Mushrooms as efficient biocontrol agents against the root-knot nematode, *meloidogyne incognita*

Gaziea Soliman[a], Waill Elkhateeb[b], Ting-Chi Wen[c,d,e], Ghoson Daba[b]

[a]Department of Plant Pathology, Nematology Unit, Agriculture and Biology Research Division, [b]Department of Chemistry of Natural and Microbial Products, Pharmaceutical Industries Researches Institute, National Research Centre, Giza, Egypt, [c]State Key Laboratory Breeding Base of Green Pesticide and Agricultural Bioengineering, Key Laboratory of Green Pesticide and Agricultural Bioengineering, [d]The Engineering Research Center of Southwest Bio-Pharmaceutical Resources, Ministry of Education, [e]The Mushroom Research Centre, Guizhou University, Guiyang, China

Correspondence to Waill Elkhateeb, PhD, Department of Chemistry of Natural and Microbial Products, Pharmaceutical Industries Researches Division, National Research Centre, El Buhouth Street, Dokki, Giza 12311, Egypt. Tel: +20 101 324 1936; fax: (+20) 233 387 758;
e-mail: waillahmed@yahoo.com

Received: 20 October 2021
Revised: 30 October 2021
Accepted: 1 November 2021
Published: 7 March 2022

Egyptian Pharmaceutical Journal 2022, 21:68–74

Background
Root-knot nematodes in general and *Meloidogyne incognita*, in particular, are pests that cause agricultural losses. Currently, nematode control relies on chemical nematicides, which are hazardous to the environment and human health. The increasing demand for ecofriendly nematicides has prompted researchers to look into biocontrol agents that act as efficient and long-lasting alternatives to the currently used chemicals.
Objective
The aim of the study was to evaluate the in-vitro nematicidal activity of eight mushroom (*Cordyceps militaris*, *Metacordyceps neogunnii*, *Hericium erinaceus*, *Dictyophora indusiata*, *Cerioporus squamosus*, *Tirmania nivea*, *Tirmania pinoyi*, and *Agaricus impudicus*) extracts against *M. incognita* juveniles and eggs.
Materials and methods
Hydromethanolic extracts were prepared from the fruiting bodies of mushrooms under investigation. Then the obtained extracts were evaluated for their in-vitro nematicidal activity against *M. incognita* juveniles' second stage after 24, 48, and 72 h of treatment, also against their eggs.
Results and conclusion
All tested mushroom extracts were capable of inducing mortality in *M. incognita* second stage juveniles with mortality percentages ranging from 79.3 to 97%. On the other hand, the tested mushroom extracts exhibited some nematostatic and nematicidal activity against *M. incognita* eggs as compared with the control after 7 days using 80 µl/ml concentration. The tested mushroom extracts caused suppression in *M. incognita* eggs hatching with inhibition rates that ranged from 59.38 to 81.25%. *A. impudicus* hydromethanolic extract showed the highest inhibition as compared with the control and other tested mushroom extracts as it caused a relative suppression that reached 81.25% against *M. incognita* eggs after 7 days of exposure. The same mushroom extract has achieved a juveniles mortality of 97%.
A. impudicus extract is nominated as a potential nematicidal agent. Further studies are required to confirm the potency of this extract and analyze its chemical profile.

Keywords:
biocontrol, egg hatching, juveniles, *Meloidogyne incognita*, mushroom extracts, nematicidal

Egypt Pharmaceut J 21:68–74

1687-4315

Introduction

Root-knot nematodes, *Meloidogyne* spp., are serious pests that cause wide-ranging losses tto different crops worldwide, and are ranked among the most difficult agricultural pests to control due to their wide host range, short life cycles, and high reproduction rate [1,2]. *M*eloidogyne *incognita* is a highly polyphagous nematode that affects crops in warmer temperatures and can be found in greenhouses in both hot and temperate countries [3]. In past decades, the use of chemical nematicides was one of the primary means of controlling root-knot nematodes [4]. However, chemical nematicide strategies are losing favor among researchers worldwide due to their high cost, effect on human health, resulting pollution, and serious environmental impacts, which led to searching for alternative approaches for sustainable crop production, control plant diseases, and plant-parasitic nematodes. Biological control is a sustainable alternative to chemical control including using natural and environment-friendly components to reduce plant-parasitic nematodes [5–10]. Mushrooms are generous sources of compounds that can act as anticancer, antioxidant hypocholesterolemic, and wound-healing agents [11–13]. Moreover, many mushrooms have a vital role in the biological control of plant-parasitic nematodes as alternatives to chemical nematicides due to their nematicidal properties, as it is easy to obtain and cultivate, as well as being ecofriendly

