

Selected nutritional values of field cricket (*Gryllus assimilis*) and its possible use as a human food

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The nutritional values of the field cricket (*Gryllus assimilis* (Fabricius, 1775)) were analysed for dry matter using infrared scales, crude protein using Kjeldahl method, fat by Soxhlet method and fatty acid composition using GC-MS. The average dry matter content of analysed insect was 22.6 %, content of crude protein was 55.6 %, fat content was 11.8 %. The analysis of the fatty acid profile shows that the most abundant fatty acids were C18:2, C16:0 and C18:1 and the atherogenic index was 0.55. Microbial analyses were also part of the research, with evaluation of the total content of bacteria ($3.3 \cdot 10^6$ CFU g⁻¹), coli form bacteria ($3.5 \cdot 10^4$ CFU g⁻¹) and lactic acid bacteria ($5.8 \cdot 10^6$ CFU g⁻¹) and of yeasts and moulds ($4.4 \cdot 10^5$ CFU g⁻¹). For the high content of lipids and proteins edible insects could be a good alternative future source of crude protein and fat. EFSA also deals with obtaining sufficient data about the nutritional value and safety of consuming edible insects.

Keywords: Edible insects, Field cricket, Fats, Fatty acid profile, Crude protein, Microbiology

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According to the theories of English economist Thomas Malthus published at the beginning of the 19th century, the amount of food resources is growing linearly, while the population is growing exponentially. The number of people is thus increasing faster than the food provision. With current population growth, in 2050 the world population will be approximately 9 billion people¹. Thus, it is necessary to almost double the current food production. The first step to solve this problem is to eradicate malnutrition and famine - one of the global problems of mankind. Edible insects such as mealworm (*Tenebrio molitor*) could contribute to the fight with malnutrition thanks to its nutritional qualities¹. Another way to solve this problem is to change the overall look on the use of food resources, reduce wasting and streamline the food crop production².

Due to limited areas for plant cultivation and limited possibilities for livestock production it is necessary to

find other ways to cut the crop amount needed for livestock production. A possible solution could be the inclusion of insects with higher feed conversion than animals into the diet of the population of the industrialized Western countries, as it is in Africa, Asia and South America²⁻⁴. One of species, which is analysed for food purposes in Europe, is the field cricket [*Gryllus assimilis* (Fabricius, 1775)] (Fig. 1), whose benefits are the high protein content (59.2 % in the dry matter) and a significant presence of both essential and non-essential amino acids, as stated by Bednářová *et al.*⁵. Fat content 34.3 % in the dry matter of field cricket was measured by Bednářová *et al.*⁵. Finke⁶ stated only 14.4 % in a similar species – the house cricket (*Acheta domestica*), while Yi *et al.*⁷, stated 12.3 % (recalculated from the live weight). In this work, we analysed selected nutritional values of the field cricket reared on a farm in the Czech Republic. Subsequently, these samples were compared with the commodities of animal origin and with the results from other countries.

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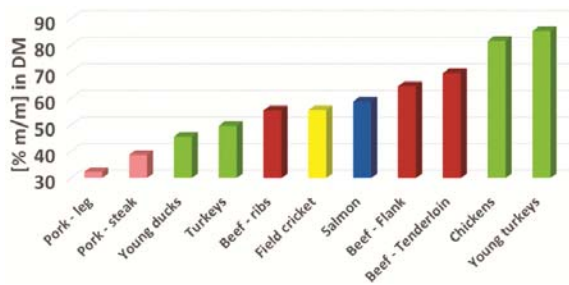


