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RESEARCH PAPER

Kaplan-meier-based survival analysis of *Cylas formicarius* following exposure to stored *Metarhizium anisopliae* formulations

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ABSTRACT

The sweet potato weevil (*Cylas formicarius*) is a major pest of sweet potato crops, particularly in tropical regions. The entomopathogenic fungus *Metarhizium anisopliae* is a promising biological control agent; however, its application is often constrained by reduced viability during storage. This study evaluated the efficacy of a dry powder formulation of *M. anisopliae* stored under ambient tropical conditions for 1 to 6 months. Bioassays assessed conidial density, adult mortality, lethal time to kill 50% of the population (LT_{50}), and survival probability using Kaplan–Meier analysis. Formulations stored for up to 3 months maintained high efficacy, with mortality rates exceeding 82.9%, conidial densities above 2.5×10^8 conidia/g, and LT_{50} values below 120 hours. In contrast, storage beyond three months significantly reduced conidial viability, increased LT_{50}, and decreased mortality. Kaplan–Meier survival curves showed a clear decline in virulence with increasing storage duration, with the 6-month formulation exhibiting the slowest mortality progression. Significant differences in survival probabilities among storage durations were confirmed statistically ($p < 0.05$). Morphological observations confirmed fungal-induced mortality, characterized by cuticle darkening, tissue softening, mummification, and external sporulation. Conidial deterioration over time was likely associated with physiological factors such as oxidative stress, lipid peroxidation, and depletion of protective compounds including trehalose and mannitol. Overall, storage duration critically affected the bioefficacy of *M. anisopliae.* It is therefore recommended that dry formulations be used within three months of production to ensure optimal pest control. These findings provide practical guidance for improving fungal biopesticide shelf life in tropical integrated pest management programs.

Keywords: Biological control, conidial viability, dry powder formulation, entomopathogenic fungi, Kaplan-Meier method

INTRODUCTION

In tropical and subtropical countries, including Indonesia, sweet potatoes (*Ipomoea batatas* L.) is a strategically important food crop due to its high economic value and significant contribution to food security (Kim et al., 2018). However, *Cylas formicarius*, a major pest of sweet potato, causes severe damage both in the field and during post-harvest storage, posing a serious threat to crop productivity (Schloemer et al., 2025). Larvae of *C. formicarius* burrow into the storage roots, causing direct physical damage and facilitating secondary infections, while adults feed on vines and roots, thereby exacerbating yield losses.

Most current control strategies rely on synthetic chemical pesticides, which, although effective, pose substantial health and environmental risks. These include the development of insect resistance, residue accumulation in food products, and adverse effects on non-target organisms (Khan et al., 2020). Consequently, there is growing interest in environmentally friendly and effective alternative pest management strategiess.

Entomopathogenic fungi such as *Metarhizium anisopliae* have been widely recognized as promising biological control agents for managing insect pests, including coleopterans such as *C. formicarius* (Reddy et al., 2014). Previous studies have demonstrated the entomopathogenic potential of *M. anisopliae*, including its effects on insect mortality and lethal time (LT_{50}). Prastowo et al. (2024) reported increased mortality and reduced LT_{50} in *Plutella xylostella* following application of *M. anisopliae* combined with botanical extracts, highlighting the importance of formulation components and external factors in determining fungal efficacy.

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However, their practical application is often constrained by limited shelf life and reduced viability during storage. Several studies have demonstrated that storage conditions strongly influence fungal persistence and virulence. For example, the viability of *M. anisopliae* decreases markedly under unfavorable storage temperatures (Jeong et al., 2022), while formulation and packaging methods significantly affect long-term stability (Elkhateeb et al., 2021; Teixidó et al., 2022). Consequently, physiological deterioration of conidia during storage remains a major challenge for large-scale application (Mascarin et al., 2024).

Dry powder formulations have been proposed to enhance shelf life and facilitate handling of entomopathogenic fungi. However, information on the effect of storage duration of dry powder formulations of *M. anisopliae* under tropical ambient conditions on their bioefficacy against *C. formicarius* remains limited, particularly under environments characterized by fluctuating temperature and humidity.

Therefore, this study evaluated the effect of storage duration (one to six months) on the efficacy of a dry powder formulation of *M. anisopliae* against adult *C. formicarius*. The findings are expected to provide valuable insights into formulation stability and to support the development of more reliable fungal biological control agents for use in integrated pest management programs.

MATERIALS AND METHODS

Research Site. This study was conducted at the Kupang State Agricultural Polytechnic, Indonesia, from June to September 2024. Field sampling of *C. formicarius* was carried out in several sweet potato farms located in South Central Timor Regency, East Nusa Tenggara, Indonesia. Laboratory activities, including fungal cultivation, bioformulation preparation, and bioassay experiments, were performed at the Plant Protection Laboratory, Department of Food Crops and Horticulture, Kupang State Agricultural Polytechnic.

