799 by René Desfontaines in his Flora Atlantica (1798-1799). Although there is no real flora of lichens in Algeria. Thus, the current knowledge of lichens in Algeria is still mainly based on the multitude of works carried out during the colonial period including: Montagne (1838); Durieu de Maisonneuve & Montagne (1846); Nylander (1854); Flagey (1892, 1895, 1896); Werner (1938, 1949); Faurel & al. (1951, 1953). Since the independence of the country, several studies have been interested in lichens: Ozenda & Clauzade (1970); Hertel (1971, 1987); Schwarz (1976); Leuckert & al. (1977); Clauzade & Roux (1984); Esnault (1985); Esnault & Roux (1987). The work of the

natives on this flora began with the study of Zouaoui (1989) on the lichens of Akfadou, then Semadi & al. (1997) in the region of Annaba and that of Rahali (2003) in the region of Algiers. Subsequently, some studies have been published mainly in the west of the country Bendaikha (2006); Ait Hammou & al. (2008, 2011, 2013); Khedim (2012); Khedim & al. (2013, 2018). Some studies have also been published in the east of the country (Rebbas & al. 2011; Serradj & al. 2013; Slimani & al. 2013; Ali Ahmed & al. 2018). Finally, we have to underline the very important recently published synthesis studies (Ait Hammou & al. 2014; Amrani & al. 2015; Amrani & al. 2018).

Specifically, Algeria, the largest country in Africa (2381741 km²) is rather poorly explored from a lichenological point of view and almost certainly contains many lichen taxa yet to be discovered (Ait Hammou 2014; Amrani & al. 2015).

The data on the Algerian lichen flora is still very incomplete and the revision of the lichen flora of Algeria must be encouraged (Khedim & al. 2018).

The region of the Mount of Tessala in western Algeria which containing a very rich flora (Bouiadjra & al. 2011) is floristically one of the least explored areas of the country (Saidi 2017). Indeed, the lichen flora of this region remains totally unknown and to our knowledge, no research study on lichens of this region has been published.

So, this study was considered in the main objective to bring new data to the country's lichen flora through the proposal of a first checklist of lichens of the Tessala Mountain, then to look for possible novelties within this list in particular in terms of chorology of taxa.

The Study area

The forest of Tessala Mountain also called "forest of El Attouche" is located in the North West of the country (Fig. 1) between three wilayas "provences": Sidi Bel Abbès, Oran and Ain Témouchent. It is part of the commune of Tessala located at 20 km from Sidi Bel Abbès city and covers an area of 2036 ha. The altitudes vary between 200 and 600 m a.s.l. on the dominant medium Mountains and up to 1060 m on the top of the Mountain (Bouiadjra & al. 2011).

The forest of Tessala includes vegetation dominated by holm oak. It is contains the following formations: preforests of *Quercus ilex* subsp. *ballota* (Desf.) Samp. and *Q. coccifera* L. in the East and South-East; scrubs formations of *Q. coccifera*, *Calycotome intermedia* Rchb.f. and *Chamaerops humilis* L. in the West and Southwest; degraded scrubs of *Calycotome intermedia*, *Chamaerops humilis* and *Ampelodesmos mauritanicus* T.Durand & Schinz in the center, East and in the extreme North-East; a clear forest of *Pinus halepensis* Mill. and *Quercus ilex* subsp. *ballota* on the high peaks (Saidi 2017).

The climate of the Tessala Mountains is Mediterranean as well as all the West Northern Algeria. It is characterized by concentrated rains during the cold period (autumn and winter, monthly average = 40.9 mm) and a marked drought during the warmer months (summer, monthly average = 1.76 mm). The bioclimatic levels vary according to the altitude from the arid to the sub-humid while the dominant level is the semi-arid (Ferka-Zazou 2006).

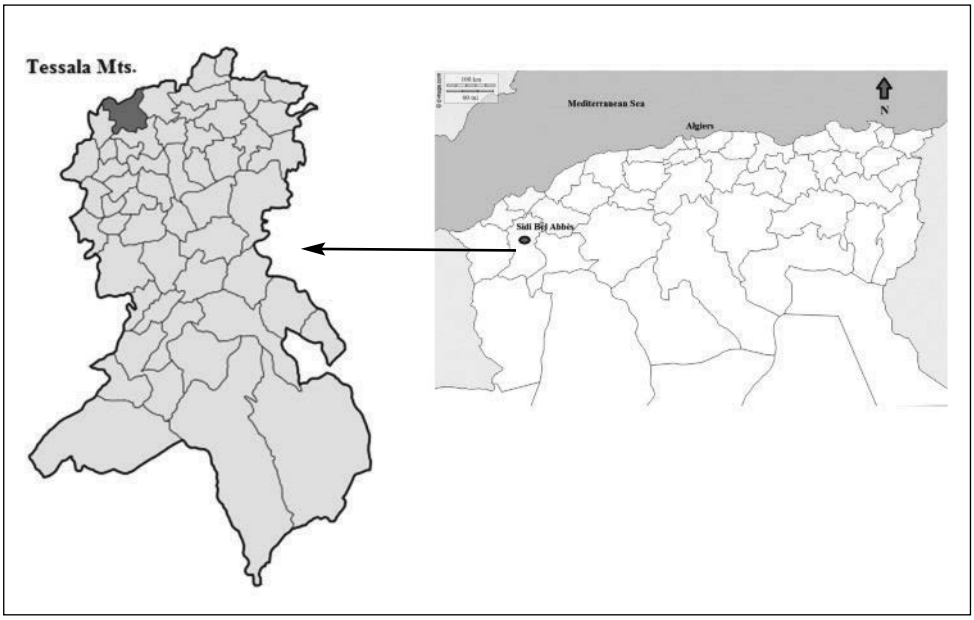

Fig.1. Location map of the study area.

Materials and Methods

Field work was started in 2013 using a stratified sampling method (Gounot 1969). Initially, we consulted the aerial photography of the region in order to explore the relief. The layer of vegetation being homogeneous (stands of holm oaks); we proceeded to the definition of the different environments of the site "strata": e.g. altitudes, exposure, topography. Then, in the field, we went through the zones previously defined. This allowed us to carry out sufficiently large lichen samplings in nine stations of one hectare (10000 m²) each "square 100 m long and 100 m wide". At these stations, survey corresponds to a part of a tree, mainly the trunk and the branches. According to Roux (1990), a maximum number of species are recorded in the field and sufficient samples are collected, especially those difficult to detect with the naked eye, and each phorophyt was georeferenced while the several parameters were recorded (height, circumference, position, density, slope, exposure, altitude, pH, sunshine). Within each station, terricolous, saxicolous and muscicolous lichens were also considered. Several special floras have been used for the botanical identification of taxa harvested in the field, including: Ozenda & Clauzade (1970); Boistel (1986); Hale (1990); Kirschbaum & Wirth (1997); Serusiaux & al. (2004); Jahns (2007); Aptroot & Schumm (2008); Krzewicka & al. (2009); Aptroot (2009) and Haluwyn & al. (2009, 2012). The nomenclature follows Roux & coll. (2017), while the collected specimens have been deposited in the herbarium of the laboratory of botany, the University of Tiaret.

In order to be able to search for possible new taxonomic and chorological novelties, we compare our results with the published works in the NW of Africa (Algeria, Morocco and Tunisia), by using all the available bibliography for Algeria: Durieu de Maisonneuve & Montagne (1846); Nylander (1854); Jourdan (1867); Flagey (1896); Zablbruckner (1904); Maheu (1928); Bouly de Lesdain (1907, 1911, 1939); Maire & Senevet (1928); Zahlbruckner (1930, 1931); Werner (1931, 1955); Reichert (1937); Dubuis & Faurel (1945); Faurel & al. (1951, 1953); Semadi (1989); Djebar & Fradjia (1992); Djellil (1989); Egea (1996); Boutabia (2000); Hamrlaine (2013); Bendaikha (2006); Mosbah (2007); Ait Hammou & al. (2008, 2011, 2014); Rebbas & al. (2011); Fadel & al. (2012); Khedim (2012); Slimani & al. (2013); Serradj (2013); Khedim & al. (2018); Ali Ahmed & Al (2018) and Amrani & al. (2018). For neighboring countries, we used mainly: Seaward (1996); El Mokni & al. (2015); Guttová & al. (2015) for Tunisia and Ajaj & al. (2013) for Morocco.

GPS limitation of sampled stations

Station 1

35°16’19.1”N 0°47’12.9”W
35°16’15.1”N 0°47’06.5”W
35°16’10.5”N 0°47’12.4”W
35°16’15.6”N 0°47’17.5”W

Station 2

35°16’20.7”N 0°47’17.5”W
35°16’26.7”N 0°47’20.7”W
35°16’24.3”N 0°47’28.5”W
35°16’19.3”N 0°47’23.4”W

Station 3

35°16’23.4”N 0°47’14.2”W
35°16’27.9”N 0°47’18.2”W
35°16’32.5”N 0°47’13.0”W
35°16’29.3”N 0°47’05.4”W

Station 4

35°16’34.3”N 0°47’04.0”W
35°16’27.4”N 0°47’05.1”W
35°16’25.0”N 0°46’56.6”W
35°16’31.6”N 0°46’54.2”W

Station 5
35°17'04.7"N 0°47'21.2"W
35°17'01.4"N 0°47'19.7"W
35°16'59.7"N 0°47'26.0"W
35°17'03.4"N 0°47'30.5"W

Station 6
35°17'00.4"N 0°47'32.4"W
35°16'55.3"N 0°47'34.5"W
35°16'57.1"N 0°47'42.2"W
35°17'02.5"N 0°47'43.4"W

Station 7
35°16'59.5"N 0°47'49.8"W
35°17'05.1"N 0°47'46.2"W
35°17'03.2"N 0°47'58.7"W
35°17'08.1"N 0°47'54.4"W

Station 8
35°16'51.4"N 0°47'46.4"W
35°16'44.3"N 0°47'43.3"W
35°16'42.5"N 0°47'52.6"W
35°16'52.3"N 0°47'53.6"W

Station 9
35°16'47.4"N 0°47'28.9"W
35°16'52.4"N 0°47'24.4"W
35°16'49.2"N 0°47'16.6"W
35°16'43.2"N 0°47'21.0"W

List of substrates and their abbreviations:
Co: Corticolous on *Quercus ilex* subsp. *ballota* (holm oak)
Te: Terricolous
Sa: Saxicolous
Mu: Muscicolous

Results and discussion

Checklist of lichen and lichenicolous fungi of Tessala Mountains

The presented list includes 53 species recorded in the study area and contains the following information: species name, botanical family, station numbers in which the lichen was recorded and substrate "in brackets", information on published data. For each species,

the known distribution in NW Africa (Algeria, Tunisia, and Morocco) is given with references. The new species reported for the first in NW Africa with asterisks (*):

Amygdalaria continua Brodo & Hertel
Lecideaceae; 5 (Sa); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Hamralaine (2013). No data published in Morocco and Tunisia.

Anaptychia ciliaris (L.) Körb. ex A. Massal.
Physciaceae; 1, 2 , 3, 4, 6, 9, 8 (Co); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Ait Hammou (2013); Bendaikha (2006); Boutabia (2000); Djellil (1989); Hamralaine (2007, 2013); Khedim (2012); Mosbah (2007); Slimani & al. (2013); Werner (1940, 1949, 1955); Alonso & Egea (2003); Fadel & al. (2012); **Tun**, Seaward (1996); El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).
Buellia coniops (Wahlenb.) Th. Fr.
Caliciaceae; 5 (Sa); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Hamralaine (2013). No data published in Morocco and Tunisia.
Caloplaca cerina (Ehrh. ex Hedw.) Th. Fr.
Teloschistaceae; 1, 9 (Co); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Haina & Bendechach (2004); Hamralaine (2007); Hamralaine (2013); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).
Caloplaca flavorubescens (Huds.) J.R. Laundon
Teloschistaceae; 4 (Co); Published data in NW Africa: **Alg**, Egea & al. (1990); Faurel & al. (1953); Haina & Bendechach (2004); Hamralaine (2013); **Mor**, Ajaj & al. (2013); **Tun**, Guttová & al. (2015).
Candelariella superdistans (Nyl.) Malme
Candelariaceae; 3 (Co); Published data in NW Africa: **Alg**, Hamralaine (2013); Nylander (1854); No data published in Morocco and Tunisia.
Collema furfuraceum (Arnold) Du Rietz
Collemataceae; 4, 5, 7 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Boutabia, 2000; Djellil (1989); Alonso & Egea (2003); Haina & Bendechach (2004); Hamralaine (2007, 2013); Rahali (2003); Semadi (1989); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).
Evernia prunastri (L.) Ach.
Parmeliaceae; 2, 4, 5, 7, 9 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Ait Hammou & al. (2011); Haina & Bendechach (2004); Hamralaine (2007, 2013); Khedim (2012); Rahali (2003); Rebbas & al. (2011); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).
Flakea papillata O.E. Erikss.
Verrucariaceae; 6 (Co); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Hamralaine (2013). No data published in Morocco and Tunisia.
Hypogymnia physodes (L.) Nyl. (= *Parmelia physodes* (L.) Ach.)
Parmeliaceae; 2, 3, 4, 6, 8, 9 (Co); Published data in NW Africa: **Alg**, Khedim & al. (2018); Brongniart (1882); Dubuis & Faurel (1945); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Mosbah (2007); Nylander (1854); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Lathagrium auriforme (With.) Otálora, P.M. Jørg. & Wedin

Collemataceae; 5, 6 (Co, Mu); Published data in NW Africa: **Alg**, Rebbas & al. (2011), Slimani & al. (2013), Amrani & al. (2018); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Lecanora allophana Nyl.

Lecanoraceae; 2, 4 (Co); Published data in NW Africa: **Alg,** Hamralaine (2013); Rebbas & al. (2011); Rehali (2003); **Tun**, El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Lecanora carpinea (L.) Vainio

Lecanoraceae; 3, 4, 6 (Co); Published data in NW Africa: **Alg**, Serradj & al. (2013); Boutabia (2000); Djellil (1989); Flagey (1896); Haina & Bendechach (2004); Merabti (2008); Mosbah (2007); Rebbas & al. (2011); Slimani & al. (2013); Werner (1949, 1955); **Tun**, Seaward (1996); El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Lecanora glabrata (Ach.) Malme

Lecanoraceae; 2, 3, 4, 6, 7, 9 (Co); Published data in NW Africa: **Alg,** Alonso & Egea (2003); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Werner (1940); **Mor**, Ajaj & al. (2013).

Lecanora dispersa (Pers.) Sommerf.

Lecanoraceae; 4, 7 (Sa); Published data in NW Africa: **Alg,** Ait Hammou & al. (2011); Bendaikha (2006); Egea & al. (1990); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2007, 2013); Merabti (2008); Mosbah (2007); Rahali (2003); Semadi (1989); Werner (1949, 1955); **Tun**, Seaward (1996); **Mor**, Ajaj & al. (2013).

Lecanora pulicaris (Pers.) Ach.

Lecanoraceae; 1, 2, 3, 4, 7, 8, 9 (Co); Published data in NW Africa: Alg, Bendaikha (2006); Djellil (1989); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Semadi (1989); Rahali (2003); Mor, Ajaj & al. (2013).

****Lecanora praesistens*** Nyl.

Lecanoraceae; 4, 3 (Co); No data published in NW Africa**.**

Lecanora rupicola (L.) Zahlbr.

Lecanoraceae; 6 (Sa); Published data in NW Africa: **Alg,** Djellil (1989); Dubuis & Faurel (1945); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Nylander (1854); Steiner (1902); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Lecidea exigua Chaub.

Lecideaceae; 5, 6 (Co); Published data in NW Africa: **Alg**, Boutabia & al. (2015), Amrani & al. (2018). No data published in Morocco and Tunisia.

Lecidella euphorea (Flörke) Hertel

Lecanoraceae; 6 (Co); Published data in NW Africa: **Alg,** Djellil (1989); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Merabti (2008); Rahali (2003); Semadi (1989); **Tun**, Seaward (1996); **Mor**, Ajaj & al. (2013).

Lepraria incana (L.) Ach.

Stereocaulaceae; 2, 3, 4, 6, 7, 8, 9 (Co); Published data in NW Africa: **Alg,** Serradj (2013); Bendaikha (2006); Alonso & Egea (2003); Haina & Bendechach (2004); Hamralaine (2013); Khedim (2012); Merabti (2008); Rebbas & al. (2011); Rahali (2003); Semadi (1989); Slimani & al. (2013); **Tun**, El Mokni & al. (2015).

Lichenomphalia umbellifera (L.: Fr.) Redhead, Lutzoni, Moncalvo & Vilgalys
Hygrophoraceae; 6, 7 (Co, Te); Published data in NW Africa: **Alg,** Khedim & al. (2018); Hamralaine (2013). No data published in Morocco and Tunisia.
****Multiclavula vernalis*** (Schwein.) R.H. Petersen
Clavulinaceae; 5 (Mu); No data published in NW Africa.
Opegrapha varia Pers.
Roccellaceae; 3, 4, 9 (Co); Published data in NW Africa: **Alg,** Bendaikha (2006); Haina & Bendechach (2004); Hamralaine (2013); Merabti (2008); **Tun**, Seaward (1996).
Parmelia horrescens (Taylor) Elix & Hale
Parmeliaceae; 2, 3, 4, 5, 6, 7, 8 (Co); Published data in NW Africa: **Alg,** Djellil (1989); Haina & Bendechach (2004); Hamralaine (2013); Werner (1955); **Tun**, El Mokni & al. (2015).
Parmelina carporrhizans (Taylor) Poelt & Vězda
Parmeliaceae; 1, 3, 4, 5, 6, 7, 9 (Co); Published data in NW Africa: **Alg,** Slimani & al. (2013); Hamralaine (2013); **Mor**, Ajaj & al. (2013).
Parmelina quercina (Willd.) Hale
Parmeliaceae; 2, 5 (Co); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Hamralaine (2013). No data published in Morocco and Tunisia.
Phaeophyscia hirsuta (Merschk.) Moberg
Physciaceae; 1, 2, 3, 4, 6, 7, 8, 9 (Co); Published data in NW Africa: **Alg,** Boutabia (2000); Alonso & Egea (2003); Fadel & al. (2012); Hamralaine (2013); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).
Phaeophyscia orbicularis (Neck.) Moberg
Physciaceae; 2, 3, 4, 5, 6, 7 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Boutabia (2000); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Mosbah (2007); Semadi (1989); **Tun**, Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).
Phlyctis argena (Sprengel) Flotow
Phlyctidaceae; 4, 5, 7 (Co); Published data in NW Africa: **Alg,** Serradj & al. (2013); Djellil (1989); Fadel & al. (2012); Haina & Bendechach (2004); Hamralaine (2013); Merabti (2008); Slimani & al. (2013); **Tun**, El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).
Physcia adscendens (Fr.) H. Olivier
Physciaceae; 1, 2, 3, 4, 5, 6, 7, 8, 9 (Co); Published data in NW Africa: **Alg,** Bendaikha (2006); Boutabia (2000); Fadel & al. (2012); Egea & al. (1990); Egea & Llimona (1991); Alonso & Egea (2003); Haina & Bendechach (2004); Hamralaine (2013); Khedim (2012); Merabti (2008); Rahali (2003); Rebbas & al. (2011); Semadi (1989); Slimani & al. (2013). No data published in Morocco and Tunisia.
Physcia albinea (Ach.) Nyl.
Physciaceae; 5 (Sa); Published data in NW Africa: **Alg,** Ait Hammou & al. (2011); Bendaikha (2006); Boutabia (2000); Haina & Bendechach (2004); Hamralaine (2007, 2003); Reichert (1937); Semadi (1989); Werner (1940); **Mor**, Ajaj & al. (2013).
Physcia biziana (A. Massal.) Zahlbr.
Physciaceae; 1, 2, 8, 9 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Bendaikha (2006); Boutabia (2000); Haina & Bendechach (2004); Hamralaine (2013); Reichert (1937); Semadi (1989); Werner (1940); **Tun**, Seaward (1996); Guttová & al. (2015); **Mor**, Ajaj & al. (2013).

Physcia aipolia (Ehrht.) E. Humb.
Physciaceae; 2, 3, 4, 6, 7, 9 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Ait Hammou & al. (2011); Djellil (1989); Alonso & Egea (2003); Haina & Bendechach (2004); Hamralaine (2007, 2013); Khedim (2012); Mosbah (2007); Semadi (1989); Slimani & al. (2013); **Tun**, El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Physcia caesia (Höffm.) Fürnrohr
Physciaceae; 5, 6, 7 (Sa); Published data in NW Africa: **Alg,** Egea & Llimona (1991); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Nylander (1854); **Tun,** Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Physcia dubia (Hoffm.) Lettau
Physciaceae; 4, 5, 6, 7 (Sa); Published data in NW Africa: **Alg,** Serradj & al. (2013); Bendaikha (2006); Egea & Llimona (1991); Haina & Bendechach (2004); Hamralaine (2007, 2013); Semadi (1989); **Tun,** El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Physcia stellaris (L.) Nyl.
Physciaceae; 1, 3, 4, 7, 8, 9 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Boutabia (2000); Haina & Bendechach (2004); Hamralaine (2013); **Tun,** Seaward (1996); **Mor**, Ajaj & al. (2013).

Physcia tenella (Scop.) DC.
Physciaceae; 3, 4, 5, 8, 9 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Ait Hammou & al. (2011); Bendaikha (2006); Boutabia (2000); Djellil (1989); Egea & al. (1990); Egea (2003); Fadel & al. (2012); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2007, 2013); Khedim (2012); Mosbah (2007); Rahali (2003); Semadi (1989); Slimani & al. (2013); **Tun,** Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Physcia tribacia (Ach.) Nyl.
Physciaceae; 3, 5, 7 (Co, Sa); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Hamralaine (2013); **Mor**, Ajaj & al. (2013).

Physconia detersa (Nyl.) Poelt
Physciaceae; 5 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Dubuis & Faurel (1945); Flagey (1896); Haina & Bendechach (2004); **Mor**, Ajaj & al. (2013).

Physconia grisea (Lam.) Poelt
Physciaceae; 3, 4, 5, 7 (Co); Published data in NW Africa: **Alg,** Bouly de Lesdain (1939); Boutabia (2000); Alonso & Egea (2003); Fadel & al. (2012); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Mosbah (2007); Rahali (2003); Semadi (1989); **Tun,** El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Physconia perisidiosa (Erichs.) Moberg
Physciaceae; 1, 5, 9 (Co, Mu); Published data in NW Africa: **Alg,** Khedim & al. (2018); Alonso & Egea (2003); Hamralaine (2013); Haina & Bendechach (2004); Semadi (1989); **Tun,** Seaward (1996); **Mor**, Ajaj & al. (2013).

**** Physconia distorta*** var. ***subvenusta*** Cromb.
Physciaceae; 4, 6 (Co); No data published in NW Africa**.**

Pseudevernia intensa (Nyl.) Hale & W.L. Culb.
Parmeliaceae; 4, 6, 7 (Co); Published data in NW Africa: **Alg,** Haina & Bendechach (2004); Hamralaine (2013). No data published in Morocco and Tunisia.

Ramalina canariensis Steiner

Ramalinaceae; 4, 5 (Co); Published data in NW Africa: **Alg,** Serradj & al. (2013); Djellil (1989); Haina & Bendechach (2004); Hamralaine (2013); Semadi (1989); Slimani & al. (2013); **Tun,** Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Ramalina farinacea (L.) Ach.

Ramalinaceae; 2, 3, 4, 6, 7, 8 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Serradj & al. (2013); Alonso & Egea (2003); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Khedim (2012); Rebbas & al. (2011); Slimani & al. (2013); Werner (1940, 1949); **Tun,** Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Ramalina fastigiata (Pers.) Ach.

Ramalinaceae; 1, 2, 4, 6, 8, 9 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); Serradj & al. (2013); Alonso & Egea (2003); Flagey (1896); Haina & Bendechach (2004); Hamralaine (2013); Khedim (2012); Rebbas & al. (2011); Slimani & al. (2013); Werner (1940, 1949); **Tun,** Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Ramalina celastri (Sprengel) Krog & Swinscow

Ramalinaceae; 5, 6, 7 (Co); Published data in NW Africa: **Alg**, Ait Hammou & al. (2011); Hamralaine (2013). No data published in Morocco and Tunisia.

Ramalina reagens (B. de Lesd.) W.L. Culb.

Ramalinaceae; 4, 5 (Co); Published data in NW Africa: **Alg,** Khedim & al. (2018); **Tun**, Seaward (1996); El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Usnea florida (L.) F.H. Wigg**.**

Parmeliaceae; 2, 4 (Co); Published data in NW Africa: **Alg,** Flagey (1896); Haina & Bendechach (2004); Nylander (1854); Trabut (1928); **Mor**, Ajaj & al. (2013).

Xanthoparmelia conspersa (Ehrh. ex Ach.) Ach.

Parmeliaceae; 3, 6, 9 (Sa); Published data in NW Africa: **Alg**, Ali Ahmed & al. (2018), Amrani & al. (2018); **Tun,** El Mokni & al. (2015).

Xanthoria fallax (Hepp)

Teloschistaceae; 1, 4, 6, 8 (Co, Sa); Published data in NW Africa: **Alg**, Hamralaine (2007); **Tun,** El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Xanthoria parietina (L.) Ach.

Teloschistaceae; 1, 2, 3, 4, 5, 6, 7, 8, 9 (Co, Sa); Published data in NW Africa: **Alg**, Khedim & al. (2018); Boutabia (2000); Fadel & al. (2012); Hamralaine (2007, 2013); Khedim (2012); **Tun,** Seaward (1996), El Mokni & al. (2015); **Mor**, Ajaj & al. (2013).

Floristic richness

Field investigations lead us to sample 532 phorophyts "trees" at the 8 stations visited across the Tessala Mountains. It allowed identifying in total 53 lichen taxa growing in this region. These taxa belong to 16 different families dominated by *Physciaceae* (16 species), *Parmeliaceae* and *Lecanoraceae* (8 species each).

Although the number of 53 lichens may seem interesting for a region with a predominantly semi-arid bioclimate (Saidi 2017), the great part of these species are nitrophityc, or ubiquists lichens, generally distributed (Roux & al. 2017). Indeed, this flora is strongly masked by foreign elements of mainly temperate origin. The temperate element, which arrived before, at the

time of the glaciations or after, is substantially equal in the three territories (Algeria, Morocco and Tunisia), although at a slightly higher percentage in Algeria (Werner 1955).

Otherwise, this number is much important than that reported by Ait Hammou & al. (2013) for green oak lichens in the semi-arid region of Tiaret (29 species).

Chorology and biogeography

The analysis of the obtained data following this research allowed us to identify 3 species that we report for the first time to NW Africa. These are the following taxa: *Lecanora praesistens, Multiclavula vernalis* and *Physconia distorta* var. *subvenusta*. These reports indicate first of all the richness of the lichen flora of Algeria, but also the fact that the Algerian lichen flora is poorly known (Khedim & al. 2018).

Comments on some interesting species

Several species in our list are listed either for the first time in NW Africa (*Lecanora praesistens, Multiclavula vernalis, Physconia distorta* var. *subvenusta*) or they are cited only in Algeria and missing in neighboring countries (*Amygdalaria continua, Buellia coniops, Candelariella superdistans, Flakea papillata, Lecidea exigua, Lichenomphalia umbellifera, Intense Pseudevernia* and *Ramalina celastri*).

Lecanora praesistens is a corticolous lichen on deciduous or coniferous trees, in moderately humid, well-lit and sunny biotopes. It is characterized by ellipsoid spores, simple, colorless, 12-16, 10-17 × 6-9 µm. In the thallin rim are large crystals clearly visible in polarized light and soluble in N but not in K. It differs from *L. chlarotera* Nyl. present in Tunisia and Morocco (Seaward & al. 1996; Ajaj & al. 2013) which shows only eight spores, coarse crystals in epithecium, large crystals in the thallin (P-) edge of apothecia. *Lecanora allophana* Nyl . and *L. pulicaris* (Pers.) Ach. reported both in Morocco (Ajaj & al. 2013) are distinguished from *L. praesistens* by the fact that the first has eight spores, an epithecium devoid of crystals, small crystals in the thallin (P-) edge of apothecia, then that the second also has fine crystals in the epithecium but the spores are eight, and the thallin edge of the apothecia is most often (P +) red (AFL 2019).

Multiclavula vernalis is one of a relatively small number of lichen-forming basidiomycetes, i.e. fungi that incorporate cells of a green alga within their tissues. The species remains to be searched in neighboring countries. The genus *Multiclavula* has not been quoted before in Algeria, Morocco and Tunisia.

Physconia distorta var. *subvenusta* is distinguished from *P. distorta* (With.) J. R. Laundon by its apothecia with lobules; the underside of the lobes is black as in *P. distorta*; in *P. venusta* (Ach.) Poelt, the underside of the lobes is white.

Amygdalaria continua is a species characterized by a continuous thallus with a finely smooth, rarely cracked or cracked texture, usually thick brownish gray with black tips. It can be recognized by the absence of soredia or by Apothecia entirely immersed, sunk in the thallus and no chemical staining (K-, C-, KC-, P-). It differs from *Amygdalaria consentiens* var. *consentians* with a cracked and cracked thallus and *Amygdalaria consentiens* var. *japonica* with a cracked thallus containing stictic and constictic acidss (Brodo & Hertel 1987). According to Amrani & al. (2018), *A. continua* is one of the taxa cited in the literature for Algeria with erroneous or doubtful records unsupported by herbarium material. But here we confirm his presence in our country. Genus and

species do not appear in the lists published in the neighboring countries including Seaward & al. (1996) and Ajaj & al. (2013).

Buellia coniops is a calcareous lichen, but can be found on non-calcareous (mainly granitic) rocks on the seacoasts. It is acidophilic, nitrophilous and halophilic (Roux & al. 2017). It has spores 1 times septated with uniformly thick, finely punctuated walls, 13-18 × 7-9 µm . This species is to be distinguished from *B. atrocinerella* (Nyl.) reported in Morocco (Ajaj & al. 2013) which has spores by 8, brown, uniseptated, 14.9-18 x 9.5-10.8 µm.

With its small, scattered yellow fruits (no hint of orange) and an inapparent thallus, *Candelariella superdistans* appears superficially similar to *C. aurella* (Hoffm.) Zahlbr. present in Morocco and Tunisia (Seaward & al. 1996; Ajaj & al. 2013), but that species has a paraplectenchymatous thalline margin without protruding hyphae.

A few years after the description of *Flakea papillata* in 1992, the species was included in the genus *Agonimia* by Aptroot & al. (1997), but the molecular data of Muggia & al. (2010) show that the taxon should be kept in a separate genus. *F. papillata* differs from other *Agonimia* species by its thallus structure. These are usually squamous-leper (Eriksson 1992, Perlmutter 2006). This species remains to be found in the two neighboring countries.

The epithecium of *Lecidea exigua* is highly charged with crystals is very similar to *Lecidella elaeochroma* found in Morocco and Tunisia (Seaward & al. 1996; Ajaj & al. 2013) but apothecia and spores are smaller and especially narrower.

Lichenumphalia umbilefera is a basidiolochen with a granular, green, conspicuous thallus; the granules are agglomerates of algae (*Coccomyxa*) traversed by thick-walled hyphae (AFL, 2019). The species has only been indicated in Algeria (Khedim & al. 2018) and remains to be searched in neighboring countries. So far, there is no other species of the genus *Lichenumphalia* in NW Africa.

Pseudevernia intensa is characterized by a thallus sometimes exceeding 10 cm in length, often during, formed of 2-5 mm wide, branched strips, with a greyish upper surface and a black underside, canaliculate, with curved edges. This thallus has a dorsiventrale organization (like the foliaceous) and no isidies on its upper face. Coloring: K-, KC-, P-, C +. *Pseudovernia furfuracea* (L.) Zopf .Beih.Bot is a close species present in Morocco and Tunisia (Seaward & al. 1996; Ajaj & al. 2013) and shares the same morpholigic characteristics, but only the thallus does not stain C-, in addition to the presence of isidies (Roux, 2017).

Ramalina leptocarpha Tuck. might be confused with *R. celastri* that differs in having flat laciniae, a convex apothecial disc without pruina, lacking pseudocyphellae on the margins and never producing zeorin. In addition, blades of *R. leptocarpha* produce apothecia on both sides, but they are usually characteristically found on upper side of blades in *R. celastri*. In addition, *R. leptocarpha* looks like *R. calicaris* (L.) Fr., a European species present in Morocco and Tunisia (Seaward & al. 1996; Ajaj & al. 2013) that differs in having more protruded pseudocyphellae.

Conservation and conclusion

Of the lichens identified in this study, 17 taxa (*Amygdalaria continua, Evernia prunastri, Hypogymnia physodes, Phlyctis argena, Physcia adscendens, Physcia biziana, Physcia*

aipolia, Physcia caesia, Physcia dubia, Physcia stellaris, Physcia tenella, Physconia grisea, Physconia perisidiosa, *Physconia distorta* var. *subvenusta*, *Ramalina canariensis, Ramalina farinacea, Ramalina fastigiata*) are officially protected in Algeria (Radp 2012). They represent the 16% of all protected species in the country. This percentage indicates the importance of the lichen flora of the Tessala Mountains region. This flora seems to shelter a very significant and original lichen diversity which deserves to be protected against the different anthropic constraints. Indeed, the natural habitats of this region, especially the original vegetation of green oak covering these mountains remain very weakened and vulnerable because of a very important anthropic activity putting this floristic and ecological wealth in potential danger (Saidi 2017). Only by the presence of this large number of protected lichens by Algerian law, this region must have an official protection status. That is why we propose that the Tessala Mountain forest be granted the status of a natural reserve in order to conserve this biodiversity.

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Xanthium strumarium – a potential cheap resource of plant substances for medicinal use

Abstract

Kozuharova, E., Ionkova, I. & Spadaro, V.: *Xanthium strumarium* – a potential cheap resource of plant substances for medicinal use. — Fl. Medit. 29: 93-102. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Xanthium strumarium L. (*Asteraceae*) is an annual herb which reproduces solely by seed. So far its centre of origin was considered Central or South America. Recent archeological research revealed that the burs of *X. strumarium* were used in Yuergou site (400–200 cal BC) in the Turpan Basin of northwestern China. This plant adventive to Europe reduces germination of various crops and behaves like and aggressive invasive species. *X. strumarium* is the most frequently recorded plant in the field borders between the crop land and adjacent territories the agricultural areas in Bulgaria.

We aim of this study is to reveal the potential of *X. strumarium* as a cheap source of compounds with valuable pharmacological activities. Here we analyse: 1) the traditional ethnobotanical data from its native habitats; 2) the modern investigations of pharmacological activity and essential secondary compounds.

Traditionally the plant is used as febrifuge drug and an immunostimulant, as a diaphoretic agent and against malaria, as well as dysentry cure, astringent, sedative, analgesic, diuretic, against leucorrhoea and urinary diseases, eczema and skin disease, bleeding, insect bite, to treat boils and pimples, against smallpox and stomach diseases, earache and strumous disease, leprosy, headache, fever, etc.

X. strumarium contains sesquiterpene lactones, thiazinediones, phenolic acids etc. and posses anticancer, antitussive, antifungal, antiinflammatory, antinociceptive, hypoglycaemic, antioxidant, antitrypanosomal, and antidepressant-like activity, diuretic effects, insecticidal and herbicidal activities as well as antitrypanosomal effect. A pharmaceutical application of this plant in the future would reduce its populations and thus would contribute to the biodiversity conservation.

Key words: ethnobotany, pharmacological activity, invasive plants management, sustainable development.

Introduction

Xanthium strumarium L. **(**common cocklebur) is a coarse, erect, branching, annual herb which reproduces solely by seed (Holm & al. 1991). The geographic distribution of *X.*

strumarium extends from latitude 53°N to 33°S (Holm & al. 1977). It is most often found in the temperate zone, but also occurs in subtropical and Mediterranean climates. Love and Dansereau (1959) identified the centre of origin of *X. strumarium* in Central or South America. The native North American *Xanthium* taxa originally grew along shores and rivers and the fruits were dispersed by water or occasionally by animals (Love & Dansereau 1959). Their hard brown, ovoid fruits covered with hooked spines and with two terminal beaks, readily stick to clothing and fur, and thus are easily spread (CABI 2018).

Interestingly the statement that *X. strumarium* originates from Central or South American (Love & Dansereau 1959) needs a revision. Recently, the burs of *X. strumarium* were discovered at the Yuergou site (400–200 cal BC) in the Turpan Basin of northwestern China. It is interesting that most of these cockleburs were broken with their seeds missing. They were likely opened by humans with a knife. The cockleburs were likely used as a common medicinal resource (Sheng & al. 2018).

X. strumarium tolerates a wide variety of soil types and textures and a soil pH range of 5.2 to 8.0, as well as frequent flooding and saline conditions (Weaver & Lechowicz 1983). This plant occurs in cultivated fields, along beaches, coastal dunes, watercourses, railway embankments, roadsides, field edges, and waste places but is not common in the mountains. It prefers open communities and will disappear if shaded or crowded (Kaul 1971). *X. strumarium* reduces germination of various crops (Gilani & al. 2010; Shajie & Saffari 2007). Adventive (not native) to all continents (Kuzmanov 2012) it is regarded, invasive in UK (CABI, 2018) and potentially invasive in Bulgaria where was the most frequently recorded plant species during a survey in the field borders between the crop land and adjacent territories in representative agricultural areas (Aneva & al. 2018).

We aim of this mini-review is to reveal the potential of *Xanthium strumarium* as cheap source of compounds with valuable pharmacological activities. Here we present data on the: 1) traditional ethnobotanical data from its native habitats; 2) modern investigations of pharmacological activity and essential secondary compounds.

Material and Methods

In 2018 and in 2019, we accessed the Scolar Google, Web of Science and PubMed to identify publications with the search string: "*Xantium strumarium**", "*traditional**", "*compounds**", "*pharmacological**", "*medicinal**", "*toxic**", "*cancer**", "*inflamatory**", "*in vivo**", "*in vitro**"etc.

Ethnobotanical data on traditional medicinal use

Xantium strumarium, has been used for various medicinal purposes. The traditional medicinal applications are summarized in Table 1. Few of the traditional indications are contradictive (decreases perspiration/diaphoretic properties). Similar applications are reported for distantly situated arrears and cultures.

Table 1. Comparison between traditional medicinal applications and modern pharmacological tests of *Xanthium strumarium*.

Traditional medicinal application	Contemporary pharmacological tests
Dysentry [1, 2]	Significant inhibition of *Shigella flexneri* [22]
Stomach diseases [3], diarrhea [21]	No modern pharmacological tests
Dental sourness and toothaches [4, 5, 16]	Analgesic [26]
Astringent, bleeding [6, 9, 17, 18]	No modern pharmacological tests
Small pox [1, 3, 6]	No modern pharmacological tests
Leprosy [7]	No modern pharmacological tests
Tuberculosis [34]	No modern pharmacological tests
Eczema, skin disease, boils and pimples [8-10, 19, 20]	Antibacterial activity against *Bacillus subtilus* and *Staphylococcus aureus* [22]
Chronic malaria [8, 11]	No modern pharmacological tests
Immunostimulant [12]	Immunostimulant [12]
Decreases perspiration [13]	No modern pharmacological tests
Diaphoretic properties [8, 10]	No modern pharmacological tests
Febrifuge [4, 14]	No modern pharmacological tests
Diuretic properties [1, 6, 8, 9] as well as leucorrhoea and urinary diseases [5, 8, 10, 11, 15]	Diuretic properties [23]
Insect bite [9]	No modern pharmacological tests
Earache and strumous disease [4]	No modern pharmacological tests
Sedative [1, 6]	CNS depressant Antidepressant -like activity
To cure total paralysis [34]	No modern pharmacological tests
Headache, analgesic [5, 7]	Analgesic [26]
No anecdotal reports	Anticancer activity, antimitotic [23, 27-32]
No anecdotal reports	Antitussive [23, 24]
No anecdotal reports	Antifungal [23, 24]
No anecdotal reports	Anti-inflammatory [23, 24, 26]
No anecdotal reports	Hypoglycaemic [23, 24]
No anecdotal reports	Antioxidant [23, 24]
No anecdotal reports	Antitrypanosomal [33]

Legend: [1] Arshad & Khan (2000); [2] Gilani & al. (2014); [3] Ullah & al. (2013); [4] Ali & Qaiser (2009); [5] Gilani, & al. (2014); [6] Hussain, & al. (2008); [7] Akhtar & al. (2013); [8] Bhardwaj & (2011); [9] Yadav & al. (2015); [10] Manandhar (2003); [11] CABI (2018); [12] Alonso-Castro & al. (2016); [13] Hocking (1956); [14] Speck (1941); [15] Bocek (1984); [16] Fowler (1989); [17] Reagan (1929); [18] Swank (1932); [19] Elmore (1944); [20] Hocking (1956); [21] Curtin (1949); [22] Nasir & Khan (2012); [23] Kamboj & Saluja (2010); [24] Kim & al. (2003); [25] Keya & al. (2018); [26] Han & al. (2007); [27] Roussakis & al. (1994); [28] Saxena & Mondal (1994); [29] Kim & al. (2003); [30] Kim & al. (2005); [31] Ramirez-Erosa & al. (2007); [32] Nibret & al. (2001); [33] Talakal & al. (1995); [34] Romero (1954).

North and Central America

According to the traditional, folk and herbal medicine of native tribes of North and Central America *Xantium strumarium* is used for various health conditions. Navaho used the burs to decrease perspiration (Hocking 1956). Houma used it as febrifuge drug - decoction of root taken for high fever (Speck 1941). Costanoan used it as urinary aid - a decoction of seeds was used for bladder ailments (Bocek 1984). Lakota used *X. strumarium* as a medicine in ceremonies, although detailed information was not reported (Rogers 1980). Paiute, Northern applied it as oral aid - burs rubbed on sore gums to take the pain, poison

and blood out (Fowler 1989). Apache, (White Mountain) used roots and leaves as a blood medicine while seeds were ground to make bread (Reagan 1929). Costanoan used to eat seeds in pinole (Bocek 1984). Keres (Western) used a poultice of ground, seed powder on open sores or saddle galls (Swank 1932). Mahuna used *X. strumarium* as an orthopedic aid (for total paralysis) and as a tuberculosis remedy (Romero 1954). Navajo applied *X. strumarium* as a dermatological cure - the plant was used as a liniment for the armpit to remove excessive perspiration (Elmore 1944; Hocking 1956). Pima used the plant against diarrhea as a decoction of burs (Curtin 1949). *X. strumarium* is listed among traditionally used medicinal plants from Mexico, Central America, and the Caribbean with pharmacological evidence of its immunostimulant effects (Alonso-Castro & al. 2016).

India

According to traditional, folk and herbal medicine of the Central Aravalli region of Rajasthan (North West India) *X. strumarium* is credited with powerful diaphoretic properties. The dose of half to one ounce is recommended in chronic malaria, leucorrhoea and urinary diseases. Leaves are used in eczema and leucoderma (Bhardwaj & al. 2011). The use against malaria is particularly emphasised (Bhardwaj & al. 2011; CABI 2018).

According to traditional, folk and herbal medicine of the northern part of Chhattisgarh State of India (Central India) *X. strumarium* is credited with skin disease, bleeding, diuretic, insect bite, urinary problems (Yadav & al. 2015).

Nepal

Fruit paste mixed with other plants is used to treat boils and pimples traditionally Manandhar (2005). In farwest Nepal *X. strumarium* is reported as one of the alien species used in traditional medicines although it is not indicated for the treatment of what health condition (Kunwar & al. 2015).

Pakistan

In Rawal Town, Pakistan is reported as sedative, diuretic, smallpox and dysentry cure Arshad & Khan 2000. In Wana district is reported to have the highest use value of all medicinal plants. Roots, fruit and seeds are demulcent and useful for stomach diseases and smallpox (Ullah & al 2013). It is one of the important medicinal plants of Hattar region in spring season - Whole plant is claimed to have a sedative, astringent, diuretic activity. Root is used in earache, fruit used in smallpox (Hussain & al. 2008). In Chitral valley leaves are chewed for dental sourness. Root is used in earache and strumous disease (Ali & Qaiser 2009). In the region of Swat, North Pakistan - fever, headache, analgesic (Akhtar & al 2013). In Galliat, Ayubia, Kashmir, Bahawalpur and Cholistan (Desert) area - the fruit is used in the cure of dysentery and urinary diseases. It is also useful in toothaches, headaches, and leprosy (Gilani & al. 2014).

China

The fruits (Chinese name 'Cang-Er-Zi') are used as a traditional herbal medicine in China for the treatment of nasal sinusitis, headache, urticaria and arthritis (Pharmacopoeia of P.R. China 2000).

Chemical composition of the plant

Xanthium strumarium contains vast number of metabolites (Fan & al. 2019). They fall into the following groups of compounds: terpenes, flavonoids, phenolic acids (such as: caffeic acid, potassium3-O-caffeoylquinate, 1-O-caffeoylquinic acid, chlorogenic acid, 4-O-caffeoylquinic acid, 1,4-di-O-caffeoylquinic acid, 1,5-di-O-caffeoylquinic acid, 3,5-di-O-caffeoylquinicacid, 4,5-di-O-caffeoylquinic acid, 1,3,5-tri-O-caffeoylquinic acid, 3,4,5-tri-O-caffeoylquinic acid, and cynarin, Minato & al. 1965; Winters & al. 1969; Malik & al. 1992; Marco & al. 1993; Kamboj & Saluja 2010). Cumarins (Fan & al. 2019) and thiazinediones (Ma & al. 1998; Qin & al. 2006 Fan & al. 2019) are reported from *X. strumarium*. The plant also contains sesquiterpene lactones (Malik & al. 1992; Marco & al. 1993; Kim & al. 2003). i.e. xanthinin, xanthatin (deacetylxanthinin), xanthanol, isoxanthanol, xanthinosin, xanthanolides, deacetylxanthumin, etc.

Chemical studies on the composition of the stem oil differed two main groups of compounds: monoterpenes (49.4%) and sesquiterpenes (29.1%). The leaf oil has similar composition: monoterpenes (55.8%) and sesquiterpenes (26.4%). The major components in both oils were identified as: limonene (35.0%), carveol (25.0%), α-ionone (10.5%), β-caryophyllene (6.0%) and p-cymene (5.0%) (Kamboj & Saluja 2010).

Many researchers claim importance to more phytochemical groups for the pharmacological action of the herb, but the findings are inconsistent within the sources (Kamboj & Saluja 2010). Apart from the etheric oil, sesquiterpenes and phenolics form the main pharmacologically active principles of the species. Also the sesquiterpene lactones (Fig. 1) should not be neglected (Roussakis & al. 1994; Saxena & Mondal 1994; Schmidt 1999, Kim & al. 2003; Nibret & al. 2011, Ghantous & al 2013, Amorim & al 2013, Piloto-Ferrer & al. 2019).

Toxicity

The data about toxicity are somewhat contradictive. The plant was traditionally considered toxic. During the famines of the 1950's in China, their seeds were collected and consumed by the farmers of Northeast China. Thirty-five people suffered from alimentary toxicosis, and seven individuals were killed (Shen 2005). It has been reported as fatal to cattle and pigs (Masvingwe & Mavenyengwa 1998). The chloroform and hexane soluble fractions of root extract were tested in Wistar rats and the toxicity determination was done according to the Organization for Economic Cooperation and Development guidelines. The study indicates that acute and subacute administration did not produce any toxicity to rats as evident from weight change, mortality, and limited biochemical and histopathological analysis. Hence, the extracts are considered as unclassified and are found to be safe (Aranjani & al. 2012). However kaurene glycosides that are present in extract obtained with 75% aqueous ethanol by infiltration induce hepatotoxicity in mice by way of its induction of oxidative stress as lipid peroxidation in liver, which merited further studies. Therefore, these toxic constituents explain, at least in part, the hepatotoxicity of *X. strumarium* L. (Wang & al. 2011). The presence of sesquiterpene lactones is also a hazard for toxicity (Kamboj & Saluja 2010). Sesquiterpene lactones are potent pharmacologically active principles (e.g. artemisinin and its derivatives are in use as first-line antimalarials and parthenolide, have recently reached cancer clinical trials). However, the toxicological profile of these compounds must be thoroughly characterized, since the same properties that make these compounds useful medicines can also cause severe toxicity (Schmidt 1999; Ghantous & al 2013; Amorim & al. 2013).

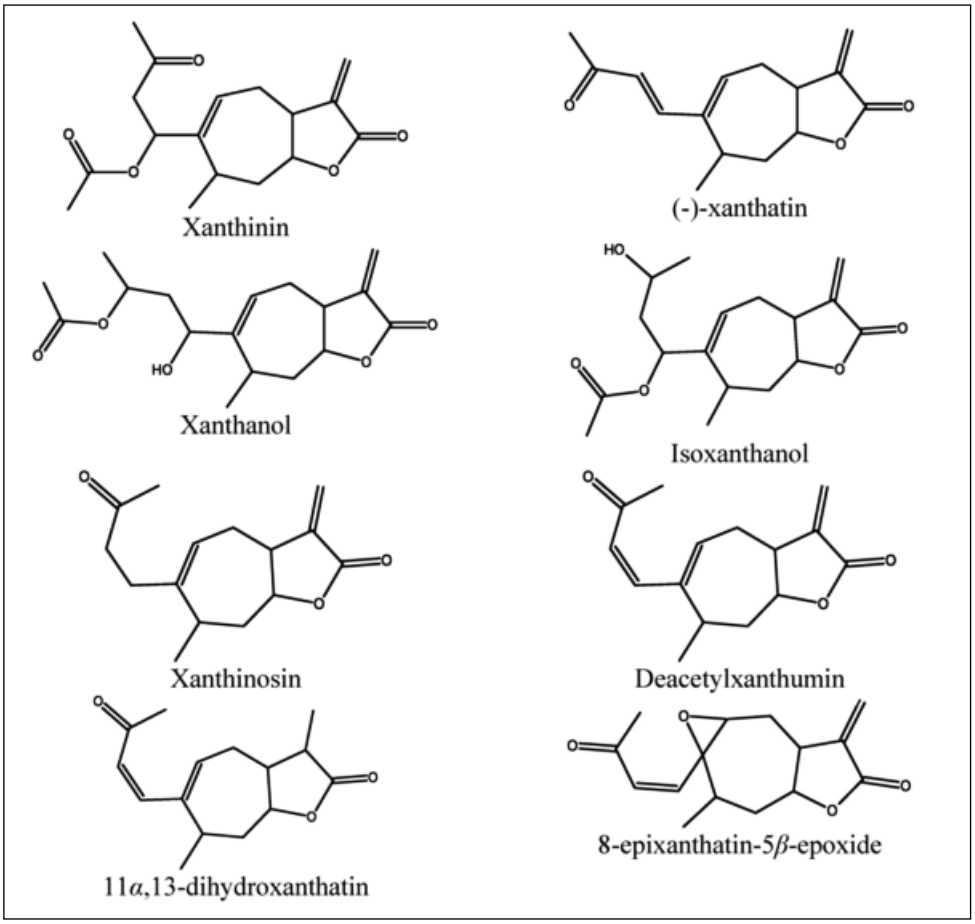

Fig. 1. Sesqiuterpene lactones in *Xanthium strumarium*.

Contemporary pharmacological investigations

Due to the presence of sesquiterpene lactones is related the potential anticancer activity (Roussakis & al. 1994; Saxena & Mondal 1994; Kim & al. 2003; Nibret & al. 2011, Piloto-Ferrer & al. 2019). For instance xanthatin and xanthinosin are two sesquiterpene lactones isolated from the burs which showed moderate to high *in vitro* cytotoxic activity in the human cancer cell lines WiDr ATCC (colon), MDA-MB-231 ATCC (breast), and NCI-417 (lung). Xanthatin and xanthinosin were purified as the result of a multi-screening bioassay-guided study of wild plant species of the family Asteraceae, collected from various sites in Saskatchewan, Canada (Ramirez-Erosa & al. 2007). Also the In vitro results show that X. strumarium, mediated by xanthatins, induces G2/M arrest and impair anaphase entrance. This leads to a significant induction of apoptotic and necrotic in CT26WT cells, demonstrating their significant anti-proliferative activity through interfering with the mitotic apparatus (Piloto-Ferrer & al. 2019).

The western herbalism practice to treat yellow diarrhea, caused by various species of Shigella with *Xanthium strumarium* was tested. Results obtained indicate that chloroform fraction from *X. strumarium* showed significant activity against *Shigella flexneri* (19 mm zone of inhibition). The same fraction revealed good antibacterial activity against *Bacillus subtilus* and *Staphylococcus aureus*, the main causative agents for skin related disorders (Nasir & Khan 2012).

The plant showed antitussive, antifungal, antiinflammatory, antinociceptive, hypoglycaemic, antimitotic, antioxidant, CNS depressant activity, diuretic effects, contact dermatitis, insecticidal and herbicidal activities (Kamboj & Saluja 2010) as well as antitrypanosomal effect (Talakal & al. 1995).

Quite interesting is the antidepressant-like activity of *X. strumarium.* The experimental data clearly demonstrate that the methanolic extract of *X. strumarium* posses antidepressant-like activity in the animal model. Administration of MEXS extract of 50, 100, and 200mg/kg significantly (p< 0.001) decreased the immobility periods of mice when compared to the control group (0.9% normal saline water), indicating significant antidepressant-like activity. The positive control imipramine hydrochloride (30mg/kg) also showed similar effect as MEXS (Keya & al. 2018).

Conclusions

Xanthium strumarium deserves further investigations as it has valuable pharmacological activities. Many of the traditional applications are confirmed with modern pharmacological tests but there are more ethnobotanical data that are still not tested. Also the data collected and analyzed in 2010 concerning the pharmacological activity (Kamboj & Saluja 2010) are extended and enriched later on (Nibret & al. 2011; Nasir & Khan 2012; Keya & al. 2018). As *X. strumarium* posses character of invasive plant species and being most often recorded weed in the agricultural lands (Aneva & al. 2018), it offers lavishly plant substance resources. Additionally a pharmaceutical application of this plant in the future would reduce its populations and thus would contribute to the biodiversity conservation.

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Short note

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L. Sánchez Villegas, E. Sánchez Gullón & A. F. Muñoz Rodríguez

Some interesting species to the flora of Huelva and Sevilla (SW Spain)

Abstract

Sánchez Villegas, L., Sánchez Gullón, E. & Muñoz Rodríguez, A. F.: Some interesting species to the flora of Huelva and Sevilla (SW Spain). — Fl. Medit. 29: 103-108. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Six new records of interesting vascular plants are described for the provinces of Huelva and Sevilla (Spain). For each taxon, details are given of distribution, habitat, ecology, previous records and the extent of naturalization for the alien species. In addition, a new spontaneous hybrid species is described in the *Linaria* Mill. (*Scrophulariaceae*) genus: *Linaria* ×*erebea* Sánchez Gullón & Muñoz Rodr., located in the province of Huelva.

Key words: floristics, Iberian Peninsula, xenophytes.

Introduction

Six new records of interesting vascular plants are described for the provinces of Huelva and Sevilla (Spain). These floristic novelties here presente are mainly the result of fielwork of first autor in Huelva and Sevilla (SW Iberian Peninsula) between 2016 and 2018. Voucher specimens of all taxa preserved in the herbaria of the University of Sevilla (SEV) and the Royal Botanical Garden of Madrid (MA).

Results

Allium pruinatum Link ex Spreng., Syst. Veg. 2: 35 (1825) (*Liliaceae*)
Huelva: Almonte, Estación Biológica Doñana, proximidades Laguna Santa Olalla 29SQA2397. 11/7/2018. *L. Sánchez Villegas* & *E. Sánchez Gullón* (SEV 287455).

Iberian endemism present in central and southern Portugal and western Andalusia (Pastor & Valdés 1983; Aedo 2013). In the village of Almonte, it has been found in sandy soil in pinewoods, outside the Doñana Natural Area (Garrido & al. 2002), making this the first reference for this geophyte within the Doñana Biological Reserve. It is located on the edges of a firebreak close to the Santa Olalla lagoon. This is a taxon classified by the UICN as DD (Magos & Kell 2011) and described as D1 Vulnerable (VU) in the Red List of

Spanish Vascular Flora (Moreno 2008); it is also mentioned in Decreto 23/2012, 14 February, which regulates the conservation and sustainable use of indigenous flora and fauna and their habitats in Andalusia.

Astragalus sesameus L., Sp. Pl. 2 (1753) (*Leguminosae*)
Huelva: Huelva, La Orden, en herbazal sobre suelos básicos con restos conchíferos 29SPB8368. 15/5/2016. *E. Sánchez Gullón* (SEV286624).

Therophyte found in dry pastures, the edges of pathways and crops, and preferably calcicolous (Rivas Goday, 1953). Endemic to the western Mediterranean, it is frequently found in the east of the Iberian Peninsula, and is rarer in western Andalusia where it has been cited in Cádiz, Córdoba and Sevilla (Vicioso 1948; Pujadas 1986; Almeida 2003; Podlech 2000). The record presented here is new for the province of Huelva, where it has never previously been cited. The habitat where it was located has limestone soil of the Huelva Sands formation (Civis & al. 1987), with silt containing numerous conchifera remains, in particular bivalves, gastropods and scaphopods from the Cenozoic era.

Lythrum portula (L.) D. A. Webb, Feddes Repert. 71: 13 (1967) (*Lythraceae*)
Sevilla: Almadén de la Plata, orilla Rivera del Cala, 29SQB5285. 6/7/2018. *L. Sánchez Villegas* & *E. Sánchez Gullón* (SEV 287457).

Aquatic terophyte widely distributed across the Iberian Peninsula, which Velayos (1997) cited in the western Andalusian provinces of Córdoba, Cádiz and Huelva. Subsequently, Morales & al. (1998) located it in the village of Constantina, in the Sierra Norte, in the province of Sevilla. The location is confirmation of its presence in this province, having been discovered in pteridophyte pastures temporarily flooded by the river course.

Solanum sisymbryfolium Lam., Tab. Encycl. 2: 25 (1794) (*Solanaceae*)
Huelva: Rociana del Condado, cuneta camino agrícola junto alcornocal psammófilo, 29SQB1030. 15/9/2018. *E. Sánchez Gullón* (SEV287456).

South American xenophyte, which is known from valleys in Cantabria and the Atlantic valleys of the Basque Country and Pontevedra (Sobrino Vesperinas & Sanz Elorza 2012), has been cited on the coast of Huelva where it appears in industrial zones around the port area (Sánchez Gullón 2000), which matches the findings of Gómez Vigide & al. (2006) who postulated its possible appearance via commercial traffic. The record presented here confirms its possible naturalization in the province of Huelva.

Teucrium pseudocamaepitys L., Sp. Pl. 562 (1753) (*Lamiaceae*)
Huelva: Ayamonte, en protosuelo calizo, 29SPB4122. 13/4/2013. *L. Sánchez Villegas* & *E. Sánchez Gullón* (MA926168).

This taxon is known in the central, southern and eastern Iberian Peninsula, and has been cited in Cádiz, Córdoba and Sevilla, in western Andalusia (Navarro 2012). This is the first

reference to it in the province of Huelva; it appears in limestone soils close to the Parador, and is associated to other calcicolous species of interest for their rarity in Huelva, such as *Picris willkommii* (Sch. Bib.) Nyman*, Centaurea bombycina* Boiss.*,* etc.

Gratiola officinalis L., Sp. Pl. 17 (1753) (*Scrophulariaceae*)
Sevilla: Almadén de la Plata, orillas de la Rivera del Cala 29SQB5285. 6/7/2018. *L. Sánchez Villegas* & *E. Sánchez Gullón* (SEV287458).

This species is dispersed across the centre, west and northeast of the Iberian Peninsula. Romero (2009) cites it in western Andalusia but only in Córdoba, although it had previously been cited in Sevilla, around the village of El Ronquillo (Pina 2000). It has recently been detected at the head of the Cala stream, in the municipality of Almadén de la Plata, which confirms its presence in the province of Sevilla, in the natural region of the Sierra Norte, where it appears associated to *Gratiola linifolia* Vahl, forming mixed hygrophila grasslands.

Linaria ×***erebea*** Sánchez Gullón & Muñoz Rodr. **nototaxon nov.** (Figura 1B). (*Scrophulariaceae*)
[*Linaria incarnata* (Vent.) Spreng. × *L. spartea* (L.) Willd.]

A new natural hybrid of the *Linaria* Mill. genus is described, formed between the parental *Linaria incarnata* (Vent.) Spreng. (Figure 1A) and *L. spartea* (L.) Willd. (Figure 1C), belonging to Subsect. *Versicolores* Benth., both distributed among pastures and scrubland clearings on sandy substrata in the west of the peninsula (Sáez & Bernal 2009). This new hybrid it appears in sandy soils along the coast and in the river Odiel basin at the Andévalo.

Holotypus: Huelva: Marismas del Odiel, en suelo arenoso ácido 29SPB8025. 15/5/2017. *E. Sánchez Gullón* (MA926163) (Fig. 1).

Description: Hybrid plant with intermediate morphological characters between both parents. Stem with unicellular non-glandular and multicellular glandular hairs. Corolla with pale coloration, from yellowish to lilac; with oblique spur. Capsules intermediate in size between its parents.

Etymology: The nothospecific name *Erebea* refers to "*Palus Erebea*" -Infernal Lagoon-, a Greek name used by the Romans when referring to the mouth of the Odiel river.

Habitats: It inhabits grasslands in sandy soils, among its parents.

Distribution area: In the litoral and Andévalo regions, province of Huelva (SW Spain).

Diagnosis: *Linaria incarnata* presents multicellular glanduliferous hairs, and *L. spartea* has non-glanduliferous unicellular hairs. The hybrid presents both types of hairs, and its glanduliferous hairs are longer than those of *L. incarnata*. *L. incarnata* has violet flo-

Fig. 1. A) *Linaria incarnata* (Vent.) Spreng.; B) *Linaria* ×*erebea* Sánchez Gullón & Muñoz Rodr.; C) *Linaria spartea* (L.) Willd.

wers and *L. spartea* has consistent yellow flowers; on the other hand, the hybrid has a corolla of pale coloration, from yellowish to lilac. The *L. spartea* spur is vertical, while that of *L. incarnata* and the hybrid is oblique. *L. spartea* has bigger capsules than *L. incarnata*, and the hybrid plants have capsules that are intermediate in size. The hybrid inhabits grasslands in sandy soils, residing between its parents.

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Article

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S. Bancheva, M. Delcheva & Tz. Kikindonov

In-situ and *ex-situ* conservation of *Spiraea crenata* (*Rosaceae*) in Bulgaria*

Abstract

Bancheva, S., Delcheva, M. & Kikindonov, Tz.: *In-situ* and *ex-situ* conservation of *Spiraea crenata* (*Rosaceae*) in Bulgaria. — Fl. Medit. 29: 109-117. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

This study presents the results of the conducted *in-situ* and *ex-situ* activities targeting the effective protection of the populations of *Spiraea crenata*, one of the rarest plant species in Bulgaria, currently known from two very restricted localities, near Kaspichan town (Northeastern Bulgaria) and near Stefanovo village (Western Bulgaria). A long-term monitoring of the two populations for identification of the main threatening factors, as well as the trends in the population state has been conducted. It is estimated that the potential for conservation *in-situ* actions exists in the locality near Kaspichan (the other locality is very small, situated in private lands and surrounded entirely by cultivated fields). The main activities conducted are the clearing of the habitat of branches of competitive trees and shrubs, as well as support of breeding abilities by *in vitro* propagation and return of acclimated plants back to the locality. Two *ex-situ* live collections of *in vitro* propagated plants were created and stored at the Institute of Biodiversity and Ecosystem research and Shumen Agricultural Institute.

Key words: breeding support, endangered plants, in vitro propagation.

Introduction

Spiraea crenata L. (Scalloped spirea) is one of the rarest species of the Bulgarian flora, known from two small localities: near Kaspichan town, Shoumen district (Northeastern Bulgaria) and near Stefanovo village, Pernik district (Western Bulgaria) (Fig. 1). Its geographic range includes Eastern and Northeastern Europe, the Balkan Peninsula and Northwestern Siberia, as the Bulgarian populations represent the southernmost point of its distribution area.

In Bulgaria this species is protected, included in Annex 3 of the Biological Diversity Law (2002, 2007), the Red List of Bulgarian vascular plants (Gussev 2009), as well as in

*Extended and enriched version of the oral presentation given at the International Symposium "Botany at the intersection of Nature, Culture, Art and Science", Selinunte, 28-30 June 2018.

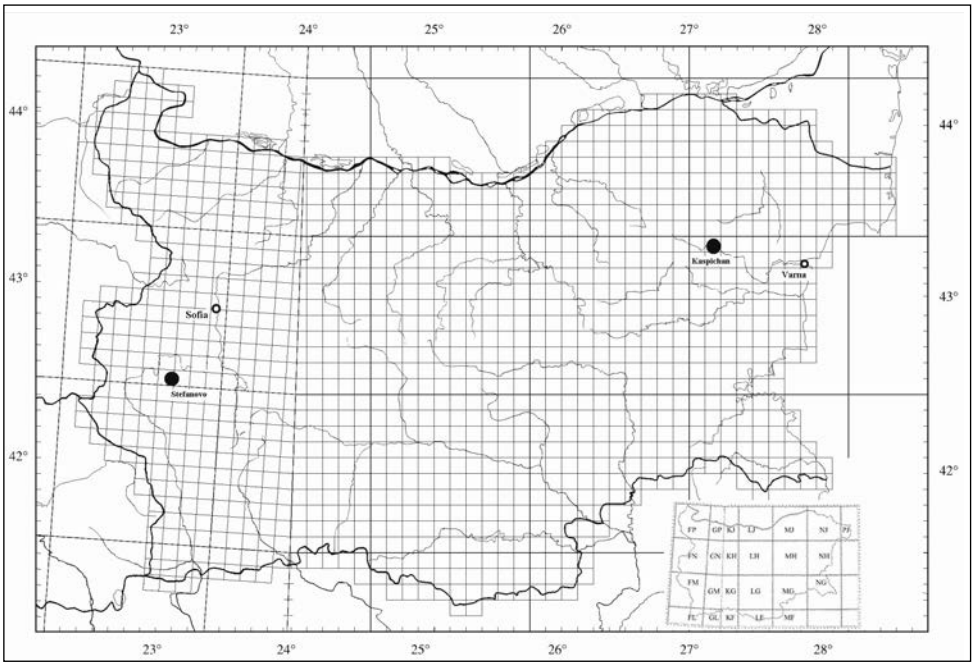

Fig. 1. Distribution map of *Spiraea crenata* L. in Bulgaria.

the Red Data Book of Bulgaria with as "Critically Endangered" (Gussev 2015). For its conservation a Protected Site "Kanyona" with an area of 17.58 ha was declared by the Minister of Environment and Water (State Gazette No 44 of 2012). Scalloped spirea belongs to the category of species for which restoration and maintenance activities are foreseen under the National Biodiversity Conservation Plan 2005-2010 (2010).

The biological characteristics and specific ecological requirements of the species, together with the existing threats to the populations and their habitats, determine the need for prompt protection measures.

This survey was conducted between 2010 and 2018 and aims to provide effective protection for populations of *S. crenata* by applying appropriate *in-situ* and *ex-situ* activities. The following tasks are included: 1. Study of the status of the species populations in Bulgaria and identification of the threatening factors; 2. Development of *in vitro* multiplication technique; 3. Implementation of *in-situ* activities for strengthening of the population of the species in the locality of Kaspichan; 4. Carry out long-term monitoring to track the effect of the activities carried out.

Material and methods

The present study focuses on the two populations of the critically endangered species *S. crenata* in Bulgaria and was implemented in the period 2010 – 2018.

Experimental work for obtaining authentic material for *in vitro* reproduction of *S. crenata* L. has been conducted at the Tissue Culture Laboratory of the Shumen Agricultural Institute. Stem cuttings of *S. crenata* were collected in 2010 from the locality near Kaspichan town. Apical meristems of plants are sterilized by plunging into a solution of 0.04% $HgCl_2$ and 0.1% Tween for 120 minutes and, subsequently by washing them 3 times with sterile distillated water (each of the tips for 30 minutes). In a sterile box under binocular are separated the inner parts of the apical meristems and put into a media for induction in thermostat for 15 days at 32-34˚C. The meristems were sub-cultivated six times on different induction media. The most intensive development of clonal regenerates from explants and whole plants are registered on medium, following Nanova (2007), which is a modification of the LePoivre (LP) nutrient medium (Quorin & al. 1977). In the phase of *in vitro* rooting a rooting media supplemented with macro salts (NH_4NO_3, KNO_3, $MgSO_4$, etc.), microelements ($ZnSO_4$, H_3BO_3, $MnSO_4$, $CuSO_4$, etc.), vitamins (C, B8), etc, was used (Slavova 1998). The *in vitro* rooted plants are taken out to controlled conditions room with temperatures of 26-28˚C and light regime with 16 hours daylight and 8 hours darkness.

The monitoring and assessment of the population state of the species were implemented in accordance with the Methodology for Monitoring of wascular plants and the Methodology for assessment of the state of vascular plants of the National system for Monitoring of the Biodiversity of Bulgaria (Executive Environment Agency-EEA) (http://eea.government.bg/en).

Results and discussion

Morphological description

S. crenata is a deciduous bush, up to 1.40 m high. Branches somewhat angulate, greyish, woody. Leaves of non-flowering shoots 2.5–3.5 cm long, about 2 cm wide, lanceolate to lanceolate-obovate, entire or crenate-serrate towards the apex, with 3 conspicuous veins running on both sides of the mid-vein from leaf base to leaf apex. Leaves of the flowering shoots smaller, oblong-obovate, and entire. Inflorescence umbellate, 10–20-flowered. Flowers small, 6–7 mm in diameter, white to yellowish-white. Follicles small, dehiscent along the ventral suture when ripe; seeds usually two in each follicle (Markova 1973). Flowering V, fruiting VI–VII. Reproduces by seeds and suckering.

