ISSN 2953-741X
Funga Latina **1** (2023) V1A3 https://doi.org/10.5281/zenodo.10214914

Improving agarics usability and sampling in conservation assessments

Gonzalo M. Romano[1], Bernando E. Lechner[2,3] and Alina G. Greslebin[1,4]

[1] Fundación Hongos de Argentina para la Sustentabilidad. Molinari 1657, Esquel, Chubut, Argentina.
[2] Universidad de Buenos Aires, Facultad de Ciencias Exactas and Naturales, Departamento de Biodiversidad y Biología Experimental (DBBE), Buenos Aires, Argentina. Intendente Guiraldes 2160, Ciudad Universitaria, Pabellón II, Piso 4, Laboratorio 7, CP1428, Ciudad Autónoma de Buenos Aires, Argentina.
[3] CONICET, Instituto de Micología y Botánica (InMiBo), Buenos Aires, Argentina. Intendente Guiraldes 2160, Ciudad Universitaria, Pabellón II, Piso 4, Laboratorio 7, CP1428, Ciudad Autónoma de Buenos Aires, Argentina.
[4] Departamento de Biología General, Facultad de Ciencias Naturales, Universidad Nacional de la Patagonia San Juan Bosco. Ruta Nacional 259, Km 16.4, CP 9200, Esquel, Chubut, Argentina.

E-mail: gonzaromano@hongos.ar

Received: 17 November 2023
Accepted for publication: 27 November 2023
Published: 8 December 2023
Editor: Carlos Rojas

Abstract: Species are not equally detectable, and this should be considered at the moment of choosing ecological indicators and considering sampling efforts. Indices that permit ranking gilled species according to their abundance, permanency and basidiome features were constructed. The same indices were used to evaluate sampling effort and efficiency: more than two hours of continuous work negatively affects the capability of finding less detectable species. Ranking species is a practical solution to organize abundance datasets and can be easily applied to find patterns of species relevance and detectability to better understand our findings and even to ensure optimum field work efficiency.

Keywords: detectability, ecology, index, sampling effort.

Introduction

Research helps to understand ecosystem complexity and factors involved in their functions and processes, by developing more and better environmental tools that can be useful in conservation assessments. Comparison of large species lists is not always appropriate to assess the conservation status of any given area. Species can be rapidly listed and ordered alphabetically or according to their abundance, but this ordination can yield partial results, as species are not equally detectable (Halme and Kotiaho 2012). This variation in detectability, defined by Lõhmus (2009) as the probability of recording the presence of a species by human observers, takes a serious relevance in organisms that can only be recorded in some seasons and are subjected to specific climate conditions, like fungal species. Due to such sensitivity to disturbance in general, fungi are considered indicators of the conservation values of forests (Abrego et al. 2018).

Fruit body surveys have been the most popular method for recording the occurrence of fungal species throughout the world. Despite the biases of this method due to the characteristics of each species,

it is one of the most used protocols in conservation studies because more accurate methods (i.e., molecular techniques) are more expensive and laborious. Given the frequent use of species presence and absence to indicate conservation status of different ecosystems, it is necessary to find criteria that help determine if species observations are equally detectable. Godeas et al. (1993) proposed an index to assign relative importance to macromycetes that allow arranging species according to their frequency and aptitude to colonize different substrates in *Nothofagus* forests of Patagonia. The authors concluded that most species have low prominence, whether it is due to their low frequency, their incapacity to colonize various substrates or their sporadic fruiting. On the contrary, species with high prominence are relatively few and rarely shared between different *Nothofagus* forests.

Lõhmus (2009) also designed an index, but with a different objective: to extract the main factors of detectability for polypore fungi, assessing factors like basidiomes longevity, size, bright coloring, as well as typical microhabitat and field identifiability. His innovative approach enabled to distinguish in a formal manner the most promising indicator species for conservation management.

In this manuscript, we propose a method to quantify features of gilled basidiomes and to construct indices that are suitable for arranging species. By applying this method, quantitative assessments of species fruiting can be conducted as well as critical evaluations on the sampling and selection of indicator species based on basidiomes.

Materials and methods

Database used

The database from Romano et al. (2020) was used for the purpose of this article. Such resource included 2380 observations of 158 gilled species sampled in autumn and spring between 2012 and 2014 from three different *Nothofagus pumilio* monospecific forests in Patagonia, Argentina. To assess abundance and diversity, basidiome production was recorded in different units randomly selected in each sampling.

