Article

Borziana 1: 67-75
https://doi.org/10.7320/Borz.001.067
Version of Record published online on 3December 2020

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Lichen diversity on Trees Outside Forest in Molise (Central Italy)

Abstract

Caporale, S. & Ravera, S.: Lichen diversity on Trees Outside Forest in Molise (Central Italy). — Borziana 1: 67-75. 2020.

The role of Trees Outside Forest (TOF) in preserving biodiversity is highlighted through the study of their associated epiphytic lichen flora. Thirty TOF units located throughout the Italian region Molise have been surveyed. 170 trees of 17 different species have been examined. A total of 111 lichen taxa has been recorded. Among these, six taxa are new to the lichen flora of Molise (*Agonimia opuntiella*, *Arthonia excipienda*, *Arthopyrenia analepta*, *Lecanora albella*, *Mycomicrothelia confusa* and *Scytinium subtile*). One species is considered vulnerable, eight near threatened in Italy and 23 taxa are non-poleotolerant species usually growing on old trees in ancient, undisturbed forests. These findings suggest that TOF can provide surrogate habitat conditions for lichens usually occurring in natural environments.

Key words: Biodiversity, Lichenized Ascomycetes, TOF.

Introduction

Trees outside forest (TOF), i.e. scattered trees, road trees, riparian rows and wooded areas with less than 5,000 m^2 area (FAO 2001; de Foresta & al. 2013), can represent multifunctional resources and sources of biodiversity in areas where tree resource in forest is not accessible (e.g. on agricultural land, in urban and settlement areas, along road, in public park and private gardens, in rows of hedges, on pastures and rangelands) providing ecological corridors and habitat for several animal and plant species (Paletto & al. 2006; Paletto & Chincarini 2012).

Even if in the last years TOF characterization in terms of biodiversity conservation has been the subject of several studies mainly at the small/local spatial scale (Idol & al. 2011; Rossi & al. 2015) and in Central and Northern Europe tree in open habitats are considered suitable phorophytes for supporting rare epiphytic lichens (Douglass & al. 2010; Lõhmus & Liira 2013; Kubiak & Osyczka 2019), investigations on the role of TOF for lichen biodiversity in Mediterranean Europe is scanty. Only a few TOF types have been the subject of large-scale study. For example, old orchards are known to host interesting species (Zarabska & al. 2009) and, particularly old chestnut orchards, *Lobarion* and *Calicion* lichen communities (Ravera & al. 2006; Matteucci & al. 2012).

In Italy atmospheric pollution biomonitoring studies in urban areas have often highlighted the exclusive presence of sensitive species on scattered trees in parks and gardens (Loppi & al. 2002; Isocrono & al. 2007; Munzi & al. 2007), but despite the high potential of TOF to implement lichen diversity, no specific research concerning the significance of these landscape elements on conservation of valuable lichens exist. In this study, we took the opportunity of a recent characterization of the TOF of Molise (Marchetti & al. 2018), to carried out a large-scale lichen surveys detecting their role in an overlooked area of Central Italy. The study of lichen flora in the Molise region, the last of the administrative regions recognized in Italy, is quite recent. In fact, there are no regional reports in the first check-list of the Italian lichens (Nimis 1993) and it remains, according to the recent national update (Nimis 2016), the last region by number of reports regarding the lichen flora with only 490 taxa recorded in 17 years (Garofalo 1999; Nimis & Tretiach 1999, 2004; Frati & al. 2004; Baruffo & al. 2006; Caporale & al. 2008, 2011; Puntillo & Puntillo 2009; Ravera & al. 2009, 2010, 2010b, 2011; Nascimbene & al. 2010; Ravera & Genovesi 2010, 2012; Paoli & al. 2011, 2015; Genovesi & Ravera 2014; Guttovà & al. 2014; Martellos & al. 2014; Brackel 2015, 2016; Cocozza & al. 2016).

The aim of this work is to investigate the role of TOF in the conservation of epiphytic lichen biodiversity on a regional scale, simultaneously contributing to improve the floristic knowledge of the Molise region.

Material and methods

Study area

The Molise region covers 446,051 ha. The landscape mostly consists of mountain (55%) and hilly areas (44%). The Adriatic coast, 40 km long, is intensively urbanized. Industrial areas, as well as the main urban areas, are located in the eastern lowlands characterized by intensive farming in the flattest area close to the coast and by natural grasslands and pastures in the inland (Sallustio & al. 2016). The western part includes high mountain areas with low anthropic density and traditional agricultural activities in small farms.

