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Use of *Metarhizium* spp. in the biological control of varroa destructor in colonies of *Apis mellifera*: a systematic literature review

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Abstract. Varroa destructor infestation represents one of the main threats to global beekeeping, causing severe damage to Apis mellifera colonies. Given the increasing resistance of the mite to synthetic acaricides and the environmental impacts associated with these products, biological control has emerged as a promising alternative. This systematic literature review aimed to gather and analyze available studies on the use of entomopathogenic fungi of the genus Metarhizium for the control of V. destructor. Searches were conducted in July 2025 in the Web of Science, PubMed, Scopus, and SciELO databases. Articles written in English, Portuguese, and Spanish that applied Metarhizium spp. directly to A. mellifera colonies were included. Abstracts, literature reviews, books, book chapters, and grey literature were excluded. As a minimum quality criterion, only peer-reviewed articles published in indexed journals were selected. In total, 11 relevant studies were included, published between 2002 and 2021 by authors from eight different countries. The results indicate that Metarhizium spp. exhibit variable efficacy in inducing V. destructor mortality, influenced by factors such as fungal strain, humidity, temperature, application method, and formulation. Despite recent advances, challenges remain regarding the standardization of protocols and the feasibility of field application. It is concluded that Metarhizium spp. show potential for integrated varroa management; however, further studies are needed to assess their effectiveness under field conditions and their effects on bee colonies.

Keywords: bees, entomopatogenic fungi, mite, biocontrol.

Introduction

Varroa destructor Anderson and Trueman, 2000 (Acari: Varroidae), popularly known as varroa, is an ectoparasitic mite that infests Apis mellifera Linnaeus, 1758 (Hymenoptera: Apidae), and Apis cerana Fabricius, 1793 (Hymenoptera: Apidae). It has historically been the greatest threat to beekeeping(Rosenkranz; Aumeier; Ziegelmann, 2010), decimating honey bee populations worldwide(Le Conte; Ellis; Ritter, 2010).

Previously, this mite was classified as jacobsoni Varroa Oudemans, 1904 (Acari: Varroidae), Anderson and Trueman but (2000)discovered that V. jacobsoni was actually a complex of at least two distinct species and proposed that the new species be named "Varroa destructor", while V. jacobsoni sensu stricto remained restricted to parasitic populations of A.

Historically, *V. destructor* had *A. cerana* bees as its natural host(Celikkol; Dogac, 2025) and

began to infest *A. mellifera* bees when they were moved to areas where *A. cerana* is endemic (Le Conte; Ellis; Ritter, 2010). The mite infests everything from adult bees to capped brood, sucking their hemolymph, which can interfere with their development and eventually decimate the entire *A. mellifera* colony when the infestation rate becomes too high(Rosenkranz; Aumeier; Ziegelmann, 2010).

For mite control, synthetic acaricides are conventionally used, such as amitraz from the formamidine group, the pyrethroids fluvalinate and flumethrin, and the organophosphate coumaphos(Maggi *et al.*, 2008). However, *V. destructor* is becoming resistant to several of these acaricides (Celikkol; Dogac, 2025; Le Conte; Ellis; Ritter, 2010)and the development of alternative and sustainable methods to combat this mite are urgent(Chandler *et al.*, 2001).

Therefore, an important alternative to replace or complement chemical control methods is the use of biological agents, such as

entomopathogenic fungi(Baker; Green; Loker, 2020; Costa *et al.*, 2022). According to Chandler *et al.* (2001), these fungi can represent a potential biological agent for the control of *V. destructor* in *A. mellifera* colonies.

The genus *Metarhizium* consists mainly of entomopathogenic fungi with some species used in mycoacaricide formulations. The species Metarhizium anisopliae (Metschnikoff, (Hypocreales: Sorokin. 1883 Clavicipitaceae), formerly known as Entomophthora anisopliae, which is used in several countries for insect control(Driver; Milner; Trueman, 2000), can infect more than 200 species of arthropods (Boaventura; Quintela, 2025) and are found in various commercial products worldwide.

However, studies by Driver, Milner e Trueman (2000)analyzed 123 fungal isolates (including *M. anisopliae, M. flavoviride* and *M. album*) through genetic sequencing and concluded that within the *M. anisopliae* clade there were four distinct varieties, namely: *M. anisopliae* var. *anisopliae*, *M. anisopliae* var. *anisopliae* var. *lepidiotae* (or *lepidiotum*) and *M. anisopliae* var. *acridum*.