The species inhabits gravelly and stony, dry, calcareous places. It is a element of the Pontic-Sarmatian shrub steppe communities, which refer to the priority habitat 40C0 Ponto-Sarmatian deciduous shrubs according to Annex I of the Directive 92/43/ EEC.

Study of the state of the populations of S. crenata *in Bulgaria*

The first population is located 2 km west of Kaspichan station, just near the railroad Kaspichan-Rousse, on the right bank of the river Kamenica (area “Canyon”) of 126 m. above sea level (Fig. 2). The population occupies an area of about 2.5 acres (Bancheva & Delcheva 2014). The species participates in the composition of mixed shrub communities dominated by Oriental Hornbeam. Projective cover of tree species is 10% with the participation of *Acer monspessulanum* L., *Carpinus orientalis* Mill., *Ulmus minor* Mill.

Fig. 2. The locality of *Spiraea crenata* near Kaspichan town, Shoumen district (Northeastern Bulgaria).

Projective cover of the bushes is 30% including *Corylus avellana* L., *Ligustrum vulgare* L., *Spiraea crenata*, etc. Projective cover of herbaceous species is 40%. As a result of the present study, the following threatening factors of anthropogenic origin have been identified: damage and/ or destruction of *S. crenata* in the cleaning of the railway's easement (high degree) and soil contamination with herbicides and fertilizers (medium degree). From the non-manageable factors key role in limited distribution of the species play a low regeneration ability of the species.

The second population is situated on the southern slopes of Golo Bardo Mountain, near the Stefanovo village, Pernik district, among cultivated fields, at 775-850 m above sea level (Fig. 3). It consists of 3 subpopulations of about 1 km distance from each other, as real the occupied area is 0.211 ha (Bancheva &Delcheva 2014). Projective cover of the bushes is 50% including *Cornus sanguinea* L.*, Crataegus monogyna* Jacq.*, Prunus spinosa* L.*, Rosa* sp.*, Spiraea crenata, Ulmus minor.* Projective cover of herbaceous species is 48% among which there are also agricultural crops. The main threatening factor in this locality is the destruction of the population of Scalloped spirea in the plowing of the surrounding fields and the passing of the dirt roads.

The study of both Bulgarian populations of *S. crenata* showed that conservation measures are urgently needed to strengthen the state of the species at national level. It is estimated that the potential for *in-situ* activities exists in the locality near Kaspichan (the other

Fig. 3. The locality of *Spiraea crenata* near Stefanovo village, Pernik district (Western Bulgaria).

locality is very small, situated in private lands and surrounded entirely by cultivated fields). The main activities that would help to strengthen the population are the clearing of the habitat of branches of competitive trees and shrubs, as well as support of breeding abilities by *in vitro* propagation and return of acclimated plants back to the locality.

Development of a technique for in vitro *propagation*

In vitro culture introduction

It was registered a weak development of the meristems, and that's why they were subcultivated again and again on different induction media. The best development was registered for the medium described by Nanova (2007).

In vitro cloning of the developed plants

For a 3 months period the developed plants were sub-cultivated repeatedly on different micro-propagation media. The most intensive development of clonal regenerates from explants and whole plants are registered on the medium, described by Nanova (2007). It should be underlined the comparatively low regeneration potential of the studied materials on the tested media. Nevertheless, enough plants for the subsequent phases of stabilization of the regenerated plants were obtained.

In vitro rooting

The most difficult appeared to be the choice of a media for *in vitro* rooting of the developed plants. After repeated sub-cultivations a rooting media supplemented with macro salts (NH_4NO_3, KNO_3, $MgSO_4$, etc.), microelements ($ZnSO_4$, H_3BO_3, $MnSO_4$, $CuSO_4$, etc.), vitamins (C, B8), etc., was chosen (Slavova 1998). The induced roots were simple and weak, which makes difficult their further development (Fig. 4).

Adaptation and acclimatization

The *in vitro* rooted plants are taken out to controlled conditions room with temperatures of 26-28˚C and light regime with 16 hours daylight and 8 hours darkness. After the careful separation from the nutrition media the plants are planted in PVC pots with 20 cm diameter in a soil substrate of peat, sand and perlite in 2:1:1 ratio. A humid camera is created via covering with a nylon bag for 1-2 weeks, which is regularly removed, so that there is no risk for over moistening and moulding of the substrate. A month later the plants form new roots, and then they are taken out consequently in a greenhouse for 2 months, and then in external conditions. The collection of *in vitro* plants number 50 individuals.

In-situ *activities for reinforcement of the population of Kaspichan locality*

In order to help strengthen the Kaspichan population, 15 of the *in vitro* propagated plants were planted in the spring of 2012 (Fig. 5). During the monitoring carried out in the

Fig. 4. *In-vitro* plantlets of *Spiraea. crenata.*

Fig. 5. Planting the *in-vitro* plants in the Kaspichan population.

autumn of the same year, 6 plants survived, while one year later, in the autumn of 2013, the number of the survived ones decreased to 3. Their number remained the same in the subsequent monitoring carried out in 2014, 2016 and 2018.

Another *in-situ* activity with a favourable effect on the state of the population of *S. crenata* was the clearing of branches of competitive trees and shrubs organized by the employees of the Bulgarian Railways serving the railway line passing through the locality of the species.

Ex-situ *activities*

For *ex-situ* conservation of the species, a live collection of 15 *in vitro* propagated individuals was created in the Greenhouse of Institute of Biodiversity and Ecosystem Research. Such a collection is also grown in the Shumen Agricultural Institute. If necessary, plants from these collections can be used to strengthen the existing populations of *S. srenata*, as well as to reintroduce the species in the locality of Kabiushka mogila (Shumen district) from where the species is considered extinct in the last decades.

Long-term monitoring of the populations' state and the effect of the activities carried out in

The monitoring of the species is based on long-term observation of selected parameters for the population status, the threats and the consequences thereof, and aims at identifying the trends in the development of the populations. Generally, carrying out systematic monitoring leads to the timely identification of negative population trends and serves as the

basis for adequate management measures. The monitoring of both species populations takes into account all parameters provided in the EEA Monitoring Methodology.

The reporting entity is a "group of bushes", because the biology of the species is such that visually cannot differentiate separate individuals.

Kaspichan's population

In 2010, 48 "groups of bushes" of the observed species were found, mostly concentrated along the edge of the canyon of the river Kamenica. The main threatening factors are damaging and/ or destroying of *S. crenata* in the cleaning of the railway's easement and soil contamination with herbicides and fertilizers. The most important factor, however, is the low regeneration ability of the species.

In the spring of 2012, two *in-situ* activities aimed at improving the state of the population were carried out: clearing the habitat from branches of trees and bushes of competing species and planting 15 *in vitro* propagated individuals (marked with permanent markers buried in the soil). In the autumn of 2013, the number of "groups of bushes" was 79, as 3 of them are the surviving *in vitro* plants. In the following observations in 2014, 2016 and 2018 the number is kept the same, which leads to the conclusion that the activities are in the right direction and the state of the population near Kaspichan can be defined as "Favorable".

Stefanovo's population

It is fragmented into 3 subpopulations about 1 km apart, surrounded by cultivated fields. The plants in each fragment are so densely positioned that individuals or "groups of bushes" can not be distinguished. The monitoring takes into account the area occupied by these fragments. In 2013 the total occupied area is 0.211 ha. In the next monitoring survey carried out in 2014, this area remains unchanged. The main threatening factor in this locality is destroying the population in plowing the surrounding fields and pave the roads. The state of the population near the village of Stefanovo is defined as "Unfavorable-bad".

Conclusions

As a result of the present study and the *in-situ* activities, the state of the Kaspichan population of the endangered species *S. crenata*, was improved. An effective method for its *in vitro* propagation has been developed. The state of the Kaspichan's population is "Favorable", while the Stefanovo's population is in "Unfavorable" state. Two *ex-situ* live collections of *in vitro* propagated plants were created and stored at the Institute of Biodiversity and Ecosystem research and Agricultural Institute in Shumen.

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Article

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Why did *Acanthus mollis*, native to West Mediterranean, become a so relevant artistic and symbolic element arising from ancient Greece?*

Abstract

Minissale, P., Magro, V. & Raimondo, F. M.: Why did *Acanthus mollis*, native to West Mediterranean, become a so relevant artistic and symbolic element arising from ancient Greece?. — Fl. Medit. 29: 119-128. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

In classical antiquity many plant species were a source of inspiration in art and architecture. An emblematic case is *Acanthus mollis*, a Western Mediterranean species, although many Mediterranean countries Floras are in contradiction with respect to its native distribution. Two subspecies are known: *A. mollis* subsp. *mollis* distributed in Italy, France and Croatia, and *A. mollis* subsp. *platyphyllus* growing in Morocco, Algeria and Tunisia. In other Mediterranean countries it should be an introduced taxon, such as in Greece and Turkey where the native species is *A. spinosus*. Although the maximum spread of the *Acanthus* leaf in architecture occurred in Roman times, the Corinthian capital was born in Greece, portraying *A. mollis*. Among the earliest examples we remember the Doric Temple of Apollo Epicurius at Bassae in Peloponnese, built in 450-425 BC. probably by Ictino, the Tholos at Epidaurus (360-330 BC.), characterized by Doric columns in the exterior, while the inner colonnade consists of 14 Corinthian columns. The leaf carved in the stone is unequivocally *A. mollis*. The invention of the Corinthian capital is attributed, without certain proof, both to Callimaco and Ictino, which operated in Athens and in the Peloponnese. As this species was not present as native in Greece at that time, it was seen and designed taking inspiration from some place in Sicily or Magna Greece or from cultivated plants originating in those areas. The first examples of this capital fall into areas of Doric ethnicity. A city founded by this ethnic group that had intense cultural exchanges with the mother country was Syracuse, where *A. mollis* is widespread. This city seems to have played an important role in the the genesis of this architectural element, which is an evidence of cultural influences implemented in the motherland starting from the colonies become autonomous.

Key words: botany and art, cultural heritage, Mediterranean plants.

Introduction

In classical antiquity many plant species were a source of inspiration in art and architecture (Caneva & Kumbaric 2010). An emblematic case is *Acanthus mollis* L. and to a

*Resulting from the merging of two posters by Minissale and Magro & Raimondo presented at thethe International Symposium "Botany at the intersection of Nature, Culture, Art and Science", Selinunte, 28-30 June 2018.

lesser extent *A. spinosus* L., both Mediterranean species used in antiquity by Greeks and Romans and later by other Mediterranean peoples above all as architectural frieze carved on capitals. *A. mollis* also has a remarkable ornamental value and therefore since ancient times it has probably been the object of cultivation and diffusion in other Mediterranean regions, today in the world (GISD 2018).

The aim of the present research is to understand with a biogeographical approach, combined with historical data, which could be the motivations that led to stylize the acanthus shape for the first time in areas where *A. mollis,* the most represented species of the genus, was not present in the native flora.

For this purpose, it is necessary to reconstruct the native distribution of the Mediterranean species of the genus *Acanthus* L. This genus is composed of 20 species distributed in Africa, southern Europe, southern Asia, and Australasia (McDade & al. 2005; Iamonico & Peruzzi 2018); in the Mediterranean area there are *A. mollis* subsp. *mollis* (Fig. 1), *A. mollis* subsp. *platyphyllus* Murb., *A. spinosus* (Fig. 2) and *A. balcanicus* Heywood & I. B. K. Richardson (Fig. 3).

Materials and methods

The research on the distribution of *Acanthus* species is based on literature material such as floras and checklists of all European and Mediterranean countries, herbarium consultations and personal distribution data. The distributive data are compared with the ancient historical sources related to key moments of displacements of peoples, in particular the Greeks and the Romans.

Historical framework

Although the maximum spread of the *Acanthus* leaf in architecture occurred in Roman times, the Corinthian capital seems to be born in Greece (Billot 1993), but through a long process concerning its stylization on the carved stone. Among the earliest examples many authors (Billot 1993, Kelly 1995, Loth 2014, Thodis 2018) remember the Doric Temple of Apollo Epicurius at Bassae in Peloponnese, built in 450-425 BC. probably by Ictino. Gros (1993) is very critical about this interpretation. As this author reminds us, the architectural formalization of the Corinthian capital takes place through the work of *Vitruvius De Architectura*. This author, lived in the second half of the first century BC, indicated in Callimachus (sculptor and architect operating during Vth century BC) the creator of the aforementioned capital; but probably his affirmation, inserted in a legendary context, only wants to ennoble its origins Inspiration would be drawn by a basket covered by a roof tile and layng on a Corintihian girl's grave by her nurse. Being placed on an acanthus root it got leaves wrapping the basket (Locatelli 2016). Anyway, Gros (1993) believes that the first, supposedly, Corinthian capital of Bassae has nothing to do with those of later centuries as the acanthus leaf is very different from the typical one and barely recognizable as such (Fig. 4). It should also be kept in mind that of this single capital, which had a particular meaning with regard to its location inside the temple (Thodis 2018), there remain only

Fig.1. *Acanthus mollis* in flower to Mount Erice (N.W. Sicily). (Photo F.M. Raimondo, June 2018).

Fig. 2. *A. spinosus* in flower, from his natural habitat in Albania (Photo F.M. Raimondo, June 2016).

Fig. 3. *Acanthus balcanicus*: particular of leaves (from Wikimedia Commons, the free media repository).

fragments, now in National Archaeological Museum of Athens, and a drawing made at the time of the discovery in 1811 (Loth 2014). This first appearance at Bassae of the acanthus leaf as decoration not only occurs on a capital but also on other architectural elements of the temple, such as simas and acroteria. Similar ornaments occur also in the temple of Poseidon at Sounion built about 440 BC. Billot (1993), recognizes at Sounion the form of *A. spinosus* but she admits that the shape is very stylized so it could also be other real or imaginary plant. The decoration with acanthus on the acroteria appears to the Parthenon of Athens and here the leaves are much more similar to those of *A. mollis* (Fig. 5a). Similar friezes are reported to the Heraion of Argos (Fig. 5b) and in Magna Graecia to Caulonia and Crotone temple of Hera Lacinia, at the Tholos of Epidaurus and at the Tholos of Delphi (Billot 1993), here with very realistic *A. mollis* friezes (Fig. 5c). The Tholos of Epidaurus, monument built in 360-330 BC., is also characterized by Doric columns in the exterior, while the inner colonnade consists of 14 Corinthian columns. The leaf carved in the stone is unequivocally *A. mollis* as you can see in the Archaeological Museum of Epidaurus where is showed a Corinthian capital unearthed below the foundations of the Tholos temple (Fig. 6).

The maximum development of the use of acanthus will take place in Roman times but always in connection with the Greek civilization; it must be remembered that since the end of the third century BC the Roman civilization undergoes a profound Hellenization especially with the fall in the Roman hands of the great cities of Magna Graecia and Sicily such as Taranto and Syracuse.

The decorative aspects of the Greek world were much appreciated and contacts with Athens were established. Artistic studios of this city were also installed in Rome (Sauron 1993) starting from 150 BC. In the same period and following, in the age of Augustus, the acanthus leaf will be a fundamental decorative element both for the Corinthian capital that will be formalized by Vitruvius and in other decorations as the extraordinary example of the Ara Pacis (13-9 BC) in Rome (Caneva 2010; Rossi 2016; Sauron 2018), where acanthus supports the architrave and it, in all its variations, will always be recognizable as *Acanthus mollis* species. Roman Art used acanthus not only in the decoration of the Corinthian capital but in the creation of friezes and pilaster strips, too. Because of its great decorative effect it was preferred in the decoration of the great columned streets ("cardus maximus*"* in Palmira, Syria, III century BC, "cardus maximus" in Gerasa, Jordan, III century BC) (Rossi 2016).

Results and Discussion

Defining the native distribution of *Acanthus mollis* is problematic as, probably, since ancient times it has been diffused from the original sites in other localities of the Mediterranean basin. The indications of the Mediterranean countries Floras and the databases are sometimes inaccurate and in contradiction with respect to its native distribution. Two subspecies are known: *A. mollis* L. subsp. *mollis* distributed in Italy (Pignatti 1982, 2018), but in the northernmost regions and Sardinia it is considered an alien species (Ballelli & Pedrotti 2009; Puddu & al. 2016); south France and Corse, and *A. mollis* subsp. *platyphyllus* Murb. growing in Morocco (Fennane & Ibn Tattou 2005), Algeria (Quézel &

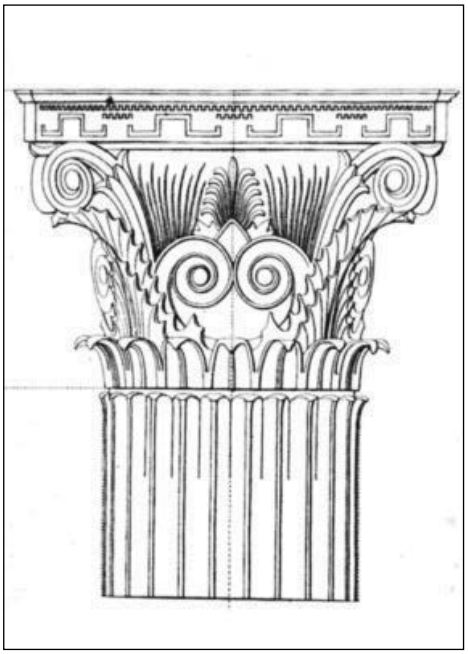

Fig. 4. Drawing of the Corinthian Capital of the Temple of Apollo Epicurius made by J.M. von Mauch for the German edition of the original work by Charles Normand (1819). The drawing is based on field notes and sketches of fragments by the archaeologist Haller von Hallerstein, highlighted during his excavation campaign carried out in Greece, at the site of Bassae (Peloponnese), between 1811 and 1812.

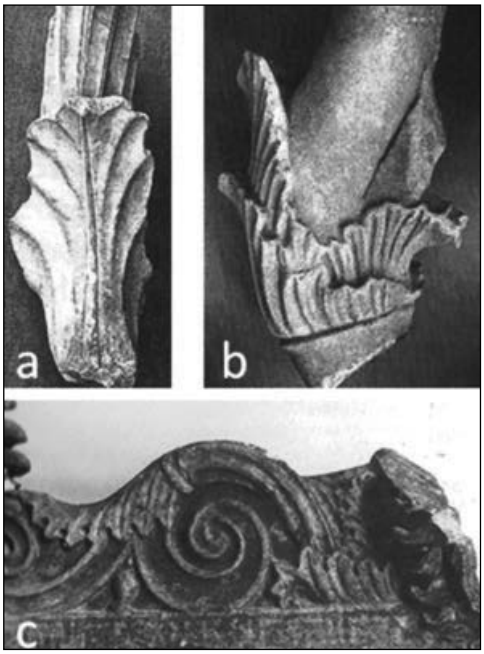

Fig. 5. *Acanthus* leaves in some ancient Greek monuments (from Billot 1993).
a. Athens; Parthenon , basal fragment of the central acroteria (Acropolis Museum – Athens).
b. Argos Heraion, classic temple, basal fragment of the central acroteria (National Museum – Athens).
c. Delphi, Tholos little sima (Archaeological Museum – Delphi.

Santa 1963), Tunisia (Pottier-Alapetite1981), Lybia (El Gadhi 1983) and south Spain (Valdés Castrillón & al. 1987). In other Mediterranean countries it should be an introduced taxon (Malcuit 1939; Barcelò 1980; Della & Iatrou 1995; Castroviejo 2001), such as in Greece (Arianoutsou & al. 2010; Dimopoulos & al. 2013, 2016) and Turkey (Davis 1984) where the native species is *A. spinosus* L. Also for these species is difficult to know the original distribution. *A. spinosus* is known in the Mediterranean and in south-eastern Europe (Italy, Croatia, Albania, Greece, Bulgaria, Crete, East Aegean islands), the Asiatic Turkey as well as in Algeria, but it is native probably only in East Mediterranean.

As this species was not present as native in Greece at that time, it was seen and designed taking inspiration from some place in Sicily or *Magna Graecia* or from cultivated plants originating in those areas. The first examples of this capital fall into areas of Doric ethnicity. A city founded by this ethnic group that had intense cultural exchanges with the mother country was Syracuse, where *A. mollis* is widespread (Minissale & al. 2015). From this city, probably, the acanthus was brought to Dalmatia. Today it is in fact present almost

Fig. 6. Corinthian capital from the tholos of Epidaurus (Argolis, Greece) clearly inspired by *Acanthus mollis* [380-375 BC. Archaeological site of Epidaurus] (from Wikimedia Commons, the free media repository).

exclusively in the Adriatic islands (Nikolić 2015) which were Syracusan colonies or in very close areas. In particular, they were Issa (today Vis Island) and the subcolonies of Lumbarda near Korkyra Melaina (Korčula Island), Tragyrion (Trogir), Epetion (Stobreč) founded from third to second century BC (Lombardo 1993). It also occurs in the nearby Pharos (Hvar Island) founded in 386 BC by Greek settlers from Island of Paros, but allied to Issa. It is surprising the overlap of the presence of the acanthus which in Dalmatia is rare, with the islands affected by the Greek colonization coming from Syracuse (Fig. 7). This circumstance proves, or at least the clue is highly realistic, that acanthus was a relevant species, perhaps also ornamental. It should be noted that Lissa is also the only Dalmatian locality of another species present in Greece and Syracuse surroundings, *Salvia fruticosa* Mill. (Radosavljević & al. 2015, 2019), also this probably was transported by the Greeks of Syracuse.

There are no clues so clear for other places, for example of Greece, but it is possible that from Syracuse or from other Greek colonies the acanthus has been made known in the motherland and, representing a much more appreciable species of the native *A. spinosus*, has become a source of artistic inspiration.

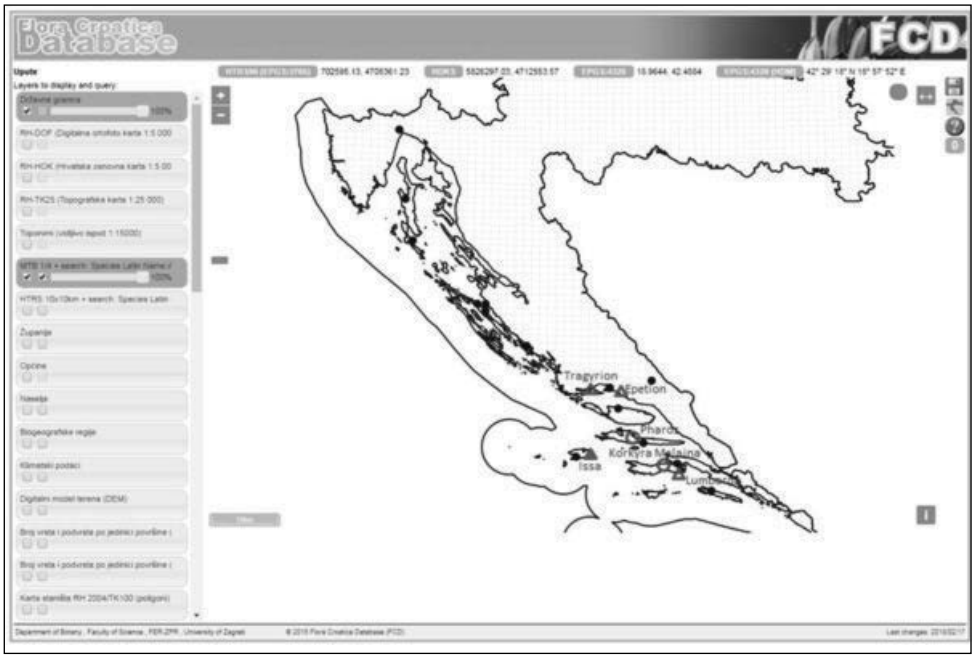

Fig. 7. *Acanthus mollis* distribution in Croatia (black dots) from Flora Croatica Database (http://hirc.botanic.hr/fcd) and the colonies founded by Syracuse. Issa (today Vis, Lissa in Italian) was the first in 390 B.C., the others are sub-colonies founded by Issa (Tragyrion, Epetion, Lumbarda) or other Greek colonies interacting with Issa (Pharos, Korkyra Melaina).

Fig. 8. Corinthian capital – probably inspired by *Acanthus spinosus* or by *A. dalmaticus* – from Ephesus (Smyrne, Turkey) [I-II Century a.C.] (from Wikimedia Commons, the free media repository).

But why was the acanthus in particular chosen? Was it just for the peculiar leaf morphology or for the plant symbolism in classical antiquity? In this regard, the acanthus symbolizes both resurrection and regeneration; in addition, as a perennial herbaceous plant – cyclically subject to dry in the summer season and come back to vegetate since the next winter starting a new cycle – it is ideally suited for being interpreted in art and architecture. Therefore, the biological form (hemicriptophyte) of the *Acanthus* species is the basis of its symbolism and its stylistic success.

But the frenetic activity of the Greek navigators remains fundamental and the consequent contaminations and cultural exchanges between colonies and motherland, contaminations that will then be absorbed and greatly developed later by the Roman civilization.

Conclusion

Contrary to common interpretations on the possibility that the Corinthian capital may be inspired by several Mediterranean species of *Acanthus,* what is just discussed opens the way in excluding or minimizing this possibility. The historical, biological and phytogeographic considerations reported, the symbolism of the plant and in particular the cultural exchanges between motherland and Magna Graecia, on the one hand, constitute the cause and effect of the anthropogenic spread of *A. mollis* in the rest of the Mediterranean area of Greek and Roman influence, on the other, of its own decorative and artistic value. All these elements allow us to affirm that the Corinthian capital, originally inspired by *A. mollis* (Fig. 1) – an easily propagated and cultivated plant and among the most decorative species – may have been inspired, subsequently, also by *A. spinosus* (Fig. 2) or ever more by *A. balcanicus* (Fig. 3); the latter – native to the Balkan peninsula, up to Dalmatia – is now also cultivated in many European and American gardens. In this different view, the cases of the capitals of the Greek Temple of Apollo Epicurius (Fig. 5) and of the columned street ("cardus maximus") of Ephesus (Fig. 8) appear very expressive and would support the thesis of the congeneric multi-specificity, of the inspirational model of the Corinthian capital.

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Article

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Belkacem Gordo & Seghir Hadjadj-Aoul

L'endémisme floristique algéro-marocain dans les monts des Ksour (Naâma, Algérie)

Abstract

Gordo, B. & Hadjadj-Aoul, S.: L'endémisme floristique algéro-marocain dans les monts des Ksour (Naâma, Algérie). — Fl. Medit. 29: 129-142. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

This study aims to focus on Algerian-Moroccan floristic endemism in the Ksour Mountains. It is based on our field surveys, which led us to identify thirty-five (35) endemic taxa. Additional information has been added to describe the habitats that support them, while updating their national distribution. The study identified six (6) exclusive taxa from the Ksour Mountains, and three that have never been reported.
For the conservation of six exclusive taxa of the Ksour Mountains, considered vulnerable, we believe it is necessary to preserve the sandstone escarpments that constitute their habitat and to include these taxa in the national list of uncultivated and protected plants.

Key words: flora, endemism, conservation, sandstone escarpments, Ksour Mountains, Aïn Sefra, Algeria.

Introduction

Considéré comme l'un des 34 points chauds de la planète (Myers & al. 2000), le bassin méditerranéen abrite 10 % des espèces végétales sur environ 1.6 % de la surface terrestre, et à un taux d'endémisme représentant 50% de la richesse floristique globale (Médail & Quézel 1997). Cette dernière a été estimée entre 25000 espèces (Quézel 1985) et 30000 espèces et sous-espèces (Greuter 1991). Sur le pourtour méditerranéen 10 points chauds ont été identifiés (Médail & Quézel 1997). D'autres ont été suggérés comme la région de Kabylie-Numidie-Kroumirie (Véla & Benhouhou 2007) et le littoral et les archipels Dalmates (Nikolić & al. 2008). En plus de ces 10 points chauds, 55 refuges putatifs ont été définis dont la moitié est associée à ces *hot spots* (Médail & Diadema 2009). Cette richesse floristique actuelle de la région méditerranéenne et ce haut niveau d'endémisme prouvent l'individualisation 'in situ' de très nombreux taxons (Quézel 1983 ; Domina & El Mokni 2019). Néanmoins, les menaces auxquelles sont confrontés les biomes méditerranéens sont énormes et peuvent mettre cette biodiversité en péril.

A l'instar des pays du Maghreb, l'Algérie est marquée par une biodiversité végétale remarquable, résultat de son orographie, de sa grande surface et de la multiplicité des paysages. Toutefois, l'information sur cette diversité végétale souffre d'un manque d'actualisation, puisque depuis la Flore de Quézel & Santa (1962-1963) peu d'initiatives ont été entreprises. Cela s'est répercuté défavorablement sur les tentatives de conservation de la biodiversité. A titre d'exemple, le nombre d'espèces endémiques et sub-endémiques demeure flou (incertain), variant entre 250 environ (Quézel 1964), 320 (Greuter 1991) et 464 (Véla & Benhouhou 2007). En Algérie septentrionale, 22 zones importantes pour les plantes ont été initialement identifiées. D'autres sites sont susceptibles d'y être inclus, si des études appropriés sont entreprises (Yahi & al. 2012), tels que Djebel Aïssa (Bouallala 2006). Le projet (CEPF 2017) avait qualifié les monts des Ksour de zones clés pour la biodiversité. Néanmoins, ces milieux arides où s'exacerbent l'ensemble des contraintes méditerranéennes par le déficit hydrique et la pression anthropique (Aidoud 1997) à l'acception des hautes altitudes où règne un bioclimat semi-aride (Djebaili 1984), sont aussi soumises au phénomène de désertisation, qui a pris une dimension inquiétante (Médail & Quézel 2003). Cela se traduit d'une part par une remontée vers le nord des espèces sahariennes (Quézel 1999) et d'autre part par une extension de paysages désertiques (Le Houérou 1995). Ces perturbations sont dues à la combinaison des effets de pâturage avec des pratiques sociales, telles que : la mise en culture, l'éradication des ligneux (Le Houérou 1993), l'augmentation de la population et du nombre de têtes de bétail (Médail & Quézel 1999). Quézel (2000) ajoute également les modifications climatiques actuelles.

Afin de sauvegarder la biodiversité végétale de cette région menacée, il est impératif d'établir un point de référence, qui consisterait à décrire la biodiversité (Primach & al. 2008), en mettant à jour la nomenclature taxonomique de la flore l'Algérie (Amirouche & Misset 2009). Cela peut se faire par la collaboration d'érudits et amateurs pour promouvoir le travail de terrain et aider à résoudre les problèmes taxonomiques (Domina & al. 2015). Ce n'est qu'à partir de ce moment là qu'il sera possible d'actualiser les statuts de plantes ; notamment, celles menacées et endémiques restreintes qui occupent une place de première importance par les programmes de conservation des ressources phyto-génétiques (Neffati & al. 2001), puisqu'elles se différencient de leurs congénères répandus par leur faible investissement dans le transfert de pollen et la production de graines (Lavergne 2004).

C'est dans ce contexte que s'inscrit le présent article dont l'objet est de rechercher les plantes endémiques algéro-marocaines des monts des Ksour en actualisant leur statut taxonomique, et leur distribution nationale et en décrivant les différents biotopes, dans lesquels nous les avons observés.

Matériel et Méthodes

Biogéographie régionale

Les monts des Ksour correspondent à l'Atlas saharien occidental telle une frontière naturelle entre le monde méditerranéen et le Sahara (Fig.1). Au nord, ces massifs sont délimités par les hautes plaines steppiques oranaises. Au sud, ils sont séparés du Sahara proprement dit par les bassins d'avant fosse nord saharienne (bassin de Béchar) ou sillon présaharien suivant la ligne approximative allant de l'oasis de Fendi à Labiadh Sidi Cheikh.

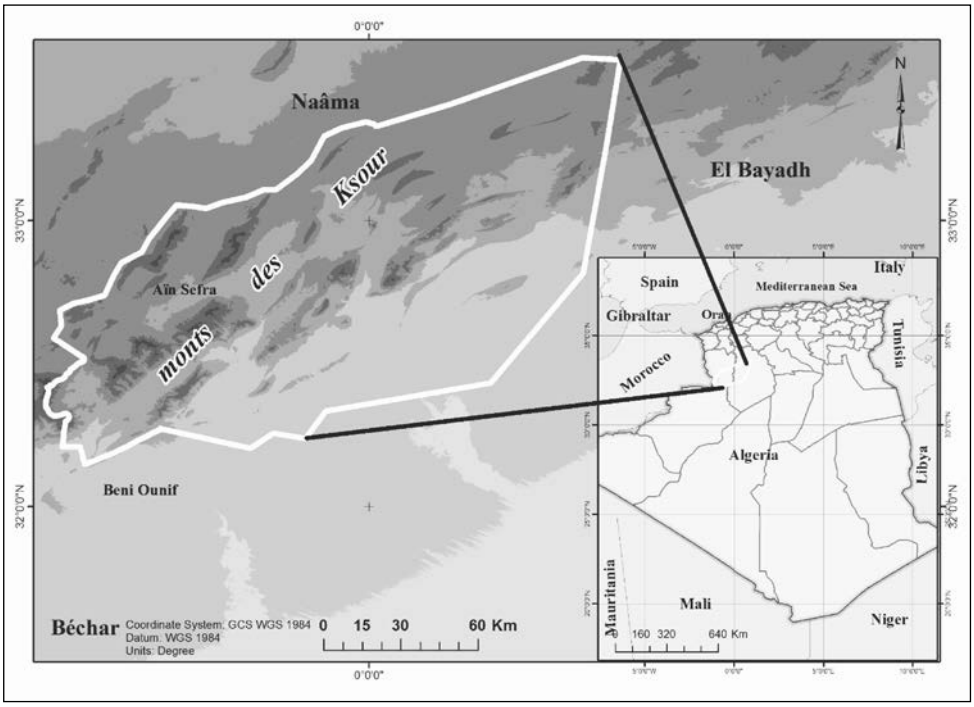

Fig. 1. Carte de localisation des monts des Ksour.

A l'ouest, ces massifs sont limités par la frontière algéro-marocaine et à l'est par le méridien d'Aïn Orak (El Bayadh).

Ce sont des chaînons orientés SW-NE, issus de la tectogenèse atlasique. Ces montagnes sont dissymétriques et couronnées de corniches de calcaire ou de grés. Ce sont les ravins d'anticlinaux et de larges synclinaux qui datent de l'ère secondaire et de l'éocène (Despois 1959). Du point de vue des substrats, l'Atlas saharien occidental est marqué par la dominance des faciès gréseux du jurassique et du crétacé inferieur (Maire 1916). Ses revers méridionaux sont formés de calcaire lacustre, de marno-calcaire et de faciès gréseux.

Sur le plan climatique, nous sommes ici sur une zone de chevauchement à la limite sud des influences du climat méditerranéen et la limite septentrionale du climat désertique chaud. D'autre part, les monts des Ksour jouissent d'une multitude de bioclimats (*sensu Emberger*). En effet, le bioclimat aride à hiver froid caractérise les hautes plaines (steppe) alors que sur les piémonts sud de cette chaine montagneuse c'est bien le bioclimat désertique tempéré qui règne. Par contre, les formations pré-forestières des hautes altitudes de montagnes sont caractérisées par le semi-aride à hiver très froid, à une altitude dépassant 2000 m (Gordo 2014).

Méthodologie

Dans le but d'étudier les groupements végétaux de cette région, nous avions à réaliser des relevés phytoécologiques. Pour cela, nos observations dans toute cette région ont commencé en 2012. Les campagnes de terrain ont été réparties au cours de l'année selon les conditions climatiques locales. En effet, les mois de mars et avril ont été réservés à la prospection des stations arides (formations steppiques) et hyper arides (faciès subdésertiques), tandis que les mois de mai à juillet ont été consacrés à l'exploration des montagnes, où domine un bioclimat semi aride très froid.

Pour identifier nos récoltes, nous avons utilisé la Nouvelle Flore de l'Algérie et des régions désertiques méridionales de Quézel et Santa (1962-1963), la Flore du Sahara d'Ozenda (1991), la Flore de l'Afrique du nord de Maire (1952-1987) et la Flore pratique du Maroc de Fennane & al. (1999-2014). Nous avons également consulté les travaux anciens, portant uniquement sur la flore des monts du sud oranais, tels que ceux de Hochreutiner (1904), Maire (1916) et Lemée (1953). De même, nous nous sommes appuyés sur, en plus de l'herbier de notre laboratoire à Oran, des herbiers disponibles en ligne tels que : Herbier du muséum national d'histoire naturelle de Paris (P), et l'Herbier de l'université de Montpellier (MPU).

Résultats et interprétations

Ainsi, nous avons réalisé environ 250 relevés phytoécologiques à travers toute la région: steppe, Atlas et désert. Cela nous a permis de réaliser un herbier personnel conservé au laboratoire ainsi qu'un catalogue floristique de 500 taxons environ.

Parmi tous les taxons endémiques du Maghreb trouvés, nous avons choisi de présenter dans cet article, les taxons dont l'aire est à cheval entre l'Algérie et le Maroc, c'est-à- dire les endémiques algéro-marocaines. La détermination de leur aire de répartition mondiale a été faite à partir des flores citées plus haut et des bases en lignes telles que : *African Plant Database* et *Euro Med Plant.*

Nous avons regroupé l'ensemble de ces taxons au nombre de 35 dans le tableau 1. En plus de la nomenclature acceptée, nous y avons rassemblé le ou les synonymes. De même, dans la troisième colonne, nous avons figuré l'aire de répartition de chaque taxon suivi par sa fréquence en Algérie ainsi que le type biologique et son habitat dans cette région.

Statut systématique et nomenclature

Concernant le nom accepté, nous avons adopté la nomenclature de *African Plant Database* (2018). Cependant, la synonymie retenue s'est inspirée de la Flore d'Algérie (1962-63) et la Flore pratique du Maroc (1999-2014).

Les divisions phytogéographiques, l'aire de répartition et la fréquence de chaque espèce dans son aire algérienne, ont été tirés de la Flore de Quézel & Santa (1962-1963). Le type biologique est déterminé au sens de Raunkier (1934).

Les 35 taxons inventoriés sont des angiospermes dicotylédones. Ils se déclinent en 26 espèces et 9 sous-espèces, réparties entre 13 familles et 30 genres (Tab.1). Les *Asteraceae* sont les plus importantes avec 15 taxons (12 espèces et 3 sous-espèces). Les familles qui suivent sont : les *Brassicaceae* avec 5 taxons (3espèces et 2 sous-espèces), les *Lamiaceae*

Tableau 1. Caractéristiques biologiques, écologiques et fréquentielles des taxons endémiques algéro-marocains.

Nom accepté	**Synonymes**	**Aires de répartition en Algérie**	**Fréq.**	**Typ. Biol.**	**Habitats dans les monts des Ksour**
Alyssum macrocalyx Coss. & Durieu	-	H1, AS1-2 & SS Ain Sefra, Laghouat, Ghardaïa, Ouargla	-	Th	Glacis septentrionaux de Dj. Mir El Djebel, Dj. Aïssa & Dj. Morghad 1200 - 1300 m
Atractylis delicatula Batt. ex L. Chevall.	-	AS1, SS Ben Zireg , Biskra jusqu' au Tademaït.	-	Th	Falaises & rocailles gréseuses, Ain Sefra jusqu'à Beni Ounif 850-1000 m
Bupleurum atlanticum subsp. *algeriense* Cauwet & Carb.	-	H1 & AS1-2	AR	Ch	Matorrals & pré-forêts monts Ksour 1600 - 2000 m
Carduus chevallieri Barratte	-	H1&AS1 Dj. Mekter, Dj.Taelbouna.	R	Hém	Matorrals, Thalwegs & versant nord Dj. Mekter 1500 m
Carthamus duvauxii (Batt.) Prain	*Carduncellus duvauxii* Batt.	AS1	RR	Hém	Voiles sableux Djeniene, Hadjret Elmguil & Beni Ounif 850 – 900 m
Carthamus cespitosus (Batt.) Greuter	*Carduncellus cespitosus* Batt.	AS1	R	Hém	Escarpements rocheux gréseux & sommets monts Ksour 1700 – 2100 m
Catananche caespitosa Desf.	-	C1,O3, H1-2 & AS3	AR	Hém	Matorrals dégradés Dj. Mekter 1500 m
Centaurea malinvaldiana Batt.	*Centaurea cossoniana Batt.* *C. granatensis subsp. battandieri* (Hochr.) Maire	AS1	R	Hém	Matorrals & préforêts Thalwegs & escarpements gréseux monts Ksour 1300-1900 m
Centaurea pomeliana subsp. rouxiana (Maire) Breitw. & Podlech	*C. pomeliana var. rouxiana* Maire	AS1 & AS2	R	Hém	Escarpements gréseux rocheux monts Ksour, 1700 – 2000 m
Centaurea pubescens subsp. saharae (Pomel) Dobignard	*C. saharae* Pomel, *C. incana* var. *saharae* (Pomel) Hochr.	Toute l'Algérie.	CC	Hém	Piémonts Dj. Aïssa, Dj. Mekter & Dj. Bou-Amoud 1300 m
Ceratolimon feei (Girard) M..B. Crespo & Lledó	*Limoniastrum feei* (Girard) Batt.	H&AS1 SS, SO	R C	Hém	Glacis méridionaux Dj. Mekter & Dj. Bou Amoud, Djeniene jusqu' à Beni Ounif 850- 950 m
Coronilla juncea subsp. pomelii Batt.	*C. juncea var. pomelii* (Batt.) Batt.	AS & SS	C	Nph	Matorrals dégradés Dj. Aïssa & Dj. Mekter 1400-1600 m
Crambe kralikii Coss. subsp. *kralikii*	-	AS1, SS& SC Hoggar	R	Ch	Rocailles gréseuses basses Dj. Mekter, oasis d'Ain Hadjadj & Moghrar Tahtani 950-1250 m
Crucianella hirta Pomel	-.	AS & SS	AC	Th	Glacis méridionaux & septentrionaux Dj. Mekter 1200-1300 m

Tableau 1. continue.

Erucastrum leucanthemum Coss. & Durieu	- .	Steppes des hautes plaines algéro-oranaises.	AC	Hém	Matorrals Dj. M'Zi, Dj. Mir El Djebel & Dj. Mekter 1400- 1600 m
Fraxinus dimorpha Coss. & Durieu	*F. xanthoxyloides* Wamm. .	K1&AS2 Bellezma, monts du Hodna & Aurès	RR R	Ph	Escarpements rocheux gréseux Dj. M'Zi, Dj. Mir El Djebel, Dj.Aissa & Dj. Mekter 1700-2000 m
Hippocrepis atlantica Ball	*H. scabra var. atlantica* (Ball) Maire	SS: Béchar Sauf dans les régions côtières.	R C	Ch	Matorrals dégradés Dj. Morghad 1600 m
Iberis atlantica (litard. & Maire) Greuter & Burdet	*I. linifolia* subsp. *atlantica* (Litard. & Maire) Maire *I. ciliata* subsp. *atlantica* Litard. & Maire	monts Tell au dessus de 900 m	R	Hém	Préforêts Dj. Morghad 1800 m
Linaria tristis subsp. *marginata* (Desf.) Maire	-	O3, A2, K1-2-3, C1, AS1-2-3.	AC	Hém	Escarpements rocheux gréseux Dj. Morghad, Dj.Mir El Djebel & Dj.Mzi 1800- 2000 m
Mauranthemum gaetulum (Batt.) Vogt & Oberpr.	*Chrysanthemum gaetulum* Batt. *Leucanthemum paludosum* subsp. *decipiens* var. *gaetulum (*Batt.) Maire	-	-	Th	Bords d'Oued dit Formene traversant Dj.Bou Amoud 1300 m
Mauranthemum reboudianum (Pomel) Vogt & Oberpr.	*Chrysanthemum reboudianum* (Pomel) Quézel & Santa *Leucanthemum. reboudianum* Pomel	AS3, C1: Boutaleb	R	Hém	Fissures des escarpements gréseux rocheux Dj. M'Zi & Dj. Mir El Djebel 1700 – 2000 m
Mecomischus halimifolius (Munby) Hochr.	*Anthemis halimifolia* Munby *Fradinia halimifolia* (Munby) Batt.	SS	R	Ch	Quelques sujets se développent au piémont nord de la dune d'Ain Sefra 1080 m
Micromeria hochreutineri (Briq.) Maire	*Satureja hochreutineri* Briq.	O3, H1 & AS1-2	R	Ch	Thalwegs & falaises gréseuses Tiout, Dj. Mekter 950-1400 m
Nepeta nepetella subsp. amethystina (Poir.) Briq.	-.	AS1-2	RR	Hém	Escarpements rocheux gréseux monts Ksour 1600-2000 m
Polycnemum fontanesii Durieu & Moq.	-	O2-3, H1 & AS1-2-3.	R	Hém	Matorrals Dj. Aïssa, Dj Mekter & Dj. Morghad 1300-1500 m
Pseudorlaya biseriata (Murb.)Sáenz de Rivas	*Daucus biseriatus* Murb.	SS1	AR	Th	Pâturages désertiques Beni Ounif 800-900 m
Rhanterium adpressum Coss. & Durieu	*R. suaveolens* subsp. *adpressum* (Coss. & Durieu) Quézel & Santa	SS&SO	R	Ch	Rocailles superficiellement ensablées piémont nord Dj.Mekter, bords Oued Bouyaala Beni Ounif 850 -1100 m.

Tableau 1. continue.

*Rhodanthemum gayanum (*Coss.& Durieu) B.H. Wilcox, K. Bremer & Humphries subsp. *gayanum*	*Leucanthemum gayanum subsp. antiatlanticum var. elatum* Maire (Maire)	AS1&AS3	R	Ch	Oued Laaouadj nord Dj. Mekter, thalweg versant sud Dj. Aissa 1200-1400 m
*Rhodanthemum maresii (*Coss.& Durieu) B.H. Wilcox, K. Bremer &Humphries	*Leucanthemum maresii* (Coss.) Maire, *Pyrethrum maresii* Coss.	AS1	R	Ch	Escarpements rocheux gréseux Dj. Mir El Djebel, Dj. M'Zi & Dj.Mekter 1700-2000 m.
Saccocalyx satureioides Coss.& Durieu	-	SS Hd & A1-2	RR R	Ch	Ouest de Mékalis près de la pinède Plantée ensablée 1250 m
Silene rouyana Batt.	-	AS1	AC	Hém	Escarpements rocheux gréseux Dj. M'Zi. 1800 -2000 m
Thesium mauritanicum Batt.	-	AS1	RR	Ch	Hautes rocailles Dj. Aïssa 1800 m
Thymelaea virescens Meisn.	-	H1-2, AS1-2-3	R	Ch	Matorrals dégradés, Dj. Morghad 1400-1600 m
Warionia saharae Benth. & Coss.	-	AS1 &SS1	R	Nph	Falaises gréseuses Tiout, Moghrar & Oued Lakhdar, jusqu' à Beni Ounif 850-1000 m
Zilla spinosa subsp. macroptera (Coss.) Maire & Weiller	-	AS1 & SS	C	Nph	Sud Dj. Aissa au sud Beni Ounif 800-1000 m

Fréq. (Fréquence); AC: Assez Commun; C: Commun; CC: Très Commun; AR: Assez Rare; R: Rare; RR: Très Rare.
Typ. Biol.: Type Biologique; Ph: Phanérophyte; Nph: Nanophanérophyte; Ch: Chaméphyte; Hém: Hémicryptophyte; Th: Thérophyte.
Secteurs biogéographiques selon Quézel & Santa (1962); O1, O2, O3: Oranie ; A1, A2 : Algérois; K1, K2, K3: Kabylie; Hd: Hodna; AS: Atlas saharien; AS1: Atlas saharien occidental (monts des Ksour); AS2: Atlas saharien central; AS3: Atlas Saharien Oriental; SS: Sahara Septentrional; SS1: Sahara Nord Occidental; SS2: Sahara Nord Oriental; SO: Sahara Occidental (Béchar à Tindouf); SC: Sahara Central.

avec 3 taxons (2 espèces et 1 sous-espèces), les *Apiaceae* et les Fabaceae avec 2 taxons chacune. Les familles des *Amaranthaceae*, *Caryophyllaceae*, *Oleaceae, Plantaginaceae*, *Plumbaginaceae*, *Rubiaceae, Santalaceae* et des *Thymelaeaceae* comptent 1 taxon chacune. Sur le plan générique, la famille des *Asteraceae* regroupe les genres les plus riches : *Centaurea* comprend 03 taxons, *Carthamus, Mauranthemum* et Rhodanthemum renferment 02 taxons chacun. Les genres restant sont représentés par 1 taxon chacun.

La détermination a été poussée jusqu'au niveau sous-spécifique pour *Bupleurum atlanticum* subsp. *algeriense* Cauwet & Carb., *Centaurea pomeliana* subsp. *rouxiana* (Maire) Breitw. & Podlech, et *Crambe kralikii* Coss. subsp. *kralikii*, reconnus par les anciens

auteurs (Hochreutiner 1904; Maire 1916; Quézel & Santa 1926-63; Ozenda 1991) au niveau spécifique seulement.

Distribution des taxons

A l'échelle algérienne, la distribution de ces taxons peut se présenter en 4 catégories d'aires (Tab.1).

-Dix-huit taxons (18) se répartissent dans les monts des Ksour et dans plusieurs secteurs limitrophes et / ou plus ou moins lointains,

-Six taxons (6) : *Carthamus duvauxii*, *Carthamus cespitosus*, *Centaurea malinvaldiana, Rhodanthemum maresii*, *Silene rouyana*, *Thesium mauritanicum*, ne se développent en Algérie que dans les monts des Ksour,

-Huit taxons (8) ont été observés par Hochreutiner (1904), Maire (1916), Battandier (1922) et Lemée (1953) et n'ont été signalés ni par Quézel et Santa (1962-63), ni par Ozenda (1991) dans la région. Ce sont : *Erucastrum leucanthemum*, *Fraxinus dimorpha*, *Hippocrepis atlantica*, *Mauranthemum gaetulum*, *Mauranthemum reboudianum*, *Mecomischus halimifolius*, *Pseudorlaya biseriata*, *Rhanterium adpressum*.

-Trois taxons (3) : *Catananche caespitosa*, *Iberis atlantica*, *Saccocalyx satureioides* n'ont jamais été signalés dans les monts des Ksour.

Rareté (sensu Quézel & Santa 1962)

Les espèces rares à différent degré (AR, R, RR) sont prépondérantes par rapport à celles considérées comme à peu près communes (AC, C, CC). Les trois taxons suivants : *Alyssum macrocalyx*, *Mauranthemum gaetulum, Atractylis delicatula* ne disposent pas de fréquences ; le deuxième ne figure ni dans la flore de Ozenda (1991) ni dans celle de Quézel & Santa (1962-63).

Statut légal des taxons

Parmi tous ces taxons, 4 espèces seulement sont considérées comme protégées par la législation algérienne (DE 2012) : *Mauranthemum reboudianum*, *Crambe kralikii* subsp. *kralikii, Fraxinus dimorpha, Saccocalyx satureioides*.

Ecologie des taxons

La diversité géomorphologique et orographique du secteur étudié a induit une remarquable complexité d'écosystèmes, avec des bioclimats caractéristiques et plusieurs types de substrats. Cela a donné naissance à l'installation d'une gamme de végétation diversifiée. Les montagnes par leur multiplicité d'exposition, d'orographie et d'altitudes : glacis, piémont, thalweg et escarpements, abritent des formations végétales à physionomie variable. Les préforêts, matorral et pelouse dominent comme biotopes hébergeant la majorité des taxons. Ils sont suivis par les rocailles gréseuses et falaises, ensuite viennent les milieux ensablés.

La répartition des types biologiques parmi les taxons rencontrés montre que les hémicryptophytes (15) représentent près de la moitié des taxons, suivis par les chaméphytes (11), les thérophytes (5), les nanophanérophytes (3) et les phanérophytes avec un seul taxon.

Discussion

De par sa position biogéographique particulière qui se combine avec la diversité d'habitats et de bioclimats, l'Atlas saharien occidental se caractérise par une flore riche et un endémisme transfrontalier algéro-marocain remarquable. Ce type d'endémisme se singularise nettement à l'échelle nationale par quelques taxons exclusifs des monts des Ksour, présents au Maroc. Cela reflète les similitudes floristiques, géomorphologiques et orographiques existantes. En effet, l'Atlas saharien occidental (AS1) est l'extension vers l'est de l'Atlas saharien marocain. Des travaux récents (CEPF 2017) ont confirmé ces liens géographiques, qui ont abouti à la formation de corridors écologiques, qualifiés de zone clés de la biodiversité. D'après Ben El Mostafa & al. (2001), le nombre élevé des taxons endémiques algéro-marocains existants au Maroc oriental est révélateur, car il confirme ce type de connections botaniques partagées. Les mêmes similitudes floristiques ont été également démontrées dans les monts des Traras (Tlemcen) qui constituent le prolongement naturel vers l'est du massif de Beni-Snassen du côté marocain (Sekkal & al. 2018).

Par ailleurs, tous les pays nord-africains notamment ceux du Maghreb, sont considérés comme réservoir de plantes endémiques (Quézel 1987). Les familles à forte endémicité telles que les *Asteraceae*, *Lamiaceae, Fabaceae*, *Caryophyllaceae*, *Brassicaceae,* sont également partagées au niveau des pays du Maghreb (Quézel 1964 ; Le Houérou 1995). Cette ressemblance est le résultat de leur histoire commune dont résulte une unité biogéographique très homogène (Quézel 1964).

Mauranthemum gaetulum (Batt.) Vogt & Oberpr., est une *Asteraceae* peu connue. Nous l'avons observé une seule fois à Oued Formene (Dj. Bou Amoud). Elle a été décrite sous le nom *Chrysanthemum gaetulum* pour la première fois par Battandier (1922) à Ben Zireg dans le sud oranais. La même plante a été ensuite signalée en Algérie par Jahandiez & Maire (1934) sous la combinaison *Leucanthemum paludosum subsp. decipiens var. gaetulum*. Nous n'avons pu l'identifier qu'en se référant à la Flore pratique du Maroc de Fennane & al. (1999-2014).

La comparaison de *Centaurea malinvaldiana* Batt., observé dans tous les monts des Ksour, avec le spécimen de (MPU), nous a permis de déduire que la photo de la plante qui figure sur African Plant Database (2018) ne correspond pas à ce taxon. Nos observations corroborent les résultats de Garcia Jacas & Susanna (1995) qui ont montré l'analogie phénotypique entre les individus de *Centaurea malinvaldiana,* provenant de l'Atlas saharien occidental.

Alors que les botanistes qui ont visité par le passé la région se sont arrêtés au niveau de l'espèce, le niveau sous-espèce a pu être délicatement défini pour 2 taxons : *Bupleurum atlanticum* subsp. *algeriense* et *Centaurea pomeliana* subsp. *rouxiana*. Cette identification a été rendu possible grâce aux révisions taxonomiques de Cauwet & Carbonnier (1977) pour la première espèce. Quant à la deuxième, nous nous sommes référés à l'herbier (P), dans lequel figure un spécimen collecté au Djebel M'Zi sur des rochers gréseux, en 1913 ; et à l'herbier (MPU), où Pomel (1860) et Roux (1880) l'ont récolté plus à l'est sur les monts des Amour à Ain Sebgag pour le premier et à Ain Berber (oued Sebgag) pour le second. La révision taxonomique de cette dernière espèce (Breitwieser, 1985) a permis d'identifier nos échantillons comme appartenant à la nouvelle sous-espèce *rouxiana*. De même, Fennane & Ibn Tattou (1998) ont signalé cette même sous-espèce dans l'Atlas saha-

rien marocain (As) au Djebel Maïz. Contrairement à *African Plan Database* (2018), la présence de subsp. *pomeliana*, nous parait incertaine dans les monts des Ksour, puisque tous nos spécimens correspondent à la sous-espèce *rouxiana.*

La diversité des expositions, de l'orographie et des substrats que nous avons observés, lors de l'analyse phytoécologique du Djebel Aïssa (2236m), un chaînon caractéristique des monts des Ksour, classé parc national en 2003, se généralise dans toute cette chaine montagneuse (Gordo 2014). Cela a été confirmé par Verlaque et al. (1997), qui ont évoqué la notion d'endémisme méditerranéen qui se concentre surtout dans les chaines montagneuses et les îles.

Concernant les taxons qui se développent sur les falaises et rochers, Quézel (2000) a confirmé leur richesse en espèces endémiques ou à aire disjointe. Le reste des taxa s'installe sur certains milieux légèrement ensablés.

La plupart des taxons inventoriés sont des hémicryptophytes. Ces derniers se caractérisent par des rosettes au ras du sol. Le développement de leurs hampes florales ne s'effectue qu'en printemps. D'après Barbéro & al. (2001), la prépondérance des hémicryptophytes dans les pays du Maghreb est due à l'humidité et à la présence de la matière organique. Cela nous semble logique, puisque la majorité de nos taxons occupent les montagnes à sols évolués. Quant aux chaméphytes, qui se placent en deuxième position, cela peut s'expliquer par la sécheresse et l'aridité qui règnent dans notre dition. Les chaméphytes sont mieux adaptés aux forts éclairements lumineux et à la sécheresse estivale (Danin & Orshan 1990). Les thérophytes qui ne survivent ni à l'été (sécheresse) ni à l'hiver (gel), occupent la troisième place : ils sont les mieux adaptés aux conditions extrêmes (Nègre 1966). Les phanérophytes et les nanophanérophytes sont peu représentées.

Concernant la distribution de ces taxons, la comparaison entre ce qui a été mentionné dans les travaux antérieurs et ce que nous avons vu sur le terrain, nous a amené à les classer en 4 catégories définies plus haut. Rappelons que les taxons exclusifs des monts des Ksour ou les plantes endémiques algéro-marocaines spécifiques de l'AS1 pourraient servir de point de départ pour caractériser et délimiter floristiquement cette entité biogéographique.

La détermination des statuts de conservation des taxons selon les critères l'UICN est indispensable et revêt une importance primordiale, plus particulièrement ceux exclusifs des monts des Ksour. Nous reconnaissons qu'il n'est pas possible d'atteindre cet objectif, si on n'a pas davantage de données précises concernant la taille et l'aire de répartition de ces plantes. Mais d'ores et déjà, nous proposons d'ajouter les taxons exclusifs : *Carthamus duvauxii*, *Carthamus cespitosus*, *Centaurea malinvaldiana*, *Rhodanthemum maresii*, *Silene rouyana*, *Thesium mauritanicum*, à la liste des plantes non cultivées et protégées figurant dans le Décret Exécutif (2012).

Signalons que seul *Fraxinus dimorpha* est considéré par UICN (2018) comme taxon en danger à l'échelle mondiale. En Effet, parmi nos taxons rares à différents degrés signalés par Quézel & Santa (1962-1963), sept (7) sont déclarés menacés au Maroc par Fennane (2016-2018). Cet auteur a adopté des catégories personnalisées en s'appuyant sur les critères de l'UICN. Pour le Maroc, les taxons classés dans les catégories menacés sont :

-Taxons en danger critique d'extinction (CR/$CR_{(e)}$): *Mauranthemum reboudianum, Saccocalyx satureioides* et *Thesium mauritanicum*,

-Taxons en danger (EN/$EN_{(e)}$): *Carthamus cespitosus*, *Centaurea malinvaldiana*, *C. pomeliana*,

-Taxon vulnérable (VU/$VU_{(e)}$): *Silene rouyana*.

Cette évaluation concorde largement avec nos observations, puisque tous ces taxons sont limités à des biotopes bien déterminés et restreints. Pour la première catégorie, *Mauranthemum reboudianum* est un chasmophyte qui n'occupe que certaines fissures des escarpements rocheux gréseux exposées au nord ou nord-ouest dans Dj. Mir El Djebel et Dj. M'Zi, dont l'altitude varie entre 1800 et 2000 m. Les deux autres taxons, sont les plus exposés à la dégradation antropozoogène.

En ce qui concerne la deuxième catégorie, mis à part *Centaurea malinvaldiana* que nous avons observé également dans les thalwegs*, Carthamus cespitosus* et *Fraxinus dimorpha* se développent uniquement sur les escarpements gréseux et quelques hauts sommets à l'abri de l'action anthropique.

La dernière catégorie comporte *Silene rouyana*, que nous n'avons observée qu'au Dj. M'Zi dans les escarpements gréseux les plus septentrionaux et les plus accidentés. Aussi, Il convient de souligner que ces escarpements gréseux, hébergent une végétation remarquable, où le frêne dimorphe et le chêne vert dominent en formant des peuplements riches et denses dans tous les monts des Ksour compte tenu des conditions stationnelles favorables telles que l'altitude et humidité.

D'autre part, Fennane (2016-2018) a dressé une liste renfermant d'autres taxons considérés au Maroc comme quasi-menacée (NT), parmi ces espèces, figurent:

Alyssum macrocalyx, *Bupleurum atlanticum*, *Hippocrepis atlantica*, *Rhanterium adpressum* et *Warionia saharae*, que nous retrouvons dans les monts des Ksour. Notons que cette dernière espèce sur- exploitée par les herboristes, ne se maintient que dans les hautes falaises gréseuses plus au moins inaccessibles.

La présence du côté algérien de deux taxons (*Adenocarpus bacquei* et *Cytisopsis ahmedii*) dans les Djebel Beni Smir et Grouz est très probable. La visite de ces deux montagnes n'a pas été possible à cause de la situation sécuritaire qui règne cette zone frontalière avec le Maroc.

D'autres taxons endémiques tels que : *Andryala chevallieri* Barrate ex L. Chevall, *Hyssopus officinalis susbsp. austro-oranensis* Maire et *Linaria peltieri* Batt., sont à rechercher dans notre dition. Ils semblent avoir disparu ou du moins être en déclin puisque nous n'avons pas pu les retrouver dans les stations où ils avaient été observés par de nombreux auteurs. Cela peut s'expliquer par la dégradation de leurs habitats (ruisseau, source d'eau, dune), sous l'effet de l'action dévastatrice de l'homme et de ses troupeaux. A cela s'ajoute d'autres facteurs, tel que l'ensablement qui a pris des dimensions alarmantes dans la région d'Aïn Sefra (Bouarfa & Bellal 2018).

Conclusion

Cette étude avait pour but principal la connaissance de la flore des monts des Ksour. Ainsi, nous avons pu confirmer la présence de divers taxons endémiques algéro-marocains. Elle se veut aussi un complément d'information aux travaux antérieurs, venant combler certaines lacunes liées à la répartition des ces taxons, à la description de leur habitat, et à leur statut de conservation.

Les 35 taxons identifiés sont représentés par 26 espèces et 9 sous-espèces. Parmi celles-ci, 3 sont observées pour la première fois dans les monts des Ksour, et 7 y ont été confirmées.

Concernant la répartition de ces taxons à l'échelle algérienne, une nouvelle aire de distribution propre aux taxons exclusifs des monts des Ksour (AS1) est définie. Ces derniers sont proposés au classement sur la liste de plantes non cultivées et protégées figurant dans le décret exécutif (2012).

Dans le même ordre d'idée, nous insistons sur la nécessité de préserver certains habitats tels que les escarpements rocheux gréseux et les sommets des montagnes, par le biais de la création des aires protégées et/ou des mises en défends. Ces mesures s'avèrent nécessaires car la conservation des habitats est le moyen le plus efficace et le plus économique pour sauvegarder la biodiversité (Meyrs 2000). Soulignons également que les monts des Ksour en tant que zones de transitions bioclimatiques peuvent constituer des centres de spéciation suite à des micro- isolements (Amirouche & Misset 2009). A l'issue de cette étude, nous souhaitons que nos prochaines recherches apportent davantage d'informations concernant la phytosociologie et la dynamique des communautés végétales de l'Atlas saharien occidental.

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An analysis of the bryophyte flora in Sicilian archaeological areas*

Abstract

Campisi, P., Dia, M. G. & Marino, M. L.: An analysis of the bryophyte flora in Sicilian archaeological areas. — Fl. Medit. 29: 143-157. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

An analysis of the bryophyte diversity in the studied Sicilian archaeological areas was conducted, highlighting which species are more common and potentially harmful on the ruins.
The floras are much diversified and the presence of some rare taxa highlights the role of refuge carried out by these areas, especially for the species of strongly threatened coastal habitats. Attention on the complexity of the relationships between restoration interventions on lithic structures and conservation needs of the rare and interesting taxa is point out.

Key words: Mediterranean area, biodeterioration, rare bryophyte taxa.

Introduction

Among Italian regions, Sicily is certainly one of those that boasts a particularly rich archaeological heritage, consisting of over 50 sites distributed throughout the territory, in many cases subjected to specific protections (Grella & al. 1997).

As evidenced by several studies, the archaeological areas are of considerable interest also from the naturalistic point of view as they represent refuge areas for numerous species threatened by urban expansion and generally host a high floristic diversity (Celesti Grapow & al. 1993-1994; Lucchese & Pignatti Wikus 1995; Ceschin & al. 2006; Minissale & al. 2015).

In Italy the studies on the flora of the archaeological sites started in the Nineties and Sicily is one of the regions most concerned by this research activity (Domina 2018).

In addition to the vascular component, also the cryptogamic one was investigated in Sicily, in order to improve the knowledge of the main biodeteriogens on the remains of the ancient human-made structures (Raimondo & al. 2008). Several researches have focused in particular on bryophyte floras, starting from the Nineties, as was pointed out by Gueli & al. (2005). Some of these studies have highlighted the presence in Sicilian archaeological areas of very rare taxa and therefore of conservation interest (Dia & al. 2003; Campisi

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& Provenzano 2004; Dia & Campisi 2006;Campisi & al. 2008; Dia & Campisi 2009), drawing attention to the need to clarify the deteriogenic role of bryophytes to evaluate in some cases the hypothesis of their maintenance on the ruins.

Here we analyze so far studied bryophyte floras in some archaeological areas of Sicily with the aim of evaluating the bryophyte diversity of the areas and highlighting which species are more common and potentially harmful on the ruins as well as wich species are worthy of conservation.

Materials and methods

The Archaeological Parks of Solunto and of Mount Iato in Palermo province, those of Segesta and Selinunte e Cave di Cusa in Trapani province, the Neapolis Archaeological Park of Syracuse town, the Temple of Cerere in Enna province and the Greco-Roman Theatre and the Roman Amphitheatre inside Catania town have been taken into consideration in this analysis (Table 1). In view of their naturalistic interest, some of these areas, in particular Mount Iato Archaeological Park, Selinunte and Cave di Cusa Archaeological Park and Solunto Archaeological Park, are included in the list of Sites of Community Interest (ITA020027 Monte Iato, Kumeta, Maganoce e Pizzo Parrino; ITA010011 Sistema dunale Capo Granitola, Porto Palo e Foce del Belice; ITA020019 Rupi di Catalfano e Capo Zafferano).The present study was based on bibliographic and unpublished data. In particular, we have considered the papers of Privitera & al. (1996), Lo Giudice & Cristaudo (1998), Di Benedetto & Grillo (1998), Puglisi (1999), Guglielmo & al. (2003) and Aiello & al. (2003). Furthermore, the data concerning bryophyte material collected by the authors in the Sicilian archaeological areas of Selinunte and Mount Iato were also taken into consideration.

The archeological areas of Selinunte, Syracuse, Catania, Solunto and Segesta are located at lowland or hilly altitudes (10-415 m a.s.l.) on the contrary Mount Iato and Enna are located at sub-mountain altitudes (850-970 m a.s.l.). Vegetation is mostly represented by xerophilous or mesophilous grasslands and garrigues; only at Selinunte psammophilous, wetlands communities and Mediterranean maquis also occur (Table 1).

The relevant samples are kept at the *Herbarium Mediterraneum Panormitanum* (PAL).

Table 1. Studied areas and relatives data of surface, altitude and vegetation types.

Archaeological area	Abbreviation	Altitude (m a.s.l.)	Area size (ha)	Vegetation type
Mount Iato Archaeological Park (Palermo)	Ia	850	40	Mesophilous grasslands, garrigues
Segesta Archaeological Park (Trapani)	Sg	415	30	Xerophilous grasslands, garrigues
Selinunte and Cave di Cusa Archaeological Park (Trapani)	Sl	10	215	Psammophilous communities, Mediterranean maquis, wetlands communities, crops
Neapolis Archaeological Park (Syracuse)	Si	25	22	Xerophilous grasslands, crops
Solunto Archaeological Park (Palermo)	So	215	20	Xerophilous grasslands, garrigues
Greco-Roman Theatre and the Roman Amphitheatre (Catania)	Ca	25	0,6	Sinantropic communities
Temple of Ceres (Enna)	En	970	0.4	Subnitrofilous xerophilous grasslands

The nomenclature of the species comply with Söderström & al. (2016) for liverworts and hornworts and Ros & al. (2013) for mosses, with the exception of *Didymodon tophaceus* complex for which Kučera & al. (2018) is followed .

In order to characterize the floras of Sicilian archaeological areas, chorological and ecological features were compared. Chorotypes were taken from Düll (1983, 1984, 1985, 1992), where almost all taxa of studied sites are reported, with same abbreviations and for the construction of the chorological histogram they were after joined in five groups: temperate (temp, s.temp, temp-mont, subkont); oceanic-Mediterranean (oc-med, oc-submed, suboc-med, suboc-submed); Mediterranean (med, med-oc, submed, submed-oc, submed-suboc, submed-suboc-mont submed-mont); boreal (subbor, subbor-mont); oceanic (suboc).

Ellenberg's indicator values relating to light, temperature, moisture and substrate reaction, were taken from Düll (1991).

All available data were assembled in a Microsoft Access database, where for each taxon ecological indicator values, chorotypes, and occurrence in the Sicilian archaeological areas were recorded.

For each site, the ecological indicator mean values were calculated to draw radar diagram. In order to measure the level of similarity of the floras, a hierarchical cluster analysis using Jaccard's similarity index (Jaccard 1908), applied on a data matrix including presence–absence of moss and liverwort taxa, was performed with the Biodiversity Professional program (McAleece & al. 1997).

