

## 12,500+ and counting: biodiversity of the Brazilian Pampa

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## Abstract

Knowledge on biodiversity is fundamental for conservation strategies. The Brazilian Pampa region, located in subtropical southern Brazil, is neglected in terms of conservation, and knowledge of its biodiversity is fragmented. We aim to answer the question: how many, and which, species occur in the Brazilian Pampa? In a collaborative effort, we built species lists for plants, animals, bacteria, and fungi that occur in the Brazilian Pampa. We included information on distribution patterns, main habitat types, and conservation status. Our study resulted in referenced lists totaling 12,503 species (12,854 taxa, when considering infraspecific taxonomic categories [or units]). Vascular plants amount to 3,642 species (including 165 Pteridophytes), while algae have 2,046 species (2,378 taxa) and bryophytes 316 species (318 taxa). Fungi (incl. lichenized fungi) contains 1,141 species (1,144 taxa). Animals total 5,358 species (5,372 taxa). Among the latter, vertebrates comprise 1,136 species, while invertebrates are represented by 4,222 species. Our data indicate that, according to current knowledge, the Pampa holds approximately 9% of the Brazilian biodiversity in an area of little more than 2% of Brazil's total land. The proportion of species restricted to the Brazilian Pampa is low (with few groups as exceptions), as it is part of a larger grassland ecoregion and in a transitional climatic setting. Our study yielded considerably higher species numbers than previously known for many species groups; for some, it provides the first published compilation. Further efforts are needed to increase knowledge in the Pampa and other regions of Brazil. Considering the strategic importance of biodiversity and its conservation, appropriate government policies are needed to fund studies on biodiversity, create accessible and constantly updated biodiversity databases, and consider biodiversity in school curricula and other outreach activities.

## Highlights

- We present the first referenced list of species for the Brazilian Pampa region.
- The Brazilian Pampa has at least 12,503 species of plants, animals, fungi and bacteria.
- Species in the Brazilian Pampa predominantly inhabit grasslands or wetlands/aquatic ecosystems.
- The Brazilian Pampa holds, in just over 2% of Brazil's area, about 9% of its total biodiversity.
- Biogeographic surveys, such as the one presented here, are needed in other regions of Brazil and the world.

**Keywords:** animal diversity, bacteria, biodiversity assessment, biogeographic survey, Brazilian Pampa, Campos, conservation, fungi, grassland, open ecosystem, plant diversity.

## Introduction

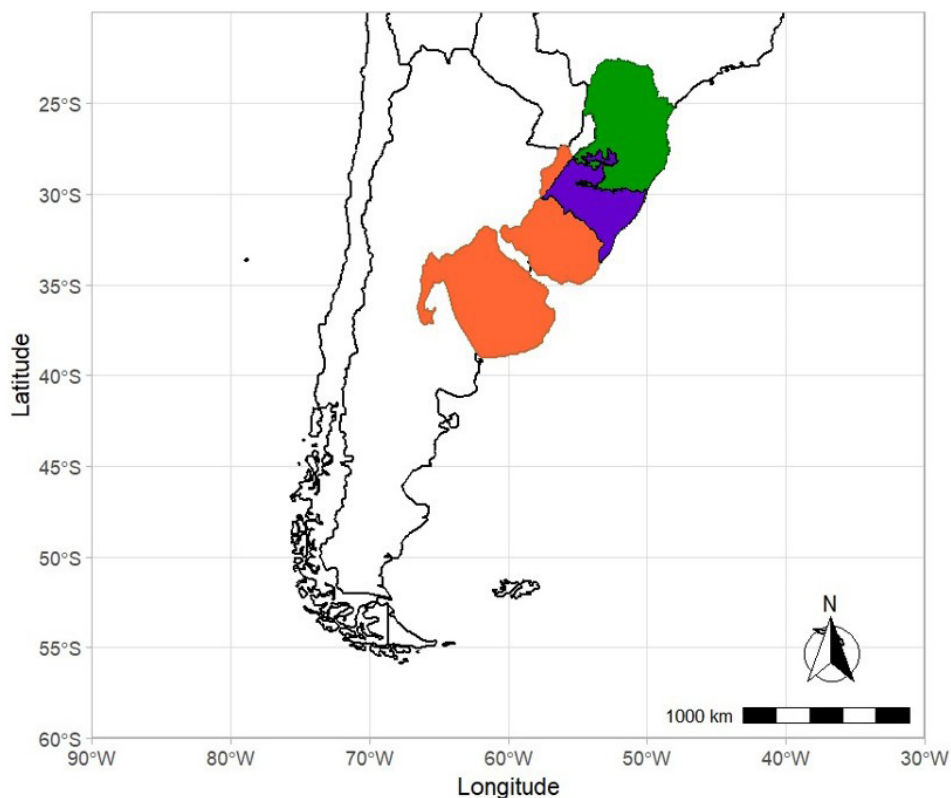
Sound knowledge of biodiversity has never been more important to support conservation strategies than it is today, in a period of accelerating human-driven changes in the distribution of biodiversity and of species losses (Dornelas et al 2014, Blowes et al. 2019). As biodiversity is important for ecosystem functions (van der Plas 2019) and the provisioning of ecosystem services that contribute to human well-being (Mace et al. 2012), knowledge on biodiversity is an important starting point for proposing sustainable land management.

