



The Soil-Dwelling Shield: Harnessing Native Entomopathogenic Fungi and Nematodes Against the White Grub Crisis

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White grubs (Coleoptera: Scarabaeidae) represent one of the most destructive subterranean pest complexes, causing multi-cropland devastation across India. Traditional chemical control strategies fail due to soil matrix buffering, pesticide binding, environmental toxicity, and destruction of soil ecology. Native entomopathogenic fungi (EPF)—such as *Metarhizium anisopliae* and *Beauveria* spp.—and entomopathogenic nematodes (EPNs)—including *Steinernema* and *Heterorhabditis* spp.—offer an eco-friendly, self-sustaining biological shield. This article explores their life cycles, pathogenesis, indigenous strain isolation, mass production technologies, advanced delivery formulations, and seamless integration into holistic White Grub Management programs.

The Hidden Crisis Below: Unmasking the White Grub Menace

Among subterranean agricultural pests, white grubs—the C-shaped larval stages of scarab beetles (Coleoptera: Scarabaeidae)—pose an unprecedented threat to global food security and farmer livelihoods. Known colloquially as 'root grubs', these soil-dwelling larvae devour the root systems of cash crops, cereals, oilseeds, pulses, commercial plantations, and turfgrasses. In India, white grubs have transitioned from localized, periodic outbreaks to a multi-regional agrarian crisis.

The covert nature of white grub damage exacerbates its severity. Because feeding occurs entirely below the soil surface, initial symptoms above ground—such as yellowing, stunting, wilting, and moisture stress—are frequently misdiagnosed as nutrient deficiencies or drought. By the time lodging or complete plant death becomes visible, the root system is irrevocably destroyed. A single mature third-instar grub per square meter can reduce groundnut yields by up to 20%, while populations exceeding 5 to 10 grubs per plant can cause total crop failure across vast contiguous field stretches.

Major Economic Species and Host Diversity in India

India hosts a rich diversity of phytophagous scarabs, with distinct species dominating specific agro-ecological zones:

- *Holotrichia consanguinea* Blanchard: Dominates the light sandy loam soils of Rajasthan, Gujarat, Haryana, Uttar Pradesh, and Punjab. Highly destructive to groundnut, bajra, sorghum, and sugarcane.
- *Holotrichia serrata* Fabricius: Prevalent across South India (Karnataka, Maharashtra, Tamil Nadu, Andhra Pradesh), attacking sugarcane, groundnut, potato, maize, and soybean.
- *Leucopholis coneophora* Burmeister & *Leucopholis lepidophora* Blanchard: Major threats in the humid, high-rainfall regions of Kerala and Coastal Karnataka, devastating arecanut, coconut, rubber, and spice plantations.
- *Anomala dimidiata* & *Phyllophaga* spp.: Predominant in the North-Western Himalayan region and North-Eastern hilly tracks, causing severe damage to potato, ginger, and temperate fruit nurseries.

Life Cycle Dynamics and Subterranean Vulnerability

The life cycle of most economic scarab species in India is univoltine (one generation per year). The adult beetles emerge from the soil en masse following the first pre-monsoon summer rains (May–June). After mating on preferred host trees (such as neem, acacia, ber, and Ailanthus), females re-enter the moist soil to deposit eggs. The emerging L1 larvae feed on fine rootlets and organic matter, progressing rapidly to L2 and L3 instars.

The L3 stage, lasting 2 to 3 months during the peak crop growth period (July–October), is the most voracious and damaging. As winter approaches, mature larvae burrow deeper into the subsoil (30–90 cm) to construct earth cells for pupation. The adults remain dormant within these subterranean pupal chambers throughout winter and summer until triggered by the next season's monsoonal rains.

