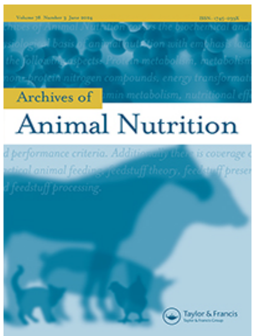

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Archives of Animal Nutrition

ISSN: (Print) (Online) Journal homepage: www.tandfonline.com/journals/gaan20

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To cite this article: Gergana Yordanova, Radka Dimova Nedeva, Apostol Petrov Apostolov, Isobel Margaret Whiting, Stephen Charles Mansbridge, Stephen Paul Rose & Vasil Radoslavov Pirgozliev (08 Nov 2024): Estimation of the digestible energy value of fat obtained from black soldier fly larvae (*Hermetia illucens*) for growing pigs, Archives of Animal Nutrition, DOI: 10.1080/1745039X.2024.2415546

To link to this article: https://doi.org/10.1080/1745039X.2024.2415546

Published online: 08 Nov 2024.

ARCHIVES OF ANIMAL NUTRITION
https://doi.org/10.1080/1745039X.2024.2415546

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RESEARCH NOTE

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Estimation of the digestible energy value of fat obtained from black soldier fly larvae (*Hermetia illucens*) for growing pigs

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ABSTRACT
An experiment was conducted to determine the digestible energy (DE) of insect fat (IF) from black soldier fly larvae (BSLF) for growing pigs. Saturated fatty acids (SFA) were the dominant group of fatty acids in the IF, with lauric acid (C12:0) and palmitic acid (C16:0) comprising the greatest concentrations in this group. Linoleic acids (C18:2) and oleic acids (C18:1) were the main unsaturated fatty acids. The IF contained 37.63 MJ/kg gross energy and 2.55 g/kg nitrogen. During the experiment, a DE bioassay was performed wherein growing pigs were fed one of the two experimental diets (either a maize–wheat–barley–soy basal diet or a diet containing 50 g/kg IF plus 950 g/kg of the basal diet). The DE of the IF was calculated based on the differences between the DE values of basal and test diet (substitution method). The DE of the IF was determined to be 36.86 MJ/kg. The IF contains a DE level comparable to vegetable oils, including soybean, rapeseed, corn and palm oils. The results showed that the examined fat from BSLF is a good source of available energy and can be incorporated in pig diets as an alternative energy source.

ARTICLE HISTORY
Received 02 October 2024
Accepted 08 October 2024

KEYWORDS
Black soldier fly larvae; insect fat; pigs; digestible energy

1. Introduction

Fats and oils are a concentrated source of energy, and increasing the amount in pig diets will increase the energy available for growth, maintenance and reproduction (Noblet and Shi 1993). Combined with volatile raw ingredient costs, there is increasing interest amongst nutritionists in maximising the use of existing and developing novel fats and oils to meet the performance targets of modern pigs (Wealleans et al. 2021). The use of insect-derived fat (IF) and meals in animal feed is a field of growing interest not only because of its energy value but also because of the possibility to apply circular economy principles (Gasco et al. 2020; Chobanova et al. 2023). Although a wide variety of edible insect species are available on the market (Gasco et al. 2020), black soldier fly larvae (*Hermetia illucens;* BSFL) are one of the most popular species. Insect fat is obtained as a by-product of full-fat insect meal

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processing and has been frequently considered a novel source of energy in poultry diets (Kierończyk et al. 2021; Kierończyk, Rawski, Mikołajczak, et al. 2022; Heuel et al. 2022). van Heugten et al. (2022) reported that supplemental BSFL oil, replacing equal amounts of corn oil, increased average daily weight gain (WG) and improved feed efficiency, resulting in increased final body weight of nursery pigs. Gasco et al. (2019) reported that fats from BSFL and yellow mealworm are suitable sources of lipids in rabbit diets to replace soybean oil without any detrimental effect on growth performance, apparent digestibility, gut mucosa traits and health. In addition, Dabbou et al. (2020) showed that these fats have antibacterial properties, positively influencing the rabbit caecal microbiota. Information on the direct use of dietary IF in growing pigs is limited, although there are data on incorporating full fat insect meal in diets (Veldkamp and Vernooij 2021; Hong and Kim 2022).