and sometimes edible [14,15]. The bioactive compounds produced by the mushrooms have a selective effect, preserving nontarget species, implying the possibility of developing environmentally friendly goods [16,17]. *Cordyceps militaris* (orange caterpillar mushroom) and *Metacordyceps neogunnii* are entomogenous medicinal mushrooms [12,18,19]. However, few studies are available reporting the biological activities of *M. neogunnii* extract. On the contrary, *C. militaris* is a well-studied mushroom that can be cultivated artificially and hence its bioactive compounds can be obtained in good quantities [20]. *Hericium erinaceus* is another well-known medicinal mushroom that exerts various biological activities such as antimicrobial, antiproliferative, antitumor, hemagglutinating, hypoglycemic, antiaging, and immunomodulatory effects [12]. *Dictyophora indusiata* is also a well-studied medicinal mushroom that is known centuries ago and has potential use as a functional food [21,22]. Both *Tirmania nivea* and *Tirmania pinoyi* are edible desert truffles (ectomycorrhizal wild mushrooms) that grow under certain environmental conditions and exist in different surrounding locations including the north coast of Egypt, Saudi Arabia, and Yemen. They are ethnically edible for their medical and nutritional values [23,24]. On the other hand, *Cerioporus squamosus* (dryad's saddle or pheasant's back mushroom) is a medicinal mushroom that has been studied and its extract showed antimicrobial, antioxidant, anticancer, and wound-healing properties [25]. The last mushroom investigated in this study is the basidiomycetes mushroom *Agaricus impudicus* (tufted wood mushroom). Studies published on this mushroom mostly discuss its habitat, taxonomy, and morphology [26]. However, publications describing the bioactivity of its extract are relatively rare.

The study aims to evaluate the *in vitro* nematicidal activity of the hydromethanolic extracts of the mushrooms *C. militaris*, *M. neogunnii*, *H. erinaceus*, *D. indusiata*, *C. squamosus*, *T. nivea*, *T. pinoyi*, and *A. impudicus* as biocontrol agents against *M. incognita* mortality and egg hatchability.

Materials and methods

Mushroom sample extraction

Mushrooms tested in this study (*C. militaris*, *M. neogunnii*, *H. erinaceus*, *D. indusiata*, *C. squamosus*, *T. nivea*, *T. pinoyi*, and *A. impudicus*) (Fig. 1) were provided by Prof. Dr Waill Elkhateeb, Prof. Dr Ting-Chi Wen, and Prof. Dr Abdu ALKolaibe. Identification of samples was conducted by Prof. Dr Waill Elkhateeb and Prof. Dr Ting-Chi Wen. Hydromethanolic extracts of mushrooms were individually prepared as described by Elkhateeb *et al.* [25]. Briefly, 250 g of fruiting bodies of each mushroom was cut into small pieces (1 cm). Then, the pieces were kept for 2 days in Erlenmeyer flasks that contain 80% methanol at room temperature. Samples were filtered and the resulting filtrates were concentrated at 37°C using a rotary evaporator (Heidolph, Germany). The obtained extracts were stored at 4°C in a clean closed container until further use.

In-vitro nematicidal activity of hydromethanolic extracts of mushrooms

Inoculum of Meloidogyne incognita

The pure population of *M. incognita* was obtained from greenhouse culture on tomato plants maintained at the Plant Pathology Department, National Research Centre, Egypt. After 40 days, the plants were uprooted, and the roots were washed free of soil and cut into 2 cm pieces. The nematode eggs were extracted from infected roots of tomato in 0.5% sodium hypochlorite on a 25-μm sieve according the method of Hussey and Barker [27]. The extracted eggs were used immediately for egg hatching ability test, or incubated for egg hatching for 72 h at the laboratory temperature (25±3°C) using the modified Baermann technique [28] to provide second-stage nematode juveniles (J_2) for juvenile's mortality test. To estimate the inoculum density, the egg suspension was poured into a measuring cylinder. The numbering of eggs was estimated in five aliquots of 1 ml in a Hawksley counting slide under a light microscope and their five means were calculated. For concentrating the egg suspension, it was left to settle down for several hours, afterward the extra water was decanted off leaving the bottom undisturbed and the same was done with J_2 to estimate the inoculum density. The percentage J_2 mortality and inhibition of egg hatch of the root-knot nematode *M. incognita* were used to test the in-vitro nematicidal activity.

