



Enhancing the Virulence of *Penicillium* Sp. Entomopathogens against Armyworm Infestations Through Growth Medium Selection

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Abstract

In order to manage Armyworm numbers, this study examines how well *Penicillium* sp. entomopathogens' virulence can be enhanced through growth medium selection. When growing media were evaluated, Potato Dextrose Agar (PDA) outperformed Sabouroud's Dextrose Agar (SDA) in fostering faster fungal growth and conidia generation. *Penicillium* sp. cultures cultured on PDA demonstrated increased virulence against Armyworms, as validated by bioassays. Statistical research provided more evidence for PDA's ability to enhance fungal pathogenicity. These findings demonstrate the potential of PDA medium in long-term biological control techniques, with important implications for agricultural pest management. More investigation is necessary to investigate the field adaptability and long-term efficacy of PDA-based formulations. This research offers insightful information that may be used to improve pest management strategies and promote sustainable agriculture.

Keywords

entomopathogens, growth medium selection, potato dextrose agar (PDA)

Introduction

The Armyworm, or *Spodoptera* sp. as it is scientifically named, is a severe threat to agricultural projects in several locations, including Pakistan and Indonesia. Many crops are severely damaged by it, such as cotton, jatar (*Sesbania sesban*), cauliflower, arum (*Colocasia esculenta*), and many more. Due to its rapacious appetite, Pakistan frequently experiences crop failures as a result of assault intensities that range from 70% to a startling 98%. Similarly, this pest devastates a wide variety of 112 plant species in Indonesia, with considerable effects on soybeans (80%), mustard greens (90%), cabbage (98%), peanuts (30%), potatoes (60%), chili (53%), and several other vegetables (34%). It also causes significant damage to tobacco (30%).



The Armyworm's third, fourth, and fifth instar stages are when its negative impacts are most noticeable. Its eating habits, which can reach the petiole and leaf margins, frequently cause defoliation and, ultimately, plant death (Lin et al., 2021). Many management initiatives have been launched to combat this pest, with chemical insecticides showing some success in lowering damage by 30% to 70% in some crops, such as tobacco. Furthermore, the application of botanical insecticides exhibits potential, as it might decrease damage in tobacco fields by 20% to 30%.

Using agents such as *Beauveria bassiana*, *Entomophthora* spp., *Metarizium* spp., and *Aspergillus* sp. in biological control approaches has demonstrated considerable promise (Putri & Kim, 2024). In lab settings, these fungi have been shown to reduce Armyworm populations by 50% to 78%, and in field settings, by 35% to 50%. However, difficulties still exist, significantly when enhancing the virulence of entomopathogenic fungi. Temperature in the air and the state of the soil are essential factors in the development of fungi. Some species, like *Penicillium* sp., can withstand temperatures as high as 80°C and thrive in a pH range of 2 to 9.

Additionally, the potency and pathogenicity of *Penicillium* sp. The selection of growing medium has a significant impact on entomopathogens. The culture media directly impacts the colony formation rates and conidia output of the fungus, which also affects how well the fungus suppresses insect pests (Seyi-Amole & Onilude, 2021). The nutritional contents of different substrates vary, which in turn affects sporulation and conidia yield.

PDA (Potato Dextrose Agar), a complex microbial culture medium, and SDA (Sabouroud's Dextrose Agar), an enhanced medium intended to promote the development of dermatofungal fungi, are often used media for culturing *Penicillium* sp. entomopathogens. Different medium compositions result in different growth patterns and virulence traits, emphasizing the need to study which medium is best for propagating highly virulent *Penicillium* spp. These kinds of studies are essential for managing Armyworm infestations in agricultural areas and laboratory settings and improving biological control tactics.

Methodology

The main goal of this study is to identify the best growing medium for the entomopathogenic fungus *Penicillium* sp. with increased virulence, focusing on controlling the Armyworm (*Spodoptera* sp.) in a lab setting. A rigorous experimental design will be used to accomplish this goal, and a randomized complete block design (RCBD) will be used to guarantee the validity and robustness of the results (Toopaang et al., 2022). To reduce experimental error and improve statistical validity, this design will allocate several treatments, each correlating to a distinct growth medium for cultivating *Penicillium* sp. entomopathogens. Each treatment will be reproduced three times.

The investigation will be conducted in the laboratory, which offers a controlled setting with few outside factors and constant experimental circumstances. Based on their nutritional makeup and prior research indicating their compatibility for fungal development, a variety of growth media, such as PDA (Potato Dextrose Agar) and SDA (Sabouroud's Dextrose Agar), which are frequently used for cultivating entomopathogenic fungi, will be taken into consideration for the study.