For sampling effort analysis, samples collected were categorized according to when they were recorded during field work. Thirteen dates and a maximum of eleven hours of fieldwork were taken into consideration. Each date was considered a replicate, to calculate proper standard deviation.

Relative abundance index

The relative abundance index (*Ri*) measures the abundance of basidiomes of each species in relation to the other species in the community. It is constructed with the number of samples per species divided by the total samples found at the same time and place:

$$Ri = \left(\frac{ni}{N}\right) * 100$$

Where *ni* = number of samples of species "*i*" in any given unit, and *N* = sum of all samples found in the unit.

Permanency index

The permanency index (*Pi*) allows ranking species according to their consistent and repeated fruiting throughout surveys:

$$Pi = si/S$$

Where *si* = number of samplings in which species "*i*" was found and *S* = total number of samplings.

Detectability index

The detectability index (*Di*) was designed based on Lõhmus (2009). It measures how detectable gilled basidiomes are, considering their inheriting features:

$$Di = Ti * (1 + BCi + VRi + Hi)$$

Where *Ti* = maximum size of the basidiome (millimeters or inches), and a series of binary factors: *BCi* = presence (1) or absence (0) of bright colours, *VRi* = presence (1) or absence (0) of veil remnants (membranaceus or fibrilous), and *Hi* = gregarious (1) or solitary (0) habit. All the latter are summed to 1, as a species without any of those features but with a large size can be easily detected.

Indicator construction

Two indicators were designed based on the indices constructed. Indicator *A* measures the relevance of each species in any given sampling, as it consists of the sum of both the relative abundance (*Ri*) and permanency (*Pi*) indices:

$$A = Ri + Pi$$

If we multiply index *A* by the detectability index, the outcome can quantify the relevance of a species according to how easily detectable it is in the field. Herein, this metric is indicator *B*:

$$B = A * Di$$

All indexes were calculated for each species found, and later ranked according to both indicators *A* and *B*. In order to simplify the interpretation, all indicators were relativized to study a 50% threshold ($B \geq 5$).

Results

Species ranking

A total of 158 gilled species were found in the established stands. According to indicator *B*, only two species showed a value higher than the 50% threshold (Table 1). To illustrate the input differences in the features of basidiomes, species with different scores of index *Di* and indicators *A* and *B* are shown in Figure 1.

Table 1. Indices and indicator values obtained for all species registered in the analyzed database.

Species	**Nutrition**	***Ri***	***Pi***	***Di***	***A***	***B***
Cortinarius magellanicus	ECM	4	3	0.76	7	5.33
Russula nothofaginea	ECM	4	1	1.00	5	5.00
Austropaxillus boletinoides	ECM	5	5	0.48	10	4.76
Pholiota baeosperma	SAP	5	5	0.48	10	4.76
Mycena galericulata	SAP	5	5	0.46	10	4.64
Pluteus spegazzinianus	SAP	5	5	0.43	10	4.29
Descolea antarctica	SAP	4	4	0.52	8	4.19
Austropaxillus statuum	ECM	5	4	0.46	9	4.18
Russula nothofaginea var. *carminea*	ECM	3	1	1.00	4	4.00
Cortinarius melleus	ECM	5	4	0.39	9	3.54
Cortinarius collariatus	ECM	2	2	0.86	4	3.43
Cortinarius parazureus	ECM	3	3	0.57	6	3.43
Hypholoma frowardii	ECM	3	2	0.67	5	3.33
Rhodocollybia butyracea	SAP	2	2	0.79	4	3.14
Crepidotus fulvifibrillosus var. *meristocystis*	ECM	4	4	0.38	8	3.05
Inocybe geophyllomorpha	ECM	5	5	0.29	10	2.86
Russula fuegiana	ECM	3	3	0.43	6	2.57
Cortinarius cf. *myxoduracinus*	ECM	3	2	0.50	5	2.50
Gymnopus fuegianus	ECM	5	5	0.24	10	2.38
Armillaria montagnei	SAP	2	1	0.76	3	2.29
Cortinarius albobrunneus	SAP	4	4	0.29	8	2.29
Cortinarius aff. *aganochrous*	SAP	5	4	0.24	9	2.14
Cortinarius albocanus	ECM	5	4	0.24	9	2.14
Cortinarius saccharatus	ECM	3	3	0.33	6	2.00
Cortinarius holojanthinus	ECM	3	2	0.39	5	1.96
Cortinarius elaphinus	ECM	4	4	0.24	8	1.90
Cortinarius albocinctus	ECM	5	4	0.19	9	1.71
Cortinarius austroduracinus	ECM	4	4	0.21	8	1.71
Cortinarius caelicolor	ECM	3	3	0.29	6	1.71
Cortinarius roseopurpurascens	ECM	1	1	0.86	2	1.71
Galerina gamundiae	SAP	5	4	0.19	9	1.71
Cortinarius bulboso-mustellinus	ECM	3	2	0.33	5	1.67
Cortinarius ocellatus	ECM	4	3	0.21	7	1.50
Entoloma patagonicum	ECM	4	3	0.21	7	1.50
Mycena pura	SAP	4	3	0.21	7	1.50
Clitocybe pleurotus	SAP	5	3	0.18	8	1.43
Cortinarius cretaceus	ECM	5	3	0.18	8	1.43
Omphalina subhepatica	SAP	3	2	0.29	5	1.43
Cortinarius darwinii	ECM	4	3	0.19	7	1.33
Inocybe neuquenensis	SAP	4	3	0.19	7	1.33
Cortinarius sp3	ECM	3	1	0.32	4	1.29
Cortinarius illitus	ECM	3	3	0.21	6	1.29
Cortinarius leucoloma	ECM	5	4	0.14	9	1.29
Mycena atroincrustata	SAP	5	4	0.14	9	1.29
Hydropus dusenii	SAP	4	3	0.17	7	1.17
Cortinarius fulvoconicus	ECM	3	3	0.19	6	1.14
Cortinarius terebripes	ECM	3	1	0.29	4	1.14

