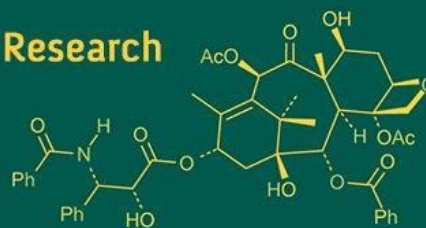


International Journal of Advanced Biochemistry Research



ISSN Print: 2617-4693
ISSN Online: 2617-4707
IJABR 2025; SP-9(5): 433-447
www.biochemjournal.com
Received: 09-02-2025
Accepted: 12-03-2025

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Biological control of storage pests: A review of current techniques

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DOI: <https://www.doi.org/10.33545/26174693.2025.v9.i5Sf.4467>

Abstract

The relentless pursuit of effective and environmentally conscious strategies for safeguarding stored products from pest infestations has propelled biological control to the forefront of agricultural research. This review delves into the cutting-edge techniques employed in the biological control of storage pests, highlighting the use of invertebrate and microbial agents, along with innovative approaches to habitat manipulation and conservation biological control, ultimately aiming to reduce our reliance on conventional chemical pesticides and foster sustainable pest management practices. The escalating demand for food security, coupled with growing concerns about the detrimental effects of synthetic pesticides on human health and the environment, has spurred intense research into alternative pest management strategies. Biological control, which harnesses the power of natural enemies to suppress pest populations, has emerged as a promising approach for protecting stored products from insect pests and other destructive organisms. The integration of available pest control options and the adoption of new crop cultivation technologies are also being recommended, particularly for small and resource-poor farmers. The review will explore the diverse array of biological control agents, encompassing parasitoids, predators, and pathogens, and examine their efficacy against a wide range of storage pests. Furthermore, it will explore the application of bio preservatives like starter cultures, or purified antimicrobial compounds to prevent microbial growth.

Keywords: Biological control, storage pests, natural enemies, microbial agents, habitat manipulation

1. Introduction

Storage pests inflict substantial post-harvest losses on agricultural commodities, necessitating effective and environmentally conscious control strategies. This review delves into cutting-edge biological control techniques aimed at mitigating storage pest infestations, highlighting the utilization of microbial antagonists, beneficial insects, and natural products to safeguard stored products. We explore the mechanisms underlying these biocontrol approaches, assess their efficacy in diverse storage environments, and discuss the challenges and opportunities associated with their implementation, offering a comprehensive overview of the current state and future directions of biological control in post-harvest pest management (Janisiewicz, 2013; Spadaro & Gullino, 2003) ^[47, 102].

The imperative to curtail post-harvest losses in fruits and vegetables, which can exceed 25% globally, has spurred the exploration of biological control strategies (Ghaouth *et al.*, 2002) ^[34]. Concerns regarding environmental pollution and the rise of fungicide-resistant pathogen strains have further accelerated the development of alternative approaches to chemical treatments, with microbial antagonists emerging as a promising avenue for managing fruit diseases across various crops (Ippolito & Nigro, 2000) ^[45]. These antagonists exhibit diverse modes of action, encompassing antibiosis, competition for nutrients and space, the induction of resistance in host tissues, and direct interaction with the pathogen, presenting a multifaceted approach to disease control (Droby *et al.*, 1996) ^[21]. The integration of microbial agents with unconventional chemicals has demonstrated synergistic effects, achieving significant levels of disease control, with the effectiveness of these combined strategies hinging on a thorough understanding of their mechanisms of action within the biocontrol system (Zhang *et al.*, 2018) ^[124].

However, the implementation of biocontrol agents as a widely adopted commercial strategy has faced obstacles, underscoring the necessity for increased governmental and industrial support, coupled with the exploration of novel research areas to establish biological control as a viable substitute for chemical interventions (Teixidó *et al.*, 2011; Vilaplana *et al.*, 2019) ^[111, 117]. A deeper understanding of the mode of action, ecology, compatibility with post-harvest commercial practices, and the effect of host physiology on their biological activity is needed to maximize the biocontrol potential of microbial antagonists (Ghaouth *et al.*, 2002) ^[34].

The application of biological control extends beyond microbial interventions, encompassing the utilization of beneficial insects and natural products to manage storage pests. Biological control offers an ecologically sound alternative, employing living organisms or their byproducts to maintain pest populations below economically damaging thresholds, thereby preserving natural enemy populations (Patil *et al.*, 2021) ^[82]. Biological control is intertwined with intricate interactions between plants and the microbes inhabiting the rhizosphere and phyllosphere, where their application minimizes disease and suppresses pests (Pandit *et al.*, 2022) ^[79]. The rise in global population has amplified the demand for high-quality agricultural products, leading to a surge in the use of chemical pesticides to combat crop diseases, which has consequently heightened concerns among consumers regarding the adverse effects of chemical fungicides on human health and the environment. Consequently, there has been a growing interest in exploring alternative solutions to safeguard crops, with biological controls employing beneficial microorganisms gaining significant traction, supported by the screening of various biological control agents, including **Bacillus**, **Pantoea**, **Streptomyces**, **Trichoderma**, **Clonostachys**, **Pseudomonas**, **Burkholderia**, and specific yeasts (Lahlali *et al.*, 2022) ^[57]. Employing biocontrol agents that are target-specific minimizes the impact on non-target organisms, thereby enhancing environmental stewardship (Singh *et al.*, 2019) ^[100]. The effectiveness of biocontrol agents is intricately linked to environmental factors, encompassing temperature, humidity, and the physiological condition of the host plant, which collectively influence the variability of their impact (Pandit *et al.*, 2022) ^[79].