Fig. 1—Cricket on butter with toasted bread

Insects bred for human consumption is considered a farm animal⁸. For this reason, all the rules and regulations for livestock breeding apply. Each farm animal has its own specific gastrointestinal tract microflora⁹, which may be both positive and negative for the human body. That is also the case of the edible insects. However, this aspect has not been thoroughly examined from the health point of view, as indicated by European Food Safety Authority (EFSA) in October 2015, EFSA conducted profiling of the risks associated with the production and consumption of insects as food¹⁰. The document suggests the lack of nutritional analyses data of edible insects in European conditions. Insect species recommended for consumption by EFSA¹⁰ are e.g. mealworm (*Tenebrio molitor*), superworm (*Zophobas morio*), field cricket (*Gryllus assimilis*), waxworm (*Galleria mellonella*), migratory locust (*Locusta migratoria*) (Fig. 2), honeybee (*Apis mellifera*), and silkworm (*Bombyx mori*). In the Czech Republic, the situation is similar to that in other European countries⁵. Although in 1920 and 1937, recipes for preparing food from insects (e.g. cockchafer soup) could be found in the Czech cookbooks^{11,12}, nowadays even in rural cultures the knowledge about how to collect and process insects has disappeared. Consumption of insects in the Czech Republic became a specialty or delicacy, which only a few enthusiasts were interested in Borkovcová *et al.*¹³. Nowadays it is possible to consume edible insects in the Czech Republic at lectures about edible insects, in the framework of scientific research and food festivals. Insects are not sold for consumption for legislative reasons. Insects bred exclusively in farms are sold only as feed. Wild insects are not sold in the Czech Republic.

The greatest risk related to the breeding and processing of edible insect product is bacterial and parasitic contamination. Part of this work is to determine the total number of bacteria, coliform and lactic acid bacteria and the number of yeasts and moulds.

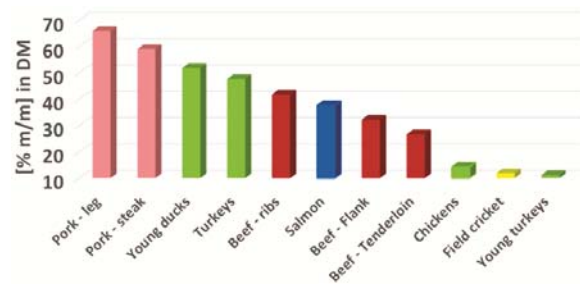


Fig. 2—Dried insects (*Tenebrio molitor*, *Gryllus assimilis*, *Locusta migratoria*, *Zophobas morio*)

Methodology

Material

Field cricket (*Gryllus assimilis*) used for the analysis was purchased in a pet store Jofikrmiva, Velvarska 105, Horomerice, Czech Republic. The insect was kept at conditions optimal for the development of each species and fed with a mixture of chopped vegetables and fruits. Prior to analysis, insect samples were adjusted as follows: after reaching adulthood, specimens were removed from breeding site. The next step was starving for 48 h, killing in boiling water (100 °C) and drying at 105 °C. Prepared samples were then homogenized, stored in refrigerator at 4-7 °C and analysed in summer 2016 in Czech University of Life Sciences Prague, Czech Republic, and Tomas Bata University in Zlin, Czech Republic.

Determination of dry matter using infrared scales

After drying, the sample was homogenized by grinding. About 0.5 g was equally spread on aluminium foil and subsequently analysed according to the standard ISO 1442:1997¹⁴ using a scale with infrared heater Precisa HA 300. Drying was carried out at 105 °C until a constant loss, which was less than 2 mg per minute.

Determination of nitrogen and crude protein using Kjeldahl method

Determination of nitrogen and the crude protein by Kjeldahl method was performed according to ISO 1871:2009¹⁵. Content of crude protein was calculated by multiplying the total nitrogen content by the coefficient 6.25.

Determination of fat content using Soxhlet method

Determining the amount of fat was carried out by continuous extraction by Soxhlet¹⁶ using the Gerhardt Soxtherm machine. The dried insect sample was homogenized, and about exactly 5 g were put into the

thimble. The sample was extracted with 150 mL of petroleum ether device SOXTHERM® (Gerhardt GmbH, Germany) using a cold extraction at 70 °C for 120 min. Then, the extraction flask was dried in an oven for 1 h at 103 °C, allowed to cool in a desiccator and weighed. The difference between total sample weight and the weight of the fat was the fat content of the sample.

