

Insect-derived Bioactive Peptides with Antioxidant Activity: Sources, Preparation, Mechanisms, and Future Perspectives

Kangwei Xie*

Wuhan Institute of Technology, Wuhan 430205, China

*Corresponding email: 22316010043@stu.wit.edu.cn

Abstract

Insect-derived bioactive peptides have attracted increasing attention as sustainable natural antioxidants. This review summarizes recent progress in antioxidant peptides obtained from edible insects, including mealworms, black soldier fly larvae, silkworm pupae, and crickets. Current studies mainly focus on enzymatic hydrolysis, simulated gastrointestinal digestion, peptide fractionation, liquid chromatography-tandem mass spectrometry (LC-MS/MS) identification, and antioxidant activity evaluation. These peptides may exert antioxidant effects through radical scavenging, metal ion chelation, enhancement of endogenous antioxidant enzymes, and regulation of oxidative stress-related pathways such as Keap1-nuclear factor erythroid 2-related factor 2 (Nrf2) and nuclear factor-kappa B (NF- κ B). However, many studies remain at the hydrolysate or peptide-fraction level, and evidence from *in vivo* validation, digestion stability, bioavailability, and structure-activity relationship analysis is still limited. Future research should emphasize sequence-level validation, multi-level biological evaluation, standardized preparation, and scalable production to support the application of insect-derived antioxidant peptides in functional foods and nutraceuticals.

Keywords

Insect-derived peptides, Antioxidant activity, Edible insects, Protein hydrolysates, Bioactive peptides

Introduction

Oxidative stress, resulting from the excessive accumulation of reactive oxygen species (ROS), is widely recognized as a key factor in lipid peroxidation, cellular damage, inflammation, and the development of various chronic diseases [1]. Maintaining the balance between ROS generation and antioxidant defense systems is therefore essential for cellular homeostasis. In recent years, increasing attention has been directed toward the identification of safe and effective natural antioxidants, particularly those derived from food sources, as alternatives to synthetic compounds.

Among these, bioactive peptides have emerged as a promising class of natural antioxidants. Owing to their relatively low molecular weight, high biological activity, and structural diversity, these peptides can interact efficiently with free radicals or metal ions and may also modulate endogenous antioxidant systems. Compared with conventional antioxidants such as polyphenols and vitamins, bioactive peptides exhibit advantages including higher stability under certain conditions and potential multifunctionality, which has stimulated

extensive research into their sources and mechanisms of action.

In this context, edible insects have gained increasing interest as sustainable and efficient protein resources. Insect proteins are typically characterized by high protein content and balanced amino acid composition, making them suitable precursors for the generation of bioactive peptides. Moreover, insects can be produced with relatively low environmental impact compared to traditional livestock, further enhancing their attractiveness as alternative protein sources. Recent studies have demonstrated that enzymatic hydrolysis or simulated gastrointestinal digestion of insect proteins can release peptide fractions with notable antioxidant properties, highlighting their potential for application in functional foods and nutraceuticals [2].

Despite the rapid growth of this field, current research remains uneven in depth and scope. Many studies focus primarily on crude protein hydrolysates or peptide fractions, with limited identification of individual active sequences. In addition, although *in vitro* antioxidant

assays are widely employed, fewer studies extend to cell-based models, in vivo validation, or detailed mechanistic analysis. A systematic review has further indicated that, among the numerous peptides identified from edible insects, only a small proportion has been validated beyond preliminary screening stages, suggesting that the understanding of structure-activity relationships and biological relevance is still incomplete [3].

Given these considerations, a concise synthesis of recent progress is necessary to clarify the current state of research and identify key directions for future work. This review therefore focuses on insect-derived bioactive peptides with antioxidant activity, summarizing their major sources, preparation and identification strategies, methods used for activity evaluation, as well as their structural characteristics and underlying mechanisms. The limitations of current studies and potential avenues for further development are also discussed.