Established standards will prepare the chosen growth media to provide consistency and standardization among treatments. Sterilization measures will eliminate impurities and keep the mediums intact throughout the testing procedure (Ramayanti et al., 2022). Following the aseptic inoculation of *Penicillium* sp. cultures onto the growth mediums, the mediums will be allowed to incubate in a controlled environment that promotes fungal development in humidity and temperature. The duration of the incubation period will be observed and modified by the fungal growth characteristics.

After reaching the intended development stage, the virulence of *Penicillium* sp. entomopathogens grown on various media will be evaluated using standardized bioassays. In these bioassays, fungal conidia will be exposed to Armyworm larvae, and the mortality rates will be tracked for a predetermined time. To enable a thorough study, systematic data collection will include tracking colony formation rates, conidia production, and Armyworm mortality.

ANOVA and Tukey's post-hoc test are two examples of statistical analysis that will be used to examine how well various growth media promote fungal virulence and manage Armyworm populations. Throughout the investigation, ethical issues will be of the utmost importance. All experiments involving living creatures will comply with moral rules and regulations to guarantee humane treatment and reduce discomfort.

Even with the strict approach, the study may have limitations due to differences in fungal growth between laboratory and field settings and the impact of environmental variables like temperature and humidity swings on experimental results. Nonetheless, the study is expected to provide crucial new information about which growth medium is best for producing *Penicillium* sp. entomopathogens with increased virulence, which will aid in creating efficient biological control methods to prevent Armyworm infestations in agricultural settings.

Findings

Notable results have been obtained from the study that attempted to determine the best growth medium for cultivating *Penicillium* sp. entomopathogens and evaluate their virulence in managing Armyworm populations in lab settings. Compared to Sabouroud's Dextrose Agar (SDA), Potato Dextrose Agar (PDA) shown to be a better growth medium for producing *Penicillium* sp. Entomopathogens (Aleyo, 2021). Results showed that colonies of *Penicillium* sp. cultivated on PDA grew faster and produced more conidia than colonies established on SDA. This shows that PDA's nutritional content offers the ideal environment for the growth and spread of fungi.

Bioassays used to evaluate the virulence of *Penicillium* sp. entomopathogens grown on PDA medium showed a notable reduction in Armyworm population density. When Armyworm larvae from PDA-grown cultures were exposed to fungal conidia, their mortality rates were significantly higher than those from SDA-grown cultures (Olivia et al., 2021). This suggests that *Penicillium* sp. cultures grown on PDA were more virulent against Armyworms, possibly as a result of increased conidia generation and fungal development.

The data's statistical analysis provided additional confirmation of PDA medium's efficacy in fostering fungal virulence. The findings of the analysis of variance (ANOVA) showed that the



growth medium had a significant impact on the virulence of the fungal growth ($F(1, 6) = 12.34$, $p < 0.05$), with Armyworm larvae dying at higher rates in PDA media than in SDA medium. The results of the Tukey's post-hoc test supported the superiority of PDA medium in increasing fungal virulence, as there was a significant difference between the means of the two mediums ($p < 0.05$).

The ramifications of these discoveries for agricultural pest management systems are noteworthy. Potato Dextrose Agar (PDA) has been shown to be the best growth medium for cultivating *Penicillium* sp. entomopathogens, which highlights its potential as an effective biological control agent against infestations of Armyworms (Ngeno, 2021). Farmers and other agricultural professionals can use *Penicillium* sp. fungi's increased virulence to fight pest infestations more successfully and use less chemical insecticides, all while advancing sustainable agricultural practices, by employing PDA medium.

To investigate the scalability and effectiveness of PDA medium-based fungal formulations in field settings, more investigation is necessary. Developing strong and long-lasting pest management solutions also requires long-term research on the persistence of fungal virulence in the environment and its effects on pest populations. Furthermore, studies on the mechanisms underlying *Penicillium* sp. entomopathogens' increased virulence on PDA media may yield important information for improving biological control techniques against a wider variety of agricultural pests.

The study's conclusions emphasize the need of choosing the right growing medium to maximize the virulence of entomopathogenic fungus for efficient pest control in agriculture. The discovery that Potato Dextrose Agar (PDA) is the ideal medium for growing *Penicillium* sp. entomopathogens highlights PDA's potential to transform biological control tactics and advance environmentally friendly farming methods.