Subsequently, Bischoff, Rehner and Humber (2009), through multigene phylogenetic and taxonomic studies, reclassified the varieties that comprised the *M. anisopliae* clade and identified new monophyletic lineages, elevating some varieties to the species category (*M. acridum*, *M. lepidiotae*, *M. majus*), in addition to proposing new species (*M. pingshaense*, *M. robertsii* and *M. globosum*), the rescue of the name *M. brunneum* for some lineages and maintaining the name *M. anisopliae* (now *sensu stricto*) for others.

Furthermore, recent studies have proposed changes in the *M. anisopliae sensu lato* species complex (Kobmoo *et al.*, 2024), with the addition of new species and the reclassification of others — such as the reclassification of *M. lepidiotae*, previously elevated to the category of distinct specie, as a synonym of *M. anisopliae sensu stricto*. The genus *Metarhizium* includes the most well-studied entomopathogenic fungi at the molecular and biochemical levels (Gao *et al.*, 2011).

Note, however, that the various taxonomic revisions in the *Metarhizium anisopliae s. l.* species complex, which have occurred in recent decades, imply that the nomenclature "*M. anisopliae*", used for fungal isolates in the articles cited in this systematic review, may not correspond to the currently recognized species — especially in publications prior to 2009.

Thus, this systematic review aims to evaluate the stage of research on the use of *Metarhizium* spp. fungi as a possible biological agent in the control of the mite *V. destructor* in *A. mellifera* colonies.

Methodology

The present study is characterized by exploratory research using qualitative methods, with

the technical procedure being a systematic literature review, following the PRISMA 2020 (Preferred Reporting Items for Systematic Reviews and Meta-Analyses)guidelines (Page *et al.*, 2021), in the Web of Science, Scopus, PubMed and SciELO databases, accessed through the CAPES Journals Portal, without a time delimitation, in order to obtain the largest possible volume of articles.

To structure this systematic review, the steps suggested by (Sampaio; Sabadini; Koller, 2022) were followed: delimitation of the research objective; choice of data sources; election of keywords for the search; search and storage of results; selection of articles according to inclusion and exclusion criteria; data extraction and evaluation of the quality of evidence in the selected articles.

The review was organized into three stages: (i) identification of records in the Web of Science, Scopus, PubMed and SciELO databases; (ii) screening of articles published in indexed journals and available for download; and (iii) inclusion of eligible full articles that were available online. In the search phase, a combination of terms in English, Portuguese and Spanish was used for searches in the databases (Table 1). Searches were conducted between July 01 and 08, 2025.

The following inclusion criteria were used: (i) must comprise studies on the use of *Metarhizium spp.* fungi for the control of the *V. destructor* mite in *A. mellifera bees*; (ii) all articles must be available for full download in PDF format and without restrictions; (iii) there will be no date limit restriction in order to obtain the largest possible number of articles.

The following exclusion criteria were used: (i) articles that do not use *Metarhizium spp.* fungi for studies on the control of the *V. destructor* mite in *A. mellifera* bees; (ii) articles that do not evaluate the effects of *Metarhizium* spp. fungi applied to *A. mellifera* bee colonies will be excluded; (iii) abstracts, extended abstracts, literature reviews, books, book chapters will be excluded. For screening and duplicate removal, Zotero[®] v. 7.0.20 was used.

As a quality criterion, it was defined that only peer-reviewed articles published in indexed journals would be selected.

Results

A total of 428 records were identified during the database search, distributed across SciELO (2), PubMed (8), Scopus (375), and Web of Science (43). Sixty duplicate records were excluded. The remaining 368 records were then screened by reading their titles and abstracts for adherence to the study's theme; in cases of doubt, the full report was reviewed.

Exclusions were made for records identified as books (11), book chapters (35), reviews (60), those not in Portuguese, English, or Spanish (10), and those outside the scope of the present study (235). After screening, 17 records remained for further investigation. One report was unavailable and subsequently removed. The remaining 16

reports were analyzed, and eligibility and quality criteria were applied. Five reports were excluded because they did not apply the *Metarhizium* spp. fungus directly to beehives, leaving 11 studies that

were included in this systematic review (Figure 1). No reports in Portuguese or Spanish were found.

Table 1. Databases and search terms used in this systematic review.