Brazil is a megadiverse country, with a wide range of ecosystems, in addition to human cultures, ethnicities, and traditions that can bring nature's contribution closer to the daily life of Brazilians (Joly et al. 2019). Nonetheless, available numbers on Brazilian biodiversity are outdated and unified species lists are missing for many taxonomic groups. According to the latest comprehensive assessment, between 169,000 and 213,000 species are known from Brazil, corresponding to about 13.1% of the world's described biodiversity (Lewinsohn and Prado 2005). A large proportion of species remains undescribed (in particular in fungi, prokaryotes, and different insect groups), and estimates of species numbers have been used to assess overall species diversity in Brazil: likely, around 1.8 million species (Lewinsohn and Prado 2005). However, even for well-known species groups,

numbers and further information are scattered across publications, limiting the use of biodiversity knowledge for conservation policies.

With over 190,000 km<sup>2</sup>, the Brazilian Pampa (Fig. 1), the northern part of the Río de la Plata grasslands region (Soriano 1992), covers 2.3% of Brazil's area and is the second smallest Brazilian biome (*sensu* IBGE 2019). The region is dominated by grasslands, but also includes other natural ecosystems, such as forest and woodland, savanna-like parklands, palm groves, rock outcrops, dunes, different types of wetlands, and water bodies. These natural ecosystems are currently under threat: land use is changing rapidly across most of the region, especially driven by the expansion of croplands and exotic tree plantations (MapBiomias 2022). The area covered by natural non-forest formations, i.e., grasslands, decreased from 9.3 million ha in 1985 to 6.6 million ha in 2020. Today, agriculture sums up to 42.7% of the Brazilian Pampa's area (MapBiomias 2022). Simultaneously, the Brazilian Pampa has one of the largest deficits in biodiversity protection when compared to other Brazilian regions (Overbeck et al. 2015), with only 3.2% of its area covered by protected areas, parts of which include degraded lands (Ribeiro et al. 2021).

One of the reasons for this lack of consideration for the Brazilian Pampa in terms of conservation value is likely the lack of awareness of its biodiversity (Overbeck et al. 2015), a worldwide phenomenon for grasslands (e.g., Veldman et al. 2015). Information on



**Figure 1.** The Brazilian Pampa (*sensu* IBGE, 2019, in purple), located within the Río de la Plata grasslands region (in orange). Also shown are the geographic regions of southern Brazil (Brazilian Pampa + area in green) and the Southern Cone (in white, including all other areas).



species richness in the region at current is available for some taxonomic groups, but often incomplete and spatially biased (e.g., concentrated in regions close to research institutions). Here, we aim to improve this situation by trying to answer a simple question that is critical to increase the consideration of its biodiversity in public policies: how many and which species occur in the Brazilian Pampa? To answer this question, we present up-to-date information on the number of described plant, fungi, bacteria and animal species recorded from the Brazilian Pampa, stemming from contributions of a large number of experts. Based on our results, we discuss the need for further systematic biodiversity assessments.

## Materials & Methods

We compiled five datasets containing valid species names (and, in some cases, infraspecific taxa) of plants, fungi, bacteria and animals occurring in the Brazilian Pampa and additional descriptive information regarding taxa occurrence, distribution and conservation status (Table 1). They were compiled by experts based on available literature, biological collections, and expert knowledge, with occurrence referenced by a voucher specimen or a literature source in most cases (89% of the total number of taxa). For each specific and/or infraspecific taxon, information was provided regarding taxonomic classification, geographic distribution, habitat of occurrence and conservation status. Geographical distribution was classified as follows: (i) restricted to the Brazilian Pampa (IBGE 2019), (ii) restricted to the Río de la Plata grasslands region (Soriano 1992), (iii) restricted to southern Brazil (Paraná, Santa Catarina, Rio Grande do Sul states), (iv) restricted to Río de la Plata grasslands region and southern Brazil, or (v) widely distributed in the Southern Cone region (South America south of the Tropic of Capricorn), if occurring in other Brazilian states or adjacent countries beyond the limits of previous categories (Fig. 1). Species were also classified according to their main habitat (aquatic, forest and grassland, mainly forest, mainly grassland, only forest, only grassland, dune, anthropic, other types of habitats, insufficient data). For animals, we added the habitat classes cave and (for Aves) savanna-like parkland. All species were classified as native or non-native to the Brazilian Pampa. Species classified as non-native were checked for their potential as invasive species according to the state list of invasive alien species (RIO GRANDE DO SUL 2013). The regional, national and global conservation status was considered according to three threatened species lists: Rio Grande do Sul state list (SEMA/RS; RIO GRANDE DO SUL 2014a,b), National list of Brazil (ICMbio/MMA 2018), and the global International Union for Conservation of Nature (IUCN) list (IUCN 2021; <https://www.iucnredlist.org/>).

