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RESEARCH ARTICLE

Collection and identification of Iranian wild mushrooms: towards establishment of a mushroom bio-bank

Sharareh Rezaeian[1], Hamid R. Pourianfar*[1], Javad Janpoor[1]

1. Industrial Fungi Biotechnology Research Department, Research Institute for Industrial Biotechnology, Iranian Academic Centre for Education, Culture and Research (ACECR), Mashhad-Branch, P.O.Box: 91775-1376, Mashhad-Iran

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Corresponding Author

Hamid R. Pourianfar

Abstract

Wild mushrooms provide a significant source of nutritional and medicinal bioactive compounds. They have been collected and consumed by people from many countries for thousands of years. However, there is a shortage of information in the literature regarding Iranian wild mushrooms. Thus, this mini-review tries to outline recent efforts made in order to collect, identify, and maintain wild mushrooms of Iran. This review may also encourage more research on collection, assessment, and biochemical analysis of Iranian wild mushrooms in order to establish a germplasm bank of wild mushrooms.

INTRODUCTION

Wild mushrooms provide a significant source of nutrients that can be used as food or in traditional medicine, particularly for local people in South East Asia, India, Europe, and Africa. In addition, there is an exponentially increasing interest in research into nutritional and medicinal properties of wild mushrooms, as compared to limited cultivated mushroom strains (Oyetayo, 2011). The literature shows a body of information on collection, medicinal properties and nutritional composition of wild mushrooms from many countries (Ferreira et al., 2009).

At the present, very little knowledge is available regarding wild-growing mushrooms in Iran. Iran has diverse climatic and geographical conditionsand thus could be a home to a broad range of mushroom species. Up to the present, several studies have been conducted in Iran to collect and identify wild mushrooms. However, this information has not been cited in the relevant review articles, where wild mushrooms and their medicinal properties have been reported from many countries (Ferreira et al., 2009). Therefore, in order to update the existing knowledge, this review will outline recent efforts on collection and identification of wild edible mushrooms growing wild in Iran.It may also encourage more efforts to collect a broader range of wild mushrooms from the country based on accurate identification approaches. This mini-review might also warrant further international research to study pharmacological and biochemical properties of Iranian wild mushrooms.

2. Wild mushrooms as potential source of nutraceuticals

Wild mushrooms had been used from ancient times so that the first record of mushroom utilization in Nigeria dates back to the Paleolithic period (7000 – 9000 years ago). In Eastern Asia, the knowledge on the use of edible and medicinal mushrooms had been documented and passed on from one generation to anothee, long before mushroom cultivation was industrialized (Oyetayo, 2011). Thus, it is not surprising that therapeutic properties of wild medicinal mushrooms such as *Ganoderma lucidum* have been indicated in ancient pharmacopoeia of countries like China (Wachtel-Galor et al., 2011).

Thus far, only around 40 edible mushroom species have been standardized for cultivation, while it has been estimated that the number of wild mushrooms could reach around 140,000 (Miles and Chang, 2004). Therefore, the number of the current cultivated mushrooms is still a very small proportion of the total number of wild mushrooms in the world. It has been well demonstrated thatwild mushrooms are one of the most prominent functional foods and a good source of nutraceuticals. They are excellent sources of fiber, essential oils, protein (including all the essential amino acids), vitamins, minerals, lectins, and bioactive compounds Accordingly, a number of pharmacological properties have been attributed to mushrooms, including antimicrobial, anticancer, antioxidant, antiviral, immunomodulatory, immunosuppressive, anti-allergic, anti-inflammatory, and anti-cholesterol activities (Rathee et al., 2012).

