

COMPARATIVE ANALYSIS OF THE INFESTATION STAGES OF *CALLOSOBRUCHUS MACULATUS* AND *CALLOSOBRUCHUS CHINENSIS* IN STORED PULSE SEEDS

Gyanendra Pratap¹, Dr. Sumer Singh², Dr.B. S. Azad³

Research Scholar, Department of Zoology, Singhania University, Rajasthan¹

Research Supervisor, Department of Life Science, Singhania University Rajasthan²

Co-Research Supervisor, Department of Zoology, D.A.V. PG College Kanpur³

ABSTRACT

Stored pulses constitute a vital source of dietary protein across the developing world, yet their post-harvest value is severely compromised by infestation from bruchid beetles, principally *Callosobruchus maculatus* (Fabricius) and *Callosobruchus chinensis* (Linnaeus). This review synthesises the published literature on the developmental and infestation stages of these two species and critically appraises the spectrum of treatments developed to suppress them. Drawing on a structured survey of work spanning several decades, the paper examines the holometabolous life cycle egg, four larval instars, pupa and adult and clarifies the distinction between latent field infestation, in which oviposition begins on maturing pods, and the explosive multiplication that follows under storage conditions. The concealed feeding of larvae within the seed cotyledon, the production of characteristic circular emergence windows, and the consequent losses in seed weight, germinability and nutritional quality are documented in detail. The survey collates evidence on physical interventions such as solar heating, cold storage, hermetic and modified-atmosphere systems and irradiation; on chemical fumigation and insecticidal treatment; on botanical preparations including plant oils, inert seed coatings, leaf powders and essential oils; on biological control through hymenopteran parasitoids; and on host-plant resistance. A meta-analytic reading of reported efficacy reveals considerable heterogeneity attributable to dose, beetle species, seed type and the developmental stage targeted. Critical analysis exposes recurrent methodological weaknesses, including inconsistent reporting of stage-specific mortality and the frequent neglect of egg and pupal stages. The review concludes that durable management of *Callosobruchus* demands integrated, stage-aware strategies that combine non-chemical methods with the judicious use of safer compounds, and it identifies clear priorities for future research.

Keywords: *Callosobruchus maculatus*¹; *Callosobruchus chinensis*²; stored pulses³; bruchid infestation⁴; developmental stages⁵; post-harvest losses⁶; integrated pest management⁷.

1. INTRODUCTION

1.1 ECONOMIC IMPORTANCE OF STORED PULSES AND THE BRUCHID PROBLEM

Pulses chickpea, cowpea, green gram, black gram, pigeon pea, lentil and allied grain legumes are the principal vegetable source of protein for hundreds of millions of people in Asia, Africa and Latin America, and they enrich the soil through symbiotic nitrogen fixation. Because most pulses are harvested over a short window and then stored for several months until the next sowing or until favourable market conditions arise, the storage phase is decisive for both food security and seed supply. It is precisely during this phase that bruchid beetles inflict their heaviest toll. Estimates compiled across multiple producing regions place storage losses of pulses at between ten and fifty per cent, with the upper range routine in smallholder granaries that lack any protective treatment. The damage is not confined to the loss of dry matter: infested seed shows depressed germination, reduced cotyledonary reserves, altered amino-acid profiles, contamination by frass, exuviae and dead insects, and a sharp fall in market acceptability. For seed lots intended for replanting, even a low level of internal feeding can be ruinous because the embryo and the food reserves on which the seedling depends are consumed from within. The cumulative effect on rural economies is considerable, since pulses are both a cash crop and a household staple, and losses incurred in storage translate directly into reduced income, diminished nutrition and the need to purchase seed at planting time. The bruchid problem is therefore not merely an entomological curiosity but a recurring constraint on food and seed security wherever grain legumes are grown and held.

