Journal of Agriculture and Food Research 19 (2025) 101637

Contents lists available at ScienceDirect

Journal of Agriculture and Food Research

journal homepage: www.sciencedirect.com/journal/journal-of-agriculture-and-food-research

Dietary inclusion of mealworm frass: Effect on blood metabolites and growth performance of rabbits

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A R T I C L E I N F O

Keywords:
Blood metabolites
Carcass
Digestibility
Mealworm frass
Rabbits
Productive performance

A B S T R A C T

The effect of incorporating mealworm frass (MWF) in the diets of growing Gabaly rabbits on nutrient digestibility, live body weight and gain, properties of the carcass, and blood parameters was investigated. Fifty-two Gabaly rabbits (6 weeks old) were selected after weaning and divided into four groups in a random fashion (13 rabbits/group) based on their weights (653 ± 15 g). The experimental period extended from 6 to 15 weeks of age. Four isocaloric, isonitrogenous and isofibrous diets in pellet shape were formulated. Mealworm frass (MWF) was incorporated at levels of 0, 1, 2, and 3 % for rabbit groups: G0 (as a control), G1, G2, and G3. At the end of the feeding experiment period (15 weeks of age), digestibility trials were conducted to determine the nutrient digestibility and nutrition values of the experimental diets. Three rabbits were slaughtered from each group to determine the carcass traits and some blood parameters. The chemical composition MWF was recorded as dry matter 87.08-, organic matter 84.98-, crude protein 22.59-, crude fibre 17.62-, ether extract 2.78-, ash17.24-, and nitrogen-free extract 41.99- %. The digestibility of dry matter, organic matter, crude protein, and crude fibre improved (P <0.05) by an average of 12.3, 10.8, 11.4, and 60.6 %, respectively, in rabbits fed diets that contained MWF (G2 and G3) compared to the control group. Moreover, the digestibility of ether extract was significantly higher in G2 (85 %) compared with G1 (76.8 %) and G0 (76 %). While the digestibility of nitrogen-free extract was higher (P <0.05) in G3 (85.2 %) compared with G0 and G1 (78.3 and 77 %, respectively). The feeding value of total digestible nutrients and digestible energy increased by an average of 9.7 %, respectively, in G2 and G3 compared to G0. Including mealworm frass in growing rabbit diets led to numerical improvements in body weight, blood parameters, and carcass traits, though these changes were not statistically significant. Increasing the MWF content exhibited improvements in both relative and economic efficiency, with gains ranging from 5 to 40 %. These results suggested that MWF can be effectively integrated into rabbit diets up to 3 % without adverse effects, promoting both animal health and economic viability in rabbit production.

1. Introduction

Feeding is the primary cost and quality aspect that influences animal production in general and rabbit production in particular, as it is widely recognized. Approximately 60–65 % of the whole production cost goes toward feeding the rabbits, making it the most expensive component. Due to its potential to recycle organic waste effectively and provide a sustainable supply of protein for feed and food, the insect production

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https://doi.org/10.1016/j.jafr.2025.101637
Received 20 October 2024; Received in revised form 14 December 2024; Accepted 5 January 2025
Available online 6 January 2025

sector is fast-expanding globally. Insect excrement, or “frass” as it is known in science, is the most common by-product of insect manufacturing. Frass is obtained in larger quantities than the actual products of insects and should be utilized to sustain a circular economy [1].

Replacing conventional feed ingredients with alternative proteins through insects grown from food waste, and/or their frass, can significantly reduce the carbon footprint. The carbon footprint of livestock production can be significantly reduced by replacing conventional livestock feed with alternative proteins such as insects grown from food waste [2]. A more sustainable aquaculture sector may arise from switching from fishmeal to insect protein [3]. The farmed salmon sector in Norway is currently testing insect protein as a substitute for fishmeal in its urgent search for alternative feed substrates [4]. Furthermore, it should be mentioned that frass has been recognized as a component of livestock feed, and promising outcomes have been observed when it is given to farmed omnivorous fish [5]. This shows that frass has potential uses beyond being a fertilizer. The composition of frass, which is the excrement of insects, can differ based on factors such as the specific insect species, the age and life stage of the insects, the insects’ diet, the conditions in which they are reared, and even the pace at which plants in the insects’ diet are fertilized [6]. Insects and their products, such as their frass, are considered valuable feed components for aquaculture and poultry, but also for rabbits. They may also be used to promote livestock health [7,8]. Ayaz et al. [9], found that feeding sheep a concentrate mixture with 25 % mealworm frass improved the growth performance and health traits. Besides providing an important protein supply in animal diets, they positively affect the animal immune system, which can lead to a reduction in the usage of antibiotics [10]. Mealworm frass is a valuable source of saturated fatty acids making it a rich resource for such nutrients [11,12]. Furthermore, there is a significant concentration of mono-unsaturated fatty acids and polyunsaturated fatty acids [13,14]. Shojaaddini [15] reported that the fatty acid composition of mealworm frass included approximately 42.8 % linoleic acid (C18:2), 38 % lauric acid (C12:0), 15 % palmitic acid (C16:0), and 10 % oleic acid (C18:1), along with linolenic acid (C18:3). In addition, the proximate analysis of mealworm frass showed that it contains 19 % crude protein, 5 % lipids, 18 % crude fiber, 5.76 % ash, and 55 % nitrogen-free extract. Moreover, Shojaaddini [15] found that the amino acids contained in mealworm frass included 1.6 % aspartic acid, 1.2 % glutamic acid and glutamine, 0.7 % serine, 0.5 % arginine, and 0.5 % alanine. According to research on the mealworm (*Tenebrio molitor*), the nitrogen level of the larval frass can vary from 2.7 % to 7.8 % based on the larvae’s diet [16]. It is quite difficult to give a complete overview of the nutritional profile of insect frass. From a practical point of view, it would be wise to concentrate on the frass produced by young insects that are currently used in industrial production. These insects should be fed organic waste, and this method is expected to be the main way of producing insect frass in the future on a large scale. A lot of recent research has focused on larvae-derived frass from the black soldier fly (*Hermetia illucens*) and from the yellow mealworm. These two insect species are widely cultivated and have significant potential to transform both food waste recycling and animal nutrition [17]. Rabbits yield approximately 1.8 million tons/year of meat. China contributes 40 % to the global production, which is significantly more than from Italy (14.6 %), Spain (3.8 %), Egypt (3.1 %), and France (2.9 %) [18]. Incorporating n-3 fatty acids into the rabbit’s diet can improve the composition of fatty acids in rabbit meat [19]. Rabbit meat and its products could be considered functional foods due to their abundance of substances with potential health benefits. They are rich in several essential elements such