The bioclimatic thermotypes were derived from the thermotypes map provided by Pesaresi & al. (2014).

Moreover, with the aim of identifying the potentially most dangerous species for the integrity of the ruins, in the sites of Selinunte, Segesta and Solunto the following data were recorded by the authors in the field and analized:

1) frequency of taxa in five classes based on percentage occurrence in the total collection points of every archaeological area (I: 0–20%, II: 21-40%, III: 41-60%, IV: 61-80%, V: 81-100%); 2) percentage cover in five classes based on percentage cover of each species on the total surface colonized by the bryophytes in 30 ×30 cm areas, in every archaeological area (I: 0–20%, II: 21-40%, III: 41-60%, IV: 61-80%, V: 81-100%); 3) sporophyte or propagule production (production was considered high (+) when their occurrence was recorded in more than 30% of plants in the collected specimens, low (-) if in 30% of plants or less).

Life strategies were taken from Dierβen (2001) with same abbreviations.

In all figures and tabular material studied sites were indicated with following abbreviations: Ca: Greco-Roman Theatre and the Roman Amphitheatre in Catania town; En: Temple of Ceres in Enna town; Ia: Mount Iato Archaeological Park; Sg: Segesta Archeological Park: Si: Neapolis Archaeological Park in Syracuse town; Sl: Selinunte and Cave di Cusa Archeological Park; So: Solunto Archaeological Park.

Results and Discussion

Overall, the bryophyte flora of the Sicilian archaeological areas studied so far includes 88 taxa (75 mosses and 13 liverworts). They are listed in Table 2, where the presence in each area, ecological indicators and chorotypes are reported.

Table 2. List of taxa of studied archaeological areas and their ecological indicators (from Düll 1991) and chorotypes (from Düll 1983, 1984, 1985, 1992). L: light; T: temperature; M: moisture; R: substrate reaction. The abbreviations of archeological areas are given in Table 1.
Temp: temperate; s.temp: south-temperate; temp-mont: temperate-montane; subkont: subcontinental; oc-med: oceanic-Mediterranean; oc-submed: oceanic-submediterranean; suboc-med: suboceanic-Mediterranean; suboc-submed: suboceanic-submediterranean; med: Mediterranean; submed: submediterranean; submed-oc: submediterranean-oceanic; submed-suboc: submediterranean-suboceanic; submed-suboc-mont: submediterranean-suboceanic-montane; submed-mont: submediterranean-montane; med-oc: Mediterranean-oceanic; subbor: subboreal; subbor-mont: subboreal-montane; suboc: suboceanic.

	Archaeological areas							**Ecological indicators**				**Chorotypes**
Taxa	**Ca**	**En**	**Ia**	**Sg**	**Sl**	**Si**	**So**	**L**	**T**	**M**	**R**	
Cephaloziella baumgartneri Schiffn.	●	x	●	●	●	x	x	5	7	5	●	oc-med
Conocephalum conicum (L.) Dumort.	●	●	●	●	●	x	●	7	3	7	7	subbor-mont
Fossombronia caespitiformis (Raddi) De Not.	●	●	●	x	x	x	x	7	9	5	8	oc-med
Fossombronia pusilla (L.) Nees	●	●	●	●	●	x	●	7	7	7	3	suboc-submed
*Lunularia cruciata (*L.) Dumort. ex Lindb.	x	●	●	●	x	x	●	7	8	6	6	oc-med
Pellia endiviifolia (Dicks.) Dumort.	●	●	●	●	●	x	●	●	4	8	9	s.temp
Riccia atromarginata Levier	●	●	●	●	●	●	x	●	●	●	●	med-oc
Riccia glauca L.	●	●	●	●	x	●	●	8	5	7	5	submed
Riccia lamellosa Raddi	●	●	●	●	x	●	x	8	9	5	●	med
Riccia sorocarpa Bisch.	●	●	●	●	x	●	●	9	●	5	5	temp
Southbya tophacea (Spruce) Spruce	●	x	●	●	●	●	●	●	●	●	●	oc-med
Sphaerocarpos michelii Bellardi	●	●	●	x	x	●	●	7	8	6	6	suboc-submed
Targionia hypophylla L.	●	●	●	●	x	●	●	6	7	4	5	oc-submed
Aloina aloides (Koch ex Schultz) Kindb.	●	●	x	x	●	x	x	7	6	4	9	temp
Aloina ambigua (Bruch & Schimp.) Limpr.	●	●	●	●	x	x	x	7	6	4	8	submed
Aloina rigida (Hedw.) Limpr	●	●	x	●	x	●	●	7	4	5	6	temp
Barbula bolleana (Müll.Hal.) Broth.	●	●	●	●	●	x	●	8	9	8	8	submed
Barbula convoluta Hedw.	●	●	x	●	x	x	x	8	●	3	6	temp
Barbula unguiculata Hedw.	x	●	x	x	x	x	x	7	●	2	7	temp
Bryum argenteum Hedw.	●	●	x	●	x	x	x	7	●	4	6	temp
Bryum canariense Brid.	x	●	●	x	●	●	●	●	●	●	●	submed
Bryum dichotomum Hedw.	●	●	●	x	x	x	x	8	6	6	5	submed
Bryum radiculosum Brid.	●	●	●	x	●	●	x	9	6	2	7	suboc-med
Crossidium crassinervium (De Not.) Jur.	●	●	●	●	x	●	x	9	8	2	8	submed

Table 2. continued.

Crossidium laxefilamentosum W.Frey & Kürschner	●	●	●	●	●	●	x	●	●	●	●	●
Crossidium squamiferum (Viv.)Jur.	●	●	●	●	x	●	x	9	8	x	8	submed
Dicranella howei Renauld & Cardot	●	●	●	x	x	●	●	9	8	5	8	oc-med
Didymodon acutus (Brid.) K.Saito	●	●	x	●	x	x	x	9	5	●	8	submed
Didymodon luridus Hornsch.	●	x	x	●	x	x	x	9	6	2	8	submed
Didymodon rigidulus Hedw.	●	x	●	●	●	●	x	5	3	4	7	temp
Didymodon tophaceus (Brid.) Lisa	●	x	x	●	●	●	●	7	●	7	7	temp
Didymodon tophaceus subsp. *sicculus* (M.J.Cano, Ros, García-Zam. & J.Guerra) Jan Kučera	●	●	●	x	●	●	x	●	●	●	●	med
Didymodon vinealis (Brid.) R.H.Zander	x		x	x	x	x	x	9	6	2	7	submed
Entosthodon muhlenbergii (Turner) Fife	●	●	●	●	x	●	●	9	6	5	8	submed-suboc-mont
Entosthodon pulchellus (H.Philib.) Brugués	●	●	●	x	x	●	●	8	8	5	8	submed-suboc
Eucladium verticillatum (With.) Bruch & Schimp.						x		5	7	7	9	submed
Fissidens crassipes Wilson ex Bruch & Schimp.	●	●	●	●	●	x	●	●	6	8	8	suboc-submed
Fissidens gracilifolius Brugg.-Nann. & Nyholm	●	●	●	●	●	x	●	3	4	6	9	temp-mont
Fissidens pusillus (Wilson) Milde	●	●	●	●	x	●	●	3	4	6	6	temp-mont
Fissidens viridulus (Sw. ex anon.) Wahlenb var. *viridulus*	●	●	●	●	x	x	x	7	5	6	8	submed
Fontinalis antipyretica Hedw.	●	●	●	●	●	x	●	8	5	9	●	subbor
Funaria hygrometrica Hedw.	●	●	x	x	x	●	●	8	●	6	6	temp
Funariella curviseta (Schwägr.) Sérgio	●	●	●	x	x	●	●	5	8	5	7	med
Gigaspermum mouretii Corb.	●	●	●	●	x	●	●	●	●	●	●	oc-med
Grimmia crinita Brid.	●	x	●	●	●	●	●	9	8	●	8	submed
Grimmia orbicularis Bruch ex Wilson	●	x	●	●	x	●	●	9	7	●	8	submed-mont
Grimmia pulvinata (Hedw.) Sm	●	x	●	●	●	●	x	9	5	●	7	temp
Gymnostomum calcareum Nees & Hornsch.	●	x	x	●	●	x	●	4	7	5	9	submed-mont
Gymnostomum viridulum Brid.	●	●	●	●	x	●	x	4	8	4	9	suboc-med
Homalothecium aureum (Spruce) H.Rob.	●	●	●	●	x	●	●	8	9	2	7	med
Homalothecium lutescens (Hedw.) H.Rob. var. *lutescens*	●	●	x	●	x	●	●	9	4	2	8	temp
Homalothecium sericeum (Hedw.) Schimp.	●	x	●	●	●	x	●	8	3	2	7	temp
Hygroamblystegium varium (Hedw.) Mönk. *var. humile* (P.Beauv.)	●	●	●	●	●	x	●	5	5	6	4	temp
Kindbergia praelonga (Hedw.) Ochyra						x		6	4	6	5	temp
Microbryum davallianum (Sm.) R.H.Zander	●	●	●	x	x	●	●	8	5	6	6	submed

Table 2. continued.

Microbryum starckeanum (Hedw.) R.H.Zander	●	●	x	x	x	●	●	9	6	7	5	submed
Orthotrichum diaphanum Schrad. ex Brid	●	●	●	●	x	●	●	8	6	2	6	temp
Oxyrrhynchium speciosum Nees var. *speciosum*	●	●	●	●	●	x	●	5	7	7	6	subkont
Pleuridium acuminatum Lindb.	●	●	●	●	●	x	●	7	5	5	4	suboc
Pohlia wahlenbergii (F.Weber & D.Mohr) A.L.Andrews var. *wahlenbergii*	●	x	●	●	x	●	●	6	●	7	6	subbor
Pseudocrossidium hornschuchianum (Schultz) R.H.Zander	x	●	x	●	x	x	x	9	5	2	7	submed-suboc
Pseudocrossidium obtusulum (Lindb.) H.A.Crum & L.E.Anderson	●	●	●	●	●	●	x	●	●	●	●	●
Pseudocrossidium replicatum (Taylor) R.H.Zander	●	●	●	●	●	●	x	7	7	3	8	temp
Pseudocrossidium revolutum (Brid.) R.H.Zander	●	●	●	●	x	●	x	7	6	3	8	oc-submed
Ptychostomum capillare (Hedw.) Holyoak & N. Pedersen	●	x	x	●	x	x	●	5	●	5	6	temp
Ptychostomum imbricatulum (Müll.Hal.) Holyoak & N.Pedersen	x		x	x	●	x	x	8	●	5	6	temp
Rhynchostegiella littorea (De Not.) Limpr.	●	●	●	●	x	●	x	4 -	5	5	8	oc-med
Rhynchostegiella tenella (Dicks.) Limpr.	●	●	●	●	x	●	●	4	5	3	8	submed-suboc
Rhynchostegium riparioides (Hedw.) Cardot	●	●	●	●	●	x	●	●	3	8	6	temp
Scleropodium touretii (Brid.) L.F.Koch.	●	●	●	●	●	x	●	8	7	3	6	oc-submed
Scorpiurium circinatum (Bruch) M.Fleisch. & Loeske	●	x	●	x	x	x	x	6	7	3	5	oc-med
Syntrichia montana Nees	●	x	●	●	●	●	●	9	6	●	8	submed-mont
Syntrichia ruralis (Hedw.) F.Weber & D.Mohr	●	●	●	●	●	x	●	9	●	2	6	temp
Timmiella anomala (Bruch & Schimp.) Limpr.	x	●	●	●	●	x	●	5	9	5	5	med
Timmiella barbuloides (Brid.) Mönk.	●	●	●	x	x	x	x	5	9	5	8	med
Tortella flavovirens (Bruch) Broth.	●	●	●	●	x	●	●	8	5	2	8	suboc-submed
Tortella inflexa (Bruch) Broth.	●	●	●	●	x	x	●	4	8	3	9	oc-med
Tortella nitida (Lindb.) Broth.	●	●	x	●	x	x	x	8	8	2	7	oc-med
Tortella squarrosa (Brid.) Limpr.	●	●	●	●	x	x	●	9	8	2	6	submed
Tortula acaulon (With.) R.H.Zander var. *pilifera* (Hedw.) R.H.Zander	●	●	●	●	x	●	●	9	6	2	7	temp
Tortula marginata (Bruch & Schimp.) Spruce	●	●	●	x	x	x	●	3	8	5	9	oc-med
Tortula muralis Hedw.	●	●	x	x	x	x	x	8	5	●	●	temp
Tortula vahliana (Schultz) Mont	●	●	●	●	x	●	●	●	●	●	●	oc-med
Trichostomum brachydontium Bruch	●	●	●	●	x	x	●	8	6	2	8	submed-mont

Table 2. continued.

Trichostomum crispulum Bruch	•	•	•	•	x	x	•	6	4	6	9	temp
Weissia condensa (Voit) Lindb. var. *condensa*	•	•	•	•	•	•	x	9	7	•	9	submed
Weissia controversa Hedw. var. *controversa*	•	x	•	•	•	x	•	7	4	4	6	temp
Weissia controversa var. *crispata* (Nees & Hornsch.) Nyholm	•	•	•	•	•	•	x	9	4	•	7	submed-mont

As shown in Table 3, bryophyte species numerosity of the studied archaeological areas is very variable and does not exclusively depend on the size of the areas. The archaeological park of Selinunte, which is also by far the largest of those studied, hosts the highest number of bryophytes (51 taxa); however, in relation to the size of the areas the species richness of the Syracuse and Solunto parks is remarkable (38 and 33 taxa respectively).

From the chorological point of view, the floras are quite diversified since the Mediterranean taxa prevail in the floras of Selinunte (43.1%), Segesta (50%) and Solunto (43,8%) parks and Catania site, while the temperate taxa are the most represented in Enna site (40%) and in Mount Iato (55.6%) and Syracuse parks (36.4%) (Fig. 1). The high incidence of the latter type in the first two archaeological areas is likely to be related to their mountain altitudes, while in Syracuse park could be due to the significant presence of wet microhabitats in it.

An analysis of the average values of the Ellenberg index (Fig. 2) shows a relative uniformity of the floras with reference to the behaviour of the species with respect to the various factors, since the average values show variations of at most 1 and 1.5 units. Some significant differences may, however, be noted such as particularly high xerophily of the Solunto and Catania floras (average values of the moisture factor 3.7), the relatively low values concerning the temperature factor at the Enna site and in Mount Iato park (average value less than 6), the low value related to the reaction of the substrate in the area of Catania (6.3 average value).

A qualitative comparison of the floras, conducted through a cluster analysis, highlighted an accentuated diversification of the floras, as demonstrated by the low levels of link in the dendrogram of Fig. 3. Furthermore, it is observed that the clusters reflect the thermoclimatic conditions of the sites. The greatest similarity is found, in fact, among the floras of

Table 3. Number of bryophyte taxa in the archaeological areas. For abbreviations see Table 1.

	Sl	Ia	Sg	Si	So	Ca	En
Mosses	44	18	18	36	28	6	13
Liverworts	7	0	2	6	4	1	2
Total bryophyte taxa	51	18	20	42	32	7	15

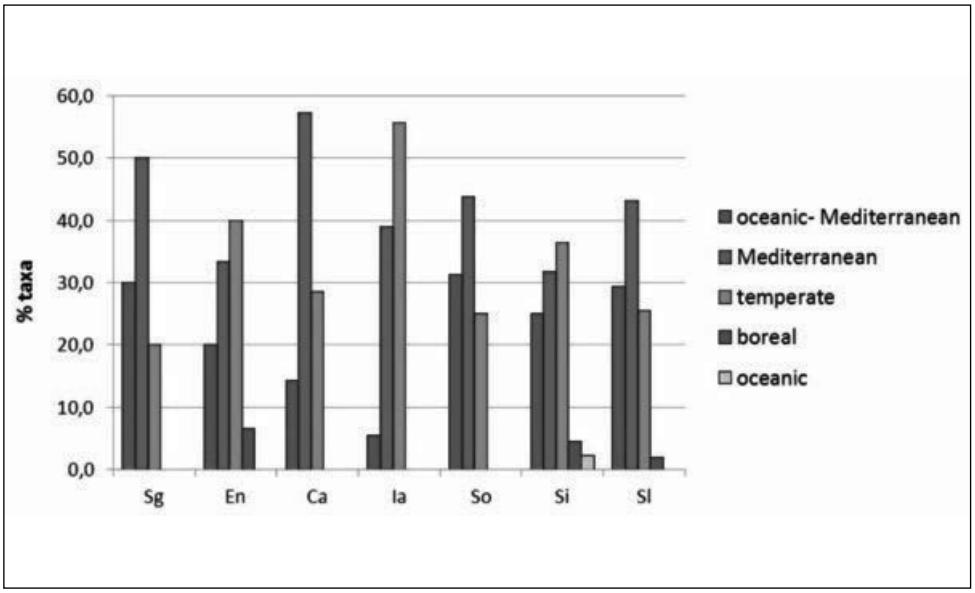

Fig. 1. Chorological Spectrum of bryofloras of studied areas. Chorological data are taken from Düll (1983, 1984, 1985, 1992). For abbreviations of archeological areas see Table 1.

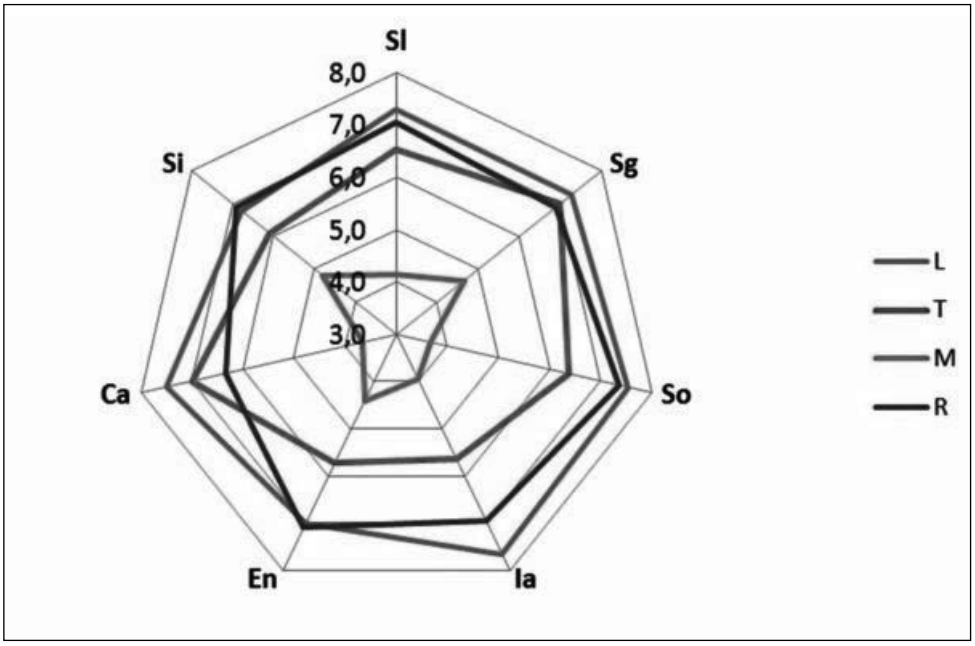

Fig. 2. Radar graph of average Ellenberg's indicator values (from Düll 1991) for taxa of studied bryofloras. The abbreviations of archeological areas are given in Table 1.

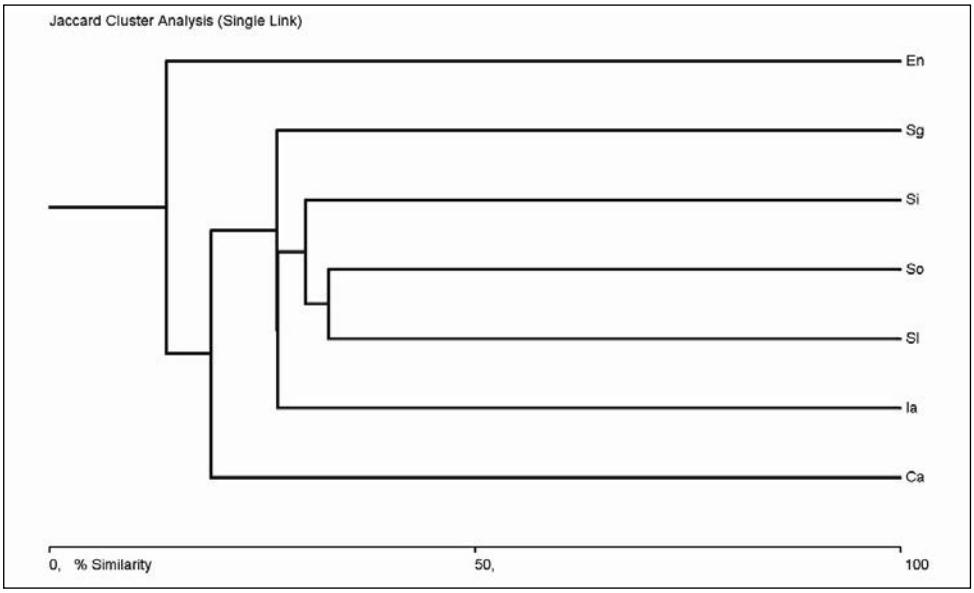

Fig. 3. Dendrogram of Jaccard's similarity (Jaccard 1908) among studied bryofloras.

Selinunte, Solunto and Syracuse, characterized by a lower thermomediterranean climate thermotype and to this cluster the floras of Mount Iato and Segesta (lower mesomediterranean climate thermotype), the flora of Catania (upper thermomediterranean climate thermotype) and, lastly, the one of Enna (upper mesomediterranean climate thermotype) are sequentially linked.

In addition to the high dissimilarity, the peculiarity of the floras of the Sicilian archaeological areas is due to the presence of taxa that be able to live almost exclusively in coastal areas with little disturbance. Among them four taxa, *Crossidium laxefilamentosum* W. Frey & Kürschner, *Gigaspermum mouretii* Corb., *Pseudocrossidium obtusulum* (Lindb.) H.A. Crum & L.E. Anderson and *P. replicatum* (Taylor) R. H. Zander., are of notable interest as they are very rare in Italy and candidates to inclusion in the European Red Data Book (Hodgetts 2015).

Pseudocrossidium replicatum was reported in Europe and Mediterranean basin only in Sicily (Solunto and Linosa) and Calabria (Dia & al. 2003; Privitera & Puglisi 2000; Ros & al. 2013; Hodgetts 2015). Elsewhere it is distributed in Central and South Africa, South-West Arabia and America (Zander 1981; Menzel 1986; Frey & Kürschner 1988a 1988b). In the archeological park of Solunto it lives on basic soil in interstices between quartz mosaic tesserae of the houses floors.

Pseudocrossidium obtusulum was reported from North America Europe, Asia and Africa, (Fedosov & Ignatova 2006; Khalil & Farag Abu-Elhamd Ali 2018). It is very rare in the Mediterranean basin, where it is only known from Andorra, Egypt, Sicily, Spain and Turkey (Ros & al. 2013; Khalil & Farag Abu-Elhamd Ali 2018). In Sicily, it was only recorded in Solunto and Linosa (Dia & Campisi 2006; Privitera & Puglisi 2009). At Solunto it lives togeth-

er with *Pseudocrossidium replicatum* in interstices between quartz mosaic tesserae of the houses floors in the archaeological park.

Gigaspermum mouretii is an oceanic-Mediterranean species scattered in Syria, Israel, Cyprus, Turkey, Crete, Sicily, Morocco, Canary Islands, Spain, and Balearic Islands (Ros & al. 2013). In Italy it was reported only from two locality of Sicily (Selinunte and Capaci) (Campisi & al. 2008). At Selinunte it grows on calcarenite plinth of the temples E and F.

Crossidium laxefilamentosum was reported from Europe , Asia (Arabian peninsula) and Africa. In the Mediterranean basin it is known from Egypt, Serbia, Sicily, Tunisia and Turkey (Ros & al. 2013). In Sicily it was recorded only in two archaeological areas (Solunto and Molino a Vento near Gela) (Dia & Campisi 2009; Puglisi & al. 2013). At Solunto it lives on exposed soil among ruins of the archaeological park.

On the basis of coverage, frequency, abundance of sporophytes and propagules data as well as the life strategies of taxa living in the Segesta, Selinunte and Solunto archaeological parks, some species of mosses were identified as a potentially great threat to the state of conservation of the ruins in consideration of the direct correlation between species diffusion and coverage on the one hand and their possible biodeteriogenic action on the other. In particular, they are *Tortula muralis* Hedw., *Scorpiurium circinatum* (Bruch) M. Fleisch. & Loeske and *Didymodon vinealis* (Brid.) R. H. Zander, which are present in all three areas, always reaching cover class II or higher. Furthermore, *Aloina ambigua* (Bruch & Schimp.) Limpr., *Barbula convoluta* Hedw., *Bryum dichotomum* Hedw., *Didymodon luridus* Hornsch., *Funariella curviseta* (Schwägr.) Sérgio, *Pseudocrossidium hornschuchianum* (Schultz) R. H. Zander and *Tortella nitida* (Lindb.) Broth. occure in at least 2 areas with high frequency or coverage class (III or more) at least in one area. All these species, with the exception of the pleurocarp moss *Scorpiurium circinatum*, are annual or colonists, biological strategies that according to Dierßen (2001) are characterized by short-lived (<1 year-few years) with more or less high reproductive effort. They present a high production of sporophytes already in the first year of growth or after 2-3 years or form also propagules in the first years of life and, therefore, tend to expand more and more on the stone substrates, continuously creating new colonies. On the contrary, the perennial species *Scorpiurium circinatum*, shows reproductive effort low and begin to form sporophytes only after several years. It forms very wide moss mats that extend above all on horizontal surfaces of archaeological structures in the studied areas.

The liverworts also show a high reproductive capacity both through the sporification and with different modality of vegetative propagation (propagules production in *Lunularia cruciata* (L.) Lindb. and y-shaped growth form in *Riccia* species). Five out of seven species (*Fossombronia caespitiformis* De Not. ex Rabenh., *Riccia glauca* L., *R. lamellosa* Raddi, *R. sorocarpa* Bisch., and *Sphaerocarpos michelii* Bellardi) present an "annual" strategy, being characterized by a very rapid growth of the vegetative body (few months) and very high formation of spores (Dierßen 2001). However, these species spread more widely on soils and, therefore, do not reach high frequency values on the ruins, where, except for *Fossombronia caespitiformis*, they have a low degree of coverage due to their small size.

Regarding the periods of maximum sporification, it is observed that most bryophytes release the spores in the spring (Table 4). In some cases the maturation of the sporophytes is a little early and occurs already in later winter (February and March), in others it is pro-

Table 4. Frequency and cover classes, sporophyte or propagule presence, time of sporophyte production and life strategies (from Dierßen 2001) of taxa growing in the sites of Segesta (Sg), Selinunte (Sl) and Solunto (So). a: annual; c: colonist; ec: ephemeral colonist; l: long-lived shuttle p: perennial; pc: competitive perennial; s: short-lived shuttle; sp: stress tolerant perennial. For abbreviations of archeological areas see Table 1.

Taxa	Frequency classes			Cover classes			Sporophyte or propagule			Sporification period	Life strategies
	Sg	Sl	So	Sg	Sl	So	Sg	Sl	So		
Hepaticae											
Cephaloziella baumgartneri	●	●	I	●	●	I	●	●	+	Late winter-early	c
Fossombronia caespitiformis	I	II	I	IV	II	II-III	+	+	+	Winter	a
Lunularia cruciata	●	I	●	●	I	●	●	+	●	●	p
Riccia atromarginata	●	●	I	●	●	II	−	●	−	Late winter-early	a
Riccia glauca	●	I	●	●	I	●	−	+	−	Late winter-early	a
Riccia lamellosa	●	I	I	●	I	II	−	+	+	Late winter-early	a
Riccia sorocarpa	●	I	●	●	I	●	−	+	−	Late winter-early	a, s
Sphaerocarpos michelii	I	I	●	I	I	●	+	+	−	Late winter-early	a
Targionia hypophylla	●	I	●	●	I	●	+	+	+	Spring-summer	l
Musci											
Aloina aloides	I	●	I	I	●	I	−	●	●	Winter-spring	c
Aloina ambigua	●	III	I	●	III	I	●	+	−	Winter-spring	c
Aloina rigida	●	I	●	●	I	●	●	●	●	Winter-spring	c
Barbula convoluta	●	III	I	●	I	I	●	●	−	●	c
Barbula unguiculata	I	I	I	I	I	I	●	●	●	Spring	●
Bryum argenteum	●	I	I	●	I	I	●	●	+	Winter-spring	c
Bryum canariense	I	●	●	II	●	●	●	●	●	●	
Bryum dichotomum	I	III	I	II	III	I	●	+	+	Spring	cp
Bryum radiculosum	I	●	I	II	●	I	+	●	+	●	ce
Crossidium crassinerve	●	I	I	●	I	II	●	●	+	Spring	c
Crossidium laxefilamentosum	●	●	I	●	●	I	●	●	●	●	c
Crossidium squamiferum	●	I	I	●	I	I	●	●	+	Spring	c
Dicranella howei	I	II	●	I	I	●	●	●	●	Winter-spring	c
Didymodon acutus	●	I	II	●	I	I-II	●	●	●	Spring	c
Didymodon luridus	●	IV	II	●	III	I-II	●	●	●	Spring	c
Didymodon rigidulus	●	●	I	●	●	I	●	●	+	●	c
Didymodon tophaceous subsp. *sicculus*	II	●	I	II	●	I	●	●	●	●	c
Didymodon tophaceus	●	I	●	●	I	●	●	●	●	Spring	●
Didymodon vinealis	II	III	II	I-II	III	I-II	●	●	●	Spring-summer	c
Entosthodon muhlenbergiii	●	I	●	●	I	●	●	●	●	Spring	a
Entosthodon pulchellus	I	I	●	I	I	●	+	+	●	Spring	a
Fissidens viridulus	●	I	I	●	I	II	●	●	+	Spring-summer	ec
Funaria hygrometrica	I	I	●	I	II	●	+	+	●	Spring-summer	f
Funariella curviseta	III	I	●	I-IV	II	●	+	+	●	Spring	a
Gigaspermum mouretii	●	I	●	●	I	●	●	●	●	Autumn	c
Grimmia orbicularis	●	I	●	●	I	●	●	+	●	Spring	c
Grimmia pulvinata	●	●	I	●	●	I	●	●	+	Winter-spring	c
Gymnostomum viridulum	●	II	I	●	II	II	●	●	+	Spring-summer	c
Homalothecium aureum	●	I	●	●	I	●	●	●	●	Spring	p
Homalothecium lutescens	●	I	●	●	I	●	●	●	●	Spring	p
Microbryum davallianum	II	I	●	I	I	●	+	+	●	Winter-spring	a
Microbryum starckeanum	I	I	●	I	I	●	+	+	●	Winter-spring	a
Orthotrichum diaphanum	●	I	●	●	I	●	●	+	●	Spring	c
Pohlia wahlenbergii	●	I	●	●	I	●	●	●	●	Spring-summer	pc
Pseudocrossidium	●	II	I	●	III	I	●	●	●	Spring	c
Pseudocrossidium replicatum	●	●	II	●	●	II	●	●	●	●	c
Pseudocrossidium revolutum	●	I	II	●	II	I	●	●	+	Spring	c
Ptychostomum capillare	●	II	●	●	I	●	●		●	Spring-summer	c
Ptychostomum imbricatulum	II	●	I	I-II	●	II	●	●	●	Spring-summer	c
Rhynchostegiella littorea	●	I	I	●	I	I	●	●	●	Spring	sp
Rhynchostegiella tenella	●	I	●	●	I	●	●	●	●	Autumn	sp
Scorpiurium circinatum	I	III	I	V	V	II-V	●	●	●	Spring	p
Timmiella barbuloides	I	I	I	V	II	III	+	●	●	Spring-summer	s
Tortella flavovirens	●	I	●	●	I	●	●	●	●	Spring	c
Tortella inflexa	●	I	●	●	I	●	●	●	●	Spring	c
Tortella nitida	●	IV	III	●	V	II-V	●	+	+	Autumn	sp
Tortella squarrosa	●	I	●	●	I	●	●	●	●	Spring	pc

Table 4. continued.

Tortula acaulon var. *pilifera*	●	I	●	●	I	●	●	●	●	Autumn-spring	a
Tortula marginata	I	I	●	I	I	●	+	−	●	Spring	c
Tortula muralis	IV	IV	IV	II-IV	III	II-IV	+	+	+	Spring-summer	c
Tortula vahliana	●	I	●	●	I	●	●	●	●	Spring	c
Trichostomum brachydontium	●	II	●	●	I	●	●	●	●	Spring	p, s
Trichostomum crispulum	●	I	●	●	I	●	●	●	●	Spring	c
Weissia condensa	●	●	I	●	●	I	●	●	−	Winter-spring	c
Weissia controversa var. *crispata*	●	●	I	●	●	I	●	●	●	Winter-spring	c

longed in the summer. Few species have autumnal sporification. Therefore, the analysis of these data suggests that the winter is the most suitable season to guarantee the effectiveness of the interventions on the ruins.

Conclusive considerations

Overall, this analysis confirms the naturalistic interest of archaeological areas for their significant floristic diversity and species richness, whose values are comparable to those recorded in Sicily in some natural areas (Campisi & al. 2006). The importance of these areas is also increased by the presence of some rare taxa in Europe for which specific attention would be required during the necessary, periodic, restorative interventions on remains of architectural structures.

These cleaning interventions should primarily be aimed at the removal of widespread bryophytes with numerous or extensive colonies, considering that the action of biodeterioration is certainly related to the degree of bryophyte coverage. Furthermore, the possibility of diffusion of spores and propaguls due to colony detachment operations should not be underestimated.

Nevertheless, it should be emphasized that further research is desirable to better clarify the biodeteriogenic role of different bryophyte taxa, so far ascertained only in some epilithic moss species (Hughes 1982; Altieri & Ricci 1994; Altieri & al. 1997). Many species on lithic structures, indeed, are not true saxicolous but they settle only in small accumulations of soil in grooves and fractures caused by the alteration usually due to other deteriogenic agents; hence, the possibility that they, like other terricolous bryophytes, can exercise, instead, a protective action on substrates cannot be excluded with certainty at least in some cases. The important role of moss coverings against soil erosion is, in fact, well known both in forest ecosystems and in arid habitats where they contribute to the formation of the biological soil crusts, very effective to counteract the action of atmospheric agents (Weber & al. 2016).

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Short note

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N. Sakhraoui, S. Metallaoui & A. Chefrour

Naturalisation d'*Anredera cordifolia* (*Basellaceae*) en Algérie

Abstract

Sakhraoui, N., Metallaoui, S. & Chefrour, A.: Naturalisation d'*Anredera cordifolia* (*Basellaceae*) en Algérie. — Fl. Medit. 29: 159-162. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

The occurrence and actual status of *Anredera cordifolia* (*Basellaceae*) in Algeria is here reported, for the first time. The species has been recorded in many localities within Skikda area. It is the only representative of *Basellaceae* in the whole country. Maintained in observation area within the native flora for many years, the plant could be considered as naturalized.

Key words: *Boussingaultia cordifolia*, alien species, degree of naturalization, Algeria.

Introduction

En période coloniale, l'Algérie a connu une introduction massive de matériel végétal pour des fins alimentaire, commerciale ou ornementale. En 1867, Hardy, le directeur du jardin d'essai du Hamma, dénombre 8214 espèces et variétés (Carra & Gueit 1952) dont plusieurs sont aujourd'hui naturalisées ou presque dans les différentes régions du pays (Meddour & al. 2016) avec un probable devenir envahissant. Les espèces envahissantes constituent un véritable danger pour la biodiversité via la réduction de la richesse spécifique autochtone (Vilà & al. 2015), la pollution génétique et l'érosion génétique. Elles sont d'ailleurs, considérées comme la deuxième cause d'érosion de la biodiversité après la destruction et la fragmentation des habitats (OTA 1993).

Au cours de la réalisation de prospections botaniques dans la wilaya de Skikda, située au NE Algérien (Sakhraoui & al. 2019), nous avons noté la présence d'une espèce exotique à léger potentiel envahissant dont l'existence et le statut en Algérie n'ont jamais été rapportés, il s'agit de la liane de Madère, *Anredera cordifolia* (Ten.) Steenis. Les relevés floristiques que nous avons réalisés tentent essentiellement à mettre le point sur les différents milieux colonisés dans un but à envisager des stratégies de lutte contre son éventuelle extension. Certaines caractéristiques biologiques observées in situ ont été notées (période de floraison, aptitude à la fructification et modalité de reproduction végétative). L'appartenance taxonomique, l'origine et la date d'introduction de cette espèce ont été établies à partir de la littérature. Le type biologique a été déterminé selon Raunkiaer (1934)

et le statut d'indigénat selon Magnanon & al. (2008). Les spécimens récoltés ont été déposés dans l'herbier de la wilaya maintenu au niveau du laboratoire de botanique du département des sciences de la nature et de la vie de l'Université de Skikda.

Description, répartition géographique et degré de naturalisation/statut actuel

Anredera cordifolia (Ten.) Steenis, Fl. Malesiana, Ser.1, Spermatoph. 5(3): 303 (1957) (=*Boussingaultia cordifolia* Ten. in Ann. Sci. Nat., Bot. sér. 3, 19: 355 (1853)).

A. cordifolia communément appelée liane de Madère, est une herbe à gros tubercule souterrain, à tiges volubiles. Feuilles alternes cordiformes ou ovales un peu charnues, entières. Les fleurs sont petites, blanches disposées en grappes terminales et axillaires (Maire 1962). La plante est une géophyte, originaire d'Amérique du Sud (Wagner & al. 1999), qui a été introduite en Algérie entre 1842 et 1867 par le jardin d'essai du Hamma (Carra & Gueit 1952).

Elle a également été introduite dans de nombreux pays en tant que plante d'ornement où elle est devenue envahissante notamment en Australie, Afrique du Sud, Nouvelle Zélande et Hawaï (Weber 2017). En méditerranée, la liane de madère est naturalisée en Italie (Galasso & al. 2018), en Grèce (Arianoutsou & al. 2010) et en Croatie (Stancic & Mihelj 2010), alors qu'elle est considérée comme envahissante avérée en Espagne (Capdevilla Argüelles & al. 2006). Dans les zones d'introduction, l'espèce semble se reproduire uniquement par voie végétative (Stancic & Mihelj 2010 ; Kottaimuthu & al. 2011**),** les nombreux tubercules aériens accrochés aux tiges semblent jouer un rôle important dans la propagation rapide de l'espèce. Ces derniers peuvent se détacher facilement et rester viables de 5 à 10 ans (Vivian-Smith & al. 2007). D'après Kolar & Lodge (2001), les probabilités d'envahissement augmentent si la plante possède des antécédents d'envahissement et une reproduction végétative.

En Afrique du Nord, sa présence comme plante d'ornement a été rapportée par Maire (1962) sans indication précise des pays dans lesquels elle a été cultivée. Plus récemment, la plante est rapportée naturalisée au Maroc (Uotila 2009; APD 2019) alors qu'en Algérie elle n'a jamais été signalée naturalisée (Quézel & Santa 1962; 1963; Dobignard & Chatelain 2011)**.**

En Algérie, et plus précisément dans notre zone d'étude, cette espèce a été retrouvée en 2014 dans plusieurs points des cités Saleh Chebel (commune de Hamadi Krouma) et Larbi Ben M'Hidi (commune de Skikda), notamment à la lisière des jardins d'où elle a tendance à échapper. A la cité Larbi Ben M'Hidi, une grande population a été observée au bord de la route du poste 3 dans un terrain abandonné où elle pousse horizontalement, formant un tapis étendu et verticalement en s'accrochant à divers arbustes avoisinants (*Pistacia lentiscus*, *Acacia karoo*, *Ephedra fragilis*, etc.). Ce point de prolifération, situé à quelques mètres du maquis arborescent à *Quercus coccifera*, *Juniperus phoenicea* et *Chamaerops humilis* qui caractérise la dune littorale, est connu d'après le témoignage des habitants depuis au moins 11 années. Elle a également été repérée, en 2015, sur le djebel Mouadher à l'Est de la ville de Skikda en face de la zone industrielle, où elle occupe une surface plus importante et y est représentée par une population plus dense. Les oliviers proliférant sur place lui servent de tuteurs. Dans les différentes localités, la floraison a été observée à partir du

mois de Juin jusqu'au mois d'Octobre de 2014 à 2018. Les fruits, par contre, n'ont pas été récoltés. La plante peut être considérée comme naturalisée dans la région de Skikda, puisqu'elle se maintient dans l'une des zones d'observation depuis au moins 11 ans et fait part de la flore indigène (Magnanon & al. 2008). *A. cordifolia* devrait être surveillée pour éviter qu'elle ne devienne envahissante avérée dans les prochaines années.

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Article

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Cristina Salmeri

Plant morphology: outdated or advanced discipline in modern plant sciences?*

Abstract

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In the last decades, with the increase of molecular studies, the study of plant forms has gone through a steady decline in interest, and researches on this topic are often neglected and underestimated. Notwithstanding, comparative morphology as integrative discipline still assumes a pivotal role in modern sciences, remaining fundamentally relevant to nearly all fields of plant biology, such as systematics, evolutionary biology, ecology, physiology, genetics, molecular biology, not to mention also agriculture, bioengineering, and forensic botany. Contrary to common belief, plant morphology is not a conservative finished science, but, like other sciences, it is open to constant innovations involving both concepts and methods. This contribution aims to promote a reflective discourse on the role of plant morphology in modern sciences and provides some examples of significant supports from plant morphology to different botanical issues.

Key words: Systematics, plant micromorphology, seed coat sculpturing, leaf anatomy, ecomorphology, climate adaptation.

Introduction

Despite the increasing societal awareness and sensitivity about the knowledge of biological diversity and ecosystem functioning as pivotal matters for nature conservation on which human health and well-being fundamentally depend, studies in morphology-based classical taxonomy have increasingly become marginalized and considered less significant than other scientific methods in plant biology. This has led to a progressive decline in attention both at research institutions and funding allocation, and nowadays most scientists and academic students think of plant morphology as just a classical and largely outdated field of research.

Plant morphology is a biological discipline that aims at understanding the biology of plant organisms on the basis of their structural appearance, so it essentially consists in the scientific investigation on the plant forms and/or structures.

*Extended and enriched version of the oral presentation given at the International Symposium "Botany at the intersection of Nature, Culture, Art and Science", Selinunte, 28-30 June 2018.

As a discipline, plant morphology can be intended in either a narrow or a broad sense. In narrow sense, plant morphology is the science of external forms and their relationships expressed at the whole plant down to the organ level of organization. In broad sense, plant morphology includes forms and structures at each level of organization, that is whole plant, organs, tissues, cells, cell organelles, etc.; in this respect plant morphology also involves micromorphology, anatomy and cell ultrastructure (Sattler 1978; Sattler & Rutishauser 1997).

Traditionally, the study of plant morphology takes its origin in the history of botany. From early times, great importance was made in the geometrical appearance of plant organs and consequently many taxonomic groups (especially genera and species) were named accordingly to their morphological characters. Similarly, also several plant families have had their taxonomic name based on characteristics of the shape of flowers or other morphological features (Betz 2006; Cervantes & de Diego 2010). Since the introduction of the term intended as a scientific discipline by Goethe (1790), modern plant morphology has resulted from an eventful history (Kaplan 2001; Claβen-Bockhoff 2001).

Contrary to a widespread misconception of being conceived as the science of static forms, plant morphology has showed an intrinsic dynamic essence. Firstly, it deals with the topological and sequential changes of plant forms and structures throughout the time, during ontogeny and phylogeny. In addition, it has dynamically changed over time and improved its theoretical and analytical approaches embracing new technologies and tools, without neglecting traditional methods (Sattler 1990; Liem 1991; Ledford 2018).

Progresses in plant morphology have influenced research in various disciplines of plant biology which fundamentally use or imply morphological concepts, such as systematics, evolutionary biology, ecology, physiology, genetics, molecular biology. Even in the current times of genomics (plus many other "omics" topics) and functional ecology, when trait-based approaches are essential for studying and understanding plant functions and species relationships, it is clear that plant morphology, used as comparative and integrative approach, still assumes a pivotal role, remaining fundamentally relevant to nearly all fields of plant science (Sattler & Rutishauser 1997; Kaplan 2001; Scotland & al. 2003; Pochynok 2012; Schönenberger & von Balthazar 2012; Schönenberger & al. 2016; Bucksch & al. 2017).

Lots of studies have demonstrated how vegetative and reproductive characters, and their anatomical and/or micro-morphological structures, can be informative for phylogenetic studies and helpful to solve systematic problems at various levels. Most phenotypic traits show adaptive variation and different range of plasticity which have proved to be of great ecological and physiological significance and useful, for instance, in reconstructing plant adaptation to past climatic conditions or establishing defence mechanisms and structural changes in response to stress, climate changes and plant invasions, which all are basic information for nature conservation. Meanwhile, understanding patterns and origins of such morphological modifications and how plant traits connect to gene activity across species is crucial to address main evo-devo questions. Not to mention the basic role of plant morphology in other applied sciences, such as agriculture, bioengineering, and forensic botany.

Notwithstanding, plant morphology and the strictly related descriptive taxonomy are often considered a lower form of science, not fashionable and very far from the biggest challenges facing humanity. If not associated to molecular and phylogenetic data, they are in general underappreciated in many highly visible journals and inadequacy of research

funding in these field is disproportionate relative to other disciplines in biology (Pyšek & al. 2013; Tahseen 2014; Coleman 2015), even though it seems that in the last decade taxonomic publications increased faster than those from other biological fields and reached high citation performances (Steiner & al. 2015).

This contribution intends to promote a reflective discourse aiming to change the current scientific culture towards a better acknowledgment and academic evaluation of research in plant morphology as the backbone of many other fields of plant biology and applied biosciences. Examples of fundamental interrelations between plant morphology and other biological disciplines are provided.

Material and Methods

Macromorphology

Studies on gross morphology were performed on both living wild plants collected from many localities of the Mediterranean area (exsiccates are preserved in CAT) and cultivated plants (10 to 30 individuals per species). Qualitative and quantitative morphological features were examined and recorded under a Zeiss Stemi SV11 Apo stereomicroscope at 6–66× magnification, on fresh samples when possible. Morphological comparison was also based on herbarium collections from various botanical museums and literature data. Both vegetative and reproductive characters were chosen according to their diagnostic value for discriminating among the investigated taxa and populations.

Micromorphology

Micromorphology was studied under a Zeiss EVO LS10 scanning electron microscope (SEM Zeiss, Oberkochen, Germany) using mature dehydrated samples set onto aluminium stubs with double adhesive tape and coated with gold prior to observation. Scanning electron micrographs were performed at an accelerating voltage of 10 kV and 18-1000× magnifications, depending on samples size. Terminology used for leaf surface and seed-coat sculpturing was based on Wilkinson (1979) and Barthlott (1981, 1984).

Leaf anatomy

Leaf blades of maximum size in their optimal vegetative development were used for the anatomical study. Cross sections 25 µm thick were made using a Leica CM 1900® cryostat (Leica, Wetzlar, Germany) at a temperature of -20 °C, then stained with 1% w/v aqueous Safranin. Vascular patterns were emphasized through leaf clarification based on the Fuchs' method (Fuchs 1963) modified by avoiding leaf tissue maceration in dried oven at 60◦C. Semipermanent slides were mounted with glycerol/water 1:1 and sealed with transparent nail polish. Photographs were taken under Zeiss Axioskope 2 light microscope equipped with digital camera.

Statistical analyses

Micro-morphological and anatomical characters were measured from five to ten different samples for each investigated species/population. Measurements were done using the Zeiss AxioVision Rel. 4.8.2 image analysis software.

Statistical analyses were computed on both quantitative and qualitative morpho-anatomical parameters and performed using XLSTAT 2018.1.1 software (Addinsoft) on Microsoft Excel platform. Simple descriptive statistics of the intra- and inter-phenetic diversity (mean, standard deviation, range, median, and so on) were calculated from the data. The statistical effect of climate conditions on leaf traits among different populations was estimated by simple and multiple linear regression models. Mean annual precipitation (MAP) and mean annual temperature (MAT), from each population site, were used as the explanatory variables. De Martonne Aridity index (expressed by the formula AI_{DM}= MAP/MAT + 10) was also calculated and tested as potential predictor for climate influence on leaf structure. Multivariate analyses were performed in order to assess the similarity or dissimilarity among populations and the degree of separation of different groups. A step-wise discriminant factorial analysis (DFA) was employed on measured data (using the method of inclusion and removal at each step) The determination of the most discriminating variables was carried out by means of Fisher's coefficient at the significant threshold value of 0.05. The posterior probability of classification of each sample (cross validation) and the Wilks' lambda value of each variable were also calculated. Principal coordinates analysis (PCoA) was carried out on general dissimilarity matrix from 23 qualitative morphological characters.

Results

Descriptive morphology and taxonomic issues

The comparative study of plant structures, at both macro- and micromorphological level, has always been the backbone of plant Systematics. There exist lots of review summarising the main role of structural aspects in systematic botany at different taxonomic levels, where the comparative morphology appear to be still necessary and helpful, and reliable determination keys based on morphological characters continue to be a major information source for species identification and distinction (e.g. Greuter 1973; Ronse & al. 2010; Endress 2011; Kendorff & al. 2015; Mannino & al. 2015; Nardi 2015; Brullo & Erben 2016; Iamonico 2016; Brullo & al. 2018; Colasante 2018). It may help in areas and at levels of the tree of life where molecular studies are difficult for some reason.

This is the case of wide complicated genera, such as the genus *Allium* whose systematic arrangement in subgenera and sections is largely based on specific combination of discriminant morphological features (Fritsch 2001; Khassanov & al. 2011; Govaerts & al. 2018; Brullo & al. 2019), which was in many cases confirmed by molecular approaches (Friesen & al. 2006; Nguien & al. 2008; Hirschegger & al. 2010; Li & al. 2010). The role of comparative morphology in *Allium* taxonomy was also essential to clarify the systematic positions and relationships within several species' complexes, many of which have proven to include cryptic species. This is, for example, the case with the *Allium cupanii* Raf. group where a distinctive combination of morphological diagnostic features, i.e. fibrous and more or less markedly reticulate outer bulb tunics, basally adherent or detached, filiform leaves, with cylindrical to semicylindrical outline, subglabrous to densely hairy leaf indumentum, persistent spathe, with 1 or 2 valves basally connate, partially sheathing the flower pedicels, few-flowered inflorescence, usually fastigiate and unilateral, arranged in

2 or 4 bostryces when the spathe is 1-valved or 2-valved respectively, perigon cylindrical to urceolate, white-pinkish to pink-purplish, simple stamen filaments included into the perigon, ovary with well-developed nectariferous pores, covered by a membranous plica, and capsule included into the perigon, suggested a more appropriate inclusion of this group in the autonomous sect. *Cupanioscordum* Chesm., also confirmed by molecular investigations; the intrinsic variability in these morphological traits allows to identify five distinct series and many new species whose populations were all formerly identified sub *A. cupanii* (bulb coats basally attached) or sub *A. hirtovaginatum* Kunth (bulb coats basally detached) (Brullo & al. 1995; Brullo & al. 2015; Salmeri & al. 2015). Another investigated critical group in the genus *Allium* was the *Allium paniculatum* L. complex. Based on literature and many herbarium collections, *A. paniculatum* was frequently conceived as having an extremely wide geographic distribution (from West to East Europe, North Africa and Asia), both in synanthropic and wild habitats, and a large morphological variability. Detailed surveys on herbarium collections, including the type specimen of the species, and on living plants from lots of different territories of the Euro-Mediterranean and Irano-Turanian regions, actually revealed that many different taxa of *A.* sect. *Codonoprasum* Rchb., all characterized by big size, diffuse and densely flowered umbrella, very long spathe valves, long pedicels, and cylindrical-campanulate perigon, have been wrongly attributed to *A. paniculatum*, thus affecting records on the geographic distribution and morphological characterization of this species. Thus, while the true *A. paniculatum* resulted to be native and circumscribed to the far eastern European territories (Ukraine and Russia), many other allied but distinct taxa (previously treated as *A. paniculatum*) have been described and well discriminated on the basis of the combination of a few relevant morphological traits (Brullo & al. 2001; Brullo & al. 2008; Salmeri & al. 2015).

Then again, comparative morphological analyses concerning vegetative and reproductive structures, such as bulbs, leaves, inflorescence and seed, were crucial to assess the current systematics of the complex genus *Scilla* L. *sensu lato*, leading to its splitting in different closely related, but taxonomically well-differentiated natural genera within the *Hyacinthaceae* family, largely confirmed by molecular data (Speta 1998; Pfosser & Speta 1999, 2004). Most of these features are easily detectable also in the field and have proven very useful in discriminating among the Italian squills (Fig. 1), for which identification keys to the existing genera and species were also implemented (Brullo & al. 2007).

Other useful examples come from leaf morphology and anatomy, which provided relevant elements for characterizing and discriminating species of the genus *Dittrichia* Greuter. This genus is represented by five distinct taxa, all distributed in the Mediterranean area, from West (*D. viscosa*, *D. revoluta*, *D. maritima*) to East (*D. orientalis*), and partly extended into the Irano-Turanian region (*D. graveolens*). Despite a certain intraspecific variability (Brullo & al. 2004), clear and stable interspecific differences were found in the leaves (but not only), regarding their shape and size, types of margin, apex, venation, and hairs, plus some anatomical aspects of palisade and spongy tissues, which can be considered valuable characters supporting the distinction at specific level (Fig. 2).

Insights from comparative micromorphology

Plant micromorphology is the study of finer details of external features, based on micro-level analyses of leaves, pollens, seeds, petals and other plant organs. The diversity of plant

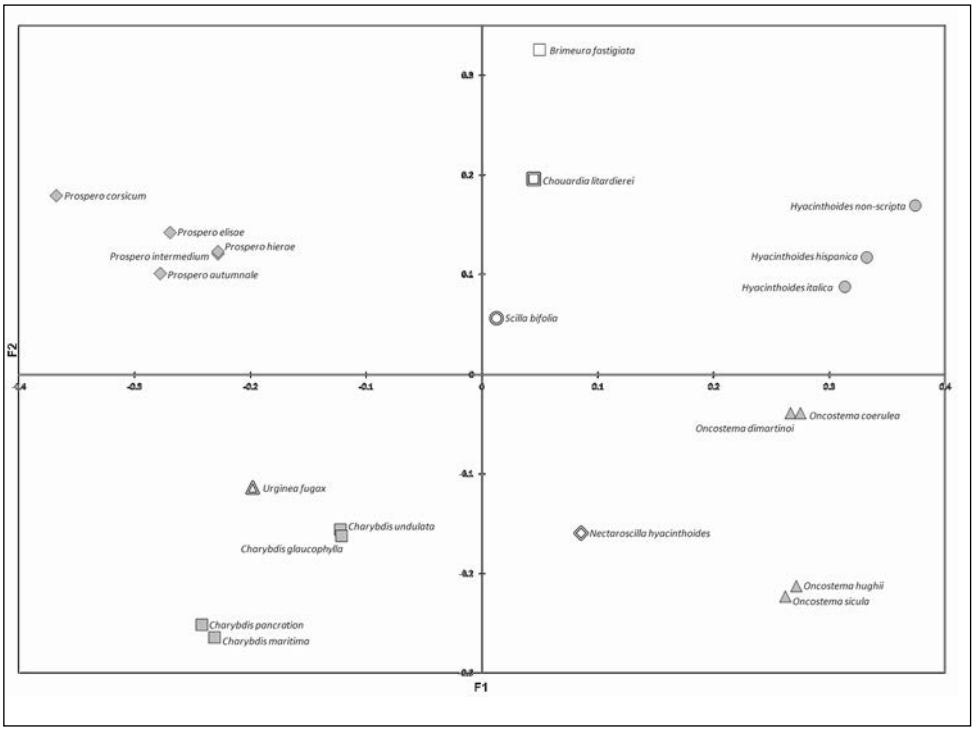

Fig. 1. Scatter plots resulting from principal coordinates analysis (PCoA) on *Scilla* s.l. species based on a data matrix of 23 morphological data.

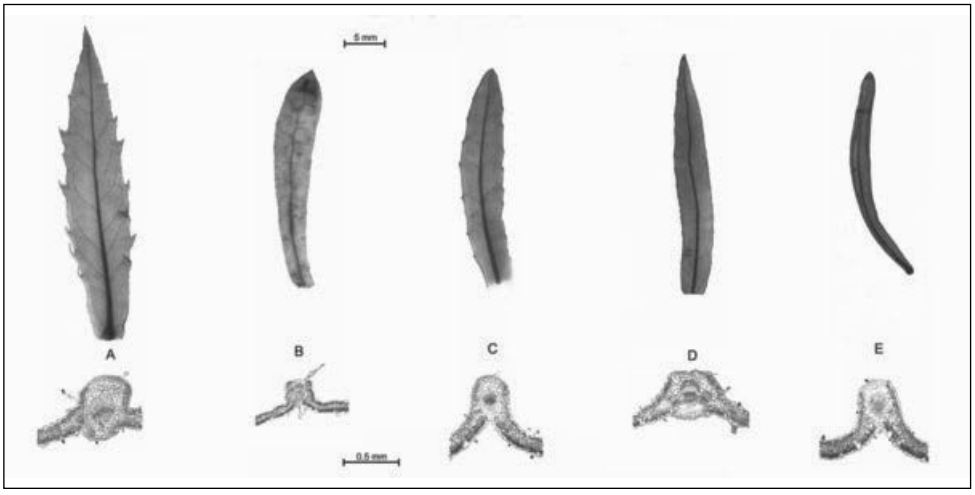

Fig. 2. Morpho-anatomical leaf variation in *Dittrichia* species: **a**, *D. viscosa*; **b**, *D. graveolens*; c, *D. maritima*; **d**, *D. orientalis*; **e**, *D. revoluta*.

surface structures arises from the variability of cell shape and size, and stratified microstructures of the cell surfaces, such as cuticle layers, epicuticular wax crystals, cuticular folds, hairs and glands. Almost unlimited different combinations of these cellular sculptures generate the high structural and functional diversity which characterizes land plants' surfaces (Barthlott & al. 2017).

The surface of plants is the critical interface for the interaction with the environment and fulfils many different functions (mechanical protection, attachment and particle adhesion, water loss reduction, light reflection, temperature control, air retention, wettability) related to ecological adaptation and/or reproductive strategies (Barthlott 1981, 1984; Bargel & al. 2004; Koch & Barthlott 2006). Furthermore, with exception of fossil pollens and spores, cuticles represent the most widespread unaltered fossil plant remains and are known from the Devonian to the recent times (Taylor & al. 1989). Thus, comparative studies of micromorphological features can provide significant insights into physiological properties and ecological responses of plants to environmental constraints and aid in systematic and evolutionary questions in extant and fossil plants, as highlighted by a vast existing literature (e.g. Stace 1965;, Jones 1986; Kessler & al. 2007; Ickert-Bond & Rydin 2010; Albert & Sharma 2013; Anil Kumar & Murugan 2015; Arabi & al. 2017; Ickert-Bond & al. 2018;, Sur & al. 2018; Scoppola & Magrini 2019).

Examples of micromorphological surveys include comparative analyses of seed coat sculpturing. The seed coat is the direct interface between embryo and external environment, acting as main modulator in the plant life cycle with key functions of regulation and protection. The taxonomic value of the macro-and micro-morphological characters of seeds and outer coats has been clearly demonstrated, being very conservative and stable features. In the genus *Brassica* L. sect. *Brassica*, with 20 taxa ten of which strictly endemic to Sicily, seed morphology and seed coat patterns provided useful information for discriminating among close taxa, especially those at subspecific level. Eighteen exomorphic parameters, including shape, size, color, surface texture, from 50 seeds for each accession (see Salmeri & al. 2011) were investigated. SEM analysis was carried out on 5-10 seeds from each sample, considering the arrangement and shape of epidermal cells, the architecture of anticlinal and periclinal cell walls (primary sculptures), their fine cuticular ornamentations (secondary sculptures).

Seed coat sculptures at low magnification (20×) showed a reticulate pattern, but higher magnifications (200-600×) revealed more complex networks, identifiable in 4 basic subtypes, i.e. simple reticulate, micro-reticulate, reticulate-foveate, reticulate-rugose, on account of their finer structuring. Significant differences were found in the overall cell shape (± polygonal or irregular) and the size, height and alignment of the meshes, which may be lax to compact, sharply angled to smooth, regular or irregular. Great variation among different taxa and populations was also observed in the anticlinal and periclinal cell walls and cell lumen, which can be straight to ± undulate, depressed, concave, flat or convex, smooth to ± markedly wrinkled, foveolate and / or papillose. Clear differences and very characteristic architectures were highlighted in most of the investigated species (Salmeri & al. 2011), with very close taxa such as the *B. villosa* and *B. rupestris* complexes (Fig. 3) characterized by well-defined sets of microsculptures valuable as discriminant features at specific and subspecific levels. In addition, seed coat microsculpturing can be helpful in the management of seed accessions in the seedbanks' collections.

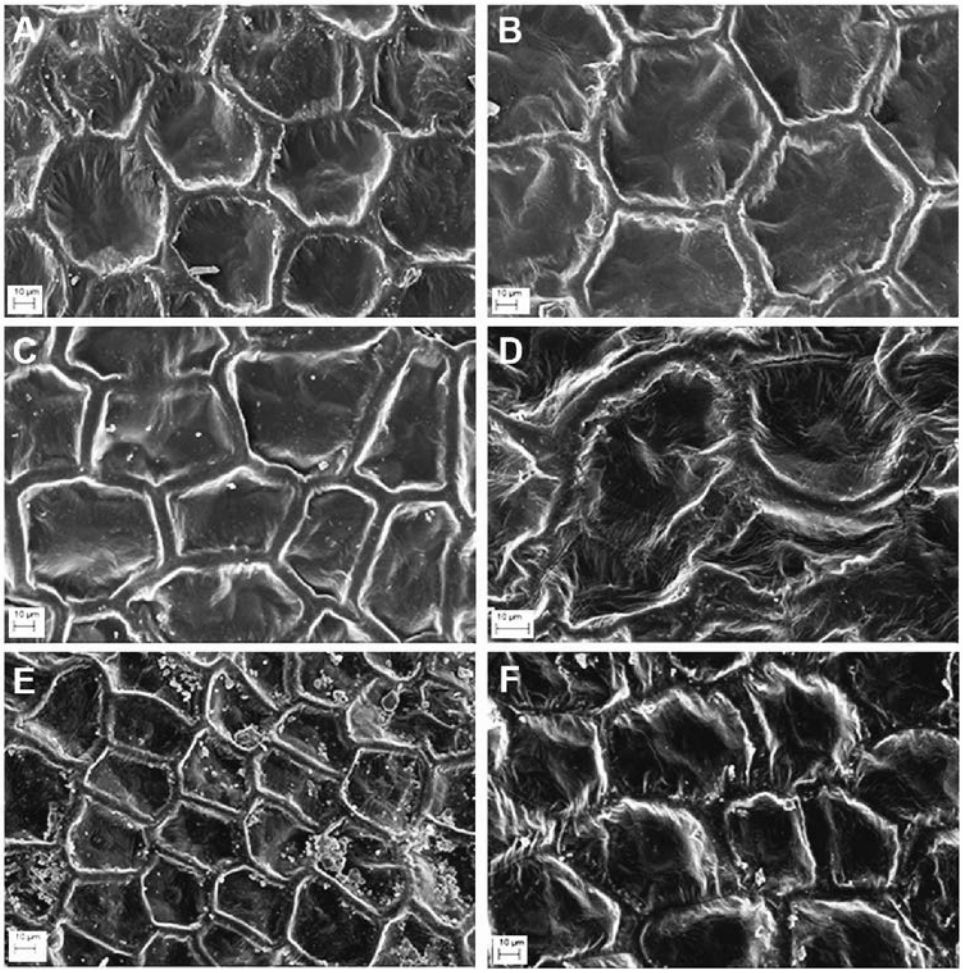

Fig. 3. Seed coat microsculpturing of some Sicilian taxa of *Brassica* sect. *Brassica* (500×): **a**, *B. villosa* subsp. *villosa*; **b**, *B. villosa* subsp. *bivoniana*; **c,** *B. villosa* subsp. *drepanensis*; **d**, *B. villosa* subsp. *tineoi*; **e**, *B. rupestris* subsp. *rupestris*; **f**, *B. rupestris* subsp. *hispida*

Comparative micromorphology of seed sculpturing is also relevant to assess dispersal strategies and infer potential and effective pathways of the gene flow within and among populations. A representative case is given by *Daucus carota* mericarps. Wild carrot fruits are oblong-ovoid schizocarps, 2-4 mm long, at maturity splitting into 2 small mericarps, with an outer convex surface provided with 5 primary short ciliate ridges and 4 secondary higher ridges with hooked prickles. Due to their shape, wild carrot diaspores can be transported by both wind and animals. Experiments revealed that especially spines decrease the fall of seeds in the air and that seeds were found to be scattered by wind over a distance on average not longer than 3 m (Lacey 1981; Manzano & Malo 2006), but they can reach

longer distances (rarely up 100 m away from the original area), especially when whole dry umbels, curled into a ball, are detached from the host plants (tumbleweed effect). Actually, the post-fertilization contraction of the umbels due to hygroscopic responses to air humidity can also act as regulator of seed dispersal (through retention or release) in local microsites (Lacey 1980; Heywood 1983). However, the commonest means of dispersal is by attachment to animal fur or human clothing. Manzano & Malo (2006) demonstrated that wild carrot mericarps attached to sheep fleece could be transported 400 km by transhumant flocks, with about 7% remaining adherent for up to 6 months so that seed dispersal continues for a greater length of time. Thus, barbs and prickles undoubtedly favour seed dispersal and the number and the distance over which propagules are scattered can differ depending on scattering medium. A preliminary survey on different Sicilian populations of wild carrot from internal and coastal sites revealed a great variation in the mass of pericarps (both size and shape), but especially in the length and density of prickles along the ridges and in the number (1 to 4-5) and orientation ($\leq/\approx/\geq$ 90°) of apical hooks (Fig. 4). These data, which still need further samplings, could provide useful information to interpret and predict gene flow among populations and hybridization patterns.

Ecomorphology and adaptive traits

Our current knowledge of biodiversity, adaptive strategies and ecosystem function is largely founded on descriptive comparative morphology, which enables our understanding of plant phenotypic plasticity and the related biological and ecological roles. The observation, description and documentation of variation in plasticity among and within populations allow to find adaptive explanations for specific forms and comprehend the ecological and evolutionary consequences of their diversity. Lots of publications discussed on the interaction of plant structures with environment or analysed how the environment conditions might modify the phenotypic expression of intrinsic features (Rotondi & al. 2003; Royer & al. 2005; Rozendaal 2003; Xu & al. 2009; Nicotra & al. 2010; Blonder & Enquist 2014; Angiolini & al. 2015; Yang & al. 2015; Mannino & Graziano 2016; Pilote & Donovan 2016; Saatkamp & al. 2018).

One investigated species showing significant levels of leaf morphometric variation across its populations was *Pancratium maritimum* L. In this widespread Mediterranean coastal species different combinations of some key leaf traits, such as thickness of epidermis components, blade tissues, features and size of stomata apparatus, and leaf venation, provide special morphological patterns which ensure populations to have a plastic ecophysiological adaptation to the local microclimatic conditions (Perrone & al. 2015). In fact, despite a main and rather stable morpho-anatomical structure, leaves in *P. maritimum* populations revealed significant differences in the size, number, and/or type of several micro-morphological and histological features. Single and multiple linear regression analyses, conducted to clarify the statistical effects of different climate parameters (mean annual precipitation, mean annual temperature, mean maximum temperature, aridity index) on leaf traits of *P. maritimum*, indicated the existence of significant correlations, positive or negative, between leaf plasticity and local climate. Thus, intra-specific variability in functional leaf traits of *P. maritimum*, especially those related to stress tolerance (thickness of cuticle and epidermis cells, cell size of palisade and spongy tissue, size and density of stomata, size and number of intercostal areas with aerenchyma or mucilage)

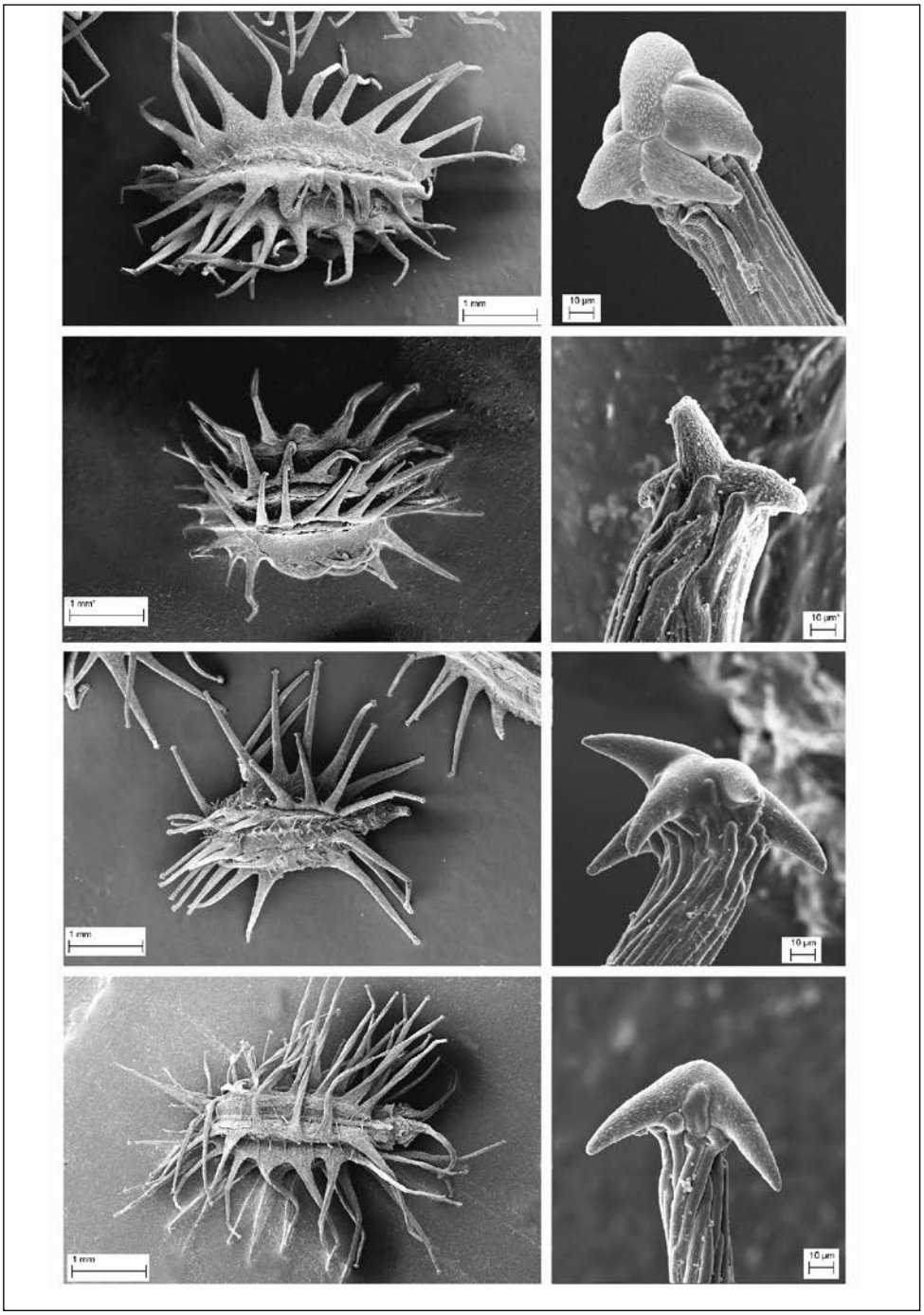

Fig. 4. Micromorphological fruit variation in some *Daucus carota* populations from Sicily: **a**, single mericarp with barbs and hooked hairs (18×); **b**, detail of the tip of hooked hairs (600×).

turned out to be a key aspect for long-term conservation, ensuring local adaptation to microsite conditions (insolation, drought, sandblasting) and increasing plant aptitude to adjust to climate changes.

