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Biodiversity of rock, beach and water fungi in Italy

S. ONOFRI[1], A. ANASTASI[2], G. DEL FRATE[3], S. DI PIAZZA[4], N. GARNERO[2], M. GUGLIELMINETTI[3], D. ISOLA[1], L. PANNO[2], C. RIPA[1], L. SELBMANN[1],G. C. VARESE[2], S. VOYRON[2], M. ZOTTI[4], & L. ZUCCONI[1]

[1] Department of Ecological and Biological Sciences, University of Tuscia, Largo dell'Universita ` snc, 01100 Viterbo, Italy;
[2] Department of Plant Biology, University of Turin, Viale Mattioli 25, 10125 Turin, Italy;
[3] Department of Earth and Environmental Sciences, Section of Mycology, University of Pavia, via S. Epifanio 14, 27100 Pavia, Italy and
[4] Mycology Laboratory, Department for Territory and its Resources Study (DIP.TE.RIS), Polo Botanico ''Hanbury'', University of Genoa, Corso Dogali 1M, I16136 Genoa, Italy

Abstract

The fungal biodiversity in its overall is mostly still unknown and the ecological role of these organisms, particularly in some border ecosystems, is often underestimated. This study aims to give both an overview of the state of the art and to present new data on the mycodiversity in some peculiar environments as rocks, beach sand, and water in Italy. Particularly, rock fungi are here reported from high mountain peaks, sea cliffs, and monuments; sand associated fungi from beach ecosystems in Puglia and Ligurian coasts; marine fungi associated with the endemic seagrass of the Mediterranean *Posidonia oceanica* L.; aquatic hyphomycetes (Ingoldian fungi) from both streams in the Ticino Natural Park and lentic water in Lago Maggiore; fungi from the water distribution system in Turin. Ecological and evolutive considerations are put forward.

Keywords: Beach fungi, Italy, marine fungi, mycodiversity, rock fungi, water fungi,

Introduction

Fungi were among the first colonizers of terrestrial habitats at least as symbionts (Heckman et al. 2001 Heckman, D S, Geiser, D M, Eidell, B R, Stauffer, R L, Kardos, N L and Hedges, S B. 2001. Molecular evidence for the early colonization of land by fungi and plants. *Science*, 293: 1129–1133. Labandeira 2005 Labandeira, C C. 2005. Invasion of the continents: cyanobacterial crusts to tree-inhabiting arthropods. *Trends Ecol Evol*, 20: 253–262.). Nowadays they colonize any kind of environment and represent one of the widest and diversified Kingdoms (Hawksworth 2001 Hawksworth, D L. 2001. The magnitude of fungal diversity: The 1.5 million species estimate revised. *Mycol Res*, 105: 1422–1432.); they may adopt different lifestyles being saprotrophs, symbionts or parasites and playing an irreplaceable role in maintaining the stability of different ecosystems. Some species are cosmopolitan while others are adapted to harsh environments precluded to other life-forms. Due to their ecological fitting (Agosta & Klemens 2008 Agosta, S J and Klemens, J A. 2008. Ecological fitting by phenotypically flexible genotypes: Implications for species associations, community assembly and evolution. *Ecol Lett*, 11: 1123–1134.) fungi may easily jump to new environments including the human body. Despite the crucial role of these organisms in any ecosystem, their biotechnological potential and their possible involvement in human health, data on their biodiversity and ecology, particularly in extreme ecosystems, are often scant. Therefore the importance of mycological studies in any ecosystem, including human environments, might not be underestimated.

Rocks are among the less investigated substrata for mycological studies. Black meristematic fungi are specialists in the extremes and thrive in and on these hard substrata. They are among the main groups of microorganisms causing weathering of rocks and biodeterioration of monuments exposed to outdoor conditions (Sterflinger & Krumbein 1997 Sterflinger, K and Krumbein, W E. 1997. Dematiaceous fungi as a major agent for biopitting on Mediterranean marbles and limestones. *Geomicrobiol J*, 14: 219–230.; Sterflinger et al. 1997 Sterflinger, K and Krumbein, W E. 1997. Dematiaceous fungi as a major agent for biopitting on Mediterranean marbles and limestones. *Geomicrobiol J*, 14: 219–230.; Wollenzien et al. 1995 Wollenzien, U, de Hoog, G S, Krumbein, W E and Urzì, C. 1995. On the isolation of microcolonial fungi occurring on and in marble and other calcareous rocks. *Sci Total Environ*, 167: 287–294., 1997; De Leo & Urzì, 2003; De Leo et al. 1999 De Leo, F, Urzì, C and de Hoog, G S. 1999. Two *Coniosporium* species from rock surfaces. *Stud Mycol*, 43: 70–79. , 2003). Recent studies based on worldwide sampling of exposed natural rocks from the Mediterranean to the Poles, including Antarctica, as well as monuments, revealed a bewildering and mostly still unknown biodiversity that is worth investigating (Diakumaku et al. 1995 Diakumaku, E, Gorbushina, A A, Krumbein, W E, Panina, L and Soukharjeski, S. 1995. Black fungi in marble and limestones: An aesthetical, chemical e physical problem for the conservation of monuments. *Sci Total Environ*, 167: 295–304.; Wollenzien et al. 1995 Wollenzien, U, de Hoog, G S, Krumbein, W E and Urzì, C. 1995. On the isolation of microcolonial fungi occurring on and in marble and other calcareous rocks. *Sci Total Environ*, 167: 287–294.; Gorbushina 2003 Gorbushina, A A. 2003. Methodologies and techniques for detecting extraterrestrial (microbial) life. Microcolonial fungi: Survival potential of terrestrial vegetative structures. *Astrobiology*, 3: 543–554.; Volkmann & Gorbushina 2006 Volkmann, M and Gorbushina, A A. 2006. A broadly applicable method for extraction and characterisation of mycosporines and mycosporine-like amino acids of terrestrial, marine and freshwater origin. *FEMS Microbiol Lett*, 255: 286–295. Isola et al. 2007 Isola, D, Onofri, S, Zucconi, L and Selbmann, L. 26–29 settembre 2007. "Caratterizzazione molecolare di funghi melanizzati isolati da rocce provenienti da ambienti estremi". In *Dati preliminari* 26–29 settembre, 328Palermo 102° Congresso Società Botanica Italiana; Sert et al. 2007 Sert, H B, Sümbül, H and Sterflinger, K. 2007. Microcolonial fungi from antique marbles in Perge/Side/Termessos (Antalya/Turkey). *Antonie van Leeuwenhoek*, 91: 217–227.; Selbmann et al. 2005 Selbmann, L, de Hoog, G S, Mazzaglia, A, Friedmann, E I and Onofri, S. 2005. Fungi at the edge of life: Cryptoendolithic black fungi from Antarctic deserts. *Stud Mycol*, 51: 1–32. , 2008; Ruibal et al. 2005 Ruibal, C, Platas, G and Bills, G F. 2005. Isolation and characterization of melanized fungi from limestone formations in Mallorca. *Mycol Prog*, 4: 23–38., 2008, 2009).