Nematicidal activity of mushroom hydromethanolic extracts on Meloidogyne incognita J_2 mortality

The nematicidal activity of hydromethanolic extracts of the eight mushrooms against *M. incognita* juveniles was tested using two concentrations (40 and 80 μl/ml). One milliliter from each concentration had 100±5 *M. incognita* J_2 in it, separately. They were arranged in a completely randomized design at 25±3°C after 24 and 72 h of exposure. All treatments were conducted in five replicates and the average results were compared with

Figure 1

Fruiting bodies used to prepare hydromethanolic extracts of mushrooms: *Agaricus impudicus* (a), *Cordyceps militaris* (b), *Dictyophora indusiata* (c), *Tirmania nivea* (d), *Tirmania pinoyi* (e), *Metacordyceps neogunnii* (f), *Hericium erinaceus* (g), and *Cerioporus squamosus* (h). Photos were taken by Prof. Dr Waill Elkhateeb, Prof. Dr Ting-Chi Wen, and Dr Ghoson Daba.

distilled water as control. Also, after 72 h of exposure, the juveniles were washed in distilled water and transferred to aerated distilled water for 24 h. The juveniles that did not regain their activities and those do not move when probed with a fine needle were considered 'dead.' On the other hand, the juveniles were considered active when they were visibly flexible and then the average percentages of nematode recoveries were determined [29]. The number of active and inactive J_2 was counted with the aid of a microscope and the percentages of inactive larvae were calculated to evaluate the percentage of juveniles' mortality.

The percentages of nematode mortality were calculated according to Abbott's formula [30] as follows:

Juvenile mortality (%)=$(m-n)/(100-n)\times100$, where m and n indicate the percentages of mortality in treatments and control, respectively.

Effect of mushroom extracts on the inhibition of Meloidogyne incognita egg hatch

In this experiment, the direct effect of the eight mushroom hydromethanolic extracts on *M. incognita* egg suppression was investigated at one concentration (80 µl/ml). One milliliter contain about 100±5 *M. incognita* J_2 with an 80 µl/ml final concentration. Plain water in the test tube supplied with nematode suspension was served as control. Treatments were conducted in five biological replicates compared with the negative control. The test tubes were kept at room temperature 25±3°C. Hatching eggs were examined, after 7 days of exposure; the number of hatched eggs was counted under a stereomicroscope by using a special microscopic slide for counting nematodes. After 1 week, the total number of J_2 was calculated. The relative suppression rate was calculated as follows: relative suppression rate (%)=(number of J_2 in control–number of J_2 in treatment)/number of J_2 in control×100.

Data analysis

Data obtained from laboratory bioassays for hatched eggs and mortality (%) were subjected to analysis of variance and means compared according to Duncan's multiple range test [31].

Results

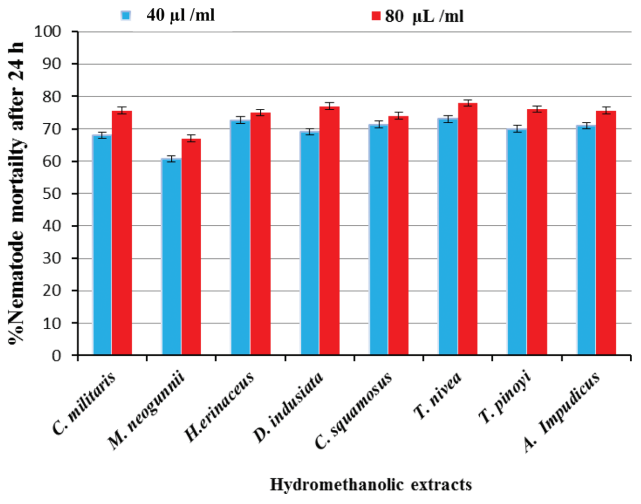
Nematicidal potential of the mushroom species

The nematicidal activity of eight mushroom extract species against *M. incognita* juveniles were tested at two concentrations (40 and 80 µl/ml) in *in vitro* was tests. The obtained results, after 24 h of exposure, revealed that all mushroom extracts caused significant mortality in *M. incognita* J_2 compared with control. The percentage of mortality ranged between 67 and 78.00% at 80 µl/ml concentration in all mushroom extracts, while the untreated control achieved zero. *T. nivea* achieved the highest percentage of mortality, 73.00 and 78.00%, at two concentrations, 40 and 80 µl/ml, respectively (Fig. 2). After 48 h, the results showed that the tested concentrations were extremely hazardous to nematode juveniles. *T. pinoyi* extract had the highest mortality of 88.67% when tested at 80 µl/ml. After 72 h of exposure, all mushroom extracts cause significant mortality in *M. incognita* J_2 as compared with control. The percentage of mortality ranged between 79.3 and 97% on using 80 µl/ml concentration in all mushroom extracts tested. *C. militaris* had the highest percentage of mortality, 92.0 and 97%, at two concentrations of 40 and 80 µl/ml, respectively (Figs 3 and 4). The percentage of nematode mortality differed with concentrations of mushroom extracts and exposure time. The percentage mortality of second-stage juveniles (J_2) exposed to highly concentrated mushroom extracts was markedly greater than that of those exposed to low-concentration mushroom extracts and controls after 24, 48, and 72 h of incubation. Microscopic investigation after treatment with extracts of *C. militaris*, *M. neogunnii*, *H. erinaceus*, *D. indusiata*, *C. squamosus*, *T. nivea*, *T. pinoyi*, and *A. impudicus* revealed no changes in the nematode bodies of juveniles treated with mushroom extracts (data not shown).

