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Original Article

Effects of housefly larvae (*Musca domestica*) dehydrated meal on production performances and sensory properties of broiler meat

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Abstract

The trial included 100 one-day-old broilers, Cobb 500, of both sexes, divided into the two groups, 50 broilers in each. The experiment lasted 42 days and was conducted in three phases: days 1-7, days 8-21 and days 22-42. The control group of broilers (C) was fed a mixture of standard composition (basal diet), while the experimental group (O-I) received the same diet but with the addition of dehydrated housefly larvae in the amount of 5% (% as fed basis) in the starter diet and 4% in grower and finisher diets. Production results (body weight, body weight gain, feed consumption and feed to gain ratio) were determined during the trial. The broilers fed larvae meal achieved statistically higher ($p < 0.001$) average body weight and daily weight gain with superior feed to gain ratio, compared to the control group, but also higher feed consumption and feed conversion. Meat samples obtained from the broilers fed larvae meal were graded as more acceptable by panelists, in comparison to those obtained from the control group. The achieved results point to the fact that dehydrated housefly larvae meal can be used as an alternative source of proteins in broiler nutrition.

Keywords: alternative protein feed, broilers, insects, larvae meal

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Introduction

In poultry production, feed accounts for about 70% of the total production costs (Moreki et al., 2012). Furthermore, protein costs represent approximately 15% of total feed costs (Roy et al., 2010). The high costs and scarcity of the protein sources, such as soybean cake, groundnut cake and fish meal, are the major challenges which commercial poultry production is confronting (Adeniji, 2007). Animal protein sources are preferred over plant protein sources due to the good balance of essential amino acid and vitamin content (Šefer et al., 2015; Saima et al., 2008).

Currently, insects are considered to be a new, alternative protein source for animal nutrition (Muros et al., 2014) not only because of the high content of protein, from 39.16% (Atteh and Ologbenla, 1993) to 64% (Hwangbo et al., 2009), but also because of the high balance of essential amino acids. Insect rearing could be one of the ways to enhance food and feed security (Al-Qazzaz et al., 2016). The ability of housefly larvae (*Musca domestica Linnaeus*) to grow on many different substrates makes them useful to turn waste (e.g. poultry manure) into a valuable biomass. The high content of proteins and fat, the low level of fibers and the amino acid composition which is close to the ideal protein for poultry make this feed comparable to the most common protein sources in poultry nutrition such as soybean meal, fish and meat-bone meal (Pretorius et al., 2011).

It was considered that the proximate composition of dehydrated larvae meal (DLM) is variable according to the processing method (Aniebo and Owen, 2010). It is important to point out that the dehydrated larvae of housefly *Musca domestica L.* used in this trial is a novel product on the market, therefore worth examining, and that production technology may impact the characteristics of the DLM product.

The objective of this study was to examine the effects of housefly larvae (*Musca domestica Linnaeus*) in the form of dehydrated meal on growth performances and sensory properties of broiler meat.

Materials and Methods

Birds, management and experimental diets: The trial included 100 one-day-old broilers (Cobb 500), of both sexes, from the same hatchery. The broilers were weighed individually and randomly divided into two groups, 50 birds in each. There were 10 replicates of 5 birds in each group. In that manner, measurement was enabled at the level of each replicate because one replicate served as an experimental unit for statistical analyses in the growth study. The broilers were reared in a temperature and humidity controlled room with 24 h constant lighting. Treatment of broilers in terms of care, zoo hygienic and micro-climatic conditions was adapted for this hybrid and floor breeding method. The experiment lasted 42 days. Feed and water were provided for *ad libitum* consumption. During the trial three feed mixtures were used (starter, from days 1-7; grower, from days 8-21; and finisher, from days 22-42) and formulated to fully meet the requirements of broilers recommended by NRC (1994). The control group of broilers (C) was fed a mixture of standard composition (basal diet), while the experimental group (O-I) received the same diet but with the addition of dehydrated larvae of housefly *Musca domestica L.* (DLM) in the amount of 5% (% as fed basis) of the starter diet and 4% of the grower and finisher diets in accordance with the manufacturer's recommendations. The commercial preparation Musca's (Altamed, LLC, Beograd) was used as a source of the housefly larvae. The inclusion of DLM has been done by partially replacing soybean meal and soybean full fat meal in the basal diet, in order to minimally affect the chemical composition and nutritional value of meals. Chemical analysis of the used product Musca's, and composition and chemical analysis of the experimental diets, are presented in Tables 1 and 2.