Experimental Design. The bioassay experiment was arranged in a completely randomised design (CRD) with a single factor, namely storage duration of the bioinsecticide formulation. Six treatment levels were applied: P1, P2, P3, P4, P5, and P6, corresponding to storage periods of one, two, three, four, five, and six months, respectively. Each treatment was replicated four times, resulting in a total of 24 experimental units. Each unit consisted of ten adult *C. formicarius*, giving a total of 240 insects used in the experiment. Insect mortality data were recorded following exposure to each formulation and used to evaluate the effect of storage duration on bioinsecticide efficacy.

Insect Collection and Rearing. Adult *C. formicarius* were collected from infested sweet potato fields to establish the experimental population. Although both larval and adult stages were encountered, only adults were used as parent stock. Field-collected adults were mass-reared for one laboratory generation (F_1) on healthy sweet potato tubers placed in plastic containers (20 cm diameter × 20 cm height). Rearing for one generation was intended to obtain a physiologically uniform and laboratory-adapted population, thereby minimizing variability associated with field-collected insects. Adults of uniform age from the F_1 generation were subsequently selected and used as test insects in the bioassay.

Preparation and Storage of Bioinsecticide Formulation. The bioinsecticide substrate consisted of 100 g of rice bran sterilized by autoclaving at 121 ºC and 15 psi for 20 min. After cooling to room temperature, the sterilized rice bran was inoculated with *M. anisopliae* culture and incubated until complete fungal colonization was achieved. The colonized substrate was then dried and milled to obtain a dry powder formulation of *M. anisopliae*.

The dry powder formulation was stored at room temperature for different durations, ranging from one to six months. Storage periods were designated as P1 to P6, representing one, two, three, four, five, and six months of storage, respectively.

Conidial Density. Conidial density of the bioinsecticide formulation was determined using a haemocytometer under a light microscope at 400× magnification. A 1 g sample of fungal biomass was suspended in 100 mL of sterile distilled water, homogenized, and filtered. A 1 mL aliquot of the suspension was loaded into the haemocytometer, and conidia were counted in five large squares and sixteen small squares. Each sample was analyzed in duplicate. Conidial density was calculated using the standard haemocytometer formula:

$$C = \frac{t \times d}{0.25 \times n} \times 100\%$$

C = Conidial density;
t = Total number of conidia counted;
n = Number of squares counted;
d = Dilution factor (10^7);
0.25 = Constant.

Bioassay Procedure. Bioassays were conducted using the CRD described above. Adult *C. formicarius* were exposed to the dry powder formulation corresponding to each storage duration. Mortality was recorded daily, and cumulative mortality data were used to assess the efficacy of the stored formulations.

Observed Parameters. The primary parameter observed was adult *C. formicarius* mortality. Mortality was calculated based on the number of dead and surviving individuals and expressed as a percentage using the following formula:

$$M = \frac{r}{n} \times 100\%$$

M= Mortality;
r = Number of dead insects;
n = Total number of insects.

To correct for natural mortality in the control group, mortality data were adjusted using Abbott's formula (Subagiya et al., 2009):

$$CM = \frac{P - P_0}{100 - P_0} \times 100\%$$

CM= Corrected mortality;
P = The percent mortality of treated insects;
P_0 = The percent mortality of insects in the untreated control.

The LT_{50} values were estimated based on cumulative corrected mortality data using probit analysis.

Infection Symptoms and Confirmation of Mortality. Infection symptoms were observed to confirm fungal-induced mortality in adult *C. formicarius*. Typical symptoms included cuticle darkening, softening of body tissues, and visible fungal growth on the insect surface. Infected individuals often exhibited mummification, indicating systemic infection by *M. anisopliae*. The presence of fungal sporulation on insect cadavers was used as confirmation that mortality resulted from fungal infection.

Data Analysis. All quantitative data were analyzed using analysis of variance (ANOVA). When significant differences among treatments were detected, mean values were separated using the Least Significant Difference (LSD) test at a 5% significance level (α = 0.05). The lethal time required to kill 50% of the insect population (LT_{50}) was estimated using probit analysis based on corrected cumulative mortality data.

Mortality dynamics over time were analyzed using the Kaplan–Meier survival method based on daily mortality observations from day 0 (day of application) to day 5 post-treatment. Survival probabilities were calculated stepwise, with the initial survival probability defined as $S(t_0) = 1$ at day 0, indicating that all individuals were alive at the beginning of the observation period. Kaplan–Meier survival curves were generated to compare the virulence of *M. anisopliae* formulations stored for different durations (1–6 months).

To statistically compare survival distributions among the six storage treatments, log-rank (Mantel–Cox) tests were performed. Pairwise comparisons were conducted to assess whether prolonged storage significantly reduced the virulence of *M. anisopliae*. A significance level of $\alpha = 0.05$ was applied for all statistical tests.