2. Significance of Storage Pests

Storage pests, encompassing a diverse array of insects, mites, and rodents, pose a persistent threat to the integrity and economic value of stored agricultural commodities. These pests inflict considerable post-harvest losses through direct consumption, contamination with excrement and cast skins, and the creation of conditions conducive to microbial growth, all of which compromise the marketability and nutritional quality of stored products (Ehinmitan *et al.*, 2024) ^[23]. The ramifications of storage pest infestations extend beyond mere economic losses, impacting food security, trade, and the livelihoods of farmers and food processors. The globalization of trade and the increasing interconnectedness of supply chains have amplified the risk of pest introductions and their subsequent spread across geographical regions, exacerbating the challenges associated with storage pest management. The reliance on synthetic pesticides for storage pest control has raised concerns regarding environmental contamination, human health risks, and the development of pesticide resistance in pest

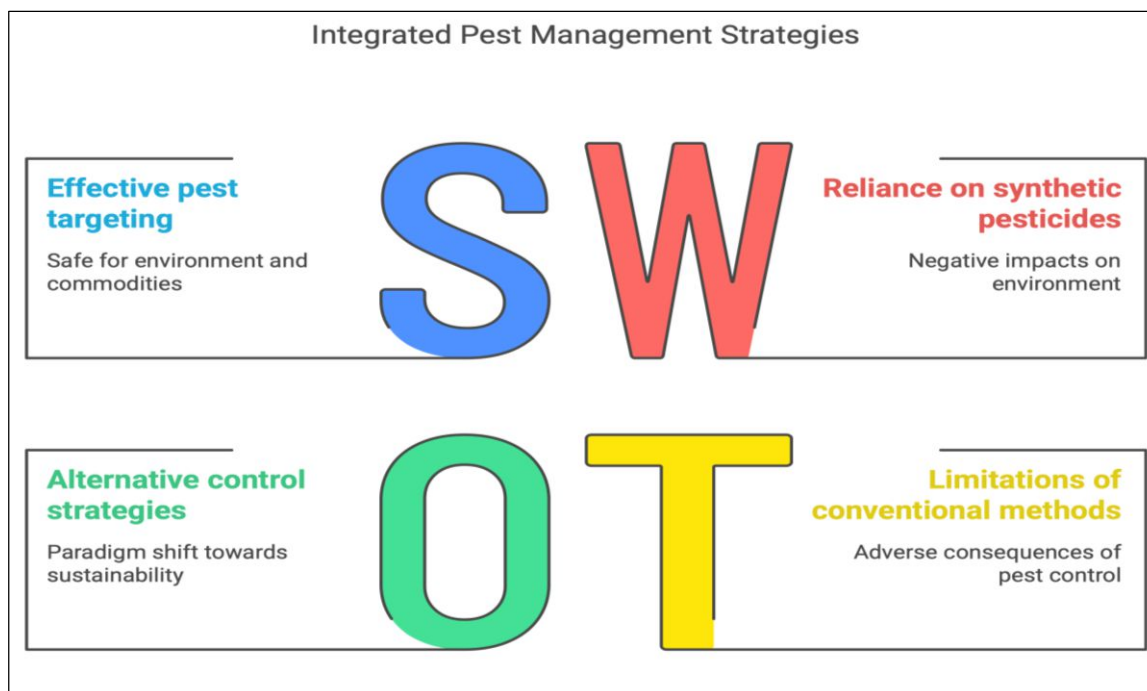
populations (Barzman *et al.*, 2015) ^[9]. Integrated pest management programs based on safer compounds are readily degradable in the environment (Khalil, 2012) ^[52]. This has led to a growing demand for safer and more sustainable pest management strategies, with biological control emerging as a promising alternative.

2.2 Conventional Control Methods and Their Limitations

Traditional storage pest control strategies predominantly rely on synthetic insecticides, fumigants, and protectants to suppress pest populations and minimize post-harvest losses. While these chemical interventions can offer rapid and effective pest control, their continued use has raised significant concerns regarding environmental sustainability, human health, and the evolution of pesticide resistance in storage pests. The widespread application of broad-spectrum insecticides can disrupt the natural balance of storage ecosystems, decimating beneficial insect predators and parasitoids that play a crucial role in regulating pest populations. The application of synthetic insecticides, modification of agronomic practices, use of resistant varieties, application of botanicals, biological and mechanical controls are some of the empirically verified methods that can be used to mitigate the impact of insect pests (Ofuya *et al.*, 2023) ^[74]. Fumigants, such as methyl bromide and phosphine, have been widely used for disinfesting stored commodities, but their ozone-depleting potential and toxicity to non-target organisms have prompted restrictions and phase-out efforts. The exposure of storage pests to sublethal doses of insecticides can trigger physiological and behavioral changes that lead to the development of resistance mechanisms, rendering these chemicals ineffective over time (Wondimagegn, 2022) ^[119]. The accumulation of pesticide residues in stored products poses a potential health risk to consumers, particularly with chronic exposure to low levels of these chemicals (Desai *et al.*, 2014) ^[119]. The development of chemical pesticides enabled farmers to abandon more traditional control strategies, but an over-reliance on the use of chemicals led to problems of resistance and environmental damage (Leake, 2000) ^[58]. The intensive use of chemical pesticides has led to the development of resistant pests and pathogens, necessitating the use of higher dosages, increasing the health- and environment-associated risks (Ehinmitan *et al.*, 2024) ^[23].

2.3 Need for Alternative Control Strategies

The limitations and adverse consequences associated with conventional storage pest control methods have created a compelling need for alternative strategies that are environmentally sustainable, economically viable, and effective in the long term. Integrated pest management emphasizes the coordinated use of multiple tactics for optimizing the control of all classes of pests in an ecologically and economically sound manner (Damos, 2004) ^[16]. Biological control, which involves the use of living organisms to suppress pest populations, offers a promising approach for managing storage pests while minimizing the risks associated with synthetic pesticides (Acharya *et al.*, 2017) ^[2]. The public is becoming increasingly aware of the impact of pesticide application on food quality and safety, human health, and environmental contamination. This concern has driven the search for pest control alternatives to synthetic pesticides.



Biological control agents, such as insect predators, parasitoids, and microbial pathogens, can effectively target storage pests without harming non-target organisms or leaving harmful residues in stored commodities. The application of biological control strategies can enhance the resilience of storage ecosystems, promoting long-term pest suppression and reducing the reliance on synthetic pesticides. This paradigm shift towards alternative control strategies is driven by increasing concerns about the negative impacts of synthetic pesticides on the environment, human health, and the sustainability of agricultural systems. Botanical plants could offer a better alternative to synthetic pesticides and effective to control many pests (Wondimamegn, 2022) ^[119].