Samples and measurement: The broilers were weighed individually and feed consumption was measured on the 1st, 7th, 21st and 42nd days of the trial. Based on the obtained results, average body weight, total body weight gain, daily weight gain, feed intake and feed to gain ratio were calculated separately for each phase and for the entire trial. During the trial, health status and mortality were monitored and recorded daily. At the end of the trial 6 broilers from each group were randomly selected and slaughtered in order to collect meat samples (breasts and legs with thighs) for sensory tests.

Research protocols of the use of animals followed the guidelines of Ethical Committee of the Faculty of Veterinary Medicine, Belgrade University, Belgrade, Serbia as well as the EU Directive 2010/63/EU for animal experiments.

Laboratory analysis: The diets were analyzed for dry matter, fat, cellulose, proteins and crude ash (AOAC, 1997). Dietary gross energy (MJ/kg), concentrations of lysine, methionine and cysteine were calculated according to INRA-AFZ (2004). Fatty acid composition of the dehydrated larvae of housefly *Musca domestica L.* (DLM) was determined by gas-liquid chromatography of the fatty acid methyl esters, on Hewlett Packard 6890 (AOAC, 1997).

Sensory analyses: Sensory analyses were carried out in the Sensory Laboratory equipped with individual booths. Each booth is made up of three walls on each side, in order to prevent panelists influencing each other. All booths were provided with florescent lights to mask color differences between the samples. Sensory tests were performed at room temperature (22-24˚C).

After a cooling period (at 4˚C) of 24 h all tested samples were marked by random three-digit numbers. All meat samples were subjected to equal heat treatment in an electric oven, at 170˚C, until constant internal temperature of 80˚C was reached. The meat samples were cut to approximately 2.5×2.5×2.5 cm. The samples were served twice, individually to each panelist, in plastic bowls, at internal meat temperature of 60±5˚C, immediately after cooking.

The order of presentation of the samples was randomized to ten trained evaluators from the Department of Food Hygiene and Technology, Faculty of Veterinary Medicine, University of Belgrade. Selection of evaluators was conducted according to

ISO standard (ISO 8586, 1993). Tap water was provided to rinse the palate between samples. Intensities of tenderness, juiciness, flavor, aroma and desirability were evaluated. A scale from one to five was used for evaluation of sensory properties, ranging from the lowest intensity of each attributes (score 1) to the highest intensity (score 5). Differences in the overall acceptability of meat samples were determined by using a paired comparison test (ISO 5495, 1983).

Table 1 Composition and chemical analysis of experimental diets

Composition (% as-fed basis)	Experimental period (days)					
	I (days 1-7)		II (days 8-21)		III (days 22-42)	
	C	O-I	C	O-I	C	O-I
Maize	50.00	52.00	58.00	60.00	63.00	65.00
Soybean meal	25.00	21.00	17.00	14.00	14.00	13.00
Soybean	20.00	17.00	20.00	17.00	18.00	13.00
Larvae meal	–	5.00	–	4.00	–	4.00
Mineral-vitamin premix[a]	5.00	5.00	5.00	5.00	5.00	5.00
Analyzed and calculated composition (% DM)						
Moisture	11.00	11.05	11.24	11.30	11.43	11.51
Ash	5.55	4.00	2.87	3.14	6.05	5.96
Crude protein	22.85	22.84	19.90	19.80	18.29	18.28
Crude fat	6.05	6.19	6.09	6.24	2.73	3.06
Crude fiber	2.85	3.10	2.87	3.14	2.73	3.06
Ca	1.01	1.20	0.98	1.14	0.92	1.12
P	0.68	0.70	0.62	0.63	0.60	0.61
Lysine[b]	1.33	1.34	1.10	1.12	0.99	1.00
Methionine and cysteine[b]	0.84	0.85	0.71	0.72	0.66	0.67
Gross energy (MJ/kg)[b]	12.70	12.80	13.00	13.10	13.15	13.17