Plant and algae species were organized into three datasets (Tables S1-S3; Supplemental Appendix S1), one including all vascular species (Angiosperms [Eudicotyledons, Monocotyledons, Magnoliids, Nymphaeales], Gymnosperms, and Pteridophytes

[Lycophytes, Monilophytes]), a second one including avascular terrestrial plant species (i.e., Bryophytes [Anthocerotophyta, Bryophyta, Marchantiophyta]) and a third one containing the artificial group Algae (Bigyra, Cercozoa, Charophyta, Chlorophyta, Choanozoa, Cryptophyta, Cyanobacteria, Euglenozoa, Miozoa, Ochrophyta, Rhodophyta). Although algae is an artificial group, we chose to consider it as a unit for user's convenience and analytical simplicity. The vascular plants dataset originated from the comparison of two other datasets: (1) the list of vascular species of the Río de la Plata grasslands region (Andrade et al. 2018) and (2) the Brazilian Flora 2020 (BFG 2021), followed by a careful review by experts and consultation to taxonomic literature. The avascular terrestrial plants and algae datasets were based mainly on Brazilian Flora 2020 (BFG 2021) and extensive review of taxonomic literature, including monographs, and floristic and phytosociological studies. In cases where we found divergences among databases or references concerning species and author names, we opted to follow the International Plant Name Index (IPNI [IK]; <https://www.ipni.org>) and Algaebase (<https://www.algaebase.org/>). Species distribution data was obtained or confirmed by consulting taxonomic and ecological studies, as well as online databases such as Global Biodiversity Information Facility (GBIF; <https://www.gbif.org>) and SpeciesLink network (<http://www.splink.org.br/index>).

The fungi dataset is organized at the species level (Table S4; Supplemental Appendix S1). Fungi were categorized into lichenized and non-lichenized species. The classification of fungi orders follows Index Fungorum (<http://www.indexfungorum.org>), MycoBank (<https://www.mycobank.org>) and reviews of taxonomic literature and monographs. For this dataset, the Brazilian Flora 2020 (BFG 2021) was also important for its development.

High-level classification for fungi, gymnosperms, angiosperms and pteridophytes follows Tedersoo et al. 2018, Yang et al. 2022, APG IV (APG IV 2016), and PPG I (PPG I 2016), respectively. For algae, we used specific classification systems for each division (Cavalier-Smith and Chao 1996, Cavalier-Smith 1998, Lewis and McCourt 2004, Cavalier-Smith 2016, Kamiya et al. 2017, Garcia-Pichel et al. 2019) following Algaebase (Guiry and Guiry 2015). Cultivated species were not included, unless occurring spontaneously in natural areas.

Animal species were divided into two main groups (Table S5; Supplemental Appendix S1): vertebrates (i.e., Amphibia, Aves, Mammalia, Actinopterygii, Chondrichthyes, and Reptilia) and invertebrates (i.e., Acari, Annelida, Araneae, Crustacea, Diplopoda, Insecta [Coleoptera, Diptera, Ephemeroptera, Hemiptera, Hymenoptera, Isoptera, Lepidoptera, Odonata, Orthoptera, Plecoptera, Strepsiptera, Thysanoptera, Trichoptera], Mollusca, Platyhelminthes, and Porifera). Species information from both vertebrate and invertebrate groups was compiled from published and unpublished (e.g., scientific collections) sources, as well as online databases (GBIF and SpeciesLink network, plus the citizen-science platform WikiAves for Aves).

**Table 1.** Species richness of the Brazilian Pampa. Besides the currently known number (second column, our study), we also indicate prior knowledge for the Brazilian Pampa (third column) as well as current estimates for each species group for Brazil (fourth column) and the world (fifth column). In the third column, NA: no prior knowledge. In the twelfth column, CR: Critically Endangered, EN: Endangered, and VU: Vulnerable. References for prior knowledge of the Pampa and estimated numbers for Brazil and the world are in Supplemental Appendix S3. \*Diptera [Chironomidae, Culicidae], \*\*Coleoptera [Dytiscidae, Elmidae, Hydrophilidae, Noteridae], ■ Number does not include aquatic insects/gall-inducing insects which are listed as separate groups.

