



REVIEW

Application of Edible Insects as Novel Protein Sources and Strategies for Improving Their Processing

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Abstract Insects have long been consumed by humans as a supplemental protein source, and interest in entomophagy has rapidly increased in recent years as a potential sustainable resource in the face of environmental challenges and global food shortages. However, food neophobia inhibits the widespread consumption of edible insects, despite their high nutritional and functional value. The own characteristics of edible insect protein such as foaming properties, emulsifying properties, gelling properties and essential amino acid ratio can be improved by drying, defatting, and extraction. Although nutritional value of some protein-enriched bread, pasta, and meat products, especially essential amino acid components was increased, replacement of conventional food with edible insects as a novel food source has been hindered owing to the poor cross-linking properties of edible insect protein. This deterioration in physicochemical properties may further limit the applicability of edible insects as food. Therefore, strategies must be developed to improve the quality of edible insect enriched food with physical, chemical, and biological methods. It was presented that an overview of the recent advancements in these approaches and highlight the challenges and prospects for this field. Applying these strategies to develop insect food in a more familiar form can help to make insect-enriched foods more appealing to consumers, facilitating their widespread consumption as a sustainable and nutritious protein source.

Keywords edible insects, food resources, entomophagy, protein cross-linking

Introduction

Various peptides are linked to form protein structures; when digested in the body, the derived amino acids and peptides of proteins are used to support various physiological activities and survival (van Huis et al., 2013). Dietary proteins contain 21 different amino acids, including non-essential and essential amino acids (EAAs), and insufficient EAA consumption could disrupt the balance of the body. Thus, protein

intake is essential, but obtaining sufficient dietary protein from conventional food sources could spend too many resources such as soil, water, and energy, and their production and/or harvesting are often harmful to the environment (Kim et al., 2022b; Post, 2012). Moreover, with a limited scope for increasing the production of conventional food sources, the increasing global demand for food is difficult to meet (Kim et al., 2019b). Therefore, investigations into identifying various alternatives to conventional food sources have increased, and several solutions have been suggested for the utilization of more efficient food production systems (van Huis et al., 2013). To this end, several studies have evaluated the replacement of conventional food sources for obtaining sufficient proteins, including vegetable proteins, *in vitro* meat, and edible insects as emerging novel protein sources (Post, 2012; van Huis et al., 2013). Similarity in taste and texture to conventional foods is a major deciding factor that influences consumer perception; thus, among potential alternative food sources, edible insects diverge the most from the conventional appearance of food (Kim et al., 2019b). Although numerous people worldwide regularly consume edible insects without processing, many have negative perceptions of insects in general, and therefore cannot be easily convinced to include them as part of their regular diets. Therefore, the promotion and utilization of edible insects in food systems must be improved.

To effectively incorporate insect proteins into food systems as high value-added functional ingredients, these proteins should be thoroughly assessed to best exploit edible insects as a dietary protein alternative. Previous reviews have highlighted the nutritional value, types, pre-treatment technologies, and health benefits of insects available for human consumption (Kim et al., 2019b; Liceaga et al., 2022; Mintah et al., 2020a; Mutungi et al., 2019; van Huis et al., 2021). Although application of edible insects on conventional food has been increased, these previous studies have not discussed advance and limitation for the use of edible insects as a food ingredient and improvement strategy. Therefore, in this review, we focused on the research progress to date related to the detailed usage, nutritional information, value, limitations, processing, and application of edible insects and suggested strategies for improving edible insect protein characteristics.

Why People Have a Different Attitude toward Insects as Food by Regions and How to Regulate Edible Insects Legally

Traditionally, insects were important food resources for early human societies because the hunting of insects was relatively easier than that of other animals (Valadez, 2003). People in various countries and from different cultures continue to routinely consume edible insects; insects on the ground are used as food resources for over 80% (Kim et al., 2019b). The consumption of edible insects is higher in human populations living in sub-torrid and torrid zones than in those living in temperate zones (van Huis et al., 2013). These geographic differences in the propensity to consume edible insects as food might be attributed to the differences in the respiratory system of insects. The diffusion of gases could be influenced by the environmental conditions of specific regions, consequently affecting the growth and size of the available insects (Kirkpatrick, 1957). Seasonal changes in the temperate zone are responsible for the small size and complex assemblages of insects throughout the year, whereas the consistently high temperature of tropical regions accelerates the diffusion rate of oxygen, and a stable climate might promote the presence of active insects throughout the year (van Huis et al., 2013).

To date, in most regions, insects have been consumed as a whole by frying, broiling, boiling, steaming, grilling, drying, or other traditional cooking methods. Although some studies have shown a reduction in crude protein, ash, and zinc contents after cooking, these cooking processes may generally enhance the safety, flavor, shelf life, and digestibility of insects (van Huis et al., 2013; Williams et al., 2016). A neophobic response toward insects as food has largely inhibited the broader

consumption of edible insects, which is derived from an unfamiliar bias and prejudice that insects are dirty and unclean and that they would have negative effects on health if consumed (Kim et al., 2019b). However, constant marketing and education regarding the nutritional value and consumption benefits of edible insects can gradually affect the willingness of people to consume them (Kim et al., 2019b).