3. Germplasm collection of wild mushrooms

According to the importance of conservation of genetic diversity of wild mushrooms, there are several initiatives or programs on collection and maintenance of mushroom germplasms in the world. The main objectives of these programs have been to study diversity, acquisition, preservation, characterization and distribution of novel germplasms of wild mushrooms. The Agaricus Resource Program (ARP) in the US (founded in 1988) is one of the most important germplasm banksof *Agaricus* spp. mushrooms (Kerrigan et al., 1991). In this collection, hundreds of wild specimens are maintained, collected from Europe, the Mediterranean region, and North America. Another American example has been reported from the Black Hills of South Dakota and Bear Lodge Mountains of Wyoming, containing 260 macro-fungi species collected during 1998–2002 (Gabel et al., 2004). Mushroom germplasm banks have also been reported from some other countries, such as Turkey (Türkoğlu, 2008). In this country, totally 125 taxa belonging to *Ascomycetes* and *Basidiomycetes* were identified.

In addition, global efforts have also been made to compile macro-fungi (including wild mushrooms) related to different regions of the world. In a study, totally 21,679 names of macro-fungi were compiled; half of which were from North America and Western Europe. In addition, approximately 35,000 macro-fungal species were estimated to be ‘‘unknown’’ by the contributing authors, giving an estimated total of 56,679 macro-fungi (Mueller et al., 2007).

In addition to the germplasm banks of wild mushrooms, utilization of bio-diversity of wild mushrooms in pharmacological research has also been taken into consideration. In this regard, medicinal properties of wild mushrooms have been studied in many countries, including India, Taiwan, Turkey, Spain, China, Korea, Portugal, Finland, and Brazil (reviewed by Ferreira et al., 2009). In addition, more recent research articles have reported gathering information on collection and medicinal uses of wild mushrooms from many other countries or regions, including Nigeria (Oyetayo, 2011), Ghana (Obodai et al., 2014), Kenya (Wandati et al., 2013), Tanzania (Hussein et al., 2015), Australia (Zeng et al., 2012), the middle-east (Hamza et al., 2013), Pakistan (Badshah et al., 2015), Europe (Kalač et al., 2009), and Mexico (Quiñónez-Martínez et al., 2014).

4. Collection of wild mushrooms in Iran

As opposed to a great history of medicinal use of herbs and plants in Iran, wild mushrooms do not have a significant place in the folk medicine. Despite significant advances made in mushroom cultivation technology, there is apparently a low public knowledge on medicinaland nutritional properties of wild mushrooms.

It has been estimated that there might be around 3500 fungi species in Iran (Rahnama et al., 2015). During the past decade, efforts have been made to collect and identify wild-growing mushrooms. However, the most of these studies have been limited to the Northern Iran (inprovinces that lie along the Caspian Sea), whilst various geographical and climatic zones could be found in the country that has a total land area of 1,648,195 square kilometers. The following is a compilation of research studies conducted during the past recent years in Iran to identify and collect wild mushroom species. Small-scale studies that have reported collection of a limited sample of wild mushroomsto assess their medicinal properties have not been included.

It is well-known that the Arasbaran forest (located in Northwestern Iran) has a rich flora of macro-fungi. Collection of macro-fungi from this region has been begun since 2003. In a study, collection and identification of several species of the genus *Cortinarius* were reported. The genus *Cortinarius* is the largest group of Agarics (Asef et al., 2007).

In 2007, collection and identification of Iranian species of medicinally important *Ganoderma* spp. was reported (Moradali et al., 2007). The identification method was based on micro- and macro-morphology and host relationships. Seven species of *Ganoderma* were found and identified; including three non-laccate species *G. applanatum*, *G. adspersum*, and *G. colossus* and the four laccate species *G. lucidum*, *G. resinaceum*,*G. tsugae*, and *G. manoutchehrii* (Moradali et al., 2007).

Olfati et al. (2009) reported collection of several wild mushroom species from Guilan province (one of the Iran's Northern provinces), including *Morchella* spp., *Cantharellus*cibarius, *Pleurotus ostreatus*, *Amanita caesarea*, and *Russulapaludosa*. Morphological markers were utilized to identify mushroom species.

Diversity and distribution of macro-fungi associated with the Beech Forests of Northern Iran (in Mazandaran province) was reported (Borhani et al., 2010). In this study, 100 taxa were identified and collected, of which 41 were new to Mazandaran, 24 were new Iranian fungus-beech association, and 11 had not previously been reported from Iran (Borhani et al., 2010).