Database	Search terms English/Portuguese/Spanish		
Scopus	(("varroa") or ("varroosis") or ("varroose") or ("varroatose")) and ("metarhizium") and (("biocontrol") or ("biological control") or ("controlebiologico"))		
Web of Science	(all=(varroa) or all=(varroosis) or all=(varroose) or all=(varroatose)) and all=(metarhizium) and (all=(biocontrol) or all=(biological control) or all=(controlebiologico))		
SciELO	((varroa) or (varroosis) or (varroose) or (varroatose)) and (metarhizium) and ((biocontrol) or (biological control) or (controlebiologico))		
PubMED	((varroa) or (varroosis) or (varroose) or (varroatose)) and (metarhizium) and ((biocontrol) or (biological control) or (controlebiologico))		

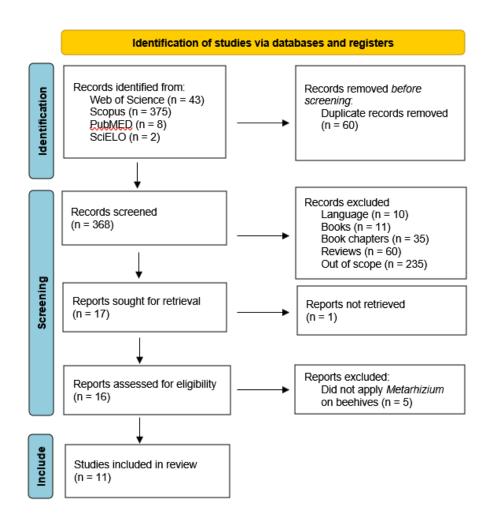


Figure 1. Flowchart of this systematic review based on the PRISMA 2020 protocol.

The 11 identified studies included in this review after screening were categorized as field or laboratory research, by publication year, and country of origin (Table 2). Authors from 8 different countries contributed to these studies on the application of *Metarhizium* spp. fungus for controlling the *V. destructor* mite in *A. mellifera* bees. The majority of studies were conducted by

researchers in the United States of America (4), with three articles sharing the same lead author(Kanga; Jones; James, 2003; Kanga; Jones; Gracia, 2007; Kanga *et al.*, 2010), followed by researchers from Canada (2).

No articles carried out in Brazilian territory that met the eligibility criteria defined in this systematic review were found in the databases used. In addition, 3 articles included in this review have more than 60 citations each(Hamiduzzaman et al., 2012; Kanga; Jones; James, 2003; Shaw et al., 2002), and all other articles have already been cited at least 6 times each by July 08, 2025. The articles

found were published between 2002 and 2021 (Table 2), which indicates a very recent period for studies on the use of *M. anisopliae* in an attempt to control the *V. destructor* mite.

Table 2. List of articles included in this systematic review, including year of publication, country of origin, type of research

(laboratory or field), authors, and article title.

Publication Year	Country of Origin	Research Type	Authors	Title
2002	UK	Laboratory	Shaw <i>et al.</i> (2002)	Laboratory bioassays to assess the pathogenicity of mitosporic fungi to <i>Varroa destructor</i> (Acari: Mesostigmata), an ectoparasitic mite of the honeybee, <i>Apis mellifera</i>
2003	USA	Field	Kanga; Jones; James (2003)	Field trials using the fungal pathogen, <i>Metarhizium anisopliae</i> (Deuteromycetes: Hyphomycetes) to control the ectoparasitic mite, <i>Varroa destructor</i> (Acari: Varroidae) in honey bee, <i>Apis mellifera</i> (Hymenoptera: Apidae) colonies
2007	USA	Field	Kanga; Jones; Gracia (2007)	Efficacy of strips coated with <i>Metarhizium anisopliae</i> for control of <i>Varroa destructor</i> (Acari: Varroidae) in honey bee colonies in Texas and Florida
2009	Chile	Laboratory /Field	Rodríguez <i>et</i> al.(2009)	Evaluation of <i>Metarhizium anisopliae</i> var. <i>anisopliae</i> Qu- M845 isolate to control <i>Varroa destructor</i> (Acari: Varroidae) in laboratory and field trials
2010	USA	Field	Kanga <i>et al.</i> (2010)	Development of a user-friendly delivery method for the fungus <i>Metarhizium anisopliae</i> to control the ectoparasitic mite <i>Varroa destructor</i> in honey bee, <i>Apis mellifera</i> , colonies
2012	Canada	Laboratory	Hamiduzzaman et al. (2012)	Entomopathogenic fungi as potential biocontrol agents of the ecto-parasitic mite, <i>Varroa destructor</i> , and their effect on the immune response of honey bees (<i>Apis mellifera</i> L.)
2013	Egypt	Field	A. Ahmed; K. Abd-Elh (2013)	Efficacy of two fungus-based biopesticide against the honeybee ectoparasitic mite, Varroa destructor
2013	Iran/Japan	Field	Pirali- Kheirabadi <i>et</i> <i>al.</i> (2013)	A field experiment to assess the rate of infestation in honey bee populations of two <i>Metarhizium anisopliae</i> isolates on <i>Varroa destructor</i> (Acari: Mesostigmata)
2018	Canada	Field	Sinia; Guzman- Novoa (2018)	Evaluation of the entomopathogenic fungi <i>Beauveria</i> bassiana GHA and <i>Metarhizium anisopliae</i> UAMH 9198 alone or in combination with thymol for the control of <i>Varroa</i> destructor in honey bee (<i>Apis mellifera</i>) colonies
2020	Italy	Field	Fernandez Ferrari <i>et al.</i> (2020)	Application of <i>Metarhizium anisopliae</i> as a potential biological control of <i>Varroa destructor</i> in Italy
2021	USA	Field	Han <i>et al.</i> (2021)	Directed evolution of <i>Metarhizium</i> fungus improves its biocontrol efficacy against <i>Varroa</i> mites in honey bee colonies