In poultry, the effect of IF inclusion has been investigated as a partial or total replacement of soybean oil (Kierończyk et al. 2021 Kierończyk, Rawski, Stuper-Szablewska, et al. 2022; Hartinger et al. 2022; Schäfer, Grundmann, Friedrichs, et al. 2023; Schäfer, Grundmann, Maheshwari, et al. 2023), poultry fat and palm oil (Benzertiha et al. 2019). Information on the available/metabolisable energy (ME) of IF for poultry is growing, although similar information for pigs is still not available. Digestible energy (DE) is the basic system historically used to describe the available energy in pig diets (BSAS 2003). However, ME can also be estimated by applying a correction factor of 0.92 to 0.98 to DE, depending on breed and class of pig, or determined through the addition of urine collection (NRC 2012). Directly added fat is used in swine diets as an energy source when the cost is economically advantageous (Kellner and Patience 2017), though knowledge of available energy (e.g. DE) of all feeding stuff used is important information for accurately formulating practical diets. Incorrect energy values of feed ingredients could lead to economic losses for pig farmers due to incorrect costing in formulations and poor performance outcomes. Given the increasing interest in using IF in pig diet formulations, information on its DE value is needed. Thus, the aim of the study was to determine the DE of an IF sample obtained from BSFL when fed to pigs. Pig body weight (BW), daily feed intake (FI), daily WG and feed conversion ratio (FCR) were determined as baseline pig performance metrics.

2. Materials and methods

2.1. Insect fat and experimental diets

The IF sample used in this experiment was from larvae (no older than 17 d) of the BSF and was purchased from Hexafly™ (Navan, Co., Meath, Ireland). The production process followed EC regulations EU 142/2011 (Commission Regulation, 2011), whereby the product was pre-washed and then oven-cooked for between 15 and 20 min at 120°C to kill all microbiological contaminants. Following this step, the product was transferred to a 60°C oven for conditioning for 40 min before fat separation. The dry product then was further processed through a screw press (OLEXA®, Arras, France) to separate the dry meal from the fat component (IF).

A basal feed (BF; Table 1) containing maize, wheat, barley and SBM was formulated in mash form to meet pig nutrient recommendations (Agricultural Institute, Shumen, Bulgaria). Five g/kg of titanium dioxide (TiO_2) was added to the feed as an indigestible

Table 1. Ingredient composition and analysis of the basal feed (g/kg, as fed) used in the experiment.

Ingredients	[g/kg]
Maize	200.00
Wheat	348.00
Barley	238.00
Soybean meal [46% CP]	190.00
Lysine	0.70
NaCL	2.00
Calcium carbonate	6.00
Dicalcium phosphate	12.50
Vitamin and mineral premix[a]	2.50
Calculated analysis (as fed):	
Crude protein [g/kg]	163.3
Metabolisable energy [MJ/kg]	12.92
Digestible energy [MJ/kg]	13.46
Lysine [g/kg]	8.0
Calcium [g/kg]	7.7
Phosphorus [g/kg]	5.7
Analyzed composition (as fed)[b]:	
Gross energy [MJ/kg]	16.93
Dry matter [g/kg]	852.0
Crude protein [g/kg]	158.1
Ether extract [g/kg]	30.3
Calcium [g/kg]	7.7
Phosphorus [g/kg]	6.7

[a]Supplied the following amounts per kilogram premix (MeloVit 14PG 0,25%): 2600000 IU of vitamin A 700,000 IU of vitamin D3, 20000 mg of vitamin E as tocopherol acetate, 800 mg of vitamin K3, 800 mg vitamin B1, 7200 mg vitamin B3 (niacin), 4000 mg vitamin 5 (calcium D-pantothenate), 400000 µg folic acid 100,000 µg biotin 10,000 µg vitamin B12, Fe (Fe-II-sulphate) 35000 mg, Zn (Zn oxide) 30000 mg, Mn (Mn oxide) 18000 mg, Cu (Cu-II-sulphate) 6000 mg, Se (sodium selenite) 120 mg, I (calcium iodide) 100 mg.
[b]Analysed in technical duplicates.

feed marker during mixing. The BF was then split into two batches, with one batch used as the BF control diet and the second batch comprising 950 g/kg BF +50 g/kg IF (diet BFIF), giving a total of two experimental diets.

