parasitologia

MDPI

Review

Biocontrol of Avian Gastrointestinal Parasites Using Predatory Fungi: Current Status, Challenges, and Opportunities

João Lozano [1,2,*], Cristina Almeida [3], Manuela Oliveira [1,2], Adolfo Paz-Silva [4] and Luís Madeira de Carvalho [1,2]

1 CIISA—Centre for Interdisciplinary Research in Animal Health, Faculty of Veterinary Medicine, University of Lisbon, Avenida da Universidade Técnica, 1300-477 Lisbon, Portugal; moliveira@fmv.ulisboa.pt (M.O.); madeiradecarvalho@fmv.ulisboa.pt (L.M.d.C.)
2 Associate Laboratory for Animal and Veterinary Sciences (AL4AnimalS), 1300-477 Lisbon, Portugal
3 Exoclinic—Clínica Veterinária de Aves e Exóticos, Quinta de Santo António, 1495-049 Miraflores, Portugal; cristinaalmeida@mail.com
4 Control of Parasites Research Group (COPAR, GI-2120), Department of Animal Pathology, Faculty of Veterinary, University of Santiago de Compostela, 27142 Lugo, Spain; adolfo.paz@usc.es
* Correspondence: jlozano@fmv.ulisboa.pt

Abstract: This review describes the current research status regarding the implementation of predatory fungi in the biological control approach of bird gastrointestinal (GI) parasitosis. The main GI parasites of Galliformes (e.g., broilers, layers, peacocks, pheasants) and Ratites (e.g., ostriches, emus, rheas) are addressed, as well as their impact on farms, zoos, and private collections. The main characteristics regarding biocontrol with predatory fungi are briefly described, such as their mode of action and efficacy against GI parasites of different animal hosts. The state of the art regarding the use of predatory fungi in birds is reviewed here by describing all associated articles already published in the main databases, techniques, and their main findings. Ovicidal fungi such as *Pochonia chlamydosporia*, *Metarhizium* spp. and *Acremonium* spp., and larvicidal fungi, namely *Duddingtonia flagrans*, *Arthrobotrys* spp. and *Monacrosporium thaumasium*, have shown promising predacious activity against ascarid eggs and nematode larvae from chickens and ostriches, both in vitro and in vivo, also revealing tolerance to the GI passage in chickens and maintenance of predacious capacity. Further studies are needed to understand the fungi–parasite–host gut microbiota interactions and target other avian GI parasitic species, such as nematodes, coccidia, cestodes, and trematodes.

Keywords: birds; intestinal parasites; biological control; predatory fungi

Citation: Lozano, J.; Almeida, C.; Oliveira, M.; Paz-Silva, A.; Madeira de Carvalho, L. Biocontrol of Avian Gastrointestinal Parasites Using Predatory Fungi: Current Status, Challenges, and Opportunities. *Parasitologia* **2022**, *2*, 37–44. https://doi.org/10.3390/parasitologia2010004

Academic Editor: Theo De Waal

Received: 25 January 2022
Accepted: 27 February 2022
Published: 1 March 2022

1. Gastrointestinal Parasites of Galliformes and Ratites

Domestic and exotic birds are commonly exposed to a wide variety of generalist or host-specific gastrointestinal (GI) parasites, with different life cycles and levels of pathogenicity [1–8].

In Galliformes kept on free-range farms, zoos, and public gardens (e.g., broilers, layers, peacocks, pheasants), coccidia infections caused by *Eimeria* spp. and *Isospora* spp. can reach prevalence and shedding values up to 80% and 15,000 oocysts per gram of feces (OPG), respectively [6,9–14] and are currently responsible for average losses of approximately 12 billion € annually worldwide in the poultry industry [5,15]. Nematode infections are also a serious problem in Galliformes, being ascarids (e.g., *Ascaridia galli*), heterakids (e.g., *Heterakis gallinarum* and *H. isolonche*), capillarids (e.g., *Capillaria* spp.), strongyles (e.g., *Trichostrongylus tenuis*), and *Strongyloides* spp., the most frequent and pathogenic species [6,8–11,13,16,17].

Larger birds like Ratites (e.g., ostriches, emus and rheas), which are commonly kept in zoos worldwide for ornamental exhibition and occasionally in farms for production purposes, are also susceptible to GI parasitic infections, and nematodes belonging to

the genera *Libyostrongylus* and *Codiostomum* are of most clinical importance, especially *Libyostrongylus douglassii*, which is responsible for the rotten stomach disease [6,18–22].

The control of these agents based solely on the administration of antiparasitic compounds (e.g., anticoccidials and anthelminthics) is of limited utility, since they do not act on the environmental forms of the parasites. In addition, common drug misuse in livestock farms often leads to efficacies lower than expected, appearance of drug resistance, and potential contamination of the environment with drug residues [23–26].

