Utilization of Edible-Insects as Protein Substitute in Food and Effects of Processing on Their Nutrient Contents and Protein Functionalities

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ABSTRACT

Population growth, poverty and climate change dictate the need for additional protein sources. Edible insects are potential protein substitutes and can provide both humans and animals with the required amount of protein, essential amino acids, and other indispensable nutrients. Incorporating edible-insects into familiar products and subjecting them to adequate processing that masks their natural appearance will improve their consumption. This article provides insights on the potentials of edible-insects as novel ingredients in food processing and its various benefits. Effects of processing on their nutritional and functional properties were also discussed. Literature was gathered through an online search on the Science Direct database and Google Scholar. Edibleinsect powders, protein isolates and concentrates and oils were reported to be incorporated into bugger, chips, chocolate, bread, cookies, and other baked snacks. The addition of insect products improves protein, fat, fiber, and minerals contents. Insects also contain healthier lipids when compared with conventional proteins. Edible-insects will provide essential nutrients to the rapidly growing world population when more attention is given to their production, processing, safety, and marketing. Efforts need to be strengthened to secure global acceptance of insect protein since the conventional sources are not reliable and cannot satisfy the world population in decades to come. Keywords: entomophagy, food processing, meat alternative, protein substitute, unconventional protein.

INTRODUCTION

Insects eating (entomophagy) was dated back to antiquity (Cartay *et al.*, 2020; Liu and Zhao, 2019; Sun-Waterhouse *et al.*, 2016) and the use of insects as food is as old as the history of mankind (Das *et al.*, 2019). Insects can be processed into food that can

provide essential nutrients (Cartay *et al.*, 2020; Dube *et al.*, 2013).

They are also fundamental in the pollination process (Cartay *et al.*, 2020), production of honey, silk, natural colouring agents, (Testa *et al.*, 2016), nutrients cycling and biological control (Dzerefos and

Witkowski, 2014), and improvement of soil fertility through waste bioconversion (van Huis *et al.*, 2013).

There is an increasing demand for protein-rich foods, more attention is given to the novel sources of protein, researchers and marketers are putting more efforts into promoting edible-insects among new consumers to meet the obligatory need to diversify protein sources (Pippinato et al., 2020). The desire to reconsider discarded eating habits and food sources into modern recipes has fascinated the interest of researchers, chefs, and businessmen on edible-insect potentials as an alternative protein source (Testa et al., 2016).

Reports from many researchers shown that insect proteins can be used as a substitute for conventional protein in many products without adverse effects on their sensory qualities. Their inclusion in food has proven to improve the nutritional values of many processed foods (Imathiu, 2020).

Research in entomophagy covered many areas of study including food science, human and animal nutrition, health, entomology, ecology, agriculture, biochemistry, anthropology, sociology, and history (Yen, 2015).

The most studied insect in recent years were cricket, silkworm pupae, housefly (Testa *et al.*, 2016), lepidopteran larvae, coleopteran larvae, different species of grasshoppers, locusts, and termites (Kelemu *et al.*, 2015).

This article intended to provides insights on the potentials of edible-insects as novel ingredients in food processing and its various benefits. Effects of processing on their nutritional and functional properties were also discussed.

EDIBLE-INSECTS AS CONVENTIONAL PROTEINS SUBSTITUTE

Edible-insects and their products are used as ingredients in food processing, insect powders, protein isolates and concentrates, and oils were reported to be incorporated into burger, chips, chocolate, bread, cookies, and other baked snacks (Table 1).

The addition of insect products improved protein, fat, fibre, and minerals contents without affecting functional properties and sensory qualities. It also improved dough characteristics and baking properties (Table 1).

Gravel and Doyen (2020) reported that insect proteins are potential functional ingredients in foods and can play important roles in large scale food production. Food scientists are working hard on the possibilities of incorporating insects into some staple foods. At present, more attention is given to bakery goods. Substitution of wheat flour with 5 % insect flour improves the protein and fibre content of bread without properties affecting the dough and acceptability of the bread (González et al., 2019).

Insect flour at a 7.5 % level of substitution improves the nutritional content of corn flour extrudates snack mixture without affecting extrusion parameters (Igual *et al.*, 2020). Extrusion cooking of soy concentrate with honeybee drone brood produced protein-rich snacks with 66 % protein content (Ulmer *et al.*, 2020). Inclusion of 25 % crude black soldier fly larvae fat as a butter substitute in the production of cakes and cookies and 50 % in the production of waffles have no effects on the texture, colour, and overall liking of these products (Delicato *et al.*, 2020).