Beaches are further niches scarcely considered by mycologists. Nonetheless the quality of beach sediments has been considered of primary importance by World Health Organization (WHO 2003 WHO. 2003. *Guidelines for safe recreational water environments, coastal and fresh waters*, vol. 1, Geneva: WHO Regional Office for EU.), which highlighted the existence of potential microbiological contamination of the sediment matrix. Furthermore, the presence of some types of fungi has been suggested as an indicator of environmentally clean beaches (Enríquez et al. 2009 Enríquez, D L, Minter, D W and González, M C. 2009. *IMI descriptions of fungi and bacteria set 181*, Wallingford, UK: CAB International.). Therefore recent researches in Italy have tried to assess the level of fungal and microbial contamination and the potential sanitary hazards for humans (Mancini et al. 2004 Mancini, L, D'Angelo, A, Pierdominici, E, Ferrari, C, Venturi, LFormichetti, P. 2004. Microbiological quality of Italian beach sands. *Microchem J*, 79: 257–261.; Soussi et al. 2007 Soussi, M, Boutayeb, H and Guessous, N. 2007. Fungal flora from the sand of two beaches of Casablanca: Analysis and epidemiological corollary. *J Mycol Med*, 17: 58–62.).

Little is known also on fungi colonizing marine habitats and freshwater despite their pivotal role in these ecosystems as parasites, symbionts and saprotrophs, nutrient recycling and energy flow. Recent molecular studies on marine fungi demonstrated that mycodiversity in marine environments is much higher than previously thought (Koch et al. 2007 Koch, J, Pang, K L and Jones, E BG. 2007. *Rostrupiella danica* gen. et sp. nov., a *Lulworthia*-like marine lignicolous species from Denmark and the USA. *Bot Mar*, 50: 1–8.); studies on freshwater fungi are scant and mycologists have focused on this topic only discontinuously in the time, while recent investigations on drinking water highlighted that fungi are currently present as contaminants (Hageskal et al. 2009 Hageskal, G, Lima, N and Skaar, I. 2009. The study of fungi in drinking water. A review. *Mycol Res*, 113: 165–172.).

Fungi are organisms with wide metabolic competences: the above listed environments, still scarcely investigated, may represent an invaluable source of fungi of still unexplored biotechnological potential.

Black fungi from rocks and monuments in Italy

Rock is a challenging substratum to colonize, due to scarcity of nutrients and water, temperature fluctuations, exposure to solar radiation. Therefore rock inhabitants are obliged to endure long periods of stress-induced inactivity, and active life is possible only during brief periods when conditions become more permissive (Palmer et al. 1987 Palmer, F E, Emery, D R, Stemmler, J and Staley, J T. 1987. Survival and growth of microcolonial rock fungi as affected by temperature and humidity. *New Phytol*, 107: 155–162.; Gorbushina et al. 2008 Gorbushina, A A, Kotlova, E R and Sherstneva, O A. 2008. Cellular responses of microcolonial rock fungi to long-term desiccation and subsequent rehydration. *Stud Mycol*, 61: 91–97.). Black meristematic fungi, also called black yeasts or MCF (Micro-Colonial Fungi, Sterflinger 2005 Sterflinger, K. 2005. “Black yeasts and meristematic fungi: Ecology, diversity and identification”. In *Yeast handbook: Biodiversity and ecophysiology of yeasts*, Edited by: Rosa, C and Gabor, P. 505–518. New York, NY: Springer.), are a polyphyletic group of fungi sharing basic features, as melanized cell walls and meristematic development, enabling them to tolerate diverse stressful environmental conditions. They commonly live in saltpans, acidic environments, and above all, on both natural rocks and stone monuments (Staley et al. 1982 Staley, J T, Palmer, F and Adams, B. 1982. Microcolonial fungi: Common inhabitants on desert rocks?. *Science*, 215: 1093–1095.; Wollenzien et al. 1997 Wollenzien, U, de Hoog, G S, Krumbein, W E and Uijthof, J MJ. 1997. *Sarcinomyces petricola*, a new microcolonial fungus from marble in the Mediterranean basin. *Antonie van Leeuwenhoek*, 71: 281–288.; Sterflinger & Krumbein 1997 Sterflinger, K and Krumbein, W E. 1997. Dematiaceous fungi as a major agent for biopitting on Mediterranean marbles and limestones. *Geomicrobiol J*, 14: 219–230.; Zalar et al. 1999 Zalar, P, de Hoog, G S and Gunde-Cimerman, N. 1999. Ecology of halotolerant dothideaceous black yeasts. *Stud Mycol*, 43: 38–48. ; Ruibal et al. 2005 Ruibal, C, Platas, G and Bills, G F. 2005. Isolation and characterization of melanized fungi from limestone formations in Mallorca. *Mycol Prog*, 4: 23–38. ; Selbmann et al. 2005 Selbmann, L, de Hoog, G S, Mazzaglia, A, Friedmann, E I and Onofri, S. 2005. Fungi at the edge of life: Cryptoendolithic black fungi from Antarctic deserts. *Stud Mycol*, 51: 1–32. , 2008).

Rock black fungi have been isolated from monuments (Diakumaku et al. 1995 Diakumaku, E, Gorbushina, A A, Krumbein, W E, Panina, L and Soukharjeski, S. 1995. Black fungi in marble and limestones: An aesthetical, chemical e physical problem for the conservation of monuments. *Sci Total Environ*, 167: 295–304.; Wollenzien et al. 1995 Wollenzien, U, de Hoog, G S, Krumbein, W E and Urzì, C. 1995. On the isolation of microcolonial fungi occurring on and in marble and other calcareous rocks. *Sci Total Environ*, 167: 287–294.; Sterflinger 2000 Sterflinger, K. 2000. Fungi as geologic agents. *Geomicrobiol J*, 17: 97–124. [Taylor & Francis Online], [Web of Science ®]; Gorbushina 2003 Gorbushina, A A. 2003. Methodologies and techniques for detecting extraterrestrial (microbial) life. Microcolonial fungi: Survival potential of terrestrial vegetative structures. *Astrobiology*, 3: 543–554. [CrossRef], [PubMed], [Web of Science ®]; Volkmann & Gorbushina 2006 Volkmann, M and Gorbushina, A A. 2006. A broadly applicable method for extraction and characterisation of mycosporines and mycosporine-like amino acids of terrestrial, marine and freshwater origin. *FEMS Microbiol Lett*, 255: 286–295.; Sert et al. 2007 Sert, H B, Sümbül, H and Sterflinger, K. 2007. Microcolonial fungi from antique marbles in Perge/Side/Termessos (Antalya/Turkey). *Antonie van Leeuwenhoek*, 91: 217–227.) and natural rocks both in dry and extremely cold or extremely hot climates, ranging from the Mediterranean (Ruibal et al. 2005 Ruibal, C, Platas, G and Bills, G F. 2005. Isolation and characterization of melanized fungi from limestone formations in Mallorca. *Mycol Prog*, 4: 23–38. 2008) to the Antarctic (Selbmann et al. 2005 Selbmann, L, de Hoog, G S, Mazzaglia, A, Friedmann, E I and Onofri, S. 2005. Fungi at the edge of life: Cryptoendolithic black fungi from Antarctic deserts. *Stud Mycol*, 51: 1–32. , 2008), as well as from high mountain tops in the Andes and Alps (Isola et al. 2007 Isola, D, Onofri, S, Zucconi, L and Selbmann, L. 26–29 settembre 2007. "Caratterizzazione molecolare di funghi melanizzati isolati da rocce provenienti da ambienti estremi". In *Dati preliminari* 26–29 settembre, 328Palermo 102° Congresso Società Botanica Italiana; Selbmann et al. 2008 Selbmann, L, de Hoog, G S, Zucconi, L, Isola, D, Ruisi, SGerrits van den Ende, A HG. 2008. Drought meets acid: three new genera in a dothidealean clade of extremotolerant fungi. *Stud Mycol*, 61: 1–20.).