Sampling and analysis

This project is part of a long-term program to characterize the TOF of the Molise region. The census started in 2008 and was completed in 2013 on the basis of orthophotos interpretation and subsequent field checks. The images were originally collected in 2000 (Marchetti & al. 2018). The sampling design comprised two levels of analysis (for details see Fattorini & al. 2016): the purpose of the first-level was to carry out a large-scale TOF inventory in the Molise region, based on the types classified in accordance with the Italian National Forest Inventory (available from www.infc.it). In the second-level phase, a subsample of 45 was randomly selected out of the total of 52,796 TOF units on the basis of a probabilistic sampling in order to describe plant diversity and structure of the TOF (Marchetti & al. 2018).

After a preliminary in situ check, 30 of these TOF units (Figure 1, Table 1) were found to be congruent with the ortophotos interpretation and accessible. These selected TOF span an altitudinal range between 65 and 925 meters above sea level. Following the phytoclimatic subdivision of Martellos & al. (2020), they belong almost exclusively to the dry submediterranean and the montane ecoregions representing the greatest part of the regional territory.

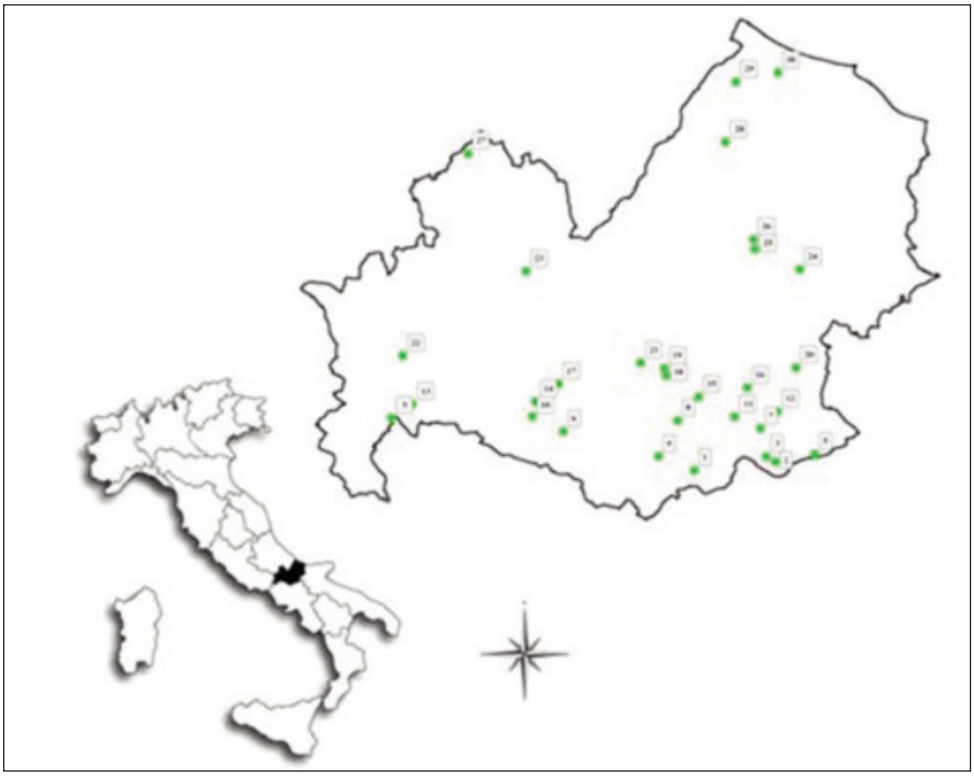

Fig. 1. Location of the 30 TOF units in the Molise region.

The lichen survey was carried out during summer 2017. In each TOF unit, lichens have been recorded on roots, stems and branches (between 0 and 200 cm from the ground) of all the trees with circumference ≥40 cm, trunk inclination less than 20°, disturbed surface less than 20 % of the total. A total of 170 trees were surveyed, belonging to 17 different tree species (Table 2) with a circumference range between 40 and 271 cm.

Some species were directly identified in the field using a magnifying glass and chemical spot tests K (a solution of 10 % potassium hydroxide), C (sodium hypochlorite solution), KC, while specimens of critical species were collected for correct identification in the laboratory using a dissecting microscope for macroscopic structures such as rhizines, eyelashes or hairs. A biological microscope, with ×5, ×10, ×20, ×40, ×100 magnification (with micrometric lens and possibility of oil immersion) was used for evaluating microscopic characters, such as asci, spores and paraphyses. Several identification keys were simultaneously used, mainly Clauzade & Roux (1985) and Smith & al. (2009). Nomenclature refers to the Index Fungorum database (2020). Ecological characterization of the species follows Nimis (2016). Specimens are stored in the personal herbarium of SC. The complete species list is reported in Annex I.