New complementary strategies are being proposed for integrated GI parasite control in domestic and wild animals kept in captivity, namely the use of predatory fungi as an accurate, innovative, natural, and sustainable tool [27,28].

2. Biocontrol of GI Parasites Using Predatory Fungi

Over the past 20 years, there has been an increasing interest in research regarding the use of predatory fungi (also referred as "nematophagous fungi", or more recently "helminthophagous fungi") for the biocontrol of animal gastrointestinal parasites, in complement with drug treatments.

These are saprophytic filamentous fungi belonging mainly to the phyla Ascomycota and Mucoromycota, often found in agricultural soil and organic decaying matter, which play a role in the recycling of carbon, nitrogen, and other elements originating from nematode degradation [29]. Besides their common saprophytic characteristics, these fungi also have the ability to predate intestinal parasites of animals, especially the eggs and larvae, which serve as an additional source of nutrients for fungal growth. Their tolerance to the animal's gastrointestinal transit has already been demonstrated, being expelled with feces to the soil, where they start predating parasitic forms, especially in micro-fecal and peri-fecal environments [30].

There are three main groups of predatory fungi, defined according to their mode of action: larvicidal, ovicidal, and endoparasitic, the first two being the most commonly used in biocontrol trials. For larvicidal fungi such as *Duddingtonia flagrans*, *Arthrobotrys* spp., and *Monacrosporium thaumasium,* the main feature is the production of a wide diversity of traps (e.g., constricting rings, non-constricting rings, adhesive nodules, and ramifications), whose formation is stimulated by the presence of helminth larvae. For ovicidal fungi, namely *Mucor circinelloides*, *Pochonia chlamydosporia*, *Verticillium* spp., *Purpureocillium lilacinum* (formerly known as *Paecilomyces lilacinus*) and *Trichoderma* spp., the main characteristic consists of their ability to predate helminth eggs, and it is the presence of parasite eggs that triggers fungal hyphae migration towards their cuticula, in which mechanic and enzymatic activity are developed [29].

Both larvicidal and ovicidal fungi have been used in several in vitro and in vivo experiments, being unanimously considered an accurate and sustainable tool for the control of GI parasites, resulting in a reduction in the number of eggs per gram of feces (EPG) of 60–97% in field trials with grazing animals [28,31–35]. The lack of adverse effects of *D. flagrans* on soil nematodes [36], as well as the innocuousness of *M. circinelloides* and *D. flagrans* on several animal species [35,37] should also be underlined.

These fungi have already been isolated in America [38–42], Europe [43], Asia [44,45], Oceania [46,47], and even in Antarctica [48], and two commercial formulations of *D. flagrans* are already commercially available in Australia and New Zealand (BioWorma®—NCIMB 30336, BioWorma, Sydney, Australia) and in Brazil (Bioverm®—AC001, GhenVet Saúde Animal, Paulínia, Brazil).

3. Testing the Use of Predatory Fungi against Avian GI Parasites: State of the Art

Despite the increasing number of studies in this topic, most of them are focused on the biocontrol of intestinal parasites affecting ruminants and horses, and there is a lack of research regarding the use of predatory fungi in other animals, such as birds.

A literature search was performed in November 2021, in PubMed, Scopus, Web of Science and Google Scholar databases, using the search string "(predatory fungi OR

predacious fungi OR duddingtonia OR arthrobotrys OR monacrosporium OR mucor OR pochonia OR verticillium OR paecilomyces OR trichoderma) AND (coccidia OR helminth OR nematode)". Title and abstract analysis were performed, only research articles in English and published from 1990 until 2021 were included, and other types of publications (e.g., reviews, letters, and editorials) were excluded. It was found that only 5 publications were related to in vitro and in vivo experiments using predatory fungi against avian GI parasites (4 original research articles and 1 research note), carried out in Brazil and Denmark (Table 1).

Table 1. In vitro and in vivo research performed with predatory fungi against avian GI parasites.

Type of Assay	Fungal Species (Biotype)	Target Organism	Study Objectives	Reference
In vitro	*D. flagrans* (AC001; CG722) *A. cladodes* (CG719)	*L. douglassii*	Test larvicidal activity against L3 larvae	[49]
	P. chlamydosporia (Biotype 10) *Me. brunneum* (KVL04-57; KVL16-26) *Me. carneum* (KVL16-33) *Acremonium* sp. (KVL16-34)	*A. galli* *H. gallinarum*	Test ovicidal activity in different soil types; isolate native ovicidal fungi	[50]
In vivo	*D. flagrans* (AC001; CG722) *M. thaumasium* (NF34A)	*Panagrellus* spp.	Test GI passage in chickens and evaluate the maintenance of germination and larvicidal capacities	[51]
	P. chlamydosporia (VC4)	*A. galli* *H. gallinarum*	Test GI passage in chickens and evaluate the maintenance of germination and ovicidal capacities	[52]
	P. chlamydosporia (Biotype 10)	*A. galli* *H. gallinarum*	Test ovicidal activity in different soil types; evaluate the interaction soil-fungi in birds worm population and burdens, and egg counting	[53]

