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Blanching pre-treatment on quality characteristics of cricket powder

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Abstract

Cricket powder (CP) is a valuable cricket-based product that has recently been developed and applied in many food products. Postharvest insect processing has a significant impact on CP quality. In this study, the effects of blanching pre-treatment at different temperatures (80, 90, and 100°C) and times (5, 10, and 15 mins) were investigated on the physicochemical and microbiological properties of two edible crickets (*Gryllus bimaculatus* and *Acheta domesticus*) powders. CP preparation without blanching treatment served as the control. CP obtained from blanching at 80°C for 10 mins increased protein and fiber content but decreased fat content compared to unblanched CP in both species. CP contained histamine at lower than 4 µg/g. Blanching at 80°C for 10 mins gave the highest total phenolic content (TPC) (150.0 and 258.4 mg GAE/g for *G. bimaculatus* and *A. domesticus*, respectively) and the highest antioxidant activities in all assays (DPPH, ABTS•+, and FRAP). Compared to unblanched CP, blanching crickets also reduced the total microbial load in both CPs. Results suggested that blanching as a pre-treatment improved the safety, functional properties, antioxidant activity of cricket powder.

1. Introduction

The global population is steadily increasing and is expected to reach 8.6 billion by 2030 and 9.8 billion by 2050 (United Nations, 2017), leading to food shortages for both humans and animals. The consumption of insects is therefore proposed as an alternative future food source that is high in proteins, fats and vitamins and can replace animal protein (FAO/INFOODS, 2013). Edible insects present a novel sustainable source of protein for human and animal diets (House, 2018). Insects are widely consumed in Asia, North and South America, and Africa comprising 1,900 different species (FAO/INFOODS, 2013). Crickets are a promising species for rearing because of their attractive nutritional profile and high protein content. They are easy to find locally and inexpensive (FAO/INFOODS, 2013). The house cricket, *Acheta domesticus*, a valuable edible species for human diets, has been authorized as a novel food ingredient for the EU market (Van Huis *et al*., 2013; Sogari, 2015; European Commission, 2022). Cricket powder is a rich source of proteins, unsaturated fatty acids, probiotic fibers, vitamins and minerals as a food additive, with huge value-added processing potential for hybrid meat products (Bawa *et al*., 2020a). *Yarrowia lipolytica* and *Debaryomyces hansenii* strain hydrolysates show potential as food ingredients to improve food safety, functionality and sensory and technological properties for food formulation (Patrignani *et al*., 2020). Cricket production and the market for trading insects for human food have shown significant increases every year.

Currently, the most common form of cricket products is powder or flour in bread, protein bars and snacks. Postharvest treatment is a key step to control cricket powder quality. Microbiologically hazard-free harvesting of insects is difficult to achieve. Using insects as food also has potential microbiological risks as insects can transport pathogenic microorganisms into humans, or animals. The European Food Safety Authority carried out a microbiological risk assessment for the production and consumption of insects as food and feed, with results equivalent to other unprocessed animal protein sources. Microbial contamination of insects can occur from

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farming, production and storage processes (Finke *et al*., 2015). Diets used for feeding insects and maintaining hygienic conditions during rearing and processing all influence the microbiology of edible insects (van der Fels-Klerx *et al*., 2018). Therefore, optimal heat treatment and storage conditions for edible insects are recommended before marketing (FASFC, 2014).

Extensive studies have assessed the presence and prevalence of pathogenic microorganisms in insects used as human food and animal feed and protecting consumers from potential health hazards is essential. Currently, no specific microbiological criteria for edible insects are enforced in the European Union (Fernandez-Cassi *et al*., 2019). Histamine causes food poisoning and allergic reactions. High concentrations are risk factors for food intoxication, whereas moderate levels may lead to food intolerance (Bodmer *et al*., 1999). Many methods including modified atmosphere packaging, irradiation, high hydrostatic pressure and food additives have been used to regulate the buildup of histamine in food products (Naila *et al*., 2010). These methods inhibit the growth of histamine-producing bacteria and histamine decarboxylase activities. Chung *et al*. (2017) determined that frying and grilling increased histamine levels in foods, whereas boiling and fermenting were more effective in reducing histamine content. Suitable insect postharvest processing methods are necessary to maintain the safety and hygiene standards of high-quality food ingredients as the final products.