Differences in survival distributions among treatments were statistically evaluated using the log-rank (Mantel–Cox) test. Pairwise comparisons were performed to assess the effect of prolonged storage on fungal virulence. All statistical analyses were conducted at a significance level of $\alpha = 0.05$. Survival analysis and curve plotting were performed using Microsoft Excel.

RESULTS AND DISCUSSION

Insect Mortality. The mortality of adult *C. formicarius* varied significantly among storage durations of the *M. anisopliae* dry powder formulations (Figure 1). The highest mortality was observed in the 1-month storage treatment (P1) (82.93%), followed by the 2-month (71.48%) and 3-month (66.53%) treatments. Prolonged storage resulted in a gradual decline in mortality, with the lowest value recorded in the 6-month treatment (P6) (54.98%).

The progressive reduction in insect mortality indicates a decline in conidial viability and virulence during storage. Physiological deterioration processes, such as desiccation stress, oxidative damage, and depletion of endogenous nutrient reserves, are known to impair conidial germination and host infection capacity (Mascarin et al., 2024). Similar reductions in the efficacy of entomopathogenic fungi following extended storage have been reported previously (Elkhateeb et al., 2021; Jeong et al., 2022). In contrast, the incorporation of protective additives, such as chitin, has been shown to enhance fungal virulence and insect mortality, as reported for *Beauveria bassiana* against *C. formicarius* (Saputro et al., 2019).

Effects of Formulation and Carrier Materials. The dry powder formulation used in this study employed carrier materials such as kaolin, zeolite, and maize flour, which are commonly used in fungal bioinsecticide formulations. These materials differ in their capacity to protect fungal conidia during storage. Kaolin provides physical separation of particles but offers limited protection against moisture fluctuations. Zeolite possesses moisture-adsorbing properties that may help stabilize conidia under humid conditions, whereas maize flour, although functioning as a filler and dispersant, may increase the risk of microbial contamination if inadequately sterilized.

Previous studies have demonstrated that improved formulation strategies, including microencapsulation, the use of hydrophobic polymers, and the incorporation of protective compounds such as trehalose or glycerol, can significantly enhance fungal shelf life by reducing desiccation stress and metabolic degradation (Moeini et al., 2022; Řepka et al., 2023; Baghi et al., 2022). These approaches may represent promising options for improving the stability of *M. anisopliae* formulations under tropical storage conditions

Comparison with Similar Studies. The mortality patterns observed in this study are consistent with previous reports demonstrating the sensitivity of entomopathogenic fungi to storage conditions. Modified atmosphere packaging (MAP) using ethylene vinyl alcohol (EVOH) film with a gas composition of 30% CO_2 and 70% N_2 was reported to maintain 80.5% viability of dried *M. anisopliae* conidia after 28 days (Jeong et al., 2022). When combined with cold storage (4 ºC), conidial shelf life was further extended, highlighting the importance of atmospheric composition and temperature control in preserving fungal viability.

In contrast, storage under ambient and uncontrolled conditions resulted in a significant decline in conidial viability and virulence (Jeong et al., 2022; Elkhateeb et al., 2021). These findings are in agreement with the gradual reduction in insect mortality observed in the present study as storage duration increased. Similar trends have been reported for other microbial-based products, where modified atmospheres stabilized microbial populations and improved storage performance (Ratajczak et al., 2022; Rovira et al., 2023), supporting the broader applicability of MAP technology in maintaining biological product quality.

This study demonstrates how entomopathogenic fungi, like strains of *B. bassiana* and *Metarhizium* strains, can be used to effectively manage pests. It was discovered that using local isolates and optimising the substrate greatly increased bioefficacy. For instance, chitin-enriched *B. bassiana* cultures achieved a 91.67% mortality rate against *C. formicarius*, and maintained 82.93% efficacy even after one month of non-refrigerated storage (Chaithra et al., 2022; Teixidó et al., 2022). Likewise, *Metarhizium* strains grown on rice and sorghum substrates caused 100% mortality in *Coptotermes*, highlighting the critical role of substrate selection (Chaithra et al., 2022).

These comparison observations support the study's conclusions while highlighting areas that could want more optimisation, particularly with regard to formulation design and storage conditions.

