INSECTS AS INGREDIENTS FOR BAKERY GOODS. A COMPARISON STUDY OF *H. illucens*, *A. domestica* and *T. molitor* FLOURS

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Abstract

Due to a rising demand for proteins, food industry is considering new alternative protein sources that can be used for human food. The aim of this research was to explore the potential use of insects' flour as protein-rich ingredient for bakery products. *Hermetia illucens*, *Acheta domestica* and *Tenebrio molitor* were ground and used to replace 5% wheat flour in doughs and breads. The protein content of the insect flours ranged from 45% to 57% (d.m) and fat content from 27% to 36% (d.m.). The inclusion of insects' flour affected the rheological properties (water absorption and stability), of dough during mixing, having less water adsorption. Breadmaking process could be carried out with all the composite flours. Breads containing *A. domestica* flour showed similar specific volume and texture parameters than wheat bread, but with higher content of proteins and fibers. Globally, results confirmed the usefulness of insects' flour for making breads with improved nutritional value.

Industrial Relevance

This study evaluated the potential application of three different insects as protein source ingredients for bakery products. Results confirm that insects flour could be added to replace wheat flour in breads without significantly affecting dough properties and

leading to breads with acceptable technological quality and improved nutritional profile.

Keywords: insects, proteins, bread, dough, rheology, flour.

1. Introduction

Considering the rising population worldwide and the increasing demand for additional sources of proteins, insects are seen as an economical alternative (Ryan, 2014). Entomophagy, the eating of insects, has been part of the world habits for centuries. Only recently, it has gained interest in Western countries (Balzan, Fasolato, Maniero, & Novelli, 2016). In Europe, certain insect species are reckoned for partial replacement of conventional sources of animal proteins in food and feed (Rumpold & Schluter, 2013 a). The most commonly eaten insects are members of the *Coleoptera* (beetles), *Lepidoptera* (caterpillars of butterflies and moths), *Hymenoptera* (bees, wasps, ants), *Orthoptera* (grasshoppers, locusts, crickets, termites), *Hemiptera* (cicadas, leaf and plant hoppers, true bugs, scale insects), *Odonata* (dragonflies) and *Diptera* (flies) families, although crickets, locusts and mealworms (larval form of the mealworm beetle) are the most commonly farmed (Sogari, 2015). A main advantage of insects is that they have high food and feed conversion efficiency and their feed can be limited to industrial by-products and cereal grains (van Broekhoven, Oonincx, van Huis, & van Loon, 2015). Likewise, the production of particular insects is more sustainable than conventional sources of proteins, considering water use and greenhouse gas emissions (van Zanten, Mollenhorst, Oonincx, Bikker, Meerburg, & de Boer, 2015). All those environmental advantages have prompted a growing interest for edible insect, because of that industrial farming of domesticated insects has been quickly expanding, ensuring consistent quality, hygienic production and cost effectiveness, which allows greater security for human consumption (H. W. Kim, Setyabrata, Lee, Jones, & Kim, 2017).

Despite the sustainable benefits, western consumers could be reluctant to accept insects as a legitimate protein source because they have never played a significant role in their food culture (Balzan, Fasolato, Maniero, & Novelli, 2016). However, people would be willing to eat them in a less visible form in modified products indistinguishable from familiar ones (Schosler, de Boer, & Boersema, 2012). Certain insect species could be targeted for being incorporated as a powder in food products, providing additional nutrients. Besides, the new Novel Foods Regulation (regulation 2015/2283), which entered into force on January 2018, has given the green light for every insect-based foodstuff by the European Food Safety Authority (EFSA), after which it can legally be sold in all EU Member States. It has been reported that insects contain around 35-61%, 15-40% and 3-10% of proteins, fats and minerals, respectively (Rumpold & Schlüter, 2013 b). Currently, many commercial food products are enriched with proteins derived from other cereal grains such as rye or oats, or legumes (Al-Attabi, Merghani, Ali, & Rahman, 2017). Likewise, insects are richer in protein than beans (23.5%), lentils (26.7%) or soybean (41.1%) (Zielinska, Baraniak, Karas, Rybczynska, & Jakubczyk, 2015).