Traditionally, popular methods for processing crickets include roasting, toasting, frying, smoking and sun drying or their combinations (Simpanya *et al*., 2000; Caparros Megido *et al*., 2017; Mutungi *et al*., 2019). These approaches reduce microbial hazards but have undesirable results such as loss of thermally sensitive nutritional and bioactive components and lipid oxidation (Klunder *et al*., 2012). These changes impact sensory characteristics such as odor and color which are crucial for consumer acceptance of insect-based products. Browning is a key factor relating to the quality of insect-based products during processing (Mancini *et al*., 2019a; Tonneijck-Srpová *et al*., 2019; Ssepuuya *et al*., 2020). Several enzymes play a role in browning including phenoloxidase, laccase, tyrosine hydroxylase, decarboxylase and peroxidase (Andersen, 2012). Most phenolic compounds in insects are derived from l-tyrosine through the shikimic acid pathway using enzymatic reactions. Phenolic compounds can react with proteins and amino acids forming dark compounds (melanins). Janssen *et al*. (2017) reported enzymatic browning in three insect species (*Tenebrio molitor, Alphitobius diaperinus, Hermetia illuce*ns).

Blanching is a food processing pre-treatment that deactivates enzymes and maintains the quality of food products. Mancini *et al*. (2019a) successfully applied blanching at 60°C for 5 mins to reduce phenoloxidase in mealworm larvae. Similarly, blanching heat treatment also reduced microbial loads and increased product safety (Fellows, 2009; Wang *et al*., 2017). Mancini *et al*. (2019a) showed that blanching at 60°C for 5 mins or at 90°C for 2.5 mins reduced the microbial population in mealworm larvae. Moreover, reduced bacterial counts in *Alphitobius diaperinus* and *Tenebrio molitor* larvae were achieved by blanching at 88°C for 5 mins and 100°C for 10, 20 and 40 s, respectively (Vandeweyer *et al*., 2017; Wynants *et al*., 2018). Boiling decreased the amount of ash, protein and insoluble nitrogen, with a decrease in protein digestibility. Boiled or fried and dried Sudanese locust (*Anacridium melanorhodon*) was low in digestible protein (El Hassan *et al*., 2008). This study was carried out to investigate the effects of blanching temperature (80, 90 and 100°C) and time (5, 10, and 15 mins) on the physicochemical and microbiological properties of two edible crickets (*G. bimaculatus* and *A. domesticus*) powders.

2. Materials and methods

2.1 Materials

Two species of frozen cricket (*G. bimaculatus* and *A. domesticus*) were purchased from a local farm following Standard Agricultural Practices (GAP) (Maha Sarakham, Thailand). All cricket samples were harvested at 40–42 days old, cleaned and washed in ice water before packing in plastic bags (1 kg /bag) and frozen (-18±1°C). Frozen cricket samples were thawed by immersing in ice water at 2±1°C for 60 mins before the blanching process.

2.2 Blanching and preparation of cricket powders

Cricket samples were blanched at 80, 90 and 100°C in hot water at a cricket-water ratio of 1:10 (w/w). Samples were randomly taken at 5 mins intervals (0, 5, 10 and 15 mins) at each temperature and immediately cooled in an ice bath. Fresh (unblanched) and blanched cricket samples were dehydrated using a tray dryer (Memmert, OLM-500, Germany) at 80±2°C for 10 hrs until moisture content was lower than 10±1% (%wet basis). To obtain full-fat cricket powder (CP), dried crickets were finely ground using a coffee grinder before passing through an 80-mesh sieve. All full-fat cricket powder samples were vacuum-packed in aluminum foil zip-lock bags and kept frozen (-18±1°C) until used.

2.3 Proximate composition of cricket powder

Moisture content, crude protein, ash, crude fat, and total dietary fiber of all CP samples were determined in

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triplicate using the standard AOAC (2000) procedures as follows:

2.3.1 Determination of moisture

Briefly, a 2.0 g sample of each CP sample was placed in a crucible and heated in an oven at 105°C for 6 hrs (AOAC, 2000). The dried sample was placed in a desiccator, allowed to cool, and then weighed again. Moisture content was determined for all CP samples, and proximate composition was calculated using a wet weight basis of g/100 g.

2.3.2 Determination of crude fat

Crude fat content was determined using the Soxhlet equipment with petroleum ether (AOAC, 2000). In brief, a 2.0 g sample of CP was placed in a fat-free thimble and then placed in an extraction chamber equipped with the Soxhlet apparatus. Petroleum ether was added to the extraction device and extracted for 2 hrs at a drip rate of 10 ml/min. Following extraction, the fat-containing solvent chamber and fat-free sample thimbles were removed from the system. The chamber was placed in an oven at 105°C for 24 hrs before being cooled in a desiccator. The crude fat content was calculated on a wet weight basis as g/100 g.

2.3.3 Determination of crude ash

Briefly, a CP sample (1.0 g) was weighed in constant weight crucibles, placed in a muffle furnace, and burned at 550°C for 24 hrs (AOAC, 2000). After that, the sample was cooled in a desiccator and weighed until a steady weight was achieved. The ash content was expressed in grams per 100 grams of wet weight.