Conidial Density. Storage duration had a significant effect on conidial density, expressed as the number of viable conidia per gram of formulation (Figure 2). The highest conidial density was observed in the 1-month

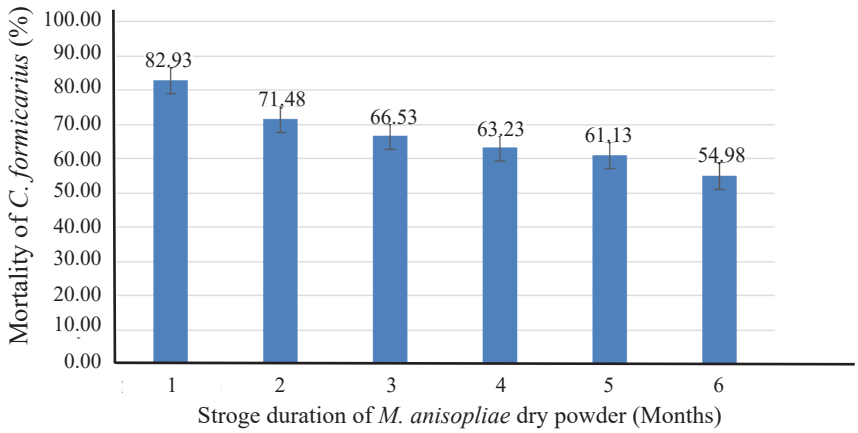

Figure 1. Mortality of *C. formicarius* following application of *M. anisopliae* dry powder formulations stored for different durations.

storage treatment (2.50×10^8 conidia/g), whereas a marked decline occurred with extended storage, reaching the lowest value in the 3-month treatment (1.36×10^8 conidia/g).

Reduced conidial density directly influences the pathogenic potential of fungal formulations, as higher spore concentrations increase the probability of host adhesion, germination, and cuticle penetration (Deka et al., 2021). The decline in conidial density observed in this study likely contributed to the reduced mortality of *C. formicarius* with increasing storage duration. Similar relationships between conidial density and bioefficacy have been reported for *Metarhizium* isolates cultivated on low-cost substrates, which achieved high conidial yields and strong pathogenicity (Zulfiana et al., 2020).

Lethal Time (LT_{50}) Values. Storage duration significantly affected the LT_{50} values of the dry powder formulation of *M. anisopliae* against adult *C. formicarius* (Figure 3). The shortest LT_{50} was recorded in the 1-month storage treatment (96 hours), indicating rapid insect mortality associated with high conidial infectivity. In contrast, the LT_{50} increased to 126.5 hours after 3 months of storage, reflecting a decline in fungal virulence as storage time increased.

Lower LT_{50} values are generally associated with higher conidial viability and faster host colonization. *M. anisopliae* infects insect hosts through cuticular penetration and produces extracellular enzymes and secondary metabolites, including destruxins, which disrupt cellular homeostasis and suppress insect immune responses, ultimately leading to host mortality (Williams et al., 2021; Hughes et al., 2022). Prolonged storage may reduce conidial physiological quality and metabolic activity, thereby delaying infection processes and increasing LT_{50} values.

In this study, LT_{50} estimation was based on a 5-day observation period. In several treatments, cumulative mortality did not reach the optimal 10–90% range required for probit analysis. Extending the observation period could have introduced bias due to natural mortality unrelated to fungal infection. Therefore, LT_{50} values were interpreted in conjunction with Kaplan–Meier survival analysis to provide a more reliable description of mortality dynamics among

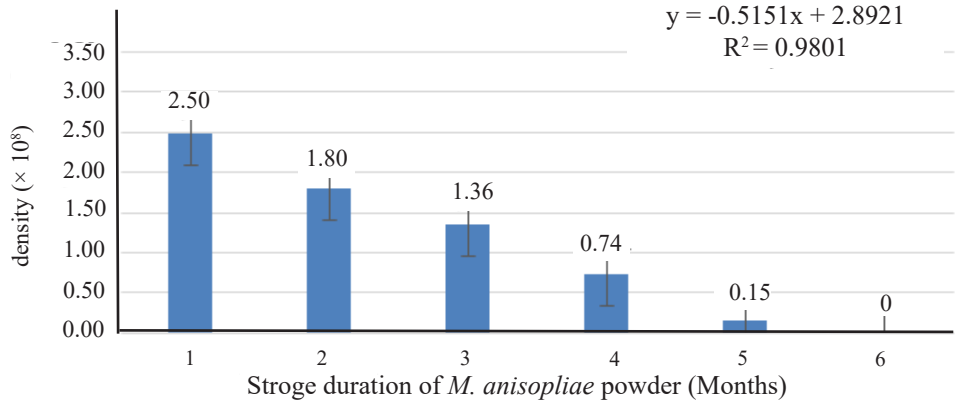

Figure 2. Conidial density of *M. anisopliae* dry formulations after various storage durations.