In spite of the mentioned advantages, up to now very few applications of insects flours in foods have been reported. A very recent proposal has been the inclusion of insect's flours from mealworm larvae and silkworm pupae to replace 10% lean pork in emulsion sausages, which increased the cooking yield and hardness of emulsion sausages, confirming the possible application of those insects flours as a novel protein ingredient (H.-W. Kim, Setyabrata, Lee, Jones, & Kim, 2016). Nevertheless, in cereal based products, to authors knowledge, only maize tortilla has been enriched in proteins by adding larva of the *Tenebrio molitor*, bringing about tortillas supplemented with larvae powder which protein content increased by 2% (Aguilar-Miranda, Lopez, Escamilla-

Santana, & de la Rosa, 2002). The aim of this research was to study the incorporation of flours from three different insects into wheat bread, by following dough behavior and bread quality regarding technological properties and chemical composition. *Hermetia illucens* (Diptera, Black Soldier Fly larvae) and *Tenebrio molitor* (Coleoptera, yellow mealworm), and adult *Acheta domestica* (Orthoptera, crickets) were the insects selected.

2. Material and methods

2.1 Material

Commercial bread-making flour was supplied by Harinera La Meta (Barcelona, Spain). Salt and compressed yeast were purchased from the local market. *Hermetia illucens* larvae were provided by Bioflytech (Alicante, Spain) and *Acheta domestica* or house cricket flour and mealworm beetle, *Tenebrio molitor* were provided by Insect side (Alicante, Spain). Some adults of *A. domestica* were also provided for the morphological characterization. Larvae samples were frozen at -40 °C and freeze dried in a VirTis Genesis 35EL (SP Scientific, Gardiner, NY), under vacuum conditions of 8 mbar. Then, they were milled in an IKA M20 (Wilmington, USA). The powder was kept at -20 °C until used.

2.2 Insects characterization: morphology and composition

Insect images were captured by an HP 6 Scanjet G3110 Flatbed Scanner (Hewlett-Packard, USA) with a resolution of 600 dpi. Images were analyzed by ImageJ program (National Institutes of Health, Bethesda, MD, EEUU) and area, width and height of insects' central body were determined.

Proximate analysis of insect, wheat flours and bread samples was determined according to AACCI International (2012) standard procedures, which were adapted whenever

required for these new food matrices. Analysis included moisture (method 44-15A), ash (method 08-01), fat (method 30-25), crude protein (Kjeldahl method) using $N \times 6.25$ and dietary fiber (method 32-07.01). Non-protein nitrogen was determined after precipitating the proteins with TCA (Trichloroacetic acid) and then evaluated using the Kjeldahl method. Carbohydrates were determined by difference $[100 - (\text{protein} + \text{fat} + \text{ash})]$ and expressed as percentage.

Insect chitin was estimated in the insects defatted flours, following the method of Black and Schwartz (1950) with some modifications. Briefly, method involved two important steps: demineralization and deproteinization.

Demineralization. In order to remove catechols, samples were treated with 1 M HCl at 85-90 °C for 50 min under constant stirring. The ratio of raw sample to acid solution during the extraction was 1/20 (w/v). Then, it was centrifuged at 3,000 g for 5 min and washed with distilled water to remove the excess of HCl.

Deproteinization. Sediment from the previous step was suspended 1/20 (w/v) in alkali (1 M NaOH) and kept at 85-90 °C for 35 min under constant stirring to remove proteins completely. The mixture was vacuum filtrated in a Buchner funnel with filter paper (pore size 20-25 μm), washed several times with deionized water to remove the excess of NaOH, and then dried in an oven at 100 °C overnight. The residue obtained was designated as a purified insect chitin in the form of a very light brown powder and calculated by weight.

2.3 Rheological analysis of dough

Mixing behavior of doughs obtained from wheat flour and blends were studied using the Mixolab (Chopin, Tripette et Renaud, Paris, France). It measures in real time the torque

(expressed in Nm) produced by passage of dough between the two kneading arms, hence allowing the study of its rheological behavior (Rosell, Collar, & Haros, 2007). For the assays, 50 g of wheat flour were placed into the Mixolab bowl and mixed with the amount of water needed to reach goal consistency (1.1 Nm). When insect flours were tested, 5% wheat flour was replaced by insect flours. The specific protocol (Mixolab Standard) used for dough testing consisted of 30 min mixing at 30 °C and the mixing speed during the entire assay was 80 rpm. Two batches were carried out for each sample. Parameters recorded from the mixolab curve included: water absorption or the amount of water required to reach the consistency of 1.1 Nm (expressed as milliliters per 100 g of flour at 14.0 % mass fraction moisture content), dough development time or time to reach the maximum consistency, and stability of the dough during mixing that indicates the elapsed time at which dough kept the maximum consistency.