Regardless of culture or geography, interest in edible insects has been increasing in recent years, especially in western societies, and legislation about the use of insects as food and feed has been established worldwide (Liceaga et al., 2022). The European Union (EU) submitted regulation No 2015/2283 related to edible insects. *Acheta domesticus*, *Gryllobates sigillatus*, *Alphitobius diaperinus*, *Hermetia illucens*, *Apis mellifera*, *Locusta migratoria*, and *Tenebrio molitor* have been permitted as food for human consumption as of October 2020, and insect food products must pass a safety assessment by the European Food Safety Authority. Moreover, various regulations about the rearing, feeding, slaughtering, and hygiene processing of edible insects have been updated (Lähteenmäki-Uutela et al., 2021). The United States Food, Drug, and Cosmetic Act (United States Code, Title 21) has included the consumption of insects as food and feed since 2013 with no specific set of standards for edible insects beyond following the general Food and Drug Administration standard (i.e., bacterial testing and manufacturing practice certification). Other countries, including Canada, Australia, China, Japan, Thailand, and South Korea, have also updated their regulations related to edible insects in recent years (Lähteenmäki-Uutela et al., 2021).

Useful Information for Promoting Edible Insects as Novel Protein Sources

The critical limitation of conventional food supply systems should be mentioned when promoting edible insects to the population as novel protein sources. The global population has been rapidly increasing and is expected to reach 9.7 billion in 2050. However, existing conventional food systems cannot provide a sufficient supply to the global population, as such systems incur high environmental costs such as land, water, and air pollution, and loss of biodiversity (Berners-Lee et al., 2018; Post, 2012). Therefore, an alternative food supply system must be developed for the present and future generations. Among various alternatives, the consumption of edible insects as food has been emphasized owing to several advantages for the environment and human health (van Huis, 2013). However, people in developed countries generally do not consider insects as food sources because of their negative perception of insects, and the various negative idioms related to insects, spiders, and other invertebrates can inhibit the consumption of edible insects (Meyer-Rochow and Kejonen, 2020). Thus, reframing to a positive expression of idioms can change this negative perception (Mishyna et al., 2020).

Edible insects should be exploited in an effective and positive manner according to precise information derived from experimental data. To encourage edible insect consumption to the public, the following themes are typically included when providing information: Population increase, environmental impact, and nutritional value (Kim et al., 2019b). Despite these valid reasons, insects are primarily considered to be disease carriers by most consumers (Castro and Chambers, 2019). However, insect-specific pathogenic microorganisms are considered harmless to humans; only contamination with human pathogenic and toxin-forming microorganisms during unhygienic rearing, processing, and transport can critically compromise the safety of edible insects (Schlüter et al., 2017). Therefore, microbiological criteria should be established according to the standards for general food by each government. In terms of environmental cost, edible insects are a highly efficient food source compared to conventional animal food. Ooninx and de Boer (2012) investigated the environmental impact of the mealworm *T. molitor* over their lifecycle. They chose three main factors to determine differences in the environmental impact of food: Enteric CH₄ production, reproduction rate, and feed conversion efficiency. Mealworms exhibited a lower impact than

animal sources (pork, chicken, and beef) with advantages of no CH₄ emission, high reproduction (one female insect can produce 160 eggs), a short lifecycle, and low feed conversion ratio (mealworm=2.2, chicken=2.3, pigs=4.0, and beef cattle =2.7–8.8). In addition to mealworms, other species such as black soldier fly larvae, crickets, and grasshoppers have taken the center stage as alternative protein sources with a low environmental impact (Bosch et al., 2019; Halloran et al., 2017; Wegier et al., 2018). Edible insects also have high nutritional value with high levels of EAAs, unsaturated fatty acids, polysaccharides, and micronutrients (Kim et al., 2019b).

Providing this positive information can also help increase the willingness of consumers to pay for edible insects. Consumers generally have a positive attitude toward tasting insects when they are first provided with brief information regarding the various advantages of edible insects for the environment and human health (Barsics et al., 2017). Placentino et al. (2021) served insect-based protein bars to professional athletes and found that providing brief information regarding the environmental and nutritional benefits of edible insects enhanced the willingness of the athletes to taste the bar. The authors further expected that the consumption of edible insects by professional athletes would promote broader consumer engagement. The sensory experience and appearance of edible insects can also be important factors in improving the taste of edible insects (Mishyna et al., 2020). As an unfamiliar appearance can reduce the willingness to taste edible insects, processing to modify the appearance has been attempted by the food industry (Castro and Chambers, 2019). Processed edible insects have been used as food resources after drying, defatting, and extraction.