2.3.4 Determination of crude protein

The protein content of all CP samples was determined using the Kjeldahl method (AOAC, 2000). A 0.3 g CP sample was weighed and placed in the digestion tube. Then, 0.5 g of digesting mixed catalyst and 3 mL of concentrated H_2SO_4 were added to each sample. The mixture was heated to begin digestion until a clear green color was achieved. After cooling, the digests were diluted with distilled water, and the contents were transported to micro distillation equipment. Each digest in the distillation chamber received about 10 mL of 40% NaOH. A conical flask with 6 mL of 4% boric acid solution was placed under the condenser. The distillation process continued until 30 mL of the distillate was caught in the boric acid solution. The distillates were then titrated with 0.1 N HCl, and the crude protein content was calculated using a nitrogen conversion factor of 6.25 and expressed in grams per 100 grams of wet weight.

2.3.5 Determination of crude fiber

The crude dietary fiber content was determined using a crude fiber extractor (AOAC, 2000). Each sample (2.0 g) was placed in a beaker, mixed with 200 mL of 1.25% H_2SO_4, and boiled in a fiber extractor. Then, 200 mL of distilled water was added and filtered using constant-weight filter paper. The extract was transferred to clean, dry, weighted crucibles. The crucibles were placed in an oven at 105°C for 8 hrs. The sample was cooled with a desiccator. The crude fiber content was calculated on a wet weight basis of g/100 g.

2.3.6 Histamine content

The histamine content in the CP was determined using a Histamine test kit-AOAC Performance Tested Method (Kikkoman Biochemifa, Japan) according to the AOAC (2000) standard.

2.4 Water absorption index and water solubility index analysis

Water absorption index (WAI) and water solubility index (WSI) analysis of the CP samples were determined following the method of Anderson *et al*. (1970). A 0.5 g CP sample was placed into a tube containing 15 mL of distilled water. The tube was then capped, placed in a water bath and shaken vigorously at 5 mins intervals for 30 mins at 30°C before centrifuging at 3000×*g* for 15 mins at 20°C. The supernatant was removed and dehydrated using a hot air oven at 100°C until reaching constant final moisture content. The tubes were drained before being reweighed. The WAI and WSI were calculated using the equations:

Water absorption index (WAI) = Weight of sediment/ Weight of dry solid

Water solubility index (WSI) = (Weight of dissolved solid in supernatant/ Weight of dry solid) × 100

2.5 Total viable bacterial count

A 5 g sample was mixed with 45 mL sterile PPS (peptone physiological salt, 1 g/L bacteriological peptone (Oxoid LP0034) and 8.5 g/L NaCl) in a sterile filter stomacher bag and homogenized at normal speed for 1 min. The homogenate was then diluted with PPS and the desired decimal dilutions were poured onto agar plates. Each CP sample was measured in triplicate. The plates were counted using a colony counter for each microorganism, with counts expressed as colony forming units per gram (CFU/g).

2.6 Antioxidant activity analyses

The CP samples were extracted following the

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method of Moongngarm *et al*. (2022) with slight modifications. A 5.0 g aliquot of each CP sample was extracted in 80% methanol (25 mL) by placing the mixture in a shaking water bath at room temperature for 2 hrs. The mixture was filtered and the residue was then subjected to the same procedure twice. The residue was extracted with 0.15 mol/L hydrochloric acid. The extracts were combined, filtered through a filter paper and evaporated to dryness under reduced pressure at 45°C by a rotary evaporator (Buchi, Switzerland). The extract samples were used for antioxidant activity and total phenolic content analyses.

2.6.1 Diphenyl-2-picrylhydrazyl assay

The antioxidant properties of all CP extracts were determined using the method of Dasgupta and De (2004) for free radical scavenging on DPPH radical. Each extract solution (100 µL) was mixed with 900 µL of DPPH solution (0.1 mM in methanol). The mixture was allowed to stand in the dark for 30 mins before measuring absorbance at 517 nm with a UV-Vis Spectrophotometer. Results were compared with 6-hydroxy-2,5,7,8-tetra-methylchromane-2-carboxylic acid (Trolox) standard and expressed as µg Trolox equivalent/ g sample.