They are phylogenetically quite heterogeneous (Ruibal et al. 2009 Ruibal, C, Gueidan, C, Selbmann, L, Gorbushina, A A, Crous, P WGroenewald, J Z. 2009. Phylogeny of rock-inhabiting fungi related to Dothideomycetes. *Stud Mycol*, 64: 123–133.) and the morphological similarities between distant groups might be considered a consequence of convergent evolution (Ruibal et al. 2008 Ruibal, C, Platas, G and Bills, G F. 2008. High diversity and morphological convergence among melanised fungi from rock formations in the Central Mountain System of Spain. *Persoonia*, 21: 93–110.). There is no clear correlation between particular fungal lineages and the rock substratum (Ruibal 2004 Ruibal, C. 2004. *Isolation and characterization of melanized, slow-growing fungi from semiarid rock surfaces of central Spain and Mallorca*, Madrid, Spain: Universidad Autónoma de Madrid/Merck, Sharp & Dohme de España. Ph.D. dissertation) but, to date, despite the remarkable range of environments from which colonized rocks have been collected, the possible relation between phylogeny and the physical and chemical parameters characterizing the source location (for example temperature and salinity) has not yet been studied. This contribution aims to highlight the specificity, if any, between rock fungal genotypes and particular environmental conditions focusing on a wide selection of rock isolates from Italy preserved in the CCFEE (Culture Collection of Fungi from Extreme Environments, University of Viterbo, Italy), Mycological section of the Italian National Antarctic Museum "Felice Ippolito". Italy is, in fact, a perfect laboratory for such studies because of its geographic diversity and richness of historical heritages. Black fungi were isolated according to methods described by Selbmann et al. (2005 Selbmann, L, de Hoog, G S, Mazzaglia, A, Friedmann, E I and Onofri, S. 2005. Fungi at the edge of life: Cryptoendolithic black fungi from Antarctic deserts. *Stud Mycol*, 51: 1–32.) or by using a sterile needle (Wollenzien et al. 1995 Wollenzien, U, de Hoog, G S, Krumbein, W E and Urzì, C. 1995. On the isolation of microcolonial fungi occurring on and in marble and other calcareous rocks. *Sci Total Environ*, 167: 287–294.). The medium used for isolation was Dichloran Rose-Bengal *Chloramphenicol Agar* (DRBC Agar, Oxoid Ltd., Basingstoke, Hampshire, UK). Fifty-four strains of rock black fungi isolated from 19 rock samples collected in six different locations of the Alps and three in the Apennines are here considered; in addition, 10 fungal strains from four locations on the coasts (sea cliffs), 22 directly from monuments in Cagliari (Sardinia), one from Sangallo Fort in Civita Castellana (VT) and 10 from a cave in Vallerano (VT) were studied. ITS rDNAs were sequenced for the selected strains and compared in public domain (NCBI, http://blast. ncbi.nlm.nih.gov/Blast.cgi) using the BlastN algorithm. Strains with the same, or very close, match in the GenBank were grouped in the same genotype and, although with some variations within individual groups, pooled into 15 different genotypes. Results are reported in Table I. All strains fall in two classes of *Ascomycota*: *Dothideomycetes* and *Eurotiomycetes* and the most representative order is *Capnodiales*.

Table I. Similarity searches and genotypes of rock black fungi isolated from mountains, coasts and monuments

	Location	CCFEE n.[a]	Genotype	BlastN search results[b]
Alps	Stolemberg (AO) – Mt Rosa. 3200 m a.s.l 45°53′25.73″N 7°52′0.13″E	5386; 5400; 5398	1	*Cladosporium* sp. CBS 282.49 (89)
		5385; 5498	2	*Capnobotryella* sp. MA 4701 (100)
		Strain A29	2	*Capnobotryella* sp. MA 4701 (97)
		5416; 5461; 5462; 5465; 5466; 5469; 5608	3	Uncultured fungus clone K5-16 (96–97)
		5478; 5493	4	*Phaeococcomyces chersonesos* Ch49 (85–86)
		5411; 5412; 5607	5	Uncultured Herpotrichiellaceae (88–91)
		5394; 5401	6	*Catenulostroma protearum* CPC 15370 (91–93)
	P.ta Indren (AO) – Mt Rosa. 3300 m a.s.l 45° 53′59.73″ N 7° 52′ 33.44″ E	5414	1	*Cladosporium* sp. CBS 282.49 (89)
		5456	1	Fungal sp. TRN 43[f] (97)
		5393; 5468	2	*Capnobotryella* sp. MA 4701 (100)
		Strain O24	3	Uncultured fungus clone K5-16 (97)
		5469	3	Uncultured fungus clone K5-16 (97)
		5391	*Recurvomyces mirabilis*	*Recurvomyces mirabilis* (99)[c]
		5392; 5500	8	*Penidiella ellipsoidea* CBS 128773 (91)
		5459; 5463	9	*Capnobotryella* sp. MA 4899 (96)
	Colle delle Pisse (AO) – Mt Rosa. 3112 m a.s.l 45°52′38.52″N 7°′53’9.38″E	5388	1	Fungal sp. TRN 43 (97)
		5460; 5477; 5496; 5547	3	Uncultured fungus clone K5-16 (97–99)
		5389; 5499	6	*Catenulostroma protearum* CPC 15370 (91)
		5554	10	*Cryomyces antarcticus* CBS 116301 (90)
	Corno Bianco (BZ) – Dolomites. 3100 m a.s.l 45°49′32.17″N 7°52′18.14″E	5545	9	*Capnobotryella* sp. MA 4899 (96)
	Alpe di Siusi – Val Gardena (BZ). 2010 m a.s.l 46°33′24.61″N 11°39′7.49″E	5395; 5396	1	Fungal sp. TRN 43 (97)
	Stelvio National Park (BZ) – Val De la Mare. 3000 m a.s.l 46°24′43.87″N 10°38′29.92″E	5415	1	*Cladosporium* sp. CBS 282.49 (87–90)
		5458	1	Fungal sp. TRN 43 (97)
		5470	3	Uncultured fungus clone K5-16 (97)
		5476	10	*Cryomyces antarcticus* CBS 116301 (91)
		5417; 5464	11	*Cladophialophora humicolae* CBS 117536 (93–95)