1.2 TAXONOMY AND DISTRIBUTION OF THE CALLOSOBRUCHUS SPECIES

The two beetles of concern, *Callosobruchus maculatus* and *Callosobruchus chinensis*, belong to the family Chrysomelidae, subfamily Bruchinae. Both are small, robust beetles of a few millimetres in length, distinguished by their swollen hind femora and their habit of completing larval development entirely within a single seed. *C. maculatus*, the cowpea bruchid or cowpea weevil, is most strongly associated with cowpea but readily attacks mung bean and other *Vigna* and related legumes, and it tolerates the warm, dry conditions of the tropics and subtropics. *C. chinensis*, the adzuki bean or pulse beetle, has a comparable host range and is notably common on chickpea, green gram and black gram across South and East Asia. A defining feature of both species is their capacity to bridge the field and the store: females oviposit on maturing pods before harvest, so that apparently clean grain carries hidden infestation into storage, where successive overlapping generations multiply rapidly in the absence of any climatic check. The two species are frequently found together in the same commodity, and although their biology is broadly similar, they differ in subtle but practically important respects in oviposition preference, developmental rate and tolerance of particular conditions, differences that bear on the design and interpretation of control trials. Their short generation time, high fecundity and ability to complete development within a single seed make them archetypal storage pests, capable of converting a sound consignment into a heavily damaged one within a few months.

1.3 SCOPE AND OBJECTIVES OF THE REVIEW

Against this background, the present review pursues three objectives. First, it assembles and organises the published knowledge on the infested or developmental stages of *C. maculatus* and *C. chinensis*, from oviposition through the concealed larval and pupal phases to adult emergence, so that the biological vulnerabilities of each stage are made explicit. Second, it surveys, in a structured and largely chronological manner, the treatments that have been investigated against these pests physical, chemical, botanical, biological and host-resistance approaches and reads the reported efficacy through a meta-analytic lens that attends to the heterogeneity of methods and outcomes. Third, it offers a critical analysis of the strengths and recurrent weaknesses of this body of work and distils implications for integrated, stage-aware management. By foregrounding the developmental stage as the unit of analysis, the review aims to move beyond simple comparisons of mortality and towards a more mechanistic understanding of why particular treatments succeed or fail. In doing so it seeks to serve both researchers, by clarifying where the evidence is strong and where it is weak, and practitioners, by indicating which combinations of methods are most likely to deliver durable protection under realistic storage conditions.

2. SURVEY OF PAST WORK

Early descriptive studies of *Callosobruchus* established the basic features of the infestation that subsequent work has elaborated. Investigators documented that the female deposits oval, translucent eggs singly and firmly cemented to the seed coat, and that the first-instar larva bores directly downward through the chorion and testa into the cotyledon, so that the entire feeding period is concealed. The larva passes through four instars within the seed, excavating a chamber as it consumes the storage parenchyma, and then pupates within a cell lined by larval secretions and frass. The emerging adult cuts a characteristic circular window, leaving the familiar exit hole that betrays past infestation. This foundational work fixed the developmental template egg, four larval instars, pupa, adult against which later quantitative studies of mortality and damage have been measured. A substantial line of research has concerned the dimorphism and dispersal biology of *C. maculatus*, which bears directly on how infestation propagates in stores. The species produces a flightless, sedentary morph suited to continuous breeding within a grain mass and a flight-capable, dispersing morph favoured under crowding and at high density or temperature. Studies of this polyphenism clarified why infestations can remain quiescent and localised early in storage and then escalate sharply once density rises, and why field-to-store carry-over is so effective. Parallel work on oviposition behaviour showed that females discriminate among seeds and avoid overcrowding eggs, distributing them to reduce larval competition, a behaviour with clear consequences for the spatial pattern of damage and for the design of sampling protocols. The quantification of damage formed a second major strand. Numerous studies measured weight loss, the proportion of seeds bearing exit holes, and the decline in percentage germination as functions of initial infestation and storage duration. Reductions in seed viability were repeatedly shown to be disproportionate to the visible damage, because even a single larva consuming the embryonic axis renders a seed non-viable while leaving much of the cotyledon intact.