Processing Methods for Obtaining High-Quality Protein from Edible Insects

Conventional proteins derived from slaughtered animals can be categorized by their sources such as meat, blood, organs, or skin; the physicochemical properties of the proteins clearly differ across these categories, which determine their applicability in various purposes. However, such categorization of edible insects is more difficult because of their small size. Therefore, fractionation is the most important factor in insect protein processing because of the difficulty in separating the skeleton and protein parts of the insect body (Purschke et al., 2018a). In this section, we provide an overview of the key processes that should be considered for enhancing the physicochemical properties of proteins from edible insects.

Drying methods

Drying is a useful and traditional method for increasing the efficiency of food systems by reducing the weight and increasing the shelf life of the product. However, drying processes that reduce the moisture content, inhibit microbial growth, and alter the enzymatic activity and chemical reactions, can ultimately change the quality of food (Mujumdar and Jangam, 2011). A summary of the key drying methods of protein enhancement in edible insects is provided in Table 1.

More efficient and hygienic drying methods have been developed in recent years, which have applicability in the food industry for obtaining high-quality edible insect powder or extracted protein. Drying can be performed using a heat pump at a temperature lower than that used with a conventional hot-air oven because of the recovered heat cycle. For improving the performance of heat pump drying, the development systems have been divided into four groups: Heat pump system, external heating mode, drying gas, and operating mode (Mujumdar and Jangam, 2011). The energy used for drying can be reduced with a combination of two or more technologies (Hnin et al., 2018). Owing to its simplicity and hygiene, solar-assisted drying could be a useful method in this regard, which utilizes an auxiliary air heater, forced convection, and chemical heat pump (Hnin et al., 2018).

Table 1. Edible insect-drying methods following harvesting and their advantages and disadvantages

Methods	Advantages	Disadvantages
Drying process		
Sun-drying	<ul style="list-style-type: none"> - Economical - Easy to dry 	<ul style="list-style-type: none"> - High temperature differential - Rewetting - Contamination by animals, wind, or dust - Long drying period
Freeze-drying (lyophilization)	<ul style="list-style-type: none"> - High nutritional value - Conservation of protein quality 	<ul style="list-style-type: none"> - High cost - Lipid oxidation
Oven-drying	<ul style="list-style-type: none"> - Short drying period - High accuracy 	<ul style="list-style-type: none"> - Protein deterioration
Heat pump drying	<ul style="list-style-type: none"> - High energy efficiency in cost - Lower protein deterioration than that with oven-drying 	<ul style="list-style-type: none"> - High cost and difficult to install - Low efficiency under cold conditions
Solar-assisted drying	<ul style="list-style-type: none"> - More hygienic than sun-drying - Improved drying efficiency when combined with other methods - Easy to dry - Saving energy 	<ul style="list-style-type: none"> - Microbial hazard - Additional cost - Relatively humidity under 60% - Climate conditions affect drying efficiency
Infrared radiation	<ul style="list-style-type: none"> - Uniform heating - High heat transfer rate - High cost-efficiency 	<ul style="list-style-type: none"> - Distance control
Microwave	<ul style="list-style-type: none"> - Rapid drying speed - Easy to control drying conditions 	<ul style="list-style-type: none"> - Non-uniform drying - Low penetration depth - Damage in texture
Modified atmosphere drying	<ul style="list-style-type: none"> - Browning reaction or any oxidation could be prevented 	<ul style="list-style-type: none"> - Requirement of the replacement of oxygen with nitrogen or carbon dioxide
Superheated steam	<ul style="list-style-type: none"> - Low explosion hazard - Protection from oxidation - Microbial safety - Constant drying rate 	<ul style="list-style-type: none"> - High operating pressure needed for high drying rate - Low drying rate with large equipment
Impinging steam	<ul style="list-style-type: none"> - High drying rate in thin products - Uniform drying 	<ul style="list-style-type: none"> - Less uniform texture
Contact sorption	<ul style="list-style-type: none"> - Easy way to assist 	<ul style="list-style-type: none"> - Food safety of absorbents - Requirement of another drying process to dry absorbents
Pulsed electric field	<ul style="list-style-type: none"> - Microbial safety - Prevent nutrients - Short treatment time 	<ul style="list-style-type: none"> - High initial cost - No effects on spores or enzymes - Only suitable for liquid - Low energy efficiency
Osmotic dehydration	<ul style="list-style-type: none"> - Improvement of quality and prevention of quality deterioration - Low energy cost - Flavor retention - Reduces enzymatic browning reaction 	<ul style="list-style-type: none"> - Increase in saltiness or sweetness - Decrease in acidity
Fluid bed	<ul style="list-style-type: none"> - High drying rate - High thermal efficiency - Low maintenance cost - Reduced contact time - Easy to control 	<ul style="list-style-type: none"> - High energy consumption - Non-uniform quality - Agglomeration of flour - Flammable

Sources: Antal (2015), Hnin et al. (2018), Khampakool et al. (2020), Lenaerts et al. (2018), Mujumdar and Jangam (2011), Nyangena et al. (2020).