Fatty acid profile

The fatty acid profile was measured using a gas chromatograph with mass spectrometry (GC-MS) Agilent 7890. The fat re-esterification was performed with a load of 0.5 g of fat using 0.25M methanolic KOH according to ISO 12966-4:2015¹⁷. Detector temperature was 250 °C. The feed had a temperature of 225 °C, with a split ratio 1:50. The temperature program was set to 70 °C (holding for 2 min), and then the temperature was increased by 5 °C per minute up to a temperature of 225 °C (holding for 9 min). Subsequently, the gradient was 5 °C/min till a temperature of 240 °C was reached (holding for 15 min). The analysis lasted 60 min totally. Carrier gas was helium, flow rate 1.2 mL/min. The column used for gas chromatography was Rt® - 2560, size 100 m x 0.25 mm x 0.2 µ. The results of the fatty acid profile were expressed using internal standardization, identified by FAME Mix (37 components) (Restek) standard. Detection was also performed using a spectrum library of the National Institute of Standards and Technology Library (NIST, USA). The fatty acid profile was complemented by the calculation of the atherogenic index by Chilliard & Ferlay¹⁸.

Microbial analyses

Freshly killed insects was aseptically weighed and divided into homogeniser bag, to which 50 mL of PPS (1 g peptone (Hi Media Laboratories Pvt. Ltd, India) and 8.5 g of NaCl (Penta, CR) dissolved in 1000 mL of distilled water and sterilized at 121 °C for 20 min) was added. Subsequently, homogenization was performed for 2 min and a dilution from 10⁰ to 10⁻⁵. From each dilution vaccination was done by plating 0.1 mL of inoculum to the respective agar. Enumeration of bacteria was carried out using a PCA (Plate Count Agar (Hi Media Laboratories Pvt. Ltd, India)) nutrient medium. Cultivation was carried out for 48 h at 30 °C. Coliform bacteria were determined using VRBA (Violet Red Bile Agar (Hi Media Laboratories Pvt. Ltd, India)). Cultivation was

carried out for 24 h at 37 °C. MRS agar soil (de Man Rogosa Sharpe Agar (Oxoid Ltd., UK)) was used for the determination of lactic acid bacteria, cultivation was done at 37 °C for 48 h. To detect the presence of yeasts and moulds CHYGA soil [chloramphenicol Yeast Glucose Agar (Oxoid Ltd., UK)] was used. Cultivation was done at 25 °C for 5 days. All culture media were prepared according to the manufacturer's instructions. After cultivation period expiration, colonies grown in the nutrient medium were counted.

Statistical analysis

The data was processed using Excel 2013 (Microsoft Corporation, USA). Results were expressed by average ± standard deviation.

Results and discussion

Dry matter content

Two samples of insect were used to determine dry weight on infrared scales. The average dry matter content in the evaluated insects was 22.6 ± 1.0 %. Bednářová *et al.*⁵ stated the average dry matter content in field cricket nymphs 33.3 ± 6.3 %. Higher values may be caused by different developmental stage of insects, as adult specimens were analysed.

Protein content

Crude protein content was 54.5 % in sample 1, and 56.6 % in sample 2. The average crude protein content was 55.6 ± 1.1 %.

The average content of crude protein in insects reported in the literature varied from 15 - 81 %^{2,5,19,20}. For Orthoptera, Rumpold & Schluter¹⁹ reported an average protein content of 61.3 %. Values measured in this study (56.6 %) are comparable with literature. Difference from these values and our results, may be caused by the fact that we used insect reared in farms, which focus on quantitative production of crickets for feeding purposes, while insect used in research used for comparison was of wild origin. As presented by Adámková²¹, Oonincx *et al.*²² and van Broekhoven *et al.*²³, the nutritional composition of insects depends on nutrition and temperature. In nature, insects feed on found and occasional food, while farm insects are supplied with all necessary nutrients for rapid growth.

Therefore, probably the influence of different feed showed higher content of proteins for example, dragonflies containing 60 % or mayflies with 66 %. Finke⁶ mentioned the protein content in the range 9 - 25 % of fresh insect weight, depending on the species.

Protein content in edible insect is comparable to other commodities of animal origin. For example, livestock meat contains approximately 18-22.3 % protein in fresh weight^{24,25}. An important indicator of protein quality is its digestibility. Digestibility of insect protein is reported in the range of 86-89 %²⁶, or even up to 96 %²⁷. For comparison, digestibility of egg white is 95 % and beef 98 %²⁶.

Comparison of the crude protein content in the field cricket (*Gryllus assimilis*) and animal commodities is shown in Fig. 3^{28,29}. From this figure, it is obvious that the content of crude protein in field cricket is comparable with the salmon and beef ribs.