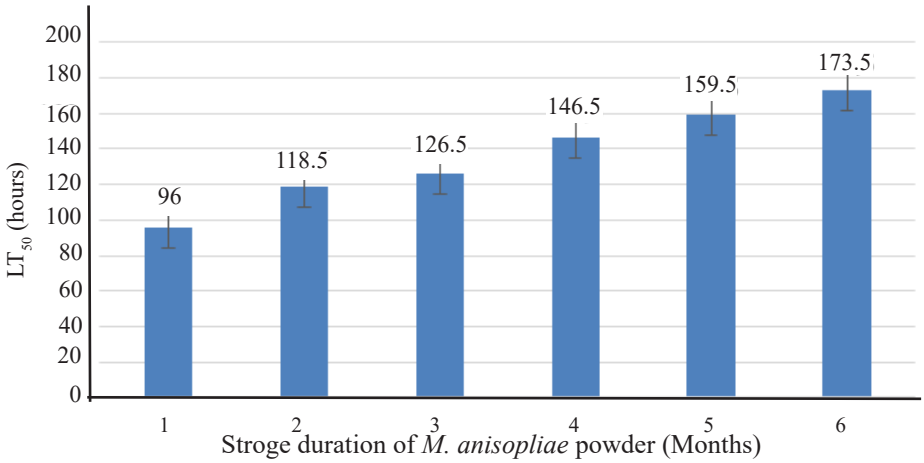

Figure 3. LT_{50} values of *M. anisopliae* formulations against *C. formicarius* after different storage durations.

storage treatments.

Morphological Characteristics of Infected Insects. Infection by *M. anisopliae* caused distinct morphological changes in adult *C. formicarius* (Table 1). Early symptoms included reduced mobility and darkening of the cuticle. These changes were followed by tissue softening, progressive blackening, and eventual mummification of the insect body. At the final stage of infection, mycelial growth and sporulation were observed on the insect surface, indicating complete fungal colonization and host death.

The observed morphological alterations reflect the successful infection process of *M. anisopliae*, beginning with conidial adhesion and cuticle penetration, followed by hyphal proliferation within the hemocoel. During this stage, the fungus exploits host nutrients and causes extensive tissue degradation, leading to systemic physiological disruption and insect mortality (Kiruthiga et al., 2022; Liu et al., 2023). Reduced movement and lethargy in infected insects indicate impairment of neuromuscular and metabolic functions as the infection progresses.

External sporulation observed at later stages of infection is typical of entomopathogenic fungi under favorable humidity conditions and facilitates horizontal transmission to other hosts (Lei et al., 2021; Zhang et al., 2021). However, the effectiveness of this infection process is strongly influenced by conidial physiological quality. Prolonged storage may induce oxidative stress and depletion of endogenous protective compounds such as trehalose and mannitol, resulting in reduced membrane integrity, lower germination capacity, and diminished infectivity (Chen et al., 2022; Wu & Wong, 2022). Under ambient tropical conditions, fungal conidia are particularly susceptible to gradual physiological deterioration during storage. Similar longevity and dormancy-related mechanisms have been reported in other fungal groups, where metabolic downregulation during storage supports spore survival but is often accompanied by a progressive loss of infectivity over time (Shemesh et al., 2023).

Trehalose and mannitol play important roles in stabilizing proteins and membranes and function as osmoprotectants during desiccation and thermal stress. Declining levels of these metabolites during prolonged storage increase conidial susceptibility to denaturation and mechanical damage. Trehalose accumulation has been associated with improved stress tolerance and survival in fungi and insects, while mannitol has been shown to enhance spore yield and tolerance to heat and osmotic stress in *Trichoderma harzianum* and other fungal species (Zhu et al., 2022; Guo et al., 2022).

Collectively, these physiological changes explain the longer LT_{50} values and lower mortality rates observed in older formulations, indicating a measurable decline in conidial viability and virulence. These findings highlight the importance of formulation and storage optimization, including the use of protective additives or controlled storage environments, to improve the shelf life and field performance of fungal biopesticides.

Survival Analysis of *C. formicarius* Based on Kaplan-Meier Curves. Kaplan–Meier survival analysis was used to describe the temporal dynamics of *C. formicarius* mortality following exposure to dry powder formulations of *M. anisopliae* stored for different durations (1–6 months) (Figure 4). Survival probabilities were calculated from daily mortality records to evaluate changes in fungal virulence over time.

The Kaplan–Meier curves showed a clear decline in bioefficacy with increasing storage duration. The

Table 1. Progression of fungal infection symptoms in *C. formicarius* after treatment

Post-inoculation time	Visible symptoms	Notes
0–24 hours	No obvious physical changes	Adaptation phase; conidia begin to adhere to the insect cuticle.
24–48 hours	Initial body color change: light brown → dark brown	Conidia begin to germinate and penetrate the cuticle with hydrolytic enzymes (proteases, lipases, chitinases).
48–72 hours	Body begins to soften and appears lethargic	Haemocoel invasion begins; the fungus spreads systemically.
72–96 hours	Body turns black; integument becomes fragile	Blastospore production and internal sporulation increase.
96–120 hours	Body becomes dark black and hardens (mummy)	Final infection phase; death occurs, and the fungus is ready for external sporulation.

formulation stored for one month (P1) caused a rapid decrease in survival probability, reaching complete mortality (S(t) = 0.0) by day 4 (96 h), indicating high conidial viability and strong infectivity. In contrast, insects exposed to the six-month-old formulation (P6) exhibited substantially higher survival, with approximately 45% of individuals still alive at day 5, reflecting reduced pathogenicity and slower infection rates. Intermediate storage periods (2–5 months) showed a gradual and proportional reduction in survival, consistent with progressive loss of conidial quality. Similar approaches linking virulence and time-to-death parameters were applied by Ponijan et al. (2023) in stink bug mortality assessments under different bioinsecticide treatments, supporting the use of Kaplan–Meier survival analysis as a robust tool for evaluating temporal differences in bioinsecticide efficacy.