Why Chemical Magic Bullets Fail in the Subsurface

For decades, chemical insecticides such as chlorpyrifos, imidacloprid, fipronil, and phorate were considered the primary defense against white grubs. However, conventional chemical drenches and soil granules face severe physical, chemical, and biological barriers in subterranean ecosystems:

1. **Soil Matrix Buffering and Adsorption:** Soil is a complex matrix of clay minerals, organic matter, and soil moisture. High organic carbon and clay contents tightly bind synthetic insecticide molecules, dramatically reducing their bioavailability in the active root zone where grubs reside.
2. **Leaching and Groundwater Contamination:** Highly soluble systemic chemicals (e.g., neonicotinoids) leach rapidly through light sandy soils during heavy monsoons, contaminating rural drinking water aquifers and leaving the upper root zone unprotected.
3. **Depth Paradox and Application Timing:** L2 and L3 grubs frequently move dynamically up and down the soil profile depending on moisture and temperature. Liquid chemical drenches rarely penetrate beyond 5–10 cm depth, failing to reach larvae feeding deeper in the rhizosphere.
4. **Ecological Collateral Damage:** Broad-spectrum chemical insecticides destroy beneficial non-target soil fauna, including earthworms, predatory mites, ground beetles, and native mycorrhizal fungal networks, leading to soil biological dead zones.
5. **Resistance and Residue Accumulation:** Rapid degradation of repeated chemical applications by soil microbes (enhanced microbial degradation) combined with selection pressure leads to insecticide resistance and toxic residue accumulation in harvested tubers and pods.

The Native Biological Arsenal: Unveiling Entomopathogenic Fungi (EPF)

In response to chemical control failures, native Entomopathogenic Fungi (EPF) have emerged as highly potent, ecologically sound bio-pesticides. Unlike bacteria or viruses that must be ingested, EPF infect host insects through direct cuticle contact and enzymatic penetration, making them uniquely suited for soft-bodied subterranean pests like white grubs.

Mode of Infection and Pathogenesis

The infectious cycle of EPF on white grubs follows a rigorous biochemical sequence:

- a. **Adhesion:** Asexual fungal spores (conidia) attach to the insect cuticle via hydrophobic and electrostatic interactions.
- b. **Germination & Appressorium Formation:** Under adequate soil humidity (>80% RH), conidia germinate and form appressoria—specialized penetration structures.
- c. **Enzymatic Degradation:** The fungus secretes a cocktail of cuticle-degrading enzymes, including PR1 proteases, endochitinases, and esterases, breaching the tough chitin-protein insect integument.
- d. **Invasion & Toxin Production:** Hyphae enter the hemocoel, converting into blastospores. The fungus releases lethal secondary metabolites—such as destruxins (in *Metarhizium*) and beauvericin/bassianolide (in *Beauveria*)—which paralyze the insect, disrupt immune responses, and cause systemic organ failure.

e. Muscardine Symptom & Sporulation: Post-death, fungal hyphae emerge through intersegmental membranes, coating the cadaver in a fluffy mycelial mat (green or white muscardine) and releasing millions of new infective conidia into the soil matrix.

Key Native EPF Species for White Grub Suppression

Metarhizium anisopliae (Metschn.) Sorokin (Green Muscardine Fungus): Considered the premiere fungal pathogen against scarabs worldwide. Indigenous strains isolated from grub-infested fields exhibit remarkable thermotolerance and soil persistence. Infected cadavers exhibit a characteristic dark green conidial crust.

Beauveria brongniartii (Sacc.) Petch: A specialized pathogen of scarabaeid larvae, particularly effective against *Holotrichia serrata* and *Leucopholis* spp. It produces dense white chalky blooms over dead grubs and exhibits high virulence at cooler soil temperatures.

Beauveria bassiana (Bals.-Criv.) Vuill.: While broadly active against foliar pests, native soil-dwelling strains of *B. bassiana* demonstrate high infectivity against early L1 and L2 instar white grubs.

The Superiority of Native Strains over Exotic Isolates

Native EPF isolates are naturally adapted to local soil pH ranges, temperature fluctuations, and soil microbial competition. They persist in the soil bank as natural regulators, establishing long-term equilibrium without altering indigenous microbial biodiversity.

The Microscopic Seekers: Entomopathogenic Nematodes (EPNs)

While EPF act as sessile biological agents waiting for physical contact, Entomopathogenic Nematodes (EPNs) from the families Steinernematidae and Heterorhabditidae function as active, lethal microscopic hunters. EPNs exist in an obligate mutualistic symbiosis with insect-pathogenic bacteria (*Xenorhabdus* spp. for *Steinernema*; *Photorhabdus* spp. for *Heterorhabditis*).