Fat content

Fat content in sample 1 was 11.4 % of dry matter and in sample 2 % was 12.3 %. An average fat content in analysed insect was 11.9 ± 0.5 %. This



Fig. 3—Comparison of the crude protein content in field cricket (*Gryllus assimilis*) and animal commodities (Pipek, 1995; Steinhauser, 1995).

value coincides with the fat content in the house cricket (*Acheta domestica*), wherein the fat constitutes from 9.8 % to 22.8 % and the average value for genus *Orthoptera* is 13.4 %¹⁹. Bednářová *et al.*⁵ stated the fat content of most insect species from 10 % to 50 %; measured content therefore coincides also with this range.

Comparison of the fat content in the field cricket (*Gryllus assimilis*) and animal commodities is shown in Fig. 4^{28,29}. The figure shows that, the fat content in field cricket is comparable with the meat from chickens and young turkeys.

Fatty acid profile

Fatty acid profile shown in Table 1 shows that, the largest part is formed by these three acids: C18:2



Fig. 4—Comparison of the fat content in field cricket (*Gryllus assimilis*) and animal commodities (Pipek, 1995; Steinhauser, 1995).

Table 1—Fatty acid profile in field cricket (*Gryllus assimilis*)

Fatty acid	Average content [%]	SD	Fatty acid	Average content [%]	SD
SFA			MUFA		
C4:0	0.01	0.000	C14:1 (cis-9)	0.06	0.000
C6:0	0.01	0.000	C16:1 (cis-9)	1.92	0.005
C8:0	0.01	0.000	C17:1 (cis-10)	0.19	0.000
C10:0	0.03	0.005	C18:1 (cis-9)	25.03	0.105
C12:0	0.12	0.000	C20:1 (cis-11)	0.24	0.000
C13:0	0.02	0.000	C22:1 (cis-13)	0.05	0.005
C14:0	1.28	0.005	Total MUFA	27.49	
C15:0	0.37	0.005	PUFA		
C16:0	25.85	0.060	C18:2 (cis-9, 12)	26.13	0.175
C17:0	0.57	0.005	C18:3 (cis-9, 12, 15)	1.60	0.005
C18:0	14.07	0.025	C20:2 (cis-11, 14)	0.44	0.005
C20:0	0.56	0.005	C20:3 (cis-11, 14, 17)	0.01	0.000
C21:0	0.03	0.000	C20:4 (cis-5,8,11,14)	0.21	0.015
C22:0	0.57	0.000	C22:2 (cis-13,16)	0.03	0.000
C23:0	0.22	0.005	C20:5 (cis-5,8,11,14,17)	0.38	0.020
Total SFA	43.72		Total PUFA	28.80	
Calculations					
Atherogenic index	0.55		Total n-3	1.99	
			Total n-6	26.81	
			Total n-9	25.32	

(cis-9, 12), C16:0 and C18:1 (cis-9). Together they form 77 % of total fatty acids. The highest average content in the analysed samples had acid C18:2 (cis-9, 12) with 26.1 %. Another significantly present is the acid C16:0 with 25.9 %. The acid C18:1 (cis-9) had slightly lower content - 25.0 %. Lower content, but still significantly above average, had the acid C18:0 - 14.1 %. The values of the remaining fatty acids, except for C16:1 (cis-9), C18:3 (cis-9, 12, 15) and C14:0 (1.9 %, 1.6 % and 1.3 %), did not exceed one percent. Unsaturated fatty acids accounted for 56.3 %. Monounsaturated fatty acid content in the analysed sample was 27.5 %. The total content of saturated fatty acids was 43.7 %.

Rumpold & Schluter¹⁹ state the content of monounsaturated fatty acids in the fatty acid profile within the range of 22.0 % to 46.6 %. These authors detected oleic acid C18: 1 (cis-9) and palmitoleic acid C16: 1 (cis-9), as the most abundant among monounsaturated fatty acids.

The average content of polyene fatty acids in literature is between 15.9 % to 39.7 %, which also corresponds to the value measured in this work. In accordance with this is the main linoleic acid C18:2 (cis 9, 12), which is important for physiological processes and the formation of polyene fatty acids. Significant representation in terms of health influence has also linolenic acid (1.6 %) having anticarcinogenic and anti-inflammatory effects³⁰.

