

Full Length Research Paper**Research on truffles: Scientific journals analysis****Małgorzata Gajos¹ and Dorota Hilszczańska^{2*}**¹Department of Biomedical Computer Systems, University of Silesia, Będzińska 39, 41-200 Sosnowiec, Poland.²Department of Forest Ecology, Forest Research Institute, Braci Leśnej 3, Sękocin Stary, 05-090 Raszyn, Poland.

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This analysis aims to provide an update in the recent truffle research through the chosen articles published in scientific journals. The journals were chosen based upon journal profiles and scientific prestige. Authors have considered publications from: Applied and Environmental Microbiology, Biochemical Journal, BMC Bioinformatics, BMC Genomics, Current Genetics, Environmental Microbiology, Eukaryotic Cell, FEMS Microbiology Ecology, Food Chemistry, Fungal Diversity, Fungal Genetics and Biology, Journal of Agricultural and Food Chemistry, Mycological Research (Fungal Biology), Mycorrhiza and New Phytologist. The number of analysed 124 articles shows the results over the twenty years period (1993-2012). Critical analysis has been used to determine thematic scope, whilst bibliometric study identifies development directions.

Key words: Truffles, mycorrhiza, chemical and biological properties.**INTRODUCTION**

Truffles are the macrofungi that form underground (hypogeous) fruit bodies. These true truffles of *Tuber* genus belong to ascomycetes (spore sac fungi). The fungi live in ectomycorrhizal association with a broad variety of gymnosperm and angiosperm hosts in a variety of habitats including subtropical cloud forests, temperate forests, boreal forests, floodplains, tree nurseries, restoration sites and Mediterranean woodlands (Bonito et al., 2010). These ectomycorrhizal fungi are of great interest for the ecosystems they colonise because of the mutualistic associations and the advantages they provide to host plants (Pacioni and Comandini, 1999). The geographic distribution of known truffle species (about 100) mainly covers the temperate zones of the northern hemisphere, with at least three differentiation areas: Europe, South-East Asia and North America. The geographical distribution patterns of truffles are not fully understood, although recently it has been shown that evolutionary lineages can be related to the biogeographical origin of the host species (Bertould et al., 1998, 2001; Martin et al., 2002; Moyersoen et al., 2003; Murat et al., 2004).

Several members of the true truffle genus *Tuber* are

highly praised and priced gourmet food, their aroma and taste are known worldwide (Rubini et al., 2007). Some of the most precious edible fungi are found amongst scented truffles: *Tuber magnatum* Pico - the white truffle and *Tuber melanosporum* Vittad - the Perigord black truffle. Although the species are mainly limited to the Mediterranean region, another one species, highly appreciated as delicacies - Burgundy truffle (*Tuber aestivum* Vitt. syn. *Tuber uncinatum* Chat.) is found throughout Europe. The natural geographical distribution of *T. aestivum* ranges from North Africa to Sweden and from Ireland to Russia (Chevalier and Frochot, 1997; Wedén et al., 2001). The presence of *T. aestivum* in some parts of Poland was mentioned by Lubelska (1953). However, due to lack of any herbarium specimens, the identity of this species could not be confirmed. Few years ago, new data on the distribution of *T. aestivum* and other species of truffles have been reported from Poland (Hilszczańska et al., 2008), Slovakia (Gazo et al., 2005), Slovenia (Piltaver and Ratosa, 2006) and Gotland (Wedén et al., 2001, 2004a, b).

The Burgundy truffle forms ectomycorrhiza with, for example, beech (*Fagus sylvatica*), birch (*Betula*), common

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hornbeam (*Carpinus betulus*), hop hornbeam (*Ostrya carpinifolia*, particularly important in Italy), hazelnut (*Corylus avellana*), oak (*Quercus robur*). Inventory made by Hilszczajska et al. (2008) showed that, in Poland, host species other than *Q. robur* and *C. avellana* and including *F. sylvatica*, *C. betulus* and *Tilia cordata* are of great importance in regard to the *Tuber* species. These findings are in accordance with a rich body of literature (Chevalier and Frochot, 1997; Chevalier et al., 2002; Gazo et al., 2005; Pacioni and Comandini, 1999). *T. aestivum* was not associated with any ground-layer vegetation species or communities.