Literature review

Insect sources of antioxidant bioactive peptides

The reported sources of insect-derived antioxidant peptides are not evenly distributed across edible insect species. Current studies are mainly concentrated on mealworms, black soldier fly larvae, silkworm pupae, and crickets, reflecting both their protein availability and their different research values. Systematic reviews indicate that edible insects can generate a broad range of bioactive peptides after hydrolysis or gastrointestinal digestion, but only a limited number of insect species have been studied in sufficient depth for antioxidant peptide discovery.

Mealworms, especially *Tenebrio molitor*, represent one of the most frequently investigated sources. Their value lies not only in their high protein content, but also in their suitability for digestion-based peptide release and bioaccessibility studies. Lee et al. showed that digests from *T. molitor* and *Zophobas atratus* proteins displayed high bioaccessibility and antioxidant activity, and several digestion-derived peptides showed strong binding affinity toward myeloperoxidase in molecular docking analysis [4]. This suggests that mealworm proteins are useful not merely as hydrolysis substrates, but also as a model for linking digestion, peptide release, and potential antioxidant targets.

Black soldier fly larvae have received increasing attention because they combine functional peptide

potential with waste bioconversion value. Early work by Zhu et al. showed that alcalase hydrolysis followed by ultrafiltration yielded a <3 kDa fraction with stronger 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), hydroxyl radical, and superoxide radical scavenging activities, and 17 peptide sequences were identified by LC-MS/MS [5]. More recent research has moved beyond activity screening. Meng et al. isolated Pep6 from black soldier fly larvae and demonstrated its cytoprotective effect in HepG2 cells, with molecular docking suggesting competitive binding to Keap1 and possible involvement of the Keap1-Nrf2 pathway [6]. Thus, black soldier fly larvae are particularly valuable for mechanism-oriented studies.

Silkworm pupae constitute another important source, largely because they are protein-rich by-products of the silk industry. Their advantage is the relatively clear progression from hydrolysates to identified peptides. Zhang et al. identified Phe-Lys-Gly-Pro-Ala-Cys-Ala (FKGPACA) and Ser-Val-Leu-Gly-Thr-Gly-Cys (SVL-GTGC) from silkworm pupae protein hydrolysates and further indicated that the Ala-Cys-Ala (ACA) and Thr-Gly-Cys (TGC) fragments may contribute substantially to ABTS radical scavenging activity [7]. Cermenon et al. further identified peptides including Ser-Trp-Phe-Val-Thr-Pro-Phe (SWFVTPF) and Asn-Asp-Val-Leu-Phe-Phe (NDVLFF), which could reduce ROS accumulation and enhance antioxidant responses in HepG2 cells [8]. These findings render silkworm pupae ideal for analyzing sequence characteristics, active peptide fragments and structural stability.

Cricket provides a different perspective. Their studies often emphasize protein fractionation and enzyme selection rather than total protein hydrolysis. Fashakin et al. separated cricket proteins into albumin, globulin, glutelin, and prolamin fractions, then identified antioxidant peptides from enzymatic hydrolysates, showing that protein fraction composition affects peptide release and activity [9]. Yu et al. also demonstrated that different proteases produced distinct structural and antioxidant properties in house cricket hydrolysates [10]. Therefore, crickets are useful for analyzing how substrate composition and processing strategy influence antioxidant peptide generation.

Overall, these insects represent different research

directions: mealworms are suitable for digestion and bioaccessibility studies, black soldier fly larvae for high-value peptide discovery and mechanism analysis, silkworm pupae for sequence identification and stability evaluation, and crickets for protein fractionation and enzyme-selection studies. This distribution shows that the source insect is not a neutral variable; it determines the protein substrate, hydrolysis behavior, peptide profile, and depth of antioxidant mechanism exploration.