Substitution of wheat flour with grasshopper powder at 100 g/kg grasshopper powder has no effects on the sensory qualities of the grasshopper-based bread, the substitution increased the protein content by 60 % and softness of the bread (Haber *et al.*, 2019). Cricket powder at 15 % inclusion

enhanced bread dough stability and reduced the degree of softening (Cappelli *et al.*, 2020).

Leavened crunchy snacks produced with 30 % lesser mealworm powder exhibit low water activity (aw) and have pleasant sensory attributes and better mineral, protein, and amino acid contents than that produced from 100 % wheat flour (Roncolini *et al.*, 2020). The low aw exhibited by lesser mealworm powder reveal the potential of using insect protein as a preservative in food.

Mulberry silkworm pupae and African palm weevil larvae can be used as a beef substitute in the production of filling for snacks and cuisines (Akande *et al.*, 2020a). Skim milk can be replaced with locust and mulberry silkworm pupae powder in the production of high-energy biscuits, at 15 % inclusion, the nutritional value is above the minimum level prescribed by USAID (Akande *et al.*, 2020b).

Another potential area of using insects is in improving the sensory properties of healthy foods with acceptance problems, silkworm powder was used by Biró et al. (2019) to mask the unpleasant sensory characteristic of buckwheat. Insects proteins are potential functional ingredients in food processing, the findings of Gravel and Doven (Gravel and Doyen, 2020) revealed that insects protein concentrate and isolate are suitable ingredients with excellent functionality. The textural properties of insect proteins are amazing in 3D printing, their consumption can be improved through the production of foods with attractive, shapes, structure, and texture (Caporizzi et al., 2019).

PROCESSING OF EDIBLE INSECTS

Insects are consumed either raw, fried, or roasted (Das *et al.*, 2019). The most common processing methods for insects are trimming, boiling, sun-drying, roasting, and pan-frying (Akullo *et al.*, 2017; Ebenebe *et*

al., 2017). In India, insect meals are prepared by boiling and roasting, sometimes with spices (Chakravorty, 2014), other processing methods are sand-roasting and osmotic drying using rock salt (Bose, 2020). Surplus catches during the season are preserved by sun-drying and stored for future use (Akullo et al., 2017), this ensures the continuous supply of insect protein year-round (Dube et al., 2013). The crude techniques used in the processing and preparation of insects improve their safety, shelf-life, nutritional value, and palatability (Akullo et al., 2017; Dube et al., 2013; Mézes and Erdélyi, 2020). Köhler et al. (2019) reported that insects purchased from the supermarket have better nutrient content than those gathered from the therefore, adopting street. modern techniques in insect processing will further improve their safety, commercial value, and versatility (Akullo et al., 2017). Edible insects were reported to contain many important nutrients but there is little information regarding the effects of different processing on their nutritional qualities, nutrients bioavailability, and toxicity (Ayensu et al., 2019).

Effects of Processing on the Qualities and Functionalities of Insect Products

The findings in Table 2 revealed that cooking affects nutrient contents and their bioavailability, digestibility, pH. and functional properties. Thermal processing of insects generally lower microbial counts and affects their distribution and activities during storage. The higher temperatures involved in cooking increase acidity due to the breaking down of glycogen into lactic acid. Higher temperature and low pH affect gelling and foaming properties, trigger coagulation, and increase surface hydrophobicity. Hydrolyses of both protein and lipid were observed during prolonged heating. Cooking has no effects on fatty acids composition and protein secondary structure, but heating with



NaCl decreases α -helix structure and surface hydrophobicity. Drying affects macronutrient contents, protein solubility, and oxidation (Table 2). Thermal processing of improved their insects safety. acceptability, digestibility, palatability (Garofalo et al., 2019; Megido et al., 2018; Murefu et al., 2019) nutritional quality, and bioavailability of sulfur-containing amino acids (Poelaert et al., 2018). Higher temperatures during cooking can have damaging effects on lipid and protein (Megido et al., 2018). Cooking methods can nutrients affect content and their bioavailability in insect, boiling, and vacuum maintains good levels cooking of polyunsaturated fatty acids and proteins in mealworms (Megido et al., 2018).