Strong ecological relationships exist between *Tuber* spp., host plants and soil type (Lulli et al., 1999). Truffles mostly grow at various altitudes (from a few meters above sea level to between 800 to 1000 m), in calcareous soils with a sub-alkaline pH (7 to 8), although a few species, such as *T. mesentericum*, are adapted to slightly acidic soils (pH 6 to 7) (Pacioni and Comandini, 1999). Soils must be permeable, well-aerated and well-drained, without stagnant water and also poor, in terms of N, P and Fe, but with a good content of Ca, K and S (Granetti, 1994; Bencivenga, 1998; Pacioni and Comandini, 1999).

Requirements as to pH differ between species, especially those linked to coniferous trees. The environmental patterns of *Tuber* have been studied mainly in economically important species such as: *T. melanosporum*, *T. aestivum* and *T. magnatum*. The first two species prefer well drained, stony soils, while the latter one grows in deeper, fresh, sandy-clay soils (Bencivenga and Granetti, 1990). Slight changes in water potential, pH, nutrient and ion concentration can favour the ectomycorrhiza of one species and repress of another, especially in black truffles species. Study on *T. aestivum* showed a good adaptability to different pedoclimatic conditions and a higher resistance to dryness in respect to other black truffles as *T. brumale* and *T. melanosporum* (Pomarico et al., 2007).

Some species of truffles, e.g. *T. melanosporum* and *T. aestivum*, produce a change in the rhizosphere of their host trees, creating an area around the tree, known as a "brule" or "pianello", which is almost circular and looks as if it has been a burnt area.

This is due to allelopathic and chemical effects of some volatile organic compounds they produce (Pacioni and Comandini, 1999; Splivallo et al., 2007b; Streiblová et al., 2012).

Study of volatile organic compounds (VOCs) emitted by truffles revealed their phytotoxic effect (Pacioni and Pomponi, 1991). To date, around 200 VOCs from various truffle species have been identified, some of them described for the first time (Splivallo et al., 2007a). Their number is likely to continue growing (Tarkka and Piechulla, 2007). They are generally simple aliphatic containing functional groups such as alcohols, aldehydes, aromatic compounds, esters, furans, hydrocarbons,

ketones, nitrogen and sulphur-containing compounds (Splivallo et al., 2007a). VOCs are released in the course of the entire truffle life cycle, including free-living mycelia, mycorrhizae, and ascocarps (Talou et al., 1989; Zeppa et al., 2004; Splivallo et al., 2007a, 2009, 2012). Profound metabolic differences between vegetative growth and the fructification stage are documented by aromatic spectra that differ in free-living mycelia versus ascocarps (Splivallo et al., 2007a).

Truffles' vegetative stage based on *T. melanosporum* is represented by a primary free-living mycelium (Paolocci et al., 2006) derived from spores. This mycelium is a facultative saprotrophic that obtains nutrients by decomposing dead and decaying organic matter from the soil. It seems that the saprotrophic ability of the free-living mycelium is low and its survival very limited and intended to give rise to mutualistic symbiotic associations in contact with roots of a wide range of host plants (Martin et al., 2010; Martin, 2011; Rubini et al., 2011a, b). The switch to the symbiotic stage is seen by the formation of ectomycorrhizae that consist of hyphae intimately associated with the intercellular space of the peripheral tissue of apical root tips. Partners exchange commodities required for their proper growth and survival. Carbon compounds of photosynthetic origin from the host plant are directed to the fungus, and various nutrients, especially inorganic nitrogen and phosphate, are transported from the fungus to the symbiotic plant (Frank, 2005). Moreover, the fungus helps the host tree to support high calcium levels and offers a source of moisture (e.g. Smith and Read, 2008). Thanks to new molecular data, the mutualistic exchange of nutrients was shown to involve specific genes/proteins in combination with environmental factors, such as nutrient shortage (Martin et al., 2002; Martin and Nehls, 2009). Publishing of *T. melanosporum* genome (Martin et al., 2010) has given researchers unique opportunities to learn more about the biology of the fungus. Large scale biochemical analyses combined with transcript profiling can be used to identify specific gene networks controlling metabolic pathways (Agostini et al., 2001; Ambra et al., 2004; Splivallo et al., 2011).