Apennines	Mt Camicia (AQ) – 1600 m a.s.l 42°26′22.63″ N 13°43′ 0.3.93″E	5584; 5588	9	*Capnobotryella* sp. MA 4899 (97)
	Campo Imperatore (AQ) – Gran Sasso. 2200 m a.s.l 42°26′29.3″ N 13°33′34.6″ E	5556	12[d]	*Phaeococcomyces chersonesos* Ch49 (94)
	Gran Sasso (AQ) – Corno Piccolo. 2566 m a.s.l 42°28′28.12″N 13°34′11.26″ E	5596	13	*Coniosporium* sp. MA 4597 (90)[e]
Coasts	Bosa – Torre Argentina's beach (OR). s.l 40°19′08.96″ N 8°26′ 38.54″E	5555	12	*Phaeococcomyces chersonesos* Ch49 (99)
		5569	13	*Coniosporium* sp. MA 4597 (94)[e]
	Cagliari – St. Elia's beach s.l 39°11′06.67″ N 9° 08′41.93″E	5551	2	*Capnobotryella* sp. MA 4701 (100)
		5540; 5559; 5579	*Phaeotheca triangularis*	*Phaeotheca triangularis* (100)
		5552	12[d]	*Sarcinomyces petricola* OUCMBI101244 (97)
	Cagliari – Calamosca's cliffs. 5 m a.s.l 39°10′ 55.26″ N 9°09′ 30.81′ E	5580; 5581	*Phaeotheca triangularis*	*Phaeotheca triangularis* (100)
	Isola d'Elba – Barbarossa Bay (LI). s.l 42°46′08.43″N 10°24′32.05″E	5666	12	*Phaeococcomyces chersonesos* Ch49 (96)
Monuments	Cagliari – Torre dell'Elefante. 60 m a.s.l 39°13′05.08″N 9°06′53.66″E	5564	12	*Phaeococcomyces chersonesos* Ch49 (96)
	Cagliari – Lion sculpture in front of St Mary's Cathedral. 60 m a.s.l. 39°13′ 07.51″ N 9°06′ 59.36″ E	5657; 5658; 5659; 5715; 5718	9	*Capnobotryella* sp. MA 4899 (96)
		5656; 5716; 5717	12	*Phaeococcomyces chersonesos* Ch49 (96)
		5660	14	*Coniosporium uncinatum* CBS 100219 (98)
	Cagliari – Porta Cristina. 60 m a.s.l. 39°13′ 18.03″ N 9° 06′ 59.40″ E	5730	12	*Phaeococcomyces chersonesos* Ch49 (99)
	Cagliari – Monumental Cemetery of Our Lady of Bonaria. s.l. – 39°12′ 36.96″ N 9°07′ 26.13″ E	5703	*Phaeotheca triangularis*	*Phaeotheca triangularis* (100)
		5714; 5770	9	*Capnobotryella* sp. MA 4899 (96)
		5702; 5705	12	*Phaeococcomyces chersonesos* Ch49 (99)
	Cagliari – Seasons sculptures of Boyl Palace. 60 m a.s.l. 39°13′ 00.17′ N 9°06′ 57.78″ E	5722; 5725; 5726; 5728; 5729; 5732	12	*Phaeococcomyces chersonesos* Ch49 (96)
	Civita Castellana (VT) – Sangallo Fort 147 m a.s.l. 42°17′ 18.82″ N 12°24′ 30.28″ E	5719	12	*Phaeococcomyces chersonesos* Ch49 (96)
	Vallerano (VT) – Holy Saviours Cave 356 m a.s.l. 42°23′24.22′ N 12°16′03.01″E	5671; 5672; 5673; 5674; 5675; 5676; 5680; 5681; 5684; 5685	15	*Devriesia lagerstroemiae* CPC 14403 (89–95)

Notes: [a]CCFEE – Colture Collection of Fungi from Extreme Environments. [b]The values in parentheses indicate the percentage of similarities in GenBank, in all cases the *E*-value was 0.0. [c]CCFEE 5391 is described as *Recurvomyces mirabilis* in Selbmann et al. (2008). [d]*Phaeococcomyces chersonesos*, *Sarcinomyces petricola* grouped in the genotype 12 because conspecific (Tsuneda et al. 2011). [e]*Coniosporium* sp. MA 4597 is the holotype of *C. sümbülii* (Sert & Sterflinger 2010). [f]Strain TRN 43 belong to the Family of *Davidiellaceae* (Ruibal et al. 2008) and has been grouped in genotype 1 (*Cladosporium* sp. CBS 282.49).

The only strains with a certain identification are CCFEE 5391, identified as *Recurvomyces mirabilis* Selbmann & de Hoog, and CCFEE 5540, 5559, 5579, 5580, 5581, 5703 from coastal sites of Sardinia and the monumental Cemetery of Cagliari with 100% identity with *Phaeotheca triangularis* de Hoog & Beguin. *R. mirabilis* was repeatedly isolated from Antarctica and the Alps (Selbmann et al. 2008 Selbmann, L, de Hoog, G S, Zucconi, L, Isola, D, Ruisi, SGerrits van den Ende, A HG. 2008. Drought meets acid: three new genera in a dothidealean clade of extremotolerant fungi. *Stud Mycol*, 61: 1–20.) while *P. triangularis* was described from humidifiers of air-conditioning systems (de Hoog et al. 1997 de Hoog, G S, Beguin, H and van de Batenburg-Vegte, W H. 1997. *Phaeotheca triangularis*, a new meristematic black yeast from humidifier. *Antonie van Leeuwenhoek*, 71: 289–295.) and, more recently, isolated from saltpans (Butinar et al. 2005 Butinar, L, Sonjak, S, Zalar, P, Plemenitaš, A and Gunde-Cimerman, N. 2005. Melanized halophilic fungi are eukaryotic members of microbial communities in hypersaline waters of solar salterns. *Bot Mar*, 48: 73–79.) indicating an ecological preference for moist, oligotrophic conditions and salinity.