Gymnopus fuscopurpureus	SAP	5	4	0.12	9	1.07
Cortinarius nothoanomalus	ECM	4	3	0.14	7	1.00
Collybia platensis	SAP	5	5	0.10	10	0.95
Cortinarius napivolvatus	ECM	3	1	0.24	4	0.95
Cortinarius succineus	ECM	5	4	0.10	9	0.86
Cortinarius xylocinnamomeus var. *xylocinnamomeus*	ECM	2	2	0.21	4	0.86
Crepidotus applanatus	ECM	4	3	0.12	7	0.83
Cortinarius concolor	ECM	3	3	0.13	6	0.79
Cortinarius mustellinus	ECM	1	1	0.38	2	0.76
Cortinarius scabrosporus	ECM	4	4	0.10	8	0.76
Inocybe cerasphora	ECM	2	2	0.19	4	0.76
Pholiota privigna	SAP	3	1	0.19	4	0.76
Cortinarius permagnificus	ECM	1	1	0.36	2	0.71
Cortinarius inocybiphyllus	ECM	4	2	0.12	6	0.71
Cortinarius xanthocholus	ECM	3	3	0.12	6	0.71
Melanoleuca lapataiae	SAP	2	1	0.24	3	0.71
Psathyrella sp1	SAP	2	1	0.24	3	0.71
Inocybe fuscocinnamomea	ECM	4	3	0.10	7	0.67
Pholiota cf. *aurantioalbida*	SAP	1	1	0.32	2	0.64
Cortinarius simplex	ECM	5	4	0.07	9	0.64
Galerina hypnorum	SAP	5	4	0.07	9	0.64
Galerina riparia	SAP	2	1	0.21	3	0.64
Schizophyllum commune	ECM	3	3	0.11	6	0.64
Cortinarius pseudotriumphans	ECM	1	1	0.31	2	0.62
Cortinarius coleopus	ECM	1	1	0.29	2	0.57
Cortinarius obesus	ECM	2	1	0.19	3	0.57
Cortinarius occentus	ECM	2	1	0.19	3	0.57
Cortinarius sp1	ECM	1	1	0.29	2	0.57
Cortinarius tricholomoides	ECM	1	1	0.29	2	0.57
Cortinarius variegatulus	ECM	3	1	0.14	4	0.57
Cuphophyllus adonis	SAP	2	1	0.19	3	0.57
Inocybe bridgesiana	ECM	4	2	0.10	6	0.57
Lepiota subgracilis	ECM	4	2	0.10	6	0.57
Mycena helminthobasis	SAP	2	2	0.14	4	0.57
Porpoloma sejunctum	ECM	1	1	0.29	2	0.57
Protostropharia semiglobata	SAP	1	1	0.29	2	0.57
Cortinarius gayi	ECM	3	2	0.11	5	0.54
Cortinarius interlectus	ECM	2	1	0.17	3	0.50
Cortinarius myxoclaricolor	ECM	2	1	0.17	3	0.50
Mycena falsidica	SAP	4	3	0.07	7	0.50
Entoloma mesites	SAP	3	2	0.10	5	0.48
Psathyrella falklandica	SAP	4	4	0.06	8	0.48
Clitocybe suaveolens	SAP	5	4	0.05	9	0.43
Cortinarius austrolimonius var. *ochrovelatus*	ECM	2	1	0.14	3	0.43
Cortinarius egenus	ECM	4	5	0.05	9	0.43
Crepidotus brunswickianus	ECM	5	3	0.05	8	0.38
Melanoleuca cf. *melaleuca*	SAP	1	1	0.19	2	0.38
Galerina sp1	SAP	2	1	0.12	3	0.36
Melanoleuca sp1	SAP	2	1	0.12	3	0.36
Pholiota sp1	SAP	2	1	0.12	3	0.36