Species group	Total number of species	Prior knowledge (species number) for the Brazilian Pampa (Refs. in Supplemental Appendix S3)	Available species richness estimates for Brazil (Refs. in Supplemental Appendix S3)	Available species richness estimates for the world (Refs. in Supplemental Appendix S3)	Native species (%)	Invasive species (%)	Species restricted to Brazilian Pampa (%)	Species restricted to Rio de la Plata grasslands region (%)	Species that only occur in grasslands (%)	Species that mainly occur in grasslands (%)	Species listed as CR, EN, VU in any of the three lists of endangered species (see Methods)	Species considered extinct, regionally extinct, or probably extinct in any of the three lists (see Methods)
Angiosperms	3472	2825	35540	295383	92.5	1.2	3.7	8.8	31.8	28.1	430	11
Gymnosperms	5	3	114	1079	60	40	0	0	0	20	2	0
Pteridophytes (Lycophytes, Monilophytes)	165	119	1405	11916	97.6	0	0	1.8	13.9	0	9	6
Bryophytes (Anthocerotophyta, Bryophyta, Marchantiophyta)	316	125	1681	21925	100	0	0	0.3	0.3	19.3	19	0
Algae	2046	947	4993	44000	99.8	0.05	0.6	0.9	0	0	0	0
Fungi (Lichenized)	933	136	6320	115209	100	0	4.0	6.8	12.9	1	1	0
Fungi (Non-lichenized)	208	229	3148	36213	97.1	0	4.3	4.3	4.3	4.8	1	0
Amphibia	62	50	1188	8454	98.4	1.6	0	22.6	27.4	32.3	5	0
Aves	567	480	1971	11051	98.6	0.9	0	0.2	4.1	8.6	61	5
Fish (Actinopterygii, Chondrichthyes)	297	229	3148	36213	96.6	5.1	13.1	36.7	0	0	33	0
Mammals	120	110	541	6495	97.5	2.5	4.2	11.7	6.7	15.0	28	1
Reptiles	90	67	842	11733	97.8	2.2	3.3	17.8	12.2	34.4	10	0
Acarí	108	NA	NA	55000	100	0	8.3	8.3	3.7	3.7	0	0
Annelida (aquatic species)	21	20	86	1800	100	0	0	0	0	0	0	0
Annelida (terrestrial species)	51	NA	424	5663	41.2	0	5.9	5.9	3.9	0	0	0
Aquatic Insecta (Hemiptera)	82	71	554	4810	100	0	1.2	3.6	0	0	0	0
Aquatic Insecta (Ephemeroptera)	19	NA	426	3500	100	0	5.3	5.3	0	0	0	0
Aquatic Insecta (Plecoptera)	5	NA	199	3718	100	0	20	40	0	0	0	0
Aquatic Insecta (Trichoptera)	6	NA	864	NA	100	0	0	0	0	0	0	0
Aquatic Insecta (Diptera)*	71	NA	1183	NA	93	7	2.8	2.8	0	0	0	0

Table 1. Continued...

Species group	Total number of species	Prior knowledge (species number) for the Brazilian Pampa (Refs. in Supplemental Appendix S3)	Available species richness estimates for Brazil (Refs. in Supplemental Appendix S3)	Available species richness estimates for the world (Refs. in Supplemental Appendix S3)	Native species (%)	Invasive species (%)	Species restricted to Pampa (%)	Species restricted to Rio de la Plata grasslands region (%)	Species that only occur in grasslands (%)	Species that mainly occur in grasslands (%)	Species listed as CR, EN, VU in any of the three lists of endangered species (see Methods)	Species considered extinct, regionally extinct, or probably extinct in any of the three lists (see Methods)
Aquatic Insecta (Coleoptera)**	24	34	909	18000	100	0	0	20.8	0	0	0	0
Aquatic Insecta (Odonata)	105	NA	873	6363	100	0	1	2	0	0	1	0
Araneae	393	NA	6050	50000	100	0	9.7	12.7	0.3	7.6	0	0
Coleoptera	1007	126	34676	360000	99.7	0	4.8	6.6	0.4	3.3	0	0
Crustacea (aquatic species)	27	NA	2040	10200	100	0	14.8	14.8	0	0	5	0
Crustacea (terrestrial species)	22	NA		4000	100	0	4.5	4.5	0	0	0	0
Diplopoda	23	NA	541	13000	100	0	8.7	8.7	0	17.4	0	0
Diptera	542	29	11475	150000	95.9	0	0.6	1.8	0.6	11.4	0	0
Gall Inducing Insects	24	NA	NA	13000	100	0	4.2	12.5	0	8.3	0	0
Hemiptera (Auchenorrhyncha)	122	17	1653	43024	100	0	7.4	9	0	0	0	0
Hymenoptera (ants)	192	NA	1518	13964	96.9	0	0	3.1	0.5	17.2	1	0
Hymenoptera (bees)	274	NA	1576	20507	99.3	0	15	15.7	0	16.1	7	0
Hymenoptera (social wasps)	24	NA	381	1017	100	0	0	4.2	0	0	0	0
Isoptera	16	4	344	2919	93.8	0	0	0	12.5	25	0	0
Lepidoptera (butterflies)	513	NA	13018	135016	100	0	0	3.3	5.7	12.7	6	0
Lepidoptera (tiger moths)	212	NA			100	0	6.1	6.1	0	0.5	0	0
Lepidoptera (micromoths)	16	NA			87.5	0	12.5	12.5	0	0	0	0
Mollusca (aquatic species)	75	17	3165	5100	94.7	5.3	0	10.7	0	0	0	0
Mollusca (terrestrial species)	22	19		24380	90.9	9.1	22.7	31.8	0	4.5	0	0
Orthoptera (Grylloidea)	16	NA	287	5966	100	0	0	0	56.3	12.5	0	0
Platyhelminthes	66	21	2188	30000	42.4	0	4.5	4.5	0	0	0	0
Porifera	21	1	603	8899	100	0	4.8	9.5	0	0	4	0
Strepsiptera	2	NA	33	599	100	0	50	50	0	0	0	0
Thysanoptera	121	83	616	6375	100	0	5.8	9.1	17.4	12.4	0	0
TOTAL	12503	5533	143425	1560278							623	23

Prokaryote species were included in part considering species identity (for aquatic species), and also as operational taxonomic units (OTUs), for soil bacteria. For soil bacteria, the studies of Beneduzi et al. (2019), Girão (2019) and Granada et al. (2019) were used as a basis (Supplemental Appendix S2).