T. melanosporum was thought to be homothallic or exclusively selfing (Bertauld et al., 1998). To date, molecular investigations have brought a profound re-evaluation of the sexuality of *T. melanosporum* having also implication for other truffle species. The current concept envisages a haploid extraradical mycelium composed of opposite sexual polarity hyphae, which are non-uniformly distributed in their natural localities, and after obligate outcrossing (Martin et al., 2010; Rubini et al., 2011a, b), they give rise to subterranean hypogeous ascocarps. It is speculated that control of outcrossing by mating (MAT) genes and most sex- and fruiting-related pathways identified in *T. melanosporum* is comparable with other heterothallic ascomycetes (Paolocci et al., 2006; Rubini et al., 2007, 2011a; Murat and Martin, 2008;

Riccioni et al., 2008a).

Except for well-known nutritional importance and unique aroma of truffles, their reported biological activities have also drawn scientific attention as they are believed to have positive effects in the development of truffle-related products. Some of their bioactivities include antiviral and antimicrobial activities (Hussan and Al-Ruqaie, 1999; Janakat et al., 2004, 2005), antioxidant capacities, hepatoprotective activity, anti-mutagenic properties as well as anti-inflammatory effects. Both antiviral and antimicrobial activities have traditionally been the most studied biological activities of truffles with most researchers limiting their scope to desert truffles. The term "desert truffles" comprises species of different hypogeous Ascomycetes genera, such as *Terfezia*, *Balsamia*, *Delastreopsis*, *Delastria*, *Leucangium*, *Mattirolomyces*, *Phaeangium*, *Picoa*, *Tirmania* and some *Tuber* species (Honrubia et al., 1992).

Hussan and Al-Ruqaie (1999) reported that truffles (*Terfeziaspp.*) possessed antiviral activity which may have potential for the treatment of eye and skin diseases. Janakat et al. (2005) found antimicrobial activity in *T. claveryi* extracts against *Pseudomonas aeruginosa*.

Antimutagenc acivities of black truffles (*T. aestivum*) from Italy were reported by Fratianni et al. (2007). This research group found that both aqueous and ethanol fresh as well as irradiated *T. aestivum* possessed inhibitory effects against the standard mutagens. The aqueous extracts from fresh truffles showed a stronger inhibitory effect against *S. typhimurium* TA98 compared to the other irradiated truffles. This study demonstrated the therapeutic potentials of some truffles. It should be noted that most of these activities were relatively weak. It is unclear if one or two particular bioactive compounds in the truffles have significant contributions on their particular activity.

The knowledge of both the spatial distribution and genetic diversity of the most relevant truffle taxa can provide a tool for planning *in situ* conservation programs. It is possible to identify a reserve by selecting the territory capturing the maximum of diversity within the least number of grid cells (Rebelo and Sigfried, 1992). Nonetheless, the assessment of *Tubertaxa* spatial distribution is difficult given that the underground ascomata can be localized only using specifically trained dogs. Given the last point, building a new tool that could help in detecting localities of truffles occurrence seems to be out of question.

The aim of the research is to determine the state of art and investigation research directions about truffles. Fungi truffles have been selected as the research subject for their unique values as foodstuffs and their positive impact on human health and well-being (Patel, 2012).

MATERIALS AND METHODS

Scientific journals play an important role in the promotion of science and offer in themselves a source of data for research into the way

research workers develop their interests. To study this, both qualitative and quantitative methodologies are necessary.

A detailed analysis was done of articles about truffles in selected journals. The research proceedings relied upon article categorisation intended to determine research directions. Categorization is the process in which ideas and objects are recognized, differentiated, and understood. Category illuminates a relationship between the subjects and objects of knowledge (Categorisation). To run the categorisation process, the body of literature analysis and critics method was used. The literature analysis as a scientific examination method is used to review scientific works and for peer review (Ankem, 2008). To present the results the bibliometric research method was used. This method is a statistical application for quantitative studies of facts, phenomena and processes related to texts and information (Diodato, 1994).