2.4 Breadmaking

The following recipe on flour basis was used: 100% of wheat flour, 1.5% of salt, 2% of compressed yeast and the amount of water corresponding to the water adsorption obtained from Mixolab. Whenever present the insect flour, wheat flour was replaced up to 5% level. After 5 min mixing, dough was divided in 50 g portions and placed into a tray of silicon pans. Pans were leavened in a proofing chamber at 30 °C for 90 min and then baked in an electric oven (F106, FM Industrial, Córdoba, Spain) at 185 °C for 20 min. Breads were left cooling down at room temperature until reaching 25 °C in the center of the loaf before slicing them into 10-mm thickness.

2.5 Bread characterization

Breads were evaluated by assessing technological properties (volume, texture and color) and proximate composition. Volume was determined by the rapeseed displacement method (AACC International Method 10–05). Central slices (10 mm thick) were subjected to the texture profile analyses (TPA) using a TA.XTPlus Texture Analyzer equipped with a 5 kg load cell (Stable Micro Systems Ltd., Godalming, UK). Slices were placed in the middle of the base plate and then compressed with P/25 probe. During the test, the probe double compresses the center of the crumb at a crosshead speed of 1 mm/s and 30 s gap between compressions, providing insight into how samples behave when chewed. The compression strain was 60% (penetration of its original height) and the averages of at least ten analyses were calculated. Hardness, springiness, cohesiveness, chewiness and resilience were recorded from the TPA measurement. Crumb colors were measured using the CIE- $L^*a^*b^*$ uniform color space by the means of a Minolta colorimeter (Chromameter CR-400/410, Konica Minolta, Tokyo, Japan) (D65 illuminant and 10° viewing angle) after standardization with a white calibration plate ($L^* = 96.9$, $a^* = -0.04$, $b^* = 1.84$). Measurements were carried out with a 30 mm diameter diaphragm inset with optical glass. Color parameters indicate: L^* the lightness, a^* the hue on a green (-) to red (+) axis and b^* the hue on a blue (-) to yellow (+) axis (Matos & Rosell, 2012). Three measurements were made at different points in each crumb. The results were the average of four slices from each batch. Proximate composition of breads was determined as previously described.

2.6 Statistical analyses

Experimental data were subjected to an analysis of variance (ANOVA) using Statgraphics Centurion XV (Statistical Graphics Corporation, city, UK). Statistical

analyses were carried out with Fisher's least significant differences test with a significance level of 0.05. All measurements were performed at least in triplicate.

3. Results and Discussion

3.1 Insects morphology and proximate composition

Edible insects used as a source of proteins to fortify breads were morphologically and chemically characterized (Figure 1. **Insects morphology. a) *H. illucens*; b) *A. domestica*; c) *T. molitor*.**

, Table 1). Area, width and height were measured. *H. illucens* and *T. molitor* were used as larvae stage, while *A. domestica* was in adult stage. As it was expected, *A. domestica*, commonly known as house cricket, had the greatest area (95.28 mm²) compared to the larvae forms of *H. illucens* (56.79 mm²) and *T. molitor* or mealworms (30.04 mm²). Larvae from *H. illucens* showed a golden brown exoskeleton that curls into a "C" shape, which was 13.79 mm long and 3.86 mm thick. Larvae from *T. molitor* displayed a darken color with a yellowish brown shell that measures 14.26 mm x 1.74 mm (length x width). *A. domestica* showed a brown-black color with a size around 19.12 mm length and 3.86 mm width.

Proximate composition (expressed in dry basis) of wheat flour and the resulting flour after insects grinding is summarized in Table 1. Due to insects could be used as an alternative protein source, to avoid an overestimation in the protein content, the non-protein nitrogen (NPN) was determined and subtracted to the total nitrogen for the quantification of protein content in whole insect. Insects' flours showed a more complete and rich nutritional profile than the wheat flour, with exception of carbohydrates. Flours from insects were rich in proteins, followed by fat. However, there were significant differences among insects, specifically flour from *A. domestica* had the highest content of proteins and *H. illucens* flour had the highest fat content.