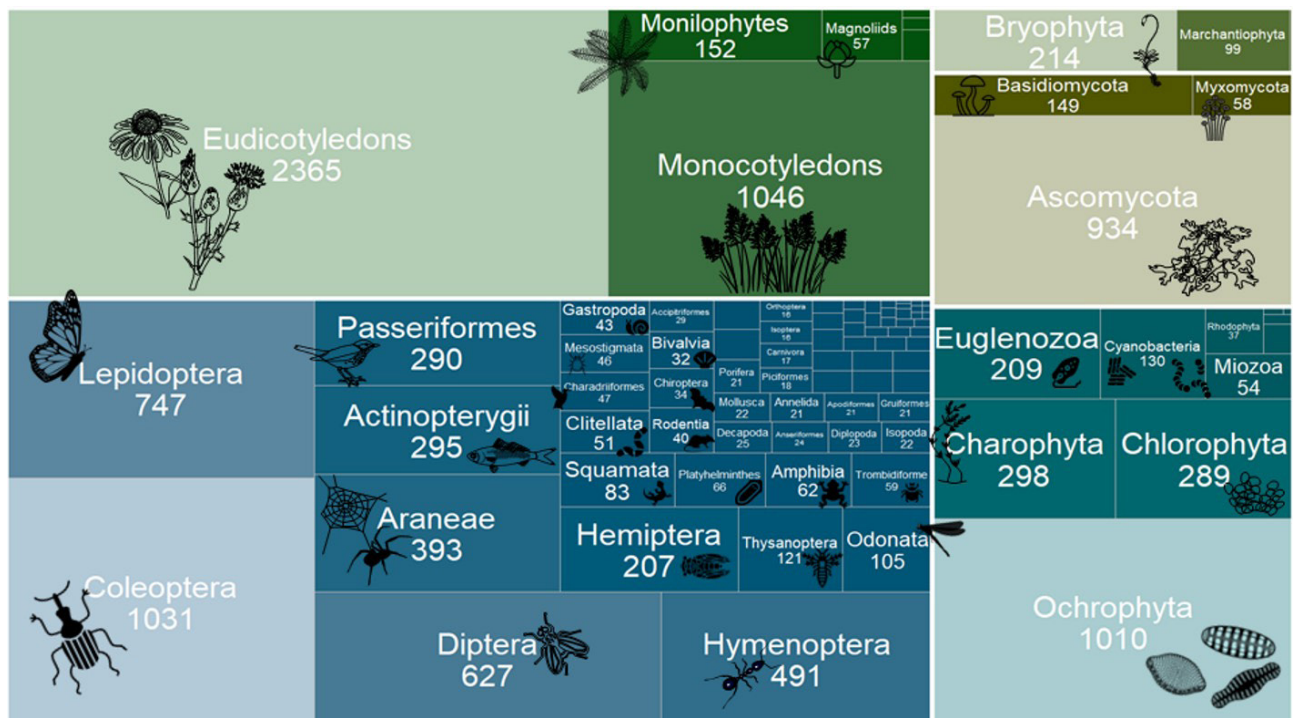
The vascular plants dataset is at the species level, while the other four datasets are organized at the infraspecific level. All calculations were made at species and infraspecific level. We used R 4.1.1 (R Core Team 2013) – loaded with the packages dplyr (Wickham et al. 2021), flora (Carvalho 2022), ggplot2 (Wickham 2016), rgbif (Chamberlain et al. 2022), redlist (Chamberlain 2020) and treemapify (Wilkins 2021) – for data preparation and manipulation, data querying, analysis, and visualization.

### Results

A total of 12,503 species (12,851 taxa) of plants, fungi (incl. lichenized fungi), bacteria and animals, from 1,025 families and 4,661 genera, are currently known to occur in the Brazilian Pampa (Table 1; Tables S1–S5, Supplemental Appendix S1). We organized the data into five datasets (Fig. 2; Tables S1–S5); metadata information is in Supplemental Appendix S1. These species are mostly native (97%), widely distributed in the Southern Cone region (83%). About half of the species occupy predominantly or exclusively grassland ecosystems (23%) or wetlands and aquatic habitats (26%). Only 3.5% and 6.7% of the species are restricted to the Brazilian Pampa and the Río de la Plata grassland

region, respectively. About a third of the species known for the Brazilian Pampa have been studied in sufficient detail to determine whether they are threatened with extinction at the regional, national, or global level. Overall, 622 species (5%) are classified as Critically Endangered (CR), Endangered (EN) or Vulnerable (VU) in at least one of the lists of threatened species considered (i.e., IUCN, ICMBio/MMA, and SEMA/RS). A total of 23 species have been declared extinct or probably extinct in the region.