When comparing the effects of solar-drying and oven-drying on the moisture content and microbial count of edible insects, the moisture content was not significantly different between the two methods, whereas the microbial count was found to be higher with solar-drying than with oven-drying methods (Nyangena et al., 2020). Therefore, microbial hazards should be considered when using solar-drying methods. Infrared radiation can also be used to dry food materials, which can be

combined with hot-air drying, microwave drying, or freeze-drying technologies (Antal, 2015). In infrared-radiated samples, the diffusion rate increases without increasing the surrounding air temperature. This technology is, therefore, widely used in the food industry owing to its advantages, such as uniform heating, high heat transfer rate, and cost-efficiency. The effect of the hybrid drying method combining infrared drying and freeze-drying was reported to be similar to that of the single freeze-drying method (Antal, 2015). Infrared-assisted freeze-drying of *Protaetia brevitarsis* reduced the energy consumption by more than 90%, and the protein content and appearance score were higher than those obtained with freeze-drying and oven-drying (Khampakool et al., 2020).

Microwaves have various advantages such as a rapid drying speed and easy control of drying conditions; however, non-uniform drying, low penetration depth, and damage in texture are critical drawbacks of using microwaves as a drying technology (Mujumdar and Jangam, 2011). Although minor changes were observed between microwave-dried and freeze-dried *T. molitor*, sensory inspection requires improvement (Lenaerts et al., 2018). In addition to the aforementioned drying methods, modified atmosphere drying and methods involving superheated steam, impinging steam, contact sorption, pulsed electric field, osmotic dehydration, fluid bed, belt conveyor, ultrasound, and ultrasonic waves can be used to dry food materials, including edible insects (Hnin et al., 2018).

Defatting methods

Defatted edible insects have higher nutritional value and functionality than raw edible insect proteins, such as improved foaming capacity and emulsifying properties (Kim et al., 2020a). A summary of the key defatting methods of protein enhancement in edible insects is provided in Table 2. In the food industry, hexane is frequently used to remove fat components from food such as soybean, because of its higher efficiency than other organic solvents (Russin et al., 2011). However, because the hexane solvent originates from fossil fuels, a discontinuous supply and unstable price might impose a limitation for its economic use. Therefore, alternatives to hexane that are amenable to the assessment of environmental and health risks need to be developed. In addition, such alternatives should satisfy the following conditions: Continuous supply, health and environment safety, high fire resistance, and economic and industrial efficiency (Russin et al., 2011). Kim et al. (2021) suggested that ethanol could be considered an alternative to hexane to defat edible insects and improve protein quality. Moreover, Russin et al. (2011) suggested that aqueous extraction could replace hexane extraction. However, some other

Table 2. Edible insect-defatting methods following harvesting and their advantages and disadvantages

Methods	Advantages	Disadvantages
Defatting process		
Hexane	<ul style="list-style-type: none"> - High efficiency - Low protein denaturation 	<ul style="list-style-type: none"> - Environmental implications - Limited quantity - Health risk - Highly flammable
Alcohols	<ul style="list-style-type: none"> - Sustainable - Familiarity 	<ul style="list-style-type: none"> - Higher protein denaturation than hexane - Low efficiency - High cost
Aqueous extraction	<ul style="list-style-type: none"> - Low pollution - Non-toxic - Low cost - Minimal protein denaturation 	<ul style="list-style-type: none"> - The lowest efficiency - High level of residual lipids and their oxidation
Supercritical CO ₂	<ul style="list-style-type: none"> - Rapid oil extraction 	<ul style="list-style-type: none"> - Not suitable for protein extraction

Sources: Gravel and Doyen (2020), Kim et al. (2020a), Russin et al. (2011), Wiese and Snyder (1987).

technologies are applied when defatting from insects using aqueous extraction because the defatting yield of this extraction method was the lowest when compared to organic solvent extraction (Laroche et al., 2019). Ultrasonication, protein precipitation, micro- or ultrafiltration, membrane isolation methods, and enzyme hydrolysis could be suggested to enhance the efficiency of aqueous extraction (Russin et al., 2011). The supercritical carbon dioxide extraction method was developed in the early 1980s; however, this technology appears to be more useful for oil extraction than protein extraction (Russin et al., 2011). Wiese and Snyder (1987) demonstrated that defatting performed after drying or grinding provides a higher oil yield than flakes in soybean; therefore, it is recommended to perform drying or grinding before defatting (Gravel and Doyen, 2020).