Table 1. Sampling sites.

TOF	Description	Coordinates (UTM WGS 84 32T)	Area (m^2)	Elevation (m)
1	Meso-xerophytic formation with *Quercus cerris*	474552E 4587150N	3204	670
2	Riparian formation	487461E 4588480N	2231	683
3	Mesophytic formation with *Q. cerris*	485997E 4589300N	2030	563
4	Meso-xerophytic formation with *Q. pubescens*	468842E 4589320N	3487	578
5	Meso-xerophytic formation with *Q. cerris*	493707E 4589710N	1042	780
6	Mesophytic formation with *Q. robur*	453757E 4593260N	4347	520
7	Formation with invasive hardwoods	485067E 4593720N	1606	601
8	Meso-xerophytic formation with *Q. cerris*	471914E 4594960N	1793	742
9	Riparian formation	426375E 4595390N	4272	206
10	Formation with invasive hardwoods	448790E 4595580N	3186	670
11	Thermophytic formation with *Q. pubescens*	480991E 4595560N	4531	671
12	Meso-xerophytic formation with *Q. pubescens*	487775E 4596400N	2505	410
13	Riparian formation	429729E 4597550N	4438	250
14	Riparian row	449331E 4597970N	3934	527
15	Meso-xerophytic formation with *Q. pubescens*	475233E 4598690N	2598	570
16	Meso-xerophytic formation with *Q. pubescens*	483013E 4600140N	2283	526
17	Riparian formation	452918E 4600730N	4548	750
18	Riparian formation	470042E 4602050N	1293	625
19	Riparian formation	469804E 4603160N	2056	590
20	Thermophytic formation with *Q. pubescens*	490726E 4603290N	1029	455
21	Formation with invasive hardwoods	466042E 4604020N	1020	772
22	Riparian formation	428208E 4605160N	4017	364
23	Meso-xerophytic formation with *Q. cerris*	447793E 4618440N	721	925
24	Meso-xerophytic formation with *Q. cerris*	491327E 4618720N	3184	887
25	Thermophytic formation with *Q. pubescens*	484194E 4621910N	943	352
26	Thermophytic formation with *Q. pubescens*	483978E 4623430N	4323	402
27	Riparian formation	438564E 4636730N	4301	860
28	Thermophytic formation with *Q. pubescens*	479515E 4638580N	4637	280
29	Thermophytic formation with *Q. pubescens*	481189E 4648010N	1109	110
30	Meso-xerophytic formation with *Q. cerris*	487873E 4649490N	961	65

Results and Discussion

One hundred-eleven lichens corresponding to more than 20% of the regional lichen flora and about 40% of the epiphytic lichens of Molise (Nimis 2016) were recorded.

They are mainly common and widespread lichens but six new species for Molise (*Agonimia opuntiella*, *Arthonia excipienda*, *Arthopyrenia analepta*, *Lecanora albella*, *Mycomicrothelia confusa* and *Scytinium subtile*) as well as red-listed species were also found.

Agonimia opuntiella (Buschardt & Poelt) Vězda is an epiphytic microscopic lichen characterized by small and loosely attached squamules and a hairy surface. It usually colonizes the basal part of old trees, also among mosses or detritus over soil or rock. It is a rarely reported species and little is known about its distribution in the Adriatic part of the

Table 2. Lichen host trees in the 30 TOF units.

Tree species	**Acronymo**
Ailanthus altissima (Mill.) Swingle	Aa
Acer campestre L.	Ac
Alnus glutinosa (L.) Gaertn.	Ag
Fraxinus excelsior L.	Fe
Fraxinus ornus L.	Fr
Juglans regia L.	Jr
Populus alba L.	Pa
Populus nigra L.	Po
Populus tremula L.	Pt
Prunus avium L.	Pr
Prunus dulcis (Mill.) D.A. Webb	Pd
Quercus cerris L.	Qc
Quercus pubescens Willd.	Qp
Quercus robur L.	Qr
Robinia pseudoacacia L.	Rp
Salix alba L.	Sa
Ulmus minor Mill.	Um

peninsula. The site characteristics confirm the autecology of the species with the exception of the altitude (780 m) that is significantly higher than in Nimis & Martellos (2020).

