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Optimizing the soluble tablet formulation of *Metarhizium anisopliae* for effective biological pest control

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ABSTRACT

The research focuses on soluble tablet formula of a *Metarhizium anisopliae*, an entomopathogenic fungus, for optimal biological pest control. *M. anisopliae* is well known for its broad spectrum of activity against a number of insect pests, and could be an important part of any integrated pest management (IPM) program. However, challenges are associated with stability, shelf life and environmental persistence of fungal-based biopesticides. The soluble tablet formulation solves the problem of delivery of the fungal spores by giving the fungal spores' better protection against environmental stressors and allowing the farmer a more accurate, easier approach to application. The formulated E showed the highest performance compared to the other samples, where it released the conidia within the shortest time (258 seconds) and disintegrated fully in water; it released the highest concentration of initial conidial spores of (16.24×10^8 spores/g), and it maintained pest control capacity for up to 12 weeks of storage. On the other hand, formula G recorded poor dispersion, the longest disintegration time of 529 seconds and the lowest viability. The study's findings can help overcome the current challenges related to the formulations of entomopathogenic fungi and a promising, sustainable, and ecofriendly alternative to chemical pesticides in agricultural production.

Keywords: Soluble tablet, Entomopathogenic fungi, *Metarhizium anisopliae*, Pest control, Storage viability.
Article type: Research Article.

INTRODUCTION

Globalization of farming practices leading to sustainable management of pests has become popular than using synthetic pesticides because of several effects on the environment and human health (Pathak *et al.* 2022). These chemicals are detrimental to the natural environment, freely entering the soil, water systems, pollinators, and their natural predators (Hashimi *et al.* 2020). The World Health Organization (WHO) report highlighted that at least 3 million people suffered from pesticide poisoning, and 220000 died annually, mostly from developing nations (Sharma *et al.* 2019; Garud *et al.* 2024). According to the Food and Agriculture Organization (FAO), more than 580 insect and mite species have become resistant to at least one insecticide, limiting the use of chemicals and making it costly for farmers (Venkatesan *et al.* 2022; Kulazhanov *et al.* 2024). It is a pathogenic fungus that infects a few specific species of insects, which is found to be naturally present in soil and has little impact on the environment (Moisan *et al.* 2019; Banaei *et al.* 2022). The global biopesticide market was USD 4.4 billion in 2020 and is expected to rise to USD 10.6 billion with a compound annual growth rate (CAGR) of 14.1% by 2027 (Belagalla *et al.* 2024). Further advancement in oral delivery systems, such as manufacturable soluble tablets, is a proper direction to advance the efficacy and reliability of fungal biopesticides in modern agriculture (Hegde *et al.* 2023). *Metarhizium anisopliae* is a hyphal, saprophytic and parasitic soil fungus that is used widely as a biocontrol agent of many insects including agro-economic pests such as *Spodoptera litura*, *Helicoverpa armigera*, and *Locusta migratoria* (Brunner-Mendoza *et al.* 2019). It enters the insect through the following process:

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airborne conidia nonspecifically adhere to the insect cuticle, germinate and form aspersoria that appress and penetrate the cuticle using proteases and chitinases (Sharma *et al.* 2024). *Beauveria bassiana*, *M. anisopliae* and *Isaria fumosorosea* are widely studied fungi with potential for reducing or eliminating pest populations in a sustainable way, i.e., as they are environmentally safe, and are biodegradable (Bamisile *et al.* 2021). It develops as blastopores within the host hemocoel, which releases destruxins, toxic substances to insects, and leads to death within 4-10 days (Gautam 2020). The pathogenicity of *M. anisopliae* is moderate to insects but non-virulent to beneficial insects like honey bees and lady beetles so it has undergone biosafety test by OECD and EPA (Vivekanandhan *et al.* 2021; Singh *et al.* 2024). This is a fertilizer that is eco-friendly and organic farmers recommend it because it is biodegradable (Namasivayam *et al.* 2024). Fungal-based biopesticides have great potential, but their formula and stability remain great challenges. Issues regarding the fungi shelf life, ease of application, and environmental persistence are also included (Butu *et al.* 2022). Moreover, the survival and activity of entomopathogenic fungi (EPF) are limited by environmental factors like temperature, humidity, and UV radiation, which negatively influence fungal survival and activity (Singh *et al.* 2024). There is a need to develop more stable and friendly formulations of fungal–based biopesticides to enhance performance and reliability (Avinash *et al.* 2024). This formulation should make the biocontrol agent less likely to degrade prematurely and offer a more accurate and easier application for farmers and pest management professionals (Lei *et al.* 2023). The fungal spores could be protected from environmental stressors but are still effective in soluble tablets, making handling easier (Hermann *et al.* 2023). Additionally, the release of active agents could be controlled, such that optimum pest control can be achieved over a longer period of time (Maheswari *et al.* 2023). This study investigates the formulation, stability, and efficacy of soluble tablet-based *M. anisopliae* as a biocontrol agent. The study seeks to create a user-friendly and stable product with high levels of biological activity under changing environmental conditions. The impact of the study is its ability to successfully serve as a solution to some of the problems associated with current EPF formulations, and provide a sustainable way to control pests in agriculture and other fields. Successful application of this research helps reduce dependence on chemical pesticides and encourages pesticide management practices that are friendly to the environment and enhance the sustainability of agricultural production.