Cortinarius dissimulans	ECM	4	3	0.05	7	0.33
Cortinarius fuegianus	ECM	1	1	0.17	2	0.33
Cortinarius phaeocephalus	ECM	1	1	0.17	2	0.33
Pholiota spumosa var. *crassitunica*	SAP	1	1	0.17	2	0.33
Mycena patagonica	SAP	4	4	0.04	8	0.30
Bolbitius reticulates	SAP	3	2	0.06	5	0.29
Cortinarius austrolimonius	ECM	1	1	0.14	2	0.29
Cortinarius rubrobasalis	ECM	1	1	0.14	2	0.29
Mycena haematopus	SAP	3	1	0.07	4	0.29
Psilocybe subcoprophila	SAP	3	2	0.05	5	0.24
Marasmius ushuaiensis	SAP	4	4	0.03	8	0.23
Clitocybe patagonica	SAP	5	4	0.02	9	0.21
Cortinarius maulensis	ECM	2	1	0.07	3	0.21
Entoloma cucurbita	SAP	2	1	0.07	3	0.21
Entoloma papillatum	ECM	2	1	0.07	3	0.21
Hypogaea brunnea	ECM	2	1	0.07	3	0.21
Cortinarius exilis	ECM	3	1	0.05	4	0.19
Leucopaxillus sp*1*	SAP	3	1	0.05	4	0.19
Resupinatus applicatus	SAP	3	1	0.05	4	0.19
Mycena epipterygia	SAP	4	1	0.04	5	0.18
Marasmiellus minutus	SAP	3	1	0.04	4	0.15
*Mycenella margaritis*p*ora*	SAP	5	3	0.02	8	0.15
Clitocybe subhygrophanoides	SAP	5	1	0.02	6	0.14
Cortinarius lignyotus	ECM	1	1	0.07	2	0.14
Laccaria tetraspora	SAP	1	1	0.07	2	0.14
Phaeomarasmius ciliatus	SAP	1	1	0.07	2	0.14
Simocybe curvipes	SAP	2	1	0.05	3	0.14
Mycena sp3	SAP	1	1	0.06	2	0.13
Gymnopus aff *fuscopurpureus*	SAP	1	1	0.06	2	0.12
Mycena sp1	SAP	4	1	0.02	5	0.12
Resupinatus chilensis	SAP	5	3	0.01	8	0.11
Galerina aff. *tibiicystis*	ECM	2	1	0.04	3	0.11
Galerina sp4	SAP	2	1	0.04	3	0.11
Arrhenia griseopallida	SAP	2	1	0.03	3	0.10
Marasmius sp1	SAP	4	1	0.02	5	0.10
Cortinarius surreptus	ECM	1	1	0.05	2	0.10
Galerina sp2	SAP	3	1	0.02	4	0.10
Kuehneromyces cystidiosus	ECM	1	1	0.05	2	0.10
Marasmius sp2	SAP	1	1	0.05	2	0.10
Mycena dendrocystis	SAP	2	2	0.02	4	0.10
Scytinotus longinquus	ECM	1	1	0.05	2	0.10
Simocybe cf. *curvipes*	SAP	1	1	0.05	2	0.10
Mycena sp4	SAP	3	2	0.02	5	0.08
Marasmius hemimycena	SAP	5	3	0.01	8	0.08
Psilocybe coprophila	SAP	1	1	0.04	2	0.08
Coprinellus truncorum	SAP	4	1	0.01	5	0.07
Cortinarius squamiger	ECM	1	1	0.04	2	0.07
Galerina sp3	SAP	2	1	0.02	3	0.07
Marasmius aporpus	SAP	2	1	0.02	3	0.07
Phaeomarasmius limulatellus	SAP	2	1	0.02	3	0.07
Psathyrella fuegiana	SAP	1	1	0.04	2	0.07
Cortinarius hebes	ECM	1	1	0.03	2	0.07