The vascular plants dataset (Angiosperms [Eudicotyledons, Monocotyledons, Magnoliids, Nymphaeales], Gymnosperms, and Pteridophytes [Lycophytes, Monilophytes]) contains 3,642 species from 191 families and 1,108 genera, including 266 non-native and 40 non-native invasive species (Table S1, Supplemental Appendix S1; Supplemental Appendix S4). Although Monocot species, in particular grasses, dictate the physiognomy of grassland ecosystems, most species in the Brazilian Pampa are Eudicots. Asteraceae (476 species), Poaceae (423), Fabaceae (292), Orchidaceae (162), and Cyperaceae (155) are the families with the highest number of species. The most species-rich Lycophytes families are Lycopodiaceae (7 species) and Selaginellaceae (4), while Pteridaceae (33), Polypodiaceae (23), and Aspleniaceae (13) are the most species-rich Monilophytes families. As many as 3,161 species (87% of the total) are widely distributed in the Southern Cone region, while 130 species (3.5%) are restricted to the Brazilian Pampa. 2,105 species (58% of the total) occur predominantly or exclusively



**Figure 2.** Treemap of all plants, animals, fungi and bacteria species known from the Brazilian Pampa region. The species were organized into five datasets (clockwise order): vascular plant (in green), bryophyte (in olive), fungi (in khaki), algae (in green pine), and animal (in blue) species.



in grassland ecosystems. A total of 441 species (12% of the total) are classified as Critically Endangered (CR), Endangered (EN) or Vulnerable (VU) in at least one of the threatened species lists considered.

The Bryophytes dataset (Anthocerotophyta, Bryophyta, and Marchantiophyta) has 316 species (318 taxa) from 137 genera and 69 families (Table S2, Supplemental Appendix S1; Supplemental Appendix S4). Bryophyta species predominate (214 species), followed by Marchantiophyta (99), and Anthocerotophyta (3). For Bryophyta, the most representative family is Fissidentaceae (27 species), for Marchantiophyta, Ricciaceae (23), and for Anthocerotophyta, Notothyladaceae (2). All species are native, and most are widely distributed in the Southern Cone region (94%) and are found in forest and grassland (66%). Only 19 species (6% of the total) are classified as Critically Endangered (CR), Endangered (EN) or Vulnerable (VU) by at least one of the analyzed endangered species lists.

The algae dataset contains 2,046 species and 2,378 taxa (species, varieties, and forms) from 478 genera, 213 families and 11 divisions (Table S3, Supplemental Appendix S1; Supplemental Appendix S4). The most diverse Division is Ochrophyta (1,152 species), followed by Charophyta (381), Chlorophyta (328), and Euglenozoa (275). Desmidiaceae (282 species), Euglenidae (181), Bacillariaceae (126), Naviculaceae (104), and Eunotiaceae (112) are the families with the largest number of species. Only four species are considered exotics, one of them, invasive. Most species are widely distributed in the Southern Cone region (2,028 species or 85% of the total) and all species are aquatic. Euglenidae (3 species; *Strombomonas* [3 species]) and Bacillariaceae (3; *Tryblionella* [2]) are the families and genera with the largest number of species restricted to the Brazilian Pampa.

The fungi dataset has 1,141 species from 293 genera and 107 families (Table S4, Supplemental Appendix S1; Supplemental Appendix S4). Ascomycota species predominate (934), followed by Basidiomycota (149) and Myxomycota (58). 99.5% of the species are native. The lichenized fungi represent 82% of the species in the dataset. Parmeliaceae (182 species), Graphidaceae (117), Physciaceae (70), Caliciaceae (60 species), and Ramalinaceae (59) are the families with the largest number of species. Most species are widely distributed in the Southern Cone region (1,037 species or 91% of the total) and only 13% have grassland ecosystems as their preferred habitat. Graphidaceae (12 species; *Allographa* [7 species]), Trypetheliaceae (5), and Ramalinaceae (4; *Bacidia* [4]) are the families and genera with the largest number of species restricted to the Brazilian Pampa. Only two species are classified as Critically Endangered (CR) or Vulnerable (VU) by IUCN Red List of Brazilian Threatened Species.