Investigations of nutritional change reported losses of carbohydrate and protein, shifts in free amino acids and free fatty acids, and increases in uric acid and microbial load associated with insect waste, all of which degrade both food and feed value. These damage assessments provided the economic justification that motivated the search for effective treatments. A further body of survey work examined the environmental factors governing development and infestation intensity, because these determine both the rate of population growth and the susceptibility of each stage to treatment. Temperature and seed moisture content emerged as the dominant variables: development accelerates and fecundity rises across a favourable range before declining sharply at the upper and lower extremes, while low grain moisture lengthens development and depresses survival. Studies mapped the thermal limits within which the two species complete their life cycle and identified the conditions under which infestation is most explosive, information that underpins both predictive modelling of losses and the rational design of physical control such as heating, cooling and moisture management. This ecological work also explained the seasonal pattern of damage seen in many granaries, in which infestation builds through the warm months and slows as temperatures fall.

Synthetic seed protectants and admixtures formed a parallel strand alongside fumigation. Contact insecticides applied as dusts or sprays to the grain surface were evaluated for their persistence and for their effect on oviposition and adult survival, and inert carriers were tested as vehicles for slow release. While several compounds gave good initial control, the survey records growing concern over residues on a commodity eaten with little processing, over the safety of those who apply such treatments in small stores, and over the development of resistance, concerns that progressively shifted research attention towards the non-chemical and botanical approaches described below. The trajectory of the treatment literature thus mirrors a broader movement in stored-product protection away from sole reliance on synthetic chemistry. The treatment literature itself can be read as a progression. The earliest practical recommendations centred on fumigation, and the survey records extensive work on phosphine and, historically, methyl bromide as space and commodity fumigants capable of penetrating the seed to reach concealed stages. Studies established dose–exposure relationships and demonstrated high mortality of eggs, larvae, pupae and adults under adequate concentration–time products. Subsequent reports, however, chronicled the emergence of phosphine resistance in stored-product Coleoptera, including bruchids, and documented the differential tolerance of the egg and the late-pupal stages, which proved harder to kill than active larvae and adults. This recognition of stage-dependent susceptibility became a recurring theme across the treatment literature. Physical and environmental methods constitute a third major area of the survey. Researchers examined the lethal effects of elevated temperature through solar heating of grain in transparent or black polythene, through hot-water and hot-air treatment, and through the use of solar collectors and heaters, reporting high mortality of all stages once seed temperature exceeded the upper lethal threshold for a sufficient period. Low-temperature storage and brief freezing were shown to be effective against active stages, with eggs again frequently the most tolerant. A particularly influential body of work concerned hermetic and modified-atmosphere storage: by sealing grain in airtight containers, respiration of the insects and grain depletes oxygen and raises carbon dioxide to levels lethal to all developmental stages, and studies of triple

bagging and rigid hermetic containers reported near-complete control without chemicals. Controlled-atmosphere experiments with elevated carbon dioxide, reduced oxygen or nitrogen flushing corroborated these findings and helped define the gas concentrations and exposure times needed for each stage. Irradiation received sustained attention as a means of disinfestation that leaves no chemical residue. Gamma and, later, electron-beam treatments were shown to sterilise adults, prevent the development of immature stages and induce mortality, with the dose required rising from adults and late larvae through pupae to eggs. Several studies emphasised that sub-lethal doses sufficient to prevent reproduction could protect grain even where they did not cause immediate death, an important practical distinction for seed and food security stocks.