Flours from larvae showed a carbohydrate content that ranged between 14.84% to 16.24%, and the *A. domestica*, in adult state, had lower content of carbohydrates. Components like carbohydrates are frequently referred as nitrogen-free-extract (Finke, 2002). Non-protein nitrogen in insects is related to the presence of chitin, nucleic acids, phospholipids and excretion products in the intestinal tract (Janssen, Vincken, van den Broek, Fogliano, & Lakemond, 2017). The content of NPN in the present study agrees with the values of chitin nitrogen (or nitrogen from chitin structure) reported by Janssen, Vincken, van den Broek, Fogliano, and Lakemond (2017). Thus, it seems that TCA treatment precipitated proteins and other nitrogen containing compounds leaving just the nitrogen from chitin. There were no significant differences in the chitin content of *A. domestica* and *T. molitor*, and *H. illucens* showed the lowest content (Table 1). Kaya, Baran, Erdogan, Menten, Ozusaglam, and Cakmak (2014) and Kaya, Sofi, Sargin, and Mujtaba (2016) studied the physicochemical properties of chitin at different developmental stages of *Vespa crabro* (wasp) and potato beetle, observing that chitin content increased gradually as the organism grew, in both cases. Therefore, *A. domestica*, which was expected to have greater chitin content due to its adult state, had an intermediate content. Hence, the barely differences observed in the chitin content of the three flours, regardless their development state, might be ascribed to the type of insect and taxonomy differences. Zielinska, Baraniak, Karas, Rybczynska, and Jakubczyk (2015) has reported that the main components in insects are proteins and fats, followed by fiber, non-protein-nitrogen and ash, although those in no particular order. Nevertheless, it must be stressed that composition depends on the type of species and growing stages, but also it varied due to diverse feeding and origin, as well as modifications in measuring methods (Chen, Feng, & Chen, 2009).

3.2 Dough rheological properties

Figure 2 illustrates the plots recorded during mixing in the Mixolab. The inclusion of insects' flours affected the rheological behavior of the dough during mixing. The presence of insect flour up to 5% (basis flour) produced a decrease in water absorption compared with the control containing only wheat flour ($58.80 \pm 0.12\%$), obtaining $56.90 \pm 0.10\%$, $58.20 \pm 0.15\%$ and $57.70 \pm 0.17\%$ when containing *H. illucens*, *A. domestica* and *T. molitor* flours, respectively. Generally, the addition of flours with high amount of hydrophilic proteins increases the hydration and water adsorption of dough, which has been observed with soybean flours (Lazo-Vélez, Chuck-Hernandez, & Serna-Saldívar, 2015). In addition, studies carried out replacing wheat flour with increasing amounts of barley flour showed a decrease in water absorption due to protein content reduction (Al-Attabi, Merghani, Ali, & Rahman, 2017). Insect flours do not contain starch and despite their high proteins content water adsorption was reduced. Presumably the amino acids composition of the proteins (Rumpold & Schlüter, 2013 b) were responsible of lowering the water absorption. Dough development time was slightly increased when present *A. domestica* or *H. illucens* (2.5 ± 0.2 min) compared to the control and *T. molitor* (2.0 ± 0.1 min). Stability also increased with doughs containing *A. domestica* (4.4 ± 0.3 min) or *H. illucens* flours, particularly in the presence of *H. illucens* (7.5 ± 0.3 min vs 1.1 ± 0.2 min in control). Dough development time and stability values are indicators of the flour strength, where higher values suggesting stronger doughs (Wang, Rosell, & Benedito de Barber, 2002). The high dough stability observed with the presence of *H.illucens* was initially attributed to the high fat content, but when deffated flour was tested (Figure 2), no significant differences were observed in the consistency plot. Again, proteins nature and composition should be responsible of the doughs stabilities.

3.3 Breads enriched with insects' flours

Breads containing insects' flours showed similar appearance than control bread (Figure 3), with exception of crumb color, which showed different brownish intensities. The addition of insect's flour did not impair dough fermentation, obtaining breads with acceptable volume and open crumbs. Nonetheless, the bread containing *H.illucens* exhibited a more closed crumb and lower volume. It must be stressed that during baking of breads containing *H.illucens* flour some off-flavors were released, likely due to its fat composition. Generally, the quantities of food flavor are associated more closely with lipids, mainly unsaturated lipids, than with proteins and carbohydrates. Spranghers, Ottoboni, Klootwijk, Oryn, Deboosere, De Meulenaer, et al. (2017) reported that the fatty acid profile from *H. illucens* feed with different waste comprised a pretty high content of unsaturated fatty acids, which could be related to the off-flavors smelled during baking.