Statistical comparison using the log-rank (Mantel–Cox) test revealed significant differences among survival distributions across treatments ($p < 0.05$). Pairwise comparisons indicated that the one-month formulation was significantly more effective than formulations stored for four months or longer ($p < 0.01$), confirming that prolonged storage markedly reduces fungal virulence.

These findings demonstrate that storage duration is a critical factor affecting the survival dynamics of *C. formicarius* and the overall performance of *M. anisopliae*. The Kaplan–Meier analysis supports results obtained from mortality rates, conidial density, and LT_{50} values, all of which showed declining bioefficacy with extended storage. Similar reductions in fungal viability and pathogenicity under ambient storage conditions have been reported previously (Elkhateeb et al., 2021; Jeong et al., 2022). Collectively, the results indicate that three months represents a practical threshold for maintaining effective virulence of dry powder formulations of *M. anisopliae* under tropical conditions, providing important guidance for their use in integrated pest management programs.

CONCLUSION

This study established the practical shelf life of dry powder formulations of Metarhizium anisopliae under tropical environmental conditions. By combining data on mortality, conidial density, LT_{50}, and Kaplan-Meier survival, we consistently found that storage for more than three months resulted in a significant reduction in virulence. Unlike previous studies that focused on short-term culture or cold storage, our findings identify a critical threshold of three months for maintaining viability and efficacy in fluctuating tropical environments. This provides a novel and practical contribution to fungal biopesticide research, offering clear, evidence-based guidance for their application in tropical integrated pest management (IPM) programs.

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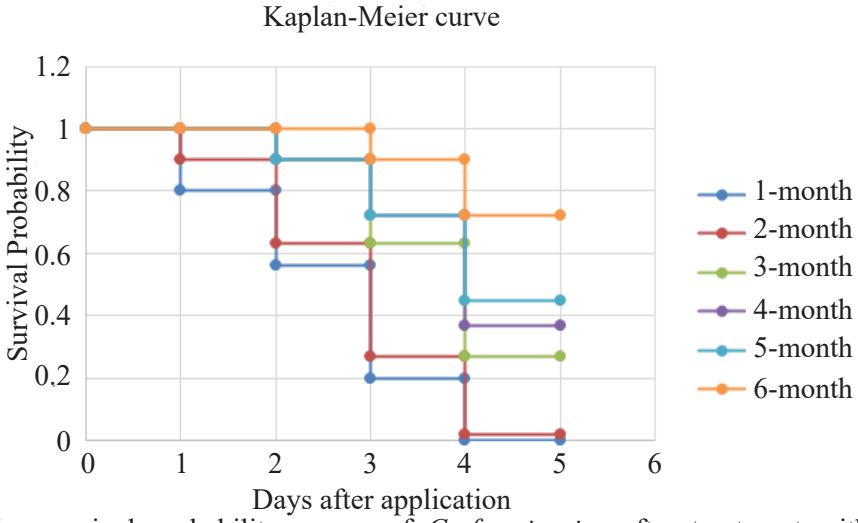

Figure 4. Kaplan-Meier survival probability curves of *C. formicarius* after treatment with *M. anisopliae* formulations stored for 1 to 6 months.

AUTHORS' CONTRIBUTIONS

YFL was primarily responsible for data collection, ensuring all relevant data for the study were gathered accurately and consistently throughout the research process. JHHS took charge of the data processing and interpretation, ensuring that the raw data collected was analyzed effectively to derive meaningful conclusions, and played a key role in interpreting the results. JAB contributed significantly to the development of the methodology, guiding the research design and ensuring the proper techniques and procedures were followed for accurate and reliable results. NJL served as the coordinator of the research, overseeing the entire project. She was responsible for writing the manuscript, including drafting the initial version, leading the revisions, and finalizing the document to ensure the research was presented clearly and coherently.

COMPETING INTEREST

The authors declare no competing interests.

