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Carrier Materials in Bio-Input Formulations: Selection Criteria, Shelf-Life Enhancement, and Field Efficacy

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INTRODUCTION

Bio-inputs are biological agents or naturally derived substances such as biopesticides, entomopathogenic microbes, botanical extracts, and biological control agents that manage insect pests while minimizing environmental harm. In modern agriculture, they address the dual challenge of enhancing food production and reducing the negative impacts of synthetic chemical inputs on the environment and human health by utilizing living organisms or their derivatives to suppress pests, thereby lowering reliance on conventional pesticides and promoting sustainability (Chandler *et al*., 2011). These inputs act through diverse mechanisms, including parasitism, predation, infection, feeding deterrence, and growth inhibition of insect species; for instance, microbial biopesticides like *Bacillus thuringiensis* and *Beauveria bassiana* infect and kill larvae and adults, while natural predators such as *Coccinella septempunctata* (ladybird beetle) and parasitoids like *Trichogramma* spp. target eggs and immature pest stages (Lacey *et al*., 2015). Similarly, botanically derived substances such as neem oil and pyrethrins offer significant control through antifeedant and insecticidal properties (Isman, 2006). Bio-inputs provide selective pest control with minimal impact on non-target organisms and integrate effectively into Integrated Pest Management (IPM) and organic farming systems, though their performance may be influenced by environmental factors and shelf-life constraints, requiring proper formulation and application (Glare *et al*., 2012). Driven by regulatory support, consumer demand for pesticide-free produce, and global emphasis on sustainable agriculture, bio-inputs are increasingly recognized as a cornerstone of pest management, with advancements in biotechnology and microbial ecology expected to further enhance their role in future farming systems (Lacey *et al*., 2015).

Carrier Materials: A carrier is a substance used to deliver microbial inoculants to plants and soil while maintaining the viability and activity of the microbes during storage and after application.

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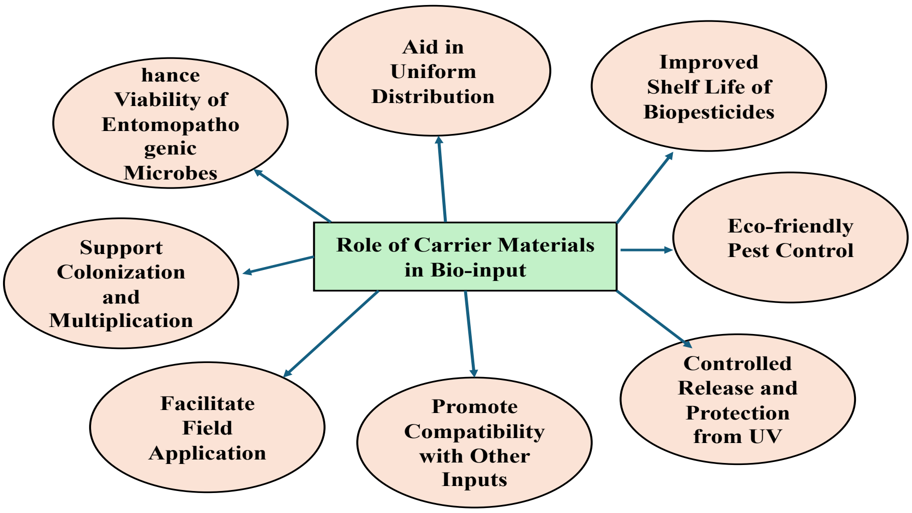
Different types of carrier materials are parent include such as,

Carrier Material	Characteristics	Remarks
Peat	High organic matter, good buffering	Traditional, excellent for rhizobia
Lignite	Good water retention, slow degradation	Used in India, economical
Talc	Inert, smooth, dry, with less microbial interaction	Common in bio-fungicides
Charcoal	Porous, retains moisture, and adsorbs toxins	Helps in long shelf-life
Vermiculite/Perlite	Lightweight, holds water well	Used in pot culture inoculants
Compost/Coir Pith	Rich in nutrients and microbes	Requires proper screening



Role of Carrier Materials:

A) Selection Criteria for Carrier Materials in Bio-input Formulations:

Carrier materials are vital components of bio-input formulations, particularly microbial biopesticides used in entomology to control insect pests. The choice of carrier material directly influences the viability, shelf life, efficacy, and ease of application of beneficial microbes such as entomopathogenic fungi (*Beauveria bassiana*, *Metarhizium anisopliae*) and bacteria (*Bacillus thuringiensis*). Carrier Materials in Bio-input Formulations include different Section criteria (Sahu & Brahmaprakash, 2016 and Malusá *et al*., 2012) such as,

a) Physicochemical Properties:

The physicochemical characteristics of a carrier material play a critical role in maintaining the survival and effectiveness of microbial biopesticides. First and foremost, the carrier should have a neutral pH ranging from 6.5 to 7.5. This near-neutral environment is ideal because many entomopathogenic microorganisms, such as *Beauveria bassiana* and *Metarhizium anisopliae*, thrive and sporulate optimally under neutral pH conditions. A carrier with too acidic or alkaline a pH may inhibit microbial growth, reducing the formulation's efficacy when applied to insect pests. For example, peat moss, which generally has a slightly acidic pH, may need treatment or buffering to adjust its pH before use as a carrier in fungal biopesticide formulations.

Another important physicochemical property is the water-holding capacity of the carrier. A carrier with at least 50% water-holding capacity helps maintain microbial moisture, which is crucial for preventing desiccation of fungal spores or bacterial cells during storage and upon field application. Without adequate moisture retention, the microbes may die or lose infectivity, rendering the bio-input ineffective. For instance, carriers like vermiculite and coir pith are preferred for their high moisture retention, which ensures sustained viability of *Beauveria bassiana* spores when sprayed on aphids or whiteflies. Moisture retention also plays a vital role in field conditions, as it can buffer against harsh environmental factors such as dry winds or intense sunlight.

The particle size of the carrier material also matters significantly. Fine particles between

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10 to 150 micrometers allow for uniform and consistent application, whether as seed coatings, sprays, or soil amendments. Smaller particles ensure better adhesion of microbial spores to insect cuticles or plant surfaces, increasing the likelihood of successful infection of target pests. For example, talc powder with a suitable particle size is often used as a carrier for *Bacillus thuringiensis* formulations because it enables easy mixing and spraying, ensuring the bacteria come in close contact with caterpillars and other insect larvae.

Lastly, good porosity and texture are essential for adequate aeration and moisture balance. Porous carriers like charcoal or expanded clay provide sufficient air spaces to allow aerobic microbes to respire effectively. This improves the shelf life and biological activity of entomopathogenic fungi, which require oxygen for spore germination and host infection. Porous carriers also prevent waterlogging, which could cause anaerobic conditions detrimental to these beneficial microbes.

b) Biological Compatibility:

Biological compatibility refers to the carrier material's ability to support the microbial inoculant without causing harm or inhibition. An ideal carrier must be non-toxic and free from heavy metals, pesticides, or other chemical contaminants that could negatively affect the survival and reproduction of the beneficial microbes. For example, if a carrier contains traces of copper or other heavy metals, these could be lethal to fungal spores or bacterial cells, limiting their pest control potential. Ensuring chemical purity is essential, especially when the goal is to target delicate insect pests such as aphids or thrips, where the microbial agent's viability is crucial for success.

Moreover, carriers should be free from pathogens or competing microorganisms. The presence of undesirable microbes could lead to competition for nutrients or the production of antagonistic compounds, which might suppress the biocontrol agents. For instance, a carrier contaminated with saprophytic fungi or bacteria could outcompete *Metarhizium anisopliae* in the formulation, reducing its effectiveness against pests like beetles or grasshoppers. Therefore, carriers often undergo sterilization to eliminate such contaminants before inoculation with beneficial microbes.