Marasmius aff. *ushuaiensis*	SAP	1	1	0.03	2	0.06
Mycena desfontainea	SAP	5	3	0.01	8	0.06
Cortinarius sp2	ECM	1	1	0.02	2	0.05
Crepidotus sp1	SAP	1	1	0.02	2	0.05
Hemimycena patagonica	SAP	1	1	0.02	2	0.04
Mycena aff. *dendrocystis*	SAP	1	1	0.02	2	0.04
Clitocybe subleptoloma	SAP	1	1	0.02	2	0.03
Cortinarius aff. *scolecinus*	SAP	1	1	0.01	2	0.02
Mycena sp2	SAP	1	1	0.01	2	0.02

Sampling effort

Index *Di* was also used for testing sampling effort in the field. The sample with the minimum value of index *Di* (among all samples taken at the same time and day) was annotated for each hour and day and plotted by hours in the field. Figure 2 shows the minimum value of species detectability that was recorded according to the accumulated sampling hours in the field. Records of species with the lowest detectability were found during the first two hours. After this period, the minimum detectability score of species found was higher.

Discussion

Godeas et al. (1993) developed an ecological index of species importance. This index was used to design two indicators of our own that not only assessed the relevance of the species found but also permitted to order them quantitatively according to research interests.

Indicator *B* was constructed by multiplying indicator *A* and the detectability index *Di*, because it is rather common that species that are difficult to detect during samplings are considered to have a low abundance (Lõhmus 2009). This can also be useful to determine which species are truly detectable and practical to be used as ecological indicators. *Pholiota baeosperma*, for example, had one of the highest values of detectability according to both indicators, and has been found to be a potential good indicator of unmanaged forests of *Nothofagus pumilio* (Romano et al. 2020). Halme et al. (2009) expressed the importance of using detectable species as indicators of any given group of interest so that they can be easily targeted for practical surveys and monitoring, and *P. baeosperma* meets this requirement.

Halme and Kotiaho (2012) used a Detectability index developed by Garrard et al. (2008) and Kéry et al. (2006), which provides essential information on number of surveys required to achieve a certain value of species detectability, but it does not provide information on species specific detectability based on its features.

Sampling effort is a measure of how much effort is needed to do sampling and, based on previous datasets, it can be used to set a standard protocol of the length of sampling sessions (Abrego et al., 2016). Overwork in sampling is more common than it is believed, especially in small research groups or if organisms studied have restricted niches (spatial and/or temporal) or are difficult to find (Lõhmus & Runnel, 2018). However, there is a limit associated with how much work can be done without losing quality data. To test our detectability index *Di* for agarics, we studied if samples were taken following any specific pattern of basidiome features. Records of species with the lowest detectability were found during the first two hours. After this period, the minimum detectability score of species found was more than

three times higher. This suggested that samples found after two hours of work were easier to find than those collected during the first hours, whether it was because of their size, bright coloring, veil remnants or habit. Moreover, rest always took place between the third and fifth hour of labor, which was reflected in a reduction of the minimum *Di* index, as observed in samples collected between the fourth and sixth hour. This means that fatigue negatively affected the capability of researchers to find basidiomes of agaric fungi, with the subsequent implications of experimental bias (Lõhmus 2009).