Figure 2. Word cloud from the keywords of the selected articles for this systematic review.

Table 3.Reference, efficacy against *V. destructor*, and safety for *A. mellifera* contained in the studies included in this systematic review.

Reference	Efficacy against V. destructor	A. mellifera safety
Shaw et al.(2002)	 M. anisopliae isolates 442.99, 443.99, 444.99 were the three most virulent, causing 100% mortality of V. destructor 7 days post-inoculation with a concentration of 1x10⁶ conidia mL⁻¹ at 30° C and 40% relative humidity. 	whole hive or field experiments
Kanga; Jones; James (2003)	Fungal powder (≈1x10¹º conidia g⁻¹), dusted into each hive using a salt shaker, caused maximum mite mortality in 3-4 days, with mites remaining infected for up to 42 days.	The treatment was harmless to the bees (adults or brood). Colony development was not affected, with only 9.9% of dead bees showing mycosis. Population decline was only observed in a high concentration treatment.
Kanga; Jones; Gracia (2007)	Tests conducted in Texas and Florida showed that the use of <i>M. anisopliae</i> for mite control in beehives was effective, though less so than the chemical product Apistan® (tau-fluvalinate). Plastic strips coated with <i>M.</i> anisopliae were effective, reducing mite counts per bee by 25 times, but less so than Apistan® (69 times). Efficacy was greater when brood production is low. Both fungal and Apistan® treatments significantly reduced mites compared to control groups, where three colonies died.	Bee populations and brood areas were similar between treatments, with treated colonies increasing their populations by more than 1.2 times. Brood production was similar to Apistan® treated groups.
Rodríguez <i>et al.</i> (2009)	Lab trials with isolate Qu-M845 produced 98% mortality at 1×10^8 conidia mL ⁻¹ . The concentration of 1×10^7 conidia mL ⁻¹ produced 72% mortality, while concentrations of 1×10^6 and 1×10^5 conidia mL ⁻¹ resulted in similar mortalities (p ≤ 0.05) of 46% and 48%, respectively. In a field study, sprinkled dry conidia reduced mite infestation by 67% after 21 days.	In autumn, 35% of dead bees showed mycosis. In spring, there were no significant differences in bee mortality and no mycosis was observed.
Kanga <i>et al.</i> (2010)	Paste formulations significantly reduced the number of mites per adult bee compared to controls. Mite infestations in capped brood were similar between fungus and Apiguard® treatments, and significantly lower than in controls. The P10-2X fungal treatment significantly reduced mite infestations in capped brood.	Adult bee populations were reduced across all treatments compared to the start of the experiments in Texas, but there was no significant difference between treatments. In Florida, there was an increase in the number of adult bees for the fungal treatments P5-3X, P10-D-2X, and P10-2X. 100% of colonies treated with P10-2X or P10-D-2X survived during the 42-day experimental period, while for the other treatment groups there were colony losses at the end of 42 days of experiment.
Hamiduzzaman et al. (2012)	All isolates caused significant mortality in lab tests at 1x10 ⁸ conidia mL ⁻¹ , with two isolates causing over 90% mortality in 7 days.	Direct inoculation of brood led to reduced emergence (75.83% ±6.8) and lower body weight in new bees compared to the control. Brood mortality (24% ±6.8) was not statistically significant.
A. Ahmed; K. Abd-Elh (2013)	for 14 days after application.	Did not infect bees at any life stage (larval, pre- pupal, pupal, oradult). Positive differences in worker body weight were observed post-treatment.
Pirali-Kheirabad et al. (2013)	spraying (5x10° conidia mL-1) were as effective as Apistan® strips in controlling mites	No adverse effects on honey bees were observed.
Sinia; Guzman- Novoa (2018)	When <i>M. anisopliae</i> UAMH 9198 was delivered by dispenser tray method caused 62% mite mortality, showing 97% higher efficacy than a manual duster.	No tests were performed on bee mortality in this study
Fernandez Ferrari <i>et al.</i> (2020)	The fungus <i>M. anisopliae</i> var. <i>anisopliae</i> BIPESCOO 5 was used. Dusted formulated product significantly lowered mite levels (<60%) compared to controls.	. The number of dead bees was higher in the treated group, but the fungus had no negative impact on colony strength, development, or brood. Pollen and honey areas were consistently larger in treated colonies
Han <i>et al.</i> (2021)	The pest control achieved with the JH1078 strain of <i>M. brunneum</i> was comparable to treatments performed with oxalic acid. Treatment with the fungus delayed the exponential increase in varroa levels, but did not fully prevent it.	No noticeable negative differences in hive populations. Fungus-treated colonies and controls entered winter with similar population estimates, but treated colonies survived significantly longer