Genotype 10 (strains CCFEE 5554, 5476) from high altitude in the Alps show significant similarity with the putative endemic Antarctic genus *Cryomyces* Selbmann, de Hoog, Mazzaglia, Friedmann & Onofri. Genotypes 5 (Mt Rosa), 11 (Stelvio National Park), and 12 (Sardinia, Elba Island, Civita Castellana and Campo Imperatore) belong to the order *Chaetothyriales*. A few genotypes have an apparently peculiar ecology being exclusive to mountains or to coasts. Genotype 1, for example, belonging to a not yet described taxon with affinities to the *Davidiellaceae*, and genotype 3 have been isolated exclusively at high altitudes in both western and eastern Alps. Genotype 15, showing similarities with *Devriesia lagerstroemiae* Crous & M.J. Wingf. CPC 14403 within the *Teratosphaeriaceae* (Crous et al. 2009 Crous, P W, Schoch, C L, Hyde, K D, Wood, A R, Gueidan, Cde Hoog, G S. 2009. Phylogenetic lineages in the *Capnodiales*. *Stud Mycol*, 64: 17–47.), was repeatedly isolated from the rural area of Vallerano on a wall painting. Genotype 2 is close to *Capnobotryella* sp. MA 4701 while genotype 9 matches with *Capnobotryella* sp. MA 4899. These two strains have been recently demonstrated to have different phylogenetic affinities: the first is related with the strain NH4-3 belonging to *C. renispora* while the second, described as new species *C. kiziroglui* Sert & Sterfl., pools in a completely separated group (Sert et al. 2011 Sert, H B, Sümbül, H and Sterflinger, K. 2011. Two new species of *Capnobotryella* from historical monuments. *Mycol Prog*, 10: 333–339.).

Fungi in the Order *Chaetothyriales* are particularly recurrent on the coasts and on monuments that are more influenced by human activities with respect to mountain sites; rock fungi in this order are known for their ability to metabolize aromatic compounds (Prenafeta-Bold[ugrave] et al. 2006) and the presence of pollutants in these sites may explain their higher presence.

Remarkably, in this study we did not find rock fungal species frequently recorded from natural rocks and monuments in the Mediterranean. *Coniosporium apollinis* Sterfl. and *C. perforans* Sterfl., for instance, were isolated from marble monuments in Greece (Sterflinger et al. 1997 Sterflinger, K and Krumbein, W E. 1997. Dematiaceous fungi as a major agent for biopitting on Mediterranean marbles and limestones. *Geomicrobiol J*, 14: 219–230.), *C. sümbülii* Sert & Sterfl. from Turkey (Sert & Sterflinger 2010 Sert, H B and Sterflinger, K. 2010. A new *Coniosporium* species from historical marble monuments. *Mycol Prog*, 9: 353–359.) and *C. uncinatum* De Leo, Urzì & de Hoog in France (De Leo et al. 1999 De Leo, F, Urzì, C and de Hoog, G S. 1999. Two *Coniosporium* species from rock surfaces. *Stud Mycol*, 43: 70–79.); *Sarcinomyces petricola* Wollenz. & de Hoog was isolated from marble and calcarenite in Greece and Italy (Wollenzien et al. 1997 Wollenzien, U, de Hoog, G S, Krumbein, W E and Uijthof, J MJ. 1997. *Sarcinomyces petricola*, a new microcolonial fungus from marble in the Mediterranean basin. *Antonie van Leeuwenhoek*, 71: 281–288.); *Pseudotaeniolina globosa* De Leo, Urzì & de Hoog was isolated from the sandstone outside wall of Santa Maria di Mili church in Mili San Pietro, Messina, Italy (De Leo et al. 2003 De Leo, F, Urzì, C and de Hoog, G S. 2003. A new meristematic fungus *Pseudotaeniolina globosa*. *Antonie van Leeuwenhoek*, 83: 351–360.); *Hormonema carpetanum* Bills, Peláez & Ruibal was isolated from granite and sandstone in Spain (Bills et al. 2004 Bills, G F, Collado, J, Ruibal, C, Peláez, F and Platas, G. 2004. *Hormonema carpentanum* sp. nov., a new lineage of dothideaceous black yeasts from Spain. *Stud Mycol*, 50: 149–157.); *Capnobotryella antalyensis* Sert & Sterfl. (Sert et al. 2007 Sert, H B, Sümbül, H and Sterflinger, K. 2007. Microcolonial fungi from antique marbles in Perge/Side/Termessos (Antalya/Turkey). *Antonie van Leeuwenhoek*, 91: 217–227.), the recently described species *C. kiziroglui* MA 4899 and *Capnobotryella erdogani* Sert & Sterfl. MA 4625 (Sert et al. 2011 Sert, H B, Sümbül, H and Sterflinger, K. 2011. Two new species of *Capnobotryella* from historical monuments. *Mycol Prog*, 10: 333–339. [CrossRef], [Web of

Science ®]) were isolated from historical marble monuments in Turkey; *Phaeococcomyces chersonesos* Bogom. & Minter, *P. exophialae* (de Hoog) de Hoog, *Phaeosclera dematioides* Sigler, Tsuneda & J.W. Carmich., *Phaeotheca fissurella* Sigler, Tsuneda & J.W. Carmich., and *Sarcinomyces petricola* were isolated from various sources in Greece and Crimea (part of Ukraine with a Mediterranean climate) (Bogomolova & Minter 2003 Bogomolova, E V and Minter, D W. 2003. *IMI Descriptions of fungi and bacteria set 156*, Wallingford, UK: CAB International.). *Pseudotaeniolina globosa* and *Sarcinomyces petricola* are the only species reported from Italy on monuments in Sicily. The first was assigned to the order *Capnodiales* on the basis of SSU and ITS phylogeny (De Leo et al. 2003 De Leo, F, Urzì, C and de Hoog, G S. 2003. A new meristematic fungus *Pseudotaeniolina globosa*. *Antonie van Leeuwenhoek*, 83: 351–360.) and the second to the *Chaetothyriales* on the basis of a four-locus phylogeny (Gueidan et al. 2008 Gueidan, C, Ruibal, C, de Hoog, G S, Gorbushina, A, Untereiner, W A and Lutzoni, F. 2008. A rock-inhabiting ancestor for mutualistic and pathogen-rich fungal lineages. *Stud Mycol*, 61: 111–119.). The fungi here studied match with many of rock species above mentioned but only with low identity; the genotype 13, for instance, shows an identity of 90–94% with *Coniosporium* sp. MA 4597 (*C. sümbülii*); genotype 12 an identity of 94–99% with *P. chersonesos* Ch49; genotype 9 shows an identity of 96–97% with *C. kiziroglui*. These low identities suggest that most of our strains belong most probably to new taxa. These data in their overall suggest that the biodiversity on rocks is still far to be fully elucidated but rocks seem to be a more attractive substratum than might be expected, representing a promising niche to search new species.

In this work it has been highlighted for the first time a clear distribution pattern of rock fungal genotypes according with the sites where the samples come from. Therefore, although the nature of rock substratum seems not to be selective for specific fungal lineages (Ruibal 2004 Ruibal, C. 2004. *Isolation and characterization of melanized, slow-growing fungi from semiarid rock surfaces of central Spain and Mallorca*, Madrid, Spain: Universidad Autónoma de Madrid/Merck, Sharp & Dohme de España. Ph.D. dissertation), the results here reported to prove that environmental factors play instead a pivotal role.

Microfungi in beach ecosystems

The beach ecosystem is an ecotone, a dynamic environment defined by sea surface waves, tide, meteorological-climatic conditions, and sediment contributions (Brown & Mclachlan 1990). Beaches can be defined on the basis of three main units: backshore, foreshore, and shoreface.