MATERIALS AND METHODS

Laboratory conditions and environmental control

The research is conducted in a laboratory environment controlled to provide optimal conditions for fungal growth and formulation development. Sterile working surfaces, controlled temperature and humidity settings for breeding *Metarhizium anisopliae* and preparing the biopesticide formulations are provided in the laboratory. The relative humidity is maintained from 60 - 70% and temperature from 25 ± 2 °C, ideal for fungal growth and production of conidia. During fungal handling and formulation processes, proper ventilation and safety measures (such as enclosed laminar flow hoods) are performed to avoid contamination (Wakil *et al.* 2022).

Isolation and culturing of *Metarhizium anisopliae*

The fungus is isolated from local field samples and maintained under sterile conditions with a pure culture of fungal strain *M. anisopliae* strain CTU17. The soil or insect samples are isolated from the specimens infected with the fungus under surface sterilization and culture in selective media. The strain is then preserved by subculturing on Sabouraud Dextrose Agar (SDA) plates and incubated at room temperature. To grow and sporulate conidia, the fungus is grown in liquid media such as glucose and peptone for large-scale culture production. Culture conditions optimized for the maximal production of the conidia with agitation and aeration are provided to achieve uniform growth and a high yield of spores. After 10–14 days of incubation, maximum spore production is at hand, and the conidia are harvested (Li *et al.* 2021).

Soluble tablet formulation process

The process of formulation of soluble tablets involves the selection of excipients which ensure the stability, solubility and controlled release of *M. anisopliae* conidia. Such excipients as sodium alginate, cornstarch, and xanthan gum have properties that help to bind and disintegrate the tablet; they are also biodegradable or not toxic; those can be chosen as suitable excipients (Table 1). A homogenous blend of the excipients and a suspension of fungal conidia are prepared. Excess moisture is then dried from the mixture and molded into tablet shapes using a tablet press. After mechanical homogenization, the mixture was transferred to a 32-mm mold diameter, frozen at -60 ^{0}C for 24 hours and lyophilized by a vacuum freezer-drying machine (BioBase) for 3 days until completely

dry at 12 mTorr pressure. This keeps the conidia viable and facilitates reconstituting the tablets easily in water at the time of application (Gao *et al.* 2025).

Table 1. Compounds of formulate the soluble tablet of *M. anisopliae.*

Formula	The ingredients of formula (%)					
	Conidia	Sodium alginate	Sodium starch glycolate	Xanthan Gum	cornstarch	Skim milk
A	50	3	3	0	0	0
B	50	3	3	1	0	0
C	50	0	0	0	3	0
D	50	3	3	1	3	0
E	50	1	3	1	3	0
F	50	0	3	1	25	0
G	50	0	0	0	0	25

Testing protocols to determine the disintegration time and dispersion properties in water are used to evaluate the effectiveness of the tablet formulations. The disintegration test is employed in which the tablet's disintegration time is measured when it breaks down completely into smaller fragments in water at room temperature. Dispersion analysis is performed to find if the fungal conidia are dispersed evenly after the tablet is dissolved in water. To observe the release of the conidia from the tablets, microscopic analysis is carried out to ensure the release of spores in a viable and uniform manner suitable for effective pest control.