In order to select journals for research into truffles knowledge development directions, a list of periodicals of international reach was taken from the Journal Citation Reports (JCR). The JCR offers a systematic, objective means to critically evaluate the world's leading journals, with quantifiable, statistical information based on citation data (Journal Citation Reports). Based on the periodical title and scientific prestige factor - impact factor (IF, 2011), 30 journals were selected: New Phytologist (6.645), Environmental Microbiology (5.843), Biochemical Journal (4.897), (Fungal Diversity) 4.769, BMC Genomics (4.073), Applied and Environmental Microbiology (3.829), Fungal Genetics and Biology (3.737), Food Chemistry (3.655), Eukaryotic Cell (3.604), FEMS Microbiology Ecology (3.408), Journal of Agricultural and Food Chemistry (2.823), Mycological Research (2.809), BMC Bioinformatics (2.751), Mycorrhiza (2.630), Process Biochemistry (2.627), Current Genetics (2.556), Fungal Ecology (2.507), Forest Ecology and Management (2.487), Postharvest Biology and Technology (2.411), Applied Soil Ecology (2.368), Biodiversity and Conservation (2.238), Phytotherapy Research (2.086), Journal of Food Composition and Analysis (2.079), Journal of Food Protection (1.937), Journal of Food Science (1.658), Fungal Biology (1.429), Plant Biosystems (1.418), Agroforestry Systems (1.378), Mycoscience (1.212), and Plant Soil and Environment (1.078). From these, the general characteristics of the research themes were examined on the journal website in order to choose journals for detailed study. Detailed characteristics of the thematic scope and IF of these journals allowed for the selection of 15 journals for scientific analysis and bibliometric research into truffles research problems (Table 1). All issues of selected journals have been analysed. Total 124 articles have been chosen (Table 1) and these show results of research for the period 1993 through 2012.

RESULTS AND DISCUSSION

The outcome of the research into the scientific journals distinguished 9 thematic categories (Table 2). The eligible number of articles in each category which set the course for studies about truffles carried out during twenty years is shown in Table 2 as well. Most articles were found in the following categories: genetic diversity, phylogeny, gene expression etc., ectomycorrhizae, biochemistry, ecology and biology.

The overview of scientific publications published in ten chosen journals indicates the great effort of mycologists to distinguish between the species (Paolocci et al., 2006; Ferdman et al., 2005; Wang et al., 2006, 2007; Wedén et al., 2004a, b). The most-studied edible truffle species are *T. melanosporum*, *T. magnatum*, *T. borchii*, *T. brumale*, *T. aestivum* and *T. indicum*.

Table 1. Journals and number of articles about truffles published in period 1993-2012.

No	Title of journal	1993	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012	Total
1	New Phytologist	-	-	-	-	1	1	-	-	1	-	1	-	-	3	3	-	1	6	1	18
2	Environmental Microbiology	-	-	-	-	-	-	-	-	1	-	-	2	-	-	-	-	-	-	3	
3	Biochemical Journal	-	-	-	-	-	-	-	-	1	-	-	1	-	-	-	-	-	-	2	
4	Fungal Diversity	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	1	-	1	3
5	BMC Genomics	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	1	
6	Applied and Environmental Microbiology	-	-	-	-	-	-	1	-	2	-	-	1	1	1	-	-	-	-	1	7
7	Fungal Genetics and Biology	-	-	-	-	1	-	-	1	1	1	2	-	1	1	-	-	-	5	-	13
8	Food Chemistry	1	-	-	1	-	-	-	-	-	-	-	-	1	2	1	-	1	1	3	11
9	Eukaryotic Cell	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	-	1	
10	FEMS Microbiology Ecology	-	-	-	-	-	1	1	-	-	-	-	-	-	-	-	-	2	1	2	7
11	Journal of Agricultural and Food Chemistry	-	-	-	-	-	-	1	1	1	-	1	3	-	-	-	-	-	1	-	8
12a	Mycological Research	-	-	-	-	-	-	-	-	-	1	2	1	1	3	1	1	-	-	-	10
12b	Fungal Biology	-	-	-	-	-	-	-	-	-	1	2	1	1	3	1	1	1	-	1	2
13	BMC Bioinformatics	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	1
14	Mycorrhiza	-	1	2	-	-	-	1	1	-	1	1	2	2	3	4	2	1	4	5	30
15	Current Genetics	-	-	-	-	-	-	-	-	-	1	-	1	1	1	-	1	-	1	7	
Total		1	1	2	1	2	2	4	3	6	5	7	11	9	16	10	3	8	18	15	124