This biological compatibility also extends to the target environment and the pest management system. The carrier must not adversely affect non-target beneficial insects such as pollinators or natural predators like ladybird beetles (*Coccinella septempunctata*), which play a vital role in integrated pest management (IPM). A carrier that supports the microbial biopesticide without harming these beneficial insects enhances the sustainability of pest control programs.

c) Sterilizability:

Sterilizability is a practical yet crucial criterion in selecting a carrier material. Before inoculation with microbial agents, carriers need to be sterilized to eliminate any unwanted microorganisms that may contaminate the bio-input and compromise its efficacy. Common sterilization methods include autoclaving (heat sterilization) and gamma radiation. The carrier material must withstand these processes without degradation or alteration of its physical and chemical properties.

For example, some organic carriers like peat may undergo structural breakdown or loss of water-holding capacity when exposed to high heat, which could reduce their suitability. In contrast, mineral-based carriers like talc or vermiculite generally retain their properties after sterilization. This resistance ensures that the carrier remains an effective medium for microbial survival and delivery post-sterilization.

Maintaining carrier integrity after sterilization is especially important for entomopathogenic fungi and bacteria, which rely on the carrier not only for protection during storage but also for successful application in the field. A damaged carrier could lead to clumping, poor dispersion, or loss of moisture, ultimately reducing the bio-insecticide's effectiveness against pests such as whiteflies or caterpillars.

d) Shelf Life and Stability:

Shelf life and stability refer to the carrier's ability to maintain microbial viability over extended periods, often 6 to 12 months, under ambient storage conditions. This feature is critical for the commercial production, storage, and distribution of microbial biopesticides, ensuring that the product remains effective until it reaches the farmer.

Carriers that provide a stable microenvironment protect microbial spores or cells from temperature fluctuations, UV radiation, and desiccation during storage. For

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instance, talc-based carriers have been widely used because they can extend the shelf life of *Bacillus thuringiensis* formulations used against lepidopteran pests. Similarly, lignite and vermiculite carriers enhance the longevity of *Beauveria bassiana* spores, facilitating their use in aphid and whitefly management over longer storage durations.

Good shelf life also enables bio-input manufacturers to produce large batches of formulations, reducing production costs and improving availability. This is especially important for entomological pest control, where timely and reliable application can significantly reduce pest populations and prevent outbreaks.

e) Environmental and Economic Considerations:

Economic feasibility and environmental sustainability are increasingly important in the selection of carrier materials. An ideal carrier should be low-cost and locally available to support widespread adoption, especially in smallholder farming systems where resources may be limited. Materials like talc, lignite, coir pith, and rice husk are often preferred because they are abundant and inexpensive in many agricultural regions.

Furthermore, carriers must be biodegradable and eco-friendly, minimizing the ecological footprint of bio-input application. Biodegradability ensures that the carrier does not accumulate in the environment, reducing soil and water contamination risks. This is particularly important in pest management strategies that emphasize conservation of beneficial organisms, including pollinators and natural predators. For example, biodegradable carriers like coir pith and peat are favoured in sustainable agriculture as they degrade naturally without harming soil health.

B) Shelf-Life Enhancement Strategies

Microbial inoculants are living organisms, and their effectiveness depends on their viability during storage. Appropriate strategies can significantly extend their shelf life without compromising efficacy, especially when used in entomological biocontrol.

a) Use of Suitable Carrier Materials

Carrier materials are critical for delivering microbial biopesticides effectively. Materials like talc and lignite are widely used in solid formulations due to their inert nature and moisture-absorbing capacity, which supports microbial survival during storage. Talc, being chemically neutral, provides a stable medium that minimizes microbial degradation, whereas lignite, an organic-rich material, enhances nutrient retention and moisture regulation, which is particularly beneficial for spores of *Beauveria bassiana* used against aphids and whiteflies (Hegde & Vijaykumar, 2022).

Charcoal serves a dual function: it protects the microbial cells from harmful metabolites due to its adsorptive surface and acts as a reservoir for nutrients. For example, formulations using charcoal carriers have shown enhanced survival of *Metarhizium anisopliae*, which is effective against soil-borne insect pests such as termites (John *et al*., 2020). These carriers not only increase the longevity of biopesticides but also make them easier to apply in field conditions.

b) Moisture Content Control

Maintaining the right moisture content is essential in solid microbial formulations. The ideal moisture range of 30–40% ensures that the microbial cells remain active but not overly hydrated, which could encourage contaminant growth. Lower moisture levels can lead to desiccation and cell death, severely limiting the effectiveness of products like *Bacillus thuringiensis*, which is widely used against caterpillars in vegetable crops (Shilpa & Brahmaprakash, 2016).