REFERENCES

Baghi F, Gharsallaoui A, Dumas E, & Ghnimi S. 2022. Advancements in biodegradable active films for food packaging: Effects of nano/microcapsule incorporation. *Foods*. 11(5): 760. https://doi.org/10.3390/foods11050760

Chaithra M, Prameeladevi T, Bhagyasree SN, Prasad L, Subramanian S, & Kamil D. 2022. Multilocus sequence analysis for population diversity of indigenous entomopathogenic fungus *Beauveria bassiana* and its bio-efficacy against the cassava mite, *Tetranychus truncatus* Ehara (Acari: Tetranychidae). *Front. Microbiol*. 13: 1007017. https://doi.org/10.3389/fmicb.2022.1007017

Chen Y, Wan Y, Cai W, Liu N, Zeng J, Liu C, Peng H, & Fu G. 2022. Effects on cell membrane integrity of *Pichia anomala* by the accumulating excessive reactive oxygen species under ethanol stress. *Foods*. 11(22): 3744. https://doi.org/10.3390/foods11223744

Deka B, Babu A, Peter AJ, Kumhar KC, Sarkar S, Rajbongshi H, Dey P, Amalraj ELD, & Talluri VR. 2021. Potential of the entomopathogenic fungus, *Metarhizium anisopliae s.l.* in controlling live-wood eating termite, *Microtermes obesi* (Holmgren) (Blattodea: Termitidae) infesting tea crop. *Egypt. J. Biol. Pest Control*. 31: 132. https://doi.org/10.1186/S41938-021-00477-4

Elkhateeb WA, Mousa KM, ELnahas MO, & Daba GM. 2021. Fungi against insects and contrariwise as biological control models. *Egypt. J. Biol. Pest Control*. 31: 13. https://doi.org/10.1186/S41938-020-00360-8

Guo H, Xiong X, Wang Y, Tian H, Zhang S, & Geng G. 2022. Integrative proteomic and physiological analyses of the molecular response to dessication-stress in *Auricularia fibrillifera*. *Front. Plant Sci*. 13: 995810. https://doi.org/10.3389/fpls.2022.995810

Hughes KM, Price D, Torriero AAJ, Symonds MRE, & Suphioglu C. 2022. Impact of fungal spores on asthma prevalence and hospitalization. *Int. J. Mol. Sci*. 23(8): 4313. https://doi.org/10.3390/ijms23084313

Jeong SG, Kim HM, Kim J, Kim JS, & Park HW. 2022. Effect of storage conditions on the shelf-life extension of fungus-colonized substrates based on *Metarhizium anisopliae* using modified atmosphere packaging. *Sci. Rep*. 12: 423. https://doi.org/10.1038/S41598-021-04232-5

Khan FA, Ibrahim AA, & Zeki AM. 2020. Environmental monitoring and disease detection of plants in smart greenhouse using internet of things. *J. Phys. Commun*. 4(5): 055008. https://doi.org/10.1088/2399-6528/ab90c1

Kim HS, Lee CJ, Kim SE, Ji CY, Kim ST, Kim JS, Kim S, & Kwak SS. 2018. Current status on global sweetpotato cultivation and its prior tasks of mass production. *J. Plant Biotechnol*. 45(3): 190–195. https://doi.org/10.5010/JPB.2018.45.3.190

Kiruthiga G, Jeyarani S, Sathiah N, Murugan M, Sivakumar U, & Uma D. 2022. Pathogenicity, ultra-structural growth and development of green muscardine fungus, *Metarhizium anisopliae* (Metschnikoff) Sorokin (Ascomycota: Hypocreales) on maize fall armyworm, *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae). *Egypt. J. Biol. Pest Control*. 32: 97. https://doi.org/10.1186/s41938-022-00596-6

Lei Y, Hussain A, Guan Z, Wang D, Jaleel W, Lyu L, & He Y. 2021. Unraveling the mode of action of *Cordyceps fumosorosea*: Potential biocontrol

agent against *Plutella xylostella* (Lepidoptera: Plutellidae). *Insects*. 12(2): 179. https://doi.org/10.3390/insects12020179

Liu D, Smagghe G, & Liu TX. 2023. Interactions between entomopathogenic fungi and insects and prospects with Glycans. *J. Fungi*. 9(5): 575. https://doi.org/10.3390/jof9050575

Mascarin GM, Golo PS, de Souza Ribeiro-Silva C, Muniz ER, de Oliveira Franco A, Kobori NN, & Fernandes ÉKK. 2024. Advances in submerged liquid fermentation and formulation of entomopathogenic fungi. *Appl. Microbiol. Biotechnol*. 108: 451. https://doi.org/10.1007/S00253-024-13287-z

Moeini A, Pedram P, Fattahi E, Cerruti P, & Santagata G. 2022. Edible polymers and secondary bioactive compounds for food packaging applications: antimicrobial, mechanical, and gas barrier properties. *Polymers*. 14(12): 2395. https://doi.org/10.3390/polym14122395