2.6.2 ABTS radical scavenging assay

The free radical scavenging activity of extract samples was evaluated using the 2,2-azino-di-(3-ethylbenzothiazoline-sulphonic acid radical cation (ABTS•+) decolorization assay (Re *et al*., 1999). In brief, the ABTS•+ stock solution (7 mM) is obtained by combining 7 mM ABTS with 2.45 mM potassium persulphate as an oxidizing agent. The working solution of ABTS•+ was prepared by diluting the stock solution in phosphate-buffered saline (PBS). The sample solution (100 µL) was mixed with 900 µL of the ABTS•+ solution. Absorbance records at 405 nm were taken at room temperature 10 mins after initial mixing. Results were calculated and expressed in µg Trolox equivalent/g sample.

2.6.3 Ferric reducing antioxidant power assay

The ability of all CP extracts to reduce Fe^{3+}-TPTZ to blue-colored Fe^{2+}-TPTZ was used to determine its reducing power using the Benzie and Strain (1996) method. The FRAP reagent was initially prepared using 300 mM acetate buffer (pH 5.6), 10 mM 2,4,6-Tripyridyl -s-triazine (TPTZ) solution in 40 mM HCl, and 20 mM $FeCl_3.6H_2O$ solution in a 10:1:1 (v/v/v) ratio. In test tubes, 300 µL extracts were combined with 2.7 mL FRAP reagent using a vortex mixer. After 30 mins of incubation, the absorbance was measured at 539 nm using a spectrophotometer. Results were expressed in µmol $FeSO_4$ equivalent/g sample.

2.7 Total phenolic content analysis

The total phenolic content (TPC) of the extracts was determined using the Folin-Ciocalteu reagent according to the method of Iqbal *et al*. (2005). A 200 µL of extract samples were placed in test tubes. After adding 750 µL of fresh Folin-Ciocalteu reagent (diluted in distilled water (1:10)) and 750 µL of 6% (w/v) sodium carbonate (Na_2CO_3), the mixture was allowed to react for 90 mins at room temperature in the dark. The absorbance of the blue-colored mixture was measured at 765 nm using a spectrophotometer. Gallic acid was used to set up the standard curve and results were calculated as GAE/g sample.

2.8 Statistical analysis

Results were reported as mean values and standard deviations from triplicate samples of each treatment for all experiments. For each cricket species, a factorial design consisting of two factors blanching temperature and holding time was applied with three temperatures 80, 90 and 100°C and three holding times 5, 10 and 15 min. One-way analysis of variance (ANOVA) was used for data comparison between cricket species. Duncan's Multiple Range Test was performed to determine significant differences between treatments. Statistical significance was declared at $p<0.05$. All experiments were conducted in triplicate.

3. Results and discussion

3.1 Effect of blanching on chemical composition

Chemical compositions of CP samples prepared from *G. bimaculatus* and *A. domesticus* were influenced by blanching under different conditions, as shown in Table 1. Results indicated interaction between blanching temperature and time for CP fat from both cricket species ($p<0.05$). The protein content was significantly affected by blanching temperature and holding time ($p<0.001$). Increasing blanching temperature lowered fat and protein contents in CP but increased fiber content in CP obtained from *A. domesticus*. Compared to unblanched CP, significantly lower fat content was observed in all blanched CP samples but higher protein content. CP prepared from *G. bimaculatus* had higher fat content and lower protein, moisture and fiber contents compared to CP from *A. domesticus*. The outer structure of the crickets was destroyed by heat, and fat leaches into the blanching water, resulting in increased protein concentration in CP. Khatun *et al*. (2021) reported that freeze-dried *A. domesticus* powder samples had significantly ($p<0.05$) lower protein content compared with oven-dried and blanched powder samples, with

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Table 1. Chemical composition of two cricket powder samples obtained from different blanching conditions.