The first in vitro experiment with predatory fungi against avian intestinal parasites was reported 9 years ago by Braga et al. [49]. The study aimed to test the larvicidal activity of two isolates of *D. flagrans* (AC001 and CG722) and one isolate of *Arthrobotrys cladodes* (CG719) on infective larvae (L3) of *L. douglassii*. The assays were performed in plates with Water-Agar medium (WA, 2%) and the number of non-preyed L3 was counted daily, for seven days of incubation, in all treated and control groups. Percentage reductions of L3 were found to be significant between test and control plates, totalizing efficacies of 85.2% (isolate AC001), 81.2% (CG722), and 89.2% (CG719). Isolates did not differ in the daily mean of non-preyed L3, but all of them differed significantly from control plates, and therefore these isolates offer potential to be used in the biocontrol of GI nematodes of ratites.

Another in vitro study was conducted in Denmark by Thapa et al. [50], which aimed to test the performance of *P. chlamydosporia* (Biotype 10) and *Metarhizium brunneum* (KVL04-57) against non-embryonated ascarid eggs (*A. galli* and *Heterakis* spp.) in sterilized and non-sterilized soils. Egg recovery was examined before and after incubation at 22 °C for 30 days. In sterilized soil, results were significantly influenced by the interaction between fungal treatment and incubation time, with egg count differing between treatments and controls after 30 days of incubation, and *P. chlamydosporia* and *Me. brunneum* showing reduction efficacies of 46% and 30%, respectively. However, in non-sterilized soil, the outcomes were slightly different, with both fungal and control plates showing significant egg recovery reductions (68–77%). In this case, only *Me. brunneum* treatment resulted in slight but significant reductions in comparison with controls and *P. chlamydosporia* plates. These results suggest that resource competition between predatory fungi and native soil microbiota may interfere negatively with the performance of fungal isolates, as well as

rejects the hypothesis of potential environmental impact on soil microbiota caused by the administration of these fungi.

In this study, the authors also aimed to evaluate the survival of ascarid eggs in different soil types, both in sterilized and non-sterilized soil, after 30 days of incubation at 22 °C. For sterilized soils, only incubation time and soil type had a significant interaction on egg recovery. For non-sterilized soils, the egg counts were significantly reduced in all soil types, ranging from 38% to 99%. Non-sterilized soils exhibiting the highest ovicidal activities were also used to isolate, identify, and test the antagonistic effect of native fungi against ascarid eggs. Fungal isolates belonged to the genera *Metarhizium* and *Acremonium*; however, none of the three isolates revealed predatory efficacies higher than 34% after 28 days of exposure. These results also suggest that soil has inherent biotic egg-degrading properties, namely due to its native microbiota.

Predatory fungi have also been tested in vivo in chickens and hens, with the first published report dating back to 2017. The study developed by Silva et al. [51] aimed to test the maintenance of germination and larvicidal capacities of *D. flagrans* (AC001; CG722) and *M. thaumasium* (NF34A) after passing through the GI tract of chickens. For this purpose, four experimental groups with two chickens were considered: three groups were provided with autoclaved concentrate feed mixed with 1 mL of an aqueous solution containing 6.4×10^4 spores of each isolate (test groups), and 1 group received feed mixed with distilled water (control group), on a daily basis. Fecal samples were collected 6, 12, 24, 48, and 74 h post-administration, and placed in Petri dishes with WA medium. Suspensions containing larvae of the free-living nematode *Panagrellus* spp. were also added to each plate, followed by incubation at 25 °C for 12 days, to test mycelial growth and average number of recovered larvae in each period of administration. Fungal structures from all isolates were observed at 6, 12, and 24 h post-administration, confirming the ability of spores to resist the GI passage in chickens. In addition, the highest percentage of reduction in the number of recovered larvae was identified at 6 h post-administration, averaging reduction rates of approximately 35% to 71%, with only isolate AC001 showing a significant reduction in comparison with the control plates. Despite larvicidal activity being tested against free-living nematodes, results from this study can be extrapolated to parasitic nematodes affecting bird species, due to a similar mode of action.

A study conducted by Valadão et al. [52] also aimed to test the maintenance of germination and ovicidal capacities of *P. chlamydosporia* (VC4) after GI transit in chickens, with an experimental design similar to the previously mentioned study. A group of 22 chickens was divided into two experimental groups: both groups received a supplementation of shredded corn for 7 days, after which only the test group started to receive the supplement inoculated with *P. chlamydosporia*. Samples were collected in each group after 0, 6, 8, 10, 12, 18, and 24 h post-administration, and placed in plates with WA medium, followed by incubation at 25 °C for 30 days, to check for the growth of *P. chlamydosporia*. The authors reported the identification of VC4 isolate only in samples from the test group, and 6 h post administration. VC4 isolates obtained after 30 days of incubation were used for further in vitro tests in WA medium, aiming to check the maintenance of ovicidal activity against *A. galli* and *H. gallinarum* eggs. A significant reduction in egg viability was observed after 74 h of incubation and the highest rates were recorded after 144 h, totalizing approximately 60% and 40% for *A. galli* and *H. gallinarum*, respectively.