Host-Seeking Strategies: Ambushers vs. Cruisers

EPN species exhibit distinct foraging behaviors essential for targeting white grubs situated at varying soil depths:

- Cruiser Foragers (*Heterorhabditis bacteriophora*, *Steinernema glaseri*, *Steinernema indica*): Cruisers actively navigate through soil pores in response to long-range host cues, including carbon dioxide (CO₂), root volatiles, and insect excretions. Cruisers excel at hunting soil-dwelling L2 and L3 grubs located deep within the root zone.
- Ambush Foragers (*Steinernema carpocapsae*): Ambushers remain stationary near the soil surface, standing on their tails (nictation) to attach to mobile insects. They are effective against surface-dwelling adult beetles during emergence or egg-laying.

Lethal Pathogenesis Cycle

The Non-Feeding Infective Juvenile (IJ or 'dauer' larva) is the only free-living soil stage. IJs enter the white grub through natural physiological openings (mouth, anus, spiracles) or directly pierce thin intersegmental membranes using a head tooth (in *Heterorhabditis*).

Once inside the hemocoel, IJs release their symbiotic bacteria (*Xenorhabdus* or *Photorhabdus*). The bacteria multiply exponentially, producing potent enterotoxins, immunosuppressive factors, and broad-spectrum antibiotics that suppress opportunistic soil microbes. The insect dies of septicemia within 24 to 48 hours. The cadaver turns characteristic colors—brick red/luminescent for *Heterorhabditis* (due to *Photorhabdus luminescens*) or brownish/ochre for *Steinernema*. Nematodes feed on bacterial bioconverted host tissues, complete 1 to 3 generations, and release tens of thousands of new IJs from the depleted cadaver back into the soil.

EPF + EPN SYNERGY IN THE RHIZOSPHERE

Combining Metarhizium anisopliae with Heterorhabditis indica produces a synergistic dual-biocontrol effect. Sub-lethal fungal infection weakens the grub's cuticle and immune response, allowing nematodes to penetrate 3x faster and achieve up to 95% grub mortality in high-infestation fields.

Mass Production, Formulation, and Quality Control

To translate laboratory efficacy into scalable field solutions, robust bioprocess engineering and quality control standards are required.

Mass Production Protocols

EPF Solid-State Fermentation: EPF are mass-produced using a two-stage diphasic system. Fungal biomass is first multiplied in liquid media (dextrose-yeast extract), then transferred to sterilized agricultural grains (broken rice, sorghum, or maize grit) in autoclavable polypropylene bags. Under controlled temperature ($25\pm 2^\circ\text{C}$) and relative humidity (80-80%), dense conidial harvest is achieved in 14-21 days.

EPN In Vivo and Liquid Culture: EPNs are mass-produced in vivo using greater wax moth larvae (*Galleria mellonella*) for small-scale operations, yielding up to 100,000 IJs per larva. For commercial scale, monoxenic liquid fermentation using three-dimensional sponge matrices or liquid bioreactors containing symbiotic bacteria provides high yield density.

Advanced Subterranean Formulations

1. **Wettable Powders (WP) & Granules (WG):** Conidial powders blended with inert talc, kaolin, or diatomaceous earth with added UV-protectants and wetting agents.

2. **Oil Suspensions (OD):** Emulsifiable vegetable-oil formulations that coat conidia, preserving moisture and extending spore viability in dry soil conditions.

3. **Infective Juvenile Hydrogel Encapsulation:** EPNs encapsulated in sodium alginate or water-retaining hydrogel beads, protecting IJs from desiccation during soil application.

Field Delivery Protocols and Best Management Practices

Biological control success depends entirely on timing, environmental moisture, and precise soil delivery. Unlike synthetic chemicals, living bio-agents must be protected from environmental extremes.

Crucial Application Guidelines

- **Target Window:** Apply bio-agents during early monsoon (June–July) when grubs are in vulnerable L1 and L2 stages near the soil surface. Mature L3 grubs require higher dosages.
- **Time of Day:** Execute soil drenching in late afternoon or evening to protect sensitive EPF spores and EPN IJs from solar ultraviolet (UV-A and UV-B) degradation.
- **Soil Moisture Management:** Irrigate fields lightly prior to and immediately following bio-agent application. Soil moisture must be maintained above 15–20% field capacity for EPN movement and fungal germination.
- **Enriched FYM / Vermicompost Method:** Mix 2.5–5.0 kg of fungal formulations (10^8 CFU/g) or EPN formulations (10^9 IJs/ha) into 250 kg of well-decomposed Farmyard Manure (FYM) or vermicompost per acre. Incubate under shade for 7 days before soil incorporation along crop rows.
- **Equipment Modifications:** Remove nozzle strainers and fine filters (<50 mesh) from knapsack sprayers when applying EPNs to prevent mechanical shear stress and clogging.