2.2. Animals, management and sample collection

The experiment was conducted at the pig research facility of the Agricultural Institute (Shumen, 9700, Bulgaria) and approved by the Academy Research Ethics Committee (Project number Ж182, Agricultural Academy, Sofia). The study was designed in compliance with the guidelines of the European and Bulgarian legislation regarding the protection of animals used for experimental and other scientific purposes (Directive 2010/63; EC, 2010 - put into law in Bulgaria with Regulation 20/2012). The manuscript has been prepared in compliance with the ARRIVE 2.0 guidelines (Percie du Sert et al. 2020). A total of 24 mixed sex, Danube White pigs were reared in groups of 8 in slated floor pens until 149 d of age. Subsequently, 16 of the pigs were selected, excluding very heavy and very light pigs, and allocated to 16 individual pens, each with 2 m^2 floor area.

The pens had a concrete floor and were equipped with an individual feeder and drinker. The ambient temperature was maintained at 18–20°C, and the relative humidity was 65–71%. The experiment started when pigs were 169 d age, providing 20 d of adaptation period to the BF. The mean weight of the pigs was 74.0 kg (SDEV ± 3.34). Each diet was fed to eight pens following randomisation. Pigs were weighed at the start (at 169 d old) and at the end of the feeding bioassay (at 177 d old), inclusively. Approximately, 3 kg of feed was offered daily to each of the pigs. The feed was split into two portions, one fed in the afternoon and the other in the morning. Two hours after the morning feeding, all feed residuals were collected and weighed to determine daily FI. If there were no refusals, the daily portion for the next day was increased. The first 5 d of the feeding bioassay were for treatment diet acclimatisation, and faeces were collected during last 4 d following standard methodology (Adeola 2001). After the collection, faeces were oven dried at 60°C, milled to pass through a 0.75 mm sieve and further analysed for the digestibility marker (TiO_2), dry matter (DM), nitrogen (N), ether extract (EE) and gross energy (GE). Daily FI, daily WG and FCR were determined as baseline pig performance metrics.

2.3. Laboratory analysis

The fatty acid composition of the IF was determined by gas chromatography (GC) (2017). Fatty acid methyl esters were obtained by pre-esterification of samples with 2% sulphuric acid in absolute methanol at 50°C (2017). The determination of the fatty acid composition was performed on an Agilent 8860 GC equipped with a DB Fast FAME capillary column (30 m × 0.25 mm × 0.25 μm film thickness) and a flame ionisation detector. The operating temperature regime was: 70°C (1 min), an increase of 6°C/min to 180°C and by 5°C/min to 250°C; injector temperature – 270°C, detector – 300°C. Carrier gas – nitrogen, carrier gas velocity 25 mL/min. The identification of fatty acids was carried out by comparison of retention times of a standard 37 component mixture of fatty acid methyl esters. The quantitative fatty acid composition was determined by the percentage ratio between the areas of the individual peaks in the chromatogram.

The DM in dietary and excreta samples was determined by drying the samples overnight in a forced draft oven set at 105°C until a constant weight was obtained (AOAC 2006: Method 934.01). To determine the total N content in feed, IF and excreta, the Dumas combustion method (Leco FP-528N, Leco Corp., St. Joseph, MI) was used with EDTA as a calibration standard (AOAC 2006: Method 968.06). Crude protein (CP) was calculated as nitrogen (N) × 6.25. Ether extract in samples was determined with petroleum ether extraction method using a Soxtec system (Foss Ltd., Warrington, UK; AOAC 2006: Method 991.36). The GE of the diets, IF and excreta samples was determined by the isoperibol bomb calorimeter (Model 6200, Parr Instrument Co., Moline, IL) with benzoic acid as an internal standard (Oduguwa et al. 2007). Titanium dioxide in samples was determined as described by Short et al. (1996).

2.4. Calculations

Dietary DE values of the experimental diets were determined as follows:

$$DE = GE_{Diet} - \frac{(GE_{Faeces} * Ti_{Diet})}{Ti_{Faeces}}$$

where GE is the gross energy in diet or faecal samples.

Total tract nutrient retention coefficients (NR) were calculated using the following equation:

$$NR = \frac{(N/Ti)_{Diet}(N/Ti)_{Faeces}}{(N/Ti)_{Diet}}$$

where (N/Ti) Diet = ratio of the respective nutrient to TiO_2 in diet, and (N/Ti) Faeces = ratio of the respective nutrient to TiO_2 in faecal samples.

The DE in IF was determined as follows:

$$DE(IF) = \frac{DE\ of\ IF\ diet - DE\ of\ BF * 0.95}{0.05}$$

where 0.95 is the proportion of BF in the IF diet and 0.05 is the proportion of IF in the IF diet.

The metabolisable (ME) and net energy (NE) values of the insect fat samples were obtained based on the determined DE value, following published methods (van Milgen et al. 2001; Kellner and Patience 2017).