Finally, a study performed by Thapa et al. [53] aimed to evaluate the performance of *P. chlamydosporia* (Biotype 10) in reducing worm burden and ascarid egg count in hens, by jointly giving the fungus with sterilized and non-sterilized soil. These soils were previously used in in vitro trials aiming to evaluate the egg recovery in sterilized and non-sterilized substrates inoculated with *P. chlamydosporia*. For the in vivo trial, birds were fed with the same soils together with the morning meal, comprising four experimental groups: sterilized control soil (SC), sterilized soil with fungus (SF), non-sterilized control soil (NC), and non-sterilized soil with fungus (NF). The study aimed to analyze worm recovery, fecal eggs counts, and *A. galli* Igγ levels after fungal administration. A significant interaction

between soil sterility and fungal treatment on ascarid worm burden was observed, which decreased significantly only in hens fed with sterilized soil inoculated with *P. chlamydosporia*, in comparison with the other three treatments. However, this scenario was completely different from that observed for egg counting, in which the overall EPG in the SF group was significantly higher than in groups SC and NC, but not versus the NF group. In addition, hens from the SF group had significant higher proportions of the three largest worm length categories (1.5–3.0 cm, 3.0–5.0 cm, 5.0–8.0 cm), in comparison with the other groups. This was an interesting result since the SF group had the lowest mean worm burden of *Ascaridia galli* and the highest abundance of mature worms, which allowed to conclude that reduced exposure modified *A. galli* populations. As stated by the authors, if all ascarid forms are not eradicated from the farm's soil or litter, the remaining eggs might therefore lead to long-term serious infection outbreaks in flocks. These results emphasize the need to optimize parasite control programs in farms, targeting the reduction of environmental contamination with eggs and thus avoiding episodes of re-infection.

4. Further Research

Although only five research articles related with the use of predatory fungi against GI parasites of birds have been published to date, overall results reveal their potential effectiveness against nematode eggs and larvae and suggest their possible use in parasite control programs for domestic and exotic birds.

Despite their promising utility, some questions remain to be addressed. One of them refers to the impact of fungal administration on bird intestinal microbiota and if it can have a potential probiotic effect, besides their activity on fecal and soil environment. Interactions between the intestinal microbiota diversity and the chicken's productivity has been demonstrated by several authors, although depending on the type of sample used for 16S rDNA sequencing (e.g., small intestine, large intestine, feces), with generally a higher bacterial diversity being found in the intestine of chickens with greater feed conversion ratio [54]. A growing number of studies aiming to characterize the relationships between parasites and the gut microbiota in several animal hosts has also been observed. For example, Huang et al. [55] demonstrated that, in chickens, coccidiosis modulated the avian gut microbiota towards a lower bacterial diversity and relative abundances of *Lactobacillus* and *Faecalibacterium*, in contrast to higher abundances of *Clostridium*, *Lysinibacillus* and *Escherichia* after fecal analysis. Therefore, it would be interesting to analyse the influence of predacious fungi administration on host intestinal microbiota, and to investigate if they can have a potential dual action on parasitism by regulating the gut microbiota and predating environmental forms.

More in vitro studies are needed to test these fungi against other bird GI parasites. Promising results already obtained against ascarid eggs and nematode larvae also reveal that it would be interesting to check the efficacy of ovicidal fungi against coccidia oocysts, cestode, and trematode eggs, as well as larvicidal fungi against L3 larvae from other nematode species. In addition, more in vivo studies using fungal formulations need to be performed in several species of domestic and exotic birds, kept in farms, zoos, or private collections, and evaluate the long-term kinetics of egg/oocyst shedding in the environment.

Since these fungi are often found in agricultural soils and animal feces, there is a great opportunity for scientific centres working on this topic to isolate native fungal species with predatory capacity and establish mycological collections, and routinely test them against GI parasites, namely from birds, both in vitro and in vivo, setting up the basis for developing more biocontrol products with market application.

Author Contributions: Conceptualization, J.L. and C.A.; methodology, J.L. and L.M.d.C.; validation, M.O., A.P.-S. and L.M.d.C.; investigation, J.L. and C.A.; resources, J.L. and L.M.d.C.; data curation, J.L.; writing—original draft preparation, J.L. and C.A.; writing—review and editing, M.O., A.P.-S. and L.M.d.C.; supervision, M.O., A.P.-S. and L.M.d.C.; project administration, L.M.d.C.; funding acquisition, J.L. and L.M.d.C. All authors have read and agreed to the published version of the manuscript.