[a]Premix provided per kg of complete feed mixture: 13000 IU vitamin A, 2500 IU vitamin D3, 30 mg vitamin E, 3 mg vitamin K3, 2.5 mg vitamin B1, 8 mg vitamin B2, 3.5 mg vitamin B6, 0.02 mg vitamin B12, 0.1 mg biotin, 15 mg calcium pantotenate, 50 mg niacin, 550 mg choline chloride, 1 mg folic acid, 20 mg vitamin C, 35 mg iron, 8 mg copper, 80 mg manganese, 50 mg zinc, 0.0125 mg iodine, 0.15 mg selenium, 0.25 mg cobalt, 1000 PU (Phytase Units) phytase, 10000 mg citric acid, 100 mg antioxidant BHT
[b]Calculations were made according to INRA-AFZ (2004).

Table 2 Chemical analysis of dehydrated larvae of housefly *Musca domestica*

Analyzed composition	% of DM (Dry matter)
Moisture	8.50
Ash	6.81
Crude protein	54.36
Crude fat (Ether extract)	16.90
Crude fiber	12.86
NFE (Nitrogen Free Extract)	0.57
Calcium	0.58
Phosphorus	1.26
Fatty acid composition (%)	
Palmitic acid	23,03
Palmitoleic acid	17,14
Stearic acid	3,41
Oleic acid	21,77
Linoleic acid	4,88

Statistical analysis: Data were analyzed by Graph Pad Prism 6.0. software (Graph Pad Software Inc., San Diego, CA, USA). Statistical analysis of intergroup differences of means was performed by unpaired t test. One replicate of five birds served as the experimental unit in the growth study, and the answer of each evaluator served as the experimental unit for sensory test. Values were expressed as mean and standard deviation. Levels of $p < 0.05$ and $p < 0.01$ were considered as significant and highly significant, respectively.

Results

During the experiment no mortality was recorded. Production results during the trial are presented in Table 3. Body weight and average daily gain did not significantly differ between the groups on day 7 of the trial. For the other examined periods (days 8-21, days 22-42 and days 1-42), the broilers from the experimental group (O-I) had significantly higher ($p < 0.001$) body weight and daily weight gain compared to the control group (C). Feed consumption was significantly different for each experimental period, as well as for the whole trial ($p < 0.005$, $p < 0.001$). Observed differences for feed conversion were significant for the whole trial ($p < 0.05$).

Results from the sensory analyses are presented in Table 4. The composition of the experimental diets had significant impact on the aroma and desirability for both types of tested samples (breasts and legs with thighs) ($p < 0.05$). Furthermore, the diet had significant impact on the flavor of the breasts and legs with thighs ($p < 0.05$ and $p < 0.01$, respectively).

Table 3 Production results of broilers during the trial

	Group	Experimental period (days)			
		Day 1	Day 7	Day 21	Day 42
Body weight (g)	C	39.92±2.81	121.46±14.32	608.32±79.68^{A}	2045.82±222.90^{A}
	O-I	39.40±2.67	140.82±20.49	754.38±96.94^{B}	2339.38±213.30^{B}
		Days 1-7	Days 8-21	Days 22-42	Days 1-42
Average daily gain (g)	C	11.649±2.044	34.776±5.803^{A}	68.452±10.781^{a}	47.760±5.333^{A}
	O-I	14.489±2.879	45.524±13.849^{B}	75.476±10.835^{b}	54.761±5.076^{B}
Average daily feed intake (g)	C	17.714±0.8181^{x}	57.434±1.317^{A}	145.189±1.631^{A}	94.692±1.321^{A}
	O-I	19.242±1.500^{y}	64.583±1.466^{B}	154.762±1.170^{B}	102.116±2.590^{B}
Feed conversion (kg)	C	1.521±0.2126	1.652±0.1576^{a}	2.121±0.2316	1.983±0.1207^{x}
	O-I	1.328±0.2123	1.419±0.1412^{b}	2.050±0.1299	1.865±0.1104^{y}