Inhibitory activity of mushroom extracts on *Meloidogyne incognita* eggs

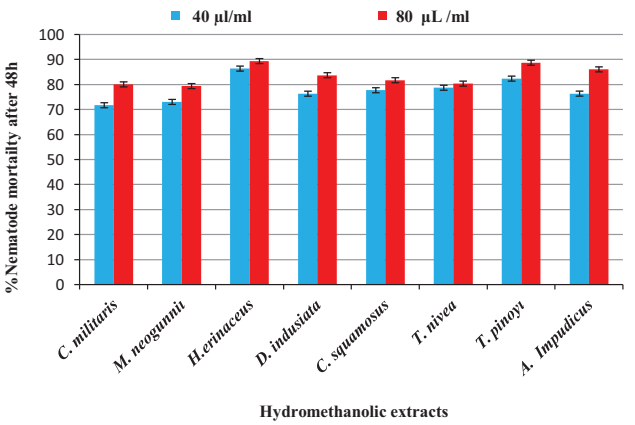
In the in-vitro experiments, all the tested mushroom hydromethanolic extracts had some nematostatic and

Figure 2



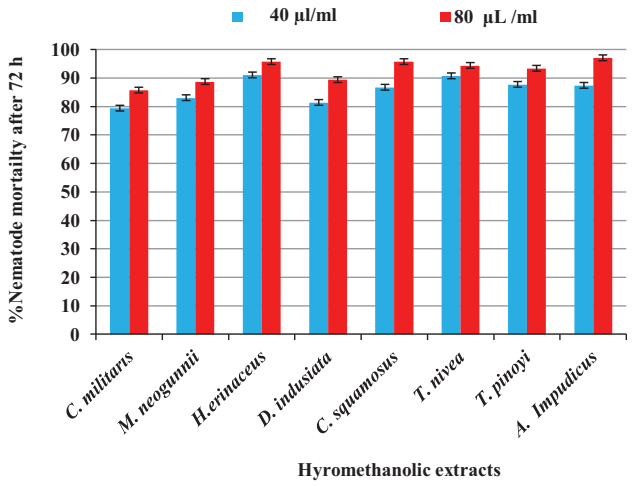
Effect of hydromethanolic extracts of mushrooms on juvenile mortality of *Meloidogyne incognita* after 24 h. Error bars represent mean ±SD.

Figure 3



Effect of hydromethanolic extracts of mushrooms on juvenile mortality of *Meloidogyne incognita* after 48 h. Error bars represent mean ±SD.

Figure 4



Effect of hydromethanolic extracts of mushrooms on juvenile mortality of *Meloidogyne incognita* after 72 h. Error bars represent mean ±SD.

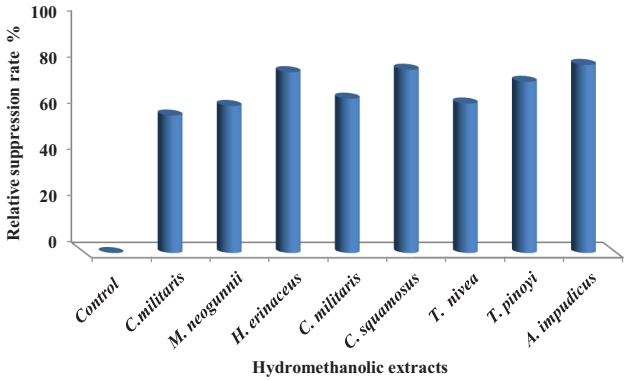
Table 1 Inhibition activity of mushroom extracts against *Meloidogyne incognita* eggs after 7 days of exposure