Alternative methods, including host plant defense, biotic agents, and the application of companion plants, are vital in pest control and global insecticide reduction (Han *et al.*, 2024) ^[40]. Integrated Pest Management is the deployment of a variety of methods of pest control designed to complement, reduce or replace the application of synthetic pesticides (Pretty & Bharucha, 2015) ^[84]. The goal of cultural control is to alter the environment or condition of the host to prevent or suppress an infestation (El-Shafie, 2019) ^[25]. Cultural practices include crop rotation, intercropping, adjusting planting and harvesting dates, sanitation, water and soil management and tillage.

2.3 Rationale for Biological Control

Biological control provides a sustainable and environmentally sound approach to manage storage pests, which minimizes the negative impacts associated with synthetic pesticides (Reis *et al.*, 2022) ^[89]. Biological control is a method of controlling pests using other living organisms. Biological pest control relies on predation, parasitism, herbivory, or other natural mechanisms, but typically also involves an active human management role. Classical biological control is used in agriculture to protect crops from damage caused by pests and involves the introduction of natural enemies that control the population of the pest. Biocontrol agents can persist in the storage

environment, providing long-term pest suppression and reducing the need for repeated interventions.

Biological control agents can be highly specific to their target pests, minimizing the risk of harming non-target organisms or disrupting the ecological balance of the storage ecosystem (Gardener & Fravel, 2002) ^[33]. Biological control strategies are often more cost-effective than synthetic pesticides in the long run, as they can reduce the need for repeated applications and minimize the development of pesticide resistance (F.D. *et al.*, 2013) ^[30]. Biological control agents can be integrated with other pest management tactics, such as sanitation and physical barriers, to create a comprehensive and sustainable pest management program. The integration of biological control agents with other pest management strategies can enhance the overall effectiveness of pest control programs.

2.4 Biological Control Methods

Biological control methods can be broadly categorized into three main approaches: conservation, augmentation, and importation. Conservation biocontrol involves preserving existing natural enemies in the storage environment by providing them with food, shelter, and protection from pesticides. Augmentation biocontrol involves releasing additional natural enemies into the storage environment to supplement the existing population and enhance pest suppression. Importation biocontrol involves introducing exotic natural enemies from other regions to control pests that are not native to the storage environment (Orr & Lahiri, 2013) ^[76].

The focus of biological control programs is to increase the effectiveness of natural enemies that already exist in an environment, either by protecting them or by increasing their numbers. The multiplication of biocontrol agents is essential for the development of augmentative technique (Mascarin *et al.*, 2022) ^[66]. Augmentation of natural enemies has long been considered a viable strategy in the field and greenhouse (Parrella *et al.*, 1992) ^[80]. Importation biological control is particularly useful when dealing with invasive pests that lack natural enemies in the storage environment.

The utilization of biopesticides presents a practical and sustainable alternative for managing pests in agricultural crops, either independently or in conjunction with reduced dosages of synthetic pesticides (Yparraguirre *et al.*, 2020) [122]. The combined application of endophytic *Beauveria bassiana* and low doses of insecticides displayed synergistic effects in reducing insect pests in rice crops. Some *Bacillus* species are effective and play a crucial role in the field of bioagents because they are systemic resistance inducer and plant growth promoter (Dadrasnia *et al.*, 2020) [14].

Biological control has been shown to be effective for plants, livestock, humans, and the ecosystem (Upadhyay *et al.*, 2021) [116].

The deployment of *Bacillus thuringiensis* strains has been observed to improve plant development and offer defense against bacterial pathogens (Ramírez-Pool *et al.*, 2024) [87]. Biopesticides, derived from natural sources like bacteria, fungi, viruses, and plant extracts, are utilized in pest management as they pose minimal risks to human health, the environment, and non-target species (Zanchi *et al.*, 2021) [123].

2.5 Types of Biocontrol Agents

A variety of biological control agents are available for use, but further research is needed to identify potential microbes and their natural products with a broad-spectrum antagonistic activity to control crop diseases (Ayaz *et al.*, 2023) [6]. These agents can be broadly classified into predators, parasitoids, and pathogens. Predators are organisms that kill and consume their prey, while parasitoids are organisms that lay their eggs in or on their host, eventually killing it. Pathogens are disease-causing organisms that can infect and kill their host pests.

Predatory insects, such as ladybugs and lacewings, can effectively control aphids, mites, and other soft-bodied pests in stored products. Parasitoid wasps, such as *Trichogramma* and *Habrobracon*, can parasitize the eggs or larvae of storage pests, preventing them from developing into adults (Sani *et al.*, 2020) [95]. Entomopathogenic fungi, such as *Beauveria bassiana* and *Metarhizium anisopliae*, can infect and kill a wide range of storage pests, including beetles, moths, and mites.

Entomopathogenic nematodes, such as *Steinernema* and *Heterorhabditis*, can also be used to control soil-dwelling storage pests. Numerous microorganisms have demonstrated efficacy against soil-borne diseases and nematodes over the past half-century, and some have even been incorporated as active ingredients in commercially available biopesticides (Jaiswal *et al.*, 2022) [46]. Botanical insecticides, derived from plant extracts, can also be used as biopesticides to control storage pests. These products are considered to be environmentally safe because of their specific action, biodegradability, and do not leave toxic residues (Syed-Ab-Rahman *et al.*, 2018) [107].

Botanical pesticides are plant extracts or compounds used to prevent, destroy, or control different types of pests (Măruțescu *et al.*, 2017) [65]. The utilization of biopesticides presents a practical and sustainable alternative for managing pests in agricultural crops, either independently or in conjunction with reduced dosages of synthetic pesticides. The source of bio-nematicides could be plants, bacteria, fungi, actinomycetes and/or any microorganism from the soil (Khalil, 2014) [53].

The search for naturally occurring compounds that will disrupt the life cycle of the pests and protect stored products has become very important. The use of microbial agents represents a considerably safer and low toxicity alternative to synthetic fungicides (Bazioli *et al.*, 2019) [10].

2.6 Application of Biocontrol Agents

The application of biological control agents in storage facilities requires careful planning and implementation to ensure their effectiveness. Factors to consider include the type of pest, the storage environment, the biological control agent, and the application method.

The application method will depend on the type of biological control agent and the storage environment. For example, predatory insects can be released directly into the storage facility, while parasitoid wasps can be introduced using release devices. Entomopathogenic fungi and nematodes can be applied as sprays or dusts. Biocontrol products are available in various formulations, including liquid concentrates, wettable powders, and granular formulations.