Preparation of insect-derived antioxidant peptides

Preparation of insect-derived antioxidant peptides generally follows a sequence of protein enrichment, hydrolysis, and fractionation, but the efficiency of peptide release and the resulting activity are highly dependent on the choice of pretreatment and hydrolysis strategy rather than a fixed protocol. In most studies, raw insect materials are first subjected to drying, grinding, and defatting to reduce lipid interference and increase protein accessibility. Protein extraction is commonly achieved by alkaline solubilization followed by isoelectric precipitation, which concentrates the protein fraction and provides a more controllable substrate for subsequent hydrolysis. This step is not merely preparative; differences in extraction conditions can alter protein conformation and thus influence enzyme accessibility and peptide profiles.

Enzymatic hydrolysis remains the dominant approach for generating antioxidant peptides from insect proteins. Proteases such as Alcalase, Flavourzyme, trypsin, and papain are widely employed, either individually or in combination, to cleave proteins into peptides of varying sizes and sequences. The effectiveness of hydrolysis is not determined solely by the degree of hydrolysis but also by the specificity of the enzyme toward particular peptide bonds. For example, studies on black soldier fly larvae proteins have shown that Alcalase tends to produce a higher proportion of low-molecular-weight peptides with stronger radical scavenging activity compared with other enzymes. Similarly, in cricket protein systems, different proteases generate distinct structural and functional outcomes, with some enzymes favoring the formation of peptides that exhibit higher reducing power or free radical scavenging capacity. These findings indicate that enzyme selection governs not only yield but also the functional characteristics of the resulting peptides.

In addition to single-enzyme systems, combined or sequential hydrolysis has been explored to broaden the peptide spectrum. Dual-enzyme treatments can expose additional cleavage sites and generate peptides that are not accessible through single-enzyme hydrolysis. However, increased hydrolysis complexity does not always translate into improved antioxidant activity, suggesting that optimal peptide profiles depend on a balance between enzyme specificity and controlled hydrolysis conditions rather than maximal degradation. Simulated gastrointestinal digestion has emerged as an alternative or complementary approach to enzymatic hydrolysis. This method aims to mimic *in vivo* digestion using enzymes such as pepsin and pancreatin, thereby producing peptides that are more relevant to actual dietary exposure. Studies have demonstrated that digestion-derived fractions often show enhanced antioxidant activity and bioaccessibility compared with undigested proteins or simple hydrolysates. This is particularly important because it links peptide generation with potential physiological functionality, rather than relying solely on *in vitro* chemical assays.

Overall, current preparation strategies reveal two key patterns. First, enzymatic hydrolysis is highly flexible but requires careful optimization of enzyme type and reaction conditions to obtain peptides with desirable antioxidant properties. Second, digestion-based approaches provide a more physiologically relevant pathway for peptide release, although they are less controllable in terms of product composition. These complementary strategies collectively define the current methodological framework for producing insect-derived antioxidant peptides.

Separation and identification of antioxidant peptides

Following hydrolysis, insect protein digests contain complex mixtures of peptides, and the identification of active components requires stepwise fractionation combined with analytical characterization. In practice, separation is typically guided by activity, with each purification step coupled with antioxidant assays to track the most effective fractions rather than relying on compositional profiling alone. This strategy reflects a key limitation of hydrolysate-based studies: Without targeted purification, it is difficult to attribute activity to specific peptide sequences.

Ultrafiltration is commonly used as the first screening

step to reduce system complexity. Fractions below 3 kDa are frequently enriched in antioxidant activity, as observed in black soldier fly larvae hydrolysates, where the <3 kDa fraction exhibited stronger radical scavenging capacity than higher-molecular-weight fractions. However, the relationship between molecular weight and activity is not strictly linear. In mealworm-derived peptides, different molecular weight fractions may exhibit optimal activity depending on the assay system, indicating that size alone cannot fully explain antioxidant performance. Therefore, ultrafiltration is more appropriately regarded as a coarse selection step rather than a definitive criterion for activity.

Further purification is usually achieved through chromatographic techniques, including gel filtration and reversed-phase high-performance liquid chromatography (RP-HPLC). Gel filtration separates peptides based on size distribution, while RP-HPLC provides higher resolution based on hydrophobicity, enabling the isolation of relatively pure peptide fractions. This combination has been widely applied in silkworm pupae studies, where sequential ultrafiltration, gel chromatography, and RP-HPLC allowed the isolation of fractions with progressively enhanced antioxidant activity. Similar multi-step purification strategies have also been employed in cricket protein systems, where prior fractionation of protein classes followed by enzymatic hydrolysis and chromatographic separation improved the efficiency of identifying active peptides.