Reports in Table 2 show that extraction conditions can affect the properties of edible-insect protein extract. The solubility of the protein extract depends on the extraction pH. Increased protein content was recorded at pH of 10 in combination with Ultrasound treatment. Pressurised-liquid assisted extraction lower cholesterol level and increases PUFAs contents. Alkaline hydrolysis lower foam stability and emulsification properties of the protein extract. Exposure to ultrasound improved antioxidant activity, solubility, and foam expansion of the protein extract over a wide range of pH (2-12) (Table 2). Liceaga (2019) reported that the functional properties of insect flour can be improved by controlled enzymatic hydrolysis. Physical properties such as turbidity, dispersibility, and particle size of insect proteins can be modified by ultra-sonication. An increase in the sulfur hydride value, dispersibility, and zeta potential, and a decrease in turbidity and particle size in protein preparations and hydrolysates were observed in soldier fly larvae subjected to ultrasonic treatment (Mintah et al., 2020). Increased foaming and emulsifying abilities, and protein value were

observed in grasshopper and honey bee brood protein extracts obtained using alkaline and sonication-assisted extraction (Mishyna *et al.*, 2019a). Mishyna *et al.* (2019b) demonstrated the mechanism of gel aggregate formation by both covalent and non-covalent intermolecular interaction occurs during heat denaturation of freezedried honey bee brood protein, this unveils the potential of using insect proteins as a gelling agent. Fermentation is a promising technique for producing a wide range of edible insect products including extracts, sauces and paste (Castro-López *et al.*, 2020).

The addition of insect proteins into wheat flour affects the dough and baking properties of baked foods. At 15 % incorporation, cricket flour increase dough tenacity and reduces extensibility, this led to poor volume in the baked product. The addition of insect flour also lowers expansion and crunchiness in extruded products. Defatting improved overall liking, digestibility, and lower hydrophobicity (Table 2)

The killing method of insects can affect the appearance and qualities of insect products. Killing generally increases the browning index and lowers radical scavenging. Killing in the absence of oxygen minimizes lipid oxidation and improves its nutritional qualities (Table 2). Killing methods affects the acceptability of insect by influencing saltiness and umami taste (Table 2). Leni et al. (2019) compare freezing (the most common killing method) with balancing and found out that the slow killing process involves in freezing gave room for reactions many enzymic including melanization reaction, killing by balancing inhibit browning, increase extractability and digestibility of protein and maintain essential amino acids. The killing method also affects the lipid composition of the insect products during storage, Caligiani et al. (2019) reported that killing by balancing provides

more stable lipid while killing by freezing causes the formation of free fatty acids during storage probably due to the activation of lipases.

OTHER INDUSTRIAL APPLICATIONS

Scientists strongly believe that insects will play a vital role when the insect industry becomes well established and it will have positive impacts in many areas including industries, government, business, and research (Dossey et al., 2016) and its financial repercussions will impact the economy in general (Costa-Neto and Dunkel, 2016). In addition to the provision of food and feed insects can convert organic waste into important industrial raw materials. Insects have been foreseen as potential raw materials for many industrial processes (Tang et al., 2019) including food fortification during processing. Insects have higher anti-microbial peptides than any other animal, therefore, can be used as a novel source of antibiotic (van Huis, 2020a). Black soldier fly is a potential candidate for the production of antibiotics, textured protein, and bioplastic (van Huis, 2020b). Insect when used as feed have positive benefits on livestock health and welfare and their use as feed can reduce the use of antibiotics in livestock production (Dicke. 2018: Veldkamp and Eilenberg, 2018). Insect postproduction waste is used as plant manure, and in an advanced way, can be used in the production of bio-methane gas using an anaerobic digestion process (Bulak et al., 2020).

RECOMMENDATIONS AND CONCLUSION

Authors recommends several points such as: at present, only few species are produced commercially, for effective promotion of entomophagy, the idea of insects gathering must be discarded, edible insects should be produced in large quantities

at an industrial scale; People involve in traditional insect gathering should be transformed into insect farmers by providing them with the required skills and capital; Understanding the interaction between insect products and other food ingredients is critical in using insects as a novel ingredient in foods; Observing good personal hygiene throughout the production chain and the use of an appropriate HACCP system can improve the safety of edible insects; Processing into delicious, attractive, and irresistible meals with no visible insects, or their parts, and the use of an appropriate and eye-catching packaging system will promote edibleinsects' acceptance.

Authors concluded that processing improves safety, stability, nutritional value, digestibility, palatability, nutrients bioavailability, acceptability, commercial value, functional properties, and versatility of edible-insects. The addition of processed insects into familiar foods improves their ingredients' nutritional qualities and functionality. Killing and processing methods affect protein functional properties, nutrient contents, and their bioavailability. Edible-insects and their products are used as ingredients in food processing. Insect powders, protein isolates, and concentrates, and oils were reported to be incorporated into burger, chips, chocolate, bread, cookies, and other baked snacks.