Inoculant carriers can fall into five categories: Plain lyophilized microbial cultures, liquid inoculants, with some chemical additives to better stickiness, stabilization, function, and dispersal inert materials (polymers), waste products of industrial and agriculture resources such as lignin, soils: coal, clays, peat and inorganic soil (Riseh *et al.*, 2021) [90]. Formulation of the biopesticides is one of the most important aspects of their successful commercialization.

Many publications have featured botanical pesticides with skewed interest towards management of insect pests (Lengai *et al.*, 2019) [59]. The difficulties presented by the formulations of botanical bioinsecticides in a large scale, have generated the search and application of new technologies (Hernández-Tenorio & Sánchez, 2020) [43].

3. Biology of Storage Pests

3.1 Major Pest Groups

Storage pests constitute a taxonomically diverse group of organisms, with insects and mites representing the predominant arthropod pests, while rodents constitute a significant vertebrate pest group. Within the insects, beetles and moths are the most commonly encountered storage pests, causing damage to a wide range of stored products.

Beetles are characterized by their hardened forewings (elytra) that protect their membranous hindwings used for flight, and their chewing mouthparts enable them to feed on whole grains, processed cereals, and other dry food products.

4.1 Microbial Antagonists

Beneficial microorganisms, including bacteria, fungi, and yeasts, have garnered considerable attention as promising biocontrol agents for storage pests.

4.2 Beneficial Insects

Predatory and parasitoid insects play a crucial role in regulating storage pest populations.

4.3 Natural Products

Plant-derived compounds, such as essential oils and extracts, exhibit insecticidal, repellent, and antifeedant

properties against storage pests, offering a sustainable alternative to synthetic insecticides.

4.4 Integrated Approaches

Combining biological control agents with other pest management strategies, such as sanitation, physical barriers, and modified atmospheres, can enhance the effectiveness and sustainability of storage pest management programs.

The integration of biological control agents with other pest management strategies offers a holistic and sustainable approach to storage pest management.

- 1 **Sanitation:** Maintaining a clean and hygienic storage environment is crucial for preventing pest infestations.
- 2 **Physical Barriers:** Implementing physical barriers, such as screens, traps, and airtight containers, can prevent pest entry and restrict their movement within the storage facility.
- 3 **Modified Atmospheres:** Altering the atmospheric composition within storage facilities, such as reducing oxygen levels or increasing carbon dioxide levels, can suppress pest development and reproduction (Stejskal *et al.*, 2019) ^[104].

These integrated approaches require a thorough understanding of pest biology, storage environment, and available control options to design and implement effective and sustainable storage pest management programs.

Several arthropod pests that cause damage to underground and aboveground plant parts are controlled using entomopathogenic bacteria, fungi, nematodes, and baculoviruses (Dara, 2016) ^[18]. Foliar and soil application of EPF formulations can be used to control many insect pests.

The use of microbial agents represents a considerably safer and low toxicity alternative to synthetic fungicides (Mbajiuka *et al.*, 2019) ^[68].

5. Major Pest Groups

Storage pests exhibit remarkable taxonomic diversity, with insects and mites representing the predominant arthropod pests, while rodents constitute a significant vertebrate pest group.

Within the insect realm, beetles and moths reign supreme as the most commonly encountered storage pests, inflicting damage upon a vast spectrum of stored commodities. Beetles, distinguished by their hardened forewings safeguarding their membranous hindwings employed for flight, and equipped with chewing mouthparts, possess the ability to consume whole grains, processed cereals, and a myriad of other desiccated food products.

5.1 Augmentation

The augmentation of biocontrol agents involves the release of additional individuals of a naturally occurring or exotic species to supplement existing populations, enhancing their pest control efficacy.

5.2 Conservation

Conservation biological control focuses on modifying storage environments to enhance the activity and survival of existing natural enemies, creating a more favorable habitat for biocontrol agents.

5.3 Importation

The importation of exotic biocontrol agents involves the

introduction of natural enemies from other regions to control storage pests that have invaded new areas.

In stored grain ecosystems, microbial biopesticides, derived from bacteria, fungi, viruses, and protozoa, have emerged as promising alternatives to synthetic insecticides (B. *et al.*, 2014) ^[8]. These biopesticides offer several advantages, including target specificity, environmental compatibility, and reduced risk of resistance development.

Among the microbial biopesticides, *Bacillus thuringiensis** (*Bt**) stands out as the most widely used and commercially successful (Ruiu, 2018) ^[92].

Entomopathogenic fungi, such as *Beauveria bassiana** and *Metarhizium anisopliae**, have also demonstrated potential for controlling storage pests.

The integration of microbial biopesticides into IPM programs for stored grains requires careful consideration of factors such as application timing, dosage, and environmental conditions to maximize their effectiveness and minimize any potential non-target effects.

The augmentation strategy relies on releasing additional individuals of a biocontrol agent to bolster existing populations and enhance pest control. Conservation biological control focuses on modifying storage environments to favor the activity and survival of natural enemies. Importation, involves introducing exotic biocontrol agents from other regions to control invasive storage pests.

The search for alternatives to chemical control has led to the exploration of biological control agents, including bacteria, as a promising strategy for postharvest disease management (Damasceno *et al.*, 2019) ^[15]. The most remarkable results in selection, mode of action, production and formulation of biocontrol agents, possible approaches to improve their viability and efficacy and the integration with other alternative methods have been described (Teixidó *et al.*, 2011) ^[111]. In light of growing environmental concerns and the increasing demand for sustainable agricultural practices, biological control has emerged as a compelling alternative to synthetic pesticides for managing storage pests (Hartmann & Proença, 2023) ^[42]. Biopesticides are gaining traction due to the expansion of integrated pest management strategies globally, with entomopathogenic fungi playing a crucial role as infective agents in the biopesticide market (Muñiz-Paredes *et al.*, 2017) ^[70]. Specifically, *Beauveria bassiana** and *Metarhizium anisopliae** are among the pioneering entomopathogenic fungi successfully employed for the biological control of insect pests (Sandhu *et al.*, 2012) ^[94].