Identification of peptide sequences is primarily conducted using liquid chromatography-tandem mass spectrometry (LC-MS/MS), which enables high-throughput characterization of peptide profiles. However, LC-MS/MS alone does not establish functional relevance. Consequently, many studies incorporate additional validation steps, including chemical synthesis of candidate peptides and re-evaluation of antioxidant activity. For example, specific peptides identified from silkworm pupae hydrolysates were synthesized and further truncated to determine active fragments, revealing that short motifs containing residues such as cysteine and alanine contributed significantly to antioxidant activity. This approach provides more direct evidence linking sequence features to function.

In recent work, bioinformatics tools have been increasingly integrated into the identification process.

Databases and predictive algorithms are used to screen peptides for potential antioxidant activity prior to experimental validation, reducing the number of candidates for synthesis. Molecular docking has also been applied to explore interactions between peptides and oxidative stress-related targets, as demonstrated in mealworm-derived peptide studies where selected sequences showed favorable binding to myeloperoxidase. While these computational methods enhance screening efficiency, their predictive nature requires experimental confirmation.

Overall, the separation and identification of insect-derived antioxidant peptides have progressed from simple fractionation to integrated workflows combining size-based screening, chromatographic purification, mass spectrometry, and functional validation. Nevertheless, the transition from complex hydrolysates to well-defined active sequences remains incomplete in many studies, and the identification of peptides with confirmed biological relevance continues to be a central challenge in this field.

Antioxidant activity and evaluation methods

Evaluation of antioxidant activity for insect-derived peptides follows a tiered framework that progresses from chemical assays to cellular systems and, less frequently, to digestion-based and in vivo validation. The choice of method strongly influences the interpretation of activity, and discrepancies across studies often arise from differences in assay principles rather than intrinsic peptide properties.

In vitro chemical assays remain the most widely applied tools because of their simplicity and comparability. Radical scavenging assays such as DPPH and ABTS are commonly used to assess the electron, or hydrogen-donating ability of peptide fractions, while hydroxyl and superoxide radical assays provide additional information on reactivity toward highly oxidative species. Reducing power and ferric reducing antioxidant power (FRAP) assays evaluate electron transfer capacity, and metal chelation assays examine the ability of peptides to bind pro-oxidant ions. Studies on various insect hydrolysates consistently show that low-molecular-weight fractions exhibit strong activity in these assays. However, the relative performance of peptide fractions often varies depending on the assay type, indicating that different mechanisms contribute to antioxidant behavior.

Therefore, relying on a single chemical assay may lead to incomplete or biased conclusions.

To address this limitation, cell-based models have been increasingly incorporated to provide biologically relevant evaluation. HepG2 hepatocytes and RAW264.7 macrophages are among the most commonly used systems, where oxidative stress is typically induced by agents such as H₂O₂ or lipopolysaccharide. In these models, antioxidant activity is assessed through intracellular ROS levels, lipid peroxidation markers such as malondialdehyde, and the activity or expression of antioxidant enzymes including superoxide dismutase and catalase. For example, peptides derived from black soldier fly larvae have been shown to reduce ROS accumulation and improve cell viability under oxidative stress conditions, indicating that their activity extends beyond direct radical scavenging to cellular protection. Similarly, silkworm pupae-derived peptides have been reported to enhance endogenous antioxidant defenses in HepG2 cells, supporting their potential physiological relevance.

Simulated gastrointestinal digestion has also become an important component of activity evaluation, particularly in studies aiming to assess dietary functionality. Digestion models using pepsin and pancreatin generate peptide mixtures that more closely resemble those available for absorption *in vivo*. In mealworm protein systems, digestion-derived fractions exhibit higher bioaccessibility and retain significant antioxidant activity, suggesting that peptide release during digestion plays a critical role in determining functional efficacy. This approach bridges the gap between *in vitro* chemical assays and potential *in vivo* effects.