Treatments	Number of J_2 (hatched eggs)	Relative suppression rate %**
Control	96a*	0.00
Cordyceps militaris	39b	59.38
Metacordyceps neogunnii	35c	63.52
Hericium erinaceus	21f	78.12
Dictyophora indusiata	32d	66.67
Cerioporus squamosus	20f	79.17
Tirmania nivea	34c	64.58
Tirmania pinoyi	25e	73.96
Agaricus impudicus	18g	81.25

*Means followed by the same letter(s) are not significantly different according to Duncan's multiple range test. Data are averages of five replicates. **Relative suppression rate (%) is calculated as mentioned in materials and methods.

nematicidal activity against *M. incognita* eggs as compared with the control. Generally, different mushroom extracts remarkably suppressed the hatching of *M. incognita* eggs. However, inhibition rates ranged from 59.38 to 81.25% after 1 week of exposure. Data presented in Table 1 revealed that *A. impudicus* caused the greatest inhibitory effect (81.25%) followed by *C. squamosus* (79.17%), *H. erinaceus* (78.12%), *C. militaris* (66.67), *T. nivea* (64.58%), *M. neogunnii* (63.52%), and *C. militaris* (59.38%), after 7 days of exposure as compared with control (Table 1 and in Fig. 5).

Figure 5



Effect of hydromethanolic extracts of mushrooms on suppression hatchability of *Meloidogyne incognita* eggs.

Discussion

M. incognita is one of the most destructive and widely distributed plant-parasitic nematodes attacking roots of different economic crops such as sugarcane, okra, potatoes, tobacco, tomato, and cotton [32]. As much as 40% yield losses in Indian tomato was caused by *M. incognita*. In 2015, *M. incognita* caused losses in American cotton yield that reached 1.35% of total cotton production [33]. The use of chemicals for controlling pests, and their impact on human health and the environment had forced scientists to search for ecofriendly alternatives to face the economic loss caused by pests without polluting the environment and compromising the whole ecosystem. Hence, screening for biocontrol solutions is of serious importance to fight such harmful pests.

In this study, we have evaluated the antinemic effect of some mushrooms' hydromethanolic extracts [34]. Microscopic investigation after treatment with extracts of *C. militaris*, *M. neogunnii*, *H. erinaceus*, *D. indusiata*, *C. squamosus*, *T. nivea*, *T. pinoyi*, and *A. impudicus* revealed no changes in the nematode bodies of juveniles treated with mushroom extracts. This indicates that mushroom extracts could have the ability to paralyze and kill plant nematode juveniles. This action could be related to the production of paralyzing compounds existing in these extracts. Similar findings were observed [34], where the mushroom *Pleurotus cystidiosus* secretes paralyzing toxins that affect the mobility of nematodes. Generally, mushrooms secrete various secondary metabolites which have nematotoxic activity [35–38]. For example, the total number of biopesticides isolated from basidiomycetes is considerably high [39]. Many *Pleurotus*-originated fatty acids were also reported for their nematicidal

action [17]. This can be causing the nematicidal effect recorded for *C. militaris*, *M. neogunnii*, *H. erinaceus*, and *D. indusiata* where fatty acids represented the majority of compounds in their chemical profiles [12,19]. On the other hand, the mushroom *Pleurotus ostreatus* was reported to produce a nematode toxin similar to peroxides that killed 95% of free-living adult worms of *Panagrellus redivivus* and phytopathogenic nematode *Bursaphelenchus xylophilus* [40]. Also, many antioxidants are produced by wild mushrooms, including phenolic acids, flavonoids, and ascorbic acid [41]. It should be noted that there is a positive relationship between phenolic compounds and nematode mortality [42–44]. The hatching inhibition potential and nematicidal effect of mushroom extracts on *M. incognita* eggs were confirmed in in-vitro experiments. Mushroom extracts and cell-free culture broth contain some bioactive compounds such as polysaccharides, proteins, fats, minerals, glycosides, alkaloids, volatile oils, phenolics, and flavonoids are reported to possess antinemic effects against plant nematodes. Accordingly, the antinemic impact of mushroom extracts or filtrate on eggs of *M. incognita* could be attributed to the intrinsic nature of toxic metabolites such as phenols and fatty acids.