Investigations on 6 different annual species of the genus *Salsola* sect. *Kali* Dumort, that is *S. kali* L., *S. tragus* L., *S. australis* R. Br., *S. squarrosa* Steven ex Moq. (formerly *S. tragus* L. subsp. *pontica* (Pall.) Rikle), and 2 taxa recently described within the genus *Kali* due to the previous taxonomic elevation of the homonymous section (i.e. *K. basalticum* Brullo & al. and *K. dodecanesicum* Brullo & al.), also revealed significant morpho-anatomical variations between species from maritime and inland areas, which clearly represent adaptations to survive under specific environmental conditions. Distinctive common features in this taxonomic group are stems rigid, not articulate, cortex green to greenish-red, with longitudinal chlorenchymatous striae, leaves linear-cylindrical, broadened at base, provided with apical spine, bracts similar to the leaves, but smaller, membranaceous perianth of 5 free segments, fruiting perianth usually winged, provided with unequal rudimentary abaxial appendices, membranaceous fruits, above flattened. Nevertheless, the species diverge in different combinations of morphological and anatomical characters, mainly related to the habit, stem, leaves and bracts, indumentum and salt glands, shape and size of flowers and fruits, which were proved to be directly involved in adaptive ecophysiological responses and/or reproductive strategies. As far as leaves are concerned, the investigated species show a cylindrical to semicylindrical outline, rather reflecting the same indumentum as the stem, no hypodermis, two concentric layers of chlorenchyma, typical of C4 Kranz anatomy, water storage tissue with mucilage in the central part, one central vascular bundle and 2 minor ones in the peripheral part, and 2 longitudinal collenchymatic ridges which interrupt palisade and Kranz cells. As showed in Fig. 5, main differences regarded the general leaf size (leaf area, leaf thickness), leaf indumentum, cell size and tissue thickness (complex cuticle-epidermis, palisade tissue, and collenchyma), which represent useful discriminant features among species (Fig. 6). Results from single and multiple linear regressions carried out on some leaf morphological characters (Fig. 7) suggested that climatic parameters have significant influence on leaf variability; in particular warming has positive relationships with leaf area and leaf thickness, while increased precipitations seem to affect negatively the leaf size. This can be explained by the fundamental role of a well-developed water storage tissue in drought conditions.

Discussion

The examples provided have demonstrated that plant morphology can and should contribute in a dynamic way to both basic and applied research, since today it has new and more opportunities than ever before, especially due to new techniques for structural research, such as SEM, confocal microscopy, microcomputer tomography and the modern morphometric analyses, which are opening possibilities for a better understanding of organisms' evolution and a further integration of comparative morphological studies and other biological disciplines.

Contrary to common belief, plant morphology is not a conservative finished science, but, like other sciences, it is open to constant innovations involving concepts and methods.

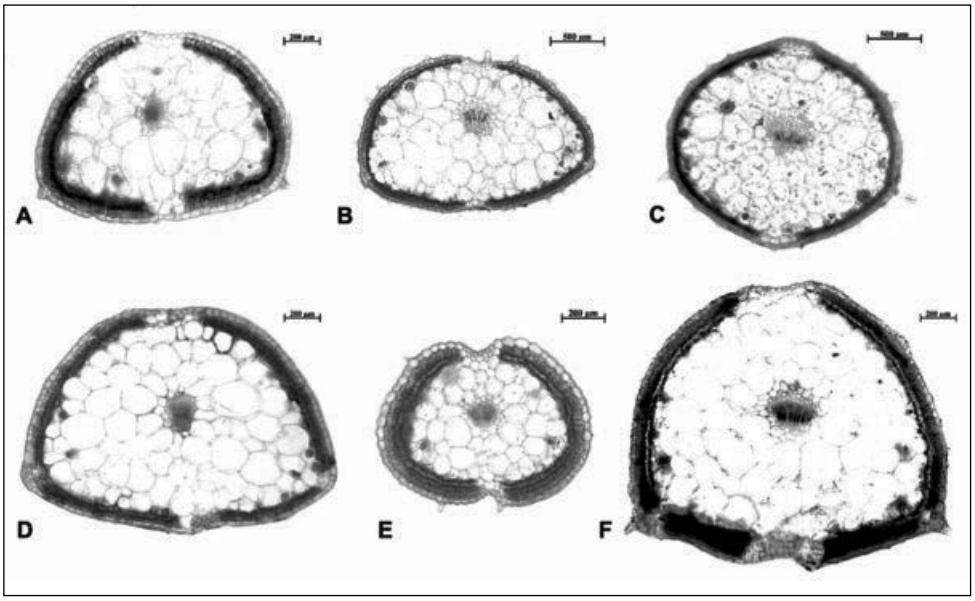

Fig. 5. Morpho-anatomical leaf variation in some annual *Salsola* species: **a**, *S. kali*; **b**, *S. tragus*; **c**, *S. dodecanesica* (*Kali dodecanesicum*); **d**, *S. squarrosa*; **e**, *S. basaltica* (*Kali basalticum*); **f**, *S. australis.*

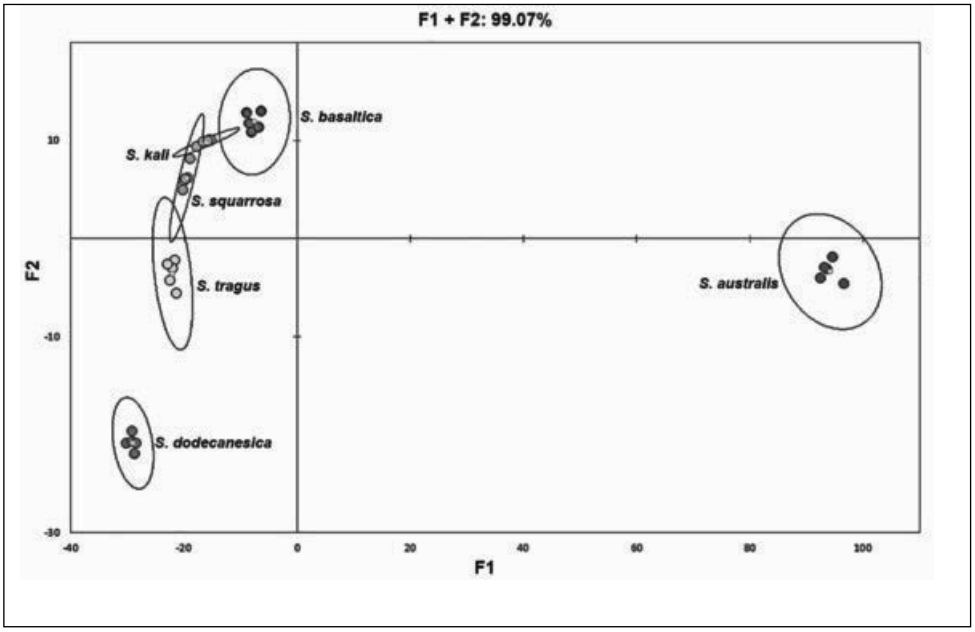

Fig. 6. Similarities among *Salsola* species resulting from discriminant factorial analysis among based on leaf parameters.

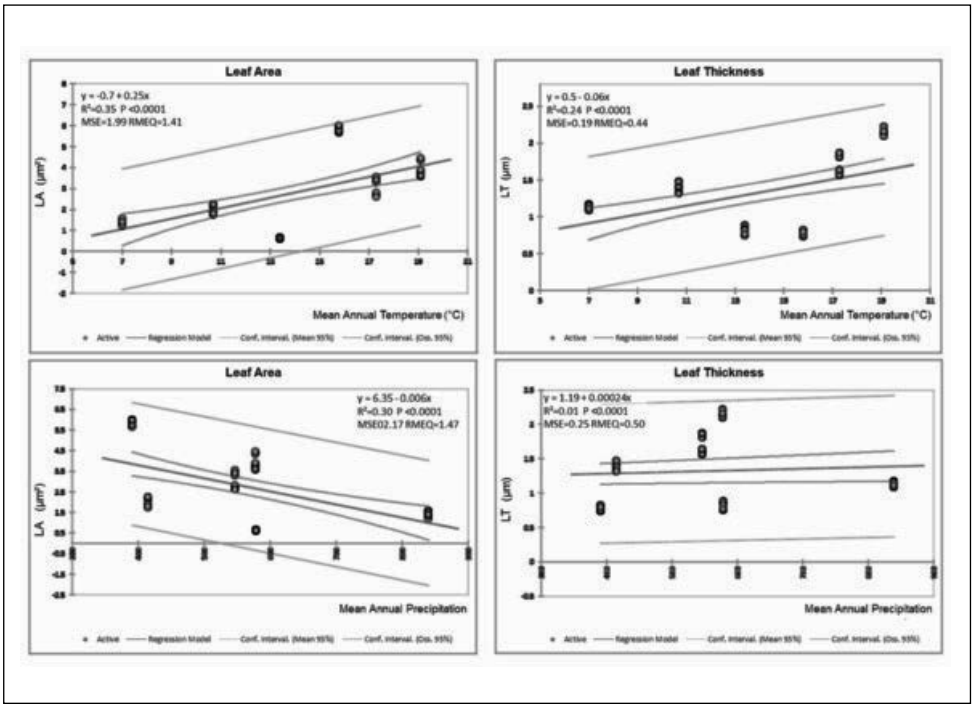

Fig. 7. Scatter plots and linear regressions indicating correlations between some leaf features and climatic parameters in *Salsola* species.

In fact, plant morphology has changed over time and improved its analytical approaches embracing new technologies and tools, without neglecting traditional methods.

Notwithstanding, morphological studies and their main related discipline, the classical alpha taxonomy, are still underestimated and somehow marginalized. Results from morphology-based research are often poorly cited in top-ranking journals, maybe because many authors tend to cite papers that support taxonomic treatments or reviews with molecular data, rather than papers only based on classical taxonomy (Pysek & al. 2013). The low number of specialists for particular limited groups further contribute to reducing the chances of plant morphology papers becoming highly cited (Krell 2002). This has progressively led to a worldwide decline in morphologists and taxonomists in general, which could have a broad impact on plant biology research and biodiversity conservation. Unfortunately, the simple assumption that biodiversity studies cannot advance without morphologists is unlikely to produce an adequate increase in public funding and broad support (Pearson & al. 2011). Thus, now it should be time to re-evaluate the contribution of plant morphology and contemporary plant morphologists at the level of modern botanical and evolutionary research in order to avoid the loss of a wide baseline expertise and favour the involvement of students and young researchers, especially through modern approaches and high technical tools, in this field of botany sciences.

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Article

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From North to South: a voyage through plant biodiversity in the Italian mountains*

Abstract

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Italy is among the European countries richest in biodiversity, mainly due to a wide variety of geomorphological and climatic conditions. Its very high plant diversity is also the result of its geographical position, acting as a bridge between Central Europe and the Mediterranean Sea and producing the coexistence of different biogeographic elements with a high contingent of endemic plant species, which amounts to more than 15%. As in many other mountains of the world, both the Alps and the Apennines host an extremely rich flora which forms peculiar plant communities characterizing several priority habitats listed in Directive 92/43/EEC and forming wonderful mountain landscapes, where nature and human work merge increasing biodiversity. The voyage from North to South through the Italian mountain plant diversity is an opportunity to observe the responses of plant species and habitats to climate and land use changes that very rapidly are transforming our mountain landscapes, not only at lower altitudes, as expected, but, surprisingly, along the whole altitudinal gradient. Recent results on changes of the summit flora (GLORIA and Summit flora projects), as well as on abandonment of the traditional grazing and forestry activities in some mountain areas and on the spread of nonnative species, produced significant changes at levels of species, habitat and landscape. On one hand the responses of plants to these changes confirm that they are a threat for plant biodiversity, but on the other hand that plants have a surprisingly rapid capacity to face abrupt climatic or land use variations.

Key words: alpine habitats, plant biodiversity, conservation.

Plant biodiversity in Italy is one of the highest of all European countries, both in terms of species richness and of vegetation communities. Knowledge in these two fields is continuously increasing and researches at the national level collect data that are the results of a great number of studies at the regional and local levels.

Examples of these studies at the national level are well represented by the recent checklist of the vascular flora native to Italy (Bartolucci & al. 2018) and the checklist of the alien flora (Galasso & al. 2018), both updating the Checklist of the Italian vascular

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flora (Conti & al. 2005), the newly published Flora d'Italia (Pignatti & al. 2017-19), a paper on endemic plant species (Peruzzi & al. 2015) and a recent book "Flora in Italia" (Blasi & Biondi 2017). To all these, and also other publications, a high number of botanists, representing all Italian regions, collaborated in order to provide an accurate analysis of the updated knowledge.

Following the Checklist of the vascular flora native to Italy 8195 taxa occur in our country and 1708 are endemic of one or more regions, highlighting an extraordinary floristic richness, due to the Italian geological, geomorphological, climatic, latitudinal and elevational variability. Some of these features have been studied and their variability has been represented on maps as in the phytoclimatic map of Italy (Blasi & Michetti 2007) or in the Map of the Important Plant Areas (IPAs) in Italy (Blasi & al. 2010) and culminated with the Map of vegetational series (Blasi, 2010) that documented the presence of more than 240 vegetational series occurring from North to South and from West to East.

All data agree on the fact that a very high floristic and vegetational richness is located in mountain areas, both in the Alps and in the Apennines, and that rare and endemic species occur mainly in the alpine belt, confirming that mountains are characterized by concentrated geological as well as climatic variability in restricted ranges, following the elevation gradient. For the above mentioned reasons mountains experienced typical climatic histories, in particular during the glacial periods, with an ice cover in some areas, in particular in the Alps and at the higher summits of the Apennines and with the absence of ice cover in other areas. These differences at least partly explain the actual distribution of rare species and, consequently, of rare plant communities.

For these reasons it is highly recommended to explore the Italian mountains, in order to have a look on some of the most interesting areas, taking time for a voyage from the Western Alps to the Southern Apennines, to observe the rich, magnificent flora but also to have a glance on the ecological observations that resulted from the analyses that have been carried out in those places.

Our voyage begins in the Western Alps, and specifically in the Aosta Valley, where the highest summits occur: Monte Bianco (Fig. 1), Monte Rosa, Gran Paradiso, Cervino, all above 4000 m a.s.l..

In this region the summit alpine flora was studied in the past by several botanists, and in particular by Lino Vaccari, who published his "Flora cacuminale della Valle d'Aosta" (Vaccari 1901) a reliable and complete study on a high number of summits. The re-survey of the same summits studied by Vaccari was carried out by Elena Barni and co-workers in the framework of the Summit Flora Project,

coordinated by Sonia Wipf, working at Davos (Steinbauer & al. 2018).

In this work the botanists used a dataset of repeated plant surveys from 302 mountain summits across Europe, spanning 145 years of observation, to assess the temporal trajectory of mountain biodiversity changes as a globally coherent imprint of the Anthropocene, and in particular of climate variability. As on the Aosta Valley summits, the authors found a continent-wide acceleration in the rate of increase in plant species richness, with five times as much species enrichment between 2007 and 2016 as fifty years ago, between 1957 and 1966. This acceleration is strikingly synchronized with accelerated global warming and is not linked to alternative global change drivers. The accelerating increases in species richness on mountain summits across this broad spatial extent demonstrate that accelera-

Fig. 1. The Mont Blanc seen from Mont La Saxe (Photo Siniscalco).

tion in climate-induced biotic change is occurring even in remote places on Earth, with potentially far-ranging consequences not only for biodiversity, but also for ecosystem functioning and services.

On ecosystem functioning several studies have been carried out in the Aosta Valley to highlight how subalpine grasslands react to the summer heat wave that more and more often occurs during August: the dry grassland vegetation, and in particular dry grasses as *Nardus stricta* L. stop their activity and only after the first rainfalls, at the end of August, become green and active, while forbs as *Trifolium alpinum* L., *Arnica montana* L., and *Geum montanum* L. continue to carry out their activity even under very dry summer conditions (Cremonese & al. 2017). Climate change causes very clear responses on the grassland community, even when snow melt begins earlier than usual in April or May because without snow the soil and plant temperature can be several degrees under zero, causing damages to the plants.

One important point in these studies is that we have to concentrate on the community but also on the single species response, trying to analyse the plant traits, in order to understand their ecological needs and to predict which will be their response to interannual climate variability.

Moving on to South, we stop in Valle di Susa, one of the most interesting endo-alpine valleys because, if compared to the others, as Engadin and Valais in Switzerland, Val

Venosta and Valle d'Aosta, its flora is enriched by several Mediterranean species, in particular in the driest areas, located in the middle of the Valley. This confirms what was observed by Aeschimann (Aeschimann & al. 2004): the richest flora in alpine areas is located where, in addition to the typical species, ranging on the chain, Mediterranean species arrived in the past, in warmer periods.

In the Xero-thermic oasis South facing slopes (SIC IT1110030 "Oasi xerotermiche della Valle di Susa - Orrido di Chianocco e Foresto) the very dry climate, less than 600 mm yearly precipitations, with strong winds blowing everyday, justify the occurrence of two priority habitats included in the Directive 92/43/EEC (Habitat Directive), "Dry semi natural grasslands (*Festuco-Brometalia*)-important orchid rich sites" (6210*), and the "Steppic, sub-pannonian grasslands" (6240*). Both grasslands need grazing to be maintained, so that a Life Project was carried out (Life 12NAT/IT/000818 Xero-Grazing) for the Semi-natural dry-grassland conservation and restoration in Valle Susa through grazing management.

The most wide spread Mediterranean species are *Euphorbia sulcata* Lens, *Asterolinon linum-stellatum* (L.) Duby, *Linum strictum* L., *Ononis reclinata* L., *Leuzea conifera* (L.) DC., *Crupina vulgaris* Cass., *Linum suffruticosum* L., *Lavandula angustifolia* Mill., *Helianthemum apenninum* (L.) Mill., *Ononis minutissima* L., *Coronilla minima* L., and *Juniperus oxycedrus* L. (Fig. 2). Moreover, 29 orchid species occur in the dry grasslands, e.g. *Ophris fuciflora* (F.W. Schmidt) Moench, *Ophrys tetraloniae* W. P. Teschner, *Neotinea tridentata* (Scop.) R. M. Bateman, Pridgeon & M.W. Chase, *Neotinea ustulata* (L.) R.M. Bateman, Pridgeon & M.W. Chase, *Anacamptis pyramidalis* (L.) Rich., *Epipactis atrorubens* (Hoffm.) Besser, *Cephalanthera longifolia* (L.) Fritsch and *Cephalanthera rubra* (L.) Rich. In the steppic, sub-pannonian grasslands, which have been observed for the first time in such Western alpine areas and were not expected to occur outside their typical range, *Stipa pennata* L. and *Cleistogenes serotina* (L.) Keng characterize physionomically the grasslands, and are to be referred to the alliance Stipo-Poion carniolicae.

This area is very interesting to visit, being an island of Mediterranean vegetation in the middle of the Alps.

Moving South in our voyage, we arrive in the Maritime Alps, one of most interesting hot spots of the Italian flora, where the influence of the Mediterranean sea avoided the presence of a continuous ice cover during the Quaternary glaciations and the formation of neoendemic species, after this period. This uncommon conditions in an area of the Alps, justifies the actual occurrence of several rare, endemic species (Fig. 3) as for example *Saxifraga florulenta* Moretti, *Saxifraga lingulata* Bellardi, *Primula allionii* Loisel, *Viola valderia* All., *Campanula alpestris* All. and *Fritillaria tubaeformis* subsp. *moggridgei* (Boiss. & Reuter ex Planch.) Rix. All these species are typical of rocky slopes or of alpine grasslands, where competition with other species is low. The researches on these and of other species allow confirming the historical, geomorphological and genetic reasons of their survival or their more recent formation in this very interesting area of the Alps.

Moving once more to South, and having an interest on the previously cited topics linked to the history of the Italian mountain flora, we arrive to the Gran Sasso and Majella groups.

The secondary prairies with dominant *Bromus erectus* Huds. are quite similar to the ones that we saw in Susa Valley, with *Helianthemum apenninum* (L.) Mill., *Medicago minima* (L.) L., *Neotinea ustulata* (L.) R.M. Bateman, Pridgeon & M.W. Chase, *Anacamptis*

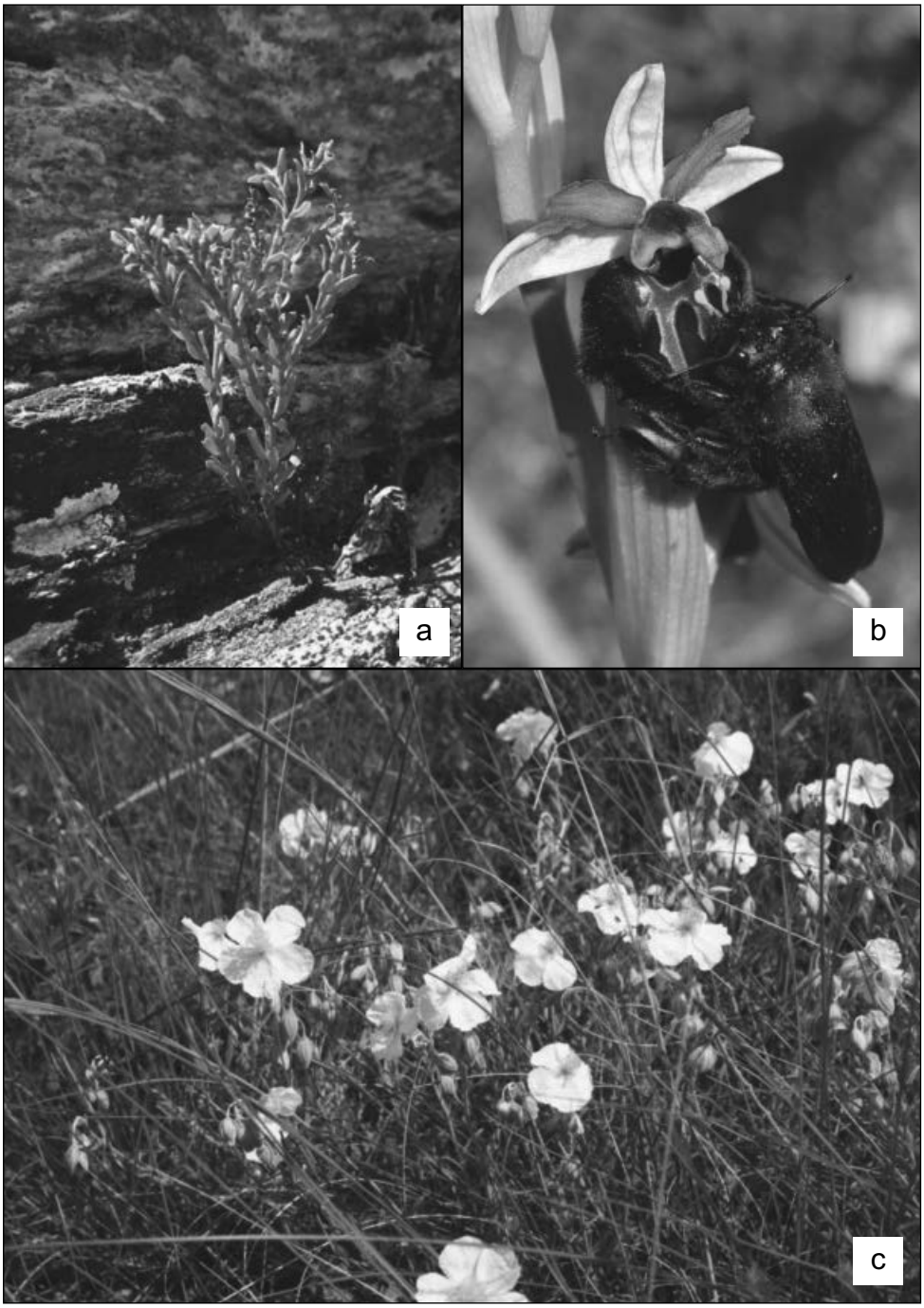

Fig. 2. a) *Euphorbia sulcata* in Susa Valley dry grasslands (Photo Gorlier); b) *Ophys fuciflora* in the Susa Valley dry grasslands (Photo Davi); c) *Helianthemum apenninum* (Photo Gorlier).

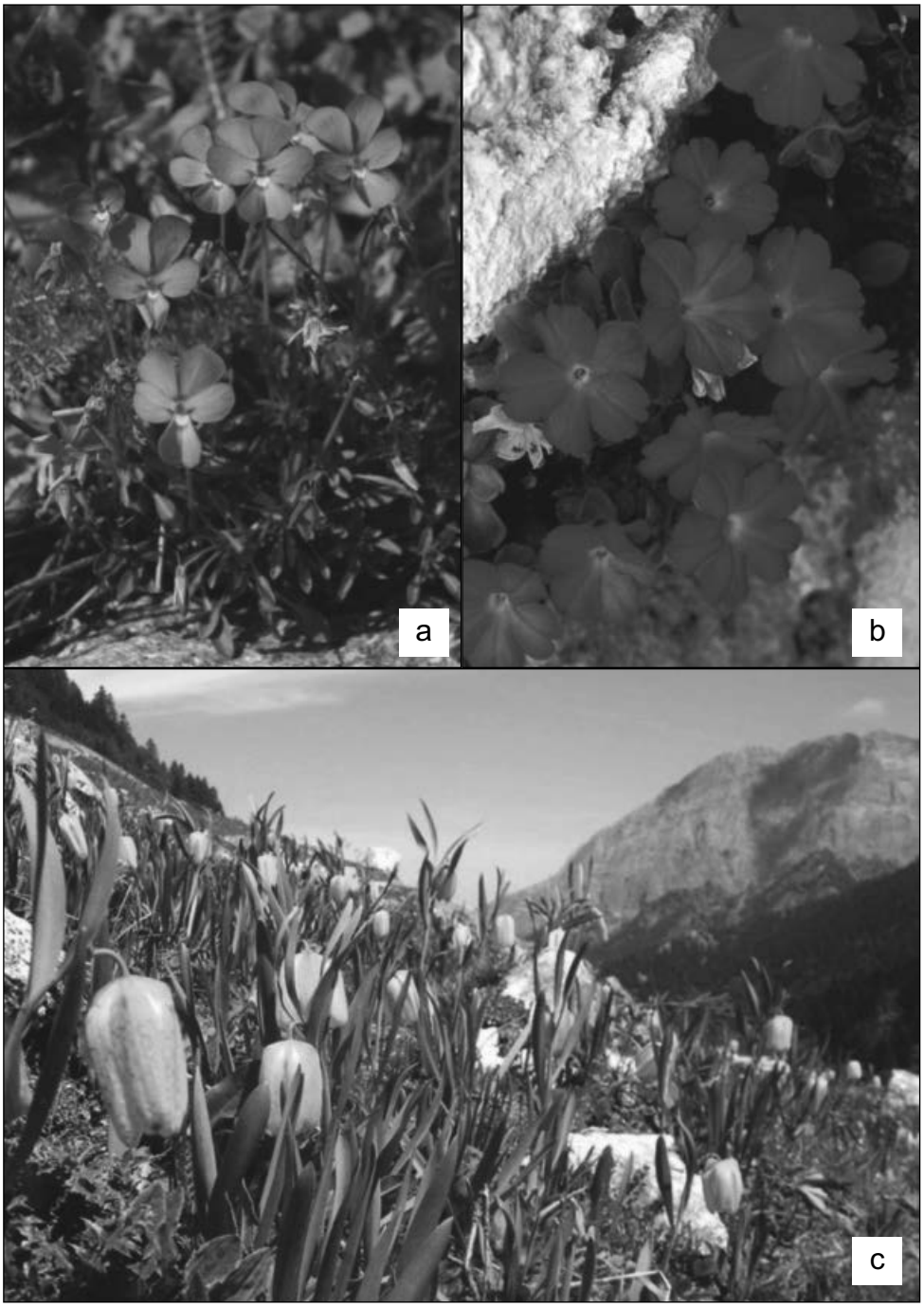

Fig. 3. a) *Viola valderia* and b*) Primula allionii* in the Maritime Alps (Photo Masante); *Fritillaria tubaeformis* subsp. *moggridgei* in the Maritime Alps (Photo Mucciarelli)

Fig. 4. a) *Ranunculus magellensis* in Majella (Photo Nicolella); b) Fig. 13. *Viola magellensis* in Majella (Photo Nicolella).

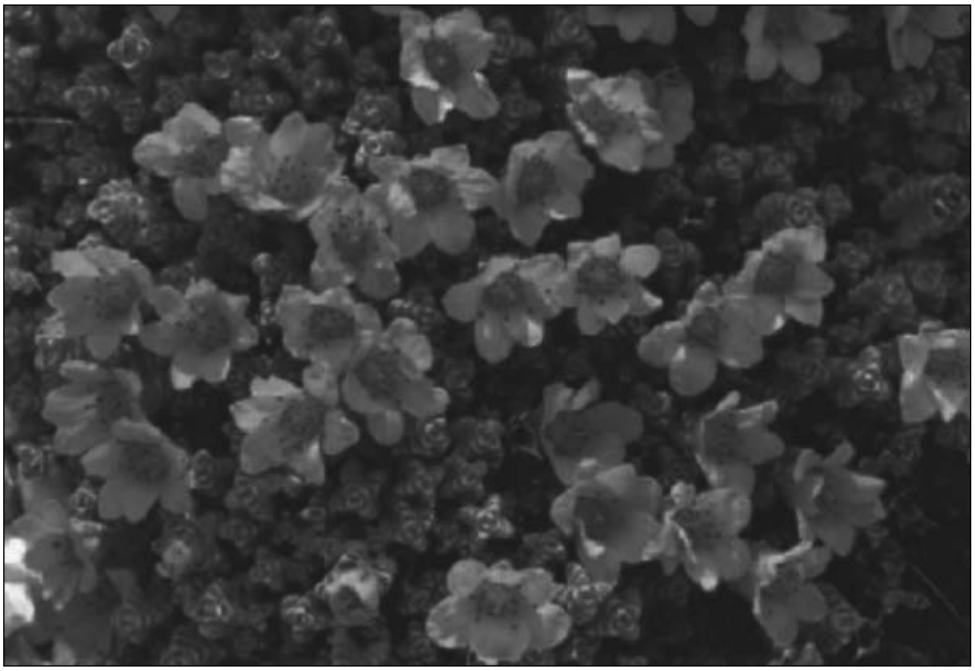

Fig. 5. *Saxifraga oppositifolia* subsp. *speciosa* (Photo Pirone).

pyramidalis (L.) Rich. and other species that very interestingly vicariate the Susa Valley ones, as *Eryngium amethystinum* L. at the place on *Eryngium campestre* L., *Trinia dalechampii* (Ten.) Janch. at the place of *Trinia glauca* (L.) Dumort.

When we arrive to the highest elevations, above the timberline, it is possible to see a magnificent flora, characterized by an extraordinary high biodiversity described by Pirone (2006) and Pirone and Frattaroli (2011). Migration, mainly from North and East, and speciation processes lead to a very rich floristic diversity that includes, beyond endemic species, a relevant number of relict species, similarly to what we have seen in the Maritime Alps. These species have wonderful flowers that evolved to respond to the scarce number and variety of pollinator insects at those elevations. An analytical analysis of these species is reported in Blasi and Biondi (2017), with notes on distribution, habitats and conservation, as for *Ranunculus magellensis* Ten., *Viola magellensis* Porta & Rigo, *Saxifraga oppositifolia* subsp. *speciosa* (Dörfl. & Hayek) Engl. & Irmsch. and *Androsace mathildae* Levier (Figs. 4-5). This last species is listed in the Habitat Directive and in Abruzzo Red List, as many other species of the high elevation habitats of Gran Sasso and Majella.

This virtual voyage that brought us to visit examples of some of the most interesting Italian mountain systems is an invitation to go and visit them on the field. These excursions confirms the extraordinary high biodiversity of the mountain flora also in terms of biogeographical as well as evolutionary value. On these bases the need is confirmed to monitor

the presence and conservation status of these species and vegetation communities in the future, in order to control the effects of climate and eventually land use changes on their survival and functionality.

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Short note

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Ridha El Mokni & Duilio Iamonico

Bassia scoparia (*Amaranthaceae* s. l.) and *Sesuvium portulacastrum* (*Aizoaceae*), two new naturalized aliens to the Tunisian flora

Abstract

El Mokni, R. & Iamonico, D.: *Bassia scoparia* (*Amaranthaceae* s. l.) and *Sesuvium portulacastrum* (*Aizocaeae*), two new naturalized aliens to the Tunisian flora — Fl. Medit. 29: 191-196. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Bassia scoparia var. *trichophylla* (*Amaranthaceae* s. l.), and *Sesuvium portulacastrum* (*Aizoaceae*) are recorded for the first time for the non-native flora of Tunisia. A morphological description, distribution and habitat, as well as taxonomic notes, are provided for each taxon.

Key words: alien species, distribution, taxonomy, xenophytes.

Introduction

Careful field surveys during the last two decades have allowed not only the improvement of the floristic and biogeographical knowledge within the Tunisian territory (see e.g., Domina & al. 2017, Domina & El Mokni 2019; El Mokni & al. 2015, 2019) but also the discover of many species new for the non-native Tunisian flora (see e.g., Iamonico & El Mokni 2016; El Mokni & Domina 2017a; El Mokni & Domina 2017b; Iamonico & El Mokni 2017a, 2017b, 2018; El Mokni & Domina 2018; El Mokni & Verloove 2019; Iamonico & El Mokni 2019a, 2019b). In this paper, we provide further results related to the families *Aizoaceae* Martinov and *Amaranthaceae* Juss. s. l. (incl. *Chenopodiaceae* Vent. *sensu* APGIV 2016). Within the first family, the genus *Sesuvium* L. is here reported for the first time to the Tunisian flora with the species *S. portulacastrum* (L.) L., whereas for the second family (*Amaranthaceae*), the genus *Bassia* All. is currently represented by only two species in Tunisia [*B. muricata* (L.) Asch., and *B. indica* (Wight) A.J.Scott] (Sukhorukov & al. 2018). We here report for the first time *Bassia scoparia* var. *trichophylla* (Voss) L.H.Bailey as an alien naturalised to the Tunisian flora.

Materials and Methods

The work is based on extensive field surveys, analysis of literature, and examination of the specimens kept in the personal collection of one of the authors (R. El Mokni) which is

deposited in the Herbarium of the Faculty of Pharmacy of Monastir and of the personal Herbarium of El Mokni. Taxa are presented alphabetically.

Records

1. ***Bassia scoparia*** var. ***trichophylla*** (Voss) L.H.Bailey, Man. Cult. Pl. 250 (1924) ≡ *Kochia trichophylla* Voss., Deutsche Gartenrat 140: 391. 1905. ≡ *Bassia scoparia* f. *trichophylla* (Voss) S.L.Welsh., Utah Fl., ed. 3.: 113. 2003.
   Lectotype: not designated.
   – *Bassia scoparia* f. *trichophylla* (Voss) S.L.Welsh., Utah Fl., ed. 5: 126. 2015, isonym (Art. 6 Note 2 of ICN; Turland & al. 2018).

Bassia scoparia, a highly variable species, and several forms, varieties, and subspecies have been described during the time. Among them, var. *trichophylla* (Voss) L.H. Bailey appears to be widespread in Tunisia. It is characterized by plants appearing ovoid or obovoid ("cypresslike") with crowded branches, and leaves narrower (Fig. 1).

Distribution and habitat

The genus *Bassia* comprises about 20 species, distributed in the western Mediterranean to eastern Asia (see e.g., Kadereit & Freitag 2011).

Bassia scoparia is a highly variable species from the morphological point of view, is native to central and eastern Europe and western Asia (see e.g., Friesen & al. 2009), and it is well naturalized as alien in Tunisia. The horticultural var. *trichophylla* is largely cultivated as ornamental throughout many regions in Tunisia and was found many times naturalized on roadsides, abandoned lands and dumps. The plant has been recorded as a weed in South and Western Australia and is declared as noxious weed in some states (Dodd & Randall 2002).

In the Mediterranean area, *B. scoparia* is reported as native to the Iberian Peninsula and as naturalized alien in France, Italy, Sicily and Malta (see e.g., Uotila 2011; Tison & De Foucault 2014; Galasso & al. 2018). The Tunisian checklist (Le Floch'h & al. 2010), the *Synonymic Index for North African Flora* (Dobignard & Chatelain 2011) and even the African Plant Database (APD 2019**)** did not list or precise this infraspecific taxon neither for Tunisia nor for North Africa but Uotila (2011) reported *B. scoparia* only as naturalized alien in Morocco and as casual alien in Algeria and Lybia. In the APD (2019) *B. scoparia* is accepted as a weed only for the Canary Islands, Morocco, Algeria and Lybia. Botanical surveys carried out since 2004, led to distinguish this taxon in some sporadic populations well naturalized, on ruderal coastal abandoned lands (from Monastir in the centre-east to Tabarka in the northern-west) of Tunisia.

Taxonomic notes

The delimitation of infraspecific taxa within *Bassia scoparia* remains highly critical and their taxonomic value is subject to questioning (see e.g. Gudžinskas & Sukhorukov 2004).

Examined specimens (new records)

TUNISIA: Bizerta (Route Corniche, NE Tunisia), 37°17'23" N, 09°52'08" E, 4 m a.s.l., 18 December 2016, *R. El Mokni s.n.*; Nabeul (Hammamet-South, CE Tunisia), 36°24'12"

N, 10°35’31” E, 8 m a.s.l., 11 December 2016, *R. El Mokni s.n.* (Herb. Univ. Monastir); Monastir (Route Khénis, CE Tunisia), 35°43’31” N, 10°49’04” E, 5 m a.s.l., 22 November 2016, *R. El Mokni s.n.* (Herb. Univ. Monastir); Jendouba (Tabarka, NW Tunisia), 36°56’57” N, 08°46’34” E, 3 m a.s.l., 05 Mai 2017, *R. El Mokni s.n.* (Herb. Univ. Monastir); Mahdia (Touristic zone, CE Tunisia), 35°30’07” N, 11°04’02” E, 01 m a.s.l., 05 February 2019, *R. El Mokni s.n.* (Herb. Univ. Monastir).

2. *Sesuvium portulacastrum* (L.) L., Syst. Nat., ed. 10(2): 1058. 1759. ≡ *Portulaca portulacastrum* L., Sp. Pl. 1: 446. 1753.
Lectotype (designated by Wijnands 1983: 175): [icon] *Portulaca corassavica Angusto longo lucidoque folio* in Hermannus (1968 : 212, image available at https://bibdigital.rjb.csic.es/viewer/13586/?offset=#page=310&viewer=picture&o=bookmark&n=0&q=).

Distribution and habitat

The genus *Sesuvium* comprises perennial and annual herbs with glabrous or papillate stems and sub-opposite, sessile or short-petiolate leaves, axillary pink or mauve flowers with five perianth lobes, five to numerous stamens and circumscissile capsule containing several or numerous black seeds completely or partially (in perennial species) covered with a translucent or whitish one-layered aril, and with a crustaceous

Fig.1. *Bassia scoparia* var. *trichophylla* in the Tabarka region (NW Tunisia) (photo R. El Mokni).

Fig. 2. *Sesuvium portulacastrum* in the Monastir region (CE Tunisia) (Photo R. El Mokni).

seed coat (Sukhorukov & al. 2017).

Sesuvium portulacastrum is an herbaceous (Fig. 2), perennial, psammophytic, dicotyledonous and facultative halophyte belonging to *Aizoaceae* (Lonard & Judd 1997; Lokhande & al. 2009). This species grows on the coastlines and it is widely distributed as a pioneer strand species on tropical and sub-tropical shores (Lonard & Judd 1997). It grows naturally in the sub-tropical, Mediterranean, coastal and warmer areas around the world (Ramani & al. 2006). Concerning North Africa, this species is currently recorded only in Morocco (see e.g., Greuter & al. 1984; Fennane & al. 1999; Dobignard & Chatelain 2011; APD 2019). It is here reported for the first time from Tunisia where it occurs as one of the floristic corteges of halophytic plants within salty water of non-permanent rivers in Medenine region (south-eastern of Tunisia) and ruderal coastal plants of Monastir's region (centre-eastern of Tunisia).

Examined specimens (new records)

TUNISIA: Medenine (Route Amra, south-eastern of Tunisia), 33°21'33.23" N, 10°30'06.30" E, 77-78 m a.s.l., 18 December 2016, *R. El Mokni s.n.* (Herb. Univ. Monastir); Monastir (Route Sousse, centre-eastern of Tunisia), 35°46'12.54" N, 10°47'14.79" E, 3 m a.s.l., 19 June 2017, *R. El Mokni s.n.* (Herb. Univ. Monastir).

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First report of *Pleurotus fuscosquamulosus* (*Pleurotaceae*, *Basidiomycota*) in Italy naturally occurring on new tropical hosts

Abstract

Torta, L., Bella, P., Conigliaro, G., Mirabile, G., Laudicina, V. A., Giambra, S., Venturella, G., Gargano, M. L.: First report of *Pleurotus fuscosquamulosus* (*Pleurotaceae*, *Basidiomycota*) in Italy naturally occurring on new tropical hosts. — Fl. Medit. 29: 197-206. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Basidiomata of a mushroom macroscopically recognised as *Pleurotus cystidosus* sensu latu were collected on the trunks of three tropical ornamental trees such as *Broussonetia papyrifera*, *Yucca elephantipes*, and *Paulownia tomentosa* in the Parco d'Orleans, an urban park in the town of Palermo (Sicily, Italy). All the plants showed cavities and white rot symptoms at the base of the trunks. Macro- and microscopic observations on both collected basidiomata and isolated colonies, as well as molecular analysis, allowed identifying the collected basidiomata as *Pleurotus fuscosquamulosus* and its anamorph as *Antromycopsis fuscosquamulosus*. These species belong to the subgenus *Coremiopleurotus* that includes different species producing synnematoid fructifications. The positive reaction of the colonies to the syringaldazine and the artificial inoculation tests on wood blocks of *B. papyrifera* confirmed its white wood rotting activity. To our knowledge, this is the first report of occurring of *P. fuscosquamulosus* in Italy on three new tropical plant hosts.

Key words: *Coremiopleurotus*, *Antromycopsis fuscosquamulosa*, white wood rot, syringaldazine.

Introduction

The genus *Pleurotus* (*Basidiomycota*) includes several genetically well-delimited saprotrophs species (Zervakis & al. 2014), which play fundamental ecological roles in the agro-forest ecosystems, and are widely cultivated for food, biotechnological and pharmaceutical purposes (Venturella & al. 2016). In the last decades fungi belonging to the genus *Pleurotus* able to synthesize lingo-cellulolytic enzymes, have been investigated for their potential applications in various industrial processes (Elisashvili & al. 2006; Sekan & al. 2019). Nevertheless, many of them are rather aggressive on woodwork and standing trees, degrading lignin and causing white rot (Bari & al. 2017). Among the ca. 30 species and subspecific taxa of *Pleurotus*, 14 were reported in Europe as frequent, occasional or, very rare species colonizing living trees, decaying wood and vegetal debris (Venturella & al. 2015).

The subgenus *Coremiopleurotus* Hilber, is characterized by the production of the coremioid anamorphic stage *Antromycopsis* Pat. & Trabut. *Pleurotus cystidiosus* O. K. Miller is the type species of the subgenus, described for the first time by Miller (1969) on red maple (*Acer rubrum* L.) in Indiana (USA). Its anamorphic state was assigned to *Antromycopsis macrocarpa* (Ellis & Everh.) Stalpers, Seifert & Samson (=*Antromycopsis broussonetiae* Pat. & Trab.; Patouillard, 1897; Stalpers & al., 199; Zervakis, 1998). Other tropical taxa are also ascribed to this subgenus , which are very similar in the morphological features, such as *Pleurotus abalonus* Y. H. Han, K.M. Chen & S. Cheng, *P. australis* Cooke & Massee, *P. cystidiosus* var. *formosensis* Moncalvo, *P. fuscosquamulosus* Reid & Eicker, *P. purpureoolivaceus* Segedin, P. K. Buchanan & J. P. Wilkie, and *P. smithii* Guzman. All these species are distributed in restricted geographical areas (Zervakis & al. 2004). Each species of *Pleurotus* correspond to a specific *Antromycopsis* anamorph.

In autumn 2016, in the town of Palermo (Sicily, Southern Italy) some gregarious gilled basidiomata, showing the main typical morphological features of *P. cystidiosus* sensu lato (Miller 1969; Guzman & al. 1991; Zervakis & al. 1992), were observed on the trunks of three tropical ornamental trees.

Materials and methods

Sampling of basidiomata

In autumn 2016, during a survey conducted in Parco d'Orleans some gregarious gilled basidiomata (5 - 15 cm wide) were observed on forty years old ornamental trees of *Broussonetia papyrifera* (L.) Vent. (paper mulberry tree), *Yucca elephantipes* Regel ex Trel. and, *Paulownia tomentosa* Steud. The basidiomata emerged from cavities localized at the base and along the trunks where white rot symptoms were also observed. No evident symptoms were observed on the aerial parts of the host plants. Samples of basidiomata from each tree species were collected and subjected to laboratory analysis according to Zervakis & al. (1992, 2004).

Morphological features

The macroscopic features of basidiomata were detected by naked eye and under a Leica stereoscope microscope. Handmade sections of gills (ca. 20 µm thin) and portions of the mycelial culture were mounted in lactophenole clear (phenol 20 g, glycerol 40 ml, lactic acid 20 ml, deionized water 20 ml) or with methyl blue (0.05%) and the slides observed at light microscope (Axioskop; Zeiss, Oberkochen, Germany) coupled to an AxioCam MRc5 (Zeiss, Oberkochen, Germany) digital camera. Images were captured using the software AxioVision 4.6 (Zeiss, Oberkochen, Germany). The collected data were shape, color and structure of the basidiomata; morphology of basidia, basidiospores and anamorphic structures; macroscopic and, microscopic colony characteristics. The size of the microscopic structures was calculated based on the arithmetic mean of the values of 50 measurements.

Fungal culture

In order to obtain the dikaryotic fungal colonies, three young basidiomata grown on each host were collected and transferred to the laboratory for isolation. In particular, after

a fast passage over the flame, each basidioma was aseptically dissected and 20 little fragments (ca. 2 mm in size) of the inner pseudo-parenchyma were removed and plated on Potato Dextrose Agar (PDA, Oxoid), in Petri dishes (5 fragments for dish). The Petri dishes were incubated at 25 °C, until the grown of the fungal colonies. Mono-hyphal pure cultures were obtained by transferring on PDA hyphal tips by means of a thin sterile needle under stereomicroscope.

Molecular and phylogenetic analysis

Genomic DNA was isolated from 1-week-old fugal isolate grown on PDA at 25°C in the dark using a standard CTAB based-protocol (Torta & al., 2015). The internal transcribed space (ITS) regions, ITS1 and ITS2, and the 5.8S gene of the ribosomal DNA (rDNA) operon, were amplified using primers ITS1F (fungal specific: 5'- CTTGGTCATTTAGAGGAAGTAA-3') (Gardes & Bruns, 1993) and ITS4 (universal: 5'-TCCTCCGCTTATTGATATGC-3') (White & al., 1990). The amplification reaction was performed in a total reaction volume of 25 µl containing 50–100 ng of DNA template, 2 mM of $MgCl_2$, 0.2 mM of each dNTP, 0.2 µM of each primer, 0.2 U of Dream Taq (Fermentas, Milan, Italy) and 1× Dream Taq buffer (Fermentas). The amplification reaction was carried out as follows: initial denaturation at 95°C for 2 min, 35 cycles of denaturation at 95°C for 30 s, annealing at 55°C for 20 s, extension at 72 °C for 1 min and final extension at 72°C for 5 min.

Results PCR products were analyzed by electrophoresis on 1,5% agarose gel, stained with SYBR Safe DNA gel stain and visualized under a UV transilluminator.

PCR products were sequenced in both directions using the same primers as for PCR reactions. Sequences were edited using the Sequencer software (Version 4.7, Gene Codes Corporation, Ann Arbor, MI) and compared to GenBank sequences through BLASTn searches. ITS sequence was aligned using MEGA6 software (Tamura & al., 2011) and manual adjustments of alignments were made when necessary. Phylogenetic analysis was performed according to Giambra & al. (2016) by using sequences of 26 *Pleurotus* species belonging to subgenus *Coremiopleurotus* retrieved from GenBank. *P. australis* (Accession number AY450342) was used as outgroup.

Enzymatic activity

The production of laccase by the fungal colonies was detected by the use of syringaldazine, an organic azo-compound oxidizable by this enzyme, turning from light yellow to dark red colour (Wilkołazka & al. 2002; Floch & al. 2007; Wang & al. 2010). Drops of a mixture containing 0.2 mM syringaldazine (dissolved in 60% ethanol) in 40 mM citrate-buffer (pH 5.6) were applied on the surface of the fungal colony. As negative control, colonies of *Fusarium oxysporum* von Schlechtendal no producing laccase were employed.

Inoculation on sapwood block

To confirm the ligninolytic activity, blocks of healthy wood of *B. papyrifera* (10 × 5 × 5 cm) were singularly put into 10 moist chambers (autoclavable plastic airtight containers of 200 ml volume, with adsorbing paper), added with 5 ml of distilled sterile water and sterilized in autolclave. Plugs (5 mm^2) of 10 days old colonies grown on PDA were inoculated singularly on 7 wood samples, and the remaining 3 samples were inoculated with plugs of sterile PDA. All the containers were incubated at 25°C in the dark for six months (Burcham & al. 2015).

Results

The young basidiomata (5-7 cm broad) caps varied from gray to light brown - pale yellow and were sub-globose or plane-convex, enrolled, with cuticle brown squamulose. The ripe basidomata (10-15 cm) were more or less depressed in the center, with involute margins. Both in young and ripe basidiomata the lamellae were thin, wide, and decurrent with some anastomoses at the top of the stipe, alternate by long lamellulae. In the older samples colour of the lamellae varied from cream white, to pale yellow or light orange ocher. Stipes, typically eccentric, in the ripe basidiomata, were fibrous, thick, fistulose, with a tapered base and light brown in colour (Fig. 1 a-c). Basidia and spores were hyaline and smooth walled, the first measuring 40 × 7 µm, and the latter, oblong, about 15 × 5 µm (Fig.1 d-e). The basidiospores, in mass, were whitish. All the macroscopic features permitted to identify the collected fungi as *P. cystidiosus* s. l.

On PDA fungal colonies were white, with radial growth and irregular margins, covering the plates in 50 days and constituted by hyaline and clamped mycelium (Fig. 2 a-b). From young colonies, whitish and cespitose coremia were produced, bringing globose black heads made up by mass of dark, cylindrical conidia originate from disarticulation of the ending hyphae, measuring 15.5 × 6 µm. All typical structures of the anamorph belonging to the genus *Antromycopsis* (Fig. 2 c-d).

Since morphological characters alone were ineffective for identifying the species within the *P. cystidiosus* s. l., a phylogenetic analysis based on ITS sequences was performed. ITS sequence of the isolate obtained in this study deposited in GenBank with the accession number MH271184 was compared with sequences of 26 isolated of the species within subgenus *Coremiopleurotus* retrieved from GenBank database. The phylogenetic tree constructed by the Maximum Likelihood method showed that the isolate obtained in this study grouped with other *P. fuscosquamulosus* isolates with high bootstrap value and was differentiated from other clusters including other *Pleutotus* species (Fig. 3).

In our study, laccase production was assayed by a colorimetric qualitative test based on syringaldazine oxidation. The quick turning from light yellow to dark red of few drops of the solution containing the syringaldazine applied on the surface of fungal colony indicated the production of the ligninolytic enzyme by the fungal isolate. (Fig. 4a). No activity was observed in the colony of the negative control (Fig. 4b). Moreover, in the inoculated wood blocks of *B. papyrifera* coarse mass of white mycelium associated with the above described coremia was observed. All the infected blocks showed symptoms of white rot (Fig. 4c), confirming the degradation activity on the lignin compounds.

Discussions and Conclusion

A basidiomycete belonging to the subgenus *Coremiopleurotus*, growing on ornamental tropical trees in a urban Park of Palermo, was identified at species level. To our knowledge, this is the first report of *P. fuscosquamulosus* in Italy. The last report of *Coremiopleurotus* in Greece (Zervakis & al. 1992) and the presence of new fructifications of *P. cystidiosus* s. l. on different trees of *B. papyrifera* observed in the same location in summer 2019, seems indicate the spreading of the subgenus also in the Mediterranean area.

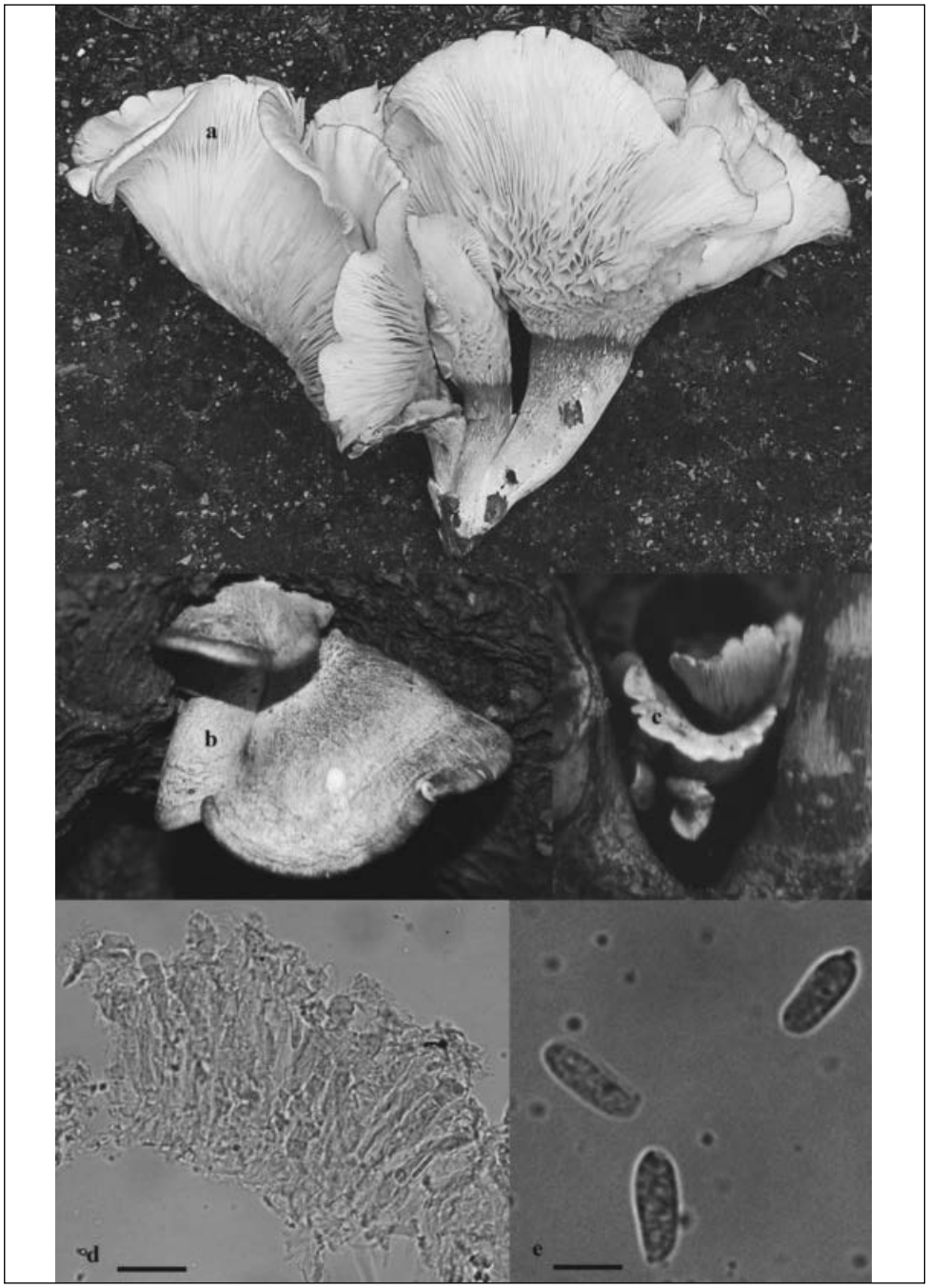

Fig. 1. Main morfological features of the collected basidioma. Fruiting bodies grown on *P. tomentosa* (a), *Y. elephantipes* (b) and *B. papyrifera* (c); microscopic features of basidia (d) and basidiospores (e). Bars: d = 20 µm; e = 10 µm.

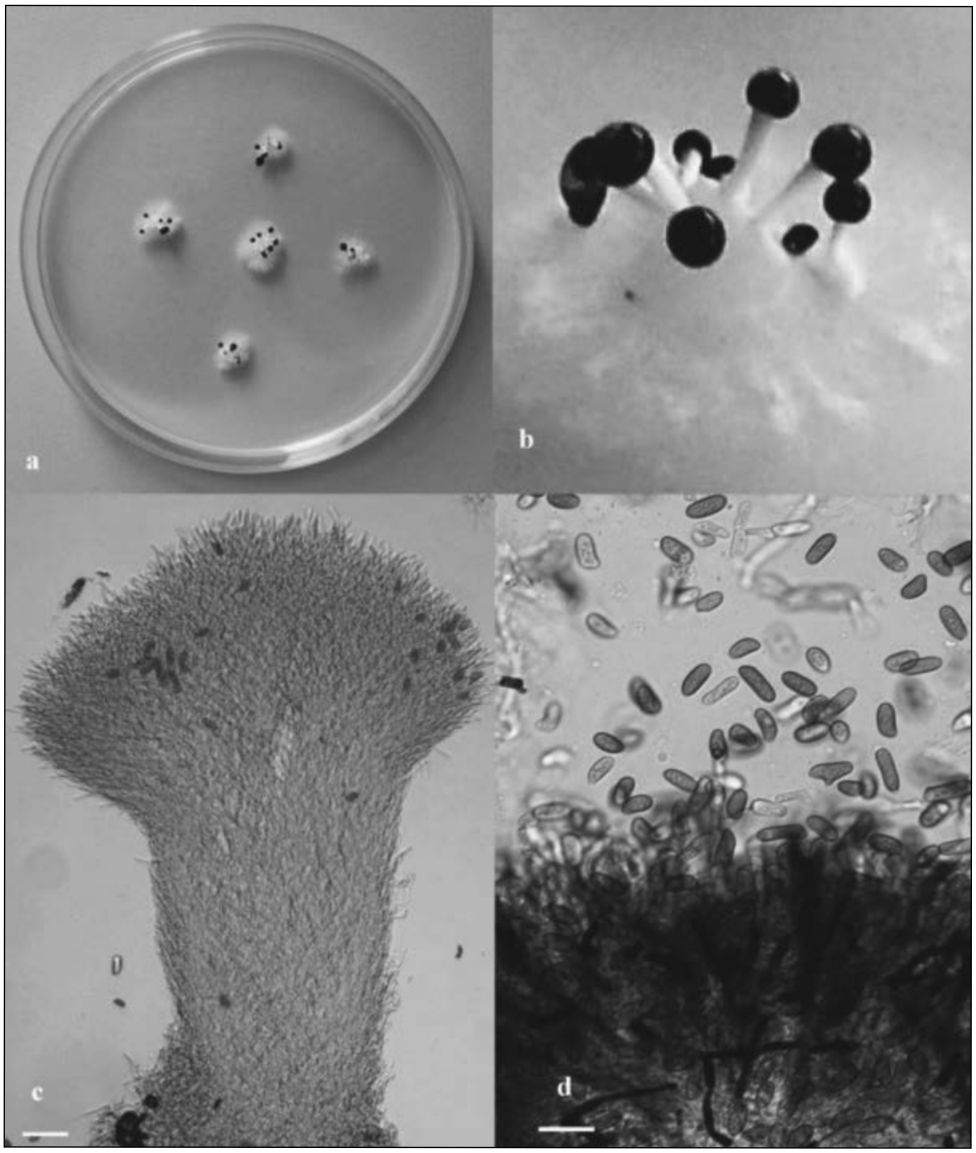

Fig. 2. Young colonies of *Pleurotus fuscosquamulosus* and coremia of *Antromicopsis fuscosquamulosus* on PDA (a, b) and at microscope (c, bar = 10 µm; d, bar = 20 µm).

P. fuscosquamulosus and the anamorphic stage *Antromycopsis fuscosquamulosus* D. A. Reid & Eicker were described for the first time by Reid and Eicker (1998) on trunk and roots of *Populus alba* L. and *Combretum molle* R. Br. ex G. Don in South Africa. More recently, Zervakis & al. (2004), reported the only presence of *P. fuscosquamulosus* also in Europe (Greece), distinguishing this species from other belonging to the

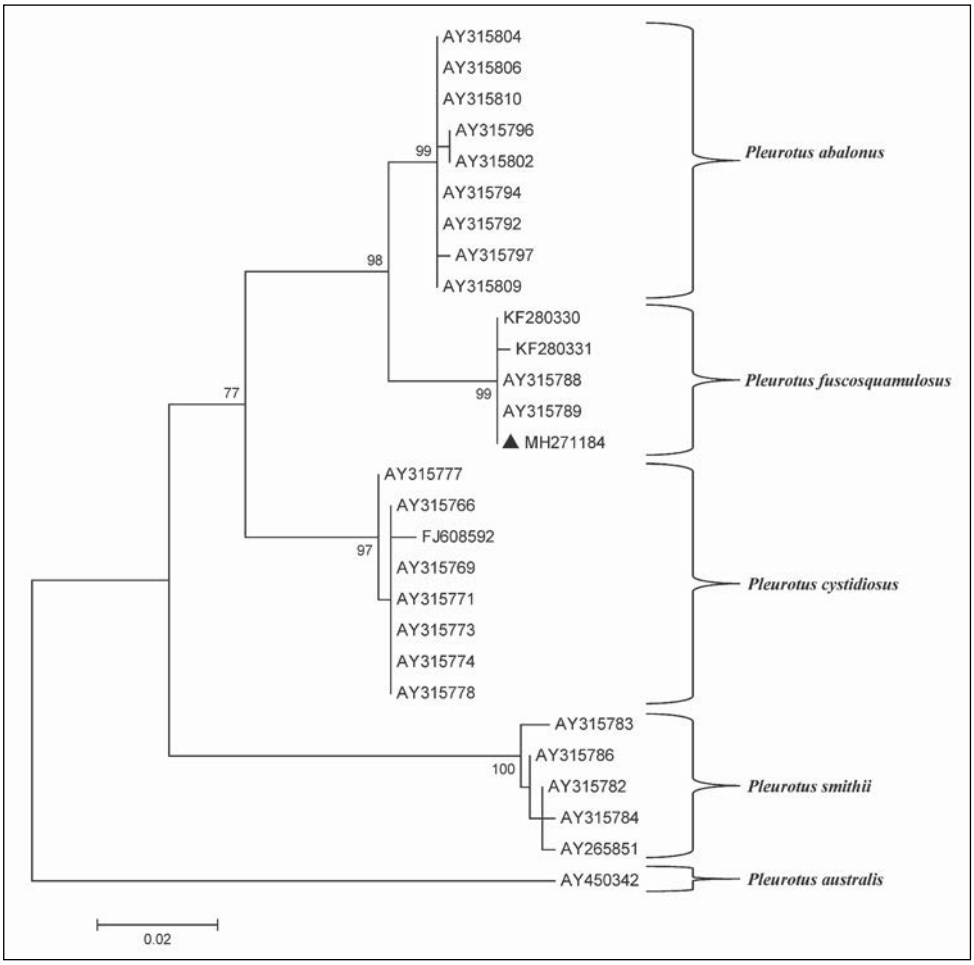

Fig. 3. Maximum likelihood trees based on ITS sequences of 26 isolates of *Pleurotus* sp. within the subgenus *Coremiopleurotus*. *P. australis* was used as outgroup. Black triangle indicates the isolate obtained during this study, whereas the other ITS sequences were retrieved from GenBank and are shown with their Accession Number. Bootstrap tests were performed with 1,000 replications.

Coremiopleurotus, based on phylogenetic species concept, molecular, mating and geographical evidence (Denchev & al. 2013). In the last few years, Menolli & al. (2014) reported the presence of the species in Brazil. Very few information is available on the host plant from which this *Coremiopleurotus* species was found. In this survey, *P. fuscosquamulosus* was reported on new tropical hosts *B. papyrifera*, *Y. elephantipes* and *P. tomentosa.* as potential white rot etiological agent.

The close resemblance between the members of the *Pleurotus* subgenus *Coremiopleurotus*, mainly in the macroscopic features, can lead to misidentification, if based only on the traditional taxonomy.

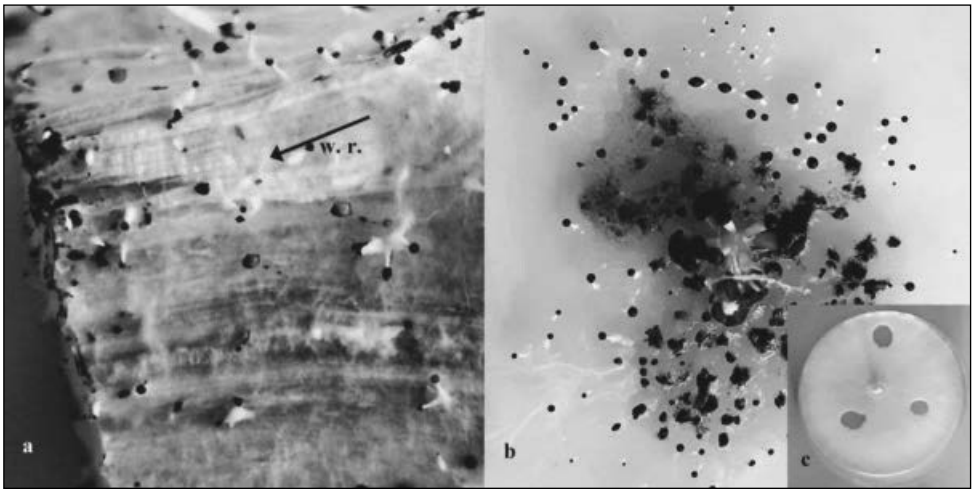

Fig. 4. Mycelial growth of *P. fuscosquamulosus* artificially inoculated on wood samples of *B. papyrifera* and symptoms of white rot (w. r.) (a). Reaction to syringaldazine in colonies of *P. fuscosquamulosus* (b) and of a not producing laccase fungus (c)

Like most *Pleurotus* species, the fungi belonging to the *Coremiopleurotus* produce good edible basidiomata. For food purposes and for the high commercial value *P. cystidiosus* and related species are widely cultivated in the United States, Canada, Europe, Asia, and, Australia (Zervakis & al. 2004). This species showed also antihypertensive hypotheses (Ching & al. 2011, 2013) and hypoglycaemic activities (Jayasuriya & al. 2015). However, data on the cultivation and medical properties of *P. fuscosquamulosus* are not known. Investigations on edible fungi producing lignin-cellulosolytic enzymes and other active secondary metabolites can be useful to the development of new strategies in food, pharmaceutical and industrial fields.

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Article

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W. Nemer, K. Rebbas & F. Krouchi

Découverte de *Cypripedium calceolus* (*Orchidaceae*) au Djurdjura (Algérie), nouvelle pour l'Afrique du Nord

Abstract

Nemer, W., Rebbas, K. & Krouchi, F.: Découverte de *Cypripedium calceolus* (*Orchidaceae*) au Djurdjura (Algérie), nouvelle pour l'Afrique du Nord. — Fl. Medit. 29: 207-214. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Discovery of *Cypripedium calceolus* (*Orchidaceae*) in Djurdjura (Algeria), news for North Africa. — *Cypripedium calceolus* L. (*Orchidaceae*) native to Eurasia is considered one of the flagship plant species of nature conservation. Because of its wide range, this species could be considered a taxon of minor concern, near threatened, vulnerable, endangered or critically endangered.
This orchid was discovered in the Djurdjura, in north-central Algeria. Ecological notes are also provided. This is the first report of this species in North Africa. This discovery will enrich the Algerian orchid in general and that of Kabylia in particular.