The dataset on animal species contains 5,358 species and 5,372 taxa (97.5% of which are native), belonging to 2,652 genera and 445 families (Table S5, Supplemental Appendix S1; Supplemental Appendix S4). Vertebrates totaled 1,136 species, while invertebrates comprised 4,222 species. Aves is the vertebrate group

with the highest number of species (567), followed by Actinopterygii (295), Mammalia (120), Reptiles (90), Amphibia (62), and Chondrichthyes (2). The richest invertebrate groups are Coleoptera (1,031 species), Lepidoptera (747), Diptera (627), and Hymenoptera (491). Vertebrates had more species classified as Critically Endangered (CR), Endangered (EN) or Vulnerable (VU) (137 species) than invertebrates (24 species) in at least one of the threatened species lists considered. Most vertebrates (82.5%) and invertebrates (70%) are widely distributed in the Southern Cone region. The proportion of vertebrate and invertebrate species restricted to the Brazilian Pampa is 4% and 5%, respectively. High proportions of species restricted to the Río de la Plata grassland region were found for fish (Actinopterygii and Chondrichthyes), amphibians, and reptiles, with 36.7%, 23.6%, and 17.8%, respectively. Regarding the main habitat type, vertebrate species are predominantly associated with aquatic (40%), forest (20%) or grassland habitats (15%), while most invertebrates have poorly known habitat preferences (28.5%) or occupy both forest and grassland ecosystems (22%).

The five sampling sites for soil microbiological diversity yielded numbers between 362 and 692 operational taxonomic units (OTUs) (Supplemental Appendix S2). Orders that were represented by more than 10% of OTUs in at least one sampling site were Acidobacteriales (Acidobacteria: Acidobacteria), Frankiales (Acidobacteria: Acidobacteria), Bacillales (Firmicutes: Bacilli), Rhizobiales (Proteobacteria: Alphaproteobacteria), Burkholderiales (Proteobacteria: Gammaproteobacteria), and Chthoniobacterales (Verrucomicrobia: Spartobacteria).

## Discussion

Our study is the first comprehensive compilation of described species for the Brazilian Pampa, providing up-to-date species richness assessments for all major biological groups in this region. While our compilation improved previously existing lists for many taxa (see Table 1), for others our efforts led to the first species lists ever compiled for the region. The number of angiosperm plants for the Brazilian Pampa increased by over 500 species in comparison with previous lists. Recorded species richness of bryophytes and algae increased by more than 60%. While the known species richness increased for all animal groups, our study is the first available species richness assessment for many groups of invertebrates whose diversity was previously only known to specialists. Only 12% of fungi species listed here are also listed in the *Flora e Funga do Brasil* project (BFG 2021). We are aware that some species groups are still missing in our compilation, for example, within Coleoptera, Hemiptera, Hymenoptera, and Orthoptera, as we did not manage to cover all groups within our group of researchers. We hope that other researchers will join us in updating this list in the future.

The total species richness revealed by our study (12,503) demonstrates that a considerable part of Brazilian biodiversity is concentrated in a relatively small portion of the country that is under-considered

in conservation strategies (Overbeck et al. 2015) and experiences rapid conversion of natural ecosystems into anthropogenic areas (MapBiomias 2022). Our data indicate that the Brazilian Pampa holds, in little more than 2% of Brazil's area, around 9% of the country's species. Species groups for which the Brazilian Pampa has an especially high contribution to Brazil's diversity (based on currently available knowledge) are Algae (41%), Aves (28.8%), mammals (22.2%), Thysanoptera (19.6%), liverworts, hornworts and mosses (18.8%), and Hymenoptera/bees (17.4%) (see Table 1). For many species groups considered here, the numbers given certainly still fall far short of representing the real species diversity (both in the Brazilian Pampa and in Brazil). This is especially true for 'invisible' taxa such as soil organisms, microorganisms or even mosses and lichens (e.g., Rocha et al. 2016), which are still poorly studied and often left out of biodiversity inventories. Importantly, these organisms are currently not considered in environmental licensing and are also poorly represented in red list assessments of threatened species. Furthermore, many groups, especially invertebrates, have a high number of undescribed species. For example, in studies on ants, approximately 40-50% of the total species sampled are still unknown (Dröse et al. 2017). Given the accelerated land use change in the region, the risk of losing species before they are known is real despite their relevance to ecosystem processes and potential use in medicine or industrial processes. For poorly described groups, especially bacteria, fungi, and other soil organisms, we still know very little about the effects of land use and other environmental changes, or about their role in maintaining overall biodiversity (see van der Heijden et al. 2008).

Across all groups, the percentage of species restricted to the Brazilian Pampa was very low, even though it needs to be acknowledged that information on distribution is not available for all groups or species. A high level of endemism to the Río de la Plata grasslands region was found for fish (Actinopterygii and Chondrichthyes; 36.7% of species restricted to this region), amphibians (23.6%), and reptiles (17.8%), revealing a high biogeographical affinity of these groups with open ecosystems in the subtropical/temperate zone. The high number of endemic species of annual fish (*Austrolebias* and *Cynopoecilus*, 30 species) that live in temporary ponds and whose life cycle depends on successive periods of drought and flood is especially remarkable, but also the endemism of several groups of fish that have diversified among rivers located in the Río de la Plata grasslands region (e.g., *Scleronema*; see Ferrer and Malabarba 2020). Further, the Río de la Plata grasslands region is considered to be the center of diversity of *Melanophryniscus* frogs (Pramuk et al. 2008, Santos et al. 2014). Similar to plants (59.9% of species occur exclusively or mainly in grasslands), amphibians and reptiles also had a high percentage of species closely associated with grasslands (grassland as exclusive or main habitat: 57.7% and 46.7% of species, respectively), emphasizing the high biodiversity of open ecosystems. The total of 623 species currently

listed as threatened (categories Critically Endangered, Endangered or Vulnerable in at least one of the lists consulted) as well as the 23 species considered extinct, regionally extinct or probably extinct, reflect the poor conservation status of the region, with ongoing and rapid land use change. Clearly, the Brazilian Pampa is suffering from Biome Awareness Disparity, i.e., poor conservation of grasslands relative to forests (Silveira et al. 2021). We hope that our new findings on the biodiversity of the Brazilian Pampa will help to change this situation.