Funding: This research was supported by CIISA—Centre for Interdisciplinary Research in Animal Health, Faculty of Veterinary Medicine, University of Lisbon, Lisbon, Portugal, Project UIDB/00276/2020 (funded by FCT). Additionally, João Lozano was awarded a PhD research fellowship 2020.09037.BD (funded by FCT).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Yazwinski, T.A.; Tucker, C.A. Nematodes and Acanthocephalans. In *Diseases of Poultry*, 12th ed.; Saif, Y.M., Ed.; Blackwell Publishing: Hoboken, NJ, USA, 2008; pp. 1025–1056.
2. Thapa, S.; Hinrichsen, L.K.; Brenninkmeyer, C.; Gunnarsson, S.; Heerkens, J.L.T.; Verwer, C.; Niebuhr, K.; Willett, A.; Grilli, G.; Thamsborg, S.M.; et al. Prevalence and magnitude of helminth infections in organic laying hens (*Gallus domesticus*) across Europe. *Vet. Parasitol.* **2015**, *214*, 118–124. [CrossRef]
3. Fatoba, A.J.; Adeleke, M.A. Diagnosis and control of chicken coccidiosis: A recent update. *J. Parasit. Dis.* **2018**, *42*, 483–493. [CrossRef]
4. Lozano, J.; Anaya, A.; Palomero Salinero, A.; Lux Hoppe, E.G.; Gomes, L.; Paz-Silva, A.; Teresa Rebelo, M.; Madeira de Carvalho, L. Gastrointestinal parasites of free-range chickens—A worldwide issue. *Bull. UASVM Vet. Med.* **2019**, *76*, 110–117. [CrossRef]
5. Attree, E.; Sanchez-Arsuaga, G.; Jones, M.; Xia, D.; Marugan-Hernandez, V.; Blake, D.; Tomley, F. Controlling the causative agents of coccidiosis in domestic chickens; an eye on the past and considerations for the future. *CABI Agric. Biosci.* **2021**, *2*, 37. [CrossRef]
6. Lozano, J.; Almeida, C.; Victório, A.C.; Melo, P.; Rodrigues, J.P.; Rinaldi, L.; Cringoli, G.; Gomes, L.; Oliveira, M.; Paz-Silva, A.; et al. Implementation of Mini-FLOTAC in Routine Diagnosis of Coccidia and Helminth Infections in Domestic and Exotic Birds. *Vet. Sci.* **2021**, *8*, 160. [CrossRef]
7. Mesa-Pineda, C.; Navarro-Ruíz, J.L.; López-Osorio, S.; Chaparro-Gutiérrez, J.J.; Gómez-Osorio, L.M. Chicken Coccidiosis: From the Parasite Lifecycle to Control of Disease. *Front. Vet. Sci.* **2021**, *8*, 787653. [CrossRef]
8. Nath, T.C.; Eom, K.S.; Choe, S.; Hm, S.; Islam, S.; Ndosi, B.A.; Kang, Y.; Bia, M.M.; Kim, S.; Eamudomkarn, C.; et al. Insight into One Health Approach: Endoparasite Infections in Captive Wildlife in Bangladesh. *Pathogens* **2021**, *10*, 250. [CrossRef]
9. Titilincu, A.; Mircean, V.; Bejan, A.; Iovu, A.; Ungureanu, R.; Cozma, V. Prevalence of endoparasites in peacocks (*Pavo cristatus*). *Sci. Parasitol.* **2009**, *10*, 101–105.
10. Papini, R.; Girivetto, M.; Marangi, M.; Mancianti, F.; Giangaspero, A. Endoparasite infections in pet and zoo birds in Italy. *Sci. World J.* **2012**, *2012*, 253127. [CrossRef]
11. Jaiswal, A.K.; Sudan, V.; Shanker, D.; Kumar, P. Endoparasitic infections in Indian peacocks (*Pavo cristatus*) of Veterinary College Campus, Mathura. *J. Parasit. Dis.* **2013**, *37*, 26–28. [CrossRef]
12. Prakashbabu, B.C.; Thenmozhi, V.; Limon, G.; Kundu, K.; Kumar, S.; Garg, R.; Clark, E.L.; Srinivasa Rao, A.S.R.; Raj, D.G.; Raman, M.; et al. *Eimeria* species occurrence varies between geographic regions and poultry production systems and may influence parasite genetic diversity. *Vet. Parasitol.* **2017**, *233*, 62–72. [CrossRef]
13. Lolli, S.; Grilli, G.; Ferrari, L.; Ferrari, P.; Ferrante, V. Effect of range use on endo- and ectoparasite infestation in italian organic egg production. *Ital. J. Anim. Sci.* **2019**, *18*, 690–695. [CrossRef]
14. Carrisosa, M.; Jin, S.; McCrea, B.A.; Macklin, K.S.; Dormitorio, T.; Hauck, R. Prevalence of select intestinal parasites in Alabama backyard poultry flocks. *Animals* **2021**, *11*, 939. [CrossRef]
15. Blake, D.P.; Knox, J.; Dehaeck, B.; Huntington, B.; Rathinam, T.; Ravipati, V.; Ayoade, S.; Gilbert, W.; Adebambo, A.O.; Jatau, I.D.; et al. Re-calculating the cost of coccidiosis in chickens. *Vet. Res.* **2020**, *51*, 115. [CrossRef]
16. Ilić, T.; Becskei, Z.; Gajić, B.; Özvegy, J.; Stepanović, P.; Nenadović, K.; Dimitrijević, S. Prevalence of endoparasitic infections of birds in zoo gardens in Serbia. *Acta Parasitol.* **2018**, *63*, 134–146. [CrossRef]
17. Valadão, M.C.; Vieira, Í.S.; Millena de Carvalho, L.; Neves, P.H.; Magalhães, R.T.; Campos, A.K.; Araújo, J. Gastrointestinal helminth parasites of *Gallus gallus* in extensive system in the city of Viçosa, Minas Gerais, Brazil. *Braz. J. Vet. Med.* **2021**, *43*, e002121. [CrossRef]
18. Jansson, D.S.; Christensson, D. Gastrointestinala parasiter hos strutsfåglar i Sverige. *Sven. Vet. Tidn.* **2000**, *52*, 621–626.
19. Ponce Gordo, F.; Herrera, S.; Castro, A.T.; García Durán, B.; Martínez Díaz, R.A. Parasites from farmed ostriches (*Struthio camelus*) and rheas (*Rhea americana*) in Europe. *Vet. Parasitol.* **2002**, *107*, 137–160. [CrossRef]
20. McKenna, P.B. *Libyostrongylus* infections in ostriches—A brief review with particular reference to their detection in New Zeland. *N. Z. Vet. J.* **2005**, *53*, 267–270. [CrossRef]
21. Kummrow, M.S. Ratites or Struthioniformes: Struthiones, Rheae, Cassuarii, Apteryges (Ostriches, Rheas, Emus, Cassowaries, and Kiwis), and Tinamiformes (Tinamous). In *Fowler's Zoo and Wild Animal Medicine*; Eric Miller, R., Fowler, M.E., Eds.; Elsevier: Amsterdam, The Netherlands, 2015; Volume 8, pp. 75–82.