Temperature (°C)	Time (min)	Moisture content (g/100 g)	Fat (g/100 g)	Protein (g/100 g)	Ash (g/100 g)	Fiber (g/100 g)	Histamine (μg/g)
G. bimaculatus							
Unblanched		1.35±0.06^{c}	21.22±0.21^{a}	55.25±0.42^{c}	4.61±0.04^{b}	6.27±0.36^{fg}	<4
80	5	1.24±0.07^{c}	19.84±0.51^{b}	59.23±1.29^{b}	3.28±0.39^{d}	7.02±0.21^{f}	<4
	10	1.21±0.01^{c}	19.67±0.50^{b}	56.83±1.25^{b}	3.37±0.16^{d}	6.56±0.91^{f}	<4
	15	1.23±0.08^{c}	19.16±0.34^{b}	56.25±1.38^{b}	3.19±0.14^{d}	6.46±0.20^{f}	<4
90	5	1.22±0.01^{c}	19.15±0.34^{b}	54.87±0.50^{c}	3.48±0.21^{cd}	6.17±1.07^{fg}	<4
	10	1.26±0.05^{c}	19.06±0.39^{b}	51.65±1.31^{cd}	3.30±0.19^{d}	5.75±0.19^{g}	<4
	15	1.21±0.01^{c}	17.84±0.41^{c}	51.43±1.40^{cd}	3.47±0.21^{cd}	5.42±0.13^{g}	<4
100	5	1.20±0.02^{c}	17.66±0.63^{c}	50.76±1.40^{d}	3.28±0.30^{d}	5.27±0.98^{g}	<4
	10	1.21±0.01^{c}	15.41±0.01^{d}	49.44±1.11^{d}	3.17±0.22^{e}	4.96±0.61^{h}	<4
	15	1.20±0.01^{c}	12.57±0.22^{e}	48.95±0.24^{d}	3.35±0.12^{d}	4.74±0.51^{h}	<4
A. domesticus							
Unblanched		2.79±0.47^{b}	19.61±1.89^{b}	69.27±1.36^{a}	4.39±0.11^{b}	12.42±0.62^{e}	<4
80	5	4.37±0.15^{a}	11.90±0.33^{ef}	70.19±2.57^{a}	3.48±0.52^{d}	14.67±0.41^{d}	<4
	10	3.59±0.11^{ab}	11.50±1.10^{ef}	71.91±4.22^{a}	4.13±0.87^{bcd}	16.27±0.29^{ab}	<4
	15	3.34±0.28^{ab}	11.96±1.68^{ef}	72.99±1.14^{a}	5.37±0.32^{a}	16.62±0.33^{a}	<4
90	5	3.95±0.71^{a}	11.97±0.35^{ef}	72.02±1.12^{a}	4.52±0.08^{b}	15.23±0.41^{bcd}	<4
	10	3.77±0.60^{ab}	10.34±1.52^{f}	73.15±1.41^{a}	4.52±0.32^{b}	15.13±0.28^{cd}	<4
	15	4.21±0.18^{a}	12.92±1.92^{e}	73.82±6.03^{a}	4.05±0.59^{bcd}	15.14±0.21^{cd}	<4
100	5	4.61±1.60^{a}	11.37±2.63^{f}	69.72±0.76^{a}	4.36±0.16^{bc}	14.87±0.33^{d}	<4
	10	4.46±1.69^{a}	9.39±1.89^{g}	23.03±2.17^{e}	3.91±0.33^{bcd}	16.14±0.67^{abc}	<4
	15	4.46±0.26^{a}	10.46±3.60^{g}	21.82±3.24^{e}	3.65±0.15^{cd}	15.91±0.47^{b}	<4
G. bimaculatus							
Temperature		ns	***	***	***	***	-
Time		ns	***	***	ns	ns	-
Temperature*Time		ns	***	ns	ns	ns	-
A. domesticus							
Temperature		ns	***	***	***	***	-
Time		*	ns	***	ns	ns	-
Temperature*Time		***	*	*	ns	ns	-

Values are presented mean±SD of triplicate samples (n = 3) on a wet weight basis. Values with different superscripts within the same column are statistically significantly different ($p<0.05$). ^{ns}no significant difference ($p\geq0.05$), *significantly different at $p<0.05$, **significantly different at $p<0.01$, ***significantly different at $p<0.001$.

protein and lipids as the major components in CP. In a similar vein, Bawa *et al*. (2020b) and Montowska *et al*. (2019) reported that crickets contained high protein (55-75%), fat (12-29%) and fiber (3.5-7%) contents. Variation in CP chemical compositions was impacted by several factors such as feed substrate, life stage, species, sex and diet (Oonincx *et al*., 2015; Bawa *et al*., 2020b).

Histamine, a chemical found in insects causing food poisoning and allergic reactions related to insect consumption, was also determined, with results showing low levels (<4 μg/g) in both blanched and unblanched CP samples, meeting the food safety standard of histamine content in fishery products (<200 μg/g) (Commission Regulation (EC) No 2073/2005).

3.2 Effect of blanching on water absorption index and water solubility index

Results indicated that blanching temperature had a significant effect ($p<0.05$) on WAI and WSI of both CP products (Figure 1). WAI and WSI in CP from *A. domesticus* increased as the blanching temperature increased. CP blanched at 90°C and 100°C had significantly highest WAI and WSI, ranging 2.11-2.45 and 2.45-2.73%, respectively ($p<0.05$) while the lowest WAI and WSI values were observed in CP without blanching (0.15±0.01 and 1.07±0.01%, respectively). These values concurred with Lee *et al*. (2020) who recorded WAI values of freeze-dried and hot air-dried *G. bimaculatus* powder as 3.6 and 3.0 g/g, respectively. The highest WAI (8.75±0.02) was found in CP from *G. bimaculatus* at a blanching temperature of 90°C followed by blanching at 100°C (6.14±0.03) and 80°C (5.30±0.01). The two crickets differed in chemical components such as protein, fat and fiber (Table 1) and heating generated by blanching changed the structure of the components in cricket powder by protein denaturation and chitin carbohydrate gelatinization, which directly affected solubility (Gutkoski and El-Dash, 1999). Both CPs blanched at 90 and 100°C showed