Shelf-life and storage stability testing

The formula shelf life is assessed by testing the viability of the conidia incorporated into the soluble tablets over time. At various intervals during the storage period, germination rate assays are performed to check the percentage of the conidia capable of successfully germinating and retaining infectivity. The storage conditions are closely monitored for temperature and humidity factors to relate to how bio-pesticide is stored in the real world. Monitoring is performed at several intervals to track conidia viability to inform upon formulation stability and long term storage with minimal reduction in efficacy. The performance of the soluble tablet formulations as insecticides is evaluated using laboratory bioassay with *Myzus persicae* (peach aphid) as a test pest in agricultural systems. Mortality rates of the aphids are evaluated under several application dosages, and the aphids are exposed to various concentrations of the reconstituted tablet formulation. Mortality rates are corrected by comparing mortality in the control group, and the effectiveness of the bio-pesticide is assessed using both time to mortality and total effect on aphid populations. Determination of the dosage and application methods necessary for adequate pest control using *M. anisopliae* tablets is essential.

RESULTS

Conidium soluble tablet disintegration and dispersion

The dispersion aspect (total dispersion, lumps presence or slow swelling) and disintegration time (min) of a conidium tablet were calculated by adding it to a Becker containing 100-mL distilled water at room temperature and monitoring the time (min). After soluble tablet disintegration, aliquots were collected and filtered to estimate conidium release (conidia mL^{-1}) by microscopic counting in a Neubauer chamber (× 400; Alves 2006). Disintegration time and aspect were determined for all formulae, using shorter disintegration time as a selection criterion. Both water dispersion speed and conidium concentration were performed for the selected formulae.

The shelf life and viability test

Conidium tablets were disintegrated in 10 mL of Tween 80® solution (0.05%). Then, serial dilutions were performed, transferring a 50 µl aliquot of this suspension to a Petri dish RODAC® containing PDA (potato dextrose agar) medium and incubating at 26 ± 2 °C, 70% relative humidity, photo-phase 12 h. After 18 h-incubation, the viability (%) was determined by evaluating the proportion between germinated and non-germinated conidia by counting under an optical microscope (× 400). This analysis with tablet samples from the different packages was performed monthly during storage. Sensory properties, water dispersion aspects, disintegration time and fungal concentration were adopted to evaluate the disintegration and dispersion characteristics of these various *M. anisopliae* tablet formulae. The results differ greatly between the formulae. Formulae B, F, D, E and A were found to disperse completely in water respectively and, hence, were suitable for

easy application and good fungal release. In this case, formula B had the longest disintegration time (508 seconds). In comparison, formula E had the shortest disintegration time (258 seconds) and was, therefore, the most efficient for rapid disintegration (Table 3).

Table 2. Disintegration and dispersion of Tablet *Metarhizium anisopliae*.

Formula	Sensory evaluation		Disintegration time **(second)**	**Concentration (10^{10} CFU g^{-1})**
	Firmness	Water dispersion aspect		
A	+	Total dispersion	345[c]	1.22
B	+	Total dispersion	508[a]	1.3
C	+	Lumps presence	86[e]	1.25
D	++	Total dispersion	415[b]	1.5
E	++	Total dispersion	258[d]	1.24
F	+	Total dispersion	347[c]	1.27
G	+++	Lumps presence	529[a]	0.61