Despite these advances, *in vivo* validation remains limited. Only a small number of studies extend to animal models, and even fewer provide detailed pharmacokinetic or bioavailability data. As highlighted in systematic analyses, the majority of identified insect-derived peptides have not been evaluated beyond preliminary screening, which restricts the interpretation of their physiological significance. Consequently, current evaluation strategies are effective for screening and comparative analysis but are insufficient for establishing comprehensive structure-function relationships or confirming health benefits.

Overall, antioxidant activity assessment in this field has

evolved toward a multi-level framework combining chemical, cellular, and digestion-based approaches. However, further integration of *in vivo* models and standardized evaluation protocols is required to improve the reliability and comparability of results.

Structural features and antioxidant mechanisms

The antioxidant performance of insect-derived peptides is governed by an interplay between physicochemical features and multiple reaction pathways rather than a single determinant. Across species and processing routes, four structural aspects recur as primary drivers-molecular size, amino acid composition, sequence motifs, and conformational properties - while the mechanisms of action span direct radical quenching, metal chelation, and regulation of cellular redox systems.

(1) Molecular size and accessibility

Peptides in the low-molecular-weight range (often <3 kDa) are frequently enriched in antioxidant activity because of higher solubility, diffusivity, and accessibility to reactive sites. In black soldier fly systems, <3 kDa fractions consistently show stronger DPPH/ABTS and ROS-scavenging capacity after enzymatic hydrolysis and ultrafiltration. However, size-activity relationships are not strictly monotonic. Mealworm-derived fractions can display assay-dependent optima, where intermediate fractions outperform the smallest peptides in certain endpoints, indicating that steric compatibility with targets and sequence chemistry modulate activity beyond size alone. Thus, molecular weight should be viewed as a facilitating factor rather than a sufficient predictor.

(2) Amino acid composition and redox chemistry

Enrichment in hydrophobic (Leu, Val, Ile), aromatic (Phe, Tyr, Trp), and sulfur-containing residues (Cys, Met) is a recurrent feature of potent antioxidant peptides. Aromatic side chains can donate electrons or hydrogen atoms to stabilize radicals, while sulfur-containing residues participate in redox cycling and nucleophilic reactions. Evidence from black soldier fly hydrolysates links higher proportions of hydrophobic and aromatic residues to enhanced radical scavenging across multiple assays. Complementary findings in cricket hydrolysates show that protease selection alters residue composition and, consequently, reducing power and radical quenching, reinforcing that composition is a controllable determinant via processing. Importantly, composition effects are context-dependent: peptides rich in basic

residues may favor metal binding, whereas those enriched in hydrophobic residues tend to interact with lipid radicals.

(3) Sequence features and active motifs

Beyond bulk composition, short sequence motifs critically determine activity. Work on silkworm pupae identified peptides such as FKGPAACA and SVLGTGC, and subsequent truncation experiments revealed that short fragments (e.g., ACA, TGC) retain or even concentrate activity, implicating specific residues and local environments as active sites. This motif-centric view explains why extensive hydrolysis can either enhance activity by exposing motifs or diminish it by over-fragmentation. In practice, activity-guided purification followed by LC-MS/MS and synthesis is necessary to resolve such sequence-function links. Recent identification efforts in *Tenebrio molitor* have yielded multifunctional peptides with concurrent antioxidant, ACE-inhibitory, and antidiabetic potentials, suggesting that certain sequences can engage multiple targets through shared physicochemical features [11].

(4) Conformation and stability

Peptide conformation in solutions and under processing conditions (heat, pH, digestion) influences both reactivity and bioavailability. Silkworm-derived peptides maintain substantial activity after thermal treatment and exhibit differential stability upon pepsin/pancreatin digestion, indicating that sequence context governs resistance to proteolysis and preservation of function. From an application standpoint, stability during digestion is as critical as intrinsic activity, because bioaccessible peptides are more likely to exert effects in vivo.