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significantly increased WAI and WSI (p<0.05). Blanched CP containing higher protein and lower fat content (Table 1) showed increased water absorption and solubility. An increase in solubility was observed after defatting yellow mealworms compared with full-fat flour (Bubler *et al*., 2016). By contrast, Mishyna *et al*. (2021) reported that protein denaturation, unfolding and exposure of internal hydrophobic groups, caused by thermal treatments, resulted in a decrease in insect protein solubility. However, the solubility of insect proteins differed between insect species and was largely affected by the type of processing treatment and its intensity (Mishyna *et al*., 2021).

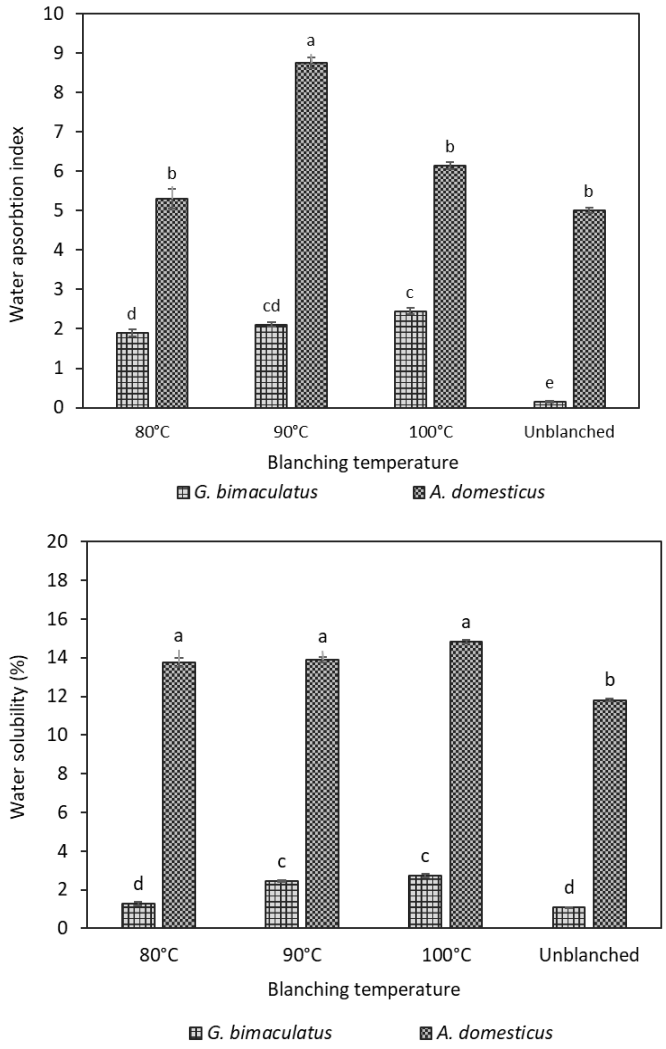

Figure 1. Water absorption index (WAI) and water solubility (WS) of cricket powder samples obtained from different blanching conditions. Bars with different notations are statistically significantly different (p<0.05). Error bars indicate the standard deviation.

3.3 Effect of blanching on total microbial count

Blanching temperatures and times had a significant effect (p<0.05) on CP microbial count (Figure 2). An increase in blanching temperature had a statistically significant effect on the reduction of CP total microbial count (p<0.05). CP blanched at 100°C showed the highest reduction of microbial count to 0.53 and 0.91 log CFU/g for *G. bimaculatus* and *A. domesticus*, respectively, while the highest microbial counts of 2.98 and 3.38 log CFU/g, respectively, were found in CP without blanching (control). Blanching at 80°C for 5 mins significantly reduced the microbial count compared with CP without blanching. In addition, the coliform bacteria and *Escherichia coli* were also determined in this study, however, no coliform bacteria and *E. coli* were found. Therefore, blanching as a pre-treatment effectively reduced the initial microbial count in CP, deactivated enzymes and maintained the quality of food products with extended shelf life. These findings concurred with Mancini *et al*. (2019a) who reported that blanching at 60°C for 5 mins successfully reduced phenoloxidase in mealworm larvae. Blanching heat treatment also reduced microbial loads and increased product safety (Fellows, 2009; Wang *et al*., 2017). Mancini *et al*. (2019a) showed that blanching at 60°C for 5 mins or at 90°C for 2.5 mins reduced the microbial population in mealworm larvae. Reductions of bacterial counts in *Alphitobius diaperinus* and *Tenebrio molitor* larvae were achieved by blanching at 88°C for 5 mins and at 100°C for 10, 20 and 40 s, respectively (Vandeweyer *et al*., 2017; Wynants *et al*., 2018). By contrast, blanching at lower temperatures did not reduce microbial loads or stop the browning effect, while higher temperatures did not improve product quality and microbiological parameters (Mancini *et al*., 2019b). However, both cricket powders (<4 log CFU/g) were safe for human consumption according to the limits established by the Thai Industrial Standards Institute (TISI) 1337/2549 (TISI, 2006).