A backshore represents the area of a shore that lies between the average high tide mark and the vegetation. Foreshore (also called intertidal zone) is the area that is above water at low tide and under water at high tide. Shoreface is the active littoral zone off the low water line.

There have been few studies of beach fungi, particularly in the last decade, and they concern backshore and foreshore zones (Vogel et al. 2007 Vogel, C, Rogerson, A, Schatz, S, Laubach, H, Tallman, A and Fell, J. 2007. Prevalence of yeasts in beach sand at three bathing beaches in South Florida. *Water Res*, 41: 1915–1920.; Cathrine & Raghukumar 2009 Cathrine, S J and Raghukumar, C. 2009. Anaerobic denitrification in fungi from the coastal marine sediments off Goa, India. *Mycol Res*, 113: 100–109.). Another source of information on this topic and on marine fungi is represented by Enríquez et al. (2009 Enríquez, D L, Minter, D W and González, M C. 2009. *IMI descriptions of fungi and bacteria set 181*, Wallingford, UK: CAB International.). The presence of marine fungi on beaches is likely correlated to the abundance of plant or algal material (Enríquez et al. 2003 Enríquez, D L, Gonzalez, M C, Ruiz, G, Nunez, R and Delgado, Y. 2003. Diversidad de Hongos Marinos en Playas de la Ciudad de La Habana. Marine fungi diversity in beaches of Havana city.

Serie Oceanografica, 1: 1–9. ; Figuera & Barata 2007 Figuera, D and Barata, M. 2007. Marine fungi from two sandy beaches in Portugal. *Mycologia*, 99: 20–23.) and this fact can motivate the scarce relevance of marine fungi on Italian beaches, which are often characterized by a high level of human disturbance due to their closeness to big towns.

The quality of beach sediments has, however, been considered of primary importance by the World Health Organization (WHO 2003 WHO. 2003. *Guidelines for safe recreational water environments, coastal and fresh waters*, vol. 1, Geneva: WHO Regional Office for EU.), which highlighted the existence of potential microbiological contamination of the sediment matrix. This explains why recent research in Italy has tried to assess the level of fungal and microbial contamination and the potential sanitary hazards for humans (Mancini et al. 2004 Mancini, L, D'Angelo, A, Pierdominici, E, Ferrari, C, Venturi, LFormichetti, P. 2004. Microbiological quality of Italian beach sands. *Microchem J*, 79: 257–261.; Soussi et al. 2007 Soussi, M, Boutayeb, H and Guessous, N. 2007. Fungal flora from the sand of two beaches of Casablanca: Analysis and epidemiological corollary. *J Mycol Med*, 17: 58–62.). It is worth noting that, for balance, the presence of some types of fungi may be considered as an indicator of environmentally clean beaches (Enríquez et al. 2009 Enríquez, D L, Minter, D W and González, M C. 2009. *IMI descriptions of fungi and bacteria set 181*, Wallingford, UK: CAB International.).

A work on this issues was performed in 12 beaches with high anthropic impact of Puglia coast (six to Adriatic and six to Ionic coast). The sands of swashed zone and emerged beach were investigated and the related analysis revealed the presence of 18 different genera of saprotrophic and opportunistic filamentous fungi such as *Acremonium, Alternaria, Chaetomium, Cladosporium, Curvularia, Fusarium, Mucor, Rhizopus, Scopulariopsis, Stachybotrys*, *Trichoderma*, *Verticillium, Penicillium* and *Aspergillus* (Online Appendix A). These latter genera showed the broadest of species with *A. niger* dominant. No yeast was found. In order to study keratinophilic fungi were employed the “modified hair-bait” technique (Orr 1969 Orr, G F. 1969. Keratinophilic fungi isolated from soil by a modified hair bait technique. *Sabouraudia*, 7: 129–134.). By this method five different genera were detected including the genus *Aphanoascus* with *A. fulvescens, A. kerathinophilum* and *A. terreus* as prevalent species (Online Appendix B). No dermatophyte species were found. No keratinophilic fungi were observed in the beaches near to great cities like Monopoli (Bari) or Brindisi, where the biodiversity of saprotrophic genera was high, probably for the severe sewage pollution of sea water. There are few qualitative differences between saprotrophic and keratinophilic fungi sampled from swashed zones and backshore.

Another study regards with Ligurian coast. The latter is among the most urbanized and industrialized in Italy: the main causes of disturbance are littoral urban development and harbour activities, the building of littoral railways and roads, and the presence of several polluting outflows. Microfungi were investigated in the Swash Zone Interstitial Water (SZIW) and sediments of an urban beach of Genoa, Italy (Vezzulli et al. 2009 Vezzulli, L, Zotti, M, Marin, V, Moreno, M, Pezzati, E and Fabiano, M. 2009. Swash zone interstitial water is a reservoir of fungal microorganisms on a Mediterranean beach (Genoa, Italy). *J Mar Biol Assoc UK 2, Marine Biodivers Rec 2: e19.*,). Fungi observed included ubiquitous species (e.g. *Acrostalagmus luteoalbus* (Link) Zare, W. Gams & Schroers, *Cladosporium cladosporioides* (Fresen.) G.A. de Vries, *Chaetomium cochlioides* Palliser, *Geotrichum candidum* Link, *Rhizopus oryzae* Went & Prins. Geerl., *Trichoderma atroviride* P. Karst., *T. hamatum* (Bonord.) Bainier, *T. harzianum* Rifai, *Ulocladium chartarum* (Preuss) E.G. Simmons), opportunistic or true pathogens (e.g. *Aspergillus fumigatus* Fresen., *A. niger* Tiegh.) and yeasts (*Candida* spp., *Rhodotorula* spp.). The results reveal that the concentration and diversity of fungi are higher in the SZIW than in beach sand, a fact apparently never previously reported in the literature. The concentration of fungi seems to be correlated with temperature and protein concentrations, but not with the concentration of faecal bacteria. The SZIW may thus constitute an important reservoir of microfungi in the beach environment. This may be due to a favourable combination of environmental factors in the matrix, such as high water content, presence of organic matter, temperature, and darkness. The promising results achieved through one year of investigation stimulated further studies on the SZIWs of three other urban beaches. Preliminary results (obtained by seasonal multiple samples starting in Spring 2010) seem to confirm the great variety of fungi present in the SZIW. Until now, 303 fungal strains have been isolated belonging to 16 genera. The species were identified by classical methodology but the identifications are waiting for being confirmed by DNA analyses that are still in progress. The most commonly occurring species (whose identification was confirmed) have been (sorted in decreasing order of occurrence) *Aspergillus oryzae* (Ahlb.) E. Cohn, *A. niger* Tiegh., *Penicillium brevicompactum* Dierckx, *Rhizopus oryzae* Went & Prins. Geerl., *Trichoderma pseudokoningii* Rifai, *T. asperellum* Samuels, Lieckf. & Nirenberg.