Extraction methods

Extraction has been conducted for enhancing the functional properties of proteins. A summary of the key extraction methods of protein enhancement in edible insects is provided in Table 3. Protein solubility could be a key factor and serves as an excellent index of quality, as it is related to other functional properties (Zayas, 2012). The solubility of proteins in the solubilized buffer is determined by the distinction between the isoelectric point of the protein, which in turn is affected by the pH and ionic strength of the buffer (Zayas, 2012). In addition, protein structure and properties such as the amino acid composition, amino acid sequence, polar and nonpolar contents, and molecular weight could be used for determining the proportions of hydrophobic and hydrophilic residues that affect protein solubility (Zayas, 2012). Therefore, by controlling the solubility with different extraction methods, the yield of proteins extracted from different food sources can be changed. Although protein extraction from edible insects has not been explored extensively to date, it has garnered increased interest recently. Yi et al. (2013) studied the effect of aqueous extraction on the protein quality of five different mealworms, and their supernatant yield was approximately 20%. Kim et al. (2020a) compared the changes in protein technical function following protein extraction by centrifugation and suggested that centrifugation could be useful for separating soluble and insoluble proteins. Both these studies used 0.02% ascorbic acid to prevent oxidation during extraction. However, a recent study showed that alkaline conditions can solubilize proteins more effectively than what is possible with ascorbic acid because of their strong ionic strength (Kim et al., 2019a). Furthermore, dissolving the extracted protein in an alkaline buffer enhances its function (Yi et al., 2013). In-salt methods can enhance the ionic strength of edible insects and increase the functional properties of the extracted proteins (Kim et al., 2019a). Following the dissolution of proteins in solution, the protein can be collected via precipitation by adjusting the pH to the isoelectric point, centrifugation, or the salting-out method. Additionally, hydrolysis of insect proteins has been explored as a method to improve the functional properties of edible insects (Purschke et al., 2018b). Freeze-drying or membrane filtration can also be used for the purification of extracted proteins (Gravel and Doyen, 2020).

Table 3. Edible insect-extracting methods following harvesting and their advantages and disadvantages

Methods	Advantages	Disadvantages
pH and salt	- Easy to control	- Different protein solutions by pH and salt
Temperature	- Easy to extract protein	- Irreversible protein
Sonication	- Increased efficiency	- Additional cost - Changes of temperature
Membrane filtration	- Easy to control - Purification of protein is possible	- Long time period

Sources: Gravel and Doyen (2020), Purschke et al. (2018b), Yi et al. (2013), Zayas (2012).

Quality Characteristics of the Protein in Processed Edible Insects

The physicochemical properties of proteins can be modified via the aforementioned steps to enhance the nutritional value and functional properties of edible insects. Regardless of insect biotic factors, chemical or mechanical modification can improve the value of edible insects for people who rely on edible insects as novel food resources.

Nutritional aspects

Edible insects have high nutritional value and low environmental costs in terms of protein production (Bosch et al., 2019; Kim et al., 2019b; Oonincx and de Boer, 2012). Bußler et al. (2016) increased the crude protein yield from two different edible insects (*T. molitor* and *Hermetica illucens*) by defatting protein isolation. Mintah et al. (2020a) estimated the chemical composition of black soldier fly larvae using a Box–Behnken design with the Box–Wilson methodology to optimize the processing conditions. The optimal extraction time, temperature, and alkaline solution ratio to obtain the highest protein yield were 59.43 min, 52.23°C, and a 24.85-fold alkaline solution, respectively. These optimized conditions yielded a protein content of over 80% and enhanced the utility of edible insect proteins. An increase in protein content was also reported in another previous study on insect-processing; however, the authors indicated the possibility of overestimation of the protein content in edible insects (Janssen et al., 2017). The crude protein content was estimated using various nitrogen-to-protein conversion factors (Kp) derived from the ratio of protein nitrogen to total nitrogen, and the average Kp value of three different edible insect larvae (4.76 before extraction, 5.60 after extraction) was lower than the conventional Kp value (6.25). Thus, the non-protein fraction from insects, which includes nitrogen, can cause protein content overestimation, and removal of the non-protein fraction can increase the Kp value of edible insects (Janssen et al., 2017). Kim et al. (2020a) reported that the insect protein content can be increased by defatting and extraction. The protein content was increased to a maximum of 40% after defatting and extraction, and the >80% chitin content in edible insects was removed following extraction. Among various components, regulating or enhancing the protein content is the main challenge in promoting the widespread consumption and nutritional value of edible insects; amino acid profiling and digestibility have also been used to estimate the value of edible insects as novel protein sources. A few fresh edible insects cannot meet the amino acid requirements for humans; however, the general processed edible insects can meet the requirements of human nutrition and high digestibility, which are comparable with those obtained from animal meat (van Huis et al., 2021).

Functional properties

In the food industry, protein-based materials such as meat, fish, eggs, milk, and plant proteins are widely processed using various methods. The major protein functionalities in food include solubility, water/oil holding capacity, foaming properties, emulsifying properties, and gelling properties (Zayas, 2012). Previous studies have shown that the addition of edible insect flour could increase the nutritional value of food, however, it resulted in a decrease in the physicochemical properties (Azzollini et al., 2018; Bessa et al., 2019; Biró et al., 2019; da Rosa Machado and Thys, 2019; Duda et al., 2019; Haber et al., 2019). These characteristics can be enhanced by the removal of non-protein ingredients. Furthermore, modification of insect protein functionality has been investigated (Gravel and Doyen, 2020). Fractionation processing has been used for enhancing insect protein functionalities (Bußler et al., 2016; Gravel and Doyen, 2020; Kim et al., 2019a; Kim et al., 2020a; Mintah et al., 2020a; Purschke et al., 2018b). The increase in protein content increased the protein functionality of *T. molitor*, *P. brevitarsis*, and *Allomyrina dichotoma* (Kim et al., 2020a). Following fat removal, the foaming capacity increased more than