In experimental trials, *Bacillus subtilis* inoculants stored at 35% moisture in lignite carriers maintained high viability for over six months, while samples with 20% moisture experienced a drastic drop in viable cell count. The regulation of moisture thus ensures longer shelf life and better field performance (Bashan *et al*., 2014).

c) Storage Temperature

Storage temperature plays a pivotal role in maintaining the viability and efficacy of microbial inoculants. Lower temperatures, typically between 4°C and 10°C, are effective in slowing down the metabolic activities of microbial cells. This reduction in metabolic rate minimizes energy consumption and the accumulation of harmful metabolic byproducts, thereby extending the shelf life of the inoculants. For instance, a study by Kurniawan *et al.* (2022) demonstrated that *Beauveria bassiana* maintained high viability when cultured on rice bran supplemented with cricket flour, suggesting that appropriate media and storage conditions can enhance fungal viability.

Moreover, refrigeration prevents the sporulation of contaminants and suppresses the rapid degradation of microbial components. In tropical climates where ambient temperatures are high, cold storage is especially vital to maintain the shelf stability of entomopathogens like *Metarhizium anisopliae*, which otherwise rapidly lose virulence (Sarma *et al*., 2023).

d) Packaging Innovations

Innovative packaging plays a pivotal role in preserving the quality of microbial biopesticides. Aluminium foil pouches and multi-layered laminated bags act as barriers against moisture and gas exchange, thereby preventing oxidation and microbial contamination. These packaging types have been shown to significantly enhance the stability of *Bacillus thuringiensis* formulations used against lepidopteran pests such as *Spodoptera litura* (Vemmer & Patel, 2013).

For instance, a study demonstrated that vacuum-packaged *B. bassiana* blastospores remained viable longer when stored at 4°C compared to 28°C, with virtually no loss in viability over nine months, regardless of the drying method used (Mascarin *et al*., 2016). Similarly, research on *T. harzianum* showed that encapsulated formulations stored at 4°C maintained viability over extended periods (Gola *et al*., 2016).

e) Additives and Protective Agents

Additives such as polymers and cryoprotectants help stabilize microbial inoculants during drying, storage, and field application. Alginate encapsulation is a common method that encases microbial cells in a polymer matrix, protecting them from UV light, desiccation, and temperature fluctuations. For example, alginate beads containing *Beauveria bassiana* showed extended viability and successful control of whiteflies (*Bemisia tabaci*) in tomato fields (Sarma *et al*., 2023).

C) Field Efficacy of Carrier-based Bio-inputs

The effectiveness of microbial-based biocontrol agents in the field depends significantly on how well the carrier supports microbial viability, activity, and adaptability under varying environmental conditions. Proper formulation using an appropriate carrier can improve pest suppression, especially in insect pest management using entomopathogenic organisms.

a) Colonization Ability

The ability of microbial inoculants to colonize target environments—particularly plant roots or phyllospheres—is critical for their effectiveness. A high viable count (≥10^8 CFU/g) in the carrier ensures enough microbial cells survive during storage and application to establish themselves in the field. For example, charcoal and talc-based carriers have shown promising results in supporting *Beauveria bassiana*, an entomopathogenic fungus, by enhancing its spore viability and colonization on plant surfaces infested with aphids.