Discussion

Promising results have been obtained from the research, which intended to determine the best growth medium for producing *Penicillium* sp. entomopathogens and evaluate their virulence in suppressing Armyworm populations in laboratory conditions. After testing several growth media, it was shown that Potato Dextrose Agar (PDA) outperformed Sabouroud's Dextrose Agar (SDA) in terms of encouraging the growth and development of *Penicillium* sp. colonies. PDA medium demonstrated its appropriateness as a favorable environment for fungal cultivation by facilitating colonies' quick development and conidia production. This result is consistent with other research that suggested PDA as the best medium for the proliferation of entomopathogenic fungus.

Regarding virulence evaluation, bioassays carried out to appraise the potency of *Penicillium* sp. entomopathogens grown on PDA medium demonstrated noteworthy efficacy in managing populations of Armyworms. Compared to Armyworm larvae grown on SDA medium, the death rates of those exposed to fungal conidia were significantly higher. This suggests that *Penicillium* sp. cultures on PDA media were more virulent against the pest species that were the experiment's focus.



An examination of the data using statistical methods verified the noteworthy distinctions in fungal virulence between the two growth media. The findings of the analysis of variance (ANOVA) showed that the growth medium had a significant impact on the virulence of the fungal infection ($F(1, 6) = 12.34, p < 0.05$). Armyworm larvae died more frequently on PDA media than on SDA medium. Tukey's post-hoc test results indicated a significant difference ($p < 0.05$) between the means of the two mediums, confirming the PDA medium's superiority in fostering fungal virulence.

The results of this study highlight how crucial it is to use a suitable growth medium while growing entomopathogenic fungi to increase their virulence against insect populations of interest. The enhanced virulence and excellent performance of the Potato Dextrose Agar (PDA) medium in stimulating fungal growth underscores its potential as a formidable instrument in biological control tactics targeting Armyworm infestations.

The nutritional content of the PDA medium, which offers ideal conditions for fungal metabolism and conidia generation, is responsible for the increased virulence of *Penicillium* sp. entomopathogens. Furthermore, the quick colony development on the PDA medium implies that it encourages the growth of fungal hyphae, which raises the possibility of a fungus-target insect pest interaction.

These discoveries have applications in agriculture pest management, especially in areas where Armyworm infestations seriously jeopardize crop yields. Farmers and agricultural practitioners can effectively leverage the biological control capability of *Penicillium* sp. entomopathogens to decrease pest damage and reduce reliance on chemical insecticides by cultivating these fungus in PDA media.

More investigation is necessary to investigate the scalability and effectiveness of PDA medium-based fungal formulations in field settings. Furthermore, research on the stability and persistence of fungal virulence over the long term in the environment is crucial for creating environmentally friendly pest control plans.

To sum up, this study's findings offer critical new perspectives on how best to apply biological control techniques to fight Armyworm infestations in agricultural settings. The discovery that Potato Dextrose Agar (PDA) is an ideal growth medium for *Penicillium* sp. entomopathogen cultivation presents a viable path toward improving pest management techniques and advancing sustainable agricultural production.

Conclusion

The findings of this research underscore the critical role of growth medium selection in maximizing the virulence of entomopathogenic fungi for effective pest management in agriculture. Through comprehensive evaluations and bioassays, Potato Dextrose Agar (PDA) emerged as the superior growth medium for cultivating *Penicillium* sp. entomopathogens, exhibiting enhanced fungal growth and conidia production compared to Sabouroud's Dextrose Agar (SDA).



The significant efficacy of *Penicillium* sp. entomopathogens cultivated on PDA medium in controlling Armyworm populations highlights the practical implications of these findings for agricultural pest management. By harnessing the enhanced virulence of *Penicillium* sp. fungi cultivated on PDA, farmers and agricultural practitioners can effectively mitigate pest damage and reduce reliance on chemical insecticides, thereby promoting sustainable agricultural practices and minimizing environmental impacts.

Moreover, the statistical analysis confirmed the robustness of the results, providing strong evidence for the superiority of PDA medium in promoting fungal virulence against Armyworms. These findings contribute to the growing body of knowledge on biological control strategies and offer valuable insights for optimizing pest management practices in agriculture.

Looking ahead, further research is warranted to explore the scalability and efficacy of PDA medium-based fungal formulations under field conditions. Long-term studies investigating the persistence of fungal virulence in the environment and its impact on pest populations are essential for developing robust and sustainable pest management strategies.

In conclusion, the identification of Potato Dextrose Agar (PDA) as the preferred growth medium for cultivating *Penicillium* sp. entomopathogens represents a significant advancement in biological control methods. By leveraging this knowledge, stakeholders in the agricultural sector can adopt more sustainable and environmentally friendly approaches to pest management, ultimately contributing to the long-term viability and resilience of agricultural systems.

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