2.5. Statistical analysis

Data were analysed by one-way ANOVA using Genstat (22^{nd} edition) statistical software (IACR Rothamsted, Hertfordshire, UK). Data were checked for homogeneity and normality of residuals prior to ANOVA. Results were considered significant at $p < 0.05$. Data are expressed as means and their pooled standard errors of means (SEM).

3. Results and discussion

The analysed chemical composition of the BF is shown in Table 1 and analysed nutrients were within the expected ranges. Saturated fatty acids (SFA) were the dominant group of fatty acids in the IF, with lauric acid (C12:0), myristic acid (C14:0) and palmitic acid (C16:0) comprising the greatest concentrations in this group (Table 2). Linoleic acids (C18:2) and oleic acids (C18:1) were the main unsaturated fatty acids. However, the concentration of polyunsaturated fatty acids was slightly greater in comparison to the monounsaturated fatty acid concentration. The overall fatty acid composition was similar to the findings of Benzertiha et al. (2020), who also reported that the main fatty acid present in BSFL oil is lauric acid (C12:0), averaging 42.4% of total fatty acids, with myristic (11.0%), palmitic (13.7%), oleic (13.9%) and linoleic (16.2%) acid representing other fatty acids present at large quantities. However, the fatty acid composition of IF can be modulated by using different rearing substrates to achieve specific targeted fatty acid composition, although lauric acid typically remains present at high concentrations (Oonincx and Finke 2021; Georgescu et al. 2022). There was no further processing/purification of the IF fraction after the screw press, thus some meal particles may have

Table 2. Fatty acid profile [g/100 g fatty acids], nitrogen, carbon and gross energy (air dried basis) of black soldier fly (*Hermetia illucens)* larvae fat used in the study[a].

Item	Amount
C 10:0	1.1
C 12:0	39.8
C 13:0	0.1
C 14:0	8.8
C 14:1	0.2
C 15:0	0.2
C 16:0	15.9
C 16:1	1.9
C 17:0	0.3
C 17:1	0.2
C 18:0	2.0
C 18:1	11.2
C 18:2 *n*-6	15.5
C 18:3 *n*-3	1.7
C 20:0	0.2
C 20:1	0.6
C 20:2 *n*-6	0.1
C 22:0	0.1
C 22:1	0.1
Saturated	68.5
Unsaturated	31.5
Monounsaturated	14.2
Polyunsaturated	17.3
Nitrogen [g/kg]	2.55
Carbon [g/kg]	509.8
Gross energy [MJ/kg]	37.63

[a]Analysed in technical duplicates.

Table 3. Effect of dietary black soldier fly larvae (*Hermetia illucens)* fat inclusion on growth performance of growing pigs, nutrient retention coefficients and dietary digestible energy.

Item	BW start [kg]	BW end [kg]	FI [kg/d]	WG [kg/d]	FCR [kg/kg]	DMR	NR	FR	DE [MJ/kg]
BF	74.2	83.1	2.85	1.12	2.65	0.743	0.649	0.866	12.45
BFIF	73.9	82.9	2.93	1.12	2.66	0.749	0.647	0.929	13.51
SEM	1.2	1.2	0.04	0.07	0.169	0.016	0.022	0.008	0.219
P	0.898	0.909	0.213	0.976	0.978	0.775	0.935	<0.001	0.004

BW: body weight; FI: daily feed intake; WG: daily weight gain; FCR: feed conversion ratio; DMR: dry matter retention; NR: nitrogen retention; FR: fat retention; DE: digestible energy; BF: basal feed; BFIF: diet containing 5% black soldier fly larvae fat + 95% BF; SEM: pooled standard error of the mean; P: probability of differences.

been left, explaining the small nitrogen content. The GE of the IF was similar to the GE of another IF batch from the same producer (Chobanova et al. 2024).

Table 3 contains information on the baseline pig performance metrics and dietary nutrient retention coefficients, in addition to DE. The growing pigs performed as expected for the breed under the research farm rearing conditions (Apostolov et al. 2015; Yordanova et al. 2017). The nutrient retention coefficients and the DE were within the expected range for fats and oils (Kellner and Patience 2017). The greater values for FR ($p < 0.001$) and DE ($p < 0.05$) in BFIF diet can be explained by the inclusion of IF.

Table 4. Comparison of the digestible (DE), metabolisable (ME) and net (NE) energy values of the commonly used dietary energy sources for pigs with black soldier fly larvae (*Hermetia illucens)* fat determined in the present study.