*Values are expressed as mean ± SD.
Means within the same column with different superscript letters differ significantly: x, y ($p < 0.05$); a, b ($p < 0.01$); A, B ($p < 0.001$).

Table 4 Sensory evaluation of broiler meat samples

Attributes	Breasts			Legs with thighs		
	Experimental group			Experimental group		
	C	O-I	P value	C	O-I	P value
Tenderness	3.6±1.075	3.7±1.16	0.8437	3.5±1.269	3.7±1.160	0.7173
Juiciness	3.1±1.197	3.2±1.317	0.8609	3.3±1.160	3.6±1.174	0.5724
Flavor	2.5±0.9178^{A}	4.1±0.9944^{A}	0.0019	2.9±1.197^{a}	3.9±0.7379^{a}	0.0373
Aroma	2.8±1.033^{a}	3.9±0.8756^{a}	0.0193	2.8±0.7888^{a}	3.6±0.8433^{a}	0.0419
Desirability	3.0±0.8165^{a}	3.9±0.8756^{a}	0.0287	2.9±0.7379^{a}	3.9±1.101^{a}	0.0282

Score ranges from 5 (highest affirmative value) to 1 (lowest value).
Values are expressed as mean ± SD.
Means within the same row with same superscript letters differ significantly: a, b ($p < 0.01$); A, B ($p < 0.001$).

Figure 1 shows results related to the overall acceptability of the meat samples (breasts and legs with thighs). The number of performed tests was 20 (10 examiners with two repetitions). The lowest number of correct answers required for reaching the level of statistical significance ($p < 0.01$) was 16. The overall acceptability of breast meat samples was rated with 16, while that of legs with thighs were rated with 17 correct answers. The obtained results show that the overall acceptability of the examined samples of breast meat, as well as legs with thighs, was significantly ($p < 0.01$) higher compared to the control group.

Discussion

During the presented trial there were neither occurrences of clinical signs of disease nor mortality, which implies that larvae ingestion had no pathological effect on the examined chicks. This is in agreement with Calvert et al. (1969) and Adeniji (2007) who reported no pathological changes in chicks fed larvae based diets.

On the other hand, potential health risks are the main concern in the use of larvae meal in farm animal diet. Bacterial hazards mainly originate from a residential microflora, related to rearing conditions, handling, processing and preservation (ANSES, 2015). Primary concerns are pathogens such as *Salmonella*, *Escherichia coli* and *Campylobacter*, but *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Aspergillus tamarii*, and *Bacillus cereus* are also isolated from flies (Lawal and Adeduji, 2005; Strother et al., 2005; Wales et al., 2010). However, that potential risks have to be overcome during the processing of insects to insect products.

The experimental group of broilers, which received larvae meal via feed, achieved higher body

weight compared to the control group in all observed periods of the experiment ($p < 0.001$). These results are in accord with results previously reported by other authors (Pretorius, 2011).

Similar to the data of average daily gain presented here, Okah and Onwujiariri (2012) reported superior growth performance (daily body weight gain and feed conversion ratio) of broilers fed larvae meal diets (1.60 and 2.00% of diet) over the control group. In a similarly designed experiment conducted by Hwangbo et al. (2009) broilers that received supplemental larvae meal showed significantly ($p < 0.05$) higher weight gain than the control group of chicks that received soybean meal. The authors explained that such improvement was caused by the enhanced nutrient composition of larvae meal compared to the soybean meal feed, especially by the protein and essential amino acid content as well as their greater digestibility.