In water dispersion, C and G formulae showed lump presence suggesting that complete dissolution did not take place to impact the uniformity of fungal application. Formula G, however, had the highest firmness (+++) along with the longest disintegration time (529 seconds) and lowest fungal concentration (0.61×10^{10} CFU g^{-1}), suggesting that this formula might have high structural stability, but at the same time, structurally unstable and thus, ineffective in a fungal release. In contrast, formula D (optimal formula) exhibited the best balance with moderate total dispersion (436%), disintegration time (415 seconds) and highest fungal concentration (1.5×10^{10} CFU g^{-1}), which make it a strong candidate for application in the field. Results from these findings suggest that rapid dispersion of biopesticides is desirable but should be counterbalanced with a suitable balance of fungal viability and concentration to use the biopesticide effectively. The images show the physical characteristics and dispersion behavior of the *M. anisopliae* tablet formulations dissolved in water. The first set of images shows the dispersion of the tablets in water (A, B, C, and D) and their improvement in structural integrity before immersion. Formula B is smooth and more compact, while A seems to have a fibrous texture of uniform green coloration. Although the formula C appears to have visible cracks in the structure, the structural weakness causes the formula to disintegrate more rapidly. Formula D has a uniform texture and a little texture on the surface. Bearing in mind their proper solubility and fungal spore release, formulae A, B, and D, upon immersion, disperse homogeneously in water. Formula C has partial dispersion, and residue is visible, so it does not dissolve completely, and uniform application becomes impossible (Fig. 1). The second set of images shows three extra-formulae consisting of the first two tablets that are well smooth and uniform and the third with a different texture that is slightly glossy. The shapes of these formulae have a very different behavior of dispersion in water. The first two exhibit relatively uniform dispersion, much like the lines of formulae A and B which exhibited in the first set. However, the third formula yields a milky less homogenous dispersion, implying differences in the binding or composition of the excipient, bearing on the effectiveness of fungal release.

Viability (%) of *Metarhizium anisopliae* conidium tablets

This measure is expressed as the viability of *M. anisopliae* conidium tablets (formulae E and G) up to week 12 (conidium concentration of 10^8 spores per gram; Fig. 2). A significantly higher conidium concentration of formula E (16.24×10^8 spores/g) than G (4.65×10^8 spores/g) was observed at the beginning of the storage period (week 0). After five days of storage at 22 °C, the viability of all conidia in both formulae had declined greatly. By week 2, formula E retained 9.95×10^8 peer-review spores/g, and G retained only 3.47×10^8 peer-review spores/g, meaning that E showed better conidial viability retention. In week 4, the viability of both formulae continued to decline, and formula E was at 5.54×10^8 spores/g while G at 2×10^8 spores/g. From week 6 onward, its decline became steeper on both the formulae, becoming close to zero viability. Formula E declined to 0.51×10^6 spores/g at week 6, whereas G carried only 0.69×10^6. By week 12, viability was lost in both formulae, with 0.02×10^8 spores/g in formula E and 0.054×10^8 spores/g in G (Table 4). These results show that formulation E had better initial survival and decayed slower than G. Both formulations suffered a large loss in viability over time. Thus

improvements in storage conditions or stability of the formulation are required to achieve a long shelf life and to ensure a long-term effect in applications for biological pest control. The data is the corrected time over life mortality of pests treated with different formulae of the *M. anisopliae* conidium tablets fired at the moment of application (newly manufactured) and twelve weeks after manufacture. Mortality rates were measured 5, 7 and 9 days after treatment (DAT) in the freshly produced formulae, and 5, 7, 9 and 11 DAT after 12 weeks of storage.