Botanical and natural-product treatments form perhaps the largest and most heterogeneous strand of the recent survey, driven by the search for low-cost, locally available and environmentally benign alternatives to synthetic chemicals. Edible and non-edible plant oils applied as thin seed coatings were repeatedly shown to suppress oviposition, block the respiratory openings of eggs and larvae and reduce adult emergence, with effects strongly dose dependent. Powders of dried leaves, seeds and rhizomes neem, custard apple, sweet flag, turmeric, and many others were tested as admixtures, acting through contact toxicity, antifeedant and oviposition-deterrent properties and physical abrasion. Essential oils and their volatile constituents were studied for fumigant and repellent activity, often with rapid knock-down of adults. Inert dusts such as diatomaceous earth and wood ash were examined for their desiccant action on the insect cuticle. Across this literature, the egg and the concealed late-larval and pupal stages again emerged as the most difficult to reach, since most botanical contact treatments act on the surface or on the exposed adult. Biological control and host-plant resistance complete the survey. Hymenopteran ectoparasitoids of bruchid larvae and pupae, attacking the immature stages through the seed coat, were investigated as augmentative agents and shown to reduce emergence under storage conditions, although their performance depends on temperature, host density and the depth of the grain mass. In parallel, screening of pulse germplasm identified accessions with resistance expressed as reduced oviposition, lower egg hatch, slower larval development, prolonged development time and lower adult emergence, traits linked to seed-coat thickness and hardness, to physical surface characters and to biochemical factors such as protease inhibitors and certain seed proteins. This resistance work pointed towards a durable, stage-targeting line of defence rooted in the seed itself and integrable with the physical and botanical methods documented above. Taken together, the surveyed literature thus spans the full arc from fundamental biology, through quantification of damage, to a diverse and expanding repertoire of control tactics, providing the raw material for the critical synthesis that follows.

Beyond single-tactic trials, a smaller but growing set of studies examined combinations of methods and the principles of integrated management for these pests. Investigations paired botanical seed coatings with hermetic storage, combined varietal resistance with admixed plant powders, or sequenced sanitation, drying and sealed storage across the post-harvest chain, generally reporting that integration gave more complete and more durable suppression than any component used alone. These studies also began to consider the practical dimensions that purely biological assays ignore, including the cost and availability of materials to smallholders, the acceptability of treated grain for consumption and sowing, and the ease of application in simple village stores. Although still

a minority of the literature, this integrative work is the most directly relevant to field practice and signals the direction in which research on *Callosobruchus* management has been moving.

3. METHODOLOGY OF THE REVIEW

This paper is a narrative review reinforced by a meta-analytic reading of the assembled evidence, and the methodology was designed to make the synthesis transparent and reproducible. Literature was gathered through systematic searching of bibliographic databases and indexing services covering agricultural entomology, stored-product protection and post-harvest science, supplemented by hand-searching the reference lists of key reviews and primary papers to capture older and regionally published work. Search terms combined the names of the two target species, *Callosobruchus maculatus* and *Callosobruchus chinensis*, together with their common synonyms and vernacular names, with terms denoting the storage context (stored pulses, grain legumes, post-harvest, granary), the biological focus (life cycle, developmental stages, oviposition, larval instar, pupa, emergence, infestation) and the intervention classes (fumigation, phosphine, irradiation, hermetic storage, controlled atmosphere, plant oil, botanical, essential oil, inert dust, parasitoid, host-plant resistance). Records were limited to peer-reviewed journal articles, authoritative reviews and standard reference works, and were screened first by title and abstract and then by full text. Selection followed explicit inclusion and exclusion criteria. Studies were included when they reported original or synthesised data on at least one developmental stage of one or both target species, or when they evaluated a defined treatment and reported a measurable outcome such as mortality, oviposition, egg hatch, adult emergence, development time, weight loss or germination. Studies were excluded when they concerned unrelated storage pests without bruchid data, when the species was not identified to the level needed for the present analysis, or when the reported outcomes could not be related to a developmental stage or a treatment dose. For each retained study the following information was extracted into a structured matrix: species and host pulse, the developmental stage examined or targeted, the treatment and its dose or exposure, the experimental conditions of temperature and humidity, the outcome measure, and the direction and approximate magnitude of effect. Because the primary studies differed widely in design, units and reporting, a formal pooled statistical meta-analysis was not attempted; instead the data were compared semi-quantitatively across stages and treatment classes to identify consistent patterns and sources of heterogeneity.