(5) Direct radical scavenging and chain-breaking effects
At the mechanistic level, many insect peptides act as direct scavengers, donating electrons or hydrogen atoms to neutralize DPPH, ABTS, hydroxyl, or superoxide radicals. Enzymatic hydrolysis of cricket proteins increases total antioxidant capacity and ferric reducing power, consistent with enhanced electron-transfer capability of the resulting peptides [12]. The diversity of assay responses across fractions underscores that different sequences preferentially engage distinct radical species, necessitating multi-assay evaluation to capture the breadth of activity.

(6) Metal ion chelation and inhibition of lipid peroxidation

Peptides containing histidine, cysteine, or acidic residues can chelate transition metals (e.g., Fe²⁺, Cu²⁺), thereby suppressing Fenton-type reactions and downstream lipid peroxidation. Although often assessed indirectly via chelation assays, this pathway is particularly relevant in complex matrices where metal-catalyzed oxidation dominates. Composition-function links (basic vs. acidic residues, presence of imidazole or thiol groups) provide a rationale for tailoring hydrolysis conditions to enrich metal-binding motifs.

(7) Cellular antioxidant responses and signaling pathways

Increasingly, studies extend beyond chemical assays to cellular mechanisms. In hepatocyte and macrophage models, insect-derived peptides reduce intracellular ROS, improve viability under oxidative stress, and modulate endogenous defense systems. A notable advance is the demonstration that selected peptides from black soldier fly larvae exert cytoprotective effects in HepG2 cells and are predicted to interact with Keap1, suggesting activation of the Nrf2 pathway and upregulation of antioxidant enzymes. Complementary evidence from silkworm-derived peptides shows decreased ROS levels and enhanced antioxidant enzyme responses in HepG2 cells, linking sequence features to cellular outcomes. Beyond Nrf2, peptides from “*Antheraea assamensis*” hydrolysates attenuate oxidative stress and inflammation by suppressing toll-like receptor 4 (TLR4)/nuclear factor-kappa B (NF-κB) signaling in macrophages, indicating crosstalk between antioxidant and anti-inflammatory pathways [13]. Such findings expand the mechanistic scope from direct radical quenching to regulation of redox-sensitive signaling networks.

(8) Integration across levels and remaining gaps

Collectively, these observations support a multi-mechanistic model: Structural features determine physicochemical reactivity (radical scavenging, chelation), which in turn influences cellular redox balance and signaling responses. However, the field still faces two constraints. First, many studies do not progress from fraction-level activity to sequence-resolved validation, limiting precise structure-activity relationships. Second, links between in vitro activity, digestion stability, and in vivo efficacy remain underdeveloped, despite emerging evidence that digestion alters both peptide composition and function.

In summary, insect-derived antioxidant peptides operate through complementary mechanisms shaped by size, composition, and sequence motifs. Advances in purification, synthesis, and cellular assays have begun to clarify these relationships, but systematic mapping of sequence features to specific mechanisms, alongside validation under physiologically relevant conditions, remains a priority for translating these peptides into functional applications.

Current limitations and future prospects

Despite the growing body of research, several limitations constrain both mechanistic interpretation and practical translation of insect-derived antioxidant peptides. A central issue is that many studies still focus on crude hydrolysates or broad molecular-weight fractions, with limited progression to sequence-level identification and functional validation. Systematic analyses indicate that, although numerous peptides have been reported from edible insects, only a small proportion has been verified as active after purification and synthesis, which restricts the development of robust structure-activity relationships. Even when peptides are identified, their contribution to the overall activity of complex mixtures is rarely quantified.