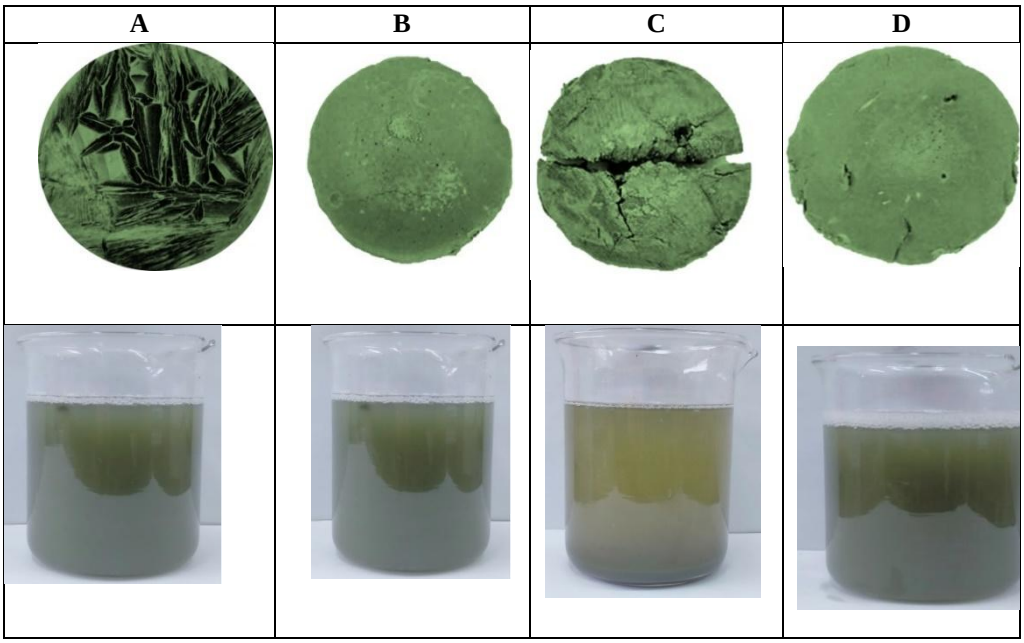

Fig. 1. Physical characteristics and dispersion behavior of *Metarhizium anisopliae* tablet.

Table 3. Viability (%) of the *Metarhizium anisopliae* conidium tablets determined by germination rate over its preservation.

Storage time (week)	conidium tablets (10^8 spores g^{-1})	
	E	G
0	16.24[a]	4.65[a]
2	9.95[b]	3.47[ab]
4	5.54[c]	2.0[bc]
6	0.51[d]	0.69[c]
8	0.39[d]	0.62[c]
10	0.06[d]	0.13[c]
12	0.02[d]	0.054[c]

Efficacy of *M. anisopliae* conidium tablets (formula E) at 12 weeks

First formula A_1 showed the highest efficacy in which mortality was 67.87, 86.11 and 100% at 5, 7 and 9 DAT, respectively. This suggests that A_1 was the best treatment shortly after being produced. The trend for formulae A_2, A_3 and A_4 was similar, albeit lower in efficacy. At 7 DAT, A_2 had 84.84% mortality, A_3 and A_4 recorded 80.24% and 76.75% mortality, respectively, whereas, at 9 DAT, both A_2 and A_3 had 93.78% and 93.82% mortality. After 12 weeks of storage, all formulae lost efficacy. Although the most effective, formula A_1 decreased the mortality rate at early time points (41.78% at 5 DAT, 50% at 7 DAT), but reached 97.53% by 11 DAT. Mortality rates as low as 36.06% and 32.53% were observed for A_2 and A_3, respectively, at 5 DAT, but provided high effectiveness by 11 DAT (93.87% and 91.4% respectively). While exhibiting the highest incidence of early mortality rates, formula A_4 recorded the lowest early mortality rates, i.e. 19.42% at 5 DAT, 38.41% at 7 DAT and 54.28% at 9 DAT, but still achieved 90.07% at 11 DAT (Table 5). Fungal biopesticide treatments were effective as treating groups showed no mortality throughout the experiment, while the control group survived as expected for the maximum test duration test. These results suggest that the *M. anisopliae* formulae retain activity with time, but activity does decrease somewhat as the formula ages. The others were consistently outdone by formula A_1,

which is perceived to be the most reliable formula for sustained pest control. All formulae eventually recovered high efficacy by 11 DAT after 12 weeks of storage; can still be used for long-term pest management. Some further improvements in formulation stability could help maintain higher early-stage efficacy after storage.