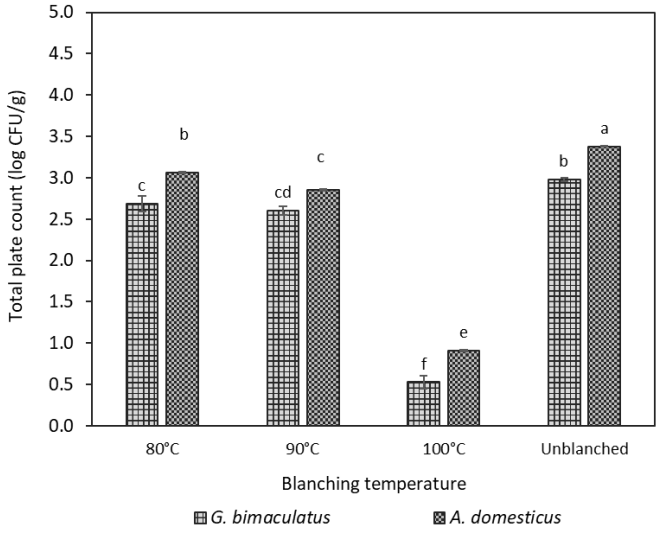

Figure 2. Total plate count of cricket powder samples obtained from different blanching conditions. Bars with different notations are statistically significantly different (p<0.05). Error bars indicate the standard deviation.

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Table 2. Antioxidant activity of cricket powder samples obtained from different blanching conditions.

Temperature (°C)	Time (min)	Antioxidant activities		
		DPPH (μg Trolox/g)	$ABTS^+$ (μg Trolox/g)	FRAP (μM $FeSO_4$/g)
G. bimaculatus				
Unblanched		74.39±1.23^{b}	20.40±2.47^{f}	49.58±0.93^{ab}
80	5	85.07±3.52^{a}	22.98±1.33^{e}	51.13±1.12^{a}
	10	84.14±3.63^{a}	21.87±1.59^{ef}	50.55±3.90^{a}
	15	84.45±1.60^{a}	21.93±1.83^{ef}	47.53±3.36^{ab}
90	5	78.03±1.47^{b}	21.22±2.71^{ef}	47.68±0.37^{ab}
	10	74.57±1.15^{b}	22.46±2.66^{ef}	47.22±3.79^{ab}
	15	71.12±1.95^{c}	20.22±2.87^{f}	43.26±1.39^{bc}
100	5	75.22±3.26^{b}	21.05±3.46^{ef}	45.47±1.47^{bc}
	10	74.57±3.73^{b}	18.29±2.40^{g}	43.46 ±1.66^{bc}
	15	70.17±1.61^{c}	18.23±2.13^{g}	46.02±2.34^{c}
A. domesticus				
Unblanched		71.40±4.24^{c}	51.21±3.03^{b}	24.33±1.52^{e}
80	5	87.36±0.62^{a}	56.13±1.44^{a}	27.85±1.83^{d}
	10	86.07±0.27^{a}	52.18±4.33^{ab}	27.15±1.54^{d}
	15	82.20±3.88^{a}	49.12±2.09^{b}	24.04±3.26^{e}
90	5	82.08±3.53^{a}	54.12±1.21^{ab}	29.28±1.43^{a}
	10	85.43±0.27^{a}	49.00±1.98^{cd}	27.62±0.54^{d}
	15	70.99±2.12^{b}	48.36±1.17^{cd}	26.15±2.99^{d}
100	5	75.54±0.83^{b}	46.95±3.10^{cd}	26.33±3.23^{d}
	10	72.55±4.53^{c}	46.56±1.64^{d}	23.16±3.81^{ef}
	15	70.52±2.64^{c}	47.95±1.28^{cd}	22.16±1.17^{f}
G. bimaculatus				
Temperature		***	***	***
Time		***	***	***
Temperature*Time		***	***	***
A. domesticus				
Temperature		***	***	***
Time		***	***	***
Temperature*Time		***	***	***

Values are presented mean±SD of triplicate samples (n = 3) on a wet weight basis. Values with different superscripts within the same column are statistically significantly different (p<0.05). *significantly different at p<0.05, **significantly different at p<0.01, ***significantly different at p<0.001.