1 -<http://onlinelibrary.wiley.com/journal/10.1111/%28ISSN%291469-8137/homepage/ProductInformation.html>2 -<http://onlinelibrary.wiley.com/journal/10.1111/%28ISSN%291462-2920/homepage/ProductInformation.html>3 - <http://www.biochemj.org/bj/toc.htm>4-<http://www.springer.com/life+sciences/ecology/journal/13225>5 - <http://www.springer.com/life+sciences/journal/12864>6-<http://aem.asm.org/>7- <http://www.journals.elsevier.com/fungal-genetics-and-biology/>8-<http://www.journals.elsevier.com/food-chemistry/>9 - <http://ec.asm.org/>10- <http://onlinelibrary.wiley.com/journal/10.1111/%28ISSN%291574-6941/homepage/ProductInformation.html>11- <http://pubs.acs.org/page/jafcau/about.html>12a - <http://www.elsevier.com/journals/mycological-research/0953-7562>12b -<http://www.journals.elsevier.com/fungal-biology>13 - <http://www.biomedcentral.com/bmcbioinformatics/>14-<http://link.springer.com/journal/572>15 - <http://www.springer.com/life+sciences/genetics+%26+genomics/journal/294>.

Source: Own elaboration.

Recently, researches focus on the role of mating types in truffle production (Linde et al., 2012; Rubini

et al., 2007, 2011b) patterns and quantification of *Tuber* mycorrhizae in truffle orchards (Baciarelli-

Falini et al., 2006; Belfiori et al., 2012; Benucci et al., 2011; 2012a, b; Garcia-Montero et al., 2007,

Table 2. Result of research on truffles based on articles from ten scientific journals presented in Table 1.