Key words: flora, floristics, Orchid, North Africa.

Introduction

La famille des orchidées est l'une des familles de plantes à graines les plus riches en espèces. Il comprend environ 880 genres comprenant plus de 25 000 espèces dans le monde (Cribb & al. 2003).

Le nom *Cypripedium* vient du grec Κυπρισ (Kupris), la déesse de Chypre, ou Aphrodite (Vénus), la déesse de l'amour des Romains et des Grecs, et du grec πεδιον (pedion), plante du pied. Un des noms français du *Cypripedium* est Sabot de Vénus, d'où le nom du genre. L'espèce "*calceolus*" signifie en latin "petite chaussure".

Bien que mondialement reconnue comme un des principaux point-chauds de biodiversité végétale (Médail & Quezel 1997 ; Médail & Myers 2004), la région méditerranéenne demeure méconnue, en particulier sur ses rives sud et est. L'ensemble de montagnes du littoral algéro-tunisien dénommé «Kabylies-Numidie-Kroumirie» ne fait pas exception avec une forte diversité végétale et un fort taux d'endémisme (Véla & Benhouhou 2007).

Malgré l'engouement énorme qu'ont suscité les orchidées européennes (cf. Delforge 2016), l'orchidoflore des rives sud de la Méditerranée demeure méconnue.

En Afrique du Nord, *Cypripedium calceolus* n'est pas citée dans l'index de Dobignard & Chatelain (2010-2013). De même aucune flore ou catalogue d'Algérie (Battandier 1888-1890; Battandier & Trabut 1905; Maire 1952-1987; Quézel & Santa 1962-1963) ni même de Tunisie (Bonnet & Barrate 1896; Pottier-Alapetite 1979-1981; Le Floc'h & al. 2010; Guittonneau 2011), ni aussi au Maroc (Fennane & Ibn Tattou 1998, 2005; Fennane & al. 2007) ne signale ce taxon. Cette orchidée n'est pas signalée dans l'Euro+Med PlantBase (Euro+Med 2006).

Le présent travail vient faire le point sur la découverte de *Cypripedium calceolus* dans le Djurdjura et son écologie.

Contexte de la découverte

Cypripedium calceolus L.
Nomencl. ref.: Sp. Pl.: 951. 1753 (WCSP 2019)

Avec une quarantaine d'individus groupés en cinq touffes, occupant une aire inférieur à 25 m², le Sabot de Vénus a été découvert en plein floraison le 7 juin 2019 par l'un de nous (W.N.) dans une station située au sud-ouest du col de Tirourda sur une pente orientée ouest à une altitude de 1485m (36.4516°N et 04.3294°E) à l'occasion d'un inventaire floristique des rochers et des falaises du Djurdjura (Kabylie, Algérie). Le 29 juin, une visite des deux auteurs (W.N. & K.R.) a permis d'observer des individus de cette orchidée en extrême fin de floraison, complètement fanées (Fig. 1 et 2). Un spécimen d'herbier collecté a été déposé dans l'herbier officiel de l'école nationale supérieure agronomique (ENSA) d'Alger le 18.9.2019. Le nouveau site algérien est représenté par une zone semi-ombragée située entre des ilots de reboisement de *Cedrus atlantica*. Le tableau 1 présente une liste de la flore inventoriée dans la station de *C. calceolus.*

Dans d'autres pays, cette orchidée occupe des habitats forestiers en Eurasie qui ont un couvert forestier semi-ouvert permettant à un peu de lumière de l'atteindre. Sa présence en Mordovie (Russie centrale) est confinée dans divers types d'habitats : forêts de feuillus, forêts de conifères et forêts mixtes. Plus un habitat de prairies pures (Baumann & Künkele 1982 ; Bournérias & Prat 2005 ; Antonelli & al. 2009 ; García & al. 2010 ; Khapugin & al. 2017).

En Europe, *C. calceolus* est une calcicole bien connue qui pousse dans des sols alcalins ou rarement neutres. En Europe centrale, il pousse dans les forêts de conifères et en bordure de forêts mixtes de feuillus, souvent dans des sites plats ou en pente, jusqu'à une altitude de 2000 m ou plus. À certains endroits, il forme de grandes colonies avec des plantes individuelles portant de nombreuses pousses (KS - RBG 2019).

Répartition géographique

Le nouveau site de *Cypripedium calceolus* en Algérie est situé à plus de 500 km au sud des sites pyrénéens.

En Europe, on trouve cette orchidée depuis le niveau de la mer au nord jusqu'à plus de 2000 m d'altitude dans les Alpes. En France, elle est toujours rare, de la Lorraine et de la Haute-Marne aux Alpes, très rare dans les Pyrénées et les grands Causses ; disparu en

Fig. 1. Illustration de *Cypripedium calceolus* dans le Djurdjura: a & b) individus en fleurs, 7.6.2019 (photos W. Nemer); c) individu fané; d) site de la découverte, 29.6.2019 (photo K. Rebbas).

Fig. 2. Localisation géographique de la station nord-africaine de *Cypripedium calceolus* dans le Djurdjura, au sud-ouest du col de Tirourda.

Tableau 1. Liste de la flore inventoriée dans la station de *C. calceolus.*

Nom scientifique	
Allium tenuiflorum Ten.	*Dianthus vulturius* Guss. & Ten
Acinos alpinus subsp. *meridionalis* (Nyman) P.W. Ball	*Epipactis tremolsii* Pau
Aegilops triuncialis L.	*Genista tricuspidata* Desf.
Allium fontanesii J. Gay	*Himantoglossum hircinum* (L.) Spreng.
Ampelodesmos mauritanicus (Poir.) T. Durand & Schinz	*Inula montana* L.
Anacamptis pyramidalis (L.) Rich.	*Juniperus communis* subsp. *hemisphaerica* (C. Presl) Nyman
Anisantha rubens (L.) Nevski	*Jurinea humilis* (Desf.) DC.
Anthyllis montana L.	*Lomelosia crenata* (Cirillo) Greuter & Burdet
Anthyllis vulneraria L.	*Mantisalca salmantica* (L.) Briq. & Cavill.
Aphanes floribunda (Murb.) Rothm.	*Orchis anthropophora* (L.) All.
Armeria plantaginea All.	*Orchis olbiensis* Gren.
Asperula hirsuta Desf.	*Origanum vulgare* subsp. *glandulosum* (Desf.) Ietsw.
Astragalus armatus subsp. *numidicus* (Murb.) Tietz	*Plantago mauritanica* Boiss. & Reut.
Bartsia trixago L.	*Poa bulbosa* L.
Briza maxima L.	*Quercus ilex* subsp. *ballota* (Desf.) Samp.
Bupleurum spinosum Gouan	*Rosa canina* L.
Carduus macrocephalus Desf.	*Sedum acre* L.
Carthamus atractyloides (Pomel) Greuter	*Sedum album* L.
Catananche caerulea L.	*Sedum caeruleum* L.
Cedrus atlantica (Endl.) Carrière	*Sedum sediforme* (Jacq.) Pau
Centaurea pubescens Willd.	*Silene coelirosa* (L.) Godr.
Crataegus laciniata Ucria	*Thymus munbyanus* subsp. *ciliatus* (Desf.) Greuter & Burdet
Crucianella angustifolia L.	*Thymus willdenowii* Boiss.
Dactylis glomerata L.	*Trifolium arvense* L.
Dactylorchis battandieri Raynaud	*Trifolium stellatum* L.
Dianthus serralatus subsp. *eu-serrulatus* Maire	*Trisetum flavescens* (L.) P. Beauv.

Belgique et au Luxembourg (Bournérias & Prat 2005). *Cypripedium calceolus* est répandu dans les Pays-Bas, Amérique du Nord, Eurasie et Japon. En Europe, il vient du sud-ouest jusqu'aux Pyrénées. Sur la péninsule, il ne pousse qu'à Huesca et à Barcelone (Bañares & al. 2004). La Suède continentale est une région qui compte les plus grandes populations européennes (Antonelli & al. 2009). En Angleterre, *Cypripedium calceolus* n'a jamais été aussi répandu et a été surexploité au point d'être presque éteint vers la première moitié du XXe siècle. Il s'est accroché sur un site et a depuis été propagé et réintroduit avec succès. Ailleurs, il est extrêmement rare (par exemple au Danemark) ou éteint (par exemple en Grèce), et atteint des populations relativement importantes (par exemple en Pologne et en Autriche).

En raison de sa large aire de répartition, cette espèce d'Eurasie pourrait être considérée comme un taxon faisant l'objet d'une préoccupation mineure, quasi menacée, vulnérable, en voie de disparition ou gravement en danger (Khapugin & al. 2017). Bien que *C. calceolus* soit inclus dans la liste rouge mondiale de l'UICN avec la caté-

gorie Préoccupation mineure (LC), il s'agit d'une des espèces d'orchidées les plus menacées d'Europe (Kull 1999). Cette orchidée est inscrite sur plusieurs listes rouges nationales comme menacée : - Régionalement éteinte au Luxembourg. - En danger critique d'extinction en Bulgarie, en Serbie et au Royaume-Uni. - En danger en Croatie, en République tchèque, en Hongrie, en Russie et en Espagne. - Vulnérable en Autriche, Biélorussie, Danemark, France, Allemagne, Lituanie, Slovaquie et Suisse. - Presque menacé en Finlande et en Norvège. - Préoccupation mineure en Suède (Rankou & Bilz 2014). - En danger critique d'extinction en Mordovia (Khapugin & al. 2017).

Conservation de *Cypripedium calceolus*

Les orchidées pantoufles sont communes dans les Amériques, ainsi qu'en Europe et dans les régions tempérées et asiatiques, au sud de l'Himalaya. Cependant, au début des années 1980, *C. calceolus*, pantoufle de la dame britannique, était réduit à un individu à floraison unique dans la nature. La collecte excessive par les amateurs d'orchidées et la disparition d'habitats convenables ont vu leur nombre diminuer jusqu'à l'extinction. En 1983, Sir Robert et Lady Sainsbury ont donc financé un projet dans l'unité de micropropagation de Kew pour tenter d'améliorer les techniques de culture d'orchidées sauvages dans des conditions artificielles (KS - RBG 2019).

L'expérience de culture en plein air de plants de *Cypripedium calceolus* L. ex vitro en République Tchèque a donné les premiers résultats positifs (Obdržálek 2009).

Des analyses d'extinction ont montré que pour une population d'une vingtaine d'individus (caractéristiques de nombreuses localités européennes), il n'y a aucune chance de survie si deux plantes seulement sont enlevées à environ cinq ans d'intervalle, un niveau d'agression qui n'est probablement pas atypique (Terschuren 1999). Cependant, pour aggraver encore la situation en matière de conservation, la pollinisation limite également le succès de reproduction des orchidées (Antonelli & al. 2009).

Il est protégé au niveau national dans la plupart des pays (France, Hongrie, par exemple) et la collecte de l'espèce est interdite (par exemple, en Lituanie). De nombreuses populations sont incluses dans des sites Natura 2000 et d'autres formes d'aires protégées. La protection des sites et une gestion appropriée sont essentielles (Rankou & Bilz 2014).

Conclusion

La poursuite de ce travail visera l'exploration botanique d'autres stations de la région de la Kabylie.

Pour la conservation durable du site jusqu'alors connu de *Cypripedium calceolus* au Djurdjura, de sévères mesures de protection semblent indispensables. Il convient d'analyser l'utilisation actuelle de la zone ainsi que les menaces éventuelles (surpâturage, incendies, réchauffement climatique) de la petite population, d'envisager la gestion future et les mesures de conservation par culture semblent être intéressantes à un stade ultérieur.

Il est important de classer cette orchidée dans la liste de plantes non cultivées et protégées figurant dans le décret exécutif algérien (J.O.R.A. 2012) et que nos prochaines explorations botaniques dans le parc national de Djurdjura apportent davantage d'informations concernant la phytosociologie et la dynamique de la population de *C. calceolus* pour son évaluation dans la liste rouge de l'UICN.

Remerciements

L'exploration à l'origine de cette découverte n'aurait pas été possible sans la complicité et l'efficacité de nos compagnons de terrain Mustapha Lateb, Massi Madouche et Youcef Rebbas. Les auteurs tiennent à remercier les relectures pour leurs remarques et leurs commentaires afin d'améliorer cet article.

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Short note

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F. Conti, F. Falcinelli, V. Giacanelli, M. Paolucci, G. Pirone, E. Proietti, A. Stinca & F. Bartolucci

New floristic data of vascular plants from central and southern Italy

Abstract

Conti, F., Falcinelli, F., Giacanelli, V., Paolucci, M., Pirone, G., Proietti, E., Stinca, A. & Bartolucci, F.: New floristic data of vascular plants from central and southern Italy. — Fl. Medit. 29: 215-222. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

Based on field, herbarium and bibliographic research, new distributional data for 18 taxa (11 native and 7 aliens) are reported for some regions of central and southern Italy. *Umbilicus luteus* is confirmed for Italy, other four taxa are new or confirmed to Abruzzo, three to Molise, two to Marche, Umbria and Campania, and one to Basilicata and Calabria respectively. In addition, new distribution data for other three rare or interesting taxa in Abruzzo are reported.

Key words: alien species, biodiversity, distribution, native species, invasiveness, Italian flora.

Introduction

The long tradition of botanical investigations in Italy, started with the botanists of the 1700s, has made possible to obtain a good knowledge of its floristic diversity. The recent checklists of native and alien vascular flora (Bartolucci & al. 2018a; Galasso & al. 2018a) placed an updated starting point for floristic and taxonomic studies in Italy. This work is a further contribution to the knowledge of the flora of central and southern Italy as other recently published (e.g. Bartolucci & al. 2019a; Conti & al. 2019; Stinca & al. 2019a, 2019b) and it is part of a series of floristic studies carried out by the authors which mainly concern the Apennines (Conti & al. 2017, 2018). It presents unpublished floristic findings, confirmations of old citations, and status changes for alien plants in central and southern Italy. The floristic novelties presented concern Umbria, Marche, Abruzzo, Molise, Campania, Basilicata and Calabria administrative regions.

Materials and methods

The floristic data are based on field investigations carried out from 1990 to 2019, and bibliographic researches. Herbarium specimens are preserved in the Herbarium Apenninicum (APP) and Herbarium Porticense (PORUN, acronyms follow to Thiers 2019).

The nomenclature follows the checklists of the vascular flora native and alien to Italy and following updated (Bartolucci & al. 2018a, 2018b, 2018c, 2019b; Galasso & al. 2018a, 2018b, 2018c, 2019).

For each taxon, ordered alphabetically, the following information is provided: currently accepted name; family; reasons for it being recorded; current invasiveness status (only for the alien taxa), information on examined herbarium specimens and any additional notes.

Results and Discussion

Adenocarpus complicatus subsp. ***samniticus*** (Brullo, De Marco & Siracusa) Peruzzi (*Fabaceae*)

Second report for Abruzzo.

Italy, Abruzzo, Tufo Basso, Carsoli (L'Aquila), sopra il paese, margine di bosco, WGS84: 42°03'06" N 13°07'31" E, 950 m, 19.03.2017, *F. Conti et V. Giacanelli* (APP n. 59972).

In Abruzzo, this endemic taxon (Peruzzi & al. 2014; Bartolucci & al. 2018a), was previously reported from Colle Centoscudi to Fosso di Patrignone close to Verrico (Montereale, L'Aquila) (Bartolucci & al. 2019a).

Allium angulosum L. (*Amaryllidaceae*)

New species for Abruzzo and peninsular Italy.

Italy, Abruzzo, Altopiano delle Rocche. Prato della Madonna, Rocca di Mezzo (L'Aquila), prato umido, WGS84: 42°13'14.34"N 13°31'22.32"E, 1264 m, 05.07.2018, *F. Conti, F. Bartolucci, A. Stinca et E. Proietti* (APP n. 59534, PORUN).

In Italy this species is recorded for the alpine regions from Piemonte to Friuli Venezia Giulia and in Emilia-Romagna. In Val d'Aosta it is extinct, while in Toscana, Marche, Umbria, Abruzzo and Puglia was indicated by mistake (Bartolucci & al. 2018a). Particularly in Abruzzo it was recorded by Gussone (1826) "*in herbosis saxorum ad rimas montium Aprutii; Majella nella valle dell'Orfenta sotto la Mucchia al nord; e nella valle dell'Inferno*". These records are probably to be referred to *A. lusitanicum* Lam. This new finding represents the southern limit of Italian distribution of this species.

Aloë maculata All. (*Asphodelaceae*)

Casual alien species new for the flora of Abruzzo.

Italy, Abruzzo, Pineta d'Avalos (Pescara), gariga a cisti, WGS84: 42°27'10.6"N 14°14'11.5"E, 4 m, 05.10.2019, *F. Conti* (APP n. 65690).

It has been observed only in a group of ten individuals.

Cardamine occulta Hornem. (*Brassicaceae*)

Casual alien species new for the flora of Abruzzo.

Italy, Abruzzo, Palombara (L'Aquila), via Bazzanese, margine stradale, WGS84: 42°20'32.09"N 13°25'44.86"E, 660 m, 31.03.2019, *F. Bartolucci* (APP n. 64917, 64918, 64919).

Only few individuals were observed.

Cotula australis (Sieber ex Spreng.) Hook.f. (*Asteraceae*)
Casual alien species new for the flora of Campania.
Italy, Campania, Napoli all'Azienda Ospedaliera Universitaria Federico II (Napoli), prato sfalciato e fessure della pavimentazione, WGS84: 40°52'4.61"N-14°13'6.93"E, 279 m, 02.03.2018, *A. Stinca*, (PORUN).

The population discovered in Campania covers an area of about 100 m^2 with many individuals producing fruits.

Eclipta prostrata (L.) L. (*Asteraceae*)
Status change from casual to naturalized alien for the flora of Abruzzo.
Italy, Abruzzo, Villa Rosa, Stabilimento Nemo, Martinsicuro (Teramo), fosso sulla spiaggia, WGS84: 42°51'55.6"N 13°55'32.0"E, 0 m, 1.08.2018, *F. Bartolucci* (APP n. 59953).

In Abruzzo this species was previously recorded only for Lago di Bomba and Selva di Altino (Conti & al. 2016).

Huperzia selago (L.) Bernh. ex Schrank & Mart. subsp. ***selago*** (*Lycopodiaceae*)
Species new for the flora of Abruzzo.
Italy, Abruzzo, Bosco Martese in loc. Ceppo, Rocca Santa Maria (Teramo), bosco di faggio, WGS84: 42°40'05.8"N 13°27'43.7"E, 1310 m, 15.09.2019, *F. Bartolucci* (APP n. 65689).

This species was recorded in central Apennine only on Monti della Laga (Lazio and Marche sectors) (Tondi & Plini 1995; Marchetti 2004; Anzalone & al. 2010). The records from Monti della Laga are at the southern limit of Italian distribution of this species.

Leontodon hispidus L. subsp. ***dubius*** (Hoppe) Pawłowska (*Asteraceae*)
Subspecies new for the flora of Marche.
Italy, Marche, Monte Vettore, presso la vetta (Arquata del Tronto, Ascoli Piceno), brecciaio, WGS84: 42°49'19.9"N 13°16'36.7"E, 2378 m, 29.07.2011, *F. Conti et P. Minghetti* (APP n. 62583, 62584, 62585, 62586).

In Italy this taxon is recorded for Lombardia, Trentino-Alto Adige, Friuli Venezia Giulia and Abruzzo (Bartolucci & al. 2018a). In central Apennine it is very rare and recorded only for Gran Sasso and Majella National Parks (Conti & Bartolucci 2016; Conti & al. 2019).

Nectaroscilla hyacinthoides (L.) Parl. (*Asparagaceae*)
Casual alien species new for the flora of Umbria.
Italy, Umbria, tra Montebello e Colonnetta (Perugia), margine stradale, suolo calcareo, WGS84: 43°4'24.31"N 12°24'4.02"E , 300 m, 28.04.2018, *F. Falcinelli* (APP n. 65273).

Pentanema bifrons (L.) D.Gut.Larr., Santos-Vicente, Anderb., E.Rico & M.M.Mart.Ort. (*Asteraceae*)
Species new for the flora of Molise.
Italy, Molise, Monte Sant'Onofrio di Agnone (Isernia), bosco di Montecastelbarone, incolto erboso, WGS84: 41°51'26.114"N 14°22'24.088 "E, 1370 m, 28.09.2019, *M. Palumbo* (APP n. 65691).

This new finding represents the southern limit of Italian distribution of this species (Bartolucci & al. 2018a).

Potamogeton pusillus L. (*Potamogetonaceae*)
Species new for the flora of Basilicata.
Italy, Basilicata, Monte Sirino, Lago Laudemio, Lagonegro (Potenza), acqua ferma, WGS84: 40°08'34.7"N 15°50'09.4"E, 1533 m, 24.08.2010, *G. Pirone* (APP n. 65131).
This species is recorded for northern and central Italy, rarest in the south where it is not confirmed in Campania and Calabria (Bartolucci & al. 2018a).

Ruppia maritima L. (*Potamogetonaceae*)
Species new for the flora of Molise.
Italy, Molise, Foce del Biferno, Campomarino (Campobasso), acqua lentamente fluente, WGS84: 41°58'39.4"N 15°01'46.1"E, 1 m, 12.07.1997, *F. Conti* (APP n. 64342, 64343); *ibidem*, 30.04.1990, *F. Conti et A. Stanisci* (APP n. 64433).

Satureja hortensis L. (*Lamiaceae*)
Status change from casual to naturalized alien for the flora of Abruzzo.
Italy, Abruzzo, Rocca di Mezzo (L'Aquila), WGS84: 42°12'16.09"N 13°31'8.68"E, 1280 m, incolti e margine stradale, 05.06.2018, *F. Bartolucci* (APP n. 65100, 65101).

Sporobolus alopecuroides (Piller & Mitterp.) P.M.Peterson (*Poaceae*)
Species confirmed for the flora of Campania.
Italy, Campania, Lago di Gallo Matese (Caserta), WGS84: 41°27.787'N 14°14.155'E, 837 m, 24.09.2006, *F. Conti* (APP n. 65027).

Sporobolus indicus (L.) R.Br. (*Poaceae*)
Naturalized alien species new for the flora of Marche.
Italy, Marche, San Benedetto del Tronto (Ascoli Piceno), giardino dell'Università, Lungomare Scipioni, WGS84: 42°56'04.5"N 13°53'33.1"E, 2 m, 1.08.2019, *F. Conti et F. Bartolucci* (APP n. 65533).
Also observed in the nearby pine plantation and in the surrounding traffic lanes.

Thymus striatus Vahl subsp. ***striatus*** (*Lamiaceae*)
Subspecies confirmed for the flora of Molise.
Italy, Molise, Pescopennataro (Isernia), WGS84: 41°52.728'N 14°17.542'E, 1142 m, 10.06.2006, *F. Conti* (APP n. 64727).
The distribution of the two recognized subspecies of *Th. striatus* (Bartolucci & al. 2013; Bartolucci & Peruzzi 2014) in central Italy is uncertain and under study (Bartolucci 2018).

Tolpis virgata (Desf.) Bertol. subsp. ***virgata*** (*Asteraceae*)
Species new for the flora of Umbria.
Italy, Umbria, Castiglione del Lago (Perugia), tra Ferretto e Pieracci nei pressi del Pod.e Querce Pendine, orlo di querceto caducifoglio, suolo calcareo, WGS84: 43°9'45.90"N 11°59'17.40"E, 282 m, 18.06.2018, *F. Falcinelli* (APP n. 65326).

Umbilicus luteus (Huds.) Webb & Berthel. (*Crassulaceae*)
Species confirmed for the flora of Calabria and Italy.
Italy, Calabria, M. Pettinascura, S. Giovanni in Fiore (Cosenza), 19.06.1990, *F. Conti* (APP n. 63491).

This species was excluded from Italy since all the previous reports were regarded as erroneous and to be referred to *U. horizontalis* (Guss.) DC. (Bartolucci & al. 2018a). The recent examination of a specimen housed in the Herbarium of Michele Guadagno preserved in PI (PI-GUAD) collected by Grande (1913) on Sila (Calabria), led Gallo (2019) to consider this species as doubtful to Italy as it was not recorded since more than one century. Therefore, the new finding confirms the presence of *U. luteus* in Italy.

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Short note

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N. Sakhraoui, A. Chefrour & S. Metallaoui

Naturalisation de *Melia azedarach* (*Meliaceae*) et premier signalement de *Canna indica* (*Cannaceae*) et *Pelargonium zonale* (*Geraniaceae*) en Algérie

Abstract

Sakhraoui, N., Chefrour, A. & Metallaoui, S.: Naturalisation de *Melia azedarach* (*Meliaceae*) et premier signalement de *Canna indica* (*Cannaceae*) et *Pelargonium zonale* (*Geraniaceae*) en Algérie. — Fl. Medit. 29: 223-226. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

In this contribution, we report the naturalization of *Melia azedarach* and the discovery of *Canna indica* and *Pelargonium zonale* growing outside the usual cultivation sites, making this work the first report of these species in Algeria. Some species must be monitored because they are invasive in the Mediterranean area.

Key words: alien flora, xenophytes, mode of reproduction, invasive character, Algeria.

Introduction

Bien qu'elles participent à l'enrichissement de la flore nationale, plusieurs espèces de la flore naturalisée, qui ne cesse d'augmenter en Algérie, sont considérées comme envahissantes ou potentiellement envahissantes (Meddour & El Mokni 2016) et peuvent de ce fait constituer un véritable danger pour la biodiversité locale et les écosystèmes en général (Shine & al. 2000).

Nous rapportons ici, la découverte de trois espèces exotiques proliférant en dehors des lieux de cultures habituels, faisant de ce travail, le premier signalement de ces espèces en Algérie. Les plantes en question, dont certaines possèdent un caractère envahissant, ont été observées lors de la réalisation de prospections floristiques dans la wilaya de Skikda (Sakhraoui & al. 2019). Cette étude met le point sur certaines de leurs caractéristiques biologiques et écologiques permettant de mieux comprendre leur comportement dans notre pays, rendant ainsi possible la prévention contre d'éventuelles futures invasions.

Matériel et Méthodes

L'identification des espèces a été effectuée grâce à la consultation de Maire (1959), Maas-Van de Kamer & Maas (2008) et Cullen & al. (2011). La présentation des familles végétales suit l'Angiosperme Phylogeny Group (APG III 2009). La nature des

milieux colonisés et le mode de reproduction ont été déterminés *in situ*. Le type biologique a été donné selon Raunkiaer (1934), quant au degré de naturalisation, il a été évalué selon Magnanon & al. (2008).

Résultats et discussion

Melia azedarach L. (*Meliaceae*)

Phanérophyte, originaire de l'Asie du sud (Yulianti & al. 2011), introduite par le jardin d'essai du Hamma avant 1843 (Payen 1845). Observée durant cinq années consécutives de 2014 à 2019 çà et là dans les ruelles de la cité Larbi Ben M'hidi et le long de la route reliant la cité à la ville de Skikda, où elle se présente souvent en pieds isolés poussant parmi la flore indigène. Les fruits ont été observés mais la germination des graines n'a pas été enregistrée. Les dimensions considérables des individus observés indiquent qu'ils se sont établis dans la région depuis plus de dix années ce qui nous permet de les considérer comme complètement naturalisés.

Canna indica L. [≡ *C. aurantiaca* Roscoe, ≡ *C. cearensis* Huber] (*Cannaceae*)

Géophyte, originaire de l'Amérique tropicale (Maire 1959), introduite en 1838 par le jardin d'essai du Hamma (Carra & Gueit 1952). Observée à plusieurs points de la cité Salah Chebel et à l'endroit appelé « El guelta » (commune de Hamadi Krouma) où de petites populations, séparées les unes des autres, prolifèrent parmi les espèces indigènes. Elle a aussi été retrouvée au bord de la route reliant Stora à la ville de Skikda. Les fruits ont été observés, mais la germination des graines n'a pas été enregistrée. En voie de naturalisation, car son apparition dans la région est récente (moins des 10 années requises).

*Pelargonium zonal*e (L.) L'Hér. [≡ *Geranium zonale* L.] (*Geraniaceae*)

Phanérophyte, originaire d'Afrique du Sud (Cullen & al. 2011), probablement introduite par le jardin d'essai du Hamma, Battandier & al. (1914) rapportent que *P. zonale* était naturalisée dans le jardin botanique d'Alger. Retrouvée en 2017 dans plusieurs points de la cité Salah Chebel (commune de Hamadi Krouma) et en 2019, au bord de la route reliant Filfilla à Hamrouche Hamoudi, et dans une décharge à la cité Larbi Ben M'Hidi. Les fruits ont été observés mais la germination des graines n'a pas été enregistrée. Subspontanée, car la plante ne se propage pas dans le milieu pour se mêler à la flore indigène.

Toutes les espèces sont considérées comme naturalisées dans d'autres régions du monde (Henderson 2007; Uludağ & al. 2017; Domina & al. 2018; Domingues De Almeida 2018; Galasso &al. 2018), où elles ont été introduites comme plantes d'ornement. En Afrique du Nord, *C. indica* est signalée naturalisée en Tunisie (El Mokni 2018), alors que *P. zonale* est signalée cultivée/parfois subspontanée à Madère et à statut problématique aux Canaries et au Maroc (Dobignard & Chatelain 2012).

En Algérie, à l'exception du *M. azedarach*, rapportée par Dobignard & Chatelain (2012) cultivée/parfois subspontanée dont nous signalons la naturalisation complète pour la première fois dans le pays, les deux autres espèces n'ont pas été signalées auparavant dans les ouvrages traitant la flore algérienne notamment Quezel & Santa (1962; 1963) et Dobignard & Chatelain (2010-2013).

C. indica et *M. azedarach* sont considérées comme envahissantes notamment dans le bassin méditerranéen (Capdevilla Argüelles & al. 2006; Arianoutsou & al. 2010; Podda & al. 2011). Bien qu'elles ne soient pas encore considérées comme invasives avérées dans notre région, elles pourraient le devenir dans les prochaines années, favorisées par le changement climatique que connait l'Algérie et l'intensification des perturbations des milieux naturels et semi naturels dans la wilaya de Skikda générée par les projets de développement local. En effet, le réchauffement climatique et les perturbations des milieux sont considérés comme des facteurs susceptibles d'amplifier les invasions biologiques (Gritti & al. 2006; Walther & al. 2009).

Ces espèces devraient donc figurer sur une liste de surveillance et être soumises à des contrôles périodiques permettant la détection précoce de toute invasion réelle.

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Article

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Le lac Fetzara (Nord-Est algérien): Biodiversité floristique et menaces potentielles

Abstract

Fetnaci, I., Beddiar, A. & Hamel, T.: Le lac Fetzara (Nord-Est algérien): Biodiversité floristique et menaces potentielles. — Fl. Medit. 29: 227-245. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

The wetlands of northeastern Algeria are rich in flora because of their biogeographical context. The Fetzara Lake, designated Ramsar site since 2002, has been the subject of multiple plant surveys conducted between 2014 and 2019 and has revealed the existence of 226 taxa belonging to 166 genera and 58 families with the dominance of therophytes (45.13%, 102 species). From the chorological point of view, the Mediterranean type was the most dominant with 128 taxa or 56.64% of the studied flora 19 of which are endemic to Algeria. Canonical analysis represented the superimposition of the three physiognomic environments of vegetation (dry lawns, amphibian meadows and semi-permanent lake) with respect to environmental variables. The results suggest that the distribution and variation of species abundance is mainly ordered according to salinity and grazing. These degradation factors therefore merit to be divulgated to arouse awareness of the public authorities and the population in order to implement appropriate protective measures.

Key words: floristic diversity, environmental variables, threats, conservation.

Introduction

Les zones humides sont considérées parmi les écosystèmes les plus riches et les plus diversifiés (Mitsch & Gosselink 2007). Ces milieux assurent plusieurs fonctions majeures telles que le stockage de l'eau, le contrôle des inondations et le piégeage des éléments chimiques notamment les toxiques (Keddy 2000; Williams 2006). Ce sont aussi des hauts lieux de biodiversité renfermant plusieurs espèces animales et végétales rares ou menacées d'extinction (Médail & al. 1998; Rhazi & al. 2001; de Bélair 2005; Ferchichi-Ben Jamaa & al. 2010; Samraoui & al. 2010). Ce sont des milieux très utiles mais aussi très menacés, ils subissent plusieurs pressions en raison des multiples activités humaines (drainage, aménagement agricole, pâturage et pollution) (Rhazi & al. 2001, 2006). Durant le dernier siècle, plus de la moitié des zones humides mondiales auraient disparues (Sajaloli 1996 ; Daumas & al. 2004 ; Hammada & al. 2004; Ferchichi-Ben Jamaa & al. 2010; Bouldjedri & al. 2011; Laribi & al. 2016).

Les zones humides des pays méditerranéens sont particulièrement touchées par ce phénomène et font face à une plus grande perte (Bonnet & al. 2005). Leur situation est donc très préoccupante étant donné que ces zones représentent un élément majeur des « points chauds » de biodiversité de la région méditerranéenne (Véla & Benhouhou 2007). Cette région est en effet considérée comme le troisième hotspot le plus riche du monde en diversité végétale (Myers & al. 2000).

L'Algérie n'est pas épargnée par ce problème, elle a été exposée au cours de ces dernières décennies à une érosion marquée des zones humides précieuses (Samraoui & al. 1992 ; de Bélair & al. 1994 ; Samraoui & al. 2011). En dépit des menaces qui pèsent sur ces écosystèmes et sur leur flore, les études phytodynamiques qui les concernent restent peu nombreuses (de Bélair 2005 ; Kadid & al. 2007 ; Bouldjedri & al 2011 ; Neffar & al. 2013). Bien que l'Algérie recèle d'un grand nombre de zones humides assez importantes concentrées principalement dans le nord-est du pays, à proximité du littoral méditerranéen. La zone phytogéographique comprenant la Kabylie et la Numidie en Algérie et la Kroumirie en Tunisie a récemment suscité la proposition de la classer en tant que point chaud de biodiversité (Véla & Benhouhou 2007).

C'est dans cette zone que se trouve le lac Fetzara dans la willaya d'Annaba, le site de la présente étude. C'est l'un des plus importants lacs de l'extrême Nord-Est Algérien, classé officiellement, en 2002, comme une zone « Ramsar » (DGF 2002: 53-55). Autrement dit, c'est une zone humide d'importance internationale car elle reçoit en moyenne 20000 oiseaux d'eau appartenant à plus de 50 espèces inféodées aux zones humides. Plusieurs études ont été effectuées sur les eaux et les sols de ce site (Marre 1992 ; Djamaï 2007 ; Habes & al. 2012 ; Zahi 2014 ; Halimi & al. 2018) et ont relevé le caractère de salinité élevée de ce dernier. Cependant, les études sur la flore de cette zone demeurent rudimentaires voire inexistantes.

Sa grande étendue et son caractère relativement temporaire font de ce site une zone humide représentative et rare de type de zone humide de la région méditerranéenne (Djamai & al. 2006). Ce lac est très affecté, d'une part par l'impact du surpâturage et des aménagements hydrauliques, d'autre part par l'augmentation de la salinité sur les parties Est et Ouest. Un déplacement des sels vers la périphérie du lac avec une dessalement du centre a été observé pendant la période sèche et durant les dernières années (Durant 1950, Ifragria 1967, Djamaï & al. 2006).

L'objectif du présent travail est (1) de dresser un premier inventaire de la flore de lac Fetzara, intégrant les espèces strictement inféodées aux zones humides et les espèces transgressives ou parfois accidentelles des milieux voisins ou apportées par un agent de dissémination, (2) de réaliser une analyse quantitative, sur la base de paramètres biogéographiques tels que la richesse spécifique, l'endémisme, la rareté et la répartition géographique, (3) évaluer l'ampleur et la nature des menaces le concernant avec la proposition des mesures de conservation et de protection.

Matériel et Méthodes

Zone d'étude

Le lac Fetzara, est une vaste dépression située dans l'extrême Nord-est algérien et se trouve à 18 km au Sud-ouest de la ville d'Annaba et 14 Km de la mer Méditerranée avec

une altitude allant de 5 à 25m (Fig. 1). Il est limité au Nord par le massif de l'Edough, par les collines d'Ain Berda au Sud et les cordons dunaires situés à l'Est et à l'Ouest. Il s'allonge sur 17 km d'Est en Ouest et sur 13 km du Nord au Sud dans sa partie la plus large, sa superficie est d'environ 18600 hectares (Djamaï & al. 2007). La partie inondée du lac est située au centre et dépend fortement de la saison des pluies, elle couvre en hiver une superficie moyenne estimée à 5800 hectares (Djamaï & al. 2006).

Les eaux du lac Fetzara proviennent des précipitations ainsi que par trois principaux oueds : l'Oued Zied, Oued El Hout et Oued El Mellah; ces cours d'eau temporaires sont torrentiels en hiver et secs en été. Un canal d'assèchement de 14 km de long achemine les eaux du Lac vers l'Oued Meboudja puis l'Oued Seybouse qui se déverse en mer Méditerranée. Ce dernier a été construit entre 1943 à 1947 par la société des mines « Mokta el Hadid » afin de récupérer des terres cultivables (Durand 1950).

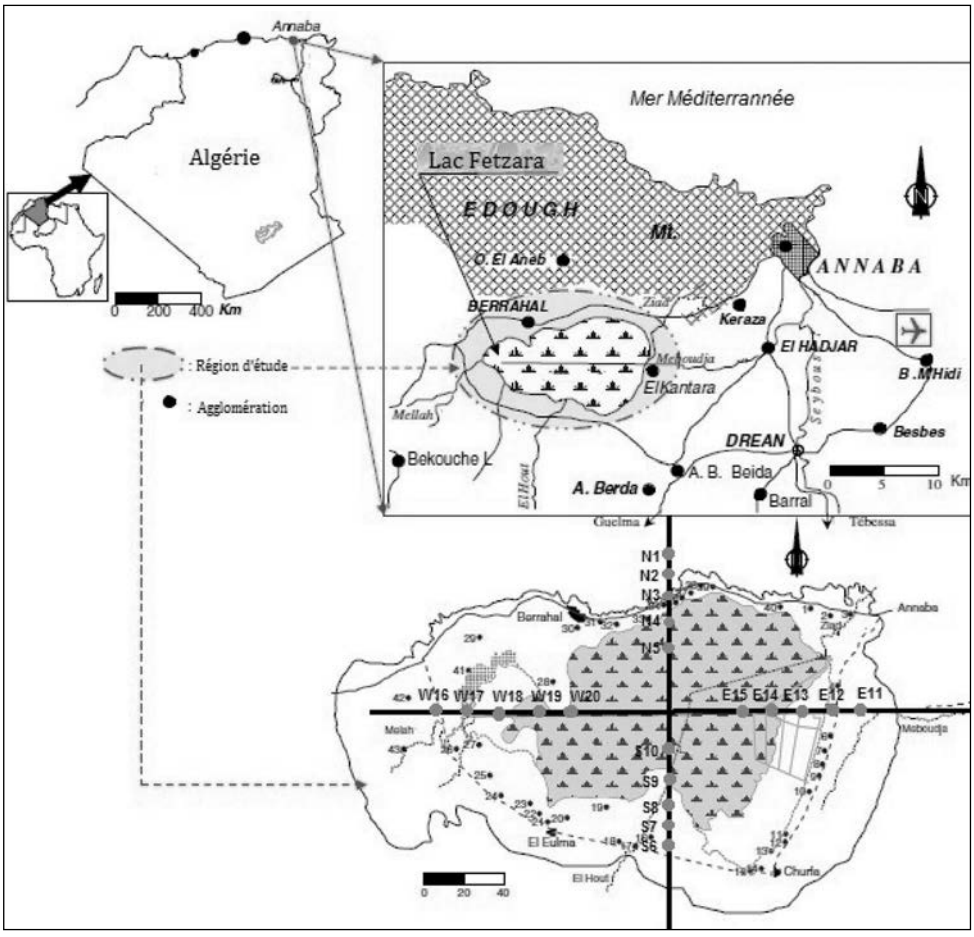

Fig. 1. Situation géographique du lac Fetzara (Nord-Est algérien).

La région d'étude est soumise à un climat méditerranéen, doux et humide en hiver et chaud et sec en été et reçoit une moyenne annuelle de pluie qui varie entre 600 mm à 700 mm. La température moyenne est de 11C° en hiver et de 25 C° en été. Bien que la température estivale peut atteindre 40 ° C (Rouabhia & al. 2012). Selon Djamaï (2007), la salinité des eaux et des sols du lac Fetzara est assez élevée. La conductivité électrique pour les sols et les eaux souterraines atteindraient respectivement des valeurs de 2,43 mS/cm et 2,1 mS/cm et seraient plus importantes au niveau de la région Sud-Est (commune de Cheurfa) et celle de la région Nord-Est du lac (commune de Oued Zied).

La géologie de cette zone comporte principalement des alluvions récentes du quaternaire constituées de limons, de sable, de gravier et de travertin (Joleaud 1936 ; Marre 1992 ; Djamaï & al. 2006; Habes & al. 2012).

Méthodologie

Etude floristique: la végétation du lac Fetzara a été étudiée à trois périodes par an (février-mars, avril-mai et juin-juillet) pendant six cycles hydrologiques (2014-2019). Cinq quadrats de superficie 100m^2 ont été appliqués par transects de longueur de 1000m depuis la zone exondée vers la zone inondée. Chaque transect est orienté en fonction de l'exposition par rapport au centre du lac (Transect Nord, Sud, Est et Ouest).

Les taxons ont été identifiés selon la flore d'Algérie (Quézel & Santa 1962-1963), la flored'Afrique du Nord (Maire 1952-1987), la flore d'Italie (Pignatti 1982) d'autre part.

La nomenclature a été mise à jour pour les espèces inventoriées en tenant compte des travauxrécents compilés dans l'index synonymique et bibliographique de la flore d'Afrique du Nord (Dobignard & Chatelain 2010-2013) et le site web de la base de données des plantes d'Afrique [http://www.ville-ge.ch/ musinfo/bd/cjb/africa/ recherche.php?langue=fr].

Les espèces recensées ont été renseignées par leur type biogéographique (Pignatti 1982 ; Blanca & al. 2009 ; Dobignard & Chatelain 2010-2013) et selon leur type biologique (Raunkier 1934; Pignatti 1982; Blanca & al. 2009; Tison & de Foucault 2014).

Analyse numérique des données floristiques: l'ensemble des relevés floristiques etenvironnementales a été soumis à une Analyse Canonique des Correspondances (ACC). Le croisement des données de la flore codées en présence-absence avec les variables environnementales (exposition, altitude, salinité, type de sol, taux de recouvrements des ligneuses et des herbacées, pâturage, incendie et activité agricole est obtenu par cette analyse (Tab. 1).

Le nuage résultant de l'ACC permet de visualiser le pourcentage explicatif d'une variable sur une autre (Ter Braak 1995). Cette analyse a été effectuée en utilisant le langage informatique R (package ade4, version 3.0.2) (R Development Core Team 2013).

Analyse pédologique : les variables pédologiques mesurées pour chaque station sont: (1) la granulométrie par la technique de Casagrande (1934); (2) la conductivité électrique; (3) le pH et le CaCO3 selon Aubert (1978); (4) la matière organique selon Afnor (1999).

Résultats

Diversité floristique

Un total de 226 espèces de plantes vasculaires appartenant à 166 genres et 58 familles ont été identifiées dans la zone du lac Fetzara (Supplément Électronique 1). La famille des

Tableau 1. Caractéristiques écologiques des différentes stations.

Station	Expo	Alti	Sali	Subs	Trli	Trhe	Patu	Ince	Acag
N1	1	4	1	1	1	5	4	1	2
N2	1	2	2	1	1	3	4	1	2
N3	1	3	2	1	1	3	3	1	1
N4	1	4	3	1	1	3	3	1	1
N5	1	4	3	1	1	2	1	1	1
S6	2	4	1	2	2	5	2	1	2
S7	2	3	1	2	1	4	3	1	2
S8	2	5	1	2	1	4	2	1	2
S9	2	3	2	3	1	3	1	1	1
S10	2	4	2	3	1	2	1	1	1
E11	3	4	2	2	2	5	4	1	2
E12	3	3	2	2	2	5	3	1	1
E13	3	2	3	3	1	4	3	2	1
E14	3	3	4	3	1	3	2	1	1
E15	3	4	4	1	1	2	2	1	1
W16	4	2	2	2	2	4	4	1	2
W17	4	1	3	2	1	3	3	1	1
W18	4	3	4	1	1	2	2	1	1
W19	4	2	4	1	1	2	2	1	1
W20	4	3	4	1	1	2	1	1	1

Transect Nord : N1, N2, N3, N4 et N5. Transect Sud: S6, S7, S8, S9 et S10. Transect Est : E11, E12, E13, E14 et E15. Transect Ouest : W16, W17, W18, W19 et W20.
Trli: taux de recouvrement des espèces ligneuses, Trhe : taux de recouvrement des espèces herbacées, Patu : pâturage, Ince : incendie, Acag : activité agricole.
[Expo : 1= Nord, 2= Sud, 3= Est, 4= Ouest/ Alti : 1= de 0 à 5m, 2= de 5 à 10m, 3= 10 à 15m, 4= de 15 à 20m, 5= de 20 à 40m/Trli et Trhe, : 1= de 0 à 5%, 2= de 5 à 10%, 3= 10 à 25%, 4= de 25 à 50%, 5= de 50 à 100%/ Patu, Ince et acag: 1= activité minime, 2= légère activité, 3= activité moyenne, 4= activité très importante/ Sali: 1= <1 mS/cm à 25°C, 2= 1-2 mS/cm à 25°C, 3= 2-4 mS/cm à 25°C, 4= 4-8 mS/cm à 25°C. Type de sol selon la carte de AJCI (1985) :1= sols halomorphes, 2= sols peu évolués, 3= sols hydromorphes].

Fabaceae était la plus importante en termes de nombre d'espèces et constitue 15.49 % des plantes identifiées (35 espèces), suivie par les *Asteraceae* avec 15.04 % (34 espèces) et les *Poaceae* avec 9,29 % (21 espèces). Ces trois familles représentent à elles seules plus d'un tiers de la flore étudiée. À côté d'elles, les *Apiaceae* (14 espèces, 6,19 %), les *Caryophylaceae* (8 espèces, 3,54 %) et les *Plantaginaceae* (8 espèces, 3,54 %) étaient assez bien représentées. Le reste des familles étaient le plus souvent monospécifique ou bien bispécifique.

La majorité des plantes recensées se concentraient dans les parties Sud et Nord autour du lac avec respectivement 193 et 207 espèces. Ces parties sont les moins touchées par le pâturage et les forts taux de salinité des sols, contrairement aux parties Est et Ouest qui

comptabilisaient beaucoup moins d'espèces (140 et 123 espèces respectivement). Un manque d'abondance a été aussi remarqué. Le nombre de taxons groupés dans chaque relevé pouvait aller de 30 à 121.

Les zones inondées se distinguent par une végétation très pauvre organisée autour des halophytes et des hélophytes (*Sarcocornia fruticosa* (L.) A. J. Scott, *Juncus acutus* L., *Cotula coronopifolia* L., *Cyperus rotundus* L. subsp. *rotundus*, *Spergula arvensis* L. et *Juncus tenageia* L. f. subsp. *tenageia*).

Trois types de milieux ont été définis sur des critères physionomiques liés à la profondeur de l'eau et au type de végétation :

- lac semi-permanent, correspondant à un plan d'eau ne s'asséchant pas totalement en période estivale. Les espèces caractéristiques sont *Ranunculus aquatilis* L., *Alisma plantago-aquatica* L., *Cotula coronopifolia* L., *Rumex crispus* L., *Myriophyllum alterniflorum* DC., *Schoenoplectus lacustris* (L.) Palla*, Lythrum junceum* Banks & Sol., *Sarcocornia fruticosa* (L.) A. J. Scott et *Juncus bufonius* L. subsp. *bufonius*.

- les prairies amphibies, correspondant à des plans d'eau peu profonds, entièrement couverts par une végétation hélophytique caractérisée par une alternance de phases sèches et inondées au cours du cycle annuel; la durée de la phase inondée est généralement supérieure à celle de la phase sèche. Nous distinguons des prairies à *Ranunculus aquatilis* L., à *Callitriche obtusangula* Le Gall et à *Alisma plantago-aquatica* L.

- les pelouses sèches développées en bordure de lac et de prairies amphibies et caractérisées par une végétation à dominance d'espèces thérophytiques (*Bellis annua* L. subsp. *annua*, *Ammi majus* L., *Daucus carota* subsp. *maximus* (Desf.) Ball, *Euphorbia helioscopia* subsp. *helioscopia*, *Medicago murex* Willd. et *Melilotus indicus* (L.) All.).

Type biologique

Les thérophytes ont été nettement le type biologique le plus abondant, représenté par 102 espèces ce qui constitue 45,13% de l'ensemble des taxons répertoriés, viennent ensuite les hémicryptophytes (60 taxons), les géophytes (27 taxons), les phanérophytes (16 taxons), les hydrophytes (13 taxons), les chamaephytes (4 taxons) et enfin les hélophytes (4 taxons).

De plus, la composition floristique a permis de distinguer des groupements végétaux aquatiques et amphibies:

- les hydrophytes représentées par: *Juncus heterophyllus* Dufour, *Myriophyllum alterniflorum* DC. et l'espèce la plus abondante *Callitriche obtusangula* Le Gall.

- les amphipytes sont largement représentées avec par exemple *Cyperus longus*; *Phragmites australis* et *Typha domingensis*, ce dernier macrophyte est le plus abondant, il a été observé sur les deux rives du lac aussi bien en milieu exondé qu'en milieu inondé.

- les hygrophytes sont représentées par *Tamarix gallica*, *Lythrum junceum* Banks & Sol. et *Mentha pulegium* L.

Type biogéographique

Les espèces recensées appartiennent à plusieurs ensembles chorologiques (Supplément Électronique 1):

- Ensemble méditerranéen : cet ensemble domine avec 128 espèces, soit 56,64 % de la flore répertoriée, dont 90 pour l'élément de liaison méditerranéen (*sensu stricto),* 26 pour

l'élément de liaison eury-méditerranéen et 12 pour l'élément de liaison méditerranéen atlantique. Dans cet ensemble, les familles les plus riches sont celles qui sont le mieux représentées dans la flore étudiée. La famille des *Fabaceae* compte 21 taxons, celles des *Asteraceae* 25 taxons, les *Poaceae* (8 taxons) et les *Apiaceae* (8 taxons). D'autres familles possèdent 4 voire 1 taxon.

- Ensemble de large répartition : cet ensemble regroupe 38 espèces, soit 16,81% de la flore du lac Fetzara. Il est présenté par 35 taxons cosmopolites (inclues les subcosmopolites) répartis en 25 familles et trois taxons d'origine tropicale (*Equisetum ramosissimum* Desf. et *Cyperus rotundus* L. subsp. *rotundus* et *Panicum repens* L.).

- Ensemble nordique : ces espèces représentent 15,04 % de la flore étudiée (34 taxons). L'élément paléotempéré est représenté par 24 taxons, suivi par l'élément eurasien avec 5 taxons. L'élément holarctique par 4 taxons, l'élément eurosibérien n'est représenté que par un seul taxon.

- Ensemble d'espèces introduites : cet ensemble est représenté par 7 espèces dont 3 sont cultivées : *Opuntia maxima* Mill., *Triticum durum* Desf. et *Vicia faba* L.

- Ensemble endémique : 19 espèces représentent cet ensemble soit 8,41 % de la flore inventoriée ; une part non négligeable qui apporte de l'intérêt à notre étude, d'autant plus avec la présence de deux endémiques algériennes (*Genista numidica* Spach subsp. *numidica* et *Carduus numidicus* Durieu). Sur les 166 genres présents, dix-sept d'entre eux possèdent des taxa endémiques. Les genres *Genista* et *Scrophularia* sont les seuls à être représenté par 2 taxons. Les pourcentages d'endémiques se répartissent particulièrement parmi les familles *Asteraceae*, *Apiaceae* et *Fabaceae*.

Rareté et endémisme

La flore rare de la région d'étude compte dix neuf espèces (*sensu* Quézel & Santa 1962-1963), parmi lesquelles, 4 se retrouvent sur la liste rouge de l'Union Internationale pour la Conservation de la Nature (UICN) avec des différents statuts : Quasi menacée pour *Bellis prostrata* Pomel, *Convolvulus durandoi*, *Juncus heterophyllus* et *Scrophularia tenuipes*. En outre, quatre espèces sont protégées selon la législation algérienne (Décret exécutif « DE » n°12/03 du 4 janvier 2012 fixant la liste des espèces végétales non cultivées protégées qui en comporte 449): *Bellis prostrata* Pomel, *Convolvulus durandoi* Pomel, *Ludwigia palustris* (L.) Elliott et *Scrophularia tenuipes* Cosson & Durieu (Tab. 2).

Les espèces rares n'ont pas toujours la même valeur patrimoniale. Certaines d'entre elles sont à la fois endémiques et rares, comme (*Convolvulus durandoi* et *Scrophularia tenuipes*).

En ce qui concerne les catégories d'endémisme, il est à noter le taux remarquable de rareté des espèces endémiques algéro-tunisiennes (50%).

Nos résultats révèlent que les milieux les plus vastes (lac) sont les plus pauvres en espèces rares et endémiques. Bien que, les prairies amphibiesrenferment le plus grand nombre en taxons rares (11 espèces) et les pelouses sèchesse distinguent par une diversité en endémisme (17 taxons).

Caractéristiques édaphiques

Les résultats obtenus par l'analyse granulométrique montrent l'importance de teneur en argile (51,55 à 76,05%) et une moyenne du total des différentes fractions granulométriques

Tableau 2. Les plantes à valeur patrimoniale.

Taxon (Dobignard & Chatelain 2010-2013)	Rareté (*sensu* Q.S. 1962-63)	Endém	JORA (2012)	UICN (2019)	Site
Bellis prostrata Pomel	RR		P	NT	PA
Carduus numidicus Durieu	R	Alg			PS
Cladium mariscus (L.) Pohl	RR				L
Convolvulus durandoi Pomel	R	Alg-Tun	P	NT	PS
Daucus virgatus (Poir.) Maire	R	Alg-Tun			PS
Eleocharis uniglumis (Link.) Schult.	R				PA
Eryngium pusillum L.	R				PA
Galactites mutabilis Durieu	AR	Alg-Tun			PS
Helosciadium crassipes W.D.J. Koch.	RR				PA
Illecebrum verticillatum L.	RR				PA
Juncus heterophyllus L. M. Dufour	R			NT	PA, L
Linum numidicum Murbeck	R	Alg-Tun			PS
Ludwigia palustris (L.) Elliott	RR		P		L
Myriophyllum alterniflorum DC.	R				L
Potamogeton trichoides Cham. & Schltdl.	AR				L, PA
Rumex crispus L.	R				PA
Salsola soda L.	RR				PS, PA
Scrophularia laevigata Vahl subsp. *laevigata*	R	Alg-Tun-Mar			PA
Scrophularia tenuipes Cosson & Durieu	R	Alg-Tun-Mar	P	NT	PA

(Endém : endémisme; AR: assez rare; R: rare; RR: très rare; NT: quasi menacée; P: protégé, L: lac semi-permanant, PA: prairies amphibies, PS: pelouse sèche).

de 62,77% ce qui confère aux sols des sites une texture argileuse (Tab. 3). Le pH est légèrement alcalin à alcalin dans l'ensemble des stations étudiées.

La conductivité électrique est de moyenne de 0,58 mS/cm dans le transect 1, dans le transect 2, elle est de moyenne de 0,83 mS/cm et dans le transect 3, elle atteigne 2,72 mS/cm de moyenne. Le transect 4 est le plus salé avec 3,96mS/cm. Ces résultats montrent que les sols de lac Fetzara sont salés à extrêmement salés.

Le taux de calcaire total reste faible dans l'ensemble dans le transect de nord et d'ouest. Dans le transect Sud et Est les teneurs de calcaire sont très élevés. Ils se situent dans la fourchette de 12,13 à 15,42%. Ce taux élevé est en relation avec l'écoulement de l'eau de Oued El Hout de coté de Sud et le canal de drainage de lac de coté de l'Est.

Celui de la matière organique est relativement faible dans la partie inondée de lac. Cette pauvreté qui peut s'expliquer par le faible taux de recouvrement par la végétation.

Tableau 3. Caractéristiques physico-chimiques du terrain des stations étudiées.

Station	pH	C E mS/cm	Granulométrie			M O (%)	CaCO3 (%)
			A (%)	L(%)	S(%)		
N1	7,81	0,19	76,05	22,91	6,14	3,15	0,67
N2	7,85	0,29	71,12	15,45	13,43	2,24	0,71
N3	7,88	0,37	69,75	15,32	14,93	1,92	0,77
N4	7,88	0,74	71,85	15,10	13,05	1,74	0,89
N5	8,13	1,27	68,77	14,12	17,11	1,22	1,48
S6	8,12	0,16	68,94	17,13	14,05	3,42	12,13
S7	8,17	0,22	70,42	15,22	14,36	3,27	13,42
S8	8,17	0,48	63,45	20,12	16,43	3,22	13,72
S9	8,22	1,22	71,12	12,13	16,75	2,87	15,15
S10	8,25	2,11	57,88	17,31	24,81	2,55	15,32
E11	8,37	2,57	52,17	19,51	28,32	1,49	12,22
E12	8,15	2,42	55,42	18,19	26,39	1,24	13,24
E13	8,15	2,53	54,12	18,77	27,11	1,22	14,25
E14	8,12	2,77	53,74	18,54	27,72	0,77	14,74
E15	8,24	2,84	51,55	19,41	29,04	0,61	15,42
W16	8,14	2,27	60,17	11,71	28,12	1,51	2,15
W17	8,12	3,55	63,03	9,66	27,31	1,22	2,27
W18	8,29	4,12	61,71	10,15	28,14	0,81	2,21
W19	8,33	4,48	60,45	11,89	27,66	0,73	2,21
W20	8,27	5,15	57,58	14,11	28,31	0,48	2,47

A = Argile ; L = Limon ; S = Sable ; pH = Potentiel hydrogène ; C E = Conductivité électrique ; CaCO3 = Calcaire total ; M O = Matière organique.

Analyse canonique des correspondances flore / environnement

L'analyse canonique des correspondances flore / environnement met en relation les 226 espèces végétales recensées et les variables environnementales. Le plan formé par le premier et le second axe totalise un taux d'inertie de 67,48% (Fig. 2). Il met en évidence la répartition des relevés des différentes stations en fonction des variables du milieu et de la physionomie de la végétation.

Le long de l'axe 1, les stations s'organisent selon un gradient de salinité, en allant des formations moins salées (côté négatif) vers les formations salées (côté positif).

Les relevés (N1, S6, S7, N3 et E11) qui se regroupent autour de variable taux de recouvrement des herbacées dominés par *Trifolium angustifolium* L., *Veronica cymbalaria* Bodard, *Trifolium scabrum* L., *Aegilops triuncialis* L., *Lagurus ovatus* L., *Daucus carota* subsp. *maximus* (Desf.) Ball, *Chamaemelum fuscatum* (Brot.) Vasc., *Trifolium stellatum* L.). De même, la présence des plantes adventices (*Vicia faba* L, *Triticum durum* Desf. et *Vitis vinifera* L.) sont le témoigne de l'activité agricole sur ces grandes formations.

Dans la partie positive de l'axe 1, les relevés d'Ouest du lac se regroupent autour des variables exposition et salinité. Ils se caractérisent par des halophytes (*Salsola soda* L.et

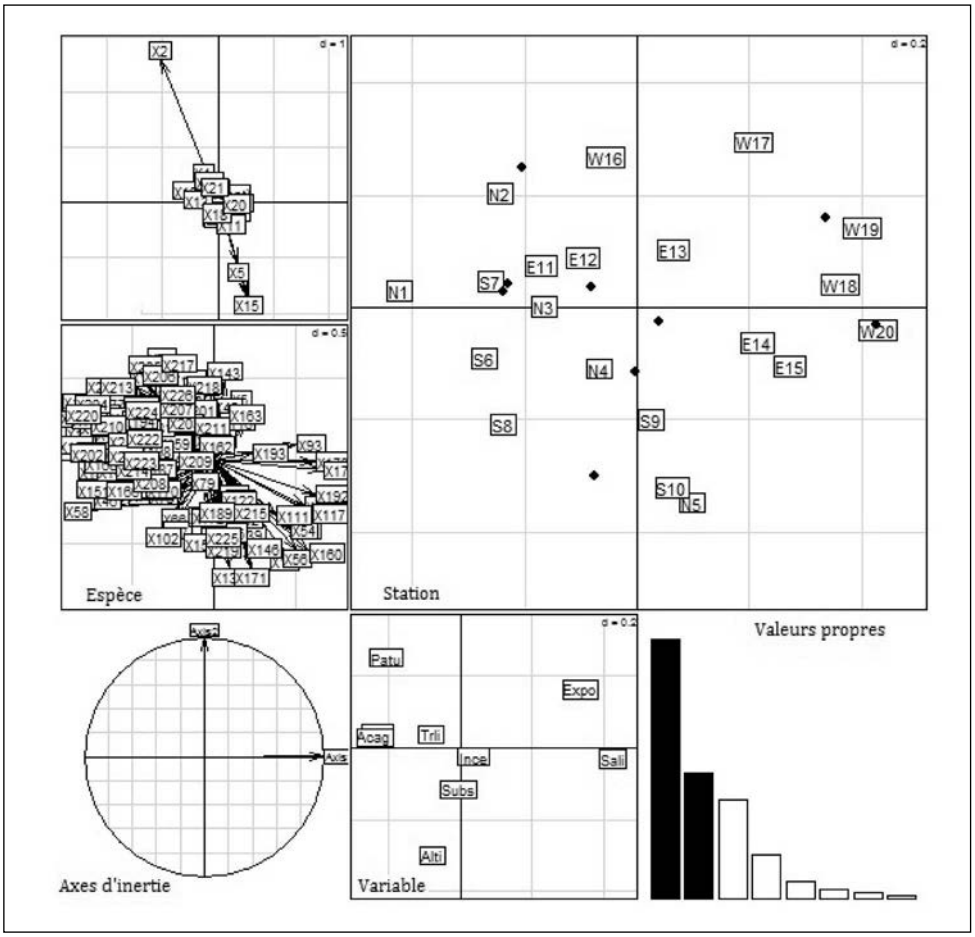

Fig. 2. Carte de l'Analyse Canonique des Correspondances (ACC) 20 stations x 211 espèces x 8 variables environnementales (Expo= exposition, Alti= altitude, Trli= taux de recouvrement des ligneuses, Trhe= taux de recouvrement des herbacées, PaIn= pâturage et incendie, Acag= activité agricole, Sali= salinité des sols, Subs= substrat).

Sarcocornia fruticosa (L.) A. J. Scott.). Ces relevés occupent les terrains argilo-sablo-limoneux et se distinguent aussi par des espèces hélophytes (*Isoetes histrix* Durieu ex Bory, *Spergularia media* (L.) C. Presl, *Spergula arvensis* L., *Lythrum junceum* Banks & Sol., *Ranunculus trilobus* Desf., *Lotus corniculatus* subsp. *preslii* (Ten.) P. Four.).

La variable pâturage est positivement corrélée à l'axe 2 avec un coefficient de corrélation de -0,98, et s'oppose aux variables topographiques substrat (Subs) et altitude (Alti) négativement corrélées à cet axe (r = - 0,47, - 0,88 respectivement).

Les groupes de relevés acquis dans le plan F2 (27,68 %) mettent en évidence une zonation de la végétation qui est tributaire de l'action de différents facteurs primordiaux. Sur la

base de la valeur écologique de ces espèces, le second axe indique la submersion du fait que, sur le côté positif, s'ordonnent des espèces caractéristiques des cuvettes et/ou des zones toujours inondées (*Rumex crispus* L.*, Veronica anagallis-aquatica* L. subsp. *aquatica*, *Zannichellia palustris* (L.) subsp. *palustris* et *Epilobium tetragonum* L. subsp. *tetragonum*). Cette formation s'installe sur des terrains argilo-limono-sableux.

Alors que, sur l'autre côté, se réunissent des espèces des pelouses sèches (*Paronychia argentea* Lam., *Verbascum sinuatum* L., *Urospermum dalechampii* (L.) F. W. Schmidt, *Trachynia distachya* (L.) Link et *Trifolium resupinatum* L.). Cette formation dégradée forme des parcours de grand nombre des cheptels ovins et caprins. Elle se traduit par la dominance des espèces annuelles héliophiles qui remplacent des espèces pérennes.

Discussion

Composante biologique

Les thérophytes (102 taxons) composent principalement le spectre biologique autour du lac Fetzara. La dominance des espèces annuelles traduit l'adaptation des communautés à l'imprévisibilité des conditions environnementales (Deil 2005 ; Williams 2006 ; Megharbi & al. 2016), en favorisant les espèces à cycle court qui investissent plus dans la reproduction sexuée que dans le développement végétatif (Rhazi & al. 2006).

Les hémicryptophytes sont aussi assez bien représentés (60 espèces). Les milieux humides sont en général favorables à leur prolifération (Belouahem & de Bélair 2009). Cette richesse peut en outre s'expliquer par l'importance des mycorhizes dans le sol (Whigham 2004).

Ainsi, la présence de nombreuses hélophytes et hydrophytes ajoutée aux types biologiques précédents vient souligner le fait d'un caractère commun qui réside dans leur exigence en eau. C'est le facteur qui fournit le milieu ambiant lui-même et qui assure les importants besoins en eau de l'appareil végétatif pour les différentes plantes hygrophiles (Belouahem-Abed & al. 2011 ; Allem & al. 2017).

Le lac n'est inondé que la moitié de l'année lors des saisons pluvieuses, cet habitat à caractère semi permanent d'immersion est propice au développement de plantes à cycle court qui croissent rapidement (Hammada & al. 2004), comme peuvent l'être essentiellement les thérophytes. Elles survivent à la sécheresse estivale sous la forme de graines. Ceci peut expliquer que les thérophytes aient été les plus abondantes (42,18%), leur abondance est donc considérée comme une stratégie d'esquive aux périodes défavorables (Daget 1980).

Bon nombre de chamaephytes sont présents. Le pâturage favorise d'une manière globale ces espèces souvent refusées par le troupeau (Benabadji & al. 2004). De plus, ce sont des espèces bien adaptées à la salinité des sols par rapport aux phanérophytes (Ghezlaoui & al. 2011).

Les géophytes et les phanérophytes s'imposent beaucoup moins, Barbéro & Quézel (1989) signalent qu'ils régressent et disparaissent dans les pelouses.

D'après nos prospections, le lac Fetzara abrite en effet 13 hydrophytes, comprenant de nombreuses espèces peu répandues, rares ou très rares (7 espèces), dont une a été observée pour la première fois à Annaba (ex Bône) dans le cadre de cette étude (*Ludwigia palustris* (L.) Elliott). Cette végétation héberge des espèces rares à haute valeur patrimoniale (Hamel & al. 2013). Toutefois, en termes de richesse spécifique les hydrophytes sont moins impor-

tantes que les xérophytes. Cependant en termes d'abondance, les hydrophytes dominent assez bien notamment dans la zone centrale du lac.

Selon Simonneau (1952) la superficie des lieux humides (lac et prairies amphibies) est inférieure à ce qu'elle devrait être par rapport la surface totale, ceci est dû à la longue durée de la période sèche qui accélère l'évaporation qui favorise l'extension des xérophytes par rapport aux autres espèces.

Diversité phytogéographique

L'examen des principaux types chorologiques rencontrés dans la région d'étude confirme la dominance de l'élément méditerranéen (56,64 %), fait souligné par Quézel (2002) pour l'ensemble des pays de l'Afrique du Nord.

Sur les 32 espèces endémiques citées pour la péninsule de l'Edough (Hamel & al. 2013), nous retrouvons 8 espèces dans la zone d'étude, soit 25%. Les familles les plus riches en espèces endémiques sont celles qui sont le mieux représentées dans la flore du lac Fetzara. Ces résultats corroborent ceux mentionnés par Le Houérou (1995) qui considère que les *Apiaceae* et les *Asteraceae* sont des familles à forte endémicité au Nord de l'Afrique.

Néanmoins, les endémiques se développant dans la région d'étude sont relativement peu nombreux par rapport à ce qui a été observé dans les mares d'Annaba, 25 taxons (Allem & al. 2017). Si, pour des raisons orographiques, les taxons endémiques sont très peu représentés dans le peuplement végétal numidien (Quézel 1964), les taxons d'origine septentrionale semblent constituer par contre une de ses composantes principales (Quézel 2002 ; Véla & Benhouhou 2007).

Le taux de rareté des espèces endémiques recensées est remarquable (7 taxons soit 36,84 %). En effet, plus des trois quarts (77,9 %) des taxons endémiques stricts d'Algérie ou sub-endémiques sont des plantes plus ou moins rares en Algérie (Quézel & Santa 1962-1963), les endémiques plus ou moins communes représentant moins du quart du total (Véla & Benhouhou 2007).

Nos données sur la répartition des plantes rares et endémiques dans la région d'étude montrent que la communauté halophile est la plus pauvre. Ce résultat confirme des données déjà obtenues en Algérie (Megharbi & al. 2016), en Tunisie (Ferchichi-Ben Jamaa & al. 2010 ; Domina & El Mokni 2019) et au Maroc (Rhazi & al. 2001).

Mosaïque de la végétation

La diversité floristique du lac Fetzara est comparable à celle des grandes zones humides emblématiques d'Afrique du Nord, telles que le lac Tonga (Kadid & al. 2007), les mares d'Annaba (Allem & al. 2017), au lac Beni-Bélaid (Bouldjedri & al. 2011) et au complexe de zones humides de Guerbès-Senhadja (Samraoui & de Bélair 1997).