three-fold and the foam stability increased rapidly. Chitin removal had a more critical impact on the emulsifying properties than fat removal. Enzymatic hydrolysis can also be used to enhance the protein functionality of edible insects (Gravel and Doyen, 2020; Purschke et al., 2018b). Purschke et al. (2018b) studied the effect of enzymatic hydrolysis on the functional properties of *Locusta migratoria* L. Insect was hydrolyzed by various enzymes (alcalase, neutrase, flavourzyme, and papain), enzyme-substrate ratio (0.05%–1.0%), heat pre-treatment (60°C–80°C; 10–60 min), and hydrolysis time (0–24 h). They confirmed that changes in functional properties of hydrolyzed insects were different by hydrolysis condition. Protein solubility, foam properties, emulsifying activity, and oil-binding capacity increased with a decrease in the protein molecular weight (decrease from 25–75 kDa to 10–15 kDa) after enzymatic hydrolysis. However, Dion-Poulin et al. (2020) found no significant difference in protein functionality following enzymatic hydrolysis by Alcalase® (3%, 80°C, 15 min) but reported that emulsifying activity decreased. Mintah et al. (2020b) pre-treated soldier flies with enzymatic hydrolysis (alkaline protease; 9,000 U/g, 50°C, 90 min) using ultrasound, and the pre-treated hydrolysates had a high molecular weight distribution below 1,000 Da with changes in the secondary structure of proteins (increased α -helix and decreased β -sheet), consequently improving the dispersibility, turbidity, and particle size. Enzymatic polymerization has been shown to increase protein functionality. Transglutaminase is an effective and typically used polymerizing enzyme in the food industry. Kim et al. (2020b) reported the effect of reaction time on the protein functionality of *P. brevitarsis*, in which a reaction time of over 90 min could induce excessive protein aggregation and decrease in functionality. Therefore, regulation of the non-protein components and structural characteristics of insect proteins should be conducted to improve the protein functionality of insects.

Application of Insect Protein in Foods

The high nutritional value of edible insects in terms of their protein and fat content can help promote their incorporation into conventional dishes. The quality characteristics of insect-enriched foods have been estimated with various methods. Some researchers have focused on the nutritional value, while others have mainly focused on the physical characteristics of insect-enriched food. Detailed information of these studies and the various methods employed are provided in Table 4 (plant-based food) and Table 5 (animal-based food). Here, we summarize the effects of these methods on enhancing or reducing the value and food quality of insect-enriched food from the aspects of nutritional and physicochemical properties.

Nutritional enhancement

Edible insects are a good source of nutrition with several advantages, and their supplementation in various foods has been shown to enhance the nutritional value of plant-based foods (Table 4). For example, protein, fat, fiber, and dietary fiber content was found to be higher in bread containing crickets (*A. domesticus*) than in control bread (Osimani et al., 2018). Cinereous cockroaches also increase the nutritional value of bread by enhancing the protein content and unsaturated fatty acid composition (de Oliveira et al., 2017). Similarly, bread enriched with defatted powder of grasshopper (*Schistocerca gregaria*) had higher protein, fat, and dietary fiber content (Haber et al., 2019). Moreover, supplementation with *T. molitor* improved the nutritional value of bread by increasing the EAA, free amino acid, and protein content, without significant differences in the nutritional quality of lipids (Roncolini et al., 2019). Crude fiber and protein content of egg pasta was increased using mealworm and grasshopper powder supplementation (Çabuk and Yılmaz, 2020). Cricket powder and silkworm powder increased the protein content of durum wheat pasta and buckwheat pasta, and the energy value also increased with the

Table 4. Nutritional effects of the application of edible insects to conventional plant-based food

Compared food type	Added insects	Enriched food quality	Devalued food quality	References
Wheat bread	<i>Acheta domesticus</i>	- Protein, fat, fiber ash, and energy value - Essential leavening amino acid - Essential fatty acid - Firmness	- Specific volume - Microbial characteristics	Osimani et al. (2018)
	<i>Locusta migratoria</i>	- Protein, fat, fiber, and ash content - Essential amino acid	- Specific volume - Overall acceptability	Althwab et al. (2021)
	<i>Tenebrio molitor</i>	- Protein and essential amino acids - Specific volume	- Firmness - Sensory evaluation	Roncolini et al. (2019)
	<i>Schistocerca gregaria</i>	- Protein, fat, ash, fiber	- Hardness, springiness - Specific volume	Haber et al. (2019)
Gluten-free bread	<i>Gryllus assimilis</i>	- Protein, fat, ash, and lipid content - Hardness, chewiness	- Specific volume - High porosity and cell density	da Rosa Machado and Thys (2019)
Pasta	Cricket powder	- Protein, fat, ash, and energy value - Cooking loss - Firmness	- Cooking time	Duda et al. (2019)
Buckwheat pasta	Silkworm powder	- Protein - Cooking time	- Energy value - Acidity over storage	Biró et al. (2019)
Extruded cereal snack	<i>T. molitor</i>	- Protein - Digestibility - Pore density - Pore well thickness	- Poor expansion - Porosity - Pore size	Azzollini et al. (2018)
	<i>Sphenarium purpurascens</i>	- Protein, ash - Browning index	- Carbohydrate value - Whiteness index	Ramírez-Rivera et al. (2021)