Marine fungi associated with the seagrass Posidonia oceanica

Marine fungi are halophilic or xerophilic organisms that form an integral part of marine ecosystems and play important ecological roles as parasites, symbionts and saprotrophs, involved in nutrient recycling, energy flow and synthesis of humic substances. Teleomorphic and anamorphic *Ascomycota* are the predominant fungi in marine environments (Raghukumar 2008 Raghukumar, C. 2008. Marine fungal biotechnology: an ecological perspective. *Fungal Divers*, 31: 19–35.). Currently, about 1500 binomials for obligate marine fungi can be found in the literature, but many are inadequately described or are merely synonyms. Information on facultative marine fungi is even more confusing (Jones & Mitchell 1996). Furthermore, molecular study based on the metagenomic approach has demonstrated that fungal diversity in marine environments is much higher than previously thought (Koch et al. 2007 Koch, J, Pang, K L and Jones, E BG. 2007. *Rostrupiella danica* gen. et sp. nov., a *Lulworthia*-like marine lignicolous species from Denmark and the USA. *Bot Mar*, 50: 1–8.).

The study of the taxonomic and functional diversity of marine fungi is very important, not only from an ecological point of view, but also for biotechnological and pharmacological applications. For example, enzymes produced by marine fungi could be important in biotechnology thanks to their greater stability in the harsh conditions of industrial processes, such as the bio-treatment of hyper-saline textile industry wastewaters. In pharmacology, in recent years, research on the chemistry of marine fungi has led to the discovery of a surprisingly large number of novel substances. Over 500 new compounds have been described so far and 15 compounds of marine origin are currently in human clinical trials (Muhammad et al. 2007 Muhammad, S, Muhammad, S A, Shafqat, H, Abdul, J, Muhammad, A and Yong, S L. 2007. Marine natural products of fungal origin. *Nat Prod Rep*, 24: 1142–1152.).

In a recent study (Panno 2009 Panno, L. 2009. “Biodiversità fungina associata alla fanerogama marina *P. oceanica*”. In *Tesi di Laurea*, 1–129. Università degli Studi di Torino, Facoltà di Scienze MMFFNN.), fungi associated with different organs (leaves, rhizomes, roots and matte-tangle of dead rhizomes and roots in which the sediment is trapped) of *Posidonia oceanica* L., an endemic seagrass of the Mediterranean Sea, were investigated. A total of 88 fungal taxa, mainly *Ascomycota*, were isolated and identified. Composition and structure of these fungal populations varied significantly between the four organs. Only two species (*Penicillium chrysogenum* Thom and *P. janczewskii* K.M. Zalessky) were isolated from all parts. This specificity may be due to multiple factors: specific environmental parameters (nutrients, light, intensity of exposure to hydrodynamic motions, etc.); presence of different antagonistic microorganisms, particularly epiphytes or herbivores; presence of toxic or repellent molecules localized in a specific organ of *P. oceanica*, e.g. the presence of high concentration of tannic acid in the leaves. The genera with the highest levels of occurrence and numbers of species were *Penicillium*, *Cladosporium* and *Acremonium*. Most of the isolated fungi, cultured at different salt concentrations, were tested for oxidoreductase (laccases, peroxidases) and tannases activity (Panno et al. 2010 Panno, L, Voyron, S, Anastasi, A and Varese, G C. 2010. Marine fungi associated with the sea grass *Posidonia oceanica* L.: A potential source of novel metabolites and enzymes. *J Biotechnol*, 150S: S1–S576.). Several fungal isolates showed strong enzyme production, many exclusively at high salt concentrations. These findings suggest that marine fungi play an important ecological role in decomposition of ligninocellulosic matrices in the marine environment and are a good source of novel extremoenzymes that can be used in different biotechnological applications (Panno et al. 2010 Panno, L, Voyron, S, Anastasi, A and Varese, G C. 2010. Marine fungi associated with the sea grass *Posidonia oceanica* L.: A potential source of novel metabolites and enzymes. *J Biotechnol*, 150S: S1–S576.). This work contributed to better knowledge of marine fungi and showed that fungi associated with *P. oceanica* are very rich and varied. *P. oceanica* is the climax vascular plant of the Mediterranean Sea and belongs to a seriously threatened plant community, being identified as a priority habitat in Annex I of the Habitats Directive. Future studies will clarify the ecological role of the fungi associated with this seagrass, increasing our knowledge of the complex interactions within this plant community, which are essential to its preservation. Finally, the creation of a collection of marine fungi provides the scientific community with a large number of isolates which will be investigated for production of secondary metabolites of pharmaceutical and biotechnological interest.

Ingoldian and other aquatic fungi in the Po river basin

Ingold (1942 Ingold, C T. 1942. Aquatic hyphomycetes of decaying older leaves. *Trans British Mycol Soc*, 25: 339–417.) first described aquatic hyphomycetes. He pioneered studies on the morphology, taxonomy and ecology, of what are now known as Ingoldian fungi. These fungi colonize submerged leaves of the riparian vegetation and have an active role in the aquatic ecosystem energy flow and in the trophic chain dynamics (Kaushik & Hynes 1971 Kaushik, N K and Hynes, H BN. 1971. The fate of dead leaves that fall into streams. *Arch Hydrobiol*, 68: 465–515.). The first Italian records were published by Ciferri (1959 Ciferri, R. 1959. Osservazioni ecologiche su ifomiceti acquatici. *Academia Republ Romaine Omagiu lui Train Savulescu*, pp. : 157–162.) who isolated 11 species from the Po and Ticino basins and irrigation canals in Pavia and its environs. No further data were published in Italy until 1983, when a study on aquatic hyphomycetes from some streams located on the plain (Ticino Park) and in mountains of northern Italy was reported (Del Frate 1983 Del Frate, G and Caretta, G. 1983. Aquatic hyphomycetes of a mountain stream in Valsesia (Piemonte). *Hydrobiologia*, 102: 69–71. [CrossRef], [Web of Science ®], [CSA]). Other studies were carried out in lentic (Lago Maggiore) and lotic environments, concerning hyphomycetes and yeasts, not all typically aquatic (Bonferoni et al. 1980 Bonferoni, B, Dacarro, C, Brandone, A, Specchiarello, M, Del Frate, G and Picco, A. 1980. Recenti acquisizioni sul livello di inquinamento del fiume Ticino nel territorio comunale di Pavia. *Inquinamento*, 3: 37–43. ; Del Frate & De Margaritis 1986 Del Frate, G and De Margaritis, B. 1986. I lieviti delle acque del Lago Maggiore. *Il Notiziario dell'Ecologia*, 3: 24–30.). In these surveys, correlation between presence of yeasts and chemical–physical parameters, were discussed in relation to anthropic activities. In a survey of the water of a small pool by plating of the membrane filtration, some keratinophilic fungi were reported by Mangiarotti and Caretta (1984 Mangiarotti, A M and Caretta, G. 1984. Keratinophilic fungi isolated from a small pool. *Mycopathol*, 85: 9–11.). Some water moulds belonged to the *Saprolegnaceae*, a group formerly included in *Phycomycetes*, but not now considered as belonging in kingdom Fungi, were recently isolated on fish carcasses in low-order streams in the north western Italian Apennines (Fenoglio et al. 2010 Fenoglio, S, Bo, T, Cammarata, M, Malacarne, G and Del Frate, G. 2010. Contribution of macro- and micro-consumers to the decomposition of fish carcasses in low-order streams: An experimental study. *Hydrobiologia*, 637: 219–228.). This study suggests that these organisms might be more important than invertebrates in carcass decomposition. Finally, *Achlya*, a water mould, was frequently isolated in rice field water in Piedmont and Lombardy, reducing seedling survival (Angelini 2008 Angelini, R. 2008. *Il Riso*, 363–364. Milano: Bayer CropScience S.r.l.).