Category	Species	Articles	No. of articles
Truffle volatiles and aroma compounds	<i>T. aestivum</i>	Díaz et al., 2002	7
	<i>Tuber</i> spp.	Tarkka and Piechulla, 2007; Splivallo et al., 2011	
	<i>T. melanosporum</i> , <i>T. indicum</i> , <i>T. borchii</i>	Splivallo et al., 2007b	
	<i>T. melanosporum</i> , <i>T. aestivum</i>	Culleré et al., 2010	
	<i>T. uncinatum</i>	Splivallo et al., 2012	
Genetic diversity, phylogeny, gene expression etc.	<i>T. melanosporum</i>	Li et al., 2012	56
	<i>T. melanosporum</i> , <i>T. borchii</i>	Mello et al., 1996	
	<i>T. borchii</i>	Bertini et al., 1998; De Bellis et al., 1998; Barbieri et al., 2000, 2002; Guidi et al., 2003, 2006; Gabella et al., 2005; Grimaldi et al., 2005; Abba' et al., 2006, 2007; Montanini et al., 2006a, b, 2011; Lazzari et al., 2007; Zeppa et al., 2010	
	<i>T. magnatum</i> , <i>T. borchii</i>	Mello et al., 1999; Lacourt et al., 2002	
	<i>T. melanosporum</i> , <i>T. indicum</i> , <i>T. brumale</i>	Paolocci et al., 1999	
	<i>T. melanosporum</i> , <i>T. indicum</i>	Paolocci et al., 2000	
	<i>T. melanosporum</i>	Séjalon-Delmas et al., 2000; Agostini et al., 2001; Murat et al., 2004, 2011; Murat and Martin, 2008; Riccioni et al., 2008a; Rubini et al., 2010; Amicucci et al., 2011; Bolchi et al., 2011; Martin, 2011; Rubini et al., 2011b; Tisserant et al., 2011; Zampieri et al., 2011; Linde and Selmes, 2012; Balestrini et al., 2012	
	<i>T. indicum</i>	Mabru et al., 2001	
	<i>T. uncinatum/aestivum</i>	Mello et al., 2002; Wedén et al., 2005	
	<i>T. borchii</i> , <i>T. oligospermum</i>	Urban et al., 2004	
	<i>Terfezia pfeilii</i>	Ferdman et al., 2005	
	<i>T. magnatum</i>	Mello et al., 2005; Rubini et al., 2005; Paolocci et al., 2006	
	<i>T. magnatum</i> , <i>T. melanosporum</i> , <i>T. indicum</i> , <i>T. aestivum</i> , <i>T. mesentericum</i>	Karkouri et al., 2007	
	<i>Tuber</i> spp.	Læssøe and Hansen, 2007; Riccioni et al., 2008b	
	<i>T. mesentericum</i>	Sica et al., 2007	
	<i>T. rufum</i> , <i>T. puberulum</i>	Wang et al., 2007	
Biology	<i>T. borchii</i>	Montanini et al., 2002; Ceccaroli et al., 2003, 2007; Ambra et al., 2004; Polidori et al., 2004; Sbrana et al., 2007	11
	<i>Tuber</i> spp.	Rubini et al. 2007; Pacioni et al., 2007; Kües and Martin, 2011	
	<i>Terfezia</i> spp., <i>Tirmania</i> spp.	Kagan-Zur et al., 2008	
	<i>T. melanosporum</i>	Iotti et al., 2012	
Ectomycorrhizae	<i>T. melanosporum</i>	Mamoun and Olivier, 1995; Sisti et al., 2003; Baciarelli-Falini et al., 2006; Pérez et al., 2007; Rubini et al., 2011a; Belfiori et al., 2012; Dominguez et al., 2012; García-Barreda and Reyna, 2012	22
	<i>Terfezia terfezioides</i>	Bratek et al., 1996	
	<i>T. claveryi</i>	Morte et al., 2000	
	<i>Tuber</i> spp.	Frank, 2005; Trappe, 2005	
	<i>T. rapeodorum</i> , <i>T. rufum</i> , <i>T. puberulum</i>	Kovács and Jakucs, 2006	
	<i>T. pseudoexcavatum</i> , <i>T. indicum</i>	García-Montero et al., 2008	
	<i>T. aestivum</i>	Pruett et al., 2008; Benucci et al., 2011	
	<i>T. indicum</i>	Geng et al., 2009	
	<i>T. borchii</i>	Iotti et al., 2010	
	<i>Terfezia boudieri</i>	Turgeman et al., 2011	
	<i>T. aestivum</i> , <i>T. borchii</i>	Benucci et al., 2012a	

Table 2. Contd.

	<i>T. macrosporum</i>	Benucci et al., 2012b	
	<i>Terfezia claveryi, Picoa lefebvrei</i>	Navarro-Ródenas et al., 2012	
Invasive species threatening truffles	<i>T. melanosporum</i>	Murat et al., 2008	1
	<i>T. texense</i>	Beuchat et al., 1993	
	<i>Terfezia claveryi</i>	Harki et al., 1997; Pérez-Gilabert et al., 2001, 2005a, b, c	
	<i>T. borchii</i>	Vallorani et al., 2002; Montanini et al., 2003; Saltarelli et al., 2003	
	<i>T. aestivum</i>	Mannina et al., 2004; Fratianni et al., 2007	
Biochemistry	<i>T. melanosporum</i>	Harki et al., 2006; Ceccaroli et al., 2011	20
	<i>Tuber spp.</i>	Nazzaro et al., 2007; Tang et al., 2011, 2012; Liu et al., 2012	
	<i>T. magnatum, T. borchii, T. melanosporum, T. aestivum</i>	Saltarelli et al., 2008	
	<i>T. magnatum</i>	Barbieri et al., 2010	
	<i>T. aestivum</i>	Wedén et al., 2004a, b	
	<i>T. indicum</i>	Wang et al., 2006; Mortimer et al., 2012	
	<i>T. melanosporum</i>	García-Montero et al., 2007; Suz et al., 2010; Streiblová et al., 2012	
	<i>T. aestivum, T. excavatum, T. rufum</i>	Hilszczńska et al., 2008	
Ecology	<i>Terfezia spp.</i>	Morte et al., 2008	14
	<i>T. brumale, T. melanosporum</i>	Valverde-Asenjo et al., 2009	
	<i>Otidea sp.</i>	Smith and Healy, 2009	
	<i>T. lyonii</i>	Bonito et al., 2011	
	<i>T. claveryi</i>	Navarro-Ródenas et al., 2011	
	<i>T. magnatum</i>	Zampieri et al., 2010	
Antioxidant activities	<i>T. indicum</i>	Guo et al., 2011	1
Source for cosmetics	<i>Tuber spp.</i>	Hyde et al., 2010	1

Source: Own elaboration.