Ponijan P, Handayani EP, Kurniawati N, Rakhmiati R, & Zulkarnaen. 2023. Joint application of *B. bassiana* and *M. anisopliae* bioinsecticides for controlling rice bugs and improving rice yields. *J. Trop. Plant Pests Dis*. 23(2): 58–64. https://doi.org/10.23960/jhptt.22358-64

Prastowo S, Wardhani WK, Pradana AP, Wafa A, Addy HS, & Hadi ZFN. 2024. *Metarhizium anisopliae* Metsch. (Moniliales: Moniliaceae) and *Aglaia odorata* Lour. (Sapindales: Maliaceae) leaf extract: A potent biopesticide cocktail for cabbage leaf caterpillar management. *J. Trop. Plant Pests Dis.* 24(2): 212–222. https://doi.org/10.23960/jhptt.224212-222

Ratajczak K, Staninska-Pięta J, Czarny J, Cyplik P, Wolko Ł, & Piotrowska-Cyplik A. 2022. Effect of processing treatment and modified atmosphere packing on carrot's microbial community structure by Illumina MiSeq sequencing. *Molecules*. 27(9): 2830. https://doi.org/10.3390/molecules27092830

Reddy GVP, Zhao Z, & Humber RA. 2014. Laboratory and field efficacy of entomopathogenic fungi for the management of the sweetpotato weevil, *Cylas formicarius* (Coleoptera: Brentidae). *J. Invertebr. Pathol*. 122: 10–15. https://doi.org/10.1016/j.jip.2014.07.009

Řepka D, Kurillová A, Murtaja Y, & Lapčík L. 2023. Application of physical-chemical approaches for encapsulation of active substances in pharmaceutical and food industries. *Foods*. 12(11): 2189. https://doi.org/10.3390/foods12112189

Rovira P, Brugnini G, Rodriguez J, Cabrera MC, Saadoun A, de Souza G, Luzardo S, & Rufo C. 2023. Microbiological changes during long-storage of beef meat under different temperature and vacuum-packaging conditions. *Foods*. 12(4): 694. https://doi.org/10.3390/foods12040694

Saputro TB, Prayogo Y, Rohman FL, & Alami NH. 2019. The virulence improvement of *Beauveria bassiana* in infecting *Cylas formicarius* modulated by various chitin based compounds. *Biodiversitas*. 20(9): 2486–2493. https://doi.org/10.13057/biodiv/D200909

Schloemer CM, Graham SH, & Lawrence KS. 2025. Sweetpotato pest challenges and management options. *J. Integ. Pest Manag*. 16(1): 4. https://doi.org/10.1093/jipm/pmaf003

Shemesh H, Bruns TD, Peay KG, Kennedy PG, & Nguyen NH. 2023. Changing balance between dormancy and mortality determines the trajectory of ectomycorrhizal fungal spore longevity over a 15-yr burial experiment. *New Phytol.* 238(1): 11–15. https://doi.org/10.1111/NPH.18677

Subagiya, Himawati MK, & Wulandari P. 2009. Pengujian toksisitas ekstrak daun wedusan (*Ageratum conyzoides*) terhadap ulat jantung kubis (Crocidolomia binotalis Zeller) [Toxicity Assessment of Wedusan Leaf Extract (*Ageratum conyzoides*) Against the Cabbage Heart Caterpillar (Crocidolomia binotalis Zeller)]. *Caraka Tani.* 24(2): 164–169

Teixidó N, Usall J, & Torres R. 2022. Insight into a successful development of biocontrol agents: Production, formulation, packaging, and shelf life as key aspects. *Horticulturae*. 8(4): 305. https://doi.org/10.3390/horticulturae8040305

Williams TJ, Gonzales-Huerta LE, & Armstrong-James D. 2021. Fungal-induced programmed cell death. *J. Fungi*. 7(3): 231. https://doi.org/10.3390/jof7030231

Wu H & Wong JWC. 2022. Temperature versus relative humidity: Which is more important for indoor mold prevention?. *J. Fungi*. 8(7): 696. https://doi.org/10.3390/jof8070696

Zhang X, Meng Y, Huang Y, Zhang D, & Fang W. 2021. A novel cascade allows *Metarhizium robertsii* to distinguish cuticle and hemocoel microenvironments during infection of insects. *PLoS Biol*. 19(8): e3001360. https://doi.org/10.1371/journal.pbio.3001360

Zhu X, Wang Y, Wang X, & Wang W. 2022. Exogenous regulators enhance the yield and stress resistance of chlamydospores of the biocontrol agent *Trichoderma harzianum* T4. *J. Fungi*. 8(10): 1017. https://doi.org/10.3390/jof8101017

Zulfiana D, Zulfitri A, Lestari AS, Krishanti NPRA, & Meisyara D. 2020. Production of conidia by entomopathogenic fungi and their pathogenicity against *Coptotermes* sp. *Biosaintifika*. 12(1): 1–9. https://doi.org/10.15294/biosaintifika.v12i1.22435