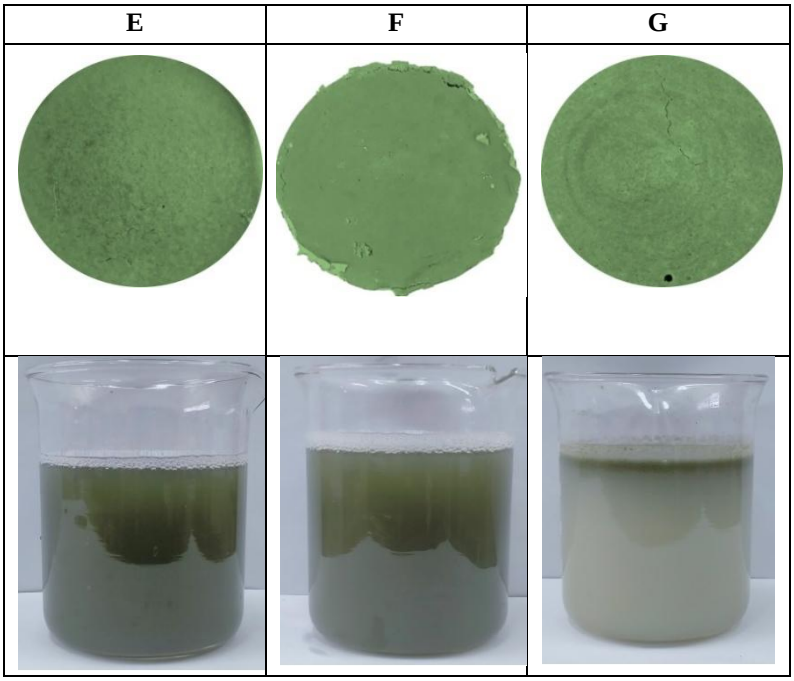

Fig. 2. Physical characteristics and dispersion behavior of *Metarhizium anisopliae* tablet.

Table 4. Efficacy of *M. anisopliae conidium* tablets (formula E) at 12 weeks' storage against aphids in laboratory.

Treatment	Corrected mortality (%)						
	After producer			After 12 weeks			
	5 DAT	7 DAT	9 DAT	5 DAT	7 DAT	9 DAT	11 DAT
A1	67.87a	86.11a	100a	41.78a	50a	65.4a	97.53a
A2	45.53a	84.84ab	93.78b	36.06a	42.94b	62.93a	93.87ab
A3	44.3a	80.24bc	93.82b	32.53a	39.52b	56.75b	91.4ab
A4	31.96ab	76.75c	93.82b	19.42ab	38.41b	54.28b	90.07b
Control	0.00b	0.00d	0.00c	0.00b	0.00c	0.00c	0.00c

DAT: Days after treatment; A_1: 1 tablet/8L of water; A_2: 1 tablet/16L of water; A_3: 1 tablet/24 L of water; A_4: 1 tablet/32 L of water

The correction was made on the mortality data of pests after 12 weeks of storage for the treatments (B_1–B_4) with the *M. anisopliae* conidium tablets. Various time points after treatment (5, 7, 9, and 11 DAT were measured for mortality rates. A comparison group was also included. The highest efficacy was noted by formula B_1 with 70.09%, 89.44% and 98.81% mortality at 5, 7 and 9 DAT, respectively. After 12 weeks of storage, B_1 performed extremely well with a corrected mortality of 55.27% at 5 DAT, increased to 79.38% at 7 DAT, 91.05% at 9 DAT, and attained almost total pest eradication (98.81%) at 11 DAT. Thus, formula B_1 keeps high effectiveness through time, being a good long term pest control option.

Efficacy of *M. anisopliae* conidium tablets (formula G) at 12 weeks

Formula B_2 trailed closely with an initial mortality of 64.38% at 5 DAT and 92.54% at 9 DAT. After 12 weeks, there was a slight loss of effect, but it remained at 54.08% mortality at 5 DAT, 65.93% at 7 DAT, 79.90% at 9 DAT and 85.15% at 11 DAT. Likewise, B3 attained a mortality degree of 65.53 % at the fifth DAT and a sharp increase to 93.78 % at 9 DAT. It dropped more significantly after 12 weeks of early-stage effectiveness after 12 weeks with a percentage of 39.47% at 5 DAT and 64.82% at 7 DAT while still having 82.74% mortality at 11 DAT. The lowest initial mortality among the treatments at 5 DAT (43.11%) and 9 DAT (90.16%) were recorded by formula B_4. After storage for 12 weeks, performance fell even further, as mortality was only 37.21% at 5 DAT and increased to 80.21% at 11 DAT. This implies that although B_4 still seems like an effective ganglion blocker,

its effectiveness decreases more quickly than that of B_1, B_2, or B3 (Table 6). The fungal biopesticide treatments proven effective were all given an additional test using uninfected ants. They were given 0% mortality throughout the experiment, ensuring that the control group was recorded over 0% mortality. The general outcomes of the results suggest that formula B_1 is the most effective and stable formula of the three over the time elapsed, with high mortality rates being observed as long as 12 weeks or even 6 weeks. Formulae B_2 and B_3 also performed well and showed slightly more reduction of their efficacy over time. In addition, B_4 had the greatest decline and needs additional optimization regarding its longevity. With this combination of formula stability, users are certain that the pest control concern is consistent and long-lasting.