Au total, avec dix neuf taxons rares au sens large qu'elle abrite, la région d'étude est faite partie d'une zone réputée pour la richesse et l'originalité de sa flore rare et endémique (Quézel 2002 ; de Bélair 2005 ; Belouahem-Abed & al. 2011 ; Hamel & al. 2013 ; Allem & al. 2017 ; Hamel & Boulemtafes 2017).

Une telle valeur conforte la position du territoire phytogéographique la Numidie au sein du point chaud « Kabylies-Numidie-Kroumirie » (Véla & Benhouhou 2007), où des mesures conservatoires doivent être mises en œuvre pour la protection de ces habitats réputés pour leur fragilité.

Cependant, contrairement à ces sites protégés Ramsar, la lac Fetzara est totalement dégradé et cela est expliqué par l'ignorance des autorités et les organisations environnementales. Bien que très perturbé, ce lac affiche toujours une structure zonale singulière pour un environnement de cette taille. Chacune de ses trois ceintures concentriques à des communautés végétales distinctes: pelouses sèches, prairies amphibies et communautés immergées.

Cette organisation spatiale de la végétation du lac révèle clairement un gradient hydrologique lié à la topographie, qui influence la durée de submersion et la granulométrie des sédiments (granulométrie décroissante de la périphérie vers le centre (Megharbi & al. 2016). Toutefois, les résultats obtenus traduisent l'influence du pâturage. Ce facteur apparaît notamment réduire l'hétérogénéité biocoenotique des ceintures externes, probablement par le tassement et l'homogénéisation du substrat, et augmenter celle des ceintures centrales, par réduction de la compétition. Il contribue en effet à limiter le développement des hélophytes, et notamment des espèces vivaces rhizomateuses très compétitives (Ferchichi-Ben Jamaa & al. 2014).

Nos données suggèrent ces effets en montrant que les pelouses sèches présentent significativement plus d'espèces annuelles que les autres types d'habitats. La moindre concurrence induite par les herbivores domestiques pourrait constituer un outil de gestion et de conservation de la biodiversité, de manière à maintenir des zones ouvertes favorables aux communautés thérophytiques patrimoniales des zones humides (Gordon & al. 1990). La présente étude montre qu'au lac Fetzara, les champs pâturés semblent contenir beaucoup plus d'espèces que les marais temporaires et les cultures inondées et confirme les conclusions de Bouldjedri & al. (2011) et de Allem & al. (2017).

Les résultats des analyses pédologiques révèlent les interférences de plusieurs paramètres que ce soit le degré de l'holomorphie et de l'hydromorphie. La basse altitude de la région d'étude est bien corrélée avec le taux de salinité. Cette salinité exerce une certaine influence sur le développement de la végétation (Djebaili 1978).

En revanche, les stations inondées sont les plus pauvres en espèces. Ces espèces dépendent de l'interaction des propriétés du sol (Conductivité électrique, calcaire total et matière organique). Dans leur étude sur les marais du Maryland, Darmody & Foss (1979) ont constaté qu'une augmentation de la teneur en sel réduirait la richesse floristique.

L'ACC sépare la végétation halophile installée sur un sol argilo-sablo-limoneux aux autres végétations, installées sur un sol argilo-limono-sableux. Les niveaux les plus élevés de la salinité du sol ont été observés dans les zones occupées par *Sarcocornia fruticosa* (L.) A. J. Scott. et *Salsola Soda* L. Ces deux espèces appartenant à un groupe écologiquement hygro-euhalophytique constituent ici deux groupements distincts dans les zones alluviales. Leur distribution suit généralement le taux des alluvions qui dépend de la durée de la période humide et de la profondeur des eaux (Khani & al. 2002).

Cependant, cette approche statististique montre que la salinité des sols est un facteur de répartition des especes autour du lac Fetzara. Nos résultats correspondent à ce qui a été observé dans les travaux de Pavoine & al. (2011) sur les liens entre les variables environnmentales et les communautés végétales à la Mafragh (Nord-est algérien).

Néanmoins, le complexe argilo-humique du sol joue un rôle major sur la salinité du lac. Il fixe les ions de Na^+ et K^+, ceci devient suffisant pour inverser l'alcalinité qui devienne négative à mesure que les solutions de sols se concentrent, conduisant l'évolution géochi-

mique des sols vers la voie saline. Ce phénomène est assez fréquent en Afrique du Nord (Sitayeb & al. 2008, Ouali & al. 2014, Megharbi & al. 2016).

Le groupement de *Juncus acutus* L., *Ranunculus aquatilis* L. et *Cyperus rotundus* L. subsp. *rotundus* a une large tolérance vis-à-vis de la salinité et une forte vulnérabilité au stress d'inondation. Il s'étend sur les milieux argileux-sableux jusqu'à l'apparition des prairies saumâtres. Cette extension est due au régime des eaux qui est considéré comme le facteur principal influençant le développement de la végétation des zones humides (Zouaidia & al. 2015).

Le groupement à *Juncus bufonius* L. subsp. *bufonius, Isoetes histrix* Durieu ex Bory, *Damasonium alisma* subsp. *bourgaei* (Coss.) Maire et *Alisma plantago-aquatica* L. occupe partiellement ou totalement les sols inondés en hiver.

Le groupement de pelouse, qui se trouve sur des substrats en faible hydromorphie et en salinité très faible, occupe la plus grande partie des terrains de lac Fetzara et forme des parcours assez importants pour l'agriculture (Djamai 2007).

Menaces et conservation

Les effets de l'urbanisation sur la zone humide sont nombreux, les rejets urbains de la nouvelle ville « Draa Errich » seront la principale cause de sa dégradation actuelle et future (Mellouk & Aroua 2015).

Les pompages intensifs, associés à la construction de barrages en amont des oueds (Oued El Hout et Oued El Mellah), sont susceptibles de modifier à court terme l'hydrologie de la plaine alluviale et de la zone humide, d'affecter le transport et le dépôt des alluvions par les cours d'eau, et d'entrainer une salinisation des sols (Djamaï 2007). De tels changements ont été mis en évidence dans la zone humide de la Macta sur l'ouest algérien (Belgherbi 2011 ; Megharbi & al. 2016) où les modifications des conditions environnementales ont entraîné des modifications de la salinité, et par voie de conséquence des cortèges floristiques (Megharbi & al. 2016) et faunistique (Ledant & Van Dijk 1977). L'influence du pâturage est également révélée dans le lac Fetzara par l'absence de stratification verticale, la petite taille des plantes, la présence de plages de sol nu et l'abondance, dans les pelouses environnantes, d'*Asphodelus ramosus* L. subsp. *ramosus*, connue pour être indicatrice de surpâturage (Pantis & Mardiris 1992). Ce qui signifie une évolution progressive du pâturage qui conduit selon Bullock & al. (2001) à un remplacement des espèces pérennes de haute taille par des annuelles avec des formes de vie variées. La protection du lac Fetzara implique, dans un premier temps, de le préserver de l'influence directe des cultures par la création d'une ceinture boisée (Brian & al. 2004). Cette ceinture pourrait également servir de barrière à la pénétration des espèces exotiques potentiellement envahissantes (Houlahan & al. 2006 ; Hamel & Azzouz 2018) et favoriser le maintien de la macrofaune (oiseaux) inféodée aux habitats riverains. Cette mesure de conservation devrait en outre impérativement être accompagnée d'une campagne de sensibilisation des populations locales sur l'intérêt de la conservation des milieux naturels, en vue de leur implication dans la gestion des sites (Allem & al. 2017).

Conclusion

La présente étude de la diversité écologique de lac Fetzara fait état de la présence de deux centvingt-six espèces de plantes appartenant à cent soixante-six genres et cinquante-huit familles botaniques et révèle donc une assez une grande richesse floristique dont un nombre important d'espèces thérophytes. Cette zone humide abrite dix neuf taxons endémiques, lui conférant une importance particulière en termes de conservation de la diversité génétique des espèces considérées. La forte pression anthropozoïque, notamment le pâturage, les incendies et les pompages intensifs de l'eau entrainant la salinisation des sols influencent négativement la flore du lac Fetzara et rend hypothétique son maintien à long terme. Il semble donc nécessaire de préserver ces zones contre le pâturage en l'intégrant par exemple durant certaines périodes de l'année dans certaines zones afin d'y maintenir un régime intermédiaire de perturbation pour rétablir l'équilibre naturel et lutter contre le phénomène de dégradation. Il faut aussi instaurer des mesures de protection de la région d'étude en encadrant les pratiques agricoles autour du lac qui peuvent porter atteinte à certaines espèces et leur habitat.

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Article

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Mehdi Trad & Mounira Harbi

Towards a bio-morphometric approach for the discrimination between wild and domesticated vines under Mediterranean environment

Abstract

Trad, M. & Harbi, M.: Towards a bio-morphometric approach for the discrimination between wild and domesticated vines under Mediterranean environment. — Fl. Medit. 29: 247-262. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

All the traditional vines, constituting the subspecies *Vitis vinifera* subsp. *vinifera*, were domesticated from the wild *V. vinifera* subsp. *silvestris*. Shape and morphometry of the seed provides information to differentiate between wild and cultivated vine populations. Measurements were made on grape seeds for the assessment of genetic diversity and for the discrimination between the two subspecies according to the Stummer index. Morphometric analysis was carried out on 340 seeds from 34 ecotypes (22 wild and 12 cultivated) originated from the north of Tunisia. In the wild vine population, seed length (L) varied between 3.2 and 5.2 mm, beak length (LS) between 0.4 and 1.7 mm and breadth (B) between 1.7 and 3.2 mm. The Stummer index (SI) was comprised between 51.43 and 88.57. Among the twenty-two wild ecotypes, seven confirmed their belonging to the spontaneous morphotype and four had 95% chance to belong to the cultivated morphotype (EC.30, EC.33, CPN5-2000 and CPN6-2000). In the cultivated vine pool, seed length ranged from 3.8 to 5.2 mm, stalk length from 0.8 to 1.4 mm and the total width from 2.3 to 3.2 mm. The Stummer index varied between 56.10 and 65.00. Among the twelve cultivars, five confirmed their belonging to the cultivated morphotype and three showed 95% chance to belong to the spontaneous type (Bidh El Hmem, Chaaraoui and Meski Rafraf). The results allowed us to relate a probable genetic proximity between spontaneous and some cultivated vines and to retrace the evolution pathway of local vine gene pool.

Key words: Discriminant function analysis, genetic variability, morphometry, plant selection; Stummer index; *Vitis vinifera*.

Introduction

Most botanists regard the wild ancestral grape *V. silvestris* as the primitive form of the cultivated grape because of the close morphological resemblance and free gene flow between them (Heywood & Zohary 1991). Since the 19th century, many researches have been carried out to clarify the evolution of species in agriculture (Zohary & Spiegel-Roy 1975; Zohary & Hopf 1988; Mc Govern & al. 1996; Terral & al. 2010).

Grapevine is one of the most important crops in the Mediterranean region and has been the subject of a number of studies aiming to discriminate between wild and cultivated

vines. Discrimination between wild and cultivated accessions used biometric approach based on the length/width ratio of the seed. This ratio, designated by the Stummer index (Stummer 1911), was the basis of the work of Marinval (1988) to establish a date of appearance of viticulture in the second Iron Age (Marinval 1988; Buxo & Capdevila 1996; Bouby & Marinval 2001). Since the work of Stummer (1911) it has been generally accepted that domestication has caused changes in the dimensions and some other morphological characteristics of the seed, so that in general it was possible to distinguish cultivated from wild vines. Stummer (1911) first and Mangafa & Kotsakis (1996) later, described seeds of wild vines as small, robust, with a rounded outline, or cordate, with short stalks. They were almost flat ventrally with sharp angles and a strongly developed chalaza. On the other hand, seeds of domesticated vines were described as large, elongated, oval or pyriform, with a longer stalk. They were more rounded ventrally and less sharply sculptured. Schieman (1953) has completed the Stummer studies by reporting that the width/length index varies with the grape variety. In the last few years, several studies have been carried out using the berry seed of the vine to classify it as cultivated or spontaneous. This provides proposals and hypotheses on vine evolution and phyllogenetic relationships between cultivated and spontaneous populations (El Oualkadi & al. 2011; Ucchesu & al. 2016; Ardenghi & al. 2017). For the description of the corpus of the seed, these researches considered qualitative criteria primarily related to the stalk, the chalaza or the fossete. Mangafa and Kotsakis (1996) proposed four formulas following the application of the four-dimensional measurement of the seed. The effectiveness of each formula varies from one vine to another. These formulas show good aptitude to recognize the seeds of the cultivated vines from those of the spontaneous vines while being only exploitable in archeobotany for Greek vines.

Tunisian gene pool of the *Vitis* genus is mostly concentrated in the north side of the country. Despite the abundance of introduced cultivars for commercial purposes, many farmers still preserve autochthonous cultivars with a strong faculty of adaptation to marginal soil and changing climate conditions. Considered as a significant part of the local vine heritage, wild grapevines were encountered in isolated forms or in groupings, in humid locations, along permanent water courses in the down side of hills and mountains showing a large polymorphism and a great morphological variability (Harbi & al. 2010; Trad & al. 2017). Their number remains limited as human activities are rapidly eroding the size of wild grapevine populations. *Vitis vinifera* subsp. *silvestris* Hegi, is a unique and valuable genetic resource for the improvement of cultivated grapevines regarding their genetic tolerance to salinity (Raymond & al. 2008), their resistance to many virus diseases and their high adaptation potential to different soil types and climates (Ocete & al. 1995; Arnold & al. 1998).

In the present work, we propose to test the accuracy and reliability of the two main methods used, namely the Stummer index and the application of the Mangafa and Kotsakis formulas for the characterization of Tunisian autochthonous vine seeds, their shapes and their dimensions, to specify their attribution to either wild (*Vitis vinifera* subsp. *silvestris*) or cultivated (*Vitis vinifera* subsp. *vinifera*) vine. Data results will help to explore the course of development and domestication of grapevines and to retrace the history of viticulture in Tunisia.

Materials and methods

Plant material.- During the two seasons 2014 and 2015, lots of grapes were picked from about 34 vines (22 wild and 12 cultivated) growing under Mediterranean conditions in the north of Tunisia. Grape clusters were collected in different says after anthesis, depending on the geographic origins of the vine plants: Nefza, Outchtata, Tabarka, Ain Draham, Cap Negro and Sejnane for spontaneous vines, and Rafraf for domesticated ones (Fig. 1). The group of local cultivars is mainly table type but also include a dual-use cultivar (Table 1). All these accessions are actually part of the national germplasm collection maintained in the vineyards of the Horticulture laboratory in the National Institute for Agricultural Research (INRAT, Tunisia) and Herbarium specimens have been deposited in the institute. A leaf comparison is provided to show variability between the grapevine genetic resources studied (Fig. 2). Seeds derived from both cultivated and wild vines were examined for their morphometric data.

Seed measurements.- Morphometric analysis covered 340 seeds from the 34 ecotypes. The twelve autochthonous vines are representative of the varietal assortment of local cultivars growing under Mediterranean conditions in the North of Tunisia. Depending on the availability of grape clusters, especially for wild vines, a minimum of 5 bunches per clone were collected. From each vine cep was collected one grape bunch. Seeds were first removed from the berry, dried at room temperature and cleaned finally from pulp trash to finally constitute a batch from which ten seeds per clone were randomly picked. The following measurements were taken: length (L), breadth (B), length of the stalk (LS), placement of chalaza, i.e. the distance from the base of chalaza to the tip of the stalk (PCH) (Fig. 3) (Mangafa & Kotsakis 1996). Then, the following indices were calculated: breath to length (B/L), length of stalk to length (LS/L) and placement of chalaza to length (PCH/L). The diagnostic features of the seeds considered these dimensions as reference to Negrul (1960) who thought that all these parameters are the most important characteristics of the seed. Two morphometric approaches were tested: the Stummer Index (IS) and the four formulas proposed by Mangafa & Kotsakis (1996). These are formulas based on four measurements performed on the grape seed. From these four measurements were calculated:

The Stummer index: SI = B/L 100

An ecotype whose Stummer index varies between 58.37 and 61.89 has 95% chance of belonging to the cultivated type. An ecotype whose Stummer index varies between 62.72 and 68.58 has 95% chance of belonging to the spontaneous type.

The indices: LS/L and PCH/L used in the formulas of Mangafa and Kotsakis according to:

$$Fomula\ 1:\ -0.3801 + (-30.2 \times LS/L) + (0.4564 \times PCH) - (1.386 \times L) + (2.88 \times PCH/L) + (9.4239 \times LS)$$

Values smaller than -0.2 indicate wild seeds and values bigger than 0.8 indicate cultivated ones. Values from -0.2 to 0.2 indicate seeds which have a 64.7% chance of being wild while values from 0.2 to 0.8 indicate seeds which have a 76.2% chance of being cultivated.

$$Formula\ 2:\ 0.2951 + (-12.64 \times PCH/L) - (1.6416 \times L) + (4.5131 \times PCH) + (9.63 \times LS/L)$$

Values smaller than -0.2 indicate wild seeds and values bigger than 0.9 indicate cultivated ones. Values from -0.2 to 0.4 indicate seeds which have a 90.1% chance of being wild while values from 0.4 to 0.9 seeds which have a 63.8% chance of being cultivated.

$$Formula\ 3:\ -7.491 + (1.7715 \times PCH) + (0.49 \times PCH/L) + (9.56 \times LS/L)$$

Values smaller than 0 indicate wild seeds and values bigger than 0.9 indicate cultivated ones. Values from 0 to 0.5 indicate seeds which have a 93.3% chance of being wild while values from 0.5 to 0.9 seeds which have a 63.3% chance of being cultivated.

$$Formula\ 4: 0.7509 + (-1.5748 \times L) + (5.297 \times PCH) - (14.47 \times PCH/L)$$

Values smaller than -0.9 indicate wild seeds and values bigger than 1.4 indicate cultivated ones. Values from -0.9 to 0.2 indicate seeds which have a 91% chance of being wild while values from 0.2 to 1.4 seeds which have a 76.5% chance of being cultivated.

Statistics.- For each variable, means, standard deviation, univariate ANOVA were calculated to assess differences (p <0.05). Discriminant function analysis was considered based on linear combinations of the predictor variables to provide the best discriminations between groups. Wilks' Lambda was used to test for significant differences between groups and summary of canonical discriminant functions was displayed with the eigenvalues and structure matrix. Classification statistics were launched to assess the how well the discriminant function works.

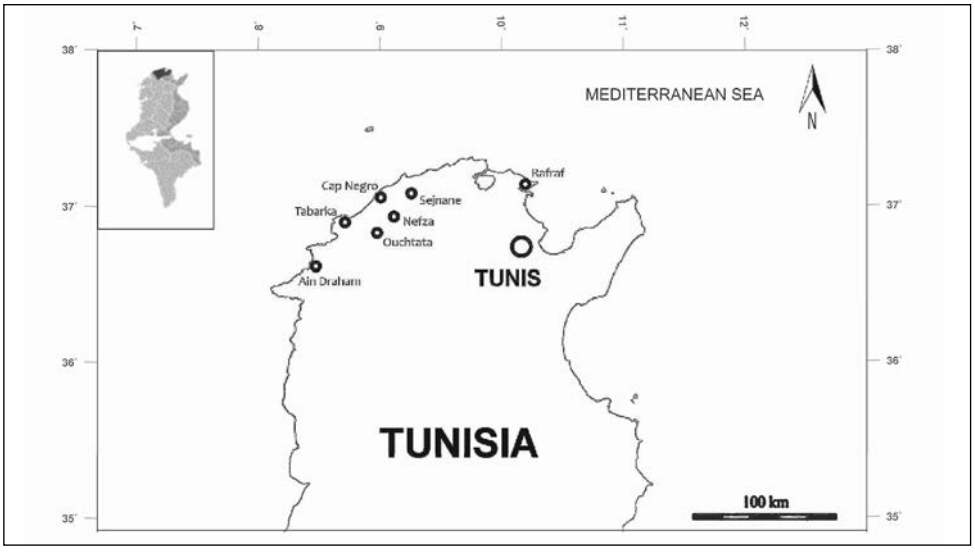

Fig. 1. Origin sites of the wild *Vitis vinifera* subsp. *silvestris* and the cultivated *V. vinifera* subsp. *vinifera* grapes collected from the north of Tunisia.

Table 1. General description and metric data of the seed in wild and cultivated grapes. (Means ± SD). L: total length, LS: length of the beack, PCH: placement of chalaza, B: total breadth, W: weight of 100 seeds. L, LS, PCH and B are expressed in (mm); W is expressed in (g)

				Berry		*Seed*				
Ecotype	Code	Origin	Main use	Shape [a]	Colour [a]	L	LS	PCH	B	W
Wild										
01	EC.01	Nefza	-	Round	Blue-black	3.5 ± 0.2	1.0 ± 0.1	2.1 ± 0.2	1.8 ± 0.1	5.88
03	EC.03	Nefza	-	Round	Blue-black	3.6 ± 0.2	0.5 ± 0.1	1.8 ± 0.1	3.1 ± 0.1	9.70
08	EC.08	Nefza	-	Round	Green-yellow	4.3 ± 0.1	0.9 ± 0.1	2.4 ± 0.1	2.4 ± 0.1	7.61
09	EC.09	Nefza	-	Round	Green-yellow	4.5 ± 0.2	1.0 ± 0.2	2.4 ± 0.1	2.6 ± 0.1	7.43
10	EC.10	Nefza	-	Round	Blue-black	3.8 ± 0.1	0.6 ± 0.1	1.8 ± 0.1	2.1 ± 0.2	8.37
13	EC.13	Nefza	-	Round	Blue-black	4.0 ± 0.2	0.7 ± 0.1	2.0 ± 0.1	2.6 ± 0.2	8.17
14	EC.14	Nefza	-	Round	Blue-black	4.3 ± 0.1	0.8 ± 0.1	1.9 ± 0.1	2.9 ± 0.1	6.57
15	EC.15	Nefza	-	Round	Blue-black	3.8 ± 0.2	0.9 ± 0.2	1.6 ± 0.1	2.5 ± 0.2	7.20
26	EC.26	Ain Draham	-	Round	Dark red-violet	3.5 ± 0.2	0.8 ± 0.1	1.7 ± 0.1	3.1 ± 0.1	7.85
30	EC.30	Tabarka	-	Round	Dark red-violet	4.6 ± 0.2	1.3 ± 0.1	2.4 ± 0.1	2.9 ± 0.2	8.44
33	EC.33	Tabarka	-	Round	Blue-black	4.2 ± 0.2	1.3 ± 0.1	2.3 ± 0.2	2.6 ± 0.1	8.45
35	EC.35	Tabarka	-	Round	Green-yellow	4.4 ± 0.3	1.3 ± 0.3	2.4 ± 0.3	2.8 ± 0.2	5.72
45	EC.45	Sejnane	-	Round	Blue-black	3.9 ± 0.1	0.9 ± 0.1	2.0 ± 0.1	2.7 ± 0.1	5.44
46	EC.46	Sejnane	-	Round	Blue-black	3.3 ± 0.2	0.6 ± 0.2	1.6 ± 0.1	2.6 ± 0.2	6.60
Cap Negro 5-2000	CPN5-2000	Cap Negro	-	Round	Blue-black	4.3 ± 0.2	1.1 ± 0.2	2.3 ± 0.2	2.6 ± 0.1	5.36
Cap Negro 6-2000	CPN6-2000	Cap Negro	-	Round	Blue-black	4.6 ± 0.1	1.4 ± 0.1	2.5 ± 0.1	2.8 ± 0.1	6.13
Cap Negro I	CPN-I	Cap Negro	-	Round	Dark red-violet	3.9 ± 0.2	0.9 ± 0.2	1.9 ± 0.1	2.8 ± 0.2	7.50
Cap Negro III	CPN-III	Cap Negro	-	Round	Dark red-violet	3.8 ± 0.3	1.5 ± 0.1	2.6 ± 0.2	2.4 ± 0.2	7.13
Cap Negro IV	CPN-IV	Cap Negro	-	Round	Dark red-violet	3.9 ± 0.1	0.8 ± 0.1	1.9 ± 0.1	2.7 ± 0.1	7.41
Ouchtata 3	OCT3	Ouchtata	-	Round	Green-yellow	3.7 ± 0.1	0.9 ± 0.1	2.0 ± 0.1	2.4 ± 0.1	6.63
Ouchtata 16	OCT16	Ouchtata	-	Round	Green-yellow	3.7 ± 0.1	0.7 ± 0.1	1.6 ± 0.2	2.6 ± 0.0	6.54
Ouchtata 17-2000	OCT17-2000	Ouchtata	-	Round	Blue-black	3.7 ± 0.1	1.1 ± 0.1	1.4 ± 0.1	2.4 ± 0.1	6.70
Cultivated										
Bidh El Hmem	BHM	Rafraf	Table	Obovate	Green-yellow	4.1 ± 0.1	1.0 ± 0.1	2.0 ± 0.1	2.8 ± 0.2	7.23
Tebourbi	TEB	Rafraf	Table	Narrow elliptic	Green-yellow	4.2 ± 0.1	1.4 ± 0.1	2.6 ± 0.1	2.4 ± 0.1	4.92
Bazzoul Khadem	BZK	Rafraf	Table	Narrow elliptic	Dark red-violet	4.7 ± 0.2	1.3 ± 0.2	2.5 ± 0.1	2.6 ± 0.2	5.11
Meski Rafraf	MKR	Rafraf	Table/wine	Ovate	Green-yellow	4.4 ± 0.1	1.2 ± 0.2	2.4 ± 0.2	2.8 ± 0.1	6.13
Chaaraoui	CHR	Rafraf	Table	Elliptic	Green-yellow	3.7 ± 0.1	1.7 ± 0.1	2.1 ± 0.1	2.4 ± 0.1	4.45
Marsaoui	MRS	Rafraf	Table	Round	Green-yellow	5.0 ± 0.2	1.1 ± 0.1	2.4 ± 0.1	2.9 ± 0.1	9.31
Farrani	FRN	Rafraf	Table	Oblate	Green-yellow	4.3 ± 0.1	1.0 ± 0.6	2.0 ± 0.1	2.6 ± 0.2	6.03
Boukhasla	BKS	Rafraf	Table	Round	Dark red-violet	4.3 ± 0.3	1.4 ± 0.2	2.3 ± 0.2	2.6 ± 0.2	7.67
Beldi	BLD	Rafraf	Table	Round	Green-yellow	3.9 ± 0.1	1.3 ± 0.1	2.1 ± 0.1	2.3 ± 0.1	4.62
Essifi	ESF	Rafraf	Table	Round	Green-yellow	4.1 ± 0.1	1.2 ± 0.1	2.4 ± 0.2	2.3 ± 0.2	6.98
Hammemi	HMM	Rafraf	Table	Obtuse-ovate	Green-yellow	4.1 ± 0.1	1.1 ± 0.1	2.2 ± 0.2	2.5 ± 0.2	7.11
Souabaa Eljia	SBE	Rafraf	Table	Arched	Green-yellow	4.8 ± 0.2	1.3 ± 0.2	2.5 ± 0.2	2.6 ± 0.2	7.04

[a] Descriptors for grapevine (IPGRI, UPOV, OIV 1997)

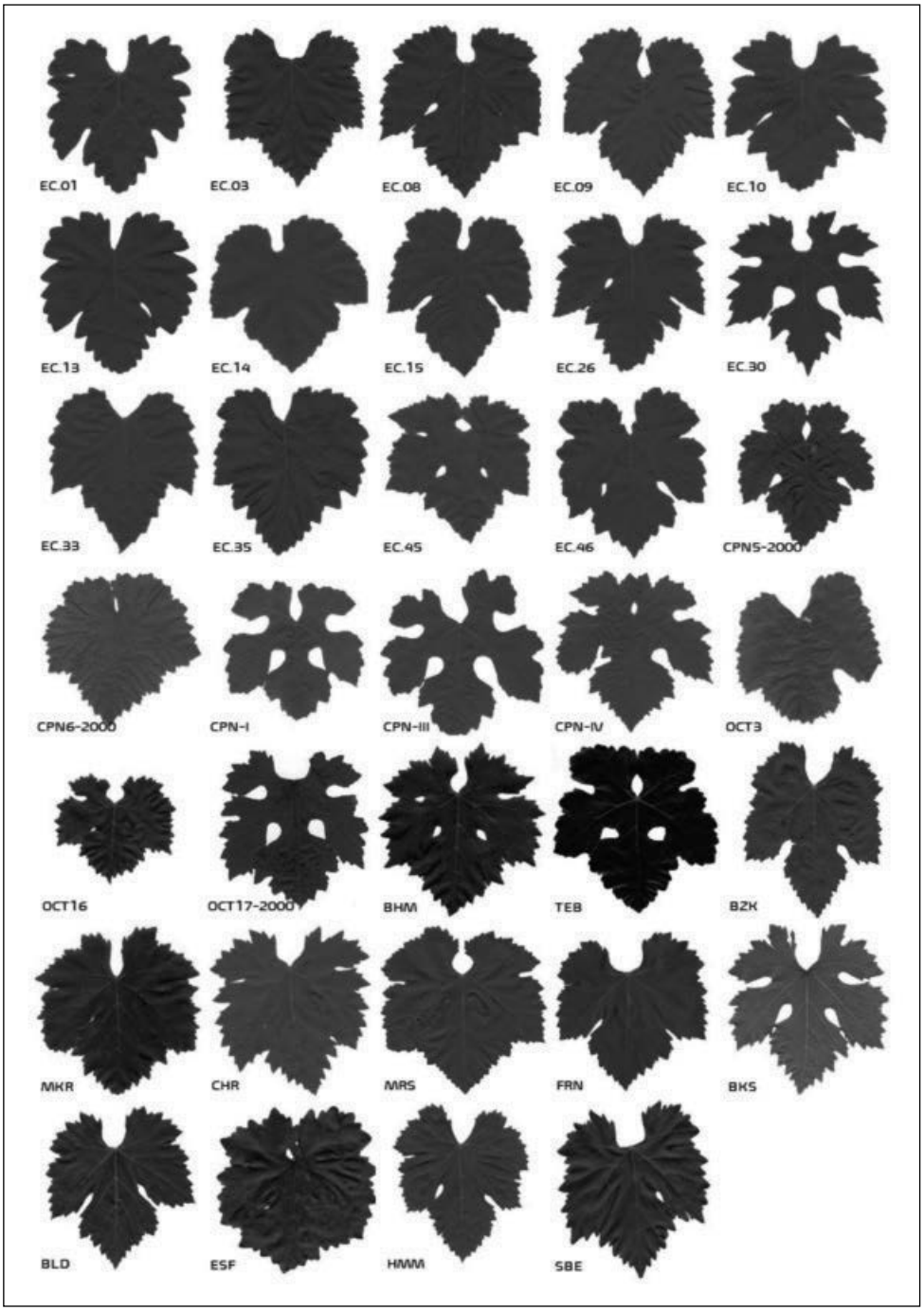

Fig. 2. Leaf variation of the spontaneous and domesticated grapevines composing the vineyard collection of the National Institute of Agricultural Research.

Multivariate analysis was tested using principal component analysis (PCA) to layout groups of vines with homogeneous metric characters of the seed. All these statistics were performed using PC software package SPSS (IBM® SPSS® Statistics, version 20.0.0).

Results

General description of the berry.- Among the 22 spontaneous grapevines, five produce green-yellow berries (Table 1). These ecotypes are originated from Nefza (2), Ouchtata (2) and Tabarka (1). Other ecotypes develop small bunches with round dark coloured berries which correspond to the general feature of wild grapes encountered in other locations from the Mediterranean basin (Levadoux 1956; Ocete & al. 1999; Cunha & al. 2007). The white-berry vines come close to *V. sativa* and could be considered as autochthonous (Levadoux 1956; Imazio & al. 2015). Local grape cultivars developed large berries with a wide range of forms, green yellow coloured for most of them, and are clearly differentiated from wild vines (Table 1). 'Meski Rafraf' is the single cultivar used for dual purposes (table and wine). The other cultivars are mainly for fresh consumption. 'Meski Rafraf' belongs to the group of Muscats. This group is widely distributed in Mediterranean area and well appreciated for the typical berry flavour with high sugar rate and low acid content (Snoussi & al. 2004; Trad & al. 2017). Characters describing the berry can therefore provide a preliminary differentiating criterion between wild and domesticated grapevines.

Seed morphometry of Vitis *grapes*.- The analysis of the 340 seeds from vines growing under Mediterranean environment showed a large variability in shape and size (Table 1). Wild grapes were characterized by spherical seeds with a small beak while cultivated grapes developed pyriform-shaped seed with a well-developed beak (Fig. 3). In wild vines, size of the seed ranged between 3.3 and 4.6 mm in length and between 1.8 and 3.1 mm in breadth. Placement of chalaza ranged between 1.4 and 2.6 mm. In *V. vinifera* subsp. *vinifera*, length of the seed varied between 3.7 and 5.0 mm and the breadth between 2.3 and 2.9 mm. Placement of chalaza varied between 2.0 and 2.6 mm.

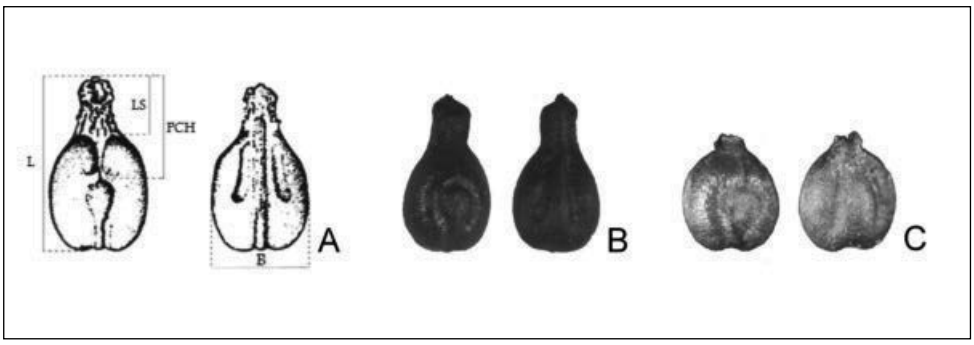

Fig. 3. (**A)** General appearance and dimensions of a grape seed. L: total length, LS: length of the beak, PCH: placement of chalaza, B: total breadth (From Mangafa & Kotsakis 1996). **(B)** Seed from cultivar 'Beldi'. **(C)** Seed from wild 'EC.46'. Left: dorsal side, right: ventral side. (Binocular × 10).

Differences between the 34 genotypes were highly significant ($p < 0.01$) for all metric parameters. The longest seed was found in cultivar 'Marsaoui' while the largest seed was identified in EC.03 and EC.26. The wild 'Cap Negro III' showed the longest beak paradoxically to most other wild genotypes for which the beak is very small. In wild populations, the length of the beak ranged between 0.5 and 1.5 mm, while it was comprised between 1.0 and 1.7 mm in domesticated grapes. Most of the wild ecotypes developed seeds with short beak compared to cultivated vines. The beak is generally considered to be a valuable characteristic, as under the effects of cultivation its length increases proportionally faster than the length of the corpus (Negrul 1960).

The average width/length ratio was 0.66 in wild seeds and reached 0.63 in domesticated grape seeds. The weight, expressed as gram per 100 seeds, was comprised between 5.36 and 9.70 g in wild vines and between 4.45 and 9.31 g in cultivated vines. In general, wild ecotypes produced heavier seeds compared to domesticated grapevines. Disparity between wild and cultivated grapes was particularly observed in seed/berry ratio. This proportion is more significant in wild grapes which produce small berries with a large seed in a tiny pulp.

Discriminant function analysis (DFA).- The test of equality of group means indicates that Stummer index and formula 4 are the most discriminating variables (Table 2). The smaller the Wilks' Lambda is, the more important the independent variable to the discriminant function (Wilks' Lambda = 0.063 and 0.099 respectively for IS and F4). Additionally, Examination of the F test leads to exclude the (F4) formula. F test value is high for Stummer index compared to the other four formulas proposed by Mangafa & Kotsakis (1996). So, the Stummer index seems to have more influence and to provide the best discrimination between wild and cultivated grape genotypes.

The summary of canonical discriminant functions displayed in Table 3 indicates that with eigenvalue of 21.08, most of the variance (67.3 %) in the dependant variable is explained by function 1 which is strongly correlated with the index of Stummer (Table 4). Thus, it becomes possible to formulate the discriminant function (DF) basing on the unstandardized discriminant function coefficients presented in Table 5.

Classification statistics showed that the model adopted correctly classified more than 68% of the genotypes. Overall, 68.6% of the grape genotypes are correctly classified. In charred grape seeds, Mangafa & Kotsakis (1996) recommended the use of the second and third formulae as more appropriate for the identification of archaeological grapevines.

Classification of Vitis *ecotypes*.- In the following, the classification will take into account the Stummer index. The Stummer index (SI) varied from 51.43 to 89.46 (variance = 80.4) for wild grapes while it was between 54.21 and 67.45 (variance = 14.7) for cultivated grapes (Fig. 4). Among the 22 wild ecotypes, seven confirmed their belonging to the spontaneous morphotype (EC.13, EC.14, EC.15, EC.35, CPN-III, OCT3 and OCT17-2000) and four had 95% chance to belong to the cultivated type (EC.30, EC.33, CPN5-2000 and CPN6-2000). The confirmed spontaneous types originated for most of them from Nefza, while the four ecotypes, presumably belonging to the cultivated type, originated from Tabarka and Cap Negro respectively i.e. 34 km away and even closer to the coast.

Table 2. Tests of equality of group means.

	Wilks' Lambda	F	df1	df2	Sig.
Index of Stummer	0.063	30.797	33	68	0.000
Formula 1	0.155	11.248	33	68	0.000
Formula 2	0.152	11.541	33	68	0.000
Formula 3	0.134	13.362	33	68	0.000
Formula 4	0.099	18.681	33	68	0.000

Table 3. Summary of canonical discriminant functions – *Eigenvalues.*

Eigenvalues				
Function	Eigenvalue	% of variance	Cumulative %	Canonical correlation
1	21.09	67.3	67.3	0.977
2	5.00	16.0	83.2	0.913
3	3.03	9.7	92.9	0.867
4	1.73	5.5	98.4	0.796
5	0.50	1.6	100.0	0.572

Table 4. Summary of canonical discriminant functions – *Structure matrix.*

	Function				
	1	**2**	**3**	**4**	**5**
Index of Stummer	0.802*	0.464	0.283	-0.169	0.182
Formula 4	-0.523	0.795*	0.170	0.153	-0.207
Formula 2	-0.398	0.265	0.762*	0.198	0.388
Formula 3	-0.445	0.332	0.748*	0.036	0.363
Formula 1	-0.401	0.480	0.449	0.300	0.564*

Pooled with-in groups correlations between discriminating variables and standardized canonical discriminant functions. Variables ordered by absolute size of correlation within function.
*: largest absolute correlation between each variable and any discriminant function.

Table 5. Canonical discriminant function coefficients.

	Function				
	1	**2**	**3**	**4**	**5**
Index of Stummer	0.336	0.197	0.119	-0.024	-0.011
Formula 1	-0.697	1.531	-3.524	0.341	4.980
Formula 2	3.721	-2.963	6.012	12.734	-6.385
Formula 3	-3.707	1.128	-1.062	-12.240	3.753
Formula 4	-0.487	3.082	0.351	2.066	-4.345
(Constant)	-23.862	-6.031	-7.424	5.246	-2.722

Unstandardized coefficients.

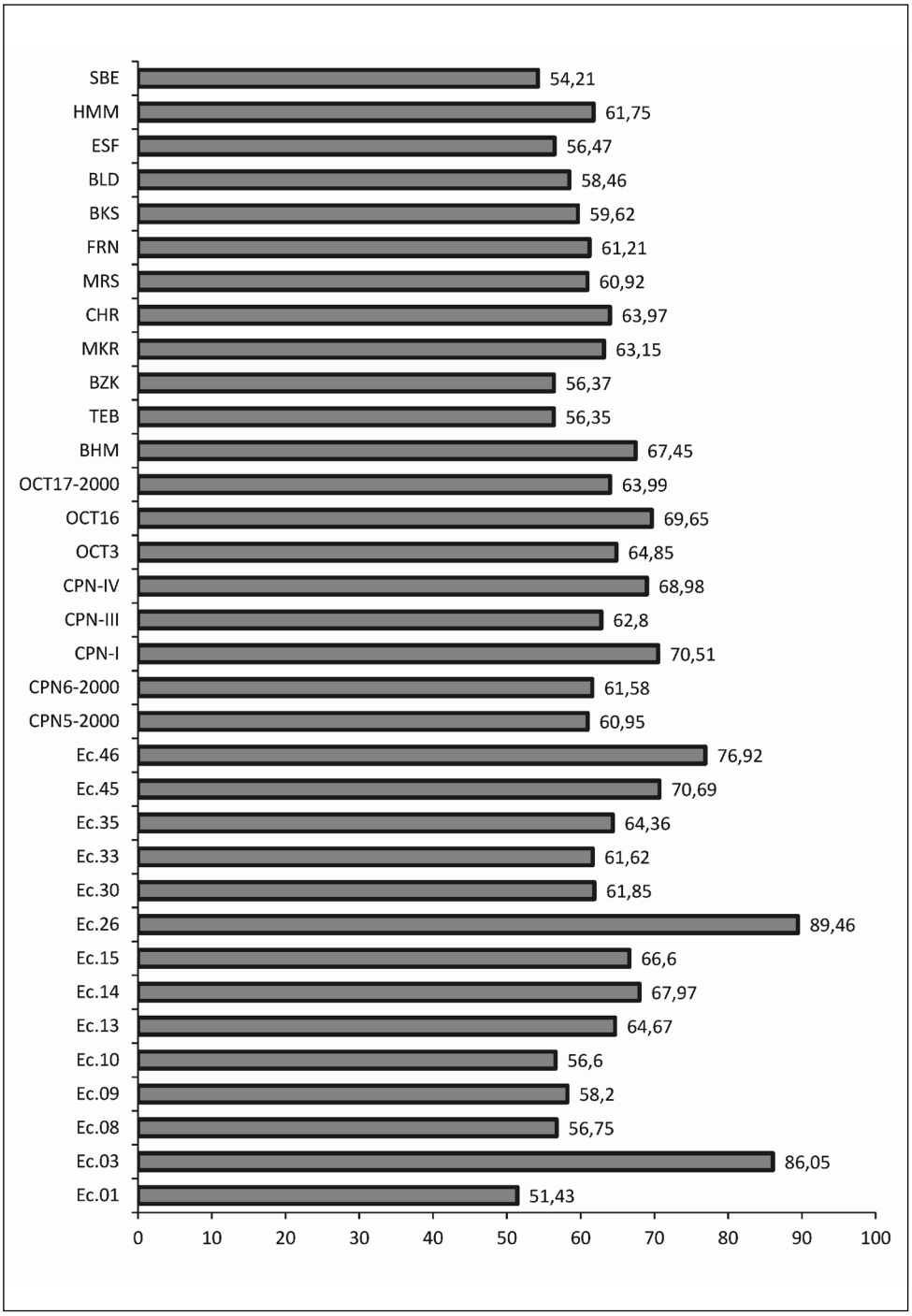

Fig. 4. Stummer index (SI) for the 34 wild and cultivated Tunisian grape ecotypes.

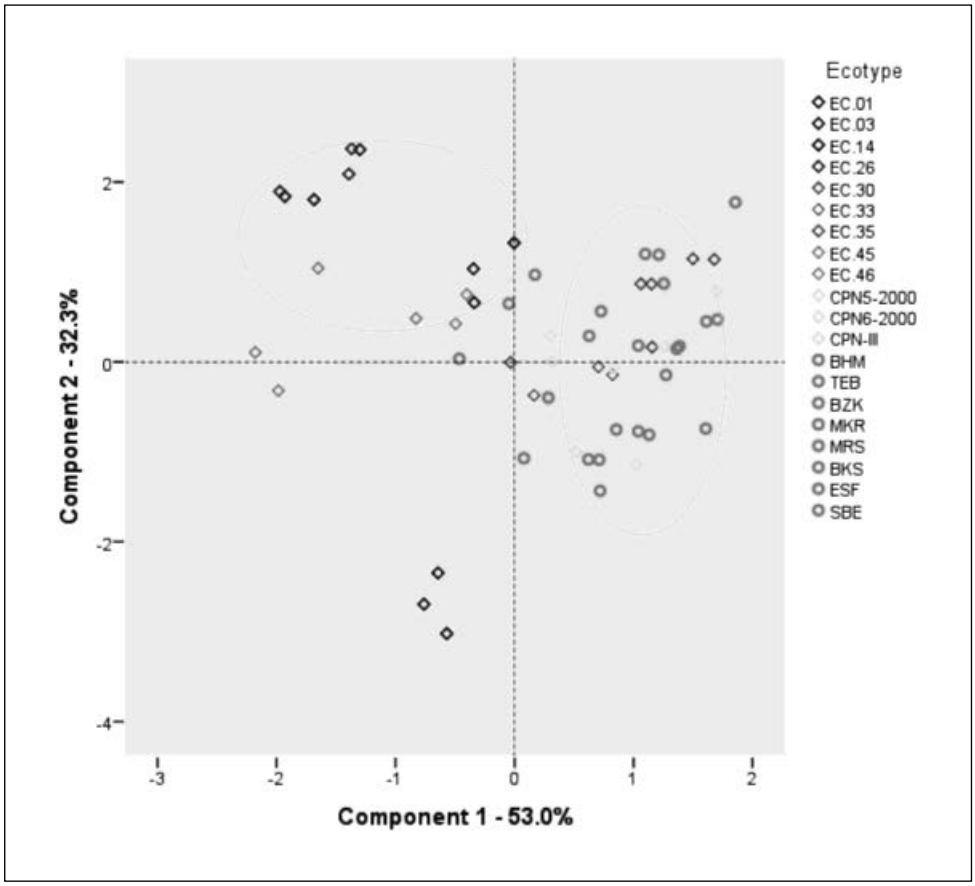

Fig. 5. PCA showing bi-dimensional discrimination between wild and domesticated grapes on the basis of morphometry and Stummer index of the seed.

From the twelve traditional varieties cropped in Rafraf, five confirmed their belonging to the cultivated morphotype (Beldi, Boukhasla, Farrani, Hammemi and Marsaoui) and three showed 95% chance of belonging to the spontaneous type (Bidh El Hmem, Chaaraoui and Meski Rafraf). The formers are definitely moving away from the *silvestris* type while the evolutionary process lasted even longer in the last cultivars (Levadoux 1956). The other unclassified grapevines could belong to either subspecies.

Principal component analysis (PCA).- Two components were extracted using PCA after varimax rotation and Kaiser Normalization. 85% of the total variance was explained by the two axes. The dispersion of the ecotypes according to axes 1 and 2 emphasized two main groups (Fig. 5): the first group is composed by cultivars ‘Tebourbi’, ‘Bazzoul Khadem’, ‘Meski Rafraf’, ‘Marsaoui’, ‘Boukhasla’, ‘Essifi’ and ‘Souabaa Eljia’. They are all vari-

eties with long seeds. The second group comprises wild ecotypes 'EC.03', 'EC.14', 'EC.26' and 'EC.46' characterized by large seeds with high Stummer index which provides information in relation with shape and curves of the seed.

Component 1 explained 53% of the variance and was well correlated with placement of chalaza, beak length and total length of the seed. This component assembled the group of local grapevines characterized by a long seed and unambiguous chalaza (Fig. 3). This axis would be a dimension axis. Component 2 explained 32% of the variance and was correlated with total breadth and Stummer index. This second component grouped four among the twelve wild ecotypes (EC.03, EC.14, EC.26 and EC.46) characterized by a large seed with high SI value. This axis would be a form axis.

The PCA classification of the collected local ecotypes reinforced their characterization and allowed the recognition of ecotypes of spontaneous vines originating from the north of Tunisia. Placement of chalaza, beak length and total length of the seed were the most predictive features for the discrimination between grape populations as they are not influenced by the year of sampling.

Discussion

Morphological data of grape seeds are important for the identity of cultivars, history, biogeography and mechanisms of grapevine domestication (Terral & al. 2010). In biological populations, shape of the seed is less variable and less subject to environmental factors than size (Mangafa & Kotsakis 1996). Morphometric analysis of grape seeds from Mediterranean locations in Tunisia has allowed us to attribute a small proportion of them to the wild *V. vinifera* subsp. *silvestris* and an equivalent proportion to the domesticated *V. vinifera* subsp. *vinifera*. This joins the idea of Bouby & al. (2010) who stated that the number of seeds attributed to the wild type is comparable to that attributed to the cultivated one whatever the discriminating method applied.

Classification based on the Stummer index showed an inter-reliability and morphological connection between seeds from wild and domesticated grapevines. In wild populations, the SI was comprised between 51 and 89. Wild grapes picked from different locations in Portugal showed seeds with SI higher than 75 in three ecotypes and between 65 and 75 in three other vines (Cunha & al. 2007). Indexes lower than 65 were not detected unlike the values reported here for some ecotypes. Schiemann (1953), working on seven Austrian grapevines, indicated an SI between 54 (cv. Sylvaner) and 65 (cv. Elbling). Such results strongly agree with those identified for local cultivars with SI ranging between 54 and 67.

Studies on seeds were carried out to allow discrimination between wild and cultivated grapes (Kislev 1988; Stummer 1911; Mangafa & Kotsakis 1996; Marinval 1997; Terral & al. 2010) and to distinguish the genetic diversity of local resources. Tunisian wild vines were clearly discriminated from cultivated vines by large spherical seeds deprived of stalk in most accessions. This genetic differentiation between wild and local cultivated populations was proved by Snoussi & al. (2004) using the genotyping of several vine accessions. Genotypic analysis of Tunisian cultivars and wild accessions at nine nuclear microsatellite loci showed that both sets of samples maintain high levels of genetic variation (Snoussi & al. 2004). This genetic diversity pattern was later observed by Oualkadi & al. (2011) when

prospecting 18 sites with 168 individual wild vines collected from the north of Morocco in comparison with populations picked from Algeria, France and Tunisia. In their study, it was demonstrated that some ecotypes from Ouchtata and Tabarka (Tunisia) were clustered with French group of wild grapevines.

The organization of genetic diversity is reminiscent of historical genetic structure originating from natural evolution, domestication and modern plant breeding (Aradhya & al. 2003; Lacombe & al. 2003). Such a structure could generally be described based on the amount and pattern of distribution of genetic diversity within and between different geographic or genetic groups. Aradhya & al. (2003) showed that measures of within-group genetic diversity merged three wild grape genotypes from Tunisia along with an accession from Corsica. These authors revealed that Tunisian grape cultivars showed close affinity to the wild progenitor subsp. *silvestris* reflecting the origin and domestication gait of many of the autochthonous cultivars. This was confirmed later by Zoghlami & al. (2009) who expected first that *V. silvestris* with small and fragmented populations would have low levels of genetic variation. Levadoux (1956) reported that foreign cultivated vines were introduced to North Africa and crosses with local populations lead to the genesis of more advanced genotypes holding the specificity of the region. According to the same author, the most advanced forms of vine seem to have appeared after the forms that come closest to spontaneous types. Thus, Tunisian local cultivars are generally not related to the still existent grapevine wild populations, suggesting that most of them could derive from materials introduced in the region in different historical times. This is a conclusion already made by Snoussi & al. (2004). According to the authors, table grape cultivars are either female or hermaphroditic self-compatible plants and most of the seeds produced by hermaphrodites result from selfing events. It is therefore possible to speculate that many of the current table grape cultivars derived from a few original ones that were imported to Tunisia as plants or raisins (Snoussi & al. 2004). The low gene flow between the wild grapevine populations and the set of Tunisian autochthonous grapevine cultivars was also revealed by Zoghlami & al. (2009). Wild grapes occur in natural closed landscape of forests, whereas the cultivated forms occur very far from the former. This constitutes a physical barrier against successful characterization of parentage between both grapevine sub-species (Zoghlami & al. 2009; 2013) taking into consideration also that the pollen cannot be dispersed far enough (Di Vecchi & al. 2007). Gristina & al. (2017) and Barbagallo & al. (2018, 2019) revealed that Sicily and its surrounding islands in the Mediterranean basin represent underexplored hotspots of genetic diversity for grapevine. These important reservoirs of potentially valuable genotypes shed light on the migration phenomenon of cultivars.

The study of *silvestris* vine has raised many prospects, in particular using them in future breeding programs to improve rusticity and disease resistance of local and introduced grape varieties. Considering that wild vines disappeared more quickly in hot and humid enough regions (Levadoux 1956), and in view of actual climate context, conservation efforts are required to maintain the genetic integrity and survival of the remnant populations (Cunha & al. 2007; Zoghlami & al. 2013). Wild grape germplasm is a potential source of unique alleles and is important for the improvement of both wine and table grapes (Aradhya & al. 2003). The spontaneous vine *V. vinifera* subsp. *silvestris* could represent a valuable gene pool to enrich diversity within the Tunisian germplasm of local vines since intensive cultivation of few grapevine varieties in extensive areas has drasti-

cally decreased genetic variability and has increased the risk of epidemic diseases (Arnold & al. 1998; Cunha & al. 2007; Harbi & al. 2010).

Conclusions

The morphometric features of the seed showed a clear difference in shape and size between wild and cultivated grapevines. Among the twenty-two wild vines, seven confirmed their belonging to the spontaneous type and are mainly originated from highlands of Nefza. Among the twelve native varieties, five confirmed their belonging to the cultivated morphotype and three suggest the wild grape. Some cultivars are definitely in more advanced form compared to others. Propagation by seedling probably contributed to the birth of these wild type cultivated forms. Certainly, the vegetative propagated varieties are and are likely to change considerably during sexual reproduction. Though, it is not to exclude the possibility that some cultivars derived from ancestral events of local domestication or cross hybridization with native wild plants.

Wild grapevines provide reservoir of useful traits and makes them particularly important sources of germplasm for breeding programs. Geometric morphometry suggests rich perspectives on the genetic and geographical characterization of cultivars. Morphometry of the seed from local grape populations showed a high level of polymorphism between wild and cultivated types. The certainty of this morphometric tool should be confronted to molecular techniques to verify the existence of a field of interaction between the two analytical methods.

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Short note

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P. Campisi, F. M. Raimondo & V. Spadaro

New floristic data of alien vascular plants from Sicily

Abstract

Campisi, P., Raimondo, F. M. & Spadaro, V.: New floristic data of alien vascular plants from Sicily. — Fl. Medit. 29: 263-267. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

New records of *Commelina communis*, *Euphorbia hypericifolia*, *Melia azedarach*, *Nicotiana tabacum,* and *Xanthosoma sorbifolium* are reported for the Sicilian flora.

Key words: xenophytes, biodiversity, plant distribution, Sicilian flora.

Introduction

Research on the urban flora of some inhabited centers of Sicily has made it possible to find alien species, infrequent not only in this island but also in the rest of the Mediterranean region. Some of them are already reported on the Island in one or a few localities, two are new for Sicily and one also for the Mediterranean and Europaean flora. These are *Commelina communis* L., *Euphorbia hypericifolia* L., *Melia azedarach* L., *Nicotiana tabacum* L. and *Xanthoceras sorbifolium* Bunge. Preliminary data on the Sicilian records of two of this taxa have been reported by Spadaro & al. (2019). In addition to brief botanical information on the taxa examined, the distribution and records data concerning Sicily are reported below.

Taxa and related Sicilian records

Commelina communis L. (*Commelinaceae*)

This Asiatic taxon has long been known in various Italian and European regions, both as a naturalized, and as a casual alien. It is a perennial herbaceous rhizomatous. It grows best in open and disturbed areas, along roadsides, and woodland borders. *C. communis* was introduced in central and south eastern Europe, and eastern North America, where it has spread to become a noxious weed. In Italy, Galasso & al (2018) report this taxon in almost all regions except Marche, Molise and Puglia. In Sicily, the species was already collected in 2017 in Palermo, on the edge of a busy street in the city center. At the same time it was

observed spontaneous in areas uncultivated within the Botanical Garden. This Sicilian record have been announced by Spadaro & al. (2019).

Specimens: Sicily, Palermo, via Nunzio Morello on the corner with via Ariosto, at the roadside, ca. 35 m a.s.l., 25.06.2017, *Spadaro* (PAL, PAL-Gr, FI); Sicily, Palermo, Botanical Garden, uncultivated space, ca. 10 m a.s.l., 12.08. 2017, *Raimondo* (PAL, PAL-Gr, FI).

Euphorbia hypericifolia L. (*Euphorbiaceae*)

Camaephyte native to the tropical and subtropical New World (i.e. USA, Mexico, Central and South America), it is also widespread in different countries on various continents (i.e. Africa, Europe, Asia). Sciandrello & al. (2016) report for the first time *E. hypericifolia* in Italy, near Taormina (Sicily). They also report the distribution data known from the literature, according to which the species is present in Singapore and Taiwan, in Egypt, Israel, Belgium, Spain, Canary Islands and Greece (Peloponnese). After Sciandrello & al (2016), a second Sicilian locality of *E. hypericifolia* is reported in Sicily in Palermo (Spadaro & Raimondo 2018). Recently, its presence was ascertained in various sites within and on the outskirts of the inhabited center: particularly in the city of Catania, Trapani and Marinella di Selinunte (Castelvetrano).

Specimens: Sicily, Castelvetrano, in Marinella di Selinunte village, ruderal environment, ca. 10 m a.s.l., 14.07.2018, *Raimondo* (PAL, PAL-Gr, FI,); Sicily, Trapani, to the edge of the square of the train station, ca. 10 m a.s.l., 11.07.2019, *Raimondo* (PAL, PAL-Gr, FI); Sicily: Catania, flower beds in the square Giovanni Verga, ca. 25 m a.s.l., 11.07.2019, *Raimondo* (PAL, PAL-Gr, FI).

Melia azedarach L. (*Meliaceae*)

Tree, native to southern Asia (Yulianti & al., 2011), is cultivated for ornamental purposes in various countries where it has also partly spontaneously grown. In Italy Galasso & al. (2018) record it in various regions including Sicily where it had been reported by Di Martino and Perrone (1962) among the species of the arboricolous flora of Palermo, a city in which it has been used extensively in the trees of streets and squares since the 1800s. Unlike its first report, the new record concerns a small uneven population, observed on the northern slopes of Monte Pellegrino where it is found at the edge of the rolling stock, at the base of an escarpment facing East. Recently for the Mediterranean area, Sakhraoui & al. (2019) report the naturalization of *M. azhedarach* in Algeria.

Specimens: Sicily, Palermo, Monte Pellegrino, East slopes in calcareos soil, ca. 200 m a.s.l., 12.9.2019, *Campisi*, (PAL, PAL-Gr, FI).

Nicotiana tabacum L. (*Solanaceae*)

The species, native to South America and cultivated in numerous countries, is known spontaneously in various regions of the world. In Italy, Galasso & al. (2018) report it in the

north, central and south, including Sicily where the species had been reported in Mazara del Vallo (Dia & Romano 1981). Recently, a rich population was found inside the former geriatric institute, in the inhabited center of the city of Trapani, in nitrified habitat.

A taxonomic study of the *exsiccata* allows to refer the Sicilian finding to *Nicotiana tabacum* var. *angustifolia* (Mill.) Aiton.

Specimens: Sicily, Trapani, nitrified space inside the ex geriatric institute, via Segesta, ca. 10 m a.s.l., 10.8.2018, *Raimondo* (PAL, PAL-Gr, FI).

Xanthoceras sorbifolium Bunge (*Sapindaceae*) (Fig. 1).

Native to China, this taxon is the only species of its genus. Introduced in some gardens of different countries, including Italy, where it is present in Sicily only. This plant has a shrubby or arborescent habit; alternate, pinnate leaves, with 10-20 lanceolate leaflets, sessile. The flowers are grouped in axillary racemes and have a pentapartite corolla, with white petals showing a small red spot at the base. The fruit is a globose, trilocular capsule, with a hook point, and yellowish when ripe. Each loculus contains 1-2 (-3) showy subspherical seeds, of dark brown colour. One individual is cultivated in the park of Villa Whitaker to Malfitano (Palermo). The site of spontaneization of *X. sorbifolium* is in the same garden where it was introduced, and cultivated. There, the population of this taxon occupies a small uncultivated space. Outside the city of Palermo, to date, there are no reports of this taxon. This record has been announced by Spadaro & al. (2019).

Specimens: Sicily, Palermo in the park of Villa Whitaker to Malfitano, in calcareos soil, ca. 35 m a.s.l., 11.8.2018, *Raimondo* (PAL, PAL-Gr, FI).

Discussion and Conclusion

According to Galasso & al. (2018), the exotic vascular flora of Sicily includes 437 taxa. Today it is enriched with two other taxa (*Commelina communis* and *Xanthosoma sorbifolium*). Furthermore, *Nicotiana tabacum* and *Melia azedarach*, previously known from a single locality, respectively in Mazara del Vallo (Trapani) and in Palermo (arboreal habitat), are confirmed: the first in another locality, in the same province, the second always in Palermo but on the soil. On the contrary, *Euphorbia hypericifolia* – recorded near Taormina, in the Messina province (Sciandrello & al. 2017) and then in Palermo (Spadaro & Raimondo 2015) – is now also reported in Catania, Trapani and, in the same province, in Marinella di Selinunte (Castelvetrano). This taxon in Sicily begins to spread into urbanized contexts. It is conceivable for this species a naturalization path that could affects all the coastal towns of the island.

The Sicilian record of *Xanthosoma sorbifolium* represents the first case of spontaneousization reported in the Mediterranean and European geographical context. We can say that today it affects the same site where the species was first introduced and cultivated in Europe for about a century, and there is reason to believe that it will remain an isolated case for a long time.

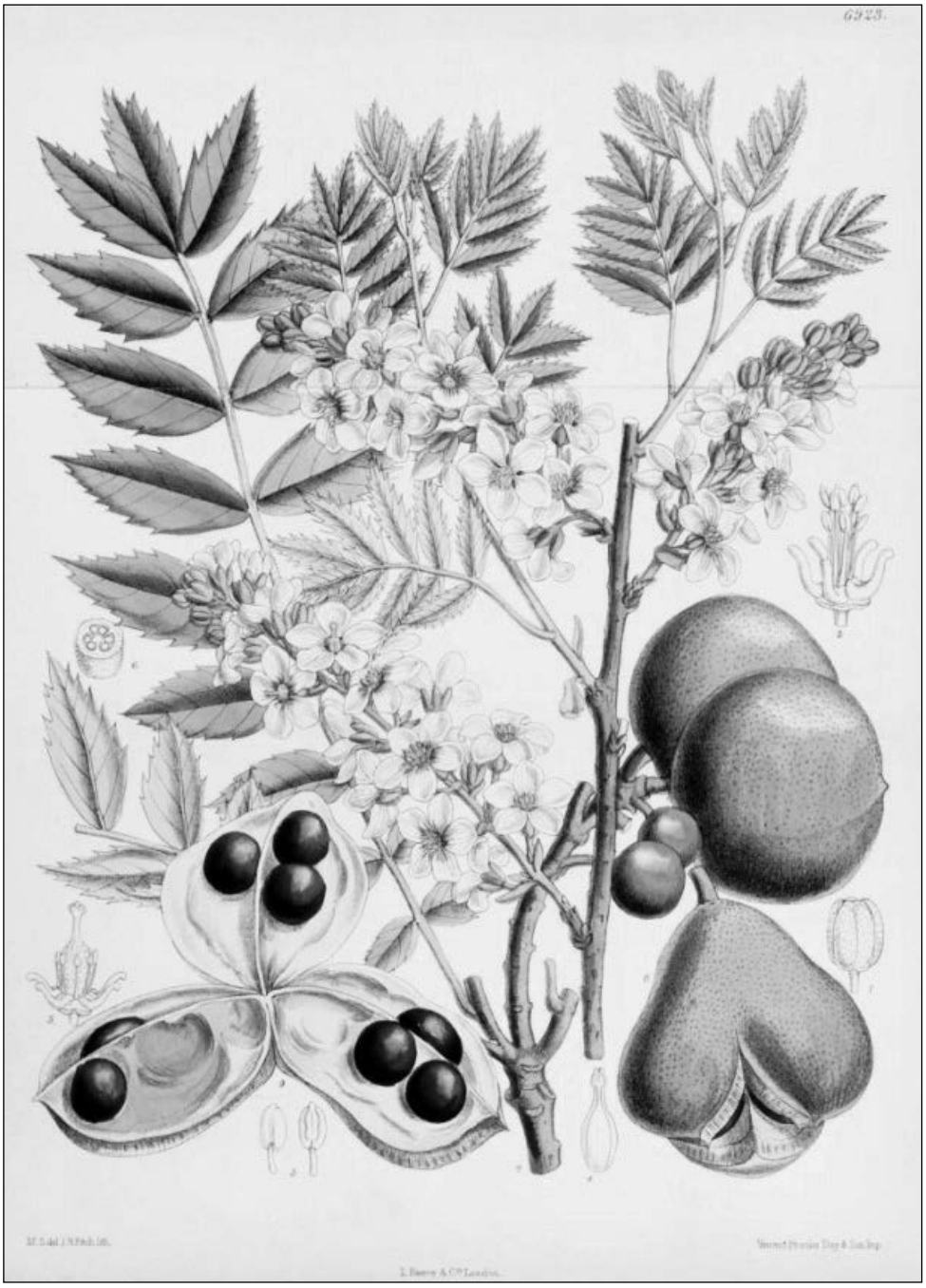

Fig. 1. *Xanthoceras sorbifolium* (Curtis illustration) - Hand-coloured lithograph of *X. sorbifolium* Bunge, after a painting by Matilda Smith (1887), taken from Curtis's Botanical Magazine 113: t. 6923 (from Kew*science,* Plants of the World *online*).

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Mediterranean plant germination reports – 1

edited by Sara Magrini & Cristina Salmeri

Abstract

Magrini, S. & Salmeri, C. S. (eds): Mediterranean plant germination reports – 1. — Fl. Medit. 29: 269-306. 2019. — ISSN: 1120-4052 printed, 2240-4538 online.

This is the first issue of a new series of germination reports from Mediterranean areas (sensu Med-Checklist). It comprises germination protocols for 18 taxa: *Abies* from Sicily by A. Scialabba (No. 1); *Centaurea*, *Cyperus*, *Launaea*, *Medicago*, *Muscari*, *Ononis*, *Pancratium*, *Plantago*, and *Thinopyrum* from Sicily by C. Salmeri & M. Trubia (Nos 2-10); *Anthemis*, *Crucianella*, *Eryngium*, and *Thinopyrum* from Sardinia by M. Porceddu & al. (Nos 11-14); *Centaurea*, *Jacobaea*, *Matthiola*, *Medicago*, *Pancratium*, and *Silene* from central Italy by S. Magrini & al. (Nos 15-20); *Sporobolus* and *Juncus* from central Italy by G. Fabrini (Nos 21-22).

Editorial

This new series of germination reports of Mediterranean plants will be published in Flora Mediterranea, as announced during the XVI OPTIMA Meeting held in Athens this year. The proposal of this new column originated by RIBES, the Italian network of seed banks, as part of its conservation strategy to 2020. The geographical coverage extends to all countries included in Med-Checklist, plus Austria, Switzerland, the Canary Islands, Madeira and the Azores.

Only germination report with high germination percentages (> 80%) which are new for any individual territory, or better than published ones, may be accepted for publication. Deviations from this standard can be taken into account if duly justified.

Each contribution should present all the relevant details of tested germination protocols, including mandatory information on:

– seed accession data, with full collection details (locality, habitat, date, collector name), the accession code and the seed-bank name;
– germination data: pre-treatments (e.g. scarification, priming, stratification, sterilization); germination media; number of seeds and replicates for each test; culture conditions (temperature and photoperiod);

– germination results summarized in a table using common germination indices.

For the preparation of the manuscript, please, follow the instructions reported below also published at http://www.herbmedit.org/guide.html.

Authors are invited to submit their manuscript in English to one of the editors.

There is currently no special column devoted to seed germination in other scientific journals, so "Mediterranean plant germination reports" could surely become a useful tool for conservationists, botanists, and other specialists. Moreover, these germination data will flow into an online, open-access database managed by RIBES association in order to make them available to a wider audience.

Instructions to author for "Mediterranean plant germination reports"

This series of articles will be a separate section published alongside the other articles of *Flora Mediterranea*. These contributions do not follow the structure of standard papers in the journal but have their own format. For the preparation of the manuscript follow the lay-out of the published contributions and the Instructions to authors of Flora Mediterranea (http://www.herbmedit.org/guide.html) for the style.

Only germination report with high germination percentages (> 80%) which are new for any individual territory (according to *Med-Checklist*), or better than published ones, may be accepted for publication. Deviations from this standard can be taken into account if justified.

Each contribution should present all the relevant details of tested germination protocols. They should include mandatory information on:

Seed accession data

The following format is to be used: Country and/or political subdivision (according to Med-Checklist): Locality (WGS84 coordinates in decimal degrees), habitat, altitude, date, *collector/s* (accession code and seed-bank name).

Germination data

Pre-treatments (e.g. scarification, priming, stratification, sterilization); germination media; number of seeds and replicates for each test; culture conditions (temperature and photoperiod); results summarized in a table through the following common germination indices:

– final germination percentage (G);
– germination delay, i.e. the first radicle emergence (T_1, in days);
– median germination time, i.e. the time to reach 50% of final/maximum germination (T_{50}, in days) estimated according to the formula: $T_{50} = [(G/2 - G_1) \times (t_2 - t_1)]/(G_2 - G_1) + t_1$, where G_1 is the germination percentage immediately lower than G/2, G_2 is the germination percentage immediately higher than G/2, t_1 and t_2 the number of days to reach G_1 and G_2, respectively;
– maximum germination time, i.e. the last radicle emergence (T_{max}, in days);

– mean time to germination (MTG, in days) estimated according to the formula: MTG = $\Sigma (n_i \times d_i)/N$, where n_i is the number of germinated seeds at day i, d_i the incubation period in days at day i, and N the total number of germinated seeds in the treatment.