3.4 Effect of blanching on antioxidant activity

Table 2 shows the interaction between blanching temperatures and times on antioxidant activity in terms of DPPH, ABTS•+ and FRAP assays (p<0.05) in all CP samples. Overall, CP antioxidant activity gradually decreased as blanching temperature and time increased. CP blanched at 80°C for a short time (5-10 mins) exhibited significantly stronger antioxidant properties in DPPH•, ABTS•+ and FRAP assays (p<0.05). CP from *G. bimaculatus* ranged 84.14-85.07 μg Trolox/g, 21.87-22.98 μg Trolox/g and 50.55-51.13 μM $FeSO_4$/g, respectively and 86.07-87.36 μg Trolox/g, 52.18-256.13 μg Trolox/g and 27.15-27.85 μM $FeSO_4$/g, respectively for *A. domesticus*. CP from *G. bimaculatus* had a significantly higher FRAP value and lower ABTS+ (p<0.05) compared to CP prepared from *A. domesticus*. Interestingly, blanching cricket at a suitable temperature/ time enhanced antioxidant activity because blanching damaged the cricket tissue structure and cell wall porosity resulting in antioxidant release. However, these effects depended on the blanching conditions used such as heating levels and blanching time. Anuduang *et al*. (2020) also reported that blanching silkworm pupae at optimal temperature increased antioxidant capacity compared to unblanched silkworm pupae. A suitable blanching process (temperature and time) can destroy the enzyme causing oxidation (Gunathilake *et al*., 2018);

https://doi.org/10.26656/fr.2017.8(6).239

however, using high temperatures and a long time (temperature 100°C, 15 mins) resulted in reduced antioxidant capacity of CP for all assays due to leaching of antioxidants into the boiling water (Kao *et al*., 2014).

3.5 Effect of blanching on total plate count

Statistical analysis results indicated an interaction effect between blanching temperatures and times on TPC (p<0.05) in all CPs, as presented in Figure 3. TPC decreased as blanching temperatures and times increased. CPs after blanching at 80°C for 5-15 mins and 90°C for 5-10 mins showed significantly highest TPC (p<0.05), ranging 258.4-266.3 mg GAE/g for *G. bimaculatus* comparable to CP without blanching. For *A. domesticus*, the highest TPC (142.3-150.0 mg GAE/g) was observed after blanching at 80°C for 5-10 mins, and higher than in CP without blanching (137.5 mg GAE/g). However, higher TPC was observed in CP from *G. bimaculatus* compared to CP prepared from *A. domesticus*. Similarly, the lowest TPC was observed in CP blanched at 100°C for 15 mins, as 162.5 and 91.2 mg GAE/g for *G. bimaculatus* and *A. domesticus*, respectively. Phenolic acids are destroyed by heat during the blanching process, resulting in decreased TPC. Similarly, Anuduang *et al*. (2020) reported that phenolic compounds in silkworm pupae decreased after immersion in hot water (90°C). Increasing heating times caused phenolic acids to leach into the blanching water (Kao *et al*., 2014). Enzymes play roles in cricket powder browning, such as phenoloxidase, laccase, tyrosine hydroxylase, decarboxylase and peroxidase (Andersen, 2012). Most phenolic compounds in insects are derived from l-tyrosine through the shikimic acid pathway using enzymatic reactions. Phenolic compounds can react with proteins and amino acids forming dark compounds (melanins). Janssen *et al*. (2017) reported enzymatic browning in three insect species (*Tenebrio molitor*, *Alphitobius diaperinus* and *Hermetia illucens*); however, suitable blanching temperature and time deactivated the enzymes responsible for oxidation, thereby increasing TPC in CP from *A. domesticus*. Most phenolic compounds in insects are derived from l-tyrosine through the shikimic acid pathway using enzymatic reactions. Phenolic compounds can react with proteins and amino acids forming dark compounds (melanins).

4. Conclusion

Results demonstrated that blanching affected CP quality in both species. CP blanched at 80°C for 10 mins had higher protein and fiber content compared to unblanched CP. The highest TPC and antioxidant activities in all assays (DPPH, ABTS•+ and FRAP) were observed under the same condition. Compared to unblanched CP, blanching also reduced significantly the total microbial load in both species. The findings suggested that blanching (at the right temperature and time) as a pre-treatment enhanced the safety and functionality of edible cricket powder.

Conflict of interest

The authors declare no conflict of interest.

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