Table 5. Efficacy of *M. anisopliae conidium* tablets (formula G) at 12 weeks' storage against aphids in laboratory.

Treatments	Corrected mortality (%)						
	After 12 weeks			After 12 weeks			
	5 DAT	7 DAT	9 DAT	5 DAT	7 DAT	9 DAT	11 DAT
B1	70.09^{a}	89.44^{a}	98.81^{a}	55.27^{a}	79.38^{a}	91.05^{a}	98.81^{a}
B2	64.38^{ab}	82.46^{ab}	92.54^{b}	54.08^{ab}	65.93^{ab}	79.90^{ab}	85.15^{b}
B3	65.53^{ab}	78.02^{b}	93.78^{b}	39.47^{abc}	64.82^{ab}	75.05^{ab}	82.72^{b}
B4	43.11^{b}	75.56^{b}	90.16^{b}	37.21^{c}	51.41^{b}	63.73^{b}	80.21^{b}
Control	0.00^{c}	0.00^{c}	0.00^{c}	0.00^{d}	0.00^{c}	0.00^{c}	0.00^{c}

DAT: Days after treatment; B_1: 1 tablet/8L of water; B_2: 1 tablet/16L of water; B_3: 1 tablet/24 L of water; B_4: 1 tablet/32 L of water.

DISCUSSION

The outcome of the general assessment of the *Metarhizium anisopliae* conidium tablet formulae and the different evaluations offered useful information on the application stability, efficacy and practicality of these formulae. Regarding disintegration and dispersion, formula G dispersed in water most quickly and could be best suited for immediate release in pest control applications because of that quality. Though it had superior water dispersion time, its shorter shelf life (6 weeks) is unsuitable for long-term storage and transport. This concurs with Stephan *et al.* (2021), highlighting the need to consider tablet excipients and structure in the formula to ensure rapid spore viability and even distribution (Stephan *et al.* 2021). On the contrary, formula E dispersed slightly slower. Still, it attained a balanced and similarly good performance with a shelf life of 12 weeks, meaning that it could undergo acceptable storage and transportation. Veronica *et al.* (2022) elaborated that differences in the type and concentration of the excipients found that the formula additives affect the physical characteristics and fungal stability (Veronica *et al.* 2022). Formula E is stable such that the biopesticide is stocked for longer periods of time without substantial loss of efficacy, making it a less risky application from the large-scale distribution and use in pest management system perspectives. Almeida *et al.* (2019) emphasized that tablets with longer disintegration time were shown to be less potent in terms of killing the fungus and in distributing it evenly in the matrix (Almeida *et al.* 2019). Based on the above viability results, the least decline in conidia viability was found for formula E, showing a small 8.25% reduction over the 12-week storage period. This also indicates that E is more resistant to environmental conditions and better preserves fungal propagates over a longer period of time to provide efficacy upon application of the product. Bharti & Ibrahim (2020) noted that the stability of fungal biopesticides depends on the formula type, selection of excipients, and environmental conditions of storage (Bharti & Ibrahim 2020). Singh *et al.* (2017) formulated biological control agents necessary to develop formulae that enhance the ability of the conidia to germinate after a long period (Singh *et al.* 2017). On the contrary, G and D had the highest falls in viability, particularly G, which had the highest drop. This implies that G began to work. Still, its shelf life has significant limiting effects with the possibility of reduced pest control efficiency if not used as soon as produced. However, formula E again had the highest immediate and stored mortality of 85% and 80% respectively. Mancera-López *et al.* (2019) also stated that conidia stored in protectant matrices at appropriate conditions maintain the reproduction capability and virulence for several months (Mancera-López *et al.* 2019). In addition, Teixidó *et al.* (2022) pointed out that formulation influences effectiveness directly in the field. Such a formula that maintains the conidial integrity during periods of storage leads to enhanced mortalities (Teixidó *et al.* 2022). The slightly lower corrected mortality rate (7%) associated with formulae A and F compared to E documents that are less reliable than E to provide consistent pest control time. Upon evaluating the commercial feasibility of these formulae, E is high performing, possible, environmentally friendly, and economically favorable. Accordingly, E