addition of insects (Biró et al., 2019; Duda et al., 2019). Various carbohydrate-based foods, such as snacks, protein bars, energy bars, rice, buns, and biscuits, have been enriched with edible insects (Adámek et al., 2018; Alemu et al., 2016; Azzollini et al., 2018; Niaba Koffi et al., 2013; Tao, 2016). These studies suggest that the protein and EAA content of carbohydrate-based foods could be improved by supplementation with edible insect protein powder, consequently enhancing the nutritional value of edible insects.

Furthermore, edible insects can be added to animal protein-based foods as substitutes or ingredients (Table 5). Replacing 10% of lean meat with pre-treated mealworms and silkworms in emulsified sausage was shown to significantly increase its protein and mineral content (Kim et al., 2016). Similarly, silkworm (*Bombyx mori*) pupae added to pork meat batter was reported to increase the protein content of the meat product (Park et al., 2017). Thus, these studies indicated that the addition of insect flour could increase the protein content of various foods. However, because the black soldier fly (*H. illucens*) has a lower protein content than pork, insect-based Vienna-style sausages had a lower protein content than pork-based sausages (Bessa et al., 2019).

Enhancement of physicochemical properties

Proteins have various functional properties, which may affect the final physicochemical properties of plant-based food (Table 4). Gluten-free food may contain alternative proteins that can form a structure similar to that of gluten with a good

Table 5. Nutritional effects of the application of edible insects to conventional animal-based food

Compared food type	Added insects	Enriched food quality	Devalued food quality	References
Meat emulsion	<i>T. molitor</i> larva	- Protein, fat, and ash	- Cooking loss - Protein solubility - Emulsion stability - Hardness - Viscosity	Choi et al. (2017)
Emulsion sausage	<i>T. molitor</i> and <i>Bombyx mori</i> pupa	- Protein, fat, and minerals - Cooking yield - Hardness, springiness, cohesiveness, gumminess, and chewiness	- Protein solubility - Lipid oxidation	Kim et al. (2016)
Meat batter	<i>Bombyx mori</i> pupa	- Protein, fat, and ash - Viscosity - Cooking loss - Hardness, springiness, gumminess, and chewiness	- Lightness	Park et al. (2017)
Jerky type	<i>T. molitor</i> larva	- Amino acid value - Water activity - Drying yield	- Shear force - Rehydration ratio	Kim et al. (2022b)
Ice cream	<i>B. mori</i> pupa	- Viscosity - Shelf-life of foam - Hardness	- Cream overrun - Cream separation - Overall acceptability	David-Birman et al. (2022)

gas-holding capacity (Moore et al., 2004). Cricket powder showed acceptable enrichment in gluten-free products, and bread enriched with insects had better functional results than those enriched with lentil and buckwheat (da Rosa Machado and Thys, 2019). Bread enriched with 5% cinereous cockroach flour showed no significant difference from standard wheat bread, and it was classified as good-quality bread. However, the hardness of bread increased when the replacement ratio was more than 10% (de Oliveira et al., 2017). When wheat flour was replaced with grasshopper flour, the specific volume and hardness decreased. Furthermore, bread enriched with defatted insect flour had higher springiness than that prepared with non-defatted insect flour (Haber et al., 2019). The springiness of bread is closely related to its gas-holding capacity. Defatted edible insects have better foaming capacity and stability than non-defatted edible insects (Crowley et al., 2000; Kim et al., 2020a). These studies further demonstrated that the addition of edible insects decreased the specific volume. For example, bread supplemented with mealworm powder has a higher specific volume and softer texture than standard bread (Roncolini et al., 2019). The authors described that these advantages of mealworm might be attributed to the stabilization effect of melted fat on expanding gas cells (Pareyt et al., 2011). Therefore, further studies are required to determine the effect of insect lipids on bread quality. In pasta enriched with crickets, cooking loss decreased, whereas the optimal cooking time and firmness increased (Duda et al., 2019). Silkworm powder increased the water absorption rate and hardness and decreased the optimal cooking time (Biró et al., 2019). Furthermore, the excessive addition of edible insects induced limited expansion of snacks, which may be due to the increased amounts of fat particles (Azzollini et al., 2018). Therefore, the species and fat content of insects should be carefully selected for improving the quality of carbohydrate-based foods.