The results of feed intake presented in this trial are consistent with data earlier reported by Ocio and Vinaras (1979), who partially replaced (12%) soybean meal with a slightly lower level (4% of the diet) of larvae and reported differences in feed intake compared to the control group of chickens (1497.6±19.5 g and 1473.6±28.2 g, respectively), without statistical significance. Similar results were reported by Adeniji (2007), who partially replaced groundnut cake with different levels of larvae (5.00, 11.00, 16.50 and 22.00% of diet) in broilers. However, the lowest feed intake was recorded in the group receiving the highest amount of meal, which was, as the author explained, due to the dull color or appearance of the diet the broilers were sensitive to. Okah and Onwujiariri (2012) reported that broilers fed basal diet consumed less feed ($p < 0.05$) than those fed larvae meal, but this effect decreased with the increase in levels of larvae meal supplementation. These authors also reported that the feed conversion of birds fed larvae meal diets (diets were formulated with larvae meal to replace fish meal at 20, 30, 40 and 50% levels) was better ($p < 0.05$) than of those fed the control diet, which is in line with the results of the present study. Better feed to gain ratio in broilers that received larvae meal (5.5% of diet) was proved by Adenijji (2007), who explained it by the higher protein retention value. Protein retention value is related to the amount of unsaturated lipids in the way that apparent digestibility of protein is always higher when diets (larvae meal) are rich in unsaturated lipids. In a trial similar to the one presented here, Ocio and Vinaras (1979) reported almost identical values of feed to gain ratio in control and experimental groups of broilers (1.93 and 1.86, respectively) that received larvae meal in the quantity of 4% of the diet as a partial replacement of soybean meal. With regard to protein quality, larvae are comparable to meat by product, bone meal as well as fish meal and are superior to soybean meal (NRC, 1994). Although larvae meal protein is characterized by high biological value, it has been proven that the addition of methionine in diet improves the feed to gain ratio, because of the low level of methionine and cysteine in larvae (Hwangbo et al., 2009; Onifade et al., 2001).

The sensory analysis of meat samples (breasts and legs with thighs) showed statistically higher ($p < 0.01$) acceptability of the samples obtained from broilers fed larvae meal compared to the control group, as well as significantly higher scores for flavor, aroma and desirability ($p < 0.05$). In accordance with this, Jiya et al. (2014) did not observed statistical differences in flavor ($p > 0.05$), but other sensory parameters (color, tenderness, juiciness and overall acceptability) were significantly ($p < 0.05$) higher in meat samples obtained from broilers fed larvae meal, compared to the control group. These results have formed a trend – as the level of larvae meal increased, the palatability score also increased, so the broilers fed the highest level of larvae (2%) had the highest values in all observed parameters. Pieterse et al. (2013) used larvae meal in the quantity of 10% of broiler diet and concluded that such inclusion had positive rather than any detrimental effects on most carcass, meat and sensory characteristics although larvae-fed samples had higher metallic aroma and aftertaste values, but these values were low and unlikely to be detected by consumers.