In light of the current difficulties in managing plant-parasitic nematodes, extracts of mushrooms could be a potential management strategy for controlling plant nematodes. Sangeetha *et al.* [45] found that the bioactive compounds of *Ophiocordyceps sinensis* inhibited egg hatching (94%) and juvenile mortality (92%) in *M. incognita* after 72 h of incubation. Similarly, the ethanolic extract of *P. ostreatus* exerted high nematicidal activity against *Meloidogyne* spp. that reached 99.1% after 1 h of treatment. On the other hand, aqueous extracts of the mushrooms *Boletus* sp., *Lactarius deliciosus* and *Amanita muscaria* caused mortality rates that ranged between 90.7 and 100.0% after 1 day of exposure [46]. In another in-vitro study, 10 mushroom aqueous extracts suppressed the hatching of second-stage nematode juveniles [47,48]. In the laboratory, the root-knot nematode, *M. incognita* juveniles were reduced by using aqueous extract of the mushroom *P. ostreatus* [15]. The use of bioactive compounds from a luminous mushroom (*Neonothopanus nambi*) for the control of plant-parasitic root-knot nematode *M. incognita* was also described [16].

Conclusions

Novel ecofriendly biocontrol agents are urgently needed to replace the currently utilized toxic nematicides, if we are to avoid further damage to the earth's ecosystems. Mushrooms are a potential alternative to these chemicals. They have also been found to protect against plant-parasitic nematodes. Results showed in the current study nominate extracts of *A. impudicus*, *C. squamosus*, and *H. erinaceus* as the most potent extracts in terms of increased percentage mortality of juveniles and reduced hatchability of eggs. After 72 h, juvenile mortality achieved 97, 95.67, and 95.67%, respectively, while, suppression rate of eggs for the same species extracts recorded 81.25, 79.17, and 78.12%, respectively, after 1 week. While these findings are encouraging, further field and mechanistic studies and chemical analyses are required to confirm the potency and mechanism of action of these extracts as nematicidal agents.

Acknowledgements

The authors thank Prof. Dr Abdu Ghalib ALKolaibe, Microbiology Department, Faculty of Science, Taiz University, Yemen, for providing some of the mushroom samples.

Availability of data and materials: The data that support the findings of this study are available from the corresponding author, Waill Elkhateeb, on reasonable request.

Authors' contributions: All the authors have read and approved the final manuscript. W.E. and G.D. have substantially contributed to the conception and design of the work. W.E.: collection, identification, and extraction of mushroom samples. T.C.W.: collection and identification of mushroom samples. G.S.: execution of nematodal experiments, analysis, and interpretation of results, writing and reviewing of the manuscript. G.D.: extraction of mushroom samples, writing, and reviewing of the manuscript.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1 Huang W, Sun J, Cui J, Wang G, Kong L, Peng H, *et al.* Efficacy evaluation of fungus *Syncephalastrum racemosum* and nematicide avermectin against the root-knot nematode *Meloidogyne incognita* on cucumber. PLoS ONE 2014; 9:e89717.

2 Archidona-Yuste A, Cantalapiedra-Navarrete C, Lie'banas G, Rapoport HF, Castillo P, Palomares-Rius JE. Diversity of root-knot nematodes of the genus *Meloidogyne* Goeldi, 1892 (Nematoda: Meloidogynidae) associated with olive plants and environmental cues regarding their distribution in southern Spain. PLoS ONE 2018; 13:e0198236.

3 Hallmann J, Kiewnick S. Virulence of *Meloidogyne incognita* populations and *Meloidogyne enterolobii* on resistant cucurbitaceous and solanaceous plant genotypes. J Plant Dis Prot 2018; 125:415–424.

4 Baidoo R, Mengistu T, McSorley RR, Stamps H, Brito J, William TC. Management of root-knot nematode (*Meloidogyne incognita*) on *Pittosporumtobira* under greenhouse, field, and on-farm conditions in Florida. J Nematol 2017; 49:133–139.

5 Qui SJ, Can JY, Liu WP, Becker JO. Degradation and absorption of fosthiazate in soil. J Agric Food Chem 2004; 52:6239–6242.

6 Haydock PP, Woods SR, Grove IG, Hare MC, Perry RN, Moens M. Chemical control of nematodes. In: Perry RN, Moens M, editors. Plant nematology. Oxford shire, UK: CABI Publishing 2006. 392–410.

7 Tanweer A, Hisamuddin A. Evaluation of *Cassia tora* for control of root-knot nematode (*Meloidogyne incognita*) in soybean. Afr J Basic Appl Sci 2012; 4:21–24.

8 Le Mire G, Nguyen ML, Fassotte B, du Jardin P, Verheggen F, Delaplace P, Jijakli H. Implementing plant biostimulants and biocontrol strategies in the agroecological management of cultivated ecosystems, a review. Biotechnol Agron Soc Environ 2016; 20:299–313.

9 Dahlin P, Hallmann J. New insights on the role of allyl is thiocyanate in controlling the root knot nematode *Meloidogyne hapla*. Plants 2020; 9:603.