The analytical strategy therefore had two layers. The first was a descriptive synthesis organised by developmental stage and by treatment class, summarising what each line of work has established. The second was a critical and meta-analytic appraisal in which the consistency, magnitude and reliability of reported effects were weighed, the methodological quality of the studies was judged against criteria such as the use of controls, replication, identification of stages and reporting of exposure conditions, and recurrent gaps were identified. Particular attention was paid to whether studies distinguished among developmental stages when reporting efficacy, since this distinction proved to be the single most informative axis along which the literature varies.

This two-layer approach allowed the review to move from cataloguing past work to interpreting why treatments differ in their effect and where the evidence base remains weakest.

4. CRITICAL ANALYSIS OF PAST WORK

Read as a whole, the literature on *Callosobruchus* is rich in descriptive and applied detail but uneven in rigour, and a critical reading exposes several systematic limitations. The most consequential is the inconsistent treatment of developmental stage as an experimental variable. The biology of these beetles makes stage central: the egg is cemented to the seed and protected by its chorion; the larva and pupa are sealed inside the cotyledon, shielded from surface treatments; and only the adult is freely exposed. Yet a large fraction of the treatment studies report a single mortality figure for a mixed-age culture, or test only on adults, without resolving the differential susceptibility of eggs, instars and pupae. Where stage-specific data do exist, they consistently show the egg and the late pupa to be the most tolerant, which means that efficacy figures derived from adult bioassays systematically overstate field performance. This conflation undermines the comparability of studies and obscures the very mechanism that determines whether a treatment can break the life cycle. A second weakness is methodological heterogeneity that frustrates synthesis. Studies differ in the host pulse used, in storage temperature and humidity, in initial infestation density, in the duration of exposure, and in the outcome measured some reporting percentage mortality, others adult emergence, oviposition deterrence, weight loss or germination. Doses of botanicals are expressed variously as weight per weight of grain, as concentrations of extract, or as undefined coatings, and the active constituents of plant materials are seldom characterised, so that nominally identical treatments may differ greatly in potency. The absence of standardised protocols means that apparent disagreements among studies may reflect differences in method rather than in biological reality, and it prevents the formal pooling of effect sizes that a quantitative meta-analysis would require. Compounding this, sample sizes are often small, replication is sometimes minimal or unreported, and statistical treatment is frequently rudimentary, so that the precision of many reported effects cannot be judged. The two target species are also not always clearly distinguished, with results occasionally generalised from one to the other despite their behavioural and physiological differences.

Third, the durability and field relevance of many reported successes are inadequately established. A large number of botanical studies demonstrate strong short-term effects in small laboratory containers but do not test persistence over a realistic storage season, do not assess whether treated seed remains fit for consumption or sowing, and do not evaluate performance at the scale and grain depth of a farm store. Repellent and oviposition-deterrent effects, while real, may simply redistribute rather than eliminate infestation. Conversely, the fumigation literature, though methodologically stronger, increasingly confronts the problem of resistance: repeated documentation of phosphine tolerance in stored-product beetles signals that chemical control alone is not sustainable, and the withdrawal of older fumigants narrows the available options. Few studies integrate resistance status into their efficacy assessments, and fewer still report whether the strains tested were of known provenance, so that the same nominal treatment may perform very differently against susceptible and tolerant

populations without this being recognised. Fourth, there is a persistent gap between the laboratory and the granary. Hermetic and controlled-atmosphere methods enjoy strong and internally consistent support, and their mode of action oxygen depletion lethal to all stages addresses the stage problem directly, yet adoption studies and evaluations under farmer management are comparatively scarce relative to the volume of efficacy testing. Biological control through parasitoids is promising in principle but constrained by the difficulty of reaching deep grain masses and by sensitivity to storage conditions, and the literature rarely reports the economics or logistics of mass release. Host-plant resistance, arguably the most durable and stage-targeting approach, is well characterised at the level of resistant accessions and associated seed traits, but the translation of these traits into widely grown, agronomically acceptable cultivars is underrepresented in the applied literature.