The keywords used by the authors of the articles had the following highest frequency words: varroa (8), destructor (8), metarhizium (7), mellifera

(7), apis (7), anisopliae (7), fungi (6), control (6), biological (5), spp (4), entomopathogenic (4), bassiana (4), beauveria (3), verticillium (1) and

varroasis (1). The article by Han et al.25 does not have keywords. The distribution of the frequency of keyword usage can be graphically visualized in Figure 2.

Efficacy data of *Metarhizium* spp. against *V. destructor* and safety for *A. mellifera* achieved by the researchers for each evaluated study were extracted from the selected articles (Table 3).

Discussion

The discussion is based firstly on the efficacy of *Metarhizium* spp. in controlling *V. destructor* and subsequently on its safety for *A. mellifera* bees.

Efficacy against V. destructor

All analyzed studies point to the viability of using *Metarhizium* spp. in controlling *V. destructor* mite infestations. However, there is a variation in the effectiveness of fungal isolates regarding their virulence against this mite.

This variation can be explained by the fact that M. anisopliae s. I. exhibits a very high level of variability among isolates(St. Leger et al., 1992). Shah and Pell (2003),comment entomopathogenic fungal isolates can behave very differently within the same species, with the range of insect hosts, infection levels, germination rates, and temperature beina variable. However. considering recent studies that reclassified the M. anisopliae s. l. species complex(Driver; Milner; Trueman, 2000; Bischoff; Rehner; Humber, 2009; Kobmoo et al. (2024), some isolates used could correspond to species distinct from M. anisopliae.

Aw and Hue (2017), cite that the culture medium used to produce *M. anisopliae* for conidia obtention was able to affect its virulence, which could also explain the variation in the effectiveness of different fungal isolates in different studies.

Furthermore, different abiotic factors, such as temperature and humidity, can influence conidia persistence, the fungus's ability to develop spores, and consequently, its ability to infect a certain type of host(Lanza; Monteiro; Malheiros, 2009), which influences the mortality rate presented. A *Metarhizium* spp. isolate that showed good efficacy under certain environmental conditions may not present the same results under other conditions, which can represent a challenge for the use of these fungi.

Another factor that can also alter the efficacy of biocontrol using entomopathogenic fungi would be the concentration of conidia used in the treatment. Rodríguez et al. (2009) found that different conidia concentrations resulted in different mite mortality levels. Kanga et al. (2010) concluded that having fungal spores with good germination, pathogenicity, and virulence, and in adequate concentrations, are important factors for successful *V. destructor* control in *A. mellifera* colonies.

The application method of fungal isolates also caused a variation in treatment effectiveness. Sinia and Guzman-Novoa (2018) found that M.

anisopliae treatments administered via dispenser trays showed 97% higher efficacy rates than those administered via manual duster. Similarly, Rodríguez et al. (2009) observed that the fungus's effectiveness varied with different application methods, noting that dry conidia sprinkled over and between the frames was the most effective approach.

The presence of beehives with high varroa infestation rates, not receiving any treatment, present in the experimental apiary, caused the popularly known effect of "Mite Bombs", where these infected colonies continuously inoculate other nearby colonies with mites and viruses transmitted by bees that transit between hives, either lost or to rob honey, which leads to health problems for all colonies in the apiary(Han *et al.*, 2021).

However, most studies pointed to good treatment efficacy, with a significant increase in mite deaths or a reduction in infestation 3 to 7 days after the application of the fungus in *A. mellifera* hives(A. Ahmed; K. Abd-Elh, 2013; Hamiduzzaman *et al.*, 2012; Han *et al.*, 2021; Kanga *et al.*, 2010; Kanga; Jones; James, 2003; Pirali-Kheirabadi *et al.*, 2013; Rodríguez *et al.*, 2009; Shaw *et al.*, 2002), depending on the fungal isolate and the application method used.