Values for saturated fatty acids were also similar. Rumpold & Schluter¹⁹ state the average content of these acids from 30.8 % to 41.9 %, which is slightly lower than the value measured in this work (43.7 %). Order of the two most abundant saturated fatty acids (palmitic acid, C16:0). Part of the fatty acid profile evaluation was the atherogenic index, calculated by Chilliard & Ferlay¹⁸. Atherogenic index is important for nutritionally-health perspective in assessing the risk of cardiovascular disease occurrence. The increasing value of this index increases the risk of atherosclerosis and other cardiovascular diseases that are the leading cause of death in Europe. 80 % of premature disease are preventable, so it is important to observe this. The measured and calculated atherogenic index is lower, compared with other animal commodities. Stajić *et al.*³¹ reported 0.7 for beef, 0.5 for poultry and 0.6 for pork. Camacho *et al.*³² calculated index in the range of 0.7 to 0.9 in sheep.

Microbial analyses

Microbial analyses were also part of the research, with evaluation of the total content of bacteria

($3.3 \cdot 10^6$ CFU g⁻¹), coliform bacteria ($3.5 \cdot 10^4$ CFU g⁻¹) and lactic acid bacteria ($5.8 \cdot 10^6$ CFU g⁻¹) and of yeasts and moulds ($4.4 \cdot 10^5$ CFU g⁻¹).

Klunder *et al.*³³ stated that the total number of microorganisms detected in house cricket nymphs was 7.2 log CFU/g, Vandeweyer *et al.*³⁴ stated a value from 8.2 to 8.5 log CFU/g. Concentration measured in this study is lower than reported in literature. This difference is due to the amount of lactic and coliform bacteria. The difference between the number of yeasts and moulds in our study and the literature is smaller. A sample of living field cricket analysed in this work contained 5.6 logs CFU/g. The available literature states the contents in similar species from 5.0 log CFU/g in the migratory locust (*Locusta migratoria*)³⁵ to 6.1 log CFU/g for house cricket (*Acheta domesticus*)³⁴.

From the available literature it is obvious, that the number of bacteria from the Enterobacteriaceae family, which includes coliform bacteria evaluated in our study, is quite volatile in edible insect samples. The numbers of Enterobacteriaceae detected by Klunder *et al.*³³ was 4.2 logs CFU/g, while Vandeweyer *et al.*³⁴ detected the values from 7.5 to 8.0 logs CFU/g. Stoops *et al.*³⁵ measured for similar species - the migratory locust value of 7.1 to 7.6 logs CFU/g. Content we detected in the field cricket was 4.5 logs CFU/g. Value is in accordance with the values given by Klunder *et al.*³³. Numbers of coliform bacteria are determined primarily by the environment in which the insect is kept.

The amount of lactic acid bacteria in a field cricket was 6.8 logs CFU/g. This result corresponds to the literature, which indicates a value from 7.3 to 7.9 logs CFU/g^{34,35}. Enzymes produced by the lactic acid bacteria convert nutrients from food into simpler substances. This helps to nourish cells. Furthermore, they produce lactic acid, which stops the reproduction of putrefactive bacteria and staphylococci³⁶. In terms of human health status, lactic acid bacteria have a positive impact especially on the intestinal microflora, where they inhibit the growth of harmful bacteria, and enhance intestinal peristalsis³⁷.

Conclusion

This research analysed essential nutritional values (dry matter, protein, fat, fatty acid profile) and microbiological attributes of the field cricket specimen reared in the Czech Republic. Based on the analyses it can be concluded that edible insects could be a good alternative source of crude protein and fat

in the future. In the fatty acid profile, the most abundant was linoleic acid, which is important for physiological processes and the formation of polyene fatty acids. A significant representation from the health point of view had also a linolenic acid with anticarcinogenic and anti-inflammatory effects. From the fatty acid profile, atherogenic index was calculated and was lower or comparable with other commodities. Finally, microbial analysis was performed, which confirmed that material from field cricket is safe and suitable for food purposes and human nutrition in Europe. Given the microbial content, the risk of microbial contamination, pinpointed by the EFSA, is obvious, and thus needs a further investigation. EFSA further states that, it is necessary to further investigate the risks and safety of insect. Legislation must also be created, to deal with the methods of obtaining or farming, feed composition and evaluation of the health safety of edible insect, including the introduction of a field cricket into the diet.

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