A study reported collection and identification of mushrooms belonging to Agaricales, Boletales, Cantharellales, Geastrales, and Russulales orders, from Northern Iran (Gorgan, ShastKalate) during 2010-2011. Identification of the collected macro-fungi was carried out according to the macroscopic and microscopic characteristics of the specimens, as well as their characteristic responses to some chemical reagents (Karim et al., 2013). A more recent study on also reported collection of several *Ganoderma* spp. from Northern Iran (Keypour et al., 2014).

The majority of the afore-mentioned studies have been based on morphological or microscopic characteristics. Our research team, for the first time in Iran, employed Internal Transcribed Spacer (ITS) sequencing analysis to accurately identify and authenticate wild mushrooms. We collected over 100 samples of wild mushrooms belonging to various genera from east-northern Iran (Khorasan Razavi Province) during the year 2012, including *Agaricus* spp., *Pleurotus* spp., *Lentinus* spp., *Flammulina* spp., and other mushroom genera. The collected mushroom specimens were then authenticated by ITS sequencing analysis (Tajalli et al., 2015). The mushroom samples are currently stored in a -80 freezer in glycerol and are also *periodically*cultured in order to maintain their viability. In addition, *Agaricus* spp. and *Pleurotus* spp. have been standardized for cultivation in synthetic compost (Rezaeian et al., 2015; Ghanbari et al., 2015). Some of the selected wild *A. bisporus* were also further subjected to mycelia growth characterization (Masoumi et al., 2015). Therefore, it was possible to maintain mycelia and fruiting bodies of the collected wild specimens and facilitate reproducibility of further pharmacological bioassays.

5. Progress in identification of wild mushrooms

Up to the present, a number of approaches have been used by researchers to identify wild mushrooms. Morphological and non-PCR molecular techniques (such as isozymes and *restriction fragment length polymorphism(*RFLP))were evolved to PCR-based methods such as r*andom amplified polymorphic DNA(*RAPD),a*mplified*fragment length polymorphisms (AFLP), simple sequence repeat (SSR), and inter-simple sequence repeats (ISSR). These PCR-based markers were further replaced with DNA barcodes. A DNA barcode is defined as 500- to 800-bp sequences to detect species of all eukaryotic kingdoms using primers that are applicable for a broad taxonomic group (Schoch et al., 2012).

Several DNA barcodes have been suggested for mushroom identification. Currently, ITS markers are frequently used by researchers for identification of wild mushrooms (Das et al; 2013). However, there are several reports showing the potential of other barcodes for mushroom identification. For example, the mitochondrial cytochrome oxidase I (COXI) was shown to be more effective than ITS in *Ascomycota* and *Basidiomycota*, particularly in *A. bisporus* (Vialle et al., 2009; Barroso et al., 2011). Our research team has demonstrated that ITS and *intergenic spacer* (IGS) regions were able to distinguish differences among species of Iranian wild *Agaricus*. On the contrary, ISSR markers were powerful enough for detection of polymorphism among closely related genotypes of within species of *Agaricus* (Malekzadeh et al., 2014). Another study reported that identification of *Pleurotus* spp. (among and within species) was efficiently carried out by IGS1 and ITS sequences (Avin et al., 2014).

7. Conclusions

Several studies have been conducted to collect Iranian wild mushrooms and evaluate their medicinal properties. However, many of these studies have not been based on well-defined objectives towards collection and maintenance of a broad range of wild mushrooms that represent the most of climatic zones of the country. Furthermore, the majority of collected mushrooms are not accessible to researchers, and their cultivation in synthetic compostshas not also beenstandardized. As a result, still there is no bank of germplasms of wild mushrooms in the country. Establishment of a bio-bank of well-authenticated wild mushrooms could guarantee conservation of genetic diversity of wildmushrooms in the country. It also would provide a reliable source of stable mother cultures for utilization by mushroom growers or researchers. These mushroom specimens should represent the most of climates and geographical zones of the country. Further studies are also warranted to investigate biochemical and nutritional composition of Iranian wild mushrooms.

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Declaration of interest
There is no conflict of interest. The authors alone are responsible for the content and writing of the paper.

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