The application of synthetic fungicides to combat fungal diseases in agriculture has led to the rise of fungicide-resistant pathogens, raising concerns about environmental and human health impacts (Ons *et al.*, 2020) ^[75]. Recent advancements in the field have explored the use of antagonistic bacteria to produce hydrolytic enzymes that degrade fungal cell walls, effectively controlling postharvest diseases. Several novel approaches are emerging as possible alternatives to synthetic fungicides (Jijakli & Lepoivre, 2006) ^[49]. Nanomaterial treatments have shown potential in reducing the use of synthetic fungicides for controlling postharvest rot in fruits and vegetables (Roberto *et al.*, 2019) ^[91]. The use of low-toxicity chemicals that have minimal toxicological effects on mammals and the environment is also gaining attention as a way to control citrus postharvest diseases. Biological control has emerged as an effective alternative, particularly for wound-invading

necrotrophic pathogens, as it allows for direct application of antagonists to the targeted area. Biological control of postharvest diseases has been recognized as a viable alternative to synthetic fungicides by the fruit industry, with registered products available for use on organically labeled fruit in the United States (Janisiewicz, 2013; Janisiewicz & Korsten, 2002) [47, 48]. Furthermore, numerous studies have explored non-chemical applications such as irradiation, natural compounds, biocontrol agents, hot water/air application, and salts, reflecting a broader effort to identify alternative control methods (Janisiewicz, 2013; Palou, 2016; Roberto *et al.*, 2019) [47, 78, 91]. *Bacillus* species have proven to be effective as bioagents and play a crucial role in disease control, offering a promising avenue for sustainable agriculture and reducing reliance on synthetic pesticides (Dadrasnia *et al.*, 2020) [14].

The alarming rise in virulent phytopathogenic fungi has spurred the exploration of innovative strategies for fungal disease control, driven by the negative impacts of synthetic fungicides on the environment and human health (El-Baky & Amara, 2021) [24]. Exploring innovative strategies for fungal disease control is paramount, given the environmental and health concerns associated with synthetic fungicides, including the rise of fungicide-resistant pathogens (El-Baky & Amara, 2021; Szczerbanik *et al.*, 2007) [24, 109]. The application of synthetic fungicides has led to the development of fungicide-resistant pathogens, raising concerns about the environmental and health impacts of these chemicals (González-Estrada *et al.*, 2019) [37]. One promising area of research focuses on the use of organic acids and plant extracts with antimicrobial properties to prevent diseases. These natural compounds can act directly on the fungus or trigger defense responses in plants by increasing the activity of antioxidant enzymes (Murmur & Mishra, 2018) [71]. Many believe the induction of resistance in harvested products, achieved through biotic or abiotic elicitors, represents a notable strategy for disease control. These elicitors can activate various defense mechanisms in the fruit, such as the production of phytoalexins, pathogenesis-related proteins, and the reinforcement of cell walls, leading to enhanced resistance against fungal pathogens.

Many fungal species can efficiently protect plants from diseases caused by plant pathogenic fungi, and man has attempted to increase crop production and control disease severity of crop plants by altering cultivation practices, which reduce both initial inoculum as well as infection rate (Thambugala *et al.*, 2020) [112]. The integration of plant extracts into disease management strategies holds immense potential for reducing reliance on synthetic fungicides and promoting environmentally sustainable agricultural practices. The use of beneficial fungi to control plant diseases is garnering significant attention, with many beneficial fungi demonstrating the ability to inhibit the growth of various plant pathogens (Ayaz *et al.*, 2023) [6]. The use of biocontrol agents to manage plant diseases has gained traction as a sustainable approach that reduces the reliance on synthetic pesticides. *Trichoderma* spp., for instance, are soil-borne fungi known for their antagonistic potential against a wide range of phytopathogenic fungi (Ratan *et al.*, 2017) [88]. Understanding the characteristics and mechanisms of action of *Trichoderma* is crucial for effectively applying them in field conditions to control phytopathogens (Mukhopadhyay & Kumar, 2020; Ratan *et*

al., 2017) [79, 88]. Several *Trichoderma* species have demonstrated remarkable capabilities in promoting plant growth and suppressing fungal phytopathogens, positioning them as promising candidates for green technologies (Tyśkiewicz *et al.*, 2022) [115].

6. Cutting-Edge Techniques in Biological Control

Trichoderma species are not only recognized for their ability to control plant diseases but also for their capacity to enhance plant growth, making them valuable tools for sustainable agriculture (Topolovec-Pintarić, 2019) [113]. In addition to their mycoparasitic and antagonistic properties, certain *Trichoderma* strains can stimulate plant growth and development, further enhancing their utility in agricultural systems (Csótó *et al.*, 2022) [13]. *Trichoderma* strains have emerged as key biocontrol agents, effectively suppressing fungal phytopathogens and promoting plant growth. Several *Trichoderma* species, including *T. harzianum**, *T. viride**, *T. virens**, and *T. atroviride**, have emerged as prominent options for the biological control of plant diseases (Jorge, 2014) [50]. *Trichoderma* exhibits good biological control effects and can reduce the use of chemical pesticides; however, more effective and suitable strains need to be found to join the biocontrol team (Yao *et al.*, 2023) [121].

Trichoderma species are ubiquitous saprotrophic fungi in terrestrial ecosystems, mediating interactions with other soil microorganisms, plants, and arthropods at multiple trophic levels (Macías-Rodríguez *et al.*, 2020) [62]. The interaction between *Trichoderma* and plants involves a complex signaling network that triggers a range of beneficial responses, including enhanced nutrient uptake, increased stress tolerance, and improved disease resistance. These fungi are known to control harmful pathogens through mechanisms like parasitism, competition, and antibiosis, making them integral to Integrated Pest Management strategies (Mukhopadhyay & Kumar, 2020) [69]. *Trichoderma* species are capable of colonizing plant roots, which allows them to establish a symbiotic relationship with the host plant. The ability of *Trichoderma* to colonize plant roots and establish symbiotic relationships further enhances their biocontrol and plant growth-promoting activities.

Mycoparasitism and Cell Wall Degradation

Trichoderma species exhibit mycoparasitism, directly attacking and feeding on other fungi, thereby reducing pathogen populations. Mycoparasitism involves the secretion of enzymes that degrade the cell walls of target fungi, such as chitinases and glucanases. The mycoparasitic activity of *Trichoderma* involves the production of cell wall-degrading enzymes, which break down the structural components of fungal pathogens, leading to their inactivation.