Fat source		DE [MJ/kg]	ME [MJ/kg]	NE [MJ/kg]	Reference
Hermetia illucens larvae fat	Pigs 83 kg BW	36.86	36.12	31.79	Present study
Soybean oil	Pigs 50 kg BW	36.05	35.32	31.09	Kellner and Patience (2017)
	Adult pigs	34.50	34.30	29.65	INRAE-CIRAD-AFZ (2022)
Rapeseed oil	Adult pigs	34.50	34.30	30.70	INRAE-CIRAD-AFZ (2022)
Canola oil	Pigs 50 kg BW	39.86	39.06	34.37	Kellner and Patience (2017)
Corn oil	Pigs 50 kg BW	33.44	32.77	28.84	Kellner and Patience (2017)
Palm oil	Pigs 50 kg BW	35.56	34.85	30.67	Kellner and Patience (2017)
	Adult pigs	34.50	34.30	30.80	INRAE-CIRAD-AFZ (2022)
Sunflower oil	Adult pigs	34.30	34.10	30.50	INRAE-CIRAD-AFZ (2022)
Poultry fat	Pigs 50 kg BW	34.04	33.36	29.36	Kellner and Patience (2017)
	Adult pigs	34.50	34.30	30.80	INRAE-CIRAD-AFZ (2022)
Pig lard	Adult pigs	33.40	33.20	29.10	INRAE-CIRAD-AFZ (2022)
Tallow	Pigs 50 kg BW	34.38	33.69	29.65	Kellner and Patience (2017)
	Adult pigs	33.40	33.20	29.80	INRAE-CIRAD-AFZ (2022)

The level of DE in a single feed material, e.g. IF, can be obtained primarily by employing ingredient substitution or linear regression methods (Wu et al. 2020; Abdollahi et al. 2021). The substitution method used in the present study was implemented by Kellner and Patience (2017) to assess the DE value of various dietary fat sources in pigs. Based on DE data, the ME and NE of the fat sources were also estimated as reference values for the industry. The results indicate that the DE of the IF was 36.86 MJ/kg, ME was 36.12 MJ/kg and NE was 31.79 MJ/kg (Table 4). In general, the calculated values are similar to experimentally obtained published reports for the available energy for pigs of various animal and vegetable fat sources (Table 4).

Recent research (Kierończyk et al. 2018) has shown that insect fat is high in available energy for poultry and can replace soybean oil in poultry diets (Hartinger et al. 2022; Schäfer, Grundmann, Friedrichs, et al. 2023; Schäfer, Grundmann, Maheshwari, et al. 2023). Research with rabbits (Gasco et al. 2019) and nursing pigs (van Heugten et al. 2022) has also shown that IF can replace soybean or corn oils, respectively, without negatively affecting growth performance and may improve health status (Dabbou et al. 2020). Partially replacing soybean products may therefore contribute to improving sustainability in animal production (Pirgozliev et al. 2023). However, factors such as dietary fat type, age (developmental stage) and genotype of pigs also affect energy and nutrient digestibility, although values vary in the literature. According to Jørgensen and Fernandez (2000), fats from plant origin are better digested than fats from animal origin due to their different fatty acid composition. Research by Kellner and Patience (2017) estimates the NE value of animal fat at 29.65 MJ/kg, while soy oil is valued at 31.09 MJ/kg. In contrast, the French INRA system (INRA CIRAD AFZ 2022) estimates the NE of soy oil at 29.65 MJ/kg, while animal fat is valued at 29.80 MJ/kg. Noblet and Shi (1993) reported that the energy value of diets or ingredients for pigs can vary with animal body weight and/or feeding level, as digestive interactions between fat and fibre can also play a role. It is therefore important to consider all relevant parameters for estimating the variations in energy value of IF for different categories of pigs to ensure precise formulations, an area which requires further research.

4. Conclusions

The present study demonstrated that fat obtained from BSFL contains a DE level comparable to vegetable oils, including soybean, rapeseed, corn and palm oils. However, the fatty acid profile differentiates it from other animal or plant products and must be considered when formulating precision diets for sustainable pig production systems. Following these considerations, IF can therefore be a valuable source of available energy for pigs.

Acknowledgments

The authors acknowledge the support of technical staff of Agricultural Academy, Agricultural Institute, 9700 Shumen, Bulgaria, for executing the animal part of the study.

Disclosure statement

No potential conflict of interest was reported by the author(s).

Funding

The project was funded by Agricultural Academy, Sofia, Bulgaria, Project number Ж182.

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