scores a perfect 5 in all three categories, thereby becoming the most practical and sustainable production and application for large-scale production. Kumar *et al.* (2021) pointed out that the effectiveness, shelf life and cost of manufacturing biopesticides for the commercial market are key factors that should be met (Kumar *et al.* 2021). Furthermore, the products have a relatively long shelf life of twelve weeks, which further lends itself to the widespread utilization of the product in pest control programs. Martínez *et al.* (2023) discussed that oil-based structures or stabilizers supported higher conidia protection because of the similar effects (Martínez *et al.* 2023). Consequently, compared to formula G, despite excellent environmental benefits and reduced cost, it has a very short shelf life which would limit its commercial application unless storage conditions can be optimized. According to Stejskal *et al.* (2021), formulae with low shelf life are usually unstable and have relatively short field performance and thus cannot be marketed (Stejskal *et al.* 2021). Formulae B and F, while having moderate environmental benefits and moderate cost effectiveness, lack their ability to maintain shelf life and are less suitable for use in commercial applications in the longer term. Formula D, with low feasibility and environmental scores and 6 weeks of shelf life appears less amenable for large-scale application. Formulae A and C are formulated to have an eight-week shelf life and to give moderate percentage effectiveness but have lower feasibility and environmental benefit scores, indicating that they may not be well suited for sustainable, cost-effective, new pest management options. Mawar *et al.* (2021) noted that biopesticides that have formula instability and poor ecological compatibility as factors cannot be accepted in the market use in the agricultural sector (Mawar *et al.* 2021). Therefore, Formula E is the most rounded and promising biopesticide formula out of the list of formulae studied; it is well-stable, efficient, environmentally sustainable, and economically affordable. Although having quick dispersion and high initial efficacy, the short shelf life of formula G prohibits its practical use. Optimization of formulae B, C and D, regarding shelf life, environmental impact and commercial feasibility in terms of scalability is required. This aligns with the suggestions made by Stejskal *et al.* (2021), underlined the need to arrive at a balanced formulation that should be active under the prevailing commercial storage conditions and, at the same time, possess an attribute of being reasonably priced as well as environmentally friendly (Stejskal *et al.* 2021). Results from overall testing show that formula E provides the best compromise in maximizing long-term effectiveness with the least impact on product sustainability and, therefore, would give a good candidate for future development and commercialization in the biopesticide market.

CONCLUSION

The study of the development and evaluation of the *Metarhizium anisopliae* conidium tablet formulae provided useful information on the effectiveness, stability and possible practical applications of the different formulae thanatology. Consistent evaluation of formula E with other formulate regarding disintegration time, viability retention, insecticidal efficacy, and their commercial feasibility further revealed that formula E always outshone other formulate in various aspects. Out of all the formulae, E possessed the most ideal shelf life (12 weeks), lowest viability loss and highest insecticidal efficacy and is the most promising for long term pest control. Although it performed very well initially, formula G has a shorter shelf life, making it less effective for use with a long shelf life. These results have significant practical applications for enhancing biopesticide technologies as replacements for chemical pesticides needed for sustainable agricultural practices. Due to the stability and efficacy of formula E, it is suitable for use in integrated pest management (IPM) systems in which short-term and long-term pest controls are required. Additionally, formulation and storage optimization are important for assuring the viability of the fungus and consistency of pest control over time. The next steps in the research would be to further optimize formula E for increased efficacy and stability. This could include testing the viability of spores in different excipients or encapsulation methods and developing control mechanisms for release. Furthermore, the formulae stability needs to investigate the influence of environmental factors (e.g. temperature, humidity and light) on the shelf-life extension. Future research can also be pursued to evaluate the field efficacy of these formulae in the field, because laboratory results are unlikely to give a realistic view of the field scenario. As for possible improvements, attention should be placed on increasing the formula's resistance to environmental stressors like UV radiation, which can harm fungal spores. Also, the storage conditions, specifically by protective packaging or other drying techniques, could preserve conidia viability for extended periods. Furthermore, optimizing and applying the specific dosage for different pests and performing more extended bioassay tests under various environmental conditions would further refine the overall effectiveness and practical application of these biopesticides for massive pest control.

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