22. Ederli, N.B.; Rodrigues de Oliveira, F.C. Gastrointestinal nematodes in ostriches, *Struthio camelus*, in different regions of the state of Rio de Janeiro, Brazil. *Braz. J. Vet. Parasitol.* **2015**, *24*, 168–173. [CrossRef]
23. Köhler, P. The biochemical basis of anthelminthic action and resistance. *Int. J. Parasitol.* **2001**, *31*, 336–345. [CrossRef]
24. Beynon, S.A. Potential environmental consequences of administration of anthelmintics to sheep. *Vet. Parasitol.* **2012**, *189*, 113–124. [CrossRef] [PubMed]
25. Noack, S.; Chapman, H.D.; Selzer, P.M. Anticoccidial drugs of the livestock industry. *Parasitol. Res.* **2019**, *118*, 2009–2026. [CrossRef]
26. Selzer, P.M.; Epe, C. Antiparasitics in Animal Health: Quo Vadis? *Trends Parasitol.* **2021**, *37*, 77–89. [CrossRef]
27. Araújo, J.V.; Braga, F.R.; Mendoza de Gives, P.; Paz-Silva, A.; Vilela, V.L.R. Recent Advances in the Control of Helminths of Domestic Animals by Helminthophagous Fungi. *Parasitologia* **2021**, *1*, 168–176. [CrossRef]
28. Canhão-Dias, M.; Paz-Silva, A.; Madeira de Carvalho, L.M. The efficacy of predatory fungi on the control of gastrointestinal parasites in domestic and wild animals—A systematic review. *Vet. Parasitol.* **2020**, *283*, 109173. [CrossRef]
29. Braga, F.R.; Araújo, J.V. Nematophagous fungi for biological control of gastrointestinal nematodes in domestic animals. *Appl. Microbiol. Biotechnol.* **2014**, *98*, 71–82. [CrossRef]
30. Madeira de Carvalho, L.M.; Bernardo, F.A.; Paz-Silva, A. The role of fungi in the control of animal parasites—classification, mode of action and practical applications. In *Fungi: Types, Environmental Impact and Role in Disease*; Paz-Silva, A., Vázquez, M.S.A., Eds.; Nova Science Publishers: Hauppauge, NY, USA, 2012; pp. 271–308.
31. Madeira de Carvalho, L.M.; Serra, P.M.; Bernardo, F.A.; Agrícola, R.; Jorge, H.; Farrim, A.P.; Fazendeiro, I.M.; Paz-Silva, A. Controlo Integrado da Estrongilidose Equina com Anti-Helmínticos Associados ao Fungo *Duddingtonia flagrans*: Aspectos da sua Utilização em Portugal. *Acta Parasitol. Port.* **2011**, *18*, 63–90.
32. Healey, K.; Lawlora, C.; Knox, M.R.; Chambers, M.; Lamb, J.; Groves, P. Field evaluation of *Duddingtonia flagrans* IAH 1297 for the reduction of worm burden in grazing animals: Pasture larval studies in horses, cattle and goats. *Vet. Parasitol.* **2018**, *258*, 124–132. [CrossRef]
33. Branco de Oliveira, L.S.S.C.; Dias, F.G.S.; Melo, A.L.T.; Millena de Carvalho, L.; Silva, E.N.; Araújo, J.V. Bioverm® in the Control of Nematodes in Beef Cattle Raised in the Central-West Region of Brazil. *Pathogens* **2021**, *10*, 548. [CrossRef]
34. Palomero, A.M.; Cazapal-Monteiro, C.F.; Viña, C.; Hernández, J.; Voinot, M.; Vilá, M.; Silva, M.I.; Paz-Silva, A.; Sánchez-Andrade, R.; Arias, M.S. Formulating fungal spores to prevent infection by trichostrongylids in a zoological park: Practical approaches to a persisting problem. *Biol. Control* **2021**, *152*, 104466. [CrossRef]