A second limitation lies in the gap between *in vitro* activity and physiological relevance. Most studies rely on chemical assays that evaluate electron transfer or radical scavenging under simplified conditions. While these assays are useful for screening, they do not necessarily reflect biological effectiveness. Evidence from digestion-based studies shows that peptide profiles and antioxidant performance can change substantially after simulated gastrointestinal processing, suggesting that bioaccessibility is a critical determinant of functionality. Multi-level assessments further demonstrate that digestion not only releases new peptide fractions but also alters their biological activity, indicating that results obtained from undigested hydrolysates may overestimate *in vivo* effects. Another challenge concerns methodological variability. Differences in insect species, pretreatment procedures, enzyme selection, hydrolysis conditions, and purification strategies result in heterogeneous peptide profiles and inconsistent activity outcomes. Studies on cricket proteins, for example, have shown that enzyme choice significantly affects both physicochemical properties and antioxidant capacity, highlighting the sensitivity of

peptide generation to processing parameters. Similarly, variations in protein fractionation strategies can influence the types of peptides released and their subsequent activity, as demonstrated in cricket protein fraction studies. Such variability complicates cross-study comparison and limits the establishment of standardized preparation protocols.

From a mechanistic perspective, current understanding remains incomplete. Although direct radical scavenging and metal chelation are widely recognized, fewer studies provide detailed insight into cellular pathways or target interactions. Some investigations have begun to address this gap by linking insect-derived peptides to signaling pathways involved in oxidative stress and inflammation. For instance, peptides derived from “*Antheraea assamensis*” hydrolysates have been shown to suppress TLR4/NF- κ B signaling and reduce pro-inflammatory mediator production in macrophages, suggesting that antioxidant effects may be closely associated with anti-inflammatory mechanisms. However, such studies remain limited, and comprehensive pathway-level analyses are still lacking.

Application-related challenges must also be considered. Although edible insects are generally regarded as sustainable protein sources, issues such as allergenicity, variability in raw materials, and potential contaminants need to be addressed for large-scale use. Reviews of insect proteins emphasize that allergenic potential and safety evaluation remain important considerations, particularly when developing functional food ingredients or nutraceuticals. In addition, current preparation processes often involve multi-step procedures that may not be economically feasible on an industrial scale. Future research should therefore move toward more integrated and standardized approaches. Priority should be given to identifying and validating specific peptide sequences with confirmed biological activity, including the use of synthetic peptides and systematic structure-activity studies. Greater emphasis should also be placed on linking *in vitro* results with digestion, absorption, and *in vivo* effects to improve physiological relevance. Advances in bioinformatics and computational prediction offer opportunities to streamline peptide discovery, but these tools must be combined with rigorous experimental validation. Finally, optimization of processing conditions and simplification of production

workflows will be essential for translating laboratory findings into practical applications.

In summary, while insect-derived antioxidant peptides have demonstrated considerable potential, current research remains fragmented across different methodological levels. Addressing limitations in peptide identification, biological validation, and process standardization will be critical for advancing both fundamental understanding and practical utilization.

Conclusion

Insect-derived bioactive peptides have emerged as promising natural antioxidants due to their favorable physicochemical properties and diverse mechanisms of action. Current research demonstrates that peptides obtained from mealworms, black soldier fly larvae, silkworm pupae, and crickets can exhibit significant radical scavenging activity, metal chelation capacity, and modulation of cellular antioxidant systems. Advances in enzymatic hydrolysis, peptide purification, and sequence identification have enabled the discovery of specific peptides with enhanced activity, while recent studies have begun to elucidate their roles in regulating oxidative stress-related signaling pathways.

However, most studies remain focused on hydrolysates or peptide fractions, and only a limited number of peptides have been fully characterized and validated under physiological conditions. In addition, the relationship between in vitro antioxidant activity and in vivo efficacy remains unclear, particularly with respect to digestion stability and bioavailability. Future work should prioritize the identification of functional peptide sequences, the integration of multi-level evaluation approaches, and the development of standardized and scalable production methods.

Overall, further efforts to bridge the gap between laboratory findings and practical applications will be essential for the effective utilization of insect-derived antioxidant peptides in functional foods and related fields.

Funding

This work was not supported by any funds.

Acknowledgements

The author would like to show sincere thanks to those techniques who have contributed to this research.