10 Ramadan WA, Soliman GM. Effect of different applications of bio-agent *Achromobacter xylosoxidans* against *Meloidogyne incognita* and gene expression in infected eggplant. Jordan J Biol Sci 2020; 13:363–370.

11 Elkhateeb WA, Daba GM, Wen TC, Thomas PW. Medicinal mushrooms as a new source of natural therapeutic bioactive compounds. Egypt Pharma J 2019; 18:88–101.

12 Daba GM, Elkhateeb WA, EL-Dein AN, Ahmed EF, EL Hagrassi AM, Fayad W, Wen TC. Therapeutic potentials of n-hexane extracts of the three medicinal mushrooms regarding their anti-colon cancer, antioxidant, and hypocholesterolemic capabilities. Biodiversitas 2020; 21:2437–2445.

13 Zhang X, Zhang X, Gu S, Pan L, Sun H, Gong E, *et al.* Structure analysis and antioxidant activity of polysaccharide-iron (III) from *Cordyceps militaris* mycelia. Int J Biol Macromol 2021; 178:170–179.

14 Elkhateeb WA, Daba GM, Soliman GM. The anti-nemic potential of mushroom against plant-parasitic nematodes. J Microbiol Biotechnol 2021; 6:1–6.

15 Youssef MMA, El-Nagdi WMA. New approach for biocontroling root-knot nematode, *Meloidogyne incognita* on cowpea by commercial fresh oyster mushroom (*Pleurotus ostreatus*). Jordan J Biol Sci 2021; 14:173–177.

16 Bua-art S, Saksirirat W, Hiransalee A, Kanokmedhaku S, Lekphrom R. Effect of bioactive compound from luminescent mushroom (*Neonothopanus nambi* Speg.) on root-knot nematode (*Meloidogyne incognita* Chitwood) and non-target organisms. KKU Res J 2011; 16:331–341.

17 Li G, Wang X, Zheng L, Li L, Huang R, Zhang K. Nematicidal metabolites from the fungus *Pleurotus* ferulae Lenzi. Ann Microbiol 2007; 57:527–529.

18 Wen TC, Xiao YP, Han YF, Huang SK, Zha LS, Hyde KD, Kang JC. Multigene phylogeny and morphology reveal that the Chinese medicinal mushroom *Metacordyceps neogunnii*. nov. Phytotaxa 2017; 302:27–39.

19 El-Hagrassi A, Daba G, Elkateeb W, Ahmed E, Negm El Dien A, Fayad W, *et al.* Invitro bioactive potential and chemical analysis of the n-hexane extract of the medicinal mushroom, Cordyceps militaris. Malays J Microbiol 2020; 16:40–48.

20 Yang CH, Kao YH, Huang KS, Wang CY, Lin LW. *Cordyceps militaris* and mycelial fermentation induced apoptosis and autophagy of human glioblastoma cells. Cell Death Dis 2012; 3:e431–e431.

21 Han S, Ma C, Hu M, Wang Y, Ma F, Tao N, Qin Z. A polysaccharide from *Dictyophora indusiata* inhibits the immunosuppressive function of cancer-associated fibroblasts. Cell Biochem Funct 2017; 35:414–419.

22 Elkhateeb WA, Elnahas MO, Thomas PW, Daba GM. *Trametes versicolor* and *Dictyophora indusiata* champions of medicinal mushrooms. J Pharma Res 2020; 4:1–7.

23 Daba GM, Elkhateeb WA, Wen TC, Thomas PW. The continuous story of truffles plant interaction. In: Kumar V, Prasad R, Kumar M, Choudhary DK (editors). Microbiome in plant health and disease – challenges and opportunities. Singapore: Springer 2019; 375–383

24 Thomas P, Elkhateeb WA, Daba GM. Truffle and truffle-like fungi from continental Africa. Acta Mycol 2019; 54:1–15.

25 Elkhateeb WA, Daba GM, Elnahas MO, Thomas PW, Emam M. Metabolic profile and skin-related bioactivities of *Cerioporus squamosus* hydromethanolic extract. Biodiversitas 2020; 21:4732–4740.

26 Cho DH, Chung JY. Study of fungal diversity of Mt. Cheonggye. Korean J Nature Conserv 2020; 19:29–60.

27 Hussey RS, Barker KR. A comparison of methods of collecting inocula of *Meloidogyne* spp., including a new technique. Plant Dis Rep 1973; 57:1025–1028.