Regarding the moisture content of breads, *H. illucens* had the highest percentage. Considering that the crust thickness of the breads was similar, a plausible explanation for this result could be that the high content of fat prevented the evaporation of water during the baking process. In fact, this assumption was confirmed by making breads with defatted *H. illucens* flour, which had 29.59% moisture content. The presence of the three insect flours affected the specific volume of bread formulations (Table 2). Breads containing insect' flours had significantly lower specific volume in comparison with the wheat control, with the exception of the bread containing *A.domestica* that had similar specific volume than the control. Different causes have been related to volume reduction, like the addition of fibers (Wang, Rosell, & Benedito de Barber, 2002) or proteins derived from different legume flours such as lentils, apart from gluten dilution

(Defloor, Nys, & Delcour, 1993). Commonly, this is attributed to reduce extensibility and weakening of the gluten network owing to dilution, reduced hydration and interactions with non-starch carbohydrates and non-gluten proteins, which also reduces the gas-retention ability. In the present case, first assumption was that the upper content of fat could be the cause, especially in *H. illucens* that presented the highest fat content, but breads made with defatted flour had similar specific volume, discarding the fat role in volume decrease.

The texture parameters including hardness, springiness, cohesiveness, chewiness and resilience of the different breads, and also specific volume, color and moisture are presented in Table 2. Breads enriched with *H. illucens* showed significant differences in all the texture parameters, having significantly harder crumb and lower springiness, cohesiveness and resilience. The lower springiness, indicative of the crumbling tendency when bread is sliced, agrees with the low cohesiveness that reflects the low internal cohesion within the crumb. Likewise, crumbs with lower number of gas cells take longer for recovering their structure after compression (Matos & Rosell, 2012). Moreover, this bread showed the highest chewiness, revealing that longer time was required for masticating a bread piece prior to swallow. No significant differences were found between the texture parameters obtained for *A.domestica* and *T. molitor* containing breads. Again, although the high amount of fat in *H.illucens* could be the cause of the texture results, breads made with defatted flour had similar texture parameters, with the exception of hardness and chewiness, bringing about softer crumbs. Therefore, the composition of *H. illucens* flour, likely regarding proteins and carbohydrates, might be responsible for interfering in the proper development of the dough during the fermentation, provoking poor gas retention ability of the resulting weak gluten networks, yielding a tight structure of bread crumb (Defloor, Nys, &

Delcour, 1993; Sui, Zhang, & Zhou, 2016). Bread making is sensitive to the substitution of wheat flour by non-gluten, non-starch flour in particular through the disruption gluten development (Villarino, Jayasena, Coorey, Chakrabarti-Bell, & Johnson, 2016). Present results agrees with the effects produced after the use of wholegrains non-wheat flours (Koletta, Irakli, Papageorgiou, & Skendi, 2014).

The color is an important feature for baked products, because together with texture and volume, they influence consumer acceptance. Overall, all formulations with insect flours led to breads with reduced luminosity (L^*), increased redness (a^*) and yellow tonality (b^*). The color changes that took place were attributed to the cooking time, temperature and the presence of insect flours. Usually the color of baked products is directly dependent on the color of the raw materials used. The color of the insects' flours was measured to find possible correlation with crumbs color. CIEL $^*a^*b^*$ of *H. illucens* (43.81, 3.38, 16.45), *A. domestica* (39.32, 5.06, 11.62) and *T. molitor* (32.38, 5.01, 6.91) flours were not directly related to the color parameters of the crumbs, suggesting that breadmaking conditions during mixing, proofing and baking affected flour constituents. Nonetheless, as it was observed in Figure 3, the presence of *H. illucens* gave the most brownish crumbs, which agrees with the highest value of b^* observed in its flour and in the bread. Similar crumb colors have been previously obtained using flours that resembled in color with insect flours, like lentils and beans (Kohajdova, Karovicova, & Magala, 2013) or chickpea (Gomez, Oliete, Rosell, Pando, & Fernandez, 2008; Mohammed, Ahmed, & Senge, 2012).