A. mellifera Safety

Most studies do not point to problems caused by the application of *Metarhizium* spp. in *A. mellifera* colonies under field conditions (A. Ahmed; K. Abd-Elh, 2013; Fernandez Ferrari *et al.*, 2020; Han *et al.*, 2021; Kanga; Jones; Gracia, 2007; Kanga; Jones; James, 2003; Pirali-Kheirabadi *et al.*, 2013).

When obtaining a high bee mortality rate in their experiment through laboratory bioassays, comment that in other studies where there was a high mortality rate in the laboratory for a given isolate, the same applied to whole colonies or in the field obtained a minimal mortality rateShaw *et al.* (2002). The authors also emphasize that the interpretation of the results obtained in their study should be cautious, given the high mortality rate in the control group, and because there are observations in other field studies that suggest a lower impact on *A. mellifera* colonies under real conditions.

The results of safety and efficacy in the application of *Metarhizium* spp. in laboratory experiments may not be consistent with reality in the field. Alves *et al.* (1996)concluded that in bioassays performed in the laboratory, bees may be subjected to stress conditions, favoring the development of pathogens, given the humidity and temperature conditions below ideal for bees.

Potrich *et al.* (2018), in laboratory bioassays performed to test the effect of different entomopathogens on Africanized *A. mellifera* bees in Brazil, observed a reduction in the lifespan of worker bees, compared to the control group, when *M. asinopliae* was applied in different ways: (i)

sprayed directly on the bees, (ii) sprayed directly into the plastic box and (iii) mixed with a honey and confectioners' sugar mixture. Only treatment (iv) applied to soybean leaves and placed with the bees in plastic boxes, showed no significant difference in lifespan alteration compared to the control group. The greatest difference in lifespan compared to the control group was observed when bees were sprayed directly with *M. anisopliae* E9 (1x10⁹ conidia mL⁻¹).

In the study by Sinia Guzman-Novoa (2018), the effects of the variety used in the study (*M. anisopliae* UAMH 9198) regarding potential problems caused in *A. mellifera* colonies were not tested. However, the authors based themselves on the concentration (1x10⁸ conidiag⁻¹) used in previous studies by the same research group that proved to be safe for bee colonies. The authors also observed that higher conidia concentrations reduced bee emergence and increased their mortality.

Kanga; Jones; James, (2003) pointed out that only 9.9% of the dead adult bees investigated showed mycosis.Rodríguez *et al.* (2009), in their study, observed that while in autumn there were 35% of bees with signs of *M. anisopliae* infection, no significant problems were detected in spring.

Hamiduzzaman et al. (2012) observed that the application of *M. anisopliae* affected larval development, resulting in newly emerged and/or worker bees with significantly lower body weight compared to those before fungal application. While Kanga; Jones; James (2003) state that there were no effects on young bees or larvae during the study with *M. anisopliae* and A. Ahmed and K. Abd-Elh (2013) found no infection in larvae and report significant weight gain in worker bees after treatment.

Conclusion

Although there are few studies on the application of *Metarhizium* spp. in *A. mellifera* colonies for *V. destructor* control, the obtained results point to the viability of its use for the integrated management of the mite, although further studies are needed regarding ideal environmental conditions, varieties or species, and application methods that would be more efficient, effective, and safe in controlling the mite.

Taking into account the analyzed studies, we can suspect that the variety of the isolate, the concentration of conidia and their form of application, as well as abiotic factors such as temperature and humidity, can influence the efficacy of the treatment and the tolerance of bees to fungi of the genus *Metarhizium*. These factors are relevant and should be considered, especially when applying the treatment in different environmental conditions and with distinct varieties or species of *Metarhizium*.

It is also worth noting that no studies on *V. destructor* control using fungi of the genus *Metarhizium* were found in Brazil in the searched databases, which may present an opportunity for future related research.

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Conflicts of Interest

The authors declare that there is no conflict of interest of a personal, commercial, academic, political, or financial nature related to the research and publication of this systematic review. They also declare that there was no funding for this systematic review. No formal assessment of the risk of bias of the included studies was performed.

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References

A. AHMED, Abdelaal; K. ABD-ELH, Hany. Efficacy of Two Fungus-based Biopesticide Against the Honeybee Ectoparasitic Mite, *Varroa destructor*. **Pakistan Journal of Biological Sciences**, [s. l.], v. 16, n. 16, p. 819–825, 2013.