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Food products	Insect/Insect product	Incorporation level	Major findings	Reference		
Cake and cookies	black soldier fly larvae fat	Addition of 25 % as butter substitute	Does not affect consumer preference	(Delicato <i>et al.</i> , 2020)		
waffles	black soldier fly larvae fat	Addition of 50 % as butter substitute	Does not affect consumer preference	(Delicato <i>et al.</i> , 2020)		
Gluten-free bread	house cricket (Acheta domesticus L.)	Addition of 5.4 %	 Does not affect fermentation process Enrich the bread with unique flavor Improved antioxidant content of the bread 	(Nissen <i>et al.</i> , 2020)		
Crunchy snacks	lesser mealworm (<i>Alphitobius</i> <i>diaperinus</i>) powder	Inclusion of 30 %	 Protein content increased by 99.3 %, also essential amino acids Increased Fe, P, and Zn, with Zn content increased by 300 % Possessed pleasant sensory attributes and low aw 	(Roncolini <i>et al.</i> , 2020)		
High-energy biscuit	Silkworm pupae (<i>Bombyx mori</i>) and locusts	Addition of 15 % as a protein substitute	 Improve in protein, fat and, fiber contents and energy value were observed The biscuits meet USAID standard No effects on the sensory qualities 	(Akande <i>et al.</i> , 2020b)		
3D printed cereal-based snacks	Yellow mealworms (<i>Tenebrio molitor</i>) larvae	Inclusion of 20 % larvae flour as a novel protein source	 Increase dough softness and flowing efficiency, these subsequently improved the geometry of the snack Microstructure was improved due to better water evaporation during baking Improvement in essential amino acids contents was observed 	(Severini <i>et al.</i> , 2018)		
Burger	Mealworm	Addition of 50 % as a meat substitute		(Megido <i>et al.</i> , 2016)		

Table 1. Insect Products as Potential Protein Substitute

Buckwheat pasta	Silkworm	Inclusion of 10 g/100g pasta	 Improve the nutritional value of the pasta without affecting its acceptability Received higher sensory scores than the control Reduced cooking time 	(Biró <i>et al.</i> , 2019)
Cookies	House cricket (Acheta domesticus L.)	Addition of 10 % as a wheat flour substitute	 Has no effects on the flavor, taste, or texture. Was accepted by more than 70 % of the panelists 	(Sogari <i>et al.</i> , 2017)
Bread	cricket (Acheta domesticus) powder	Addition of 10 % as a wheat flour substitute	 Does not affect the sensory qualities Improves protein and essential amino acids contents 	(Osimani <i>et al.</i> , 2018)
Bread	cinereous cockroach (Nauphoeta cinerea)	Addition of 10 % as a wheat flour substitute	 Has no effects on the functional and sensory properties Protein content increased by 49.16 % 	(de Oliveira <i>et al.</i> , 2017)
Tortilla chips and chocolate bars		Tortilla chips containing 15% cricket flour and chocolate bar with figs and 5.5% cricket flour	 Chocolate bar with insect flour possess better sensory qualities, followed by whole cricket. Snacks with visible whole insects were scored low 	(Cicatiello <i>et al.</i> , 2020)
Bread	Hermetia illucens, Acheta domestica and Tenebrio molitor	Addition of 5 % as a wheat flour substitute	 Hermetia illucens and Acheta domestica increased dough stability and reduced water absorption Improved protein and fibre contents 	(González <i>et al.</i> , 2019)
Extruded corn snacks	Cricket flour (Acheta domesticus)	Enrichment with 12.5 and 15 %	Improved the protein content to EC recommendation n. 1924/2006	(Igual <i>et al.</i> , 2020)