Integrated White Grub Management (IWGM) Architecture

Native EPF and EPNs serve as the biological backbone of a multi-tiered Integrated White Grub Management (IWGM) framework:

Management Tier	Action / Strategy	Target Stage & Timing
Cultural & Mechanical	Deep summer plowing to expose pupae to avian predators; installation of solar LED light traps (1 trap/ha) during first monsoon rains.	Pupae & Emerged Adults (May–June)
Host Tree Management	Spraying host trees (neem, acacia) with recommended bio-pesticides or neem oil; mechanical shaking of trees at night to collect beetles.	Adult Beetles (June)
Biological Defense (Soil)	Soil application of native <i>Metarhizium anisopliae</i> / <i>Beauveria brongniartii</i> enriched in FYM + <i>Heterorhabditis indica</i> drenching.	L1 & L2 Instar Larvae (July–August)

Management Tier	Action / Strategy	Target Stage & Timing
Botanical Support	Soil application of neem cake / mahua cake (250 kg/ha) at sowing to deter egg laying and provide slow-release nitrogen.	Eggs & Early Larvae (June–July)

Case Studies, Field Efficacy Data, and Farm-Level Successes

Multi-location field trials conducted across major agricultural universities in India demonstrate the outstanding efficacy of native EPF and EPN strains in groundnut, sugarcane, and arecanut systems.

Groundnut Eco-Zones of Rajasthan & Gujarat

In trials targeting *Holotrichia consanguinea*, field applications of native *Metarhizium anisopliae* (1×10^{12} spores/ha) combined with *Heterorhabditis indica* (2.5×10^9 IJs/ha) achieved 82.5% reduction in grub population, yielding a 38% increase in pod yield compared to chemical treatments (chlorpyrifos 20 EC), while preserving indigenous soil earthworm populations.

Sugarcane Belts of Maharashtra & Karnataka

Against *Holotrichia serrata* in sugarcane, soil incorporation of *Beauveria brongniartii* enriched in pressmud (1×10^{13} conidia/ha) resulted in 79% mortality of L2 larvae within 21 days, completely preventing root lodging and extending ratoon crop longevity.

Arecanut Plantations of Coastal Karnataka

In heavy-rainfall arecanut ecosystems infested with *Leucopholis coneophora*, application of *Steinernema carpocapsae* and native *M. anisopliae* via root zone drenching restored root density by 45% over two consecutive seasons and reduced reliance on synthetic organophosphates to zero.

Challenges and Future Research Horizons

Despite impressive field results, widespread commercial adoption of soil-dwelling bio-agents faces key technical and market challenges:

1. Shelf-Life and Storage Stability: Liquid and powder EPF/EPN formulations require cool storage (4-10°C) to maintain viability beyond 6 months, challenging rural supply chains.
2. Desiccation and Thermal Stress: Elevated surface soil temperatures (>38°C) during dry spells restrict bio-agent persistence prior to canopy closure.
3. Registration & Quality Control: Harmonizing CIB&RC registration standards for native biological strains and establishing stringent QC labs to prevent sub-standard bio-pesticide marketing.
4. Future Horizons: Genetic selection for heat and drought-tolerant EPF strains, microencapsulated delivery systems, genome sequencing of hyper-virulent *Xenorhabdus* bacterial toxins, and AI-driven precision soil drenching equipment.

Conclusion

The white grub crisis demands a paradigm shift from toxic chemical drenches toward ecologically resilient biocontrol strategies. Native Entomopathogenic Fungi and Nematodes represent a powerful, self-perpetuating subterranean shield. By establishing indigenous bio-agent mass-production facilities, refining farmer-friendly soil application techniques, and embedding biological control within Integrated White Grub Management, Indian agriculture can safeguard crop productivity, restore soil health, and secure sustainable farming livelihoods for generations to come.

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