ALVES, S. B. *et al.* Effects of some insect pathogens on the Africanized honey bee, *Apis mellifera* L. (Hym., Apidae). **Journal of Applied Entomology**, [s. *l.*], v. 120, n. 1–5, p. 559–564, 1996.

ANDERSON, D.L.; TRUEMAN, J.W.H. *Varroa jacobsoni* (Acari: Varroidae) is more than one species. **Experimental & Applied Acarology**, [s. *l.*], v. 24, n. 3, p. 165–189, 2000.

AW, Kimberly Moon San; HUE, Seow Mun. Mode of Infection of *Metarhizium* spp. Fungus and Their Potential as Biological Control Agents. **Journal of Fungi**, [s. *I.*], v. 3, n. 2, p. 30, 2017.

BAKER, Brian P.; GREEN, Thomas A.; LOKER, Ali J. Biological control and integrated pest management in organic and conventional systems. **Biological Control**, [s. *l.*], v. 140, p. 104095, 2020.

BISCHOFF, Joseph F.; REHNER, Stephen A.; HUMBER, Richard A. A multilocus phylogeny of the *Metarhizium anisopliae* lineage. **Mycologia**, [s. l.], v. 101, n. 4, p. 512–530, 2009.

BOAVENTURA, Heloiza Alves; QUINTELA, Eliane Dias. The multifunctionality of the fungus

Metarhizium spp. and its use in Brazilian agriculture. **Bragantia**, [s. *l.*], v. 84, p. e20240183, 2025.

CELIKKOL, Elif; DOGAC, Ersin. The status of pyrethroid resistance mutation frequencies in *Varroa destructor* populations in the most important beekeeping areas of Türkiye. **Experimental and Applied Acarology**, [s. *I.*], v. 94, n. 2, p. 32, 2025.

CHANDLER, D. et al. Prospective Biological Control Agents of *Varroa destructor* n. sp., an Important Pest of the European Honeybee, Apis mellifera. **Biocontrol Science and Technology**, [s. l.], v. 11, n. 4, p. 429–448, 2001.

COSTA, Antonio Felix Da *et al.*Controle alternativo e biológico de pragas e fitopatógenos: uma década de contribuição. **Pesquisa Agropecuária Pernambucana**, [s. *l.*], v. 27, n. 2, p. e2742272022, 2022.

DRIVER, Felice; MILNER, Richard J.; TRUEMAN, John W.H. A taxonomic revision of *Metarhizium* based on a phylogenetic analysis of rDNA sequence data. **Mycological Research**, [s. *l.*], v. 104, n. 2, p. 134–150, 2000.

FERNANDEZ FERRARI, Maria Celeste *et al.*Application of *Metarhizium anisopliae* as a potential biological control of *Varroa destructor* in Italy. **Journal of Apicultural Research**, [s. l.], v. 59, n. 4, p. 528–538, 2020.

GAO, Qiang *et al.* Genome Sequencing and Comparative Transcriptomics of the Model Entomopathogenic Fungi *Metarhizium anisopliae* and *M. acridum.* **PLoS Genetics**, [s. *l.*], v. 7, n. 1, p. e1001264, 2011.

HAMIDUZZAMAN, Mollah Md. *et al.* Entomopathogenic fungi as potential biocontrol agents of the ecto-parasitic mite, Varroa destructor, and their effect on the immune response of honey bees (*Apis mellifera* L.). **Journal of Invertebrate Pathology**, [s. *l.*], v. 111, n. 3, p. 237–243, 2012.

HAN, Jennifer O. *et al.* Directed evolution of Metarhizium fungus improves its biocontrol efficacy against Varroa mites in honey bee colonies. **Scientific Reports**, [s. *l.*], v. 11, n. 1, p. 10582, 2021.

KANGA, Lambert H. B. *et al.* Development of a user-friendly delivery method for the fungus *Metarhizium anisopliae* to control the ectoparasitic mite Varroa destructor in honey bee, *Apis mellifera*, colonies. **Experimental and Applied Acarology**, [s. *l.*], v. 52, n. 4, p. 327–342, 2010.

KANGA, Lambert H. B.; JONES, Walker A.; GRACIA, Carlos. Efficacy of strips coated with *Metarhizium anisopliae* for control of *Varroa*

destructor (Acari: Varroidae) in honey bee colonies in Texas and Florida. **Experimental and Applied Acarology**, [s. l.], v. 40, n. 3–4, p. 249–258, 2007.