Competition for Nutrients and Space: *Trichoderma* species compete with plant pathogens for essential resources such as nutrients and space, limiting pathogen growth and colonization. By rapidly colonizing the rhizosphere, *Trichoderma* species outcompete fungal pathogens for nutrients and space, thereby limiting their ability to infect plant roots. Furthermore, *Trichoderma*'s competitive behavior aids in reducing the incidence and severity of plant diseases by limiting pathogen access to plant tissues.

Induction of Plant Resistance: *Trichoderma* can induce systemic resistance in plants, enhancing their ability to defend against pathogen attacks. Induced systemic resistance is a defense mechanism activated in plants by beneficial microbes like *Trichoderma*, leading to enhanced resistance against a broad spectrum of pathogens. By inducing plant defense responses, *Trichoderma* not only protects against immediate pathogen attacks but also primes the plant for future encounters, resulting in long-lasting resistance (Tyśkiewicz *et al.*, 2022) ^[115]. The ability of *Trichoderma* to trigger plant defense mechanisms further contributes to its effectiveness as a biocontrol agent.

Trichoderma species produce a variety of secondary metabolites with antimicrobial properties, which inhibit the growth and development of plant pathogens (Guzmán-Guzmán *et al.*, 2023) ^[39]. These compounds act by disrupting cellular processes in target organisms, effectively suppressing their growth and spread (Vinale, 2014) ^[118]. Additionally, the production of antimicrobial metabolites contributes to the antagonistic activity of *Trichoderma* against plant pathogens. Secondary metabolites from *Trichoderma* can also influence plant development, acting as hormone-like substances that affect growth and stress responses (Vinale, 2014) ^[118].

In addition to direct antagonism, *Trichoderma* can enhance plant growth and development through various mechanisms. *Trichoderma* can improve nutrient uptake by plants, facilitating the mobilization and assimilation of essential elements from the soil (Alfiky & Weisskopf, 2021) ^[4]. Some *Trichoderma* strains are also capable of producing plant hormones, such as auxins and cytokinins, which stimulate plant growth and development. Furthermore, *Trichoderma* can enhance root development, increasing the surface area for nutrient and water absorption, leading to improved plant vigor.

Trichoderma can also improve the plant's ability to tolerate various environmental stresses, such as drought, salinity, and heavy metal toxicity. Plants colonized by *Trichoderma* often exhibit increased tolerance to drought stress due to improved water uptake and reduced transpiration rates. *Trichoderma*'s ability to enhance plant stress tolerance further underscores its significance in sustainable agriculture, particularly in regions facing environmental challenges. The use of *Trichoderma* as a biostimulant holds considerable promise for promoting sustainable agriculture by improving plant health and productivity while minimizing the reliance on chemical inputs (Esparza-Reynoso *et al.*, 2020) ^[27]. The use of *Trichoderma* as a biostimulant holds considerable promise for promoting sustainable agriculture by improving plant health and productivity while minimizing the reliance on chemical inputs (Dutta *et al.*, 2017; Palma *et al.*, 2018) ^[22, 77].

The effectiveness of *Trichoderma* as a biocontrol agent is influenced by a combination of environmental factors, including temperature, moisture, pH, and soil type.

6. Formulation and Delivery

Effective formulation and delivery systems are crucial for maintaining the viability and efficacy of biocontrol agents in storage environments.

The careful selection of appropriate formulations and application methods can enhance the persistence and impact of biocontrol agents on target pest populations.

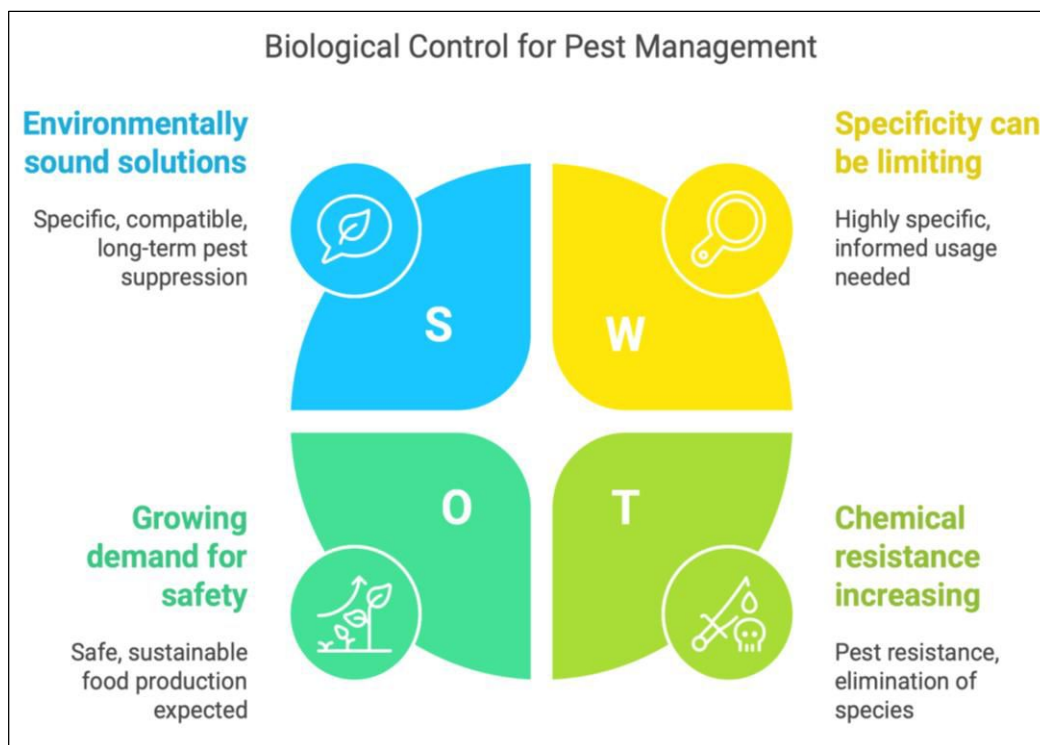
6.1 Efficacy of Biological Control Agents

The efficacy of biological control agents in storage pest management depends on factors such as the agent's specificity, application rate, and environmental conditions.

The successful implementation of biological control strategies requires a thorough understanding of pest biology and behavior.