As regards Ingoldian fungi, up to now, 45 species have been recorded from Italy (Online Appendix C), some records not yet published. New records refer to streams running through woodlands located in areas of low human impact. The most frequent species are, *Lunulospora curvula* Ingold, *Flagellospora curvula* Ingold, *Clavariopsis aquatica* De Wild., *Anguillospora crassa* Ingold, *A. longissima* (Sacc. & P. Syd.) Ingold, *Campylospora chaetocladia* Ranzoni, *Lemonniera aquatica* De Wild. A few species, found only on the plain, are considered tropical (*L. curvula* Ingold and *C. chaetocladia* Ranzoni); most, whether from the plain or from mountain streams, are typically of temperate climates; a few are isolated only in mountain streams. Quantitative data, obtained by means of membrane filtration (8 µm pore size), are reported in some other papers (Del Frate & Caretta 1983 Del Frate, G and Caretta, G. 1983. Aquatic hyphomycetes of a mountain stream in Valsesia (Piemonte). *Hydrobiologia*, 102: 69–71.; Rodino et al. 2003 Rodino, D, Tosi, S and Del Frate, G. 2003. Ifomiceti acquatici in un canale del Parco del Ticino. *Studi Trent Sci Nat, Acta Biol*, 80: 55–57. , 2004). Microscopic examination of stained membranes permits the count and identification of the star-shaped spores and long thin spores so distinctive for these fungi. High peaks of occurrence appear in autumn in Ticino Park, with more than 14,700 conidia per liter. Two peaks occur in mountain streams, the first in autumn (1850 conidia per liter) and a lower second peak in spring samples. In the Ticino Park water samples, the spring peak is very low or absent.

Fungi in water distribution systems

Some fungi are primarily adapted to aquatic environments, and are naturally found in water. These zoosporic fungi belong to phylum *Chytridiomycota*. Fungi belonging to the other phyla in the kingdom *Fungi* are primarily adapted to the terrestrial environment (Kirk et al. 2008 Kirk, P M, Cannon, P F, Minter, D W and Stalpers, J A. 2008. *Ainsworth & Bisby's dictionary of the fungi.* , 10th ed, Wallingford, Oxfordshire: CABI Publishing.). Although water is not considered to be their primary habitat, they are associated intrinsically with water, which they can enter in various ways, and there are much evidences to show that they are commonly present, with a high diversity, in surface water and groundwater. In these environments, in fact, fungi have been recorded at averages from 1000 to 10,000 CFU l^{-1} with peaks that can reach 30,000 CFU^{-1} (Göttlich et al. 2002 Göttlich, E, van der Lubbe, W, Lange, B, Fiedler, S, Melchert, I and Reifenrath, M. 2002. Fungal flora in groundwater derived public drinking water. *Int J Hyg Environ Health*, 205: 69–279.).

It is now recognized that fungi are common contaminants of water distribution systems and of all types of water (raw, treated, ultra-pure), including bottled drinking water (Hageskal et al. 2009 Hageskal, G, Lima, N and Skaar, I. 2009. The study of fungi in drinking water. A review. *Mycol Res*, 113: 165–172.). The quality of drinking water is typically expressed in terms of pathogenic microorganisms present in a given volume of water. Certain microorganisms, including various bacteria, viruses, and parasites are well-known water contaminants (Mara & Horan 2006 Mara, D and Horan, N. 2006. *The handbook of water and wastewater microbiology.* , 1st ed, 819London: Elsevier Academic Press.), but fungi rarely have been studied and characterized as water contaminants, although some authors have reported that exposure can occur through inhalation of aerosolized fungi from water. Sweden seems to be the only country in Europe that has current legislation specifying fungi: the average total charge may not exceed 100 CFU/100 ml (Anonymous, 2003 Anonymous. 2003. “Drinking water regulations (In Swedish)”. In *SLVFS*

2001:30, Stockholm, Sweden: National Food Administration.). At present, Italian legislation on water intended for human consumption (Decreto Legislativo 31/2001) does not fix a legal limit for fungi, and considers monitoring of fungi in drinking water as "incidental" and therefore not included in routine analysis. To fill this gap, between July 2006 and July 2007, a survey of the water system of Turin was conducted (Garnero 2008 Garnero, N. 2008. "I funghi delle acque destinate al consumo umano. Analisi della micoflora nelle acque di pozzo del circondario torinese". In *Tesi di Laurea*, 1–153. Università degli Studi di Torino, Facoltà di Scienze MM FFNN.). That survey demonstrated that 94% of analyzed samples contained fungal colonies with an average total fungal load ranging between 2 and 11 CFU/100 ml. Filamentous fungi were isolated from 91% of analyzed samples, while yeasts were isolated from only 17% of samples. In total 163 fungal entities belonging to 69 species were identified. Most of the isolated taxa belonged to anamorphic fungi, and among them the most frequent species were *Acremonium strictum* W. Gams, *Cadophora fastigiata* Lagerb. & Melin, *Paecilomyces lilacinus* (Thom) Samson, *Phoma leveillei* Boerema & G.J. Bollen and *Pyrenochaeta* sp. Although none of the isolated species is considered as pathogenic according to Italian law (Dlgs 81/08), 24 of the species encountered had already been reported as opportunistic pathogens of humans or animals, or as producers of mycotoxins (de Hoog & Guarro 1995 de Hoog, G S and Guarro, J. 1995. *Atlas of clinical fungi*, Baarn and Delft, the Netherlands: Centraalbureau voor Schimmelcultures. ; Richardson & Warnock 1997 Richardson, M D and Warnock, D W. 1997. *Fungal infection diagnosis and management.* , 2nd ed, Oxford, UK: Blackwell Science Ltd. ; Weinderborner 2001 Weinderborner, M. 2001. *Encyclopedia of food mycotoxins*, Berlin Heidenberg: Springer-Verlag.). The risk arising from the presence of fungi in water intended for human consumption is not known and the small amount of information available is often contradictory. Further investigation of drinking water will therefore be necessary to establish the possible adverse effects of fungi and/or their metabolites to consumers health, taking in account also the effects arising from use of contaminated water in the food chain.

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