Insects	Origin	Processing methods	Effects	Reference	
Beetle & cricket	Wild	Boiling	Reduced Fe and Zn bio-accessibility by 50 % and lower protein content and digestibility	(Manditsera <i>et al.</i> , 2019)	
mealworms	Laboratory reared	Vacuum cooking Frying Boiling Oven cooking	Lower protein, lipid, and ash contents Lower protein and ash contents and increases lipid content Increases protein and ash contents and lower lipid content Increases protein and ash contents and lower lipid content	(Megido <i>et al.</i> , 2018)	
Cricket flour (Acheta domesticus)	Farmed		Increases dough tenacity (P) and curve configuration ratio (P/L), and reduces dough extensibility (L). these subsequently lead to a fall in volume	(Cappelli <i>et al.</i> , 2020)	
Cricket flour (Acheta domesticus)	Farmed	Addition of 15 % in extruded corn snacks	 Lead to low crunchy and low expansion extrudates Modified water solubility index and improve stability 7.5 % inclusion was recommended for better extrudates parameters 	(Igual <i>et al.</i> , 2020)	
Acheta domesticus and Tenebrio molitor	Farmed	ultrasound-assisted extraction and pressurized-liquid extraction	Causes a decrease in saturated fatty acids and monounsaturated fatty acids contents and increase PUFAs contents, also lower cholesterol level	(Otero <i>et al.</i> , 2020)	
Cricket- based snacks	Farmed	Effect of degree of processing and incorporation on the acceptability	Degree of processing affects acceptability, insect-based products should contain processed insects, not whole insects	(Gmuer <i>et al.</i> , 2016)	
Protaetia brevitarsis larvae	Wild	Defatting using hexane	 decrease in hydrophobicity and tryptophan fluorescence intensity was observed digestibility is higher than that of beef 	(Lee <i>et al.</i> , 2021)	
Cricket	Farmed	Broth making by cooking in pouches at 85 °C for 1 h in a water bath	Lower pH due to the breakdown of glycogen and formation of lactic acid	(Farina, 2017)	

 Table 2. Effects of Processing on Nutritional Qualities, Functionality and Acceptability of Insect Products



<i>Hermetia</i> Lab <i>illucens</i> rear larvae	ured		Alkaline hydrolysis improved protein solubility, but lower forming stability and emulsifying properties. Ultrasound treatment improves antioxidant strength, solubility, and foam expansion properties over a wide pH range (2-12)	(Mintah <i>et al.</i> , 2019)
Honey bee brood (<i>Apis</i> <i>mellifera</i>)			Highest coagulation achieved at 85 °C and pH 5-7 The least gelation concentrations vary between 5 to 11 % at pH of 7 and 3 respectively	(Mishyna <i>et al.</i> , 2019c)
House Far cricket (Acheta domesticus)	rmed	Effects of different killing method: blanching, steaming, freezing, carbon dioxide, vacuum,	 all the killing methods were found to increase the browning index and reduce radical scavenging activity. better proximate values were obtained by carbon dioxide, vacuum, and plastic bag killing killing by freezing, carbon dioxide, vacuum, and plastic bag provide healthier lipids and minimize lipid oxidation 	(Singh <i>et al.</i> , 2020)
Mealworms Lab (<i>Tenebrio</i> rear <i>molitor</i>)	boratory ared	Comparing the effects of different drying	 all the drying methods affect the macronutrient contents protein solubility is higher in freeze-drying and lower in microwave drying, fluidized bed drying, and vacuum drying oxidative status is higher in freeze-dried samples 	(Kröncke <i>et al.</i> , 2018)
Yellow Far mealworm (<i>Tenebrio</i> <i>molitor</i>) and lesser mealworm (<i>Alphitobius</i> <i>diaperinus</i>) larvae	rmed	3 C/	Processing methods lower microbial counts and affect their distribution in the finished products and during storage under different conditions	(Stoops <i>et al.</i> , 2017)

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Black cricket (Gryllus assimilis)	Farmed	aggregation on foaming and gelling	 Heat treatment alone doesn't change the protein secondary structure but increase the surface hydrophobicity critical gelling concentration achieved at 90 °C for 15 min exposure the foamability at the treatment; 75 and 95 °C for 15 min, with or without NaCl is similar to that of whey protein isolate heating with NaCl induced decrease in α-helix structure content and surface hydrophobicity 	(Santiago 2021)	et	al.,
Yellow mealworm (<i>Tenebrio</i> <i>molitor</i> L.)	Laboratory reared		 The treatments have no effects on fatty acid profiles Hydrolysis of triacylglycerols was favoured by the low-temperature long time treatment Low-temperature long time treatment also lead to protein hydrolysis which leads to an increase in free amino acid contents 	(Melis et a	ıl., 20)18)
Tenebrio molitor	Farmed	Protein extraction using water at alkaline	1. At pH 10 and 45 °C protein content was increased by two folds	(Azagoh 2016)	et	al.,
larvae Crickets (Acheta domesticus and Gryllodes sigillatus)	Farmed	pH Effects of defatting and freeze-drying on nutritional value and sensory qualities of cereals bars enriched with insects	2. The solubility of the extracted protein depends greatly on pH Defatting improved the overall liking and willingness to consume the products	(Ribeiro 2019)	et	al.,
Crickets	Farmed	Effect of killing methods (freezing and heating) on the sensory and physiological properties of insect broth	There is significant difference between the samples on overall liking, saltiness, and umami scores	(Farina, 20)17)	