Carriers with adsorptive and porous properties, such as lignite and charcoal, provide microhabitats that protect microbial cells from desiccation and UV exposure post-application. Ghabru *et al*. (2023) reported that using carrier-based *Rhizobium* formulations maintained microbial viability during storage and improved root colonization, which is critical for successful pest suppression in the rhizosphere by secondary metabolite-producing microbes.

b) Compatibility with Application Methods

The method of applying microbial biocontrol agents depends largely on the carrier used. For seed coating, talc-based formulations offer superior adhesion to seeds, allowing entomopathogenic bacteria or fungi to germinate upon seedling emergence. In soil applications, compost and peat-based carriers enhance rhizosphere colonization, creating an environment conducive to the activity of insect-parasitic fungi like *Metarhizium anisopliae*. For foliar sprays, liquid carriers are preferred as they provide uniform distribution and maintain spore viability on plant surfaces.

A study by Yimer & Abena (2019), emphasized the importance of selecting carriers compatible with different application methods, noting that talc, peat, and liquid carriers each cater to specific microbial delivery needs in the field. For instance, when using *Beauveria bassiana* for aphid control, foliar applications with water-based carriers enhanced field persistence and efficacy compared to dry formulations.

c) Performance Under Stress

Field conditions often involve abiotic stresses like temperature extremes, salinity, or low soil moisture, which can adversely affect microbial viability and performance. Carriers that buffer

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microbes against these stresses play a crucial role in sustaining biocontrol efficacy. Biochar, for instance, improves water-holding capacity and nutrient retention in soils, supporting microbial inoculants during droughts. This resilience enhances the survival of insect-pathogenic organisms like *Bacillus thuringiensis*, increasing their efficacy in pest control.

A systematic review by Rathinapriya *et al*., (2024) found that biochar-amended carriers improved soil health and microbial survival under abiotic stress, especially salinity and drought, which are critical factors affecting microbial biocontrol activity against soil and foliar insect pests.

d) Enhancement of Plant Growth

Besides direct pest control, microbial bio-inputs in effective carriers can promote plant growth through nitrogen fixation, phosphorus solubilization, and production of phytohormones. This growth promotion also indirectly increases the plant's resistance to pest attacks. For example, formulations of *Trichoderma*, *Azotobacter*, and *Bacillus subtilis* have demonstrated improved root development and enhanced plant defense responses, which can reduce aphid infestations by strengthening host resistance.

Khandare *et al*. (2020) reported that carrier-based bio-inoculants of *Azotobacter* and phosphate-solubilizing bacteria not only enhanced wheat growth but also improved soil microbial diversity and resistance. In pest-prone conditions, these improvements create a more resilient crop system less susceptible to insect pressure, highlighting the indirect pest-suppressive effects of well-formulated bio-inputs.

Classification of Bio-input Tools in Pest Management: Categories, Types, Examples, and Target Pests:

Category	Type	Examples	Mode of Action	Target Insect Pests
Microbial Biopesticides	Bacteria	*Bacillus thuringiensis* (Bt)	Toxin production leading to gut paralysis and death	Caterpillars, mosquitoes, beetles
	Fungi	*Beauveria bassiana, Metarhizium anisopliae*	Penetrate cuticle, colonize body, cause insect death	Aphids, whiteflies, thrips, beetles
	Viruses	*Nucleopolyhedro virus (NPV), Granulo virus (GV)*	Infect midgut and multiply in cells, leading to lysis	Lepidopteran larvae (e.g., Helicoverpa)
	Nematodes	*Steinernema spp., Hetero rhabditis spp.*	Enter through natural openings and release symbiotic bacteria	Soil-dwelling insect larvae (e.g., grubs, borers)
Botanical Insecticides	Plant-based compounds	Neem oil (*Azadirachtin*), Pyrethrin	Feeding deterrence, oviposition inhibition, growth disruption	Aphids, jassids, whiteflies, beetles
Predators	Beneficial insects	*Coccinella septempunctata*, Lacewings, Predatory bugs	Consume multiple life stages of pests	Aphids, scales, mealybugs, mites
Parasitoids	Egg, larval or pupal parasitoids	*Trichogramma spp., Aphidius colemani*	Lay eggs inside/on pest eggs or larvae, causing death	Lepidopteran eggs, aphids
Pheromones & Semiochemical	Mating disruption and traps	Sex pheromones (e.g., Helilure), kairomones	Attract or confuse pests to reduce mating or monitor	Moths, fruit flies, borers

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