2008; Garcia-Barreda and Reyna, 2012; Geng et al., 2009; Iotti et al., 2010, Rubini et al., 2011a; Bonito et al., 2011; Kües and Martin, 2011), organisms associated with the fungi (Barbieri et al., 2000, 2002, 2010; Dominguez et al., 2012), genetic diversity within populations of *Tuber* spp. (Bertault et al., 1998, 2001; Mello et al., 2005; Riccioni et al., 2008a, b), and new molecular methods and challenges for further understanding of the *Tuber* life-cycle (Lacourt et al., 2002; Murat et al., 2011; Iotti et al., 2012). Improvement of our knowledge about biology of these fungi and the molecular mechanisms (Bertini et al., 1998; De Bellis et al., 1998; Ceccaroli et al., 2003, 2007, 2011; Montanini et al., 2003, 2006b; Gabella et al., 2005; Grimaldi et al., 2005; Guidi et al., 2006a, b; Abbà et al., 2006, 2007; Lazzari et al., 2007; Zeppa et al., 2010; Amiccuci et al., 2011; Bolchi et al., 2011; Balestrini et al., 2012) which govern the production and interaction at the level host-fungus is of great value for truffle protection in the wild as well as for owners of truffle plantation.

A few studies address environmental and genetic

factors influencing truffle volatiles and aroma compounds (Díaz et al., 2002; Tarkka and Piechulla, 2007; Spilvallo et al., 2007b, 2011, 2012; Culleré et al., 2010; Li et al., 2012). Noticeably, accumulated studies had an emphasis on the understanding of truffle volatile-host plant interaction or VOC related to both inter- and intra-organismic interactions (Tarkka and Piechulla, 2007). Study of nutritional contents revealed that truffles vary from species to species with the compositional details of investigated truffles (Beuchat et al., 1993; Saltarelli et al., 2008; Kagan-Zur et al., 2008).

In preservation of truffles for consuming purposes, researchers have attempted to employ combinations of different treatment to yield synergistic effects on extending the shelf-life of truffles with several combinations being recommended (Nazzaro et al., 2007; Saltarelli et al., 2008). With the recent progress in food technology, more innovative ways are expected to be found to preserve truffles whilst maintaining their biochemical properties. Challenges still exist regarding

improving the shelf life of truffles whilst safeguarding their all-important sensory and structural characteristics. The challenge of preservation and failure is one of the possible reasons that truffles become one of the most expensive products worldwide.

Possibilities on how to incorporate the truffles' chemical and biological properties into value-added truffle or truffles-related products need more attention since only two publications from the analysed journals have been found (Guo et al., 2011; Hyde et al., 2010). Some truffle species are still a conundrum to scientists as their chemical and biological functions have not been thoroughly investigated. It is also important how to use these findings to enhance truffle functionality, for example, using their bioactive compounds as potential therapeutic agents.

Conclusions

This paper provides an update in the recent truffle research with particular emphasis on the chemical properties (nutritional and aromatic profile) and their potential biological activities such as antioxidant, antiviral, anti-microbial, hepatoprotective, anti-mutagenic, anti-inflammatory, anti-carcinogenic, and anti-tuberculosis. In addition, some of the diversification patterns (e.g., biogeography, cultivar, and morphology) and preservation of truffles are briefly introduced. A few snapshot summary tables are also incorporated to give further detailed guidance for each section, spanning in particular the findings in the last twenty years (1993 to 2012). It is quite clear that further scientific studies need to pay greater attention on how to incorporate these biochemical and biological properties into the value-added truffles and truffle related products.

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