Arthonia excipienda (Nyl.) Leight. is a crustose non-lichenized species usually growing on the smooth bark of deciduous trees and shrubs in riparian montane woodlands. The species is considered "Vulnerable" in Italy (Nascimbene & al. 2013).

Arthopyrenia analepta (Ach.) Massal. is a crustose species recognized as an early coloniser of smooth bark, it is facultative lichenized or probably non-lichenized fungus, characterized by small black perithecia and an inconspicuous, hardly visible, thallus. It has a discontinuous distribution along the Italian peninsula, probably overlooked due to the difficulty of identification, so far never reported in the southern part of Adriatic Italy.

Lecanora albella (Pers.) Ach. is a crustose lichen with pruinose apothecia, found on smooth bark, with moderately acid pH, in humid-shaded habitats. It is quite similar to the more common *L. carpinea*, from which it differs by the negative C- reaction. The species is distributed throughout the Italian peninsula, mainly in the montane belt. As it is not resistant to eutrophication, it mainly occurres in natural or semi-natural habitats. Our specimen, collected in an agricultural context is worthy of interest as it confirms the potential of traditional agriculture landscapes for rare lichen species.

Mycomicrothelia confusa D. Hawksw. is a crustose species, with a thin, creamy white thallus and black globose perithecia, usually found on smooth-barked trees, generally in shady, humid and oceanic contexts. In spite of its known scarce tolerance to the anthropic impact and eutrophication, the specimen was collected near one important industrial areas of the region.

Scytinium subtile (Schrad.) Otálora, P. M. Jørg. & Wedin is a squamulose cyanolichen with a blueish grey to brown thallus, usually growing on the basal parts of old trees in mild-temperate areas. This is the southernmost record for the Italian peninsula.

More than half of the lichens recorded on TOF are crustose as usual for epiphytic lichens at these latitudes and in line with the Italian lichen flora. Among macrolichens (i.e. foliose and fruticose species), narrow-lobed foliose lichens (e.g. *Physcia* sensu lato), are the most abundant (17 taxa, 15.3%), as expected in a Mediterranean agro-pastoral landscape (Aragón & al. 2019).

From the ecological point of view, the lichens of TOF in Molise are mesophitic, moderately acidophitic, photophitic, and tolerant to eutrophication. However, there is an interesting group of species otherwise associated to natural or semi-natural habitats (29 taxa, 26.1%). Among them, five species (*Bacidia fraxinea*, *B. rosella*, *Gyalecta carneola*, *G. truncigena,* and *Leptogium cyanescens*) usually occur on old trees in ancient, undisturbed forests (Nimis 2016). A few suboceanic species have been also found: *Bacidia fraxinea*, *B. rosella*, *Physconia servitii*, *Staurolemma omphalarioides*. This element of biogeographical interest is very rare in Adriatic Italy (Nimis & Tretiach 1995, 1999).

Regarding the conservation status of the recorded taxa, in addition to the aforementioned *Arthonia excipienda* (VU), there are also 8 species (*Arthopyrenia salicis*, *Bacidia rosella*, *Buellia disciformis*, *Candelariella subdeflexa*, *Catapyrenium psoromoides*, *Gyalecta carneola*, *Leptogium cyanescens*, and *Thelenella modesta*) considered near threatened probably due to their sensitivity to human disturbance (Nascimbene & al. 2013).

Finally, it is noteworthy that, besides the finds of rare species, some are considered by different authors indicating ancient and well-preserved woods and long ecological continuity, often associated with the bryo-lichenic communities of the Lobarion (Zedda 2002, Nimis 2016, Rizzi & al. 2011) e.g., *Collema nigrescens*, *Fuscopannaria mediterranea*, *Gyalecta carneola*, *Normandina pulchella* and *Piccolia ochrophora*.

Conclusion

This study represents the first contribution the lichen flora of the TOF in Molise. In spite of the disturbance in the surrounding areas, mainly due to agricultural activities, and its relatively small extension, TOF hosts valuable species and could support the regional ecological network outside the protected areas. Rare, sensitive, and red-listed species may find in TOF a kind of "micro-refuge" which can host lichen communities that would be otherwise absent, providing surrogate habitat where more suitable and typical forest habitats are missing.

Acknowledgements

We are very grateful to Marco Di Cristofaro e Marco Ottaviano (University of Molise) for the valuable support provided during the field work.

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