28 Christie JR, Perry VG. Removing nematodes from soil. Proc Helminthol Soc Washington 1951; 18:106–108.

29 Cayrol JC, Djian C, Pijarowski L. Study of the nematocidal properties of the culture filtrate of the nematophagous fungus *Paecilomyces lilacinus*. Rev Nematol 1989; 12:331–336.

30 Abbott WS. A method of computing the effectiveness of an insecticide. J Econ Entomol 1925; 8:265–267.

31 Duncan DB. Multiple ranges and multiple F test. Biometrics 1955; 11:11–24.

32 Warmerdam S, Sterken MG, Van Schaik C, Oortwijn ME, Lozano-Torres JL, Bakker J, *et al.* Mediator of tolerance to abiotic stress ERF6 regulates susceptibility of Arabidopsis to *Meloidogyne incognita*. Mol Plant Pathol 2019; 20:137–152.

33 Lawrence KS, Lawrence GW, Faske T, Overstreet C, Wheeler T, Young H, *et al.* Beltwide nematode research and education committee 2015 nematode research report cotton varietal and nematicide responses in nematode soils. In: Proc. Beltwide Cotton Conf. Memphis, TN: National Cotton Council; 2016; 1:113–115.

34 Luo H, Mo M, Huang X, Li X, Zhang K. *Coprinus comatus*, a basidiomycete fungus forms novel spiny structures and infects nematode. Mycologia 2004; 96: 1218–1225.

35 Dong LQ, Zhang KQ. Microbial control of plant-parasitic nematodes, a five-party interaction. Plant Soil 2006; 288:31–45.

36 Degenkolb T, Vilcinskas A. Metabolites from nematophagous fungi and nematicidal natural products from fungi as alternatives for biological control. Part II: metabolites from nematophagous basidiomycetes and non nematophagous fungi. Appl Microbiol Biotechnol 2016; 100:3813–3824.

37 Tanimola AA, Adedokun OM. Nematicidal potentials of aqueous extracts of two mushroom species and spent mushroom substrate on *Meloidogyne incognita* and *Meloidogyne javanica*. Nig J Agri Food Environ 2020; 16:110–119.

38 Castañeda-Ramírez GS, Torres-Acosta JF, Sánchez JE, Mendoza-de-Gives P, González-Cortázar M, Zamilpa A, *et al.* The possible biotechnological use of edible mushroom bioproducts for controlling plant and animal parasitic nematodes. Biomed Res Int 2020; 2020: 1–12.

39 Lorenzen K, Take T. Basidiomycetes as a source for new bioactive natural products. Curr Organic Chem 1998; 2:329–364.

40 Kwok O, Plattner R, Weisleder D, Wicklow DA. Nematicidal toxin from *Pleurotus ostreatus* NRRL 3526. J Chem Ecol 1992; 18:127–136.

41 Ferreira ICF, Barros L, Abreu RM. Antioxidants in wild mushrooms. Curr Med Chem 2009; 16:1543–1560.

42 Caboni P, Saba M, Tocco G, Casu L, Murgia A, Maxia A, *et al.* Nematicidal activity of mint aqueous extracts against the root-knot nematode *Meloidogyne incognita*. J Agric Food Chem 2013; 61:9784–9788.

43 Faizi S, Fayyaz S, Bano S, Iqbal EY, LubnaSiddiqi H, Naz A. Isolation of nematicidal compounds from *Tagetes patula* L. yellow flowers: structure-activity relationship studies against cyst nematode *Heteroderazeae* infective stage larvae. J Agri Food Chem 2011; 59:9080–9093.

44 Pinho J, Rodrigues A, Dibb S. The role of corporate culture, market orientation, and organizational commitment in organizational performance. J Manage Dev 2014; 33:374–398.

45 Sangeetha C, Krishnamoorthy AS, Ramakrishnan S. Testing bioactive compounds of Chinese caterpillar fungus, *Ophiocordyceps* spp. against root knot nematode (*Meloidogyne incognita*). Res J Agri Sci 2015; 6:1129–1133.

46 Miyazaki S, Kitamura N, Nishio A, Tanaka S, Kayano T, Moryia T, *et al.* Gymnopilin – a substance produced by the hallucinogenic mushrooms, *Gymnopilus junonius* – mobilizes intracellular Ca^{2+} in dorsal root ganglion cells. Biomed Res 2012; 33: 111–118.

47 Wille CN, Gomes CB, Minotto E, Nascimento JS. Potential of aqueous extracts of basidiomycetes to control root-knot nematodes on lettuce. Horticult Brasil 2019; 37:054–059.

48 Balaeş T, Tănase C. Basidiomycetes as potential biocontrol agents against nematodes. Rom Biotechnol Lett 2016; 21:11185–11193.