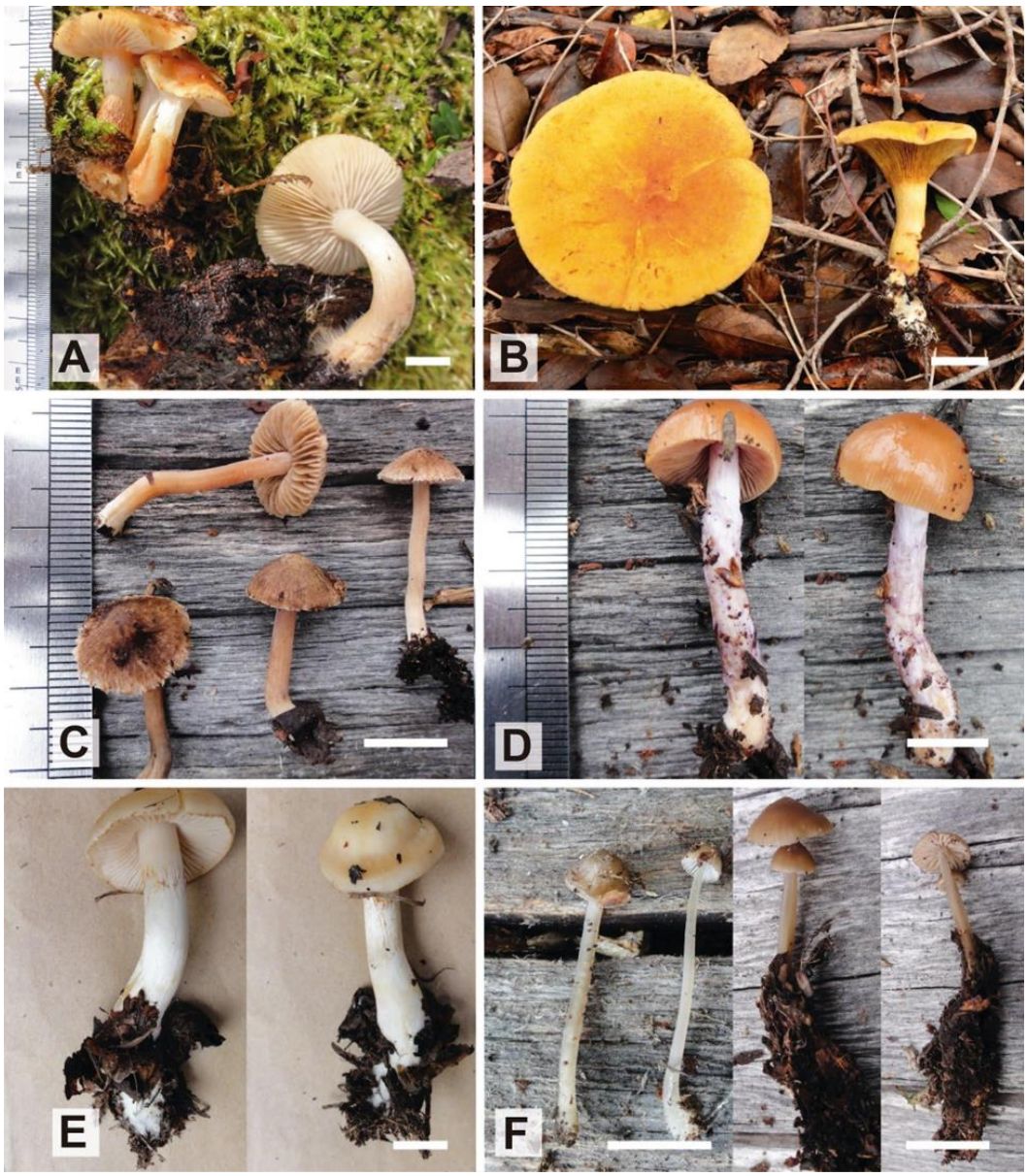

Figure 1. Species with different values of the detectability index (Di) as well as indicators A and B. A: *Pholiota baeosperma* (Di = 0.48, A = 10, B = 4.76). B: *Austropaxillus boletinoides* (Di = 0.48, A = 10, B = 4.78). C: *Inocybe geophyllomorpha* (Di = 0.29, A = 10, B = 2.86). D: *Cortinarius aff. aganochrous* (Di = 0.24, A = 9, B = 2.14). E: *Cortinarius simplex* (Di = 0.07, A = 9, B = 0. 64). F: *Mycena desfontainea* (Di = 0.01, A = 8, B = 0.06). Scale bar: 1 cm.

Indicators *A* and *B* also revealed that many relevant species are not as easily detected as other ones that are considered less important. This was the case of *Cortinarius* aff. *aganochrous, Cortinarius dissimulans, Cortinarius leucoloma, Cortinarius simplex, Galerina hypnorum, Inocybe fuscocinnamomea, Inocybe geophyllomorpha* and *Mycena desfontainiae*.