7. Future Trends and Research Directions

Further research is needed to optimize the efficacy and reliability of biological control agents in diverse storage environments (Shang *et al.*, 2024) ^[98]. Development of novel formulations and delivery systems to enhance the survival and activity of biocontrol agents is crucial. Investigating the potential of combining different biological control agents or integrating them with other pest management strategies is an important area of research. The President of China has recently started a "National research program on reduction in chemical pesticides and fertilizers in China" for the development of non-chemical control methods, showing a need for them (Lenteren *et al.*, 2017) ^[61]. Biological control is the employment of any living organism to combat a specific plant disease or pest through parasitism, antibiosis, or competition for resources or space (Pandit *et al.*, 2022) ^[79]. In recent years, the concept of biological control has become increasingly important due to legislative and consumer pressures to phase out or reduce the use of traditional chemical-based pesticides in agricultural practices (Martínez *et al.*, 2023; Pandit *et al.*, 2022) ^[64, 79]. Several tactics, including cultural control, the use of resistant types, and the employment of synthetic pesticides, are used to manage plant diseases. However, cultural control is labor-intensive and time-consuming, resistant varieties are not always available, and synthetic pesticides can harm the environment and human health. The use of biological control is a possible alternative. Assessing the economic feasibility and market potential of biological control products is important for their widespread adoption by the industry. Addressing regulatory hurdles and developing standardized protocols for the registration and use of biological control agents is essential. The growing global demand for safe and sustainable food production is expected to drive further research and development in the field of biological control for storage pests. Increased usage of chemical fertilizers and pesticides has resulted in the accumulation of leftover chemical compounds in the environment, and pathogenic microorganisms are beginning to develop resistance (Rabbee *et al.*, 2019) ^[85].



This has become a serious issue. (Barzman *et al.*, 2015) ^[9]. Over the last few decades, the use of chemical compounds, such as pesticides, has rapidly increased. Thus, the harmful effects of insecticides on non-target organisms and the environment are well documented in order to limit their use. This fact justifies the necessity for research and development of alternative approaches to balance agricultural, environmental, and health issues in crop protection (Karimi *et al.*, 2012) ^[51]. Furthermore, because of its efficacy and ease of application, chemical control has historically been the favored method of managing plant diseases; however, it is now being discouraged due to its hazards to ecological health and the buildup of pathogen resistance (Deshi *et al.*, 2014) ^[119]. Plant diseases are managed using a variety of strategies, including cultural control, resistant types, and synthetic pesticides; however, cultural control is labor-intensive, resistant varieties are not always obtainable, and synthetic pesticides have detrimental consequences for the environment and human health. The use of biological control presents a viable option and has received more attention as a result. Because of growing worries about the negative impacts of some chemicals on the environment, human health, and living things, researchers are increasingly looking at possible biological control microbes as practical alternatives for managing pests and plant pathogens (Syed-Ab-Rahman *et al.*, 2017) ^[108]. Integrated Pest Management programs rely heavily on biorational pesticides, particularly with the increased use of sustainable agriculture. The need for environmentally friendly controls is becoming more urgent, and the public is becoming more aware of them, thus there is an increasing need for the usage of botanical and biological pesticides in the management of integrated plant pests and diseases, in addition to other safe options (Ehinmitan *et al.*, 2024; Khalil, 2012; Leake, 2000; Prashar *et al.*, 2013) ^[23, 52, 58, 83]. The use of synthetic chemicals has several drawbacks, including pesticide resistance in pests, the elimination of non-target species, secondary pest outbreaks, and negative effects on human health (Wondimagegn, 2022) ^[119]. In

addition, synthetic chemicals have a chronic and acute detrimental impact on human wellness. For these reasons, using chemical insecticides does not satisfy the criteria for integrated pest management. Botanical plants may be a superior alternative to synthetic pesticides and are effective in controlling a wide range of pests, offering a solution to all of the aforementioned drawbacks of insecticides. Humans have employed plant-based insecticides against herbivores for ages (Wondimagegn, 2022) ^[119]. They stand out as potential pest management tools because of their selectivity and biodegradability. Botanical pesticides are thought to be safe for use in agricultural systems in general. As a result, they are viewed as appealing substitutes for synthetic chemical pesticides in pest control (Đăng *et al.*, 2012; Siswanto *et al.*, 2020) ^[17, 101]. Biological control agents offer numerous advantages, including their specificity, environmental compatibility, and ability to provide long-term pest suppression (Lenteren, 1988) ^[60]. Biological control tactics are generally highly specific, which makes them ecologically benign, but this specificity necessitates their use in a highly informed manner (Harper, 2006) ^[41]. In contrast to chemical pesticides, biopesticides' specificity in targeting pests may be a disadvantage if the goal is to eradicate many pests at once (Ayilara *et al.*, 2023) ^[7]. Biological control tactics are generally highly specific, which makes them ecologically benign, but this specificity necessitates their use in a highly informed manner. The advancement of biopesticides based on inert dusts, plant extracts, and essential oils offers a complementary or alternative way to use traditionally manufactured insecticides to protect intensive food production from pests (Stanković *et al.*, 2020) ^[103].

7. Novel delivery systems for biocontrol agents

The exploration of novel delivery systems for biocontrol agents represents a cutting-edge area of research with the potential to revolutionize the application and efficacy of these beneficial organisms in storage pest management. Encapsulation technologies, such as microencapsulation and

nanoencapsulation, offer a means to protect biocontrol agents from environmental stressors, enhance their targeted delivery to pest populations, and prolong their activity. These techniques involve embedding the biocontrol agent within a protective matrix, such as a polymer or lipid, which can be designed to release the agent gradually or in response to specific environmental cues. Formulation technologies play a crucial role in maintaining the viability and efficacy of biocontrol agents during storage and application (Rajput *et al.*, 2020) ^[86]. Controlled-release formulations, for example, can extend the period of pest control by gradually releasing the biocontrol agent over time (Seven *et al.*, 2019) ^[97]. Additionally, the use of attractants or carriers can improve the delivery of biocontrol agents to target pests in storage environments. Coatings made of edible polymers that can be combined with antagonists are one strategy with the most promise; this method of formulation of biocontrol products, which includes appropriate microbial protectants, can produce stable products with high microbial viability that are ready for field applications and have improved adherence and survival of the BCA after application in plants (Marin *et al.*, 2017) ^[63].