An "Observations" section may be used for additional information, also including alternative methods or comparative evaluation of existing germination data from other geographical areas.

Good quality images of germinated seeds may be added.

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*RIBES, Rete Italiana Banche del germoplasma per la conservazione Ex Situ della flora italiana, www.reteribes.it

Anna Scialabba

Seed germination in *Abies nebrodensis* (*Pinaceae*)

Abstract

Scialabba, A.: Seed germination in *Abies nebrodensis* (*Pinaceae*) [In Magrini, S. & Salmeri, C. (eds), Mediterranean plant germination reports – 1]. – Fl. Medit. 29: 272-276. https://doi.org/10.7320/FlMedit29.272

Abies nebrodensis is a strict Sicilian endemic species, listed as "Critically Endangered" in the IUCN Red List of Threatened Species. Seed production highly varies annually and among specimens. The species is affected by a very low seed germination rate, mainly depending on the large number of empty seeds (from 68 to 96%). The embryo presence in seeds increases with increasing seed size and weight. Optimal germination rate (61%) was achieved at constant 5°C in the darkness. The low proportion of embryos in seeds is a relevant threat for *A. nebrodensis* conservation, as it affects the intrapopulation genetic diversity and reduces the potential genetic variability of seed accessions preserved in a gene bank collection.

Keywords: endangered species, germplasm conservation, Sicily.

Introduction

Abies nebrodensis (Lojac.) Mattei (Nebrodi fir or Sicilian fir) is a Sicilian critically endangered species (Thomas 2011), restricted to small areas (overall c. 150 ha) located in the Vallone Madonna degli Angeli, Mt. Scalone, Mt. dei Pini and Mt. Cavallo (Madonie Natural Park, NW Sicily), at 1360-1700 m of elevation.

The natural population is currently restricted to 30 mature individuals, each labelled by a single ID number (Virgilio & al. 2000), with a very fluctuating number of juveniles derived from natural regeneration. Actually, only 24 specimens produce cones, giving low germinating seeds (Scialabba & al. 2007).

Though conservation and restoration programs have been carried out since 1992 (Venturella & al. 1997), the low sexual performance of mature individuals and the low survival rate of seedlings and saplings involve specific actions of *ex-situ* conservation supporting the *in-situ* protection strategies (Ducci 2014). To this regard, the study of seed germination behaviour and embryo growth capability are pivotal activities to ensure appropriate conservation of this relict species.

1. ***Abies nebrodensis*** (Lojac.) Mattei (*Pinaceae*)

Accession data

Si: Polizzi Generosa (Palermo), loc. Madonie, Vallone Madonna degli Angeli (WGS84: 37.845512°N, 14.020996°E), Specimen ID number 22, 1390 m a.s.l., 20 Oct 2004, *Corpo Forestale dello Stato & A. Scialabba* (SPGR/PA – Acc.1403, Sicilian Plant Germplasm Repository of Palermo University SGPR/PA, formerly HBP Bank).

Germination data

Pre-treatments: Manual removal of wings; sterilization with a solution of 30% sodium hypochlorite and Tween 20 for 5 minutes, followed by 3 rinses in sterile water; imbibition for 48 hours in distilled water.

Germination medium: 4 sheets of sterilized filter paper (Whatman 40), imbibed with 10 ml distilled water under sterile conditions.

Sample size: 75 seeds for each test (15 × 5 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
61.0%	constant 5°C	0/24h	63.0	70.0	110.0	75.24

Observations

Mature cones were collected in October from wild trees with the identification number (ID) 13, 16, 17, 22, 25, 27. The seeds were individually evaluated, measuring both the major axis and weight. These morphological parameters are useful to distinguish well-developed seeds (Fig. 1a) from empty ones (Fig. 1b), which can affect the germination rate.

As far as the seed size is concerned (Scialabba & al. 2007), there are big seeds (9-12 mm), small seeds (< 9 mm), and small flattened seeds which are always empty. Based on seed weight (Scialabba & al. 2009), 3 main classes can be distinguished: seeds < 30 mg were always empty; the highest amount of seeds (mean value of 58.4%) belonged to the 31-50 mg weight class, with a very low embryo presence, ranging from 6% (specimen ID 27) to 39.9% (specimen ID 16); while 17.3% of seeds belonged to the weight class of 51-70 mg, where the embryo presence increased but with different values among the specimens, ranging from 22% (specimen ID 13) to ca. 98% (specimens ID 16 and 22). A fourth weight class > 71 mg was only found in plant ID 13 with a high embryo presence (93.3%). Therefore, the embryo presence increases both seed size and weight, and weight higher than 40 mg represents the most suitable fraction for valuable seed bank collections. Unfortunately, seed production in *A. nebrodensis* varies annually very much among different specimens (4% ID 25 to 32% ID 22), so that it is not possible to predict the number of vigorous seeds available per year.

Different germination tests were carried out 6 months after seed harvesting, under different thermoperiod conditions, i.e. constant temperatures (5°C and 15°C) and alternating temperature (5/15°C), in full darkness, applying in the medium pure distilled water or 10^{-3} M gibberellic acid (GA_3) water solution. Not germinated seeds were checked to verify the embryo presence or absence and the endosperm presence or not within empty seeds.

The germination rate is deeply affected by big quantity of empty seeds. The individuals of *A. nebrodensis* actually produce high percentages of seeds without embryo and even without endosperm. For this reason, seedlings can be obtained only by selecting seeds to germinate by their size and weight.

In fact, optimal results were reached only on selected seeds with embryo at 5°C (Fig. 1c), but germination tests carried out at 15°C in GA_3 10^{-3} M provided similar germination percentages. Seed germination proceeded very gradually; the specimen ID 22 showed the highest germination capability and the highest production of full developed seeds, with G = 25% using randomly selected seeds. Integument rupture (IR) was also observed in not germinated seeds (IR = 80% in plant ID 25 after 23 days incubation at 5°C in pure water). Seeds not containing embryo often also lacked endosperm (63.95%) (Fig. 1b). Embryos showed five cotyledons, they were albino (mainly in ID 16, 17 and 22 respectively for 83.0, 79.7 and 75.5%), and partly green (Fig. 1d) or fully green (Fig. 1e) (mainly in ID 13 and 27 respectively for 87.1 and 83.3%).

It must be emphasized that proteomic analyses carried out on embryos and endosperm of not germinated seeds, at different incubation times (30, 60 and 130 days), revealed specific protein patterns for both embryo at different developmental stages and endosperm at different mobilization of seed storage reserves (data not shown) and indicated that a metabolic active phase occurred also in non-germinating seeds.

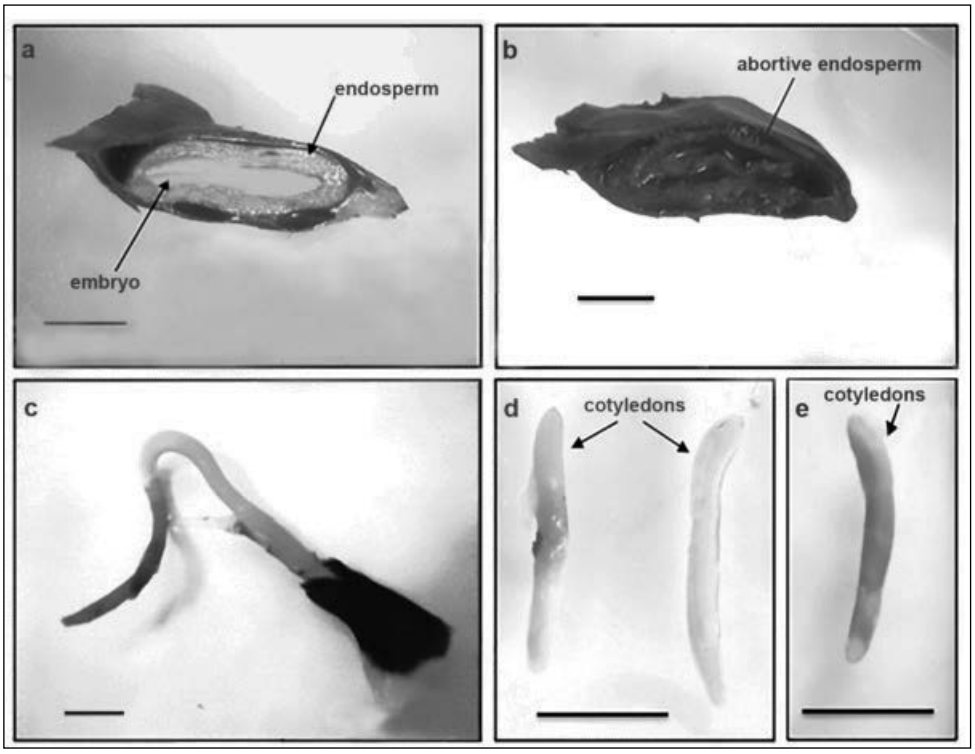

Fig. 1. Seeds of *Abies nebrodensis*: **a,** longitudinal section of fully developed seed; **b**, longitudinal section of empty seed. **c**, seedling with cotyledons covered by integument; **d**, albino embryos; **e**, green embryo. Bars: 5 mm.

The high presence of empty seeds in *A. nebrodensis* seems to be related with pro-embryo degeneration due to self-pollination or hybridization with *A. cephalonica* Loudon and *A. alba* Mill., which also occur in the area (Scialabba & al. 2005). *A. nebrodensis* appears intermixed with the other *Abies* species, supporting the hypothesis of remarkable hybridization processes within the genus *Abies* (Conte & Cristofolini 2003). The quotient of inbreeding depression (Q. I.), expressed as the ratio between developed and empty seeds and the total amount of seeds was very high in the whole population (80.24% ± 5.22) and single specimens (from 64.80% to 95.26%) of *A. nebrodensis*. These data are consistent with the low population density, as recorded in other conifers (Restoux & al. 2008), and the presence of the same haplotype, detected by cpSSRs, in at least 7 individuals (Parducci & al. 2001).

Conclusion

The low reproductive rate of the seeds in *A. nebrodensis*, mainly depending on the high amount of empty seeds, represents a relevant threat for its conservation, as previously hypothesized by Messeri (1958). The low percentage of embryos inside the seeds affects the results from germination tests, when carried out on randomly selected seeds, and decrease the genetic diversity of seed accessions. This, however, can be improved by manually selecting seeds containing embryo (based on their size/weight), thus avoiding overestimation of the genetic diversity of seed bank collections and improving the qualitative and quantitative potentiality of seed accessions.

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Cristina Salmeri & Maria Trubia

Seed germination reports for coastal sand dune species from Sicily

Abstract

Salmeri, C. & Trubia, M.: Seed germination reports for coastal sand dune species from Sicily [In Magrini, S. & Salmeri, C. (eds), Mediterranean plant germination reports – 1]. — Fl. Medit. 29: 277-287. 2019. https://doi.org/10.7320/FlMedit29.277

This study investigated seed germination in nine psammophilous species occurring in Sicilian sand dunes, some of which, such as *Muscari gussonei*, *Launaea resedifolia* and *Pancratium maritimum*, are endemic and/or rare and very scattered in the area. Different germination protocols were tested for one or more populations and the best germination results per species are provided, reporting experimental conditions and specific comments on the germination behaviour.

Key words: germination protocols, Mediterranean flora, psammophytes.

2. *Centaurea sphaerocephala* L. subsp. ***sphaerocephala*** (*Asteraceae*) (Fig. 1a)

Accession data

Si: Gela (Caltanissetta), Contrada Roccazzelle (WGS84: 37.093372°N, 14.165187°E), 5 m a.sl., 08 Jun 2010, *A. Lantieri & R. Galesi* (BGS-CT 32AL/NV/07, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Si: Sampieri (Ragusa), loc. Pineta (WGS84: 36.720606°N, 14.741661°E), 2 m a.s.l., 11 Jul 2011, *A. Lantieri & R. Galesi* (BGS-CT 01AL/RG/11, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Manual removal of pappus. Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40), imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Observations

Germination tests carried out at different constant temperatures (10°C to 25°C) indicated 20°C as optimal germination temperature, though significant interspecific variation was detected, with highly different germination rates between the two investigated populations. Samples from Roccazzelle, very close to the town of Gela, showed the highest germination

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]	Accession code
97.0%	constant 20°C	12/12h	2.0	3.4	22.0	5.0	BGS-CT 32AL/NV/07
83.0%	constant 20°C	12/12h	2.0	4.3	31.0	7.9	BGS-CT 01AL/RG/11

percentages, all exceeding 87% (88% at 10°C and 15°C, 97% at 20°C, 87% at 25°C), while seed germination values ranged from 57% (10°C) to 81% (25°C) and 83% (20°C) in the other accession from Sampieri beach. Increase in temperature enhanced the germination speed by over 70% in the first accession (T_{50} = 6.8 at 10°C *vs* T_{50} = 1.8 at 25°C) and over 55% in the second one (T_{50} = 8.8 at 10°C *vs* T_{50} = 4 at 25°C). Temperature also affected the germination rate out of the optimal level of 20°C, with lower values at both minor and major temperature for the two populations. Royal Botanic Gardens Kew (2019) reports for the species 82 % germination under 20°C and light 8/16 photoperiod.

3. *Cyperus capitatus* Vand. (*Cyperaceae*) (Fig. 1b)

Accession data

Si: Gela (Caltanissetta), Contrada Roccazzelle (WGS84: 37.093372°N, 14.165187°E), 5 m a.sl., 08 Jun 2010, *A. Lantieri & R. Galesi* (BGS-CT 07AL/RG/10, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40), imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Observations

Based on Redondo-Gómez & al. (2011), *C. capitatus* seeds from SW Spain exhibit their greatest germination values at salinity levels between 0 and 1% under a 16 h light/8 h dark photoperiod and 25/15°C thermoperiod. Salt concentration > 1% in the substrate completely inhibited seed germination. Germination results, here firstly given for Sicilian plants, agree with these data since seeds achieved the highest germination percentage in distilled water, as well as in 0.1 M NaCl water solution though with much longer germination time. Conversely, increasing NaCl concentration (0.2M, 0.3M, 0.5M) in the germination medium strongly affected seed germination up to total inhibition (G = 0%) at the higher salinity conditions. Seed germination under light conditions was just a bit lower than in full darkness, indicating that seeds are not affected by photoinhibition and light

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
100%	25/15°C	0/24h	7.0	7.7	22.0	10.3
96.7%	25/15°C	12/12h	13.0	15.3	28.0	16.7
96.7%(1)	25/15°C	0/24h	8.0	13.5	31.0	15.3
91.7%(1)	25/15°C	12/12h	16.0	25.9	35.0	27.2

(1) 0.1 M NaCl concentration in the germination medium.

only reduces the germination rate (Pi < 0.1, Carta & al. 2017). Alternating temperature also was a determinant factor for germination success, as germination tests carried out on this accession under constant temperatures (10°C, 15°C, 20°C, 25°C) showed very low germination values, ranging from 0% to 17%, except for 25°C with 57%. Constant temperatures appeared to increase photoinhibition since germination values in light-incubated seeds (12/12h) did not exceed 15%, with photoinhibition index (Pi) ranging between 0.7 and 1 that indicates strong inhibitory effects of light (Carta & al. 2017). Contrarily, Royal Botanic Gardens Kew (2019) reports 80% of germination at a constant temperature of 31°C and 12/12 h photoperiod, though only 20 seeds were tested.

4. *Launaea fragilis* (Asso) Pau (*Asteraceae*) (Fig. 1c)

Accession data

Si: Sampieri (Ragusa), loc. Pineta (WGS84: 36.720606°N, 14.741661°E), 2 m a.s.l., 11 Jul 2011, *A. Lantieri & R. Galesi* (BGS-CT 02AL/RG/11, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Manual removal of pappus. Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40), imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
99.0%	constant 25°C	12/12h	2.0	3.6	15.0	4.9
98.0%	constant 15°C	12/12h	2.0	11.2	31.0	12.4

Observations

This species is a suffruticous chamaephyte with a Saharo-Sindian distribution area, which in Italy only occurs in the sand dunes of the southern and south-eastern Sicily, where it is rather rare and grows in the *Ammophila arenaria* mobile dune community. This is the first germination report for the species. Germination tests carried out on fresh seeds provided successful germination results also at 20°C (90%), while only 70% of germination was reached at 10°C.

5. *Medicago marina* L. (*Fabaceae*) (Fig. 1d)

Accession data

Si: Sampieri (Ragusa), loc. Pineta (WGS84: 36.720606°N, 14.741661°E), 2 m a.s.l., 11 Jul 2011, *A. Lantieri & R. Galesi* (BGS-CT 03AL/RG/11, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water. Mechanical scarification by sandpaper for 5 minutes. Imbibition in distilled water for 24 h.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40) imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
98.0%	constant 10°C	12/12h	3.0	4.4	22.0	6.1
98.0%	constant 15°C	12/12h	2.0	2.7	22.0	4.4
98.0%	constant 20°C	12/12h	1.0	2.1	13.0	3.1

Observations

The examined Sicilian population of *Medicago marina* revealed an optimal temperature range of 10-20°C, which agree with Royal Botanic Gardens Kew (2019), where 100% germination was reported at both 15°C and 20°C with 12/12 photoperiod. Lower germination percentage (89%) was achieved at 25°C with the same pre-treatments. Seeds resulted positively photoblastic, because germination tests under full darkness condition provided a bit lower results, with seed germination percentages ranging from 87% to 90% at the same temperature regimes as the light-incubated seeds. Again, increase in temperature (25°C) led to lower seed germination percentage (50%), which is in contrast with the 98% germination obtained by Scippa & al. (2011) for plants from the Molise coastal dunes (Central-south Italy) under the same conditions. Ballesteros & al. (2015) reported a germination percentage of 95% for dark-incubated seeds at 20°C, but previously chemically scarified with sulfuric acid for 20 minutes.

6. *Muscari gussonei* (Parl.) Nyman (*Asparagaceae*) (Fig. 1e)

Accession data

Si: Gela (Caltanissetta), Contrada Roccazzelle (WGS84: 37.095833°N, 14.161388°E), 3 m a.s.l., 08 Jun 2010, *A. Lantieri & R. Galesi* (BGS-CT 06AL/RG/10, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40) imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
100%	constant 10°C	0/24h	11.0	13.4	35.0	15.4
98.0%	constant 15°C	12/12h	12.0	13.7	36.0	14.9

Observations

Muscari gussonei is an Endangered endemic species (Orsenigo & al. 2018) which occurs in small fragmented populations along the south-western coast of Sicily. Seed germination resulted negatively affected by temperature since it decreased with increasing

temperature (84% at 20°C, 0% at 25°C). No statistically significant effect of light was instead detected among germination percentages, which were high and similar for both light- and dark-incubated seeds under 10°C (100%, in accordance with Royal Botanic Gardens Kew 2019) and 15°C (97%).

7. *Ononis variegata* L. (*Fabaceae*) (Fig. 1f)

Accession data

Si: Gela (Caltanissetta), Contrada Roccazzelle (WGS84: 37.093372°N, 14.165187°E), 5 m a.sl., 23 Jul 2007, *A. Lantieri & R. Galesi* (BGS-CT 32AL/NV/07, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Si: Riserva Naturale Orientata Oasi del Simeto (Catania), foce del fiume Simeto (WGS84: 37.407266°N, 15.091696°E), 1 m a.s.l., 02 Jul 2010, *A. Lantieri & R. Galesi* (BGS-CT 11AL/RG/10, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water. Mechanical scarification by sandpaper for 5 minutes. Imbibition in distilled water for 24 h.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40), imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
100%	constant 10°C	12/12h	2.0	2.3	8.0	3.3
100%	constant 15°C	12/12h	1.0	2.1	7.0	3.2
99.0%	constant 20°C	12/12h	1.0	1.3	12.0	2.0

Observations

The species shows a physical dormancy linked to its hard, impermeable seed coat, which needs scarification pre-treatments. The examined Sicilian populations of *Ononis variegata* revealed a wide range of optimal germination temperature from 10°C to 25°C, with maximum germination percentages very close to 100%. These results to some extent agree with existing data from Royal Botanic Gardens Kew (2019). Germination tests carried out under 24h dark photoperiod provided germination rates similar to those of light-incubated seeds

(98-100%), but with faster germination speed (T_{50} = 1-1.1 d, MTG = 2.1-2.2 d). The population from the coastal area of Simeto River showed a very similar germination behaviour, i.e. 97-99% under 12/12h photoperiod and 97-100% under 24h dark photoperiod, at constant temperature regimes from 10°C to 25°C, but germination speed was much faster with lower values of T_{50} and MTG.

8. *Pancratium maritimum* L. (*Amaryllidaceae*) (Fig. 1g)

Accession data

Si: Marsala (Trapani), Isola Grande dello Stagnone (WGS84: 37.903868°N, 12.455125°E), 1 m a.s.l., 02 Oct 2004, *S. Pasta & L. Scuderi* (BGS-CT 087G5/04, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Si: Gela (Caltanissetta), Spiaggia di Macchitella (WGS84: 37.076439°N, 14.210627°E), 6 m a.s.l., 26 Sept 2004, *Lantieri, Sciandrello & Visalli* (BGS-CT 052G1/04, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Si: Riserva Naturale Orientata Oasi del Simeto (Catania), foce del fiume Simeto (WGS84: 37.407266°N, 15.091696°E), 28 Sept 2005, *Restuccia & Visalli* (BGS-CT 060AR/NV/05, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40), imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]	Accession code
98.0%	constant 20°C	12/12h	1.0	6.7	14.0	7.2	BGS-CT 087G5/04
98.0%	constant 20°C	12/12h	5.0	10.0	25.0	11.0	BGS-CT 052G1/04
98.0%	constant 20°C	12/12h	3.0	7.7	18.0	9.0	BGS-CT 060AR/NV/05

Observations

Three different populations of *P. maritimum* from Sicily were investigated, coming from 3 opposite areas, W Sicily (Isola Grande), S Sicily (Gela) and E Sicily (Simeto river), which are characterized by the same mean annual temperature (around 17.5°C), but by dif-

ferent mean annual rainfall and relative humidity (448.6 mm/77.5%, 354.2 mm/75.6%, and 547.2 mm/69.9%, respectively). However, each accession had its optimal germination response (98%) at a constant temperature of 20°C with 12/12 photoperiod. High levels of germination, all exceeding 92%, were also obtained at other constant temperatures: 92-95% at 10°C, 93-96% at 15°C, 92-97% at 25°C. Increasing temperatures in general enhanced seed germination speed (T_{50} of c. 25d at 10°C, c. 17d at 15°C, and c. 10d at 15°C). Seeds from all populations resulted positively photoblastic, because germination tests under full darkness provided very low germination rates at each thermoperiod, not exceeding 50%, which is in contrast with germination results from Sardinian and Tuscan populations (Bacchetta & al. 2007; Balestri & Cinelli 2004).

9. *Plantago macrorrhyza* Poir. (*Plantaginaceae*) (Fig. 1h)

Accession data

Si: Sampieri (Ragusa), loc. Pineta (WGS84: 36.720606°N, 14.741661°E), 2 m a.s.l., 11 Jul 2011, *A. Lantieri & R. Galesi* (BGS-CT 04AL/RG/11, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40), imbibed with 3 ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
100%	constant 10°C	12/12h	2.0	3.7	17.0	5.2
100%	constant 15°C	12/12h	1.0	2.3	9.0	2.9
100%	constant 20°C	12/12h	1.0	2.3	6.0	2.6

Observations

The species shows a moderate sensitivity to temperature and salinity. Temperature regimes over 25°C tended to induce seed dormancy, with delay in both germination time and final germination percentage (94% at 25°C, 50% at 30°C); a 50% germination is also reported at 25°C and 12/12 h photoperiod by Royal Botanic Gardens Kew (2019). These results are in accordance with those given by Luciani & al. (2001), who studied germina-

tion behaviour of this species related to temperature, salinity and after-ripening time, assessing an optimal germinability (98%) at 15°C (full darkness and distilled water), and a seed dormancy induction with both rising temperature (> 20°C) and salinity (> 0.1 M NaCl), but basically overcome by longer after-ripening time.

10. *Thinopyrum junceum* (L.) Á. Löve (*Poaceae*) (Fig. 1i)

Accession data

Si: Riserva Naturale Orientata Oasi del Simeto (Catania), foce del fiume Simeto (WGS84: 37.407266°N, 15.091696°E), 2 m a.s.l., 09 Aug 2010, *A. Lantieri & R. Galesi* (BGS-CT 07AL/RG/10, Banca del Germoplasma delle Specie Spontanee, Università di Catania).

Germination data

Pre-treatments: Disinfection with a 1% sodium hypochlorite (NaClO) water solution for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 3 sheets of sterilized filter paper (Whatman 40), imbibed with 3ml of sterilized distilled water.

Sample size: 100 seeds for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
78.3%	25/15°C	12/12h	6.0	7.9	23.0	9.8
73.3%	25/15°C	0/24h	6.0	4.9	16.0	6.8

Observations

Results of germination tests less than 80% were due to the persistency of embedded seeds which did not show coat rupture and radicle protrusion. Since these seeds included a vital embryo, mechanical strengths by the pericarp are plausible (Mavroeidi 2015). Tests performed with the addition of NaCl water solutions to the germination medium revealed that seed germination of Sicilian plants is negatively affected by salinity. In fact, while low NaCl concentration (0.1 M) did not alter significantly the germination rate (G = 72-75%), increasing salt quantities in the germination medium progressively delayed germination, increased MTG and reduced final germination (65% with 0.2 M NaCl, 52% with 0.3 M, and 30% with 0.5M). Light had no significant effects on final germination; tests carried out under 12/12h photoperiod provided comparable results to full darkness (from 78.3% without salt to 72% with 0.1 M NaCl or less with higher salt concentrations). Ballesteros & al. (2015) reported about 90% of germination with a 12/12 h photoperiod and 20°C constant

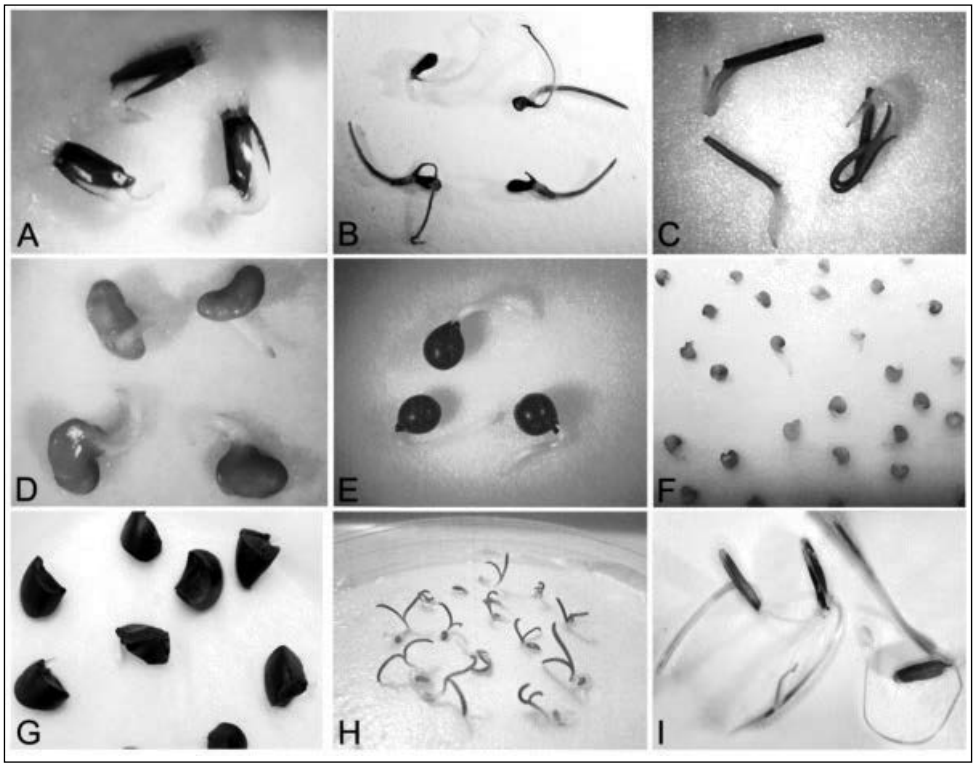

Fig. 1. Germinating seeds of: **a**, *Centaurea sphaerocephala*; **b**, *Cyperus capitatus*; **c**, *Launaea fragilis*; **d**, *Medicago marina*; **e**, *Muscari gussonei*; **f**, *Ononis variegata*; **g**, *Pancratium maritimum*; **h**, *Plantago macrorrhyza*; **i**, *Thinopyron junceum*.

temperature. However, our tests carried out in the same condition provided a final germination percentage of 53%, while it reached 67% under full darkness. Other constant temperatures (15°C and 25°C) yielded germination percentages not higher than 60%, with lower values in the presence of light. Mavroeidi (2015) obtained high germination values in plants from Crete at constant temperature (94.7% at 10°C, 100% at 15°C and 98.7 at 20°C) under full darkness; in these experiments, however, seeds were previously scarified (dispersal unit removed) and this clearly produced their higher and faster germination, because germination at 15°C decreased to 69% in seeds with pericarp.

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M. Porceddu, M. E. Boi & G. Bacchetta

Germination data of four Mediterranean species of coastal sand dunes

Abstract

Porceddu, M., Boi, M. E. & Bacchetta, G.: Germination data of four Mediterranean species of coastal sand dunes [In Magrini, S. & Salmeri, C. (eds), Mediterranean plant germination reports – 1]. Fl. Medit. 29: 288-292. 2019. https://doi.org/10.7320/FlMedit29.288

The present work gathers new germination assays of four Mediterranean species of coastal sand dunes. The studied species are: *Anthemis maritima* L. subsp. *maritima, Crucianella maritima* L., *Eryngium maritimum* L. and *Thinopyrum junceum* (L.) Á.Löve. Seeds were collected at the time of natural seed dispersal in two sites in the South of Sardinia. The germination tests were carried out at the Sardinian Germplasm Bank (BG-SAR). Our results show a high germination capability and germination rate for the tested plant species.

Key words: vascular plants, Mediterranean coasts, dune habitats, Sardinia.

Plant species who colonize coastal dune live in a sensitive equilibrium with some extreme conditions like high salinity, the lack of soil nutrients and wind exposure (Brown & McLachlan 1990). However, Mediterranean coastal dunes are threatened by several factors such as unsustainable tourism and invasive alien species. These threats can modify the dune structure, the vegetation and cause a loss of biodiversity. As a consequence, the knowledge of the best germination condition is important in the broader perspective of conservation action.

This work illustrates new germination data of four species of coastal dunes which are spread throughout the Mediterranean area: *Anthemis maritima* L. subsp. *maritima*, *Crucianella maritima* L., *Eryngium maritimum* L. and *Thinopyrum junceum* (L.) Á.Löve.

11. *Anthemis maritima* L. subsp. ***maritima*** (*Asteraceae*)

Accession data

Sa: Domus de Maria (Sud Sardegna), loc. Porto Campana, Chia (WGS84: 38.887864°N, 8.868983°E), duna costiera, 4 m a.s.l., 09 Jul 2014, *M. Orrù & M. Duran* (BG-SAR-2014-0089, Sardinian Germplasm Bank).

Germination data

Pre-treatments: no treatment.

Germination medium: 1% agar.

Sample size: 60 seeds (20 × 3 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
82.6%	constant 20°C	12/12h	3.0	6.0	35.0	9.5

Observations

Anthemis maritima subsp. *maritima* is a perennial species of white dunes distributed throughout the Mediterranean area in which contributes to coastal sand dune edification and restoration (De Lillis & al. 2004). Seeds of *A. maritima* subsp. *maritima* showed an optimal germination temperature of 20°C; in addition, high germination was obtained also at 15°C reaching value of 72.7%. The results obtained by BG-SAR, testing the seeds at both 15 and 20°C, showed higher germination percentages than reported by Benvenuti (2016) for *A. maritima* collected in Torre Mozza (Livorno, Italy). Very few germination data are present in literature for this species.

12. *Crucianella maritima* L. (*Rubiaceae*)

Accession data

Sa: Villasimius (Cagliari), loc. Simius (WGS84: 39.121822°N, 9.523533°E), duna costiera, 2 m a.s.l., 18 Jul 2011, *M. Orrù & S. Pinna* (BG-SAR-2017-0523, Sardinian Germplasm Bank).

Germination data

Pre-treatments: no treatment.

Germination medium: 1% agar.

Sample size: 100 seeds (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
95.2%	constant 15°C	0/24h	–	–	21.0	–

Observations

Crucianella maritima is a suffruticous chamaephyte considered an important species of the coastal fixed dunes habitat. As reported by Del Vecchio & al. (2012), germination of *C. maritima* seeds is characterized by photoinhibition and intraspecific differences in final germination percentages was highlighted depending on the seed provenance. Germination was scored only at the end of the test (21 days) so it was not possible to calculate T_1, T_{50} and MTG. In accordance with the previously reported data, the results shown here and

obtained from seed collected in Villasimius population highlighted high germination when tested at 15°C in dark conditions. Contrarily, percentages of 73% (at 10°C) and 57% (at 20°C) obtained under light condition (8h light/16h dark) have been reported for *C. maritima* by Royal Botanic Gardens Kew (2019).

13. *Eryngium maritimum* L. (*Apiaceae*)

Accession data

Sa: Domus de Maria (Sud Sardegna), loc. Dune Campana, Chia (WGS84: 38.887650°N, 8.869067°E), duna costiera, 13 m a.s.l., 18 Oct 2011, *M. Orrù* (BG-SAR-2017-0776, Sardinian Germplasm Bank).

Germination data

Pre-treatments: Cold stratification at 5°C for 60 days.

Germination medium: 1% agar.

Sample size: 75 seeds (25 × 3 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
85.9%	constant 15°C	12/12h	11.0	12.0	56.0	12.6

Observations

Eryngium maritimum is a psammophilous plant species growing on sand dunes; although the taxon is listed as widespread species in western and southern Europe, its populations are declining in many areas (van der Maarel & van der Maarel-Versluys 1996). Necajeva & Ievinsh (2013) reported that no seeds of *E. maritimum* from the Kurzeme coast of Latvia on the Baltic Sea germinated within two months without cold stratification and few seeds germinated after only one month of cold stratification; however, the germination percentage increased further after two, three, and four months of cold stratification. Our results are in accordance with the results reported by these authors, in which germination percentage of ca. 85% was obtained only after two months of cold stratification. In general, other studies reported very low germination percentages for the seeds of this species (Walmsley & Davy 1997; Curie & al. 2007). Royal Botanic Gardens Kew (2019) reported percentages of 51 and 67% obtained without a cold pre-treatment; however, these percentages were reached after a long time (154 and 105 days respectively), likely due to the cold stratification requirements for dormancy breaking.

14. *Thinopyrum junceum* (L.) Á.Löve (*Poaceae*)

Accession data

Sa: Domus de Maria (Sud Sardegna), loc. Su Giudeu, Chia (WGS84: 38.884236°N, 8.863153°E), duna costiera, 2 m a.s.l., 22 Jul 2011, *M. Orrù & S. Pinna* (BG-SAR-2017-0708, Sardinian Germplasm Bank).

Germination data

Pre-treatments: no treatment.

Germination medium: 1% agar.

Sample size: 80 seeds for each test (20 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
91.3%	constant 20°C	12/12h	3.0	4.0	21.0	5.5
88.8%	constant 15°C	12/12h	4.0	9.0	15.0	7.4

Observations

Thinopyrum junceum is one of the most common species of Mediterranean coastal areas growing in particular in the foredune systems (Feola & al. 2011). Our germination tests permitted to define the best germination temperatures of *T. junceum* seeds of a Sardinian population. In details, mature seeds of this species germinated without any specific treatment and high germination percentages were obtained at 15 and 20°C within 21 days.

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S. Magrini, D. Barreca, L. Neri, A. Iacomino, F. Barbarese, S. Superchi & L. Zucconi

Seed germination protocols for six dune species from the Tyrrhenian coast (central Italy)

Abstract

Magrini, S., Barreca, D., Neri, L., Iacomino, A., Barbarese, F., Superchi, S. & Zucconi, L.: Seed germination protocols for six dune species from the Tyrrhenian coast (central Italy) [In Magrini, S. & Salmeri, C. (eds), Mediterranean plant germination reports – 1]. Fl. Medit. 29: 293-302. 2019. https://doi.org/10.7320/FlMedit29.293

Successful germination protocols for the following six dune species are presented: *Centaurea sphaerocephala* subsp. *sphaerocephala*, *Jacobaea maritima* subsp. *maritima*, *Matthiola sinuata*, *Medicago marina, Pancratium maritimum*, and *Silene canescens*. Seeds were collected in sand dunes along the Tyrrhenian coasts in south Tuscany and north Latium (central Italy). The germination ability was tested at the Tuscia Germplasm Bank (BGT) at six constant temperatures (from 5 to 30°C), under both light (with a 12/12h photoperiod) and total dark. Our results show a high germination ability and germination rate for the tested species.

Key words: coastal dunes, Latium, photoinhibition, psammophytes, Tuscany.

Mediterranean coastal dunes are dynamic and threatened environments, particularly vulnerable being subjected to the strong impact of human activities and in need of conservation actions. Research focused on the assessment of germination requirements of dune species are carried out at the Tuscia Germplasm Bank where over one hundred seed accessions of thirty species collected in Central Italy dunes are long term preserved. Here we present successful germination protocols for six species occurring in the Tyrrhenian sandy dunes: *Centaurea sphaerocephala* L. subsp. *sphaerocephala*, *Medicago marina* L., *Pancratium maritimum* L., and new data for Italy of *Jacobaea maritima* (L.) Pelser & Meijden subsp. *maritima*, *Matthiola sinuata* (L.) W.T.Aiton, and *Silene canescens* Ten.

15. *Centaurea sphaerocephala* L. subsp. ***sphaerocephala*** (*Asteraceae*) (Fig. 1a)

Accession data

It: Latium. Tarquinia (Viterbo), S. Agostino (WGS84: 42.173710°N, 11.738198°E), duna costiera, 3 m a.s.l., 4 Sept 2017, *S. Magrini & D. Barreca* (BGT-A-43017, Tuscia Germplasm Bank).

Germination data
Pre-treatments: sterilization with a solution of 5% sodium hypochlorite + Tween 20 for 5 minutes, followed by 3 rinses in sterile distilled water.

Germination medium: 1% agar.

Sample size: 100 achenes for each test (20 × 5 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
100%	constant 25°C	0/24h	1.0	2.4	7.7	2.9
100%	constant 25°C	12/12h	1.0	3.4	9.0	3.9
100%	constant 20°C	12/12h	1.0	1.8	7.7	3.3
100%	constant 20°C	0/24h	1.0	3.5	8.7	3.9
96.5%	constant 15°C	0/24h	4.0	3.7	9.7	4.9
95.0%	constant 15°C	12/12h	4.0	3.8	8.7	4.9
91.7%	constant 30°C	12/12h	1.0	2.6	11.7	4.1
90.0%	constant 30°C	0/24h	1.0	2.4	12.0	3.5
88.0%	constant 10°C	0/24h	5.0	7.1	17.0	7.6

Observations
Centaurea sphaerocephala is a perennial species of foredunes and fixed dunes, quite common in Central and South Italy (Acosta & Ercole 2015). High germination (> 72%) was obtained at a wide range of temperatures (10-30°C), independently from the applied light/dark conditions. Lower germination percentages have been reported for this species, 79% by Menini & al. (2014) for Tuscany (Italy) and 82% by Royal Botanic Gardens Kew (2019), both at 20°C under an 8/16h photoperiod.

16. *Jacobaea maritima* (L.) Pelser & Meijden subsp. ***maritima*** (*Asteraceae*) (Fig. 1b)

Accession data
It: Tuscany. Grosseto (Grosseto), loc. Collelungo, Marina di Alberese (WGS84: 42.638073°N, 11.067104°E), duna costiera, 3 m a.s.l., 30 Jun 2017, *S. Magrini & D. Barreca* (BGT-A-41717, Tuscia Germplasm Bank).

Germination data
Pre-treatments: sterilization with a solution of 5% sodium hypochlorite + Tween 20 for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 1% agar.

Sample size: 100 achenes for each test (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
81.5%	constant 15°C	12/12h	4.3	6.6	21.0	8.4

Observations
Jacobaea maritima subsp. *maritima* is a perennial dune species with a West Mediterranean distribution. Here we report the first germination data for this species for Italy. Germination percentages higher than 70% were obtained only at 15-25°C in the light, with the lowest percentages at 30°C in the dark (14.6%) and at 5°C in the light (18.9%). Light-promoted seed germination highlighted by Van der Meijden & Van der Waals-Kooi (1979) for a Dutch population of *Senecio jacobaea* L. can be confirmed in this species only at temperatures higher than 15°C, while photoinhibition was found at 10°C and 5°C.

17. *Matthiola sinuata* (L.) W.T.Aiton (*Brassicaceae*) (Fig. 1c)

Accession data
It: Latium. Tarquinia (Viterbo), S. Agostino (WGS84: 42.173365°N, 11.738726°E), duna costiera, 2 m a.s.l., 4 Sept 2017, *S. Magrini & D. Barreca* (BGT-A-43117, Tuscia Germplasm Bank).
It: Tuscany. Castiglione della Pescaia (Grosseto), loc. Roccamare, presso il fosso Tonfone (WGS84: 42.773051°N, 10.823768°E), duna costiera, 1 m a.s.l., 31 Aug 2014, *S. Magrini* (BGT-A-31114, Tuscia Germplasm Bank).

Germination data
Pre-treatments: sterilization with a solution of 5% sodium hypochlorite + Tween 20 for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 1% agar.

Sample size: 100 seeds for each test (20 × 5 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]	Accession code
90.7%	constant 25°C	12/12h	2.0	3.6	12.0	3.8	BGT-A-43117
86.7%	constant 25°C	12/12h	3.0	5.3	15.0	7.6	BGT-A-31114
85.0%	constant 25°C	0/24h	1.7	3.6	17.3	4.3	BGT-A-31114

Observations

Matthiola sinuata is a short-lived (biennial) species typical of the sand beach drift-lines, occurring in Italy mainly along the Tyrrhenian coasts (Acosta & Ercole 2015). Here we report the first germination data for this species for Italy. Germination tests using six constant temperature regimes (5-30°C) showed that 25°C is the optimum temperature for both the accessions, the same temperature reported also by Pérez-García & al. (2007) for Spain. High germination (> 70%) was obtained only at warmer temperatures (20-30°C), independently from the applied photoperiod, unlike Royal Botanic Gardens Kew (2019) which reported the highest percentage (90%) at 11°C and at 25°C a lower percentage (78%) than those reported here.

Strong photoinhibition has been found in other dune species of the family *Brassicaceae*, like *Matthiola tricuspidata* (L.) R.Br., *Cakile maritima* Scop., and *Malcolmia littorea* (L.) R.Br. (Thanos & al. 1991, 1994; De Vitis & al. 2014, 2018; Carta & al. 2017). On the other hand, our results highlighted a strong photoinhibition of seed germination in *M. sinuata* exclusively at temperatures below 10°C and only a weak effect at 15-20°C.

18. *Medicago marina* L. (*Fabaceae*) (Fig. 1d)

Accession data

It: Tuscany. Grosseto (Grosseto), loc. Rosmarina, Marina di Grosseto (WGS84: 42.726583°N, 10.969202°E), duna costiera, 4 m a.s.l., 24 Jul 2016, *S. Magrini* (BGT-A-39416, Tuscia Germplasm Bank).

Germination data

Pre-treatments: manual scarification by sandpaper; washing with sterile distilled water + Tween 20 for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 1% agar.

Sample size: 100 seeds for each test (20 × 5 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
100%	constant 20°C	12/12h	5.0	5.9	20.3	9.6
100%	constant 30°C	0/24h	1.7	8.1	43.7	20.5
100%	constant 5°C	0/24h	10.0	16.9	44.7	21.1
97.6%	constant 5°C	12/12h	13.3	16.3	67.3	24.6
95.2%	constant 30°C	12/12h	3.3	6.8	28.0	10.7
92.6%	constant 10°C	12/12h	8.3	11.1	41.0	18.8
90.5%	constant 20°C	0/24h	2.0	1.9	39.3	6.9
88.1%	constant 10°C	0/24h	5.0	5.6	18.0	13.3
88.1%	constant 15°C	0/24h	2.3	2.8	39.0	7.2
87.5%	constant 15°C	12/12h	5.0	5.5	25.0	9.0
86.1%	constant 25°C	0/24h	1.0	2.0	7.0	3.3
83.9%	constant 25°C	12/12h	3.0	4.9	36.3	11.4

Observations

Medicago marina is a perennial species characteristic of the embryonic shifting dunes, common along the Italian coasts (Acosta & Ercole 2015). As for many other genera of Fabaceae, *Medicago* seeds are characterized by a physical dormancy due to the impermeable coat. In accordance with Scippa & al. (2011), mechanical scarification by sandpaper improved seed germination, with high percentages (≥ 83.9%) at all the tested conditions (six constant temperatures from 5 to 30°C, both in the light and in the dark). In particular, the fastest germination was recorded at 25°C in darkness (T_1 = 1 day, T_{50} = 2 days and a total of 7 days for the maximum germination), even if with a lower germination percentage than reported, under the same conditions, by Scippa & al. (2011) for seeds from Adriatic coastal dunes (south Italy). The slowest germination was recorded at 5°C under light condition.

19. *Pancratium maritimum* L. (*Amaryllidaceae*) (Fig. 1e)

Accession data

It: Latium. Tarquinia (Viterbo), loc. Saline (WGS84: 42.210943°N, 11.708386°E),

duna costiera, 2 m a.s.l., 25 Sept 2016, *A. Caldelli & S. Magrini* (BGT-A-39816, Tuscia Germplasm Bank).

Germination data
Pre-treatments: sterilization with a solution of 5% sodium hypochlorite + Tween 20 for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 1% agar.

Sample size: 60 seeds for each test (20 × 3 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
97.8%	constant 20°C	0/24h	12.0	15.3	23.7	14.9
97.8%	constant 15°C	12/12h	20.3	36.0	55.3	35.7
88.9%	constant 15°C	0/24h	24.3	32.3	58.3	31.9
82.2%	constant 30°C	0/24h	14.3	23.9	63.3	27.7

Observations

Pancratium maritimum is a species of coastal dunes, with a wide distribution along the Italian sandy coasts even if overcollection, urbanization, and tourism put serious threats to the species, resulting in a significant decrease of its populations (De Castro & al. 2012; Giovino & al. 2015). To date, these are the best germination data for this species for Latium. Our germination tests defined an optimal temperature range of 15–20°C, in accordance with data previously reported for Sardinia (95-98% at 15-20°C, in the dark; Bacchetta & al. 2007). Moreover, at all the temperatures in the dark (except 20°C), higher germination percentages have been recorded than those reported by Balestri & Cinelli (2004) for Tuscany (53% vs. 0% at 5°C, 80% vs. 25% at 10°C, 89% vs. 50% at 15°C, and 82% vs. 25% at 30°C) and by De Lillis & al. (2004) for Latium (60% with a longer T_1 at 22/15°C with a 14/10h photoperiod).

Our results, however, highlighted the ability of *P. maritimum* seeds to germinate in a wide range of temperatures (5-30°C) both in the dark (53-98%) and in the light (7-98%). Moreover, we observed a strong photoinhibition of seed germination exclusively at 5°C, with seeds weakly photoinhibited at T > 20°C, in accordance with Carta & al. (2017), and with no inhibition at 10-15°C.

20. *Silene canescens* Ten. (*Caryophyllaceae*) (Fig. 1f)

Accession data

It: Latium. Montalto di Castro (Viterbo), Montalto Marina (WGS84: 42.321758°N, 11.585935°E), duna costiera, 3 m a.s.l., 12 May 2016, *S. Magrini & D. Barreca* (BGT-A-36516, Tuscia Germplasm Bank).

Germination data

Pre-treatments: sterilization with a solution of 5% sodium hypochlorite + Tween 20 for 5 minutes followed by 3 rinses in sterile distilled water.

Germination medium: 1% agar.

Sample size: 100 seeds for each test (20 × 5 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
100%	constant 20°C	12/12h	1.3	1.3	6.0	2.4
98.3%	constant 20°C	0/24h	1.0	1.3	11.3	2.3
98.3%	constant 25°C	0/24h	1.0	0.5	5.3	1.4
96.7%	constant 25°C	12/12h	1.0	0.7	3.3	1.4
95.0%	constant 15°C	12/12h	1.0	1.4	16.3	2.7
93.3%	constant 15°C	0/24h	1.0	0.7	9.3	1.7
91.7%	constant 10°C	0/24h	6.0	5.6	15.0	6.7
90.2%	constant 30°C	0/24h	1.0	0.5	3.0	1.1
90.0%	constant 30°C	12/12h	1.0	0.6	10.3	1.9

Observations

Silene canescens (= *S. colorata* Poir.) is an annual Mediterranean species of foredunes and fixed dunes, quite common along the Italian sandy coasts (Acosta & Ercole 2015). Here we report the first germination data for this species for Italy. Successful and fast ger-

mination (> 90%, T_{50} = 0.5-1.4) was obtained at a wide range of constant temperatures (15-30°C), in accordance with Royal Botanic Gardens Kew (2019), with germination percentages much higher than those reported for Portugal: 0% at 15-20°C and 28% at 25°C, under light condition (Marques & al. 2007). The lowest germination percentages were recorded at 5°C, where strong photoinhibition of seed germination was observed (Carta & al. 2017).

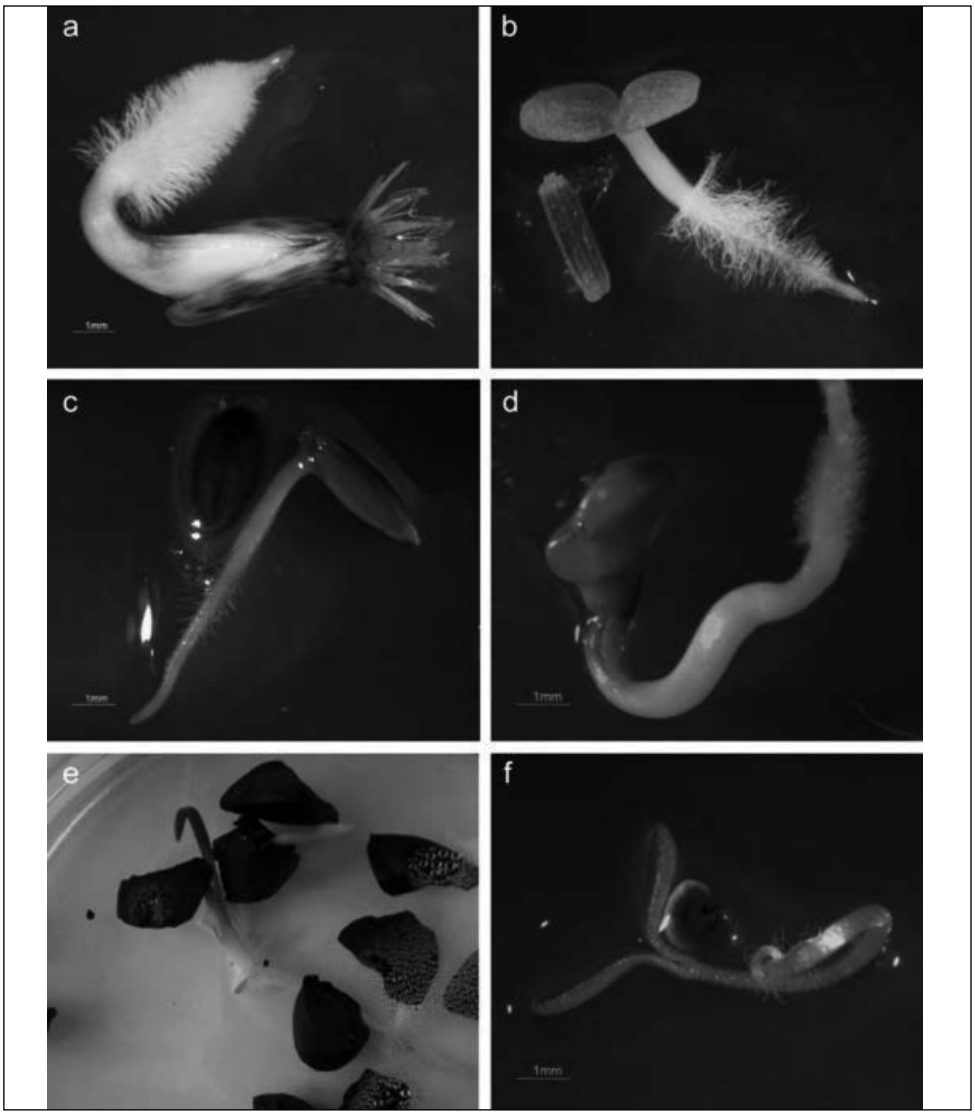

Fig. 1. Germinated seeds of: **a,** *Centaurea sphaerocephala* subsp. *sphaerocephala*; **b,** *Jacobaea maritima* subsp. *maritima*; **c,** *Matthiola sinuata*; **d,** *Medicago marina*; **e,** *Pancratium maritimum*; **f,** *Silene canescens*.

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Giuseppe Fabrini

Seed germination data of *Sporobolus aculeatus* (*Poaceae*) and *Juncus subulatus* (*Juncaceae*)

Abstract

Fabrini, G.: Seed germination data of *Sporobolus aculeatus* (*Poaceae*) and *Juncus subulatus* (*Juncaceae*) [In Magrini, S. & Salmeri, C. (eds), Mediterranean plant germination reports – 1]. Fl. Medit. 29: 303-306. 2019. https://doi.org/10.7320/FlMedit29.303

The most significant results of seed germination of *Sporobolus aculeatus* (L.) P.M.Peterson and *Juncus subulatus* Forssk. are reported. The seeds were collected from the Natural Monument "Palude di Torre Flavia" in Latium. The highest germination value (92.0%) for *S. aculeatus* seeds were obtained after 2 months of cold stratification and then incubation at an alternating temperature of 20/10°C in continuous darkness. For *J. subulatus*, the best germination value (95.0%) were obtained with alternating temperatures, 20/10°C and 15/6°C, under a 12/12h (light/dark) photoperiod.

Key words: cold stratification, degradation of wetland habitats, Latium, retrodunal humid depressions.

21. *Sporobolus aculeatus* (L.) P.M.Peterson (*Poaceae*) (Fig. 1)

Accession data

It: Latium. Ladispoli, Cerveteri (Roma), loc. Monumento Naturale "Palude di Torre Flavia" (WGS84: 41.963194°N, 12.044944°E), area umida, 0 m a.s.l., 7 Oct 2008, *M. Giovannetti & G. Fabrini* (BGR-1431028S07101014A2 12, Rome Germplasm Bank).

Germination data

Pre-treatments: cold stratification in moistened sand (0, 1, 2 months at 5°C) (Côme 1970).

Germination medium: Petri dishes with Whatman Filter Papers N.2 soaked in distilled water.

Sample size: 120 seeds for each test (30 × 4 replicates).

Germination	Stratification [months]	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
92.0%	2	20/10°C	0/24h	3.0	9.0	9.0	8.5
86.0%	1	20/10°C	12/12h	3.0	15.0	27.0	14.2
85.0%	2	20/10°C	12/12h	3.0	12.0	24.0	11.6

Observations

The degradation of wetland habitats is becoming more and more sensitive worldwide. This degradation also affects the territory included in the municipalities of Ladispoli and Cerveteri, which houses the Natural Monument "Palude di Torre Flavia" and the studied species. The progressive reclamation, urbanization and the continuous withdrawal of groundwater have created an alteration of the hydrological regime, with serious consequences for habitats and ecosystems (Battisti 2006).

The seeds of *S. aculeatus* are of the orthodox type, with a moisture content of ca. 8.4%. They are subject to a physiological dormancy (Baskin & Baskin 1998). In the studied area, the species completes the entire biological cycle between March and September. *S. aculeatus* colonizes retrodunal humid depressions areas and lives in association with many species typical of fresh and brackish water environments, in particular *Bolboschoenus maritimus* (L.) Palla.

Seed germination tests were conducted at 5°C, 20/10°C, 15/6°C, 25/15°C (12 h light/12 dark and continuous dark) (Bacchetta & al. 2006). The results obtained from this study contribute to a better knowledge of seed germination ecology of this annual species, Critically Endangered in Latium (Conti & al. 1997), as no data on its germination were found in the literature.

22. *Juncus subulatus* Forssk. (*Juncaceae*) (Fig. 2)

Accession data

It: Latium. Ladispoli, Cerveteri (Roma), loc. Monumento Naturale "Palude di Torre Flavia" (WGS84: 41.963194°N, 12.044944°E), area umida, 0 m a.s.l., 10 Oct 2010, *M. Giovannetti & G. Fabrini* (BGR-1338028S10100911 A4, Rome Germplasm Bank).

Germination data

Pre-treatments: cold stratification in moistened sand (0, 1, 2, 3 months at 5°C) (Côme 1970).

Germination medium: Petri dishes with Whatman Filter Papers N.2 soaked in distilled water.

Sample size: 100 seeds (25 × 4 replicates for each test).

Germination	Stratification [months]	Thermoperiod	Photoperiod [light/dark]	T_1 [d]	T_{50} [d]	T_{max} [d]	MTG [d]
95.0%	0	15/6°C	12/12h	12.0	12.0	24.0	13.0
95.0%	0	20/10°C	12/12h	6.0	9.0	24.0	9.4
86.0%	0	25/15°C	12/12h	6.0	6.0	12.0	8.4
85.0%	3	15/6°C	12/12h	12.0	12.0	24.0	13.9

Observations

Juncus subulatus is a perennial rhizomatous geophyte. It is found in marshes, wet sands and moist grounds in salt and freshwater. It is native to the Mediterranean and Irano-Turanian Regions and occurs in central and south Italy (Bartolucci & al. 2018). In the IUCN Red List of Threatened Species, it is listed as Least Concern (LC) at the global and Mediterranean level as it is widespread with stable populations and does not face any major threats (de Belair & Lansdown 2014).

Seed germination tests were conducted at 5°C, 20/10°C, 15/6°C and 25/15°C (12 h light/12 h dark and continuous dark) (Bacchetta & al. 2006). The obtained results showed that *J. subulatus* produces non-dormant seeds, which are also markedly affected by photoinhibition, as highlighted by the almost total absence of germination in dark conditions at all tested thermoperiods.

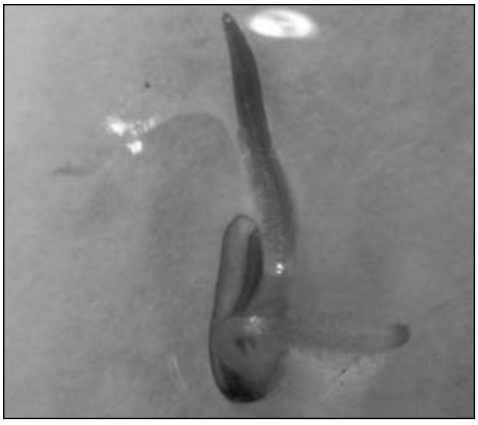

Fig. 1. Germinated seeds of *Sporobolus aculeatus*.

Fig. 2. Germinated seed of *Juncus subulatus*.

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Mediterranean plant karyological data – 29

edited by G. Kamari, C. Blanché & S. Siljak-Yakovlev

Abstract

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This is the twenty-nine of a series of karyological data from Mediterranean area, peri-Alpine communities and the Atlantic Islands, in English or French language. It comprises contributions on 20 taxa: *Allium*, *Ficaria*, *Freesia*, *Paeonia*, *Romulea* from Greece by Liveri, E., Katopodi, E. & Kamari, G. (Nos 1962-1967); *Delphinium*, *Hesperis*, *Silene* from Spain by Bosch, M., Sàez, L., Simon, J. & Blanché C. (Nos 1968-1971); *Vicia* from Morocco by Hormat, K. Tahiri, H., Guennoun, N. & Gounssa, A. (Nos 1972-1975); *Allium*, *Ptilostemon*, *Salvia* from Italy and Sicily by Salmeri, C. (Nos 1976-1981).

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Karyosystematic study of some taxa from the Ionian floristic region (Greece). II

Abstract

Liveri, E., Katopodi, E. & Kamari, G.: Karyosystematic study of some taxa from the Ionian floristic region (Greece). II. [In Kamari, G., Blanché, C. & Siljak-Yakovlev, S. (eds), Mediterranean plant karyological data - 29]. – Fl. Medit. 29: 308-320. http://dx.doi.org/10.7320/FlMedit29.308

A karyosystematic study of six interesting taxa from the Ionian Islands (Greece) is presented. The chromosome number (2*n* = 16) and karyotype analysis for two ecologically different populations of the Greek endemic *Allium callimischon* subsp. *callimischon* are given and its karyotype variation is also confirmed. The chromosome number (2*n* = 16) and karyotype morphology of *Allium flavum* subsp. *tauricum* is reported for the first time in material from Ionian Islands. Karyotype analysis of *Ficaria calthifolia* (2*n* = 16) is given for first time in Greek material. Chromosome data (2*n* = 22) for *Freesia leichtlinii* subsp. *alba* are presented for the first time for this taxon. Previous karyological references of *Paeonia mascula* subsp. *russoi* (2*n* = 10) confirm our findings from a new population from north Kefallinia isl. The first karyological attempt at *Romulea bulbocodium* in Greece is performed here, resulting in two ploidy levels (2*n* = 3x = 33 and 2*n* = 4x = 44) for, ecologically, different populations and additionally the basic chromosome number of the genus is discussed.

Keywords: *Allium callimischon*, *A. flavum*, *Ficaria calthifolia*, *Freesia leichtlinii*, *Paeonia mascula*, *Romulea bulbocodium*, karyology, distribution, taxonomy.

Introduction

The present article is the second contribution to the flora of the Ionian floristic region, under the same title, also published in Flora Mediterranea journal by the research team of D. Phitos & G. Kamari. The reasons of this effort are mentioned in the introduction of the first article (Bareka & al. 2018).We hope that this effort will be continued in a follow-up project.

1962. ***Allium callimischon*** Link subsp. ***callimischon*** — 2*n* = 2x = 16 (Figs 1A, 1B & 3A, 3B).

Gr: Ins. Lefkada: Regio insulae borealis, Gyra dicta; in stagnosis, alt. 0-1 m, 20º 40′ 54.77′′ E, 38º 50′ 20.70′′ N, 05.XI.2017, leg.: *D. Phitos, G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29383*, UPA). – Figs 1A & 3A.

— Ins. Lefkada: Mons Stavrotas, ad cacumen Elati, in petrosis calc., alt. ca. 1100 m, 20°

38′ 47.38′′ E, 38° 42′ 27.14′′ N, 27.IX.2017, leg.: *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29428*, UPA). – Figs 1B & 3B.

Allium callimischon (*Amaryllidaceae*) consists of two subspecies, according to Stearn (1978):

(a) subsp. *callimischon* (Figs 1A, 1B & 1C), endemic to Greece, described from NE Peloponnisos, has a wide distribution in the whole Peloponnisos, especially in the eastern (Constantinidis & Kalpoutzakis 2016) and southern part (Alibertis pers. com.); it also occurs on Kithira isl., W Ipiros, Ionian Islands (Tzanoudakis & al. 1991) and W Sterea Ellas (Bareka & al. 2000).

(b) subsp. *haemostictum* Stearn (Fig. 1D), restricted in Crete (Stearn 1978) and probably in SW Anatolia (Özhatay & Özhatay 1981).

The examined material of *Allium callimischon* subsp. *callimischon* was collected from Lefkada isl. The distribution of this subspecies in Lefkada is rather wide, extending from the sea level up to 1100 m. One of the observed populations occurs at the northern part of the island, specifically at the area of Gyra, in alt. 0-1 m (Fig. 1A). On Mt. Stavrotas, we observed three subpopulations: on the summit Elati (Fig. 1B), at the area of Agios Donatos and above the village Eglouvi. Moreover, in the eastern part of Lefkada isl. we also noticed two more subpopulations; above the village Nikiana (alt. 10-20 m) and in the clearing of the forest Skari (alt. ca. 600 m).

The cytotaxonomical study, hereby, includes plants from the two different habitats of the island. The first population of *A. callimischon* subsp. *callimischon* was collected at the area Gyra, on stagnate places (Fig. 1A), whereas the second population grows on the summit Elati of Mt. Stavrotas, on rocky limestone places (Fig. 1B). The above mentioned different ecological conditions of the two examined populations (i.e. altitude and substrate) have resulted in a different flowering time and size of the plants. More specifically, the plants of Elati are flowering from the end of August until the end of September and their stems size does not exceed 15 cm, whereas the plants of Gyra area are flowering from the end of October until mid-November and the stem height can reach up to 40 cm.

It is noteworthy that in the taxonomical keys for the two subspecies, given by Stearn (1978), only the difference in the shape of the spots on the upper part of the tepals is mentioned. As we have observed in a great amount of material from Lefkada, but also from all the distributional range of the taxon, the two subspecies differ additionally in the tepal shape and mainly in the ovary colour (mentioned here for the first time); green to yellow-green in the typical one (Figs 1A, 1B & 1C), whereas always dark reddish (colour of blood) in subsp. *haemostictum* (Fig. 1D).

The two karyologically examined populations of *A. callimischon* subsp. *callimischon* from Lefkada isl. are found to be diploids, with $2n = 16$ chromosomes (Figs 3A & 3B). The same chromosome number for the typical subspecies has been also given by Stearn (1978), Johnson (1982) and Iatrou (1986) in material from Peloponnisos, Tzanoudakis (1983a) and Tzanoudakis & al. (1991) in material from Peloponnisos, Lefkada, Ipiros and Kithira island and Bareka & al. (2000) in material from Sterea Ellas.

Concerning the chromosome morphology of *A. callimischon* subsp. *callimischon*, Johnson (1982), Tzanoudakis (1983a) and Bareka & al. (2000) give figures and more details. Tzanoudakis (1983a) highlights the great karyotype variation of all the examined

populations. The karyotype formula given by Bareka & al. (2000) is 2*n* = 12m + 2m/sm + 2sm = 16 chromosomes.

The karyotypes of both examined populations are symmetrical, consisting mostly of metacentric chromosomes. The karyotype of Gyra population includes two pairs of marker-chromosomes: one pair is metacentric with a clear secondary restriction close to the centromere and one satellited pair with an additional secondary restriction close to the centromere (not always visible on both chromosomes) (Fig. 3A). On the contrary, the karyotype of the population from Elati has 4 marker-chromosomes with a strong secondary restriction close to the centromere, lacking, however, the small spherical satellites (Fig. 3B). Moreover, the karyotype of Elati population consists of chromosomes varying in size from 10.23 to 15.35 μm, while the chromosomes of Gyra population's karyotype vary from 12.22 to 19.44 μm.

1963. ***Allium flavum*** subsp. ***tauricum*** (Besser ex Rchb.) K. Richt — 2*n* = 2x = 16 (Figs 1E & 3C).

Gr: Ins. Lefkada: Mons Mega Oros, supra pagum Eglouvi in regione Striftouna; in petrosis, alt. ca. 880 m, 20° 37′ 38.45′′ E, 38° 43′ 35.15′′ N, 18.VI.2016, *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29116*, UPA).

Stearn (1978) recognizes under *Allium flavum* (*Amaryllidaceae*) two subspecies:

(a) subsp. *flavum*, distributed in S Europe and C Europe, from France to Turkey (Euro+Med 2006-).

(b) subsp. *tauricum*, distributed in SE Europe from Greece to European Russia (Euro+Med 2006-).

The geographical distribution of *Allium flavum* s.l. in Greece is notably unclear. Especially, for the Ionian floristic region, Dimopoulos & al. (2013) give only the presence of the typical subspecies, whereas the Flora Ionica Working Group (2016) refers to subsp. *tauricum* in Lefkada isl. and particularly from Mt. Stavrotas.

The taxonomical examination of our studied material from Mt. Stavrotas attributes it with certainty to subsp. *tauricum* (Fig. 1E).

The karyological studies of *Allium flavum* s.l. remain similarly unresolved, like their taxonomical relationships within the species. Many chromosome data with 2*n* = 16 and 2*n* = 32 are referred to *A. flavum* s.l., without further discrimination into a subspecies. Tzanoudakis & Vosa (1988) report both diploid and tetraploid populations from N and S Greece. Populations of *A. flavum* s.l. from Serbia are given as tetraploids (2*n* = 32) with B-chromosomes observed in some individuals (Vujošević & al. 2013). In total, 26 reports for chromosome numbers of *A. flavum* s.l. have been given mainly for European populations showing both diploids (2*n* = 16+0-2B) and tetraploids (2*n* = 32) (for more references see the Missouri Botanical Garden Database - Tropicos.org & PhytoKaryon - Kamari & al. 2017).

Concerning the subsp. *tauricum*, Baltisberger & Baltisberger (1995) report in material from adjacent Albania the chromosome number 2*n* = 16+1B and give a karyotype, consisting of meta- to submetacentric chromosomes, four of which are satellited. Also,

Fig. 1. *Allium* taxa studied: **A**, *Allium callimischon* subsp. *callimischon* on stagnate place (Gyra) and **B**, on rocky limestone (Mt. Stavrotas) from Lefkada island; Inflorescences of *A. callimischon*: **C**, subsp. *callimischon* from Lakonia (Peloponnese) and **D**, subsp. *haemostictum*from Crete; **E**, Individuals of *Allium flavum* subsp. *tauricum* from Mt. Mega Oros, Lefkada. – Photos C & D by A. Alibertis.

Karavokyrou (1994) reports $2n = 16$ for populations of subsp. *tauricum* from Lesvos isl. (East Aegean). However, Özhatay (1990) describes a tetraploid karyotype ($2n = 4x = 32$), in material from Turkey, consisting of five sets of metacentrics, one set of submetacentric and two set of acrocentrics, with one of them bearing a satellite on the short arm.