Our results indicated that each sampling session should not exceed more than two hours of continuous work. A minimum of time should be used to rest every two hours: it is important not only to ensure the maximum sampling efficiency but also to guarantee the integrity of researchers and volunteers involved in the team.

Ranking species is a practical solution to organize abundance datasets and can be easily applied to find patterns of species relevance and detectability to better understand our findings and even to ensure optimum field work efficiency during sampling.

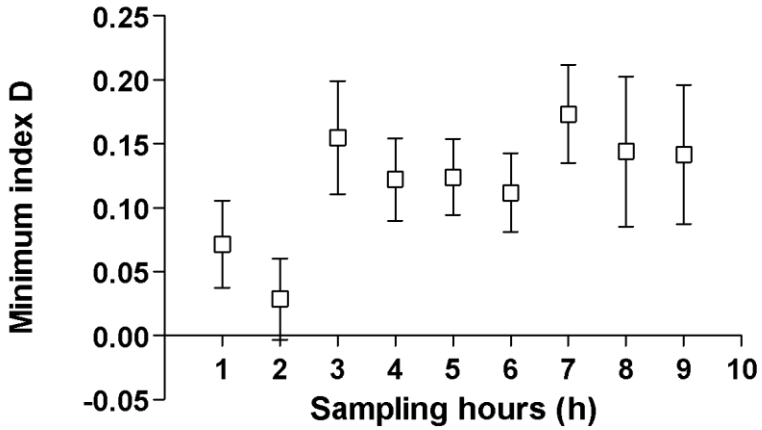

Figure 2. Minimum detectability index (D) value according to sampling hours.

Acknowledgements

This work was funded by Consejo Nacional de Investigaciones Científicas y Técnicas and Agencia Nacional de Promoción Científica y Técnica (FONCyT, PICT 1229).

References

Abrego N, Halme P, Purhonen J, Ovaskainen O. 2016. Fruit body based inventories in wood-inhabiting fungi: Should we replicate in space or time? Fungal Ecol. 20: 225-232.

Abrego N, Norros V, Halme P, Somervuo P, Ali-Kovero H, Ovaskainen O. 2018. Give me a sample of air and I will tell which species are found from your region: Molecular identification of fungi from airborne spore samples. Mol Ecol Resour. 18(3): 511-524.

Garrard GE, Bekessy SA, McCarthy MA, Wintle A. 2008. When have we looked hard enough? A novel method for setting minimum survey effort protocols for flora surveys. Austral Ecol. 33(8): 986-998.

Godeas AM, Arambarri AM, Gamundí IJ. 1993. Micosociología en los bosques de *Nothofagus* de Tierra del Fuego II. Importancia relativa de las distintas especies de Macromicetes. RACSAM Rev R Acad A. 45: 303-311.

Halme P, Kotiaho JS, Ylisirniö AL, Hottola J, Junninen K, Kouki J, Lindgren M, Mönkkönen M, Penttilä R, Renvall P, Siitonen J, Similä M. 2009. Perennial polypores as indicators of annual and red-listed polypores. Ecol Indic. 9: 256-266.

Halme P, Kotiaho JS. 2012. The importance of timing and number of surveys in fungal biodiversity research. Biodivers Conserv. 21: 205-219.

Kéry M, Spillmann JH, Truong C, Holderegger R. 2006. How biased are estimates of extinction probability in revisitation studies? J Ecol. 94(5): 980-986.

Lõhmus A, Runnel K. 2018. Assigning indicator taxa based on assemblage patterns: Beware of the effort and the objective! Biol Conserv. 219:147-152.

Lõhmus A. 2009. Factors of species-specific detectability in conservation assessments of poorly studied taxa: The case of polypore fungi. Biol Conserv. 142: 2792-2796.

Romano GM, Greslebin AG, Lechner BE. 2017. Hongos agaricoides de los bosques de *Nothofagus pumilio* (Chubut, Argentina): Clave y listado de especies. Rev Mus Argent Cienc Nat. n.s. 19(1): 39-69.

Romano GM, Lechner BE, Greslebin AG. 2020. Basidiome production of ectomycorrhizal and saprophytic agaricoid fungi respond differently to forest management. Can J For Res. 50(2): 102-112.