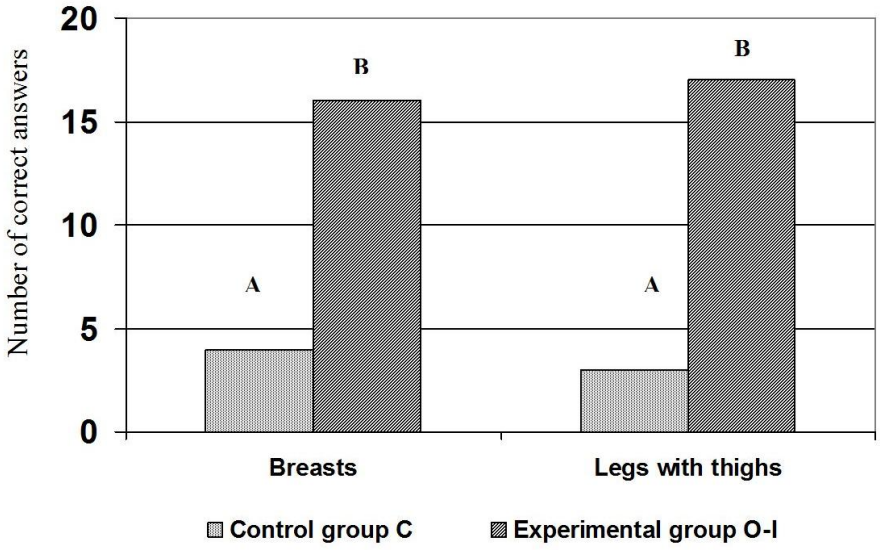

Figure 1 Sensory evaluation (overall acceptability) of broiler meat samples
Legend: Within the same tested parameter (breasts or legs with thighs) different superscript letters differ: A, B ($p < 0.01$).

The current EU regulatory framework (Regulation (EC) No 999/2001 and Commission Regulation (EU) No 56/2013) imposed a general ban on the use of processed animal proteins (PAPs) in the feeding of both ruminant and non-ruminant animals (with the exception of PAPs derived from non-ruminant animals (such as pigs and poultry) to be used in aquaculture feed), in order to control, prevent and eradicate transmissible spongiform encephalopathies. Considering safety for the use as animal feed, this ban includes insects and insect products, thus further investigations are mandatory in this field.

Based on the results of the present study it can be concluded that dehydrated housefly larvae meal is a potential protein supplement, particularly being an animal protein source, for commercial poultry diets.

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บทคัดย่อ

ผลของตัวอ่อนหนอนผีเสื้อ (*Musca domestica*) แบบอาหารแห้ง ต่อสมรรถนะการผลิตและคุณสมบัติทางประสาทสัมผัสของเนื้อไก่เนื้อ

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การทดลองในไก่เนื้อ Cobb 500 จำนวน 100 ตัว คละเพศ โดยแบ่งออกเป็นสองกลุ่ม กลุ่มละ 50 ตัว โดยการทดลองใช้เวลา 42 วัน และดำเนินการเป็น 3 ระยะ คือ วันที่ 1-7, วันที่ 8-21 และวันที่ 22-42 กลุ่มควบคุม (C) ได้รับอาหารผสมสูตรมาตรฐาน (อาหารมาตรฐาน) ในขณะที่กลุ่มทดลอง (O-I) ได้รับอาหารชนิดเดียวกัน แต่มีการเสริมหนอนตัวอ่อนแห้งจำนวน 5% (% ของอาหารมาตรฐาน) ในอาหารไก่เล็ก และ 4% ในอาหารไก่โต และไก่ระยะสุดท้าย โดยวัดผลการทดลองจากการผลิต ได้แก่ น้ำหนักตัว การเพิ่มน้ำหนักตัว การกินอาหาร และอัตราการเพิ่มน้ำหนักตัว ผลการทดลองพบว่า ไก่เนื้อที่ได้การเสริมหนอนตัวอ่อนแห้งในอาหาร มีน้ำหนักตัวเฉลี่ยและน้ำหนักตัวเพิ่มขึ้น อย่างมีนัยสำคัญทางสถิติ และมีอัตราการเพิ่มน้ำหนักตัวดีขึ้นเมื่อเทียบกับกลุ่มควบคุม แต่มีการกินอาหารและการเปลี่ยนอาหารที่เพิ่มขึ้น เช่นกัน ตัวอย่างเนื้อสัตว์ที่ได้รับจากไก่เนื้อที่เสริมหนอนตัวอ่อนแห้งในอาหาร มีผลคะแนนการประเมิน เป็นที่ยอมรับสูงขึ้น เมื่อเปรียบเทียบกับกลุ่มควบคุม ผลการทดลองชี้ให้เห็นว่า หนอนตัวอ่อนแห้งสามารถใช้เป็นแหล่งโปรตีนทดแทนในอาหารไก่เนื้อ

คำสำคัญ: อาหารโปรตีนทดแทน ไก่เนื้อ แมลง อาหารหนอนตัวอ่อน

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