Nano-formulation of essential oils can address issues with volatility, water solubility, and oxidation, protecting them from deterioration and evaporation losses, enabling regulated release, and improving handling. These formulations are anticipated to be more potent than bulk chemicals (González *et al.*, 2013) ^[36]. Nanoencapsulation, active packaging, and polymer-based coatings have demonstrated effectiveness in addressing these challenges, improving the bioefficacy and controlled release of essential oils (Maurya *et al.*, 2021) ^[67].

Nanoencapsulation, which involves encapsulating flavors in capsules with sizes ranging from 10 to 1000 nm, not only protects flavors but also significantly enhances their bactericidal, virucidal, fungicidal, antiparasitical, and insecticidal activities, as well as preserving other medicinal properties like analgesic, sedative, anti-inflammatory, and spasmolytic effects (Gupta & Variyar, 2016) ^[38]. Nanoencapsulation is a technique that involves encasing a bioactive compound in a matrix or inert material in liquid, solid, or gaseous states to preserve the coated substance, and it has grown in popularity as a response to consumers' rising concerns and demands regarding food safety and quality (Arpagaus, 2019; Estevinho *et al.*, 2013) ^[5].

The design of functional foods has recently increased in response to rising consumer demand and concerns about natural, healthy, and nutritious food products (Pateiro *et al.*, 2021) ^[81]. The incorporation of bioactive compounds in food products using micro- and nano-delivery systems may offer extra health benefits beyond basic nutrition, since their encapsulation may provide protection against undesired environmental conditions along the food chain, thus improving their bioavailability while enabling their controlled release and target delivery (Simões *et al.*, 2017) ^[99]. However, maintaining the stability and bioactivity of bioactive compounds is one of the challenges in the development of functional foods, food packaging, and preservation. These constraints undermine their advantages. Microencapsulation has been used in industry and is the subject of ongoing research for food packaging, functional foods, nutraceuticals, and pharmaceutical applications in order to get beyond unfavorable external environmental conditions, mask organoleptic characteristics, and enable the

regulated release of ingredients over time. The produced particles can range in diameter from 1 µm to 1000 µm (Coelho *et al.*, 2022) ^[11].

Essential oils' volatility, water solubility, and propensity to oxidize can all be problematic; nano-formulation can help with these issues by shielding them from deterioration and evaporation losses, enabling regulated release, and improving handling. These formulations are anticipated to be more potent than bulk chemicals (Salvia-Trujillo *et al.*, 2017) ^[93]. Several techniques for encapsulating bioactive substances have been studied; these techniques include spray drying, extrusion, coacervation, emulsification, and liposome entrapment.

These techniques offer the possibility of increasing the shelf life and efficacy of biocontrol products while minimizing their environmental impact (Días *et al.*, 2015) ^[20]. Further research is needed to optimize these delivery systems for specific biocontrol agents and storage pest scenarios, as well as to assess their cost-effectiveness and regulatory implications. The use of nano/micro delivery systems for food product development is proving to be a game changer which has raised the expectations of the researchers, food manufacturers and consumers regarding possibility of enhancing the functionality of bioactives within the fortified food products (Aditya *et al.*, 2017) ^[3]. In this direction, nano/micro delivery systems using lipids, surfactants and other materials have been fabricated to stabilize and enhance the biological activity of the bioactive compounds. Microencapsulation emerges as a potential approach to overcome these problems and, additionally, to provide controlled or targeted delivery or release (Aditya *et al.*, 2017; Días *et al.*, 2015; Fathi *et al.*, 2014) ^[3, 20, 29]. This is achieved by coating the core material or bioactive compound with another material to produce capsules ranging from several nanometers to several millimeters in size (Comunian & Fávaro-Trindade, 2016; Khan *et al.*, 2020; Maurya *et al.*, 2021; Subramani & Ganapathyswamy, 2020) ^[12, 54, 67, 105].

8. Challenges and Limitations

Biological control of storage pests, while promising, faces several challenges and limitations that need to be addressed to ensure its widespread adoption and effectiveness. One of the main challenges is the specificity of many biocontrol agents, which may limit their effectiveness against a broad range of storage pests. This specificity requires accurate identification of the target pest species and selection of the appropriate biocontrol agent, which can be time-consuming and require specialized expertise. Environmental factors, such as temperature, humidity, and aeration, can significantly impact the efficacy of biocontrol agents in storage environments (Maurya *et al.*, 2021) ^[67]. Understanding the optimal conditions for each agent is crucial for successful implementation, and fluctuations in these conditions can lead to inconsistent results. The complex interactions between biocontrol agents, storage commodities, and the surrounding environment can also pose challenges. It is challenging to create solutions that are effective across a variety of hosts and geographical locations due to the complexity of the virus and its varied interactions with biocontrol agents and animals (Fenta & Mekonnen, 2024) ^[31]. Some biocontrol agents may be sensitive to certain storage conditions or incompatible with specific commodities, limiting their applicability.

The cost of developing, producing, and applying biocontrol agents can be a barrier to their adoption, particularly for small-scale farmers and storage facilities with limited resources. Compared to synthetic insecticides, which are often readily available and relatively inexpensive, biocontrol agents may require specialized production facilities and application equipment, increasing the overall cost. The effectiveness of biocontrol agents can vary depending on the storage environment and the presence of other pest management strategies.

9. Conclusion

Biological control offers a sustainable and environmentally friendly approach to managing storage pests, reducing reliance on synthetic pesticides. The integration of biological control agents into IPM programs can enhance the long-term effectiveness of pest management strategies while minimizing adverse effects on human health and the environment.

The adoption of cutting-edge techniques in biological control holds great promise for improving the sustainability and resilience of food storage systems.

Although biological control shows effectiveness, its use is limited because ecological traits like climate circumstances and the host's genetic condition can change the desired impact of the BCA. The integration of biological control agents into integrated pest management programs can improve the long-term efficacy of pest management strategies while minimizing negative consequences for human health and the environment. The creation of biopesticides utilizing RNA interference is also an additional strategy for eliminating storage insect pests. Cultural practices and synthetic fungicides are the most often utilized control methods; nevertheless, the usage of some of these synthetic products has created a variety of issues due to environmental contamination, including consequences such as toxicity to humans and resistance of some pathogens to these fungicides. Over the last 50 years, biological control has remained a component of integrated pest management, exhibiting consistent but encouraging growth. Looking into the significance of biological control, a literature survey was conducted, and a review was made on the extent of damage caused by various pests of major vegetable crops, as well as their potential management using biological control agents.

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