Although the focus of this study is the native biodiversity of the Brazilian Pampa, and cultivated species were not included, we should not forget the importance of the region's socio-biodiversity. Before European colonization, Guaraní people cultivated species brought from other regions of South and Central America such as cassava (*Manihot esculenta*), maize (*Zea mays*), beans (*Phaseolus vulgaris*), sweet potato (*Ipomoea batatas*) and pumpkin/squash (*Cucurbita* sp.), among others. With European colonization, plants from the old world and distant parts of the new world were introduced and today local varieties are still used and traded (Barbieri et al. 2014). Several native plant species, already used by traditional communities, have been started to be commercially exploited, at least locally, in food production chains, such as pitanga (*Eugenia uniflora*), jelly palm (*Butia odorata*), araquá (*Psidium cattleianum*), alongside plants with pharmaceutical properties, such as *marcela* (*Achyrocline satureioides*), *bananinha-do-mato* (*Bromelia antiacantha*) or *carqueja* (*Baccharis crispa*), to give just a few examples (Barbieri et al. 2014, Ramos et al. 2017). On a larger scale, traditional animal husbandry on native grassland, maintaining natural biodiversity, is an important socio-environmental and economic element in the Brazilian Pampa (Nabinger et al. 2009), just as is beekeeping of native stingless bees (Meliponini), such as the locally threatened *Plebeia wittmanni* and *Melipona quadrifasciata*, among other species (Dos Santos et al. 2021). These examples are indicative of the high potential of sustainable use of natural resources to maintain biodiversity and ecosystem services (including cultural services) in a way that is also economically interesting, involves local people and maintains their cultures.

While our study considerably increases knowledge on natural biodiversity in the Brazilian Pampa, more efforts to collect biodiversity data are clearly necessary here and in other Brazilian regions. We suggest that this should be done in a combination of approaches. We need targeted sampling of understudied groups (including soil organisms) and regions for which information on species richness and distribution, as well as taxonomic knowledge, is still incipient. Data from this type of study are highly relevant to improve regional species lists, as presented here. We also need standardized biodiversity inventories that can contribute to a better understanding of biodiversity patterns in space and time (e.g., Ferreira et al. 2020, Rosa et al. 2021). Collecting and describing organisms

and sampling biodiversity are, however, only the first steps toward a better knowledge of biodiversity: information needs to be made available to other researchers and to society as a whole. While for some species – in particular, the flora – easily accessible databases exist and are constantly improved (e.g., BFG 2021), similar initiatives are still lacking for many other groups, despite efforts underway such as the *Catálogo Taxonômico da Fauna do Brasil*, or, on a global level, the Catalogue of Life (<https://www.catalogueoflife.org/>). Platforms where researchers, natural resource managers and the interested public can easily store and access biodiversity information are highly relevant to improve biodiversity management. Additional information on species, such as classifications according to habitat preference, is important to increase usability of the data for researchers, environmental managers, or the interested public. Beyond data compilation and sharing, outreach activities are necessary to spread information on biodiversity to society. All these activities need to be well integrated – ideally in networks – and to be able to count on reliable funding. Biodiversity is a strategic issue in a world undergoing global change, and biodiversity knowledge is fundamental. This needs to be reflected in appropriate government policies for: (i) funding studies on biodiversity and its distribution, as well as on conservation needs and strategies through specific calls; (ii) creating easily accessible, constantly updated and properly curated databases on biodiversity of all Brazilian regions; (iii) better consideration of biodiversity in school curricula and other educational activities.

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## Author Contributions

GEO conceived the study. BOA and WD organized data collection, managed data and conducted the analyses. GEO, BOA and WD led the writing. All authors

contributed with data and revised the text. All authors approved the final version.

## Data Accessibility Statement

All data are available in the Zenodo Repository (doi: 10.21425/F5FBG59288; <https://zenodo.org/record/7504156>). The dataset metadata, on the other hand, are in Supplemental Material (<https://escholarship.org/uc/fb>).

## Supplemental Material

The following materials are available as part of the online article at <https://escholarship.org/uc/fb>

**Appendix S1.** Dataset Metadata.

**Appendix S2.** Soil microbial diversity: methodological strategy and spatial overview.

**Appendix S3.** Table 1 References.

**Appendix S4.** Most frequent families and genera for all datasets.

**Appendix S5.** Acknowledgements from individual researchers.

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