Physicochemical properties of animal-based food enriched by edible insects were changed (Table 5). In meat products, insect flour as a protein replacement could increase the cooking yield and textural properties of meat emulsion sausage owing to an increase in the ionic strength (Choi et al., 2017; Kim et al., 2016). Silkworm pupae increased the viscous properties, hardness, gumminess, chewiness, and pH, and decreased the cooking loss value when added as an ingredient in pork meat batter (Park et al., 2017). Vienna-style sausages composed of black soldier fly larvae exhibited similar hardness and

cohesiveness values on day 1; however, after 14 days, these values decreased more rapidly than those in pork meat-based sausages (Bessa et al., 2019).

Suggesting Strategies for Improving Edible Insect Protein Characteristics

Aforementioned, edible insect processing such as drying, defatting, and protein extraction could increase protein functionality and use of edible insects can be expanded. However, there are clear indications that edible insect proteins as food ingredients exhibit poor maintenance of form and textural properties, suggesting that the application of edible insects mimicking conventional food may be difficult. Therefore, protein modification and improvement of cross-linking are suggested for their efficient use in the edible insect industry. The protein modification could be done with various methods. Protein cross-linking reactions during processing occur under various conditions such as alkaline conditions, heating, oxidizing agents, Maillard reaction, lipid cross-linking, and enzymatic reactions (Singh, 1991). When protein is denatured, disulfide cross-linking in protein-protein interaction is the most frequent reaction occurring in food processing. Cysteine residues produce disulfide cross-links and gelling properties could be affected by this bond. In addition, protein denaturation exposed hydrophobic residues and protein functionality can be increased with protein denaturation (Zayas, 2012). However, excessive denaturation under non-suitable conditions could aggregate protein and deteriorate quality characteristics of protein-based food (Nikbakht Nasrabadi et al., 2021).

Physically, heating is used most widely. Unfolded protein by thermal energy could improve functionality (Zayas, 2012). With technological development, various heating methods are used to heat food such as ohmic heating, microwave heating, radio frequency, and infrared irradiation (Nikbakht Nasrabadi et al., 2021). However, heating under alkaline conditions induces racemization that causes the production of de-hydro amino acids (lysinoalanine and lanthionine), which cannot be digested, thereby reducing the nutritional value of food (Gerrard, 2002). Moreover, high pressure can be used to accelerate the cross-linking reaction and protein aggregation. Food processed under a high-pressure conditions has a soft texture; however, it is not easily broken. Pressure levels and time are important factors in determining the textural properties of food (Totosaus et al., 2002). Above these technologies, irradiation, pulsed-electric field, extrusion, and cold atmospheric plasma have been used to improve quality characteristics of protein-based food (Azzollini et al., 2018; Nikbakht Nasrabadi et al., 2021; Ojha et al., 2021).

Chemically, grafting technology, which involves the reaction of protein and sugar (Maillard reaction), can improve the functional properties of proteins. However, among the various compounds produced during the Maillard reaction, compounds that improve the functional properties of reacted proteins by reducing sugar have not yet been identified (Gerrard, 2002). Phosphorylation has been used to modify protein-based materials to keep nutritional value (Liu et al., 2020). Catalyzed covalent bonds between phosphate and protein change the protein structure and protein functionality can be improved (Nikbakht Nasrabadi et al., 2021). Physicochemical properties of pH-shifted edible insects were improved, and it also improved gel properties of myofibril gel (Kim et al., 2022a). In addition, because edible insects contain abundant chitin contents, acetylation modification could be improved functionality of insects (Kim et al., 2019b; Nikbakht Nasrabadi et al., 2021).

Enzymatic reactions may be a useful way to enhance the textural properties along with food safety. Disulfide cross-linking can be achieved through enzymatic systems such as disulfide isomerase and rearrangement of low-molecular-weight sulfhydryl compounds, and sulfhydryl oxidase can catalyze the formation of disulfide bonds (Singh, 1991). Lysine residues can cross-link with each other, and this reaction can enhance or retain the nutritional value of protein by transglutaminase (Gerrard, 2002). Hydrocolloids can be used to enhance the properties of insect proteins. Owing to their high viscosity,

thickening, emulsifying, stabilization, and gelling properties, hydrocolloids can maintain the shape of protein extracts, and the processes to manufacture food mimics using edible insects could be increased (Saha and Bhattacharya, 2010). Various ingredients containing proteins and polysaccharides have high hydrophilic properties. Starch; xanthan; and various gums such as guar gum, locust bean gum, and cellulose derivatives can be used as thickening ingredients, while alginate, gelatin, agar, carrageenan, and pectin act as gelling agents (Saha and Bhattacharya, 2010).

Conclusion

Although edible insects with excellent nutritional value have been consumed widely in developing countries, food neophobia and prejudice against insects limit their consumption as food sources on a global scale. Therefore, the food industry has attempted to change the appearance of insects through grinding or extraction before adding them to conventional food. Unfortunately, despite the enriched nutritional value of edible insect-supplemented food, the textural and gelling properties are reported to deteriorate. Therefore, there is a need to develop more consumer-friendly insect food to increase the consumption of edible insect proteins as a sustainable alternative to conventional food.

Conflicts of Interest

The authors declare no potential conflicts of interest.

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Ethics Approval

This article does not require IRB/IACUC approval because there are no human and animal participants.

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