35. Voinot, M.; Bonilla, R.; Sousa, S.; Sanchís, J.; Canhão-Dias, M.; Delgado, J.R.; Lozano, J.; Sánchez-Andrade, R.; Arias, M.S.; Madeira de Carvalho, L. Control of Strongyles in First-Season Grazing Ewe Lambs by Integrating Deworming and Thrice-Weekly Administration of Parasiticidal Fungal Spores. *Pathogens* **2021**, *10*, 1338. [CrossRef] [PubMed]
36. Saumell, C.; Fernández, A.; Echevarria, F.; Gonçalves, I.; Iglesias, L.; Sagües, M.; Rodríguez, E. Lack of negative effects of the biological control agent *Duddingtonia flagrans* on soil nematodes and other nematophagous fungi. *J. Helminthol.* **2016**, *90*, 706–711. [CrossRef]
37. Hernández, J.; Arroyo, F.L.; Suárez, J.; Cazapal-Monteiro, C.F.; Romasanta, Á.; López-Arellano, M.E.; Pedreira, J.; Madeira de Carvalho, L.M.; Sánchez-Andrade, R.; Arias, M.S.; et al. Feeding horses with industrially manufactured pellets with fungal spores to promote nematode integrated control. *Vet. Parasitol.* **2016**, *229*, 37–44. [CrossRef] [PubMed]
38. Soto-Barrientos, N.; Oliveira, J.; Vega-Obando, R.; Montero-Caballero, D.; Vargas, B.; Hernández-Gamboa, J.; Orozco-Solano, C. In-vitro predatory activity of nematophagous fungi from Costa Rica with potential use for controlling sheep and goat parasitic nematodes. *Rev. Biol. Trop.* **2011**, *59*, 37–52. [CrossRef] [PubMed]
39. Falbo, M.K.; Soccol, V.T.; Sandini, I.E.; Vicente, V.A.; Robl, D.; Soccol, C.R. Isolation and characterization of the nematophagous fungus *Arthrobotrys conoides*. *Parasitol. Res.* **2013**, *112*, 177–185. [CrossRef]
40. Ojeda-Robertos, N.F.; Aguilar-Marcelino, L.; Olmedo-Juárez, A.; Luna-Palomera, C.; Peralta-Torres, J.A.; López-Arellano, M.E.; Mendoza de Gives, P. In vitro predatory activity of nematophagous fungi isolated from water buffalo feces and from soil in the Mexican southeastern. *Rev. Bras. Parasitol. Vet.* **2019**, *28*, 314–319. [CrossRef]
41. Arroyo-Balán, F.; Landeros-Jaime, F.; González-Garduño, R.; Cazapal-Monteiro, C.; Arias-Vázquez, M.S.; Aguilar-Tipacamú, G.; Esquivel-Naranjo, E.U.; Mosqueda, J. High Predatory Capacity of a Novel *Arthrobotrys oligospora* Variety on the Ovine Gastrointestinal Nematode *Haemonchus contortus* (Rhabditomorpha: Trichostrongylidae). *Pathogens* **2021**, *10*, 815. [CrossRef]
42. Ocampo-Gutiérrez, A.Y.; Hernández-Velázquez, V.M.; Aguilar-Marcelino, L.; Cardoso-Taketa, A.; Zamilpa, A.; López-Arellano, M.E.; González-Cortázar, M.; Hernández-Romano, J.; Reyes-Estebanez, M.; Mendoza de Gives, P. Morphological and molecular characterization, predatory behaviour and effect of organic extracts of four nematophagous fungi from Mexico. *Fungal Ecol.* **2021**, *49*, 101004. [CrossRef]
43. Hernández, J.A.; Vázquez-Ruiz, R.A.; Cazapal-Monteiro, C.F.; Valderrábano, E.; Arroyo, F.L.; Francisco, I.; Miguélez, S.; Sánchez-Andrade, R.; Paz-Silva, A.; Arias, M.S. Isolation of Ovicidal Fungi from Fecal Samples of Captive Animals Maintained in a Zoological Park. *J. Fungi.* **2017**, *3*, 29. [CrossRef]
44. Liu, W.; Han, Y.; Wang, B.-B.; Sun, L.-J.; Chen, M.-Y.; Cai, K.-Z.; Li, X.; Zhao, M.-W.; Xu, C.-L.; Xu, Q.; et al. Isolation, identification, and characterization of the nematophagous fungus *Monacrosporium salinum* from China. *J. Basic Microbiol.* **2015**, *55*, 992–1001. [CrossRef] [PubMed]