KANGA, Lambert Houssou Ble; JONES, Walker A.; JAMES, Rosalind R. Field Trials Using the Fungal Pathogen, *Metarhizium anisopliae* (Deuteromycetes: Hyphomycetes) to Control the Ectoparasitic Mite, *Varroa destructor* (Acari: Varroidae) in Honey Bee, *Apis mellifera* (Hymenoptera: Apidae) Colonies. **Journal of Economic Entomology**, [s. *l.*], v. 96, n. 4, p. 1091–1099, 2003.

KOBMOO, Noppol *et al.* Integrative taxonomy of *Metarhizium anisopliae* species complex, based on phylogenomics combined with morphometrics, metabolomics, and virulence data. **IMA Fungus**, [s. *l.*], v. 15, n. 1, p. 30, 2024.

LANZA, Lourenço Maurício; MONTEIRO, Antonio Carlos; MALHEIROS, Euclides Braga. Sensibilidade de *Metarhizium anisopliae* à temperatura e umidade em três tipos de solos. **Ciência Rural**, [s. *l.*], v. 39, n. 1, p. 6–12, 2009.

LE CONTE, Yves; ELLIS, Marion; RITTER, Wolfgang. *Varroa* mites and honey bee health: can *Varroa* explain part of the colony losses? **Apidologie**, [s. *l.*], v. 41, n. 3, p. 353–363, 2010.

MAGGI, Matías D. *et al.*LC₅₀ baseline levels of amitraz, coumaphos, fluvalinate and flumethrin in populations of *Varroa destructor* from Buenos Aires Province, Argentina. **Journal of Apicultural Research**, [s. *l.*], v. 47, n. 4, p. 292–295, 2008.

PAGE, Matthew J et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. **BMJ**, [s. l.], p. n71, 2021.

PIRALI-KHEIRABADI, Khodadad *et al.*A Field Experiment to Assess the Rate of Infestation in Honey Bee Populations of Two *Metarhizium anisopliae* Isolates on Varroa destructor (Acari: Mesostigmata). **Journal of Arthropod-Borne Diseases**, [s. l.], v. 7, n. 1, p. 15–22, 2013.

POTRICH, Michele *et al.* Effect of entomopathogens on Africanized Apis mellifera L. (Hymenoptera: Apidae). **Revista Brasileira de Entomologia**, [s. l.], v. 62, n. 1, p. 23–28, 2018.

RODRÍGUEZ, Marta *et al.*Evaluation of Metarhizium anisopliae var. anisopliae Qu-M845 Isolate to Control Varroa destructor (Acari: Varroidae) in Laboratory and Field Trials. **Chilean journal of agricultural research**, [s. *l.*], v. 69, n. 4, 2009. Disponível em: http://www.scielo.cl/scielo.php?script=sci_arttext&pi d=S0718-

58392009000400009&Ing=en&nrm=iso&tIng=en. Acesso em: 4 nov. 2025.

- ROSENKRANZ, Peter; AUMEIER, Pia; ZIEGELMANN, Bettina. Biology and control of Varroa destructor. **Journal of Invertebrate Pathology**, [s. *l.*], v. 103, p. S96–S119, 2010.
- SAMPAIO, Maria Imaculada Cardoso; SABADINI, Aparecida Angélica Zoqui Paulovic; KOLLER, Silvia Helena (org.). **Produção Científica: um Guia Prático**. [S. I.]: Universidade de São Paulo. Instituto de Psicologia, 2022. DOI: 10.11606/9786587596280Acesso em: 4 nov. 2025.
- SHAH, P. A.; PELL, J. K. Entomopathogenic fungi as biological control agents. **Applied Microbiology and Biotechnology**, [s. *l.*], v. 61, n. 5–6, p. 413–423, 2003.
- SHAW, Katie E *et al.* Laboratory bioassays to assess the pathogenicity of mitosporic fungi to *Varroa destructor* (Acari: Mesostigmata), an ectoparasitic mite of the honeybee, Apis mellifera. **Biological Control**, [s. *l.*], v. 24, n. 3, p. 266–276, 2002.
- SINIA, Alice; GUZMAN-NOVOA, Ernesto. Evaluation of the entomopathogenic fungi *Beauveria bassiana* GHA and *Metarhizium anisopliae* UAMH 9198 alone or in combination with thymol for the control of *Varroa destructor* in honey bee (*Apis mellifera*) colonies. **Journal of Apicultural Research**, [s. I.], v. 57, n. 2, p. 308–316, 2018.
- ST. LEGER, R.J. *et al.* Genetic differences in allozymes and in formation of infection structures among isolates of the entomopathogenic fungus *Metarhizium anisopliae*. **Journal of Invertebrate Pathology**, [s. *l.*], v. 60, n. 1, p. 89–101, 1992.