Finally, the meta-analytic reading reveals that the heterogeneity in reported efficacy is largely explicable once these limitations are recognised. Variation across studies aligns systematically with the developmental stage targeted, the beetle species, the host pulse and the dose or exposure achieved; differences that at first appear contradictory dissolve into a coherent picture when efficacy is interpreted relative to the stage actually reached by the treatment. The clear lesson is that future work must report stage-resolved outcomes under standardised conditions, must test persistence and grain-scale performance, and must situate single tactics within integrated programmes rather than presenting them as stand-alone solutions. Only on such a foundation can the accumulated evidence be combined into the quantitative synthesis the field still lacks.

5. DISCUSSION

The synthesis carries a consistent and actionable message: the developmental stage is the organising principle for understanding both the damage caused by *Callosobruchus* and the success of treatments against it. Because the destructive feeding and a large part of the life cycle occur hidden within the seed, treatments that act only on the surface or on the exposed adult cannot, by themselves, break the cycle. This single fact explains much of the apparent inconsistency in the literature and points towards the methods most likely to deliver durable control those that reach every stage. Hermetic and modified-atmosphere storage achieves this through oxygen depletion, fumigation through penetrating gas, lethal heating through temperature, and irradiation through radiation dose, while most botanical and physical surface treatments leave the protected egg, late larva and pupa relatively untouched.

It follows that the strongest practical strategy is integration rather than reliance on any one tactic. A stage-aware programme might combine the sanitation and varietal resistance that limit initial carry-over from the field with hermetic or controlled-atmosphere storage that suppresses all stages during the storage season, reserving botanical seed treatments and parasitoids as supplementary measures for specific contexts and reserving fumigation for situations where its use can be managed to delay resistance. Such integration aligns with the established logic of integrated pest management and offers resilience against the failure of any single component. The emphasis on resistance management is reinforced by the documented spread of phosphine

tolerance, which makes the preservation of chemical efficacy a shared responsibility rather than an assumption. Designing such a programme requires knowledge of which stage each component reaches and of how the components interact, so that gaps in coverage are closed rather than duplicated; a combination that controls only exposed adults, however many tactics it stacks, will still fail if no element reaches the protected egg and pupa. For the smallholder context in which most pulse losses occur, the discussion also highlights the importance of cost, availability and ease of use. Hermetic bagging is attractive precisely because it is chemical-free, scalable and increasingly affordable, while locally sourced plant oils and powders retain value as low-cost first-line measures despite their limitations against concealed stages. The choice of method must therefore weigh not only biological efficacy but also the fitness of treated seed for consumption and sowing, the persistence of protection over the storage period, and the practicality of application at farm scale. Bringing these considerations together, the review argues that progress now depends less on discovering new individual treatments than on rigorously evaluating, standardising and integrating those already at hand, with the developmental biology of the pest kept firmly in view.

6. CONCLUSION

Callosobruchus maculatus and *Callosobruchus chinensis* remain among the most damaging pests of stored pulses, and the substantial literature reviewed here shows both how much is understood about their biology and how unevenly that understanding has been applied to their control. The decisive insight to emerge is that the concealed developmental stages egg, larva and pupa, sealed within the seed determine the outcome of any intervention, and that efficacy must be judged stage by stage rather than as an aggregate figure. Treatments that reach all stages, notably hermetic and controlled-atmosphere storage, lethal heating, penetrating fumigation and irradiation, offer the most reliable suppression, while surface-acting botanical and physical methods, valuable for their low cost and safety, are best deployed as components of an integrated programme rather than as complete solutions. Host-plant resistance and biological control add durable, stage-targeting dimensions that deserve greater investment and translation into practice. The critical and meta-analytic appraisal identifies the path forward clearly: future studies should report stage-resolved outcomes under standardised conditions, test persistence and performance at granary scale, incorporate resistance status, and frame single tactics within integrated, resistance-conscious management. Pursued in this way, the accumulated knowledge can be consolidated into the quantitative evidence base the field still requires, and translated into practical protection of a crop on which the food and seed security of millions depend. The central recommendation is therefore both simple and demanding: treat the developmental stage as the unit of analysis, and judge every method by whether it can reach the concealed life within the seed.

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