3.4 Nutritive value of composite breads

The addition of insects' flour affected the nutritional composition of breads (Table 3). They produced statistically significant changes compared to the control, increasing its nutritional value specially regarding protein content and fat content. Breads containing *A.domestica* flour showed the highest content in proteins, which was expected since it had the major percentage in the flour. It was a significant attenuation in the total carbohydrate content of bread and also a significant increase in the total mineral content compared to the control bread. On the other hand, the TDF in enriched breads was higher than the content of the control. All insect flours provided IDF and SDF, although their contribution to the soluble digestible fiber fraction was more noticeable. There was a marked difference in IDF in the sample containing *T. molitor* comparing with the rest samples. There are great divergences in the reported content of fiber in insects, which ranged from 5% to 25% crude fiber, depending on the species and their developmental stages (Rumpold & Schlüter, 2013 b). According to the results, the inclusion of 5% *A. domestica* flour into wheat bread provided an increase in the protein content and dietary fiber, mainly of soluble nature, and only with slight increase in fat. Up to now, legumes (Mohammed, Ahmed, & Senge, 2012; Ouazib, Dura, Zaidi, & Rosell, 2016) and pseudocereals (Alvarez-Jubete, Arendt, & Gallagher, 2009; Collar & Angioloni, 2014) have been the main source of flours when searching for nutritional enrichment of bread, and lately also vegetables (Ranawana, Campbell, Bestwick, Nicol, Milne, Duthie, et al., 2016), even when they were added in small amounts. However, results obtained in the present study show the alternative of insects to be used as ingredient in baked goods.

Conclusions

Edible insects in the form of flour could be incorporated in baked goods to improve their nutritional pattern, principally regarding protein content. Insects flour composition

was dependent on the stage and taxonomic order, but they were rich in proteins and fats. Wheat flour replacement (5%) by insects flours from *H. illucens*, *A. domestica* and *T. molitor* affected the rheological properties of dough during mixing, requiring lower water adsorption and increasing dough stability in the case of *H. illucens* and *A. domestica*. Among the insects tested, *A. domestica* was the one that led to breads with technological features similar to wheat breads, but improved nutritionally regarding proteins and fiber content. Overall, results confirm the usefulness of insects' flour for protein enrichment of bread. However, further research will be undertaken to understand the interaction between cereals and insects flours and to modulate insects flour composition since a reduction in the fat content would be advisable for better nutritional balance of breads.

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FIGURE CAPTIONS

Figure 1. Insects morphology. a) *H. illucens*; b) *A. domestica*; c) *T. molitor*.

Figure 2. Mixolab curves of Control with wheat flour (black solid line), *H. illucens* (black discontinuous line), *H. illucens* (grey discontinuous line), *A. domestica* (black dotted line) and *T. molitor* (solid grey line) insect flours.

Figure 3. Cross section of breads obtained by replacing 5% wheat flour with different insect's flours.

490 **Table 1.** Nutritional composition of Wheat and Insect flours.

Samples	Wheat	<i>H. illucens</i>	<i>A. domestica</i>	<i>T. molitor</i>
Protein	12.69 ± 0.60 ^a	45.09 ± 0.82 ^b	56.58 ± 0.86 ^d	48.82 ± 0.76 ^c
NPN	- ± -	0.52 ± 0.02 ^a	0.66 ± 0.00 ^b	0.85 ± 0.07 ^c
Fat	1.19 ± 0.02 ^a	35.82 ± 0.66 ^d	27.08 ± 0.72 ^b	30.69 ± 0.80 ^c
Ash	0.64 ± 0.01 ^a	4.25 ± 0.00 ^c	4.02 ± 0.01 ^b	4.25 ± 0.01 ^c
Carbohydrates	85.57 ± 0.58 ^d	14.84 ± 0.35 ^b	12.33 ± 0.41 ^a	16.24 ± 0.63 ^c
Chitin	- ± -	3.52 ± 0.22 ^a	4.46 ± 0.44 ^b	4.73 ± 0.51 ^b

491 Means in rows followed by different letters are significantly different ($P<0.05$). Proximate values are expressed in g/100g dry matter basis. NPN:
 492 non protein nitrogen.

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495 Table 2. Effect of insect flour (5%) on textural and color parameters, moisture content and specific volume of breads.