45. Xue, Y.-J.; Li, E.-L.; Jing, C.-X.; Ma, L.; Cai, K.-Z. Isolation, identification and characterization of the nematophagous fungus *Arthrobotrys* (*Monacrosporium*) *sinense* from China. *Acta Parasitol.* **2018**, *63*, 325–332. [CrossRef] [PubMed]
46. Larsen, M.; Faedo, M.; Waller, P.J. The potential of nematophagous fungi to control the free-living stages of nematode parasites of sheep: Survey for the presence of fungi in fresh faeces of grazing livestock in Australia. *Vet. Parasitol.* **1994**, *53*, 275–281. [CrossRef]
47. Faedo, M.; Larsen, M.; Waller, P.J. The potential of nematophagous fungi to control the free-living stages of nematode parasites of sheep: Comparison between Australian isolates of *Arthrobotrys* spp. and *Duddingtonia flagrans*. *Vet. Parasitol.* **1997**, *72*, 149–155. [CrossRef]
48. Gray, N.F.; Smith, R.I.L. The distribution of nematophagous fungi in the maritime Antarctic. *Mycopathologia* **1984**, *85*, 81–92. [CrossRef]
49. Braga, F.R.; Araújo, J.V.; Tavela, A.O.; Vilela, V.L.R.; Soares, F.E.F.; Araujo, J.M.; Magalhães, L.Q.; Ferreira da Silveira, W.; Feitosa, T.F.; Dantas, E.S.; et al. First report of interaction of nematophagous fungi on *Libyostrongylus douglassii* (Nematoda: Trichostrongylidae). *Rev. Bras. Parasitol. Vet.* **2013**, *22*, 147–151. [CrossRef]
50. Thapa, S.; Mejer, H.; Thamsborg, S.M.; Lekfeldt, J.D.S.; Wang, R.; Jensen, B.; Magid, J.; Meylingb, N.V. Survival of chicken ascarid eggs exposed to different soil types and fungi. *Appl. Soil Ecol.* **2017**, *121*, 143–151. [CrossRef]
51. Silva, M.E.; Ferreira da Silveira, W.; Braga, F.R.; Araújo, J.V. Nematicide activity of microfungi (*Orbiliales*, *Orbiliaceae*) after transit through gastrointenstinal tract of "*Gallus gallus domesticus*". *Rev. Bras. Saúde Prod. Anim.* **2017**, *18*, 1–9. [CrossRef]
52. Valadão, M.C.; Millena de Carvalho, L.; Vieira, Í.S.; Neves, P.H.; Ferreira, V.M.; Campos, A.K.; Soares, F.E.F.; Ferraz, C.M.; Vilela, V.L.R.; Braga, F.R.; et al. Germination capacity of the *Pochonia chlamydosporia* fungus after its passage through the gastrointestinal tract of domestic chickens (*Gallus gallus domesticus*). *Exp. Parasitol.* **2020**, *216*, 107936. [CrossRef]
53. Thapa, S.; Thamsborg, S.M.; Wang, R.; Meyling, N.V.; Dalgaard, T.S.; Petersen, H.H.; Mejer, H. Effect of the nematophagous fungus *Pochonia chlamydosporia* on soil content of ascarid eggs and infection levels in exposed hens. *Parasites Vectors* **2018**, *11*, 319. [CrossRef]
54. Carrasco, J.M.D.; Casanova, N.A.; Miyakawa, M.E.F. Microbiota, Gut Health and Chicken Productivity: What Is the Connection? *Microorganisms* **2019**, *7*, 374. [CrossRef] [PubMed]
55. Huang, G.; Tang, X.; Bi, F.; Hao, Z.; Han, Z.; Suo, J.; Zhang, S.; Wang, S.; Duan, C.; Yu, Z.; et al. *Eimeria tenella* infection perturbs the chicken gut microbiota from the onset of oocyst shedding. *Vet. Parasitol.* **2018**, *258*, 30–37. [CrossRef] [PubMed]