Conflicts of Interest

The author declares no conflict of interest.

References

- [1] Ma, Z., Mondor, M., Valencia, F. G., Hernández-Álvarez, A. J. (2023) Current state of insect proteins: extraction technologies, bioactive peptides and allergenicity of edible insect proteins. *Food & Function*, 14(18), 8129-8156.
- [2] Quah, Y., Tong, S. R., Bojarska, J., Giller, K., Tan, S. A., Ziora, Z. M., Chai, T. T. (2023) Bioactive peptide discovery from edible insects for potential applications in human health and agriculture. *Molecules*, 28(3), 1233.
- [3] Teixeira, C. S., Villa, C., Costa, J., Ferreira, I. M., Mafra, I. (2023) Edible insects as a novel source of bioactive peptides: a systematic review. *Foods*, 12(10), 2026.
- [4] Lee, J. H., Kim, Y. J., Kang, M. C., Park, M. K., Choi, Y. S. (2025) Bioaccessibility and antioxidant activity of edible-insect protein digests from *Tenebrio molitor* and *Zophobas atratus* in an in vitro digestion model. *Food Chemistry: X*, 103251.
- [5] Zhu, D., Huang, X., Tu, F., Wang, C., Yang, F. (2020) Preparation, antioxidant activity evaluation, and identification of antioxidant peptide from black soldier fly (*Hermetia illucens* L.) larvae. *Journal of Food Biochemistry*, 44(5), e13186.
- [6] Meng, L., Yu, D., Lin, J., Hu, Y., Peng, N., Zhao, S. (2025) Structural characterization, HepG2 cell cytoprotective ability, and antioxidant mechanism of novel antioxidant peptides identified from black soldier fly larvae (*Hermetia illucens* L.). *Food Chemistry*, 463, 141462.
- [7] Zhang, Y., Wang, J., Zhu, Z., Li, X., Sun, S., Wang, W., Sadiq, F. A. (2021) Identification and characterization of two novel antioxidant peptides from silkworm pupae protein hydrolysates. *European Food Research and Technology*, 247(2), 343-352.
- [8] Cermeno, M., Bascón, C., Amigo-Benavent, M., Felix, M., FitzGerald, R. J. (2022) Identification of peptides from edible silkworm pupae (*Bombyx mori*) protein hydrolysates with antioxidant activity. *Journal of Functional Foods*, 92, 105052.
- [9] Fashakin, O. O., Tangjaidee, P., Unban, K., Klangpetch, W., Khumsap, T., Sringarm, K., Phongthai, S. (2023) Isolation and identification of antioxidant peptides derived from cricket (*Gryllus*

- bimaculatus) protein fractions. *Insects*, 14(8), 674.
- [10] Yu, T., Yuan, B., Huang, J., Wang, R., Huang, G., Huang, D., Xiao, J. (2025) Characterisation of the physicochemical, functional and antioxidant properties of house cricket (*Acheta domesticus*) protein hydrolysate processed with six proteases. *LWT - Food Science and Technology*, 227, 118007.
- [11] de Matos, F. M., de Castro, R. J. S. (2025) Characterization and identification of potential antioxidant, antidiabetic, and antihypertensive peptides from hydrolysates of *Tenebrio molitor* flour and its protein concentrate. *Journal of Food Science*, 90(10), e70595.
- [12] de Matos, F. M., Novelli, P. K., de Castro, R. J. S. (2021) Enzymatic hydrolysis of black cricket (*Gryllus assimilis*) proteins positively affects their antioxidant properties. *Journal of Food Science*, 86(2), 571-578.
- [13] Sahu, R. K., Borthakur, S., Saikia, M., Sarkar, S., Ahmed, R. S., Dasgupta, D., Kalita, J. (2023) A common edible insect (*Antheraea assamensis*) protein hydrolysate regulates LPS-induced oxidative stress and inflammation by modulating the TLR4/NF- κ β Signaling Pathway. *Journal of Insects as Food and Feed*, 10(7), 1151-1170.