	Wheat			<i>H. illucens</i>			<i>H. illucens defatted</i>			<i>A. domestica</i>			<i>T. molitor</i>		
Moisture (%)	31.73	±	1.37 ^a	35.10	±	1.24 ^b	29.59	±	1.18 ^a	30.89	±	1.15 ^a	32.06	±	1.15 ^a
Specific Volume (ml/g)	3.54	±	0.05 ^c	2.03	±	0.14 ^a	2.29	±	0.24 ^a	3.46	±	0.15 ^c	3.07	±	0.13 ^b
Texture Parameters															
Hardness (N)	3.24	±	0.65 ^a	28.39	±	4.16 ^c	12.50	±	2.48 ^b	3.99	±	0.46 ^a	3.62	±	0.23 ^a
Springiness	1.12	±	0.13 ^b	0.84	±	0.03 ^a	0.76	±	0.07 ^a	1.06	±	0.05 ^b	1.06	±	0.19 ^b
Cohesiveness	0.91	±	0.02 ^c	0.71	±	0.01 ^a	0.73	±	0.03 ^a	0.86	±	0.01 ^b	0.87	±	0.01 ^b
Chewiness (N)	3.15	±	0.85 ^a	17.67	±	2.78 ^c	6.75	±	1.60 ^b	3.70	±	0.38 ^a	3.21	±	0.45 ^a
Resilience	0.49	±	0.02 ^b	0.19	±	0.01 ^a	0.19	±	0.02 ^a	0.47	±	0.01 ^b	0.48	±	0.01 ^b
Color															
<i>L</i> *	66.51	±	1.43 ^c	57.22	±	1.56 ^a	57.79	±	0.82 ^a	60.19	±	1.06 ^b	60.66	±	1.14 ^b
<i>a</i> *	-0.67	±	0.12 ^a	3.38	±	0.18 ^c	3.22	±	0.15 ^c	1.18	±	0.24 ^b	1.15	±	0.23 ^b
<i>b</i> *	14.64	±	0.65 ^a	21.26	±	0.28 ^c	21.08	±	0.17 ^c	16.92	±	0.58 ^b	17.17	±	0.41 ^b

496 Means in rows followed by different letters are significantly different ($P<0.05$).

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Table 3. Nutritional composition of breads obtained by replacing 5% wheat flour with diverse insects' flour.

Samples	Wheat	<i>H. illucens</i>	<i>H. illucens deffated</i>	<i>A. domestica</i>	<i>T. molitor</i>
Protein	9.00 ± 0.03 ^a	9.87 ± 0.03 ^b	11.59 ± 0.17 ^c	10.43 ± 0.04 ^d	10.13 ± 0.04 ^c
Fat	0.23 ± 0.00 ^a	0.71 ± 0.01 ^b	0.20 ± 0.02 ^a	0.78 ± 0.00 ^c	0.90 ± 0.00 ^d
Ash	0.69 ± 0.04 ^a	0.76 ± 0.01 ^b	0.91 ± 0.00 ^d	0.87 ± 0.00 ^c	0.85 ± 0.02 ^c
Carbohydrates	58.35 ± 0.07 ^c	53.56 ± 0.03 ^a	57.72 ± 0.18 ^d	57.02 ± 0.04 ^c	56.06 ± 0.07 ^b
TDF	1.30 ± 0.13 ^a	2.72 ± 0.36 ^b	n.d.	2.73 ± 0.30 ^b	3.15 ± 0.03 ^c
IDF	1.16 ± 0.37 ^a	1.34 ± 0.05 ^{ab}	n.d.	1.30 ± 0.03 ^a	1.88 ± 0.39 ^c
SDF	0.13 ± 0.00 ^b	1.38 ± 0.00 ^c	n.d.	1.43 ± 0.00 ^d	1.27 ± 0.00 ^b

501 Means in rows followed by different letters are significantly different ($P<0.05$). Proximate values are expressed in g/100 g wet matter basis. TDF
502 – Total Dietary Fiber, IDF – Insoluble Dietary Fiber, SDF – Soluble Dietary Fiber. N.d. Not determined.

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Figure 1. Insects morphology. a) *H. illucens*; b) *A. domestica*; c) *T. molitor*.

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Figure 2. Mixolab curves of Control with wheat flour (black *solid line*), H. illucens (black *discontinuous line*), H.illucens (grey *discontinuous line*), A. domestica (black *dotted line*) and T. molitor (solid grey line) **insect flours**.

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