The karyological study of *A. flavum* subsp. *tauricum* from Lefkada is the first report for the Ionian floristic region. The examined material is diploid ($2n = 16$), has always a symmetrical karyotype with all chromosomes being metacentric, varying in size from 9.44 to 17.78 μm and at least two pairs bearing small spherical satellites (one of them occurring on the longer arm) not always visible. The karyotype formula is: $2n = 2x = 12m + 4m\text{-SAT} = 16$ chromosomes (Fig. 3C). No B-chromosomes have been observed in our material, but

several reports mention one or two of them (Cheshmedzhirev 1971; Loidl 1979; Baltisberger & Baltisberger 1995; Krahulcová 2003; Vujosĕvić & al. 2013).

1964. *Ficaria calthifolia* Rchb. — 2*n* = 2x = 16 (Figs 2A & 3D).

Gr: Ins. Kephallinia (Cephalonia vet.): Mons Aenos: ad cacumen Roudi, in loco Thea dicto; in apertis silvae Abietis cephalonicae, solo petroso, alt. 1055 m, 20° 36′ 59.68′′ E, 38° 11′ 24.03′′ N, 15.II.2017, leg.: *D. Phitos & D. Spanou* (Herb. Phitos & Kamari *29382*, UPA).

The genus *Ficaria* Guett. (*Ranunculaceae*) is known for its great morphological polymorphism and the subsequent confusion created by the use of different synonymies (*Ficaria* or *Ranunculus* L. etc.). Molecular data support the discrimination of *Ficaria* and *Ranunculus* into two different genera (Paun & al. 2005).

The *Ficaria verna* complex in Ionian Islands (e.g. Corfu, Lefkada, Kefallinia, etc.) is the case of a very variable group within the genus. According to Gutermann & al. (2014) and Flora Ionica Working Group (2016), at least three taxa of *Ficaria verna* agg. can be identified as being present on Ionian Islands: *F. calthifolia* as diploid, *F. verna* Huds. as tetraploid and *F. chrysocephala* (P. D. Sell) Galasso, Banfi & Soldano as tri/tetraploid.

However, the karyological reports for *F. calthifolia* in its wider distribution give several levels of polyploidy: 2*n* = 16 in material from Bosnia & Herzegovina, Croatia, Slovenia, 2*n* = 16+0-9B, 24, 40 in material from Croatia and 2*n* = 32 in material from Croatia and Slovenia (Papes & Trinajstic 1981, Druskovic & Lovka 1995, Lovka 1995). More chromosome counts are reported under the synonyms of the species e.g. 2*n* = 16 and 2*n* = 16+1-8B as *Ranunculus ficaria* subsp. *calthifolius* Arcang. in material from Armenia, Poland, Hungary, Bulgaria, Yugoslavia (Pogan & Wcisslo 1973, 1974, 1975, 1981, 1986) and Italy (Miceli & Garbari 1976, Capineri & al. 1978).

The studied population of *F. calthifolia* from the summit Roudi of Mt. Aenos shows diploid chromosome number 2*n* = 16. The karyotype is rather asymmetrical varying in size from 5.00-10.36 μm, consisting of 2*n* = 2x = 4m + 6sm + 4st + 2t-SAT= 16 chromosomes (Fig. 3D). The karyotype includes a SAT-chromosome pair, bearing large satellites that often are separated from the arm and, thus may seen as B-chromosomes. This chromosome count is the first for Greek material.

1965. *Freesia leichtlinii* subsp. ***alba*** (G. L. Mey.) J. C. Manning & Goldblatt — 2*n* = 2x = 22 (Figs 2B & 3E).

Gr: Ins. Kephallinia (Cephalonia vet.): Pars occidentalis insulae, supra sinu Lagadakia; in nanofruticetosis, terramroseam, alt. ca. 20 m, 20° 38′ 47.38′′ E, 38° 42′ 27.14′′ N, 16.II.2017, leg.: *D. Phitos& P. Minetos* (Herb. Phitos & Kamari *29385*, UPA).

The genus *Freesia* (*Iridaceae*) includes about 16 species the eastern tropical and southern Africa as their center of distribution (Goldblatt & al. 2006). *F. leichtlinii* is native to

South Africa, but it is found to grow naturalized in the wild also in other continents (Manning & Goldblatt 2001, 2010; Kim 2014). In Greece, it is also found as naturalized, usually growing at low altitude at open shiny stony places. Its occurrence in the Greek area cannot be considered frequent or at least it is not often collected. In Ionian Islands it is known from the islands Kefallinia, Lefkada, Paxi etc.

Freesia leichtlinii subsp. *alba* (Fig. 2B) is a geophyte, producing underground fleshy buds and arising from an underground corm. It grows from autumn to winter, flowering in early spring and dying back to the ground and remaining dormant in summer (Kim 2014). More information for *F. leichtlinii*, but also generally for the genus *Freesia* are offered in the exhaustive article of Manning & Goldblatt (2010).

As mentioned by previous researchers, the basic chromosome number of this genus is x = 11 (Taylor 1926, Goldblatt & Takei 1977, Wongchaochant & al. 2002, Goldblatt & al. 2006 etc.). Only one chromosome count ($2n = 22$) has been reported for *F. leichtlinii*, but no more details for its karyotype were provided then (Goldblatt 1982).

The chromosome number of the examined material is $2n = 22$ with a diploid, symmetrical karyotype, consisting of metacentric and submetacentric chromosomes; the largest pair bears satellites (Fig. 3E). The size of the chromosomes varies from 1.47 to 2.06 μm. The given chromosome data comprise the first report not only for *F. leichtlinii* subsp. *alba*, but for the whole genus in the Greek area. Probably, this happens because *Freesia* does not attract particular interest in Greece, as it is considered naturalized.

1966. *Paeonia mascula* subsp. ***russoi*** (Biv.) Cullen & Heywood — $2n = 2x = 10$ (Figs 2C & 3F).

Gr: Ins. Kephallinia (Cephalonia vet.): Inter pagum Neochori et sinum Giagana, alt. ca. 70 m, 20° 37′ 02.30′′ E, 38° 20′ 43.50′′ N, 27.IV.2015, leg.: *D. Phitos, N. Katsouni & V. Karagianni* (Herb. Phitos & Kamari *29441*, UPA).

The genus *Paeonia* L. (*Paeoniaceae*) includes ca. 35 species distributed widely in the northern hemisphere: eastern & central Asia, the western Himalayas, the Mediterranean region and the Pacific North America (Stern 1946, Tzanoudakis 1983b, Sang & al. 1997). In the Mediterranean area almost 22 species have been recorded so far (Aghababian 2011).

Paeonia mascula comprises five subspecies: (1) subsp. *mascula*, (2) subsp. *bodurii* N. Özhatay, (3) subsp. *hellenica* Tzanoud., (4) subsp. *icarica* Tzanoud. and (5) subsp. *russoi* (Aghababian 2011). All the above mentioned subspecies of *P. mascula* occur in Greece except for the Turkish endemic subsp. *bodurii*. However, the taxonomic status of the Greek subspecies is not clear. Tzanoudakis (1977, 1983b) highlights the variability within the species and recognizes the above mentioned four subspecies in the Greek area.

The plants of *Paeonia mascula* that occur on the islands of the Ionian floristic region Zakinthos, Kefallinia and Lefkada, as well as on the Akarnanika mountain range at the opposite side (W Sterea Ellada), were recognized as subsp. *russoi* (Cullen & Heywood 1964; Tzanoudakis 1977; 1983b). However, Hong & Wang (2006) and Hong (2010), based on some morphological similarities, as well as on the same chromosome number ($2n = 10$), assume that the Greek populations of subsp. *russoi* belong to *P. corsica* Sieber. The same

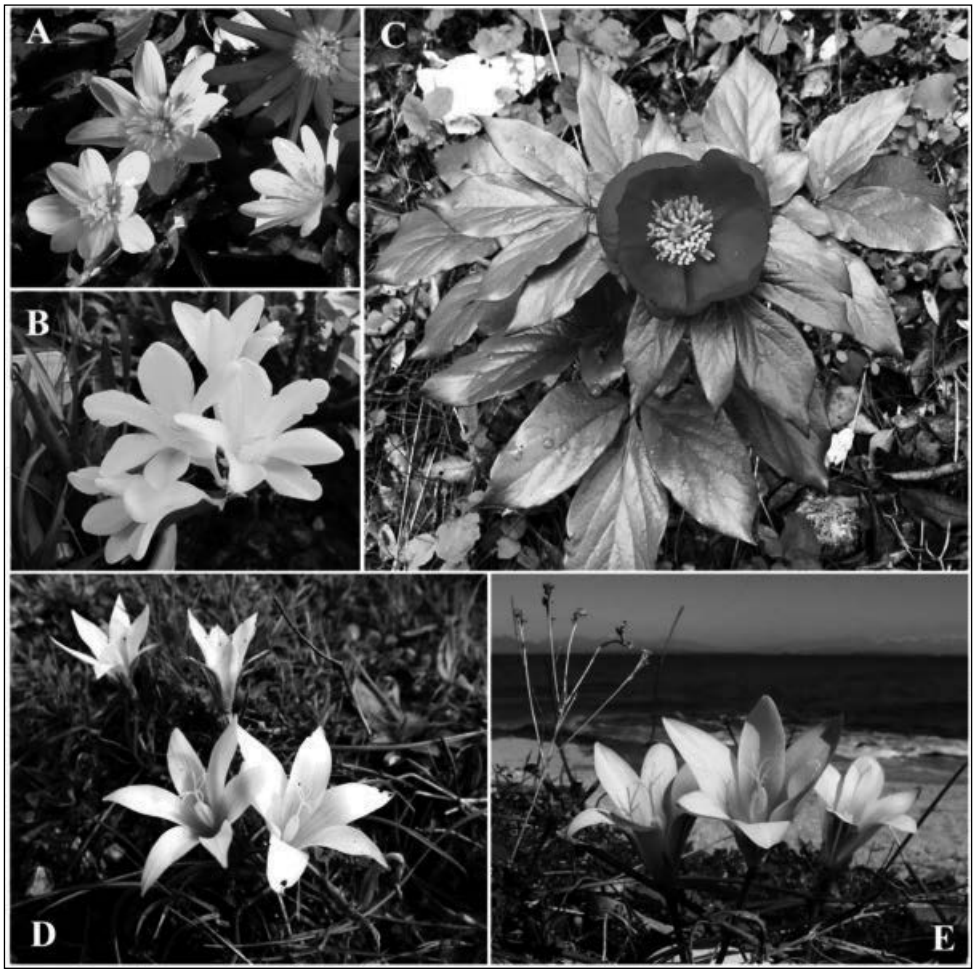

Fig. 2. Studied taxa from Kefallinia and Lefkada islands: **A**, *Ficaria calthifolia* from Mt. Aenos, summit of Mt. Roudi; **B**, Cultivated material of *Freesia leichtlinii* subsp. *alba*; **C**, individual of *Paeonia mascula* subsp. *russoi* from Giagana, N Kefallinia; *Romulea bulbocodium* from Lefkada: **D**, from Mt. Stavrotas and E, from the sand dunes of cape Gyrapetra.

name is used by Stearn & al. (2012), Dimopoulos & al. (2013) and Strid (2016). However, Phitos & al. (2015) consider the above facts insufficient to rename the plants of the Greek populations to *P. corsica*. Even less convincing is the phytogeographical distribution of *P. corsica* (Stearn & al. 2012). It is evident, mostly because of the numerous synonyms of *P. mascula* at the southern tip of the Italian Peninsula, Sicily, Corsica and Sardinia that the distinction of the representative taxa of the species has been ambiguous. For the above reasons we preserve here the previous name *P. mascula* subsp. *russoi* with its distribution in Ionian Islands and W Sterea Ellas. Moreover, molecular analysis of *P. mascula* subsp. *russoi* and

P. corsica from the whole distribution areas is under progress in order to elucidate their taxonomic relationships.

It is noteworthy that *P. mascula* subsp. *russoi* is included in the Red Data Book of Rare and Threatened Plants of Greece (Phitos & al. 2009) categorized as Near Threatened (NT), according to IUCN criteria (2001).

The evolutionary history of the genus *Paeonia* in the Mediterranean area has been closely related to polyploidy events. Concerning the ploidy levels within *P. mascula* subspecies, subsp. *russoi*, is diploid (2*n* = 2x = 10) and the remaining four are tetraploids (2*n* = 4x = 20) (Tzanoudakis 1977, 1983b; Özhatay & Özhatay 1995).

The chromosome number 2*n* = 10 has been given for *P. mascula* subsp. *russoi* by Tzanoudakis (1977, 1983b) in material from the Ionian Islands (Lefkada, Kefallinia, Zakinthos) and W Sterea Ellas (Mt. Boumistos). Additionally, Passalacqua & Bernardo (2004) reported the tetraploid chromosome number 2*n* = 4x = 20 for Italian populations under the taxonomic level of variety as *P. mascula* subsp. *mascula* var. *russoi* (Biv.) N. G. Passal. & Bernardo.

The chromosome number and the karyotype morphology, which is presented here from a new population in northern Kefallinia isl. (Fig. 3F), is in accordance with that given by Tzanoudakis (1977). The karyotype is diploid and symmetrical, consisting of 2*n* = 4m + 2m-SAT + 2sm-SAT + 2st-SAT = 10 chromosomes. The chromosome size varies from 9.30-13.95μm.

1967. *Romulea bulbocodium* (L.) Sebast. & Mauri — 2*n* = 3x = 33 (Figs 2D & 3G) & 2*n* = 4x = 44 (Figs 2E & 3H).

Gr: Ins. Lefkada: Mt. Stavrotas, ad cacumen Elati, alt. ca. 1100 m, 20° 38′ 47.38′′ E, 38° 42′ 27.14′′ N, 31.III.2016, leg.: *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari *29442*, UPA). – Figs 2D & 3G.

— Ins. Lefkada: Regio borealis insulae, ad promunturium Gyrapetra dictum; in semiarenosis et in solo humidulo, alt. 2-5 m, 20° 40′ 54.77′′ E, 38° 50′ 20.70′′ N, 02.III.2017, leg.: *D. Phitos*, *G. Kamari & E. Katopodi* (Herb. Phitos & Kamari*29210*, UPA). – Figs 2E & 3H.

The genus *Romulea* Maratti (*Iridaceae*) comprises ca. 95 taxa (Goldblatt & al. 2008). The distribution centre of the genus is found in South Africa and Arabian Peninsula, where 80 taxa occur (Manning & Goldblatt 2001, 2004, 2008). The remaining 15 taxa are endemic to the Mediterranean area (Peruzzi & al. 2011a).

Romulea is represented in Greece by five species: 1) *R. bulbocodium*, 2) *R. columnae* Sebast. & Mauri, 3) *R. linaresii* Parl., 4) *R. ramiflora* Ten. And 5) *R. tempskyana* Freyn (Dimopoulos & al. 2013). The first four species have been reported in some Ionian Islands, whereas *R. bulbocodium* occurs in most of them (see Flora Ionica Working Group 2016).

Despite the relatively wide distribution of *Romulea* species in Greece, the only taxon that has been studied karyologically is *R.* c.f. *linaresii* (2*n* = ca. 39) from Athens (Goldblatt & Takei 1997).

Concerning the ancestral basic chromosome number in *Romulea*, the opinions of the authors are not concordant. For example, de Vos (1972) suggested the number x = 12, whereas Goldblatt & Takei (1997) x = 13. Additionally, several secondary basic chromo-

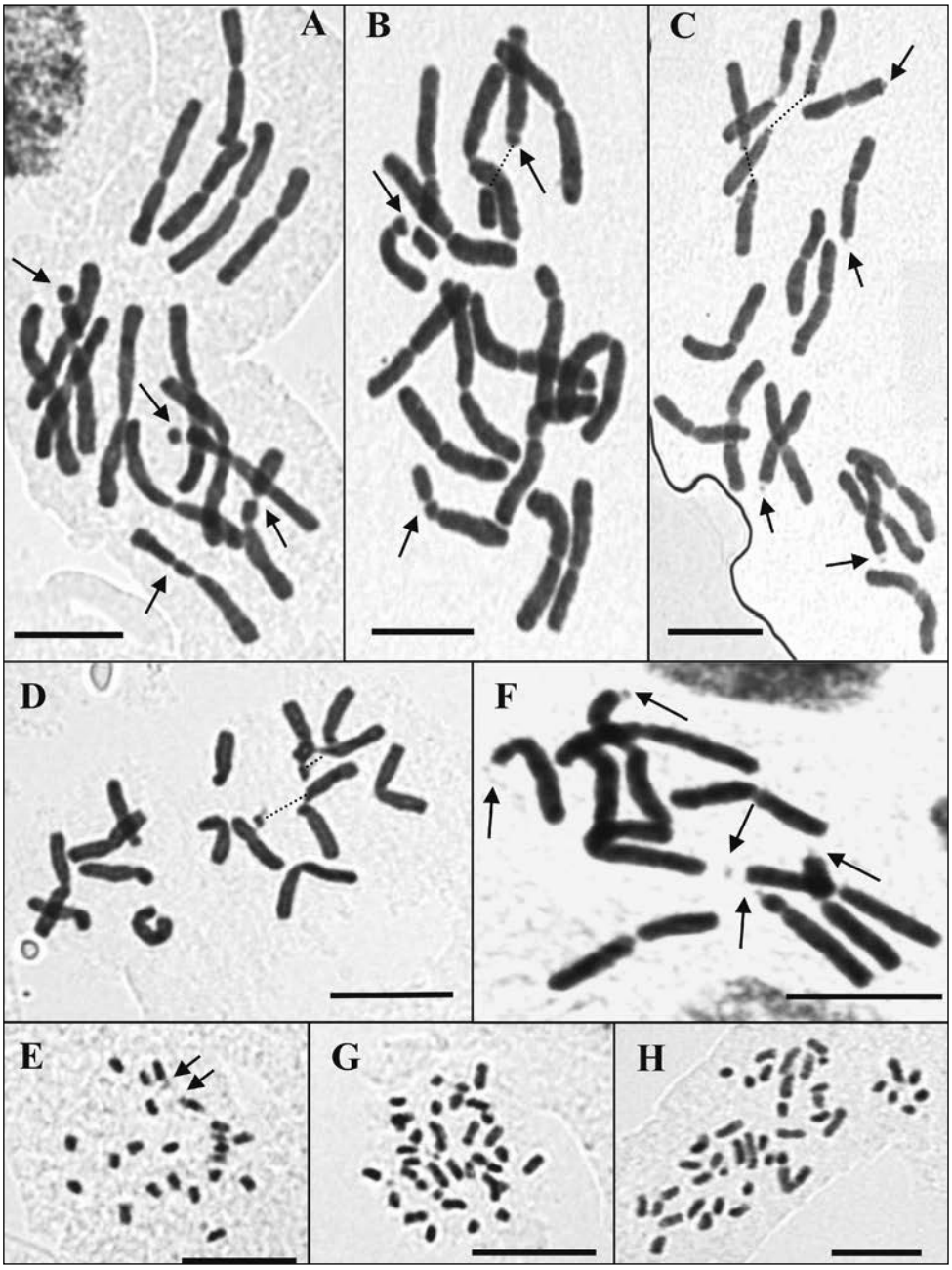

Fig. 3. Microphotographs of mitotic metaphase plates of the studied taxa: **A**, *Allium callimischon* subsp. *callimischon* from Gyra, 2*n* = 16 and **B**, from Mt. Stavrotas, 2*n* = 16; **C**, *Allium flavum* subsp. *tauricum*, 2*n* = 16; **D**, *Ficaria calthifolia*, 2*n* = 16; **E**, *Freesia leichtlinii* subsp. *alb*a, 2*n* = 22; **F**, *Paeoniamascula* subsp. *russoi*, 2*n* = 10; **G**, *Romulea bulbocodium* from Mt. Stavrotas, 2*n* = 3x = 33 and **H**, from Gyra, 2*n* = 4x = 44. – Arrows indicate satellites and/or secondary restrictions. Scale bars = 10 μm.

some numbers are reported, such as x = 9, 10, 11, 14 that are justified, due to dysploidy (de Vos 1972; Goldblatt & Takei 1997; Castro & Roselló 2005; Peruzzi & al. 2011a). Peruzzi & al. (2011a) suggest x = 9 as a shared basic chromosome number for all investigated taxa, opposing x = 14, favored by Castro & Rosello (2005). The discordant chromosome numbers given in literature are often the case for several Mediterranean *Romulea* species and could be interpreted as miscounting, due to the small chromosome size and their reluctance to stain, or as dysploid chromosome sets, realized through descending (i.e., $2n = 34$) or ascending (i.e., $2n = 56$) aneuploidy, or finally, as a possible effect of B-chromosomes (Peruzzi & al. 2011a).

The reported chromosome numbers for *Romulea bulbocodium* are: $2n = 28$ in material from Spain (Castroviejo 1984), $2n = 34$ in material from Iberian Peninsula (recognized as var. *rectifolia*; Fernandes & al. 1948) and in material from eastern Mediterranean of unknown origin (Darlington & Wylie 1955), $2n = 36$ in material from Italy, Sicily and Sardinia (Peruzzi & al. 2011a, b) and $2n = 42$ in material from Croatia (Susnik & Lovka 1973).

Our results confirm the intense karyotype variability of the species and the genus in general. One of the examined populations from Mt. Stavrotas (Fig. 2D) shows the chromosome number $2n = 3x = 33$ with chromosome size varying from 1.07-2.86 μm (Fig. 3G), whereas the second population from the sand dunes of cape Gyrapetra (Fig. 2E) shows $2n = 4x = 44$ with chromosome size from 0.88-3.53 μm (Fig. 3H). The karyological findings of the studied, here, populations support that the basic chromosome number for the species is x = 11. According to that, one population is triploid and the other one is tetraploid (Figs 3G & 3H). We present the chromosome number and a karyotype microphotograph because of the difficulty in counting and staining the material, as also mentioned by previous authors (e.g. Goldblatt & Takei 1997; Peruzzi & al. 2011a). The chromosome data given here constitute the first karyological contribution for *R. bulbocodium* from Greece.

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Karyological study of some E Iberian threatened plants

Abstract

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In the framework of cytogenetic studies of rare and threatened plant species of the Catalan Countries and surrounding territories, a karyological study of some taxa – three populations of *Delphinium bolosii* and the only known one of *D. mansanetianum* (*Ranunculaceae*), one of *Hesperis laciniata* subsp. *laciniata* (*Brassicaceae*), and one of *Silene neglecta* (*Caryophyllaceae*) – is presented here. Karyotype microphotographs and corresponding idiograms for most taxa are provided and their karyotype morphology is discussed.

Keywords: CromoCat, Catalan countries, chromosome number, karyotype, threatened plants.

Introduction

Chromosome databases are useful as both a repository of existing knowledge as well as a tool to identify gaps of karyological information. The CromoCat chromosome database gathers data from the Catalan Countries since 1998 (Simon & al. 2011). At present, 215 taxa (4,4 % of the total vascular flora, Simon & Blanché 2016) still remain karyologically unknown.

Thus, we launched a research program to obtain new counts to fill the detected needs, with priority given to, threatened species, endemics and taxa with reported variation in chromosome number. This contribution is oriented to the endemics *Delphinium bolosii* (with two previously detected cytotypes) and *D. mansanetianum* (unknown number) and to two Mediterranean species evaluated as threatened in Catalonia, *Hesperis laciniata* subsp. *laciniata* and *Silene neglecta*, with few previous karyological reports and not counted in our territory.

1968. *Delphinium bolosii* C. Blanché & Molero — 2*n* = 16.

Hs: Barcelona, Bages, Sant Llorenç del Munt i l'Obac Natural Park, near Mura, footcliff, 41° 40' 58'' N 1° 58' 48'' E, alt. 552 m, 1 Jul 2014, *M. Bosch* (BCN 113231). – Figs 1A-1B & 2A.

— Lleida, La Noguera, Alòs de Balaguer, screes and understory with strong slope, in N facing cliffs over the river Segre, 41° 54' 37'' N 0° 57' 03'' E, alt. 305 m, 13 Jun 2007, *M. Bosch, M.C. Martinell, J. Molero & A. Rovira* (BCN 127363). – Figs 1C & 2B.

— Lleida, La Noguera, Presa de Camarasa, cliffs over the river Segre, 41° 54' 14'' N 0° 53' 47'' E, 465 m, 3 Jul 2008, *M. Bosch & J. Molero* (BCN 53601). – Figs 1D & 2C.

The three surveyed populations share the same chromosomic number of 2*n* = 16 (Figs 1A-1D), which is different than the previous reports for this species of 2*n* = 18 (Blanché 1991; Simon & al. 1995; Bosch 1999), meaning the coexistence of two cytotypes in this species. It is a very rare phenomenon in the tribe *Delphinieae* only detected in 20 taxa, 4 of them in the genus *Delphinium* (Bosch & al. 2016). In the case of the population near Mura, this number is confirmed in different individuals from two different nuclei, 500 m apart (Figs 1A &1B).

The karyotype analysis (Figs 2A-2C) shows a formula with 2 pairs of large metacentric or submetacentric chromosomes and 6 smaller subacrocentric ones (Mura: 2*n* = 2x = 2m-SAT + 2sm + 10st + 2st-SAT = 16; Alòs de Balaguer: 2*n* = 2x = 2m + 2m-SAT + 10st + 2st-SAT = 16; Presa de Camarasa: 2*n* = 2x = 2m + 2m-SAT + 10st + 2st-SAT = 16), similar to the general pattern described in *Delphinieae* tribe (Epling & Lewis 1951, Blanché & al. 1997, Bosch & al. 2016). The chromosome size varies from 3.86-14.09 µm.

Satellites are located in the short arm of the first metacentric pair and in the short arm of the IV or V subacrocentric pairs (Figs 2A-2C). In the series *Fissa* B. Pawl., where this species is placed (Bosch & al. 2019), the satellites number and pair are reported in slightly variable positions. In the *D. bolosii* population from Rubió (La Noguera, Lleida), with 2*n* = 18, satellites have been reported from pairs II, IV and VI, whereas satellites are missing in the reports from the Ulldemolins (Priorat, Tarragona), also with 2*n* = 18, and the *D. fissum* subsp. *sordidum* studied populations (Blanché 1991; Bosch 1999). In *D. fissum* subsp. *fissum* French (Bosch 1999) and Greek populations (Constantinidis & al. 1997), satellites appear in one (I) or two (I and V) pairs and in *D. fissum* s.l. from Bulgaria (Koeva-Todorovska 1985), indicated from three pairs (I, IV and VIII or I, V and VIII). In all the surveyed taxa, satellites are reported from the short arms, which is the most abundant case in the tribe (Bosch & al. 2016). Attribution to distinct very similar chromosome pairs (very close in length or symmetry) could be, at least in some cases, a mere artefact, but variation in the total number of satellites (from zero to three in *D. bolosii*, i.e.) can have a more deep evolutionary significance. Notably, this species, with two cytotypes and maximum satellite diversity, also contains the maximum genetic diversity in the whole series *Fissa* and is considered as a genetic reservoir to understand the diversity pattern of this group in the W Mediterranean (Bosch & al. 2019).

Delphinium bolosii is assessed as EN ("Endangered") (Sàez & al. 2010), but currently under new evaluation to probably lower its threat category.

1969. *Delphinium mansanetianum* Pitarch, Peris & Sanchis — 2*n* = 16.

Hs: Aragón, Teruel, Mosqueruela, Masia del Matorrillo, thorny edges of Scots pine clearings, on calcareous rocky soils, 40° 22' 35'' N 0° 28' 54'' W, alt. 1640 m, 15 Jul 1997, *R. Pitarch* (MA 692377).

The count 2*n* = 16 is the first report for this species, which is only known from one location in Teruel (Aragón, Spain). This species belongs to the series *Fissa* of genus *Delphinium* and it is closely related to *D. bolosii* (Bosch & al. 2019), with two different numbers (see previous species reported). It is assessed as CR ("Critically endangered") (Bosch & al. 2019).

1970. *Hesperis laciniata* All. subsp. ***laciniata*** — 2*n* = 12.

Hs: Lleida, La Noguera, Rubió de Baix, footcliffs and shady herbaceous places below an old tower in a fluvial terrace, 41° 54' 21'' N 0° 59' 56'' E, 280 m, 9 May 2007, *C. Blanché*, *M. Bosch & J. Molero* (BCN 47270). – Figs 1E & 2D.

This reported chromosome number of 2*n* = 12 (Fig. 1E) confirms the previous counts of Ančev & Peneva (1984) and Ančev & Goranova (1997) from Bulgarian material, Constantinidis & al. (2002), from Greek material, and also with the gametophytic number (*n* = 6) provided by Ruíz de Clavijo Jiménez (1994) from material of Cordoba (S Spain). Constantinidis & Kamari (1994) found the same number for *H. laciniata* subsp. *secundiflora* (Boiss. & Spruner) Breistr., an endemic subspecies of Greece. Dvorak (1966) reported a different number 2*n* = 14 for *H. glutinosa* Vis., which is considered a partial synonym of *H. laciniata* (Marhold 2011). Both chromosome numbers (2*n* = 12, 14) occur in *Hesperis* (Rice & al. 2015).

The karyotype analysis showed a formula of 2*n* = 2x = 8m + 4sm = 12 chromosomes (Fig. 2D), the same described by *H. laciniata* ssp. *laciniata* (Ančev & Goranova 1997, Constantinidis & al. 2002), but with a small difference in *H. laciniata* subsp. *secundiflora* (Constantinidis & Kamari 1994) where one of the submetacentric pairs is a subacrocentric one. The chromosomes vary in size from 4.08 to 8.16 μm. We have not detected the presence of satellites, observed by Constantinidis & Kamari (1994) and Constantinidis & al. (2002), which mentioned that they are very small. Constantinidis & Kamari (1994) also reported frequent secondary constrictions in the short arms of metacentric chromosomes, but we could not appreciate it in our material.

H. laciniata is of S. European and N African species, threatened (VU, "Vulnerable") and very rare in Catalonia (Sàez & al. 2010), our count is from the single confirmed population.

1971. *Silene neglecta* Ten. — 2*n* = 24.

Hs: Barcelona, Garraf, Gavà, NW rocks over Ermita de Bruguers church, sandstone rocky soil, 41° 18' 52'' N 1° 57' 36'' E, alt. 265 m, 6 May 2015, *L. Sáez* (LS-7626, LS: Sáez, personal herbarium). – Figs 1F & 2E.

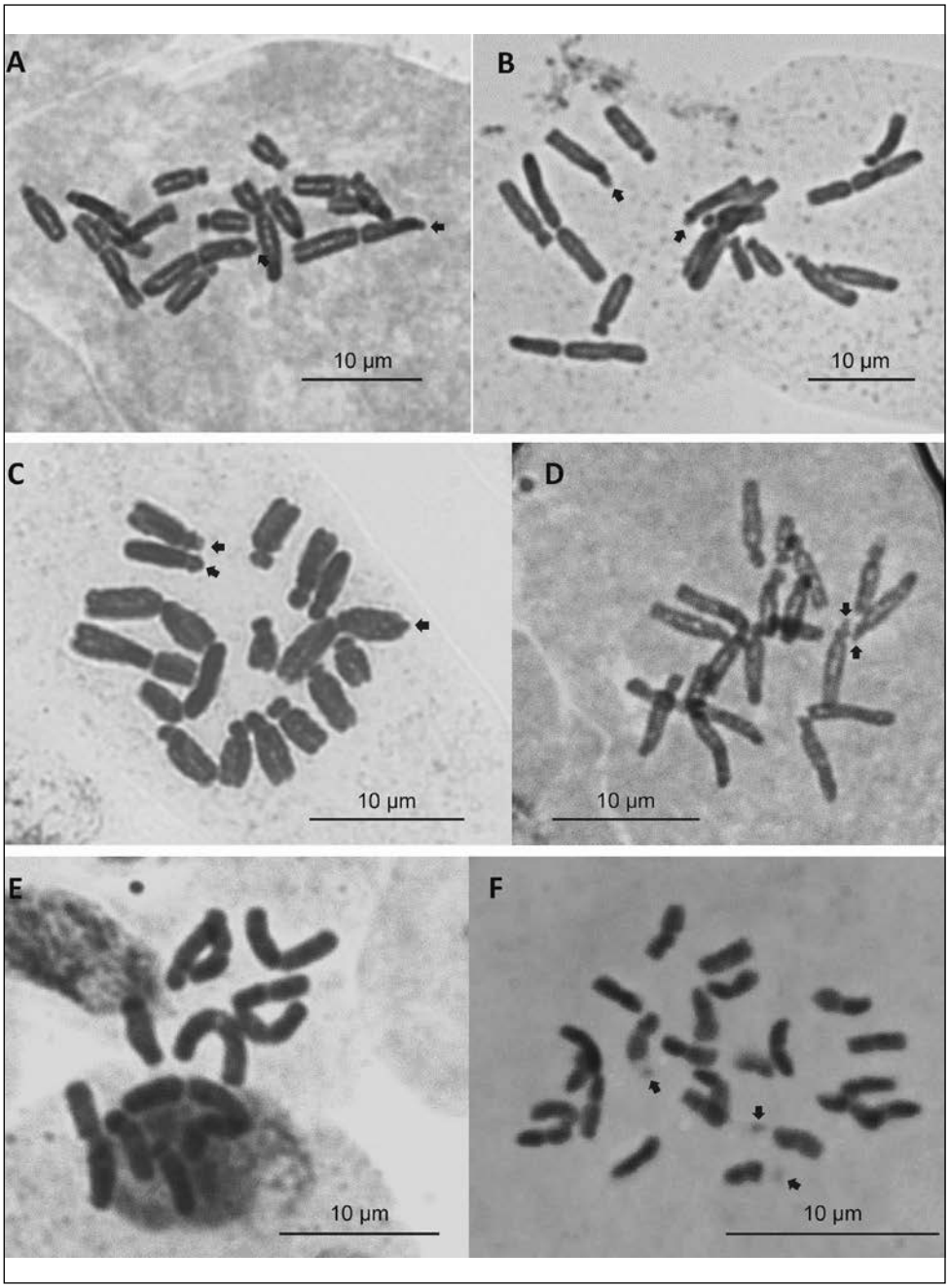

Fig. 1. Microphotographs of somatic metaphase plates of: **A-D**, *Delphinium bolosii*, 2*n* = 16: **A** & **B**, Mura (Hs: Barcelona), **C**, Alòs de Balaguer (Hs: Lleida), **D**, Presa de Camarasa (Hs: Lleida); **E**, *Hesperis laciniata* subsp. *laciniata*, 2*n* = 12: Rubió de Baix (Hs: Lleida); **F**, *Silene neglecta*, 2*n* = 24: Ermita de Bruguers (Hs: Barcelona). – Arrowheads indicate satellites.

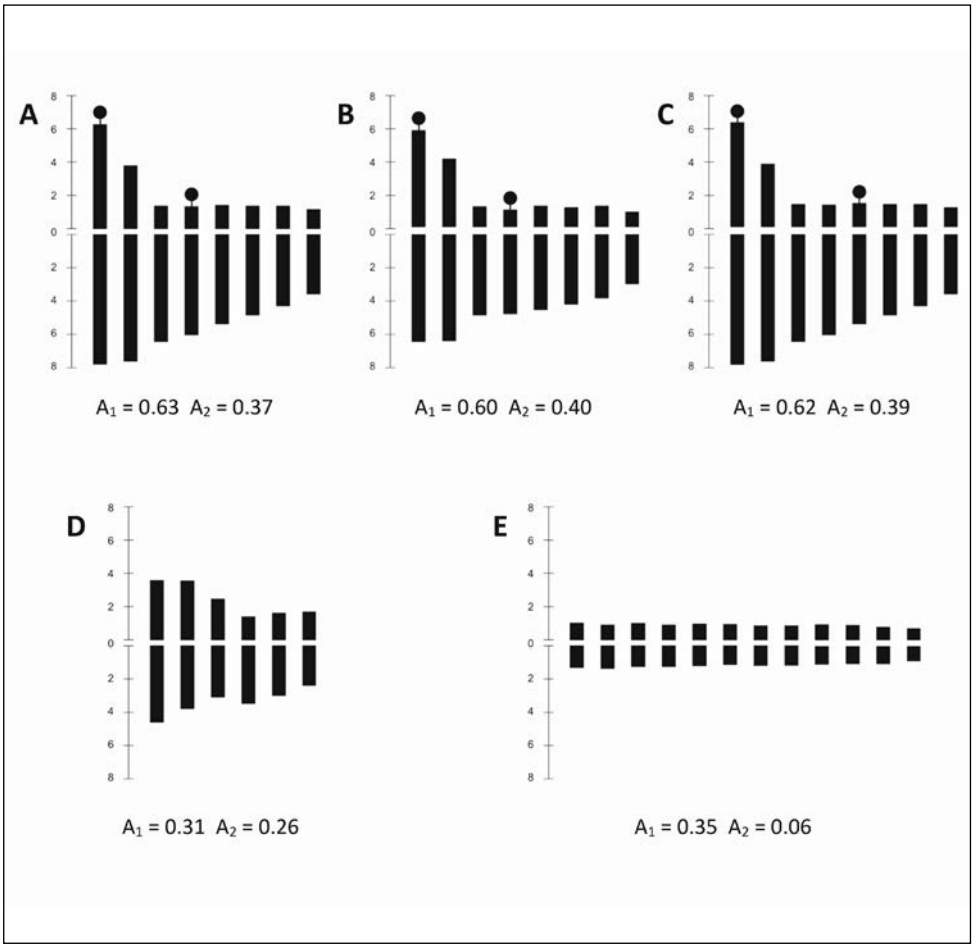

Fig. 2. Haploid idiograms obtained by measuring at least 5 good mitotic metaphases, 2 hours after pretreatment with hidroxiquinoleine 0.002 M. Asymmetry indices A1 and A2 are calculated following Romero (1986). **A-C**, *Delphinium bolosii*: **A**, Mura (Hs: Barcelona), **B**, Alòs de Balaguer (Hs: Lleida) & **C**, Presa de Camarasa (Hs: Lleida); **D**, *Hesperis laciniata* subsp. *laciniata*: Rubió de Baix (Hs: Lleida); **E**, *Silene neglecta*: Ermita de Bruguers (Hs: Barcelona). – Scale bars = 8+8 μm.

The count of $2n = 24$ (Fig. 1F) is the first report of this species for the Iberian Peninsula. This taxon has been misunderstood and confused with *Silene nocturna* L., although several reproductive characters (petals shape and colour, seed size, shape and seed-coat surface, among others) allow separating both species (Talavera 1990; Pignatti 2019).This population is the only known one in Iberian Peninsula, and locally considered severely threatened: Iberian evaluation as EN ("Endangered") (Sàez 2018). The species is mainly distributed in Italy and N Africa, with some cited populations in S France although not confirmed. This count coincides with the most frequent number of the genus *Silene* (Rice & al. 2015).

The karyological study reveals a chromosomic formula of 2*n* = 2x = 24m = 24 chromosomes (Fig. 2E), varying in size from 1.59 to 2.34 μm, which are very similar between them and very symmetrical (all are metacentric chromosomes). Presence of satellites is detected (at least 3 pairs) but it is hard to assign due to the similarity of chromosomes in size and symmetry.

The revision by Peruzzi & Carta (2013) of the *S. nocturna* group in Italy (including the first count for *S. neglecta*) gives the same chromosome number (2*n* = 24) for the components of this group: *S. capraria* Sommier (= *S. nocturna* subsp. *capraria* (Sommier) Peruzzi & Carta), *S. neglecta* and *S. nocturna*, with chromosome sizes ranging from 0.5 to 2.5 μm. The karyotype analysis of Italian populations of *S. neglecta* (from Tuscan Archipelago) is very similar in length and symmetry to our population, also confirming longer chromosomes in *S. neglecta* than in *S. capraia*. However, no satellites are reported for any of the Italian studied populations. The same number 2*n* = 24, similar karyotype and absence of satellites are reported for *S. nocturna* subsp. *boullui* (Rouy & Foucaud) Gamisans in Gamisans & D. Jeanmonod (Bachetta & al. 2014) and for another close species of the same group, as the Turkish endemic *S. muradica* Schischk., also with similar chromosome lengths (1.50 - 2.77 μm) and symmetry (all pairs are median type) (Martin & al. 2019). Thus, the three pairs with satellites seem to characterize the Catalan population. In relatively related species, we have found only a report of one pair (I) of satellites in the 2*n* = 24 common European annual *S. gallica* (Bari 1973).

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K. Hormat, H. Tahiri, N. Guennoun & A. Gounssa

Étude caryologique d'un taxon endémique du Maroc appartenant au genre *Vicia* (*Leguminosae*)

Abstract

Hormat, K. Tahiri, H., Guennoun, N. & Gounssa, A. 2019: Étude caryologique d'un taxon endémique du Maroc appartenant au genre *Vicia* (*Leguminosae*) [In Kamari, G., Blanché, C. & Siljak-Yakovlev, S. (eds), Mediterranean plant karyological data - 29]. – Fl. Medit. 29: 329-333. http://dx.doi.org/10.7320/FlMedit29.329

A systematic revision of two taxa belonging to the genus *Vicia* L. section *Cracca* S.F. Gray, *V. benghalensis* L. and *V. villosa* Roth, led us to propose, in a previous contribution, a new combination and a new status for an endemic taxon of Morocco: *V. benghalensis* subsp. *heterocalyx* var. *simulans* (Maire) Hormat. This taxon, as well as *V. benghalensis* var. *benghalensis* show a $2n = 14$ chromosome number, whereas *V. benghalensis* subsp. *heterocalyx* var. *heterocalyx* gives $2n = 12$.

Keywords: *Vicia*, *Fabaceae*, Karyosystematics, Moroccan flora.

Introduction

Vicia benghalensis subsp. *heterocalyx* var. *simulans* (Maire) Hormat, taxon endémique marocain de la région de Mehdia, a été décrit pour la première fois par René Maire (1937) et a été rattaché *à Vicia villosa* Roth en raison de son fruit totalement glabre. Raynaud (1976), dans une révision taxonomique de la section *Cracca*, a gardé ce taxon au sein de l'espèce *V. villosa* Roth. Lors de notre étude de la section (Hormat 1985, 2004) nous avons remarqué, qu'en plus de son fruit glabre, ce taxon diffère des taxons infra-spécifiques de *V. villosa* par la couleur de la corolle, ce qui nous a amené à faire une étude caryologique qui pourrait nous fournir des renseignements nouveaux pour situer ce taxon au sein de la section *Cracca*.

Matériel et méthodes

Nous avons récolté des échantillons fleuris et surtout fructifiés de *V. benghalensis* subsp. *heterocalyx* var. *simulans* à Mehdia, seul endroit où il semble exister avec *V. benghalensis* subsp. *heterocalyx* var. *heterocalyx*, que nous avons aussi récolté pour comparaison. Pour les besoins de comparaison toujours, nous avons également récolté *V. villosa* subsp. *villosa* à Tétouan, qui semble exister tout à fait au nord du Maroc et d'Algérie et *V. benghalensis* var. *benghalensis* à Tanger. Les échantillons récoltés sont déposés à l'herbier de Montpellier (MPU).

L'étude caryologique, à partir des racines obtenues par la germination de graines récoltées, a nécessité deux méthodes de colorations: la méthode au carmin acétique et la méthode à l'or-

céine acétique pour les plantes qui n'ont pas montré une bonne coloration de chromosomes avec la technique au carmin acétique. A partir des plaques métaphasiques (10 par population) nous avons établi le caryotype et l'idiogramme correspondant, suivis de la formule chromosomique. Le type chromosomique est déterminé par la position du centromère d'après la nomenclature de Levan & al. (1964). Pour préciser la dimension des chromosomes nous avons établi l'échelle des valeurs reprise de Verlaque (1983) et de Xena De Enrech (1987).

Résultats et discussion

Au cours de nos observations nous avons pu mettre en évidence deux types de chromosomes : les chromosomes A, ordinaires, qui se présentent sous forme de bâtonnets, de longueur variant de 2 à 7 µm, et de nombre variant de $2n = 12$ à $2n = 14$; et les chromosomes B, ou surnuméraires, ou accessoires, qui se présentent sous forme de très petits bâtonnets, souvent métacentriques, et dont le nombre est variable même parfois parmi les cellules d'un même individu. Il semble que ces chromosomes B proviennent des chromosomes A par l'intermédiaire de translocations, délétions ou inversions (Fernandes 1949).

1972. ***Vicia villosa*** Roth subsp. ***villosa*** — $2n = 14$.

Ma: Rif : 6 Km au sud-est de Tétouan, entre Tanger et Tétouan, 30 m, 35° 45' / 5° 18'. MPU - HERBIER HORMAT n° 157.

La méthode à l'orcéine a révélé un nombre chromosomique de $2n = 14$ qui est en accord avec les rapports des auteurs qui ont travaillé sur *V. villosa s.s.*, notamment Seen (1938), Baksay (1954), Cincura (1962) et Raina & Rees (1983), avec toujours la présence de deux chromosomes B, toutefois Baksay a noté la présence de satellites sur une paire de chromosomes. Le caryotype est composé d'une paire de chromosomes moyens, de 4,5 µm environ, à centromère subterminal, 3 paires de chromosomes moyens, de 4 µm environ, à centromère submédian, 2 paires de chromosomes moyens, de 3,5 µm environ, à centromère dans la région médiane, une paire de chromosomes très courts, de 2 µm environ, à centromère submédian. Ce caryotype est représenté par la formule chromosomique suivante: 6 M (1 st + 3 sm + 2 m) + 1 TC (sm).

1973. ***Vicia benghalensis*** L. var. ***benghalensis*** — $2n = 14$.

Ma: Rif : 30 Km au sud de Tanger, entre Asila et Tanger, dans la forêt diplomatique, 30 m, 35° 38' / 5° 57'. HERBIER HORMAT n° 149.

Selon la méthode à l'orcéine nous avons obtenu des plaques métaphasiques qui montrent $2n = 14$ chromosomes. Le nombre chromosomique de base x = 7 est en accord avec celui proposé par des auteurs qui ont travaillé sur ce taxon, notamment Senn (1938), Rajan (1952) et Raina & Rees (1983). Le caryotype est composé d'une paire de chromosomes

très longs (environ 7 µm) à centromère submédian, d'une paire de chromosomes longs (environ 6 µm) à centromère submédian, d'une paire de chromosomes longs (environ 5,5 µm) à centromère submédian, d'une paire de chromosomes longs (environ 5 µm) à centromère submédian, d'une paire de chromosomes moyens (environ 4,5 µm) à centromère dans la région médiane, d'une paire de chromosomes moyens (environ 4 µm) à centromère dans la région médiane, et d'une paire de chromosomes courts (environ 3 µm) à centromère médian. Ce caryotype est représenté par la formule chromosomique suivante: 1 TL (sm) + 3 L (sm) + 2 M (m) + 1 C (m).

On note une différence majeure avec *V. villosa* par les chromosomes qui sont beaucoup plus grands. Ils se rapprochent par la taille et par l'absence de chromosomes subtélocentriques à ceux de *V. villosa* subsp. *simulans*.

1974. ***Vicia benghalensis*** subsp. ***heterocalyx*** var. ***simulans*** (Maire) Hormat — 2*n* = 14.

Ma: Région de Kenitra : Mehdia, tout le long de la Merja de Sidi Bou-Ghaba, 60 m, 34° 14' / 6° 41'. MPU - HERBIER HORMAT n° 160.

La méthode au carmin acétique a révélé un nombre diploïde de 2*n* = 14 avec présence de chromosomes surnuméraires dont le nombre varie entre 2 et 3 chromosomes B. Ce nombre chromosomique est récent, il n'a jamais fait l'objet d'une recherche caryologique, il se trouve pour la première fois chez ce taxon endémique du Maroc. Le caryotype est composé de 2 paires de chromosomes longs de 5 µm environ, à centromère submédian, 2 paires de chromosomes moyens de 4 µm environ, à centromère submédian, 2 paires de chromosomes moyens de 3,5 µm environ, à centromère submédian (1 paire) dans la région médiane (1 paire), 1 paire de chromosomes courtsde 3 µm environ, à centromère submédian. La formule chromosomique est la suivante: 2 L (sm) + 4 M (3 sm + 1 m) + 1 C (sm).

On remarque que les chromosomes sont un peu plus grands que ceux de *V. villosa,* et qu'il y a absence de chromosomes subtélocentriques.

1975. ***Vicia benghalensis*** subsp. ***heterocalyx*** var. ***heterocalyx*** (Maire-Weiller) Hormat — 2*n* = 12.

Ma: Région de Kenitra : Mehdia, tout le long de la Merja de Sidi Bou-Ghaba, 60 m, 34° 14' / 6° 41. MPU - HERBIER HORMAT n° 150.

Selon la méthode au carmin acétique, dans les métaphases somatiques nous avons dénombré 2*n* = 12 chromosomes, et nous avons noté la présence de chromosomes surnuméraires dont le nombre est variable (5, 6 à 8 chromosomes B). Le nombre chromosomique de base x = 6 est récent et trouvé pour la première fois chez ce taxon, toutefois il a été également indiqué chez *V. benghalensis* var. *benghalensis* par Srivastava (1963). Les chromosomes sont en général moins grands que ceux de *V. benghalensis* var. *benghalensis*. Le caryotype correspondant aux idiogrammes montre: une paire de chromosomes longs d'environ 6 µm, à centromère submédian, 2 paires de chromosomes longs d'environ 5 µm,

à centromère submédian, 2 paires de chromosomes moyens (4,5 µm environ) à centromère submédian, et une paire de chromosomes moyens de 4 µm à centromère submédian. La formule chromosomique est la suivante: 3 L (sm) + 3 M (sm).

On remarque une ressemblance entre les caryotypes de *V. benghalensis* subsp. *heterocalyx* var. *heterocalyx* et de *V. benghalensis* var. *benghalensis*, sauf pour les chromosomes de la première paire et ceux de la dernière paire. On peut se demander si c'est la première paire ou si c'est la septième paire chez *V. benghalensis s.s.* qui s'est fragmentée pour donner les chromosomes B chez *V. benghalensis* subsp. *heterocalyx* var. *heterocalyx*. Les chromosomes s'approchent beaucoup de ceux de *Vicia benghalensis* subsp. *heterocalyx* var. *simulans* (Maire) Hormat par la taille et par l'absence de chromosomes subtélocentriques.

Conclusion

L'étude caryologique réalisée dans ce groupe met en évidence l'existence de deux nombres chromosomiques $2n = 12$ et $2n = 14$, avec l'absence ou la présence de chromosomes surnuméraires.

On peut penser que le nombre de base x = 6 provient d'une diminution du nombre de base x = 7 par le phénomène de la dysploidie descendante (Senn 1938; Goldblatt 1981; Polhill & al. 1981). Il faut noter aussi qu'il y a chez tous les taxons étudiés l'absence de chromosomes télocentriques (t) et de chromosomes acrocentriques (T).

L'examen des caryotypes entraine certaines constatations: *V. benghalensis s.s.* se distingue de *V. villosa s.s.* par la forme de leurs chromosomes; en effet les chromosomes les plus longs à centromère submédian ou médiane existent chez *V. benghalensis*. Chez *V. villosa* les chromosomes sont petits et la première paire de chromosomes moyens est toujours à centromère subterminal comme l'estime Roti-Michelozi (1986). *Vicia benghalensis* subsp. *heterocalyx* var. *simulans* a des chromosomes un peu plus grands que ceux de *V. villosa* et l'absence de chromosomes subtélocentriques (caryotype symétrique) le rapproche de *V. benghalensis*.

Vicia benghalensis subsp. *heterocalyx* var. *heterocalyx* possède des chromosomes relativement grands analogues à ceux de *V. benghalensis* et jamais de chromosomes subtélocentriques comme *V. benghalensis* subsp. *heterocalyx* var. *simulans*. La perte de deux chromosomes de $2n = 14$ à $2n = 12$ s'explique par des translocations inégales par lesquelles un chromosome devient si petit qu'il finit par se perdre après d'être fragmenté pour donner naissance à des chromosomes B (Favarger 1962; Hollings & Stace 1974; Bandel 1974).

En conclusion, *V. benghalensis* subsp. *heterocalyx* var. *simulans,* rattachée à l'espèce *V. villosa* par Maire (1937) en se basant sur le seul critère du fruit glabre, c'est bien qu'il soit exclue et mieux placée au sein de l'espèce *V. benghalensis*, comme indiqué in Hormat (2004), aussi par la présence de grands chromosomes et par l'absence de chromosomes subtélocentriques.

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Cristina Salmeri

Karyological data of some plant species native to South Italy

Abstract

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The somatic chromosome number, karyotype morphology, geographical distribution and ecology of five plant species from the indigenous flora of southern Italy and Sicily are presented. The study includes in particular four species of *Allium* subgen. *Allium* (*Allium agrigentinum*, *A. apulum*, *A. diomedeum, A. chamaespathum*), *Ptilostemon greuteri* and *Salvia ceratophylloides*. Five out six of these species are strict endemics and all of them are very rare and differently threatened based on the IUCN criteria. Karyotype microphotographs for all taxa are provided and their karyotype morphology is discussed.

Keywords: *Allium*, endemism, karyology, *Ptilostemon*, *Salvia*, Sicily, southern Italy.

1976. *Allium agrigentinum* Brullo & Pavone — 2*n* = 2x = 16 (Figs 1A & 2A).

Si: Sicily, Aragona presso le Maccalube, 15 Jun 1994, *Brullo s.n.* (CAT) – type locality.
— Sicily, Calanchi argillosi sotto Troina, 29 May 1993, esemplare coltivato, *Brullo s.n.* (CAT).

Allium agrigentinum is an endangered Sicilian endemic species (Troìa & al. 2018) that typically grows in clay gullies in the Mediterranean steppe-type habitats of the central part of Sicily at an elevation of 100-700 m asl.

The chromosome number of the species (2*n* = 16) was reported for the type locality by Brullo & al. (2001), but no information regarding the chromosome complement and karyotype features have yet been provided. The diploid chromosome number 2*n* = 16 is confirmed for a new locality in addition to the *locus classicus*. The karyotype mainly consists of more or less median chromosomes and 1-2 submedian pairs. Plants from the type locality have, in particular, 4 pairs of metacentric chromosomes, 3 pairs of meta-submetacentric type (arm ratio between 1.30-1.67), and one submetacentric pair. No evident satellites have been detected. Thus, the chromosome formula can be resumed as: 2*n* = 2x = 8m + 6msm + 2sm = 16.

Chromosome total lengths range on average from 9.3 ± 1 µm to 5.39 ± 0.7 µm, while the values of symmetry indices M_{CA} and Cv_{CL} are 13.5 and 17.7 respectively.

The chromosome complement of the other population (Troina, not shown) is slight different from that one of the type locality in having 2 submetacentric pairs, while the biggest and the smallest pairs in size have secondary constrictions on the short arms of the homo-

logues; thus, in this population the chromosome formula can be expressed as: 2*n* = 2x = 6m + 2m-SAT + 2msm +2msm-SAT + 4sm = 16.

1977. *Allium apulum* Brullo, Guglielmo, Pavone & Salmeri — 2*n* = 2x = 16 (Figs 1B & 2B).

It: Puglia, Torre di Inserraglio, sul litorale roccioso calcareo, 31 May1992, *Brullo & Minissale* (CAT) - type locality.
— Puglia, Monte Sant'Angelo, Gargano, esemplare coltivato, 12 Jun 1986, *Brullo s.n.* (CAT).
— Puglia, Monti sopra Manfredonia, Gargano, 3 Jun 1982, *Brullo & Signorello* (CAT).
— Puglia, Vieste, Gargano, 6 Jun 1982, *Brullo s.n.* (CAT).

Allium apulum is a rare species endemic to the Apulia region in South Italy, where it occurs in the rocky ledges on coastal cliffs, and in ephemeral meadows among Mediterranean garrigues and maquis, up to 550 m of elevation. Based on the IUCN criteria, the species is currently assessed as Least Concern (LC) in the Italian national Red List (Wagensommer & al. 2018).

The chromosome number of the species (2*n* = 16) was reported by Brullo & al. (2001), who did not present neither any metaphase plate, nor the karyogram. The diploid count 2*n* = 16 can be confirmed for some additional localities other than the type one. The karyotype is composed by 5 metacentric pairs and 3 meta-submetacentric pairs, one of which (with arm ratio 1.61) actually tending towards the submetacentric type. The smallest metacentric pair was found to be microsatellited on the short arms of the homologues in the samples from Monte Sant'Angelo (Foggia). Thus, the karyotype formula can be summarized as: 2*n* = 2x = 8m + 2m-SAT + 6msm = 16. Chromosome total lengths range on average among the studied populations from 13 to 8.8 µm for the longest chromosome and from 7.0 to 5.0 µm for the shortest chromosome, while the values of symmetry indices M_{CA} and Cv_{CL} are 11.21 and 18.81 respectively.

1978. *Allium chamaespathum* Boiss. — 2*n* = 2x = 16 (Figs 1C & 2C).

It: Calabria, Capo dell'Armi, Reggio Calabria, 18.09.2008, *C. Brullo, S. Brullo, G. Giusso, R. Guarino, C. Marcenò s.n.* (CAT).

Allium chamaespathum is a typical Balkan element. It is endemic to the SW half of Balkan peninsula and some Aegean islands. The species is apparently widespread occurring in several localities, from S Albania, to continental Greece (especially Attic peninsula and Peloponnese), Ionian islands, some W Aegean islands, Crete, with an interesting extension to the southern Italy (Calabria), which represents its westernmost geographic limit. The IUCN status (Economou 2011) for this species is currently assessed as Data Deficient (DD), due to its scattered and not well-known distribution, but in most territories the species is threatened and very rare, with isolated and restricted populations (often few

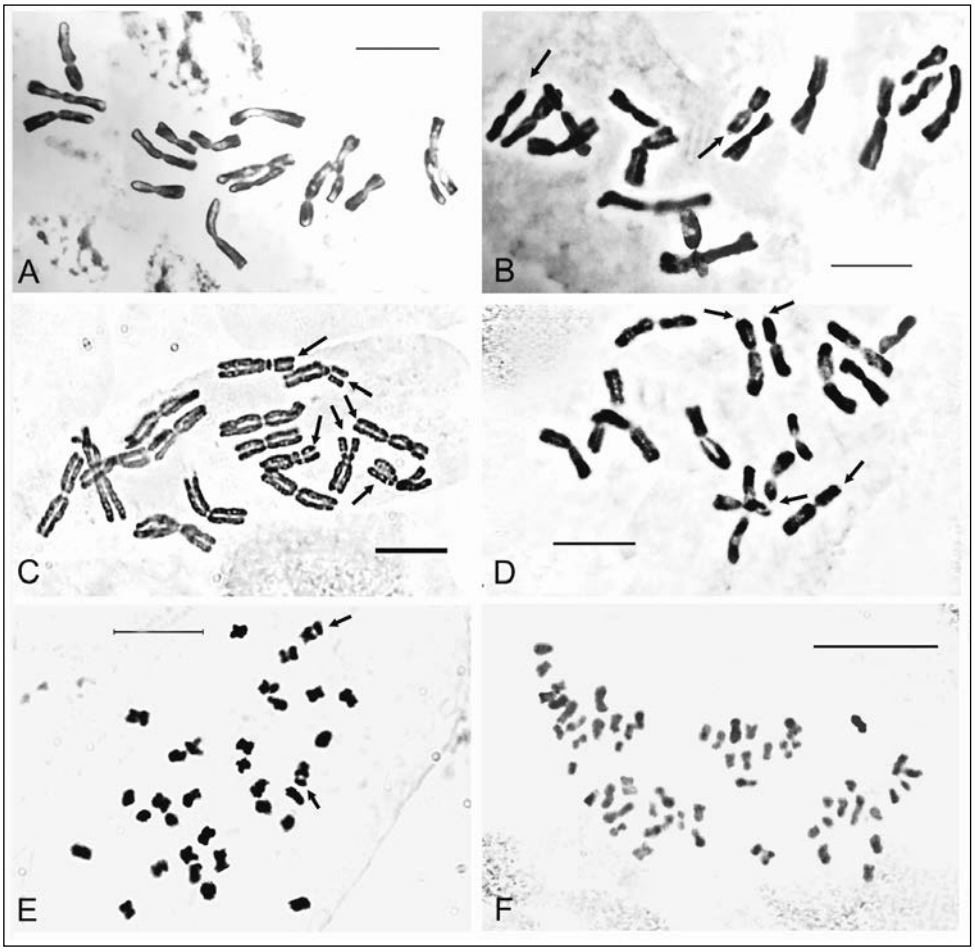

Fig. 1. Microphotographs of mitotic metaphase plates of the studied taxa: **A**, *Allium agrigentinum* from Aragona (Sicily), 2*n* = 16; **B**, *Allium apulum* from Mt. Sant'Angelo (Apulia), 2*n* = 16; **C**, *Allium chamaespathum* from Capo dell'Armi (Calabria), 2*n* = 16; **D**, *Allium diomedeum* from Ins. Tremiti (Apulia), 2*n* = 16; **E**, *Ptilostemon greuteri* from Mt. Inici (Sicily), 2*n* = 32; **F**, *Salvia ceratophylloides* from Reggio Calabria (Calabria), 2*n* = 54. – Scale bars = 10 μm.

dozens of individuals). Plants grow in rocky calcareous places (from the coasts to more than 2,000 m of elevation); habitats are usually represented by garrigues and grasslands, more rarely by open woodlands and dunes, sometimes also roadsides.

The species is known to be diploid with a somatic chromosome number 2*n* = 16 (Bothmer 1974, Tzanoudakis 1985). The Italian population was found to have the same diploid chromosome count (Brullo & al. 2010), but the karyotype features were not showed. All the investigated plants have the typical karyotype structure of the sect. *Allium* L., with chromosomes having linear satellites; in particular, there are 12 metacentric pairs

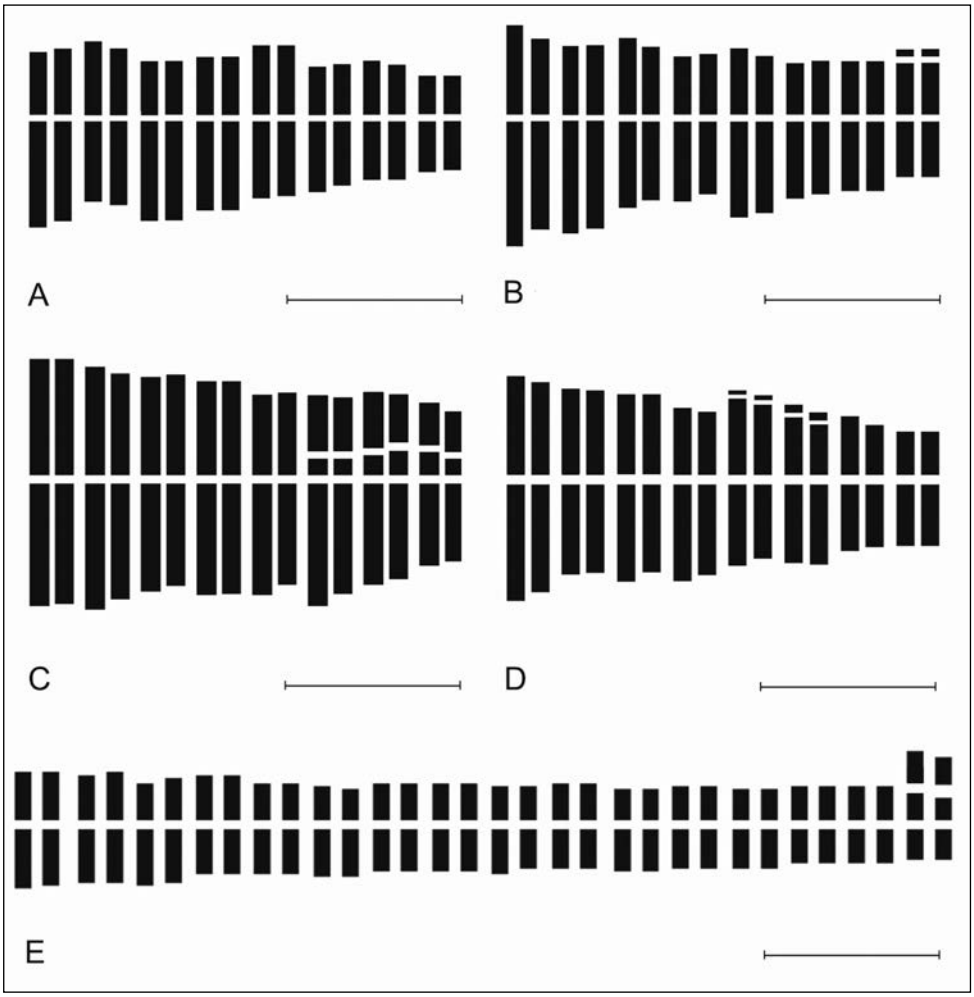

Fig. 2. Karyograms of the studied taxa: **A**, *Allium agrigentinum* from Aragona (Sicily), 2*n* = 16; **B**, *Allium apulum* from Mt. Sant'Angelo (Apulia), 2*n* = 16; **C**, *Allium chamaespathum* from Capo dell'Armi (Calabria), 2*n* = 16; **D**, *Allium diomedeum* from Ins. Tremiti (Apulia), 2*n* = 16; **E**, *Ptilostemon greuteri* from Mt. Inici (Sicily), 2*n* = 32. – Scale bars = **A-D,** 10 µm & **E,** 5 µm.

and 3 subtelocentric pairs with long linear satellites. Thus, the karyotype formula is: 2*n* = 2x = 10m + 6st-SAT = 16. The chromosomes on average vary in size from 14.1 ± 0.6 µm to 8.75 ± 1.3 µm, while the length of linear satellites is on average 3.2-2.3 µm, corresponding to c. 30% of the total chromosome length. The values of symmetry indices M_{CA} and Cv_{CL} are 28.1 and 15.4 respectively. On the whole, our results for the population studied are in accordance with previous reports from the Balkan peninsula (Bothmer 1974; Tzanoudakis 1985).

1979. *Allium diomedeum* Brullo, Guglielmo, Pavone & Salmeri — $2n = 2x = 16$ (Figs 1D & 2D).

It: Italia meridionale, Tremiti, San Domino, 18 Jul 1985, *Brullo*, *Minissale*, *Signorello & Spampinato* (CAT).

Allium diomedeum is an endangered species (Wagensommer & al. 2018) endemic to the Tremiti Islands, in the Adriatic Sea (Apulia, southern Italy). It is found in the shady rocky ledges of coastal cliffs and in the undergrowth of pine woods at the sea level. The chromosome number of the species ($2n = 16$) was reported for the type locality (San Domino, Tremiti Ins.) by Brullo & al. (2001) without further evidence of the karyotype structure. The chromosome complement of *A. diomedeum* mainly consists of more or less submedian pairs, with one pair submetacentric. Secondary constrictions were detected on the short arms of one metacentric pair and the submetacentric one. The karyotype formula can be summarized as follows: $2n = 2x = 8m + 2m\text{-SAT} + 4msm + 2sm\text{-SAT} = 16$. Chromosome total lengths range on average from 12.3 ± 0.5 µm of the longest chromosome to 6.1 ± 0.8 µm of the shortest chromosome, while the values of symmetry indices M_{CA} and Cv_{CL} are 11.20 and 20.83 respectively.

1980. *Ptilostemon greuteri* Raimondo & Domina — $2n = 4x = 32$ (Figs 1E & 2E).

Si: Castellammare del Golfo, Monte Inici, rupi calcaree della valle Il Finestrone, 07 Jul 2004, *Brullo, Campo, Musarella & Sciandrello* (CAT).

Ptilostemon greuteri is a very rare chasmophyte only known from a single locality on Mt. Inici, near Trapani, where it grows in a small valley in the NE limestone slopes. Differently from the other taxa of *Ptilostemon* occurring in Sicily [*P. niveus* (C. Presl) Greuter and *P. stellatus* (L.) Greuter], this is a perennial shrub up to 1.5 m tall, or more in cultivation, belonging to the section *Ptilostemon*. The species is currently assessed as Critically Endangered (CR) in the Italian national Red List (Rivers 2017). The chromosome complement is reported as $2n = 24$ by Raimondo & Domina (2006) being unique in *Ptilostemon* subg. *Ptilostemon*. Conversely, our investigations on the same single population revealed a tetraploid chromosome complement with $2n = 32$, the same as the other species of the sect. *Ptilostemon* (Greuter 1973; Tzanoudakis 1986; Brullo & al. 1991). The karyotype structure of this species is provided here for the first time. The karyotype is quite homogeneous and regular with very small chromosomes mainly of median type, varying in size from 3.22 to 2.06 µm; one pair is macrosatellited on the short arms of the homologues. The chromosomal formula can be represented as $2n = 4x = 24m + 2m\text{-SAT} + 6msm = 32$. It must be highlighted that the species is actually a diploidized tetraploid, with coupled homologues instead of tetraplets, as well exemplified by the occurrence of a single macrosatellited chromosome pair.

1981. ***Salvia ceratophylloides*** Ard. — 2*n* = 6x = 54 (Fig. 1F).

It: Calabria, Contrada Serro dei Morti press Puzzi (Reggio Calabria), 340 m, 24 May 2004, *Crisafulli, Maiorca & Spampinato* (CAL).

The count 2*n* = 54 is the first report for this species, which is a rare narrow endemism of southern Italy (Calabria), circumscribed to the suburban surroundings of Reggio Calabria, on coastal strip hilly ridges between 250 and 450 m of elevation (Spampinato & al. 2019). Based on the IUCN criteria, the species is currently assessed as Critically Endangered (CR) in the Italian national Red List (Spampinato & al. 2011; Rossi & al. 2014). *S. ceratophylloides* belongs to the subgenus *Sclarea* (Moench) Benth., Section *Plethiosphace* Benth., and is a part to the group of *S. pratensis* (Spampinato & al. 2019).

The chromosome number 2*n* = 54 reported in this study can be considered a hexaploid arrangement in accordance with the basic number x = 9 which is reported for some its allied taxa (*S. pratensis, S. haematodes*) resulting diploid with 2*n* = 18 (Del Carratore & Garbari 1996). A detailed analysis of the karyotype features is not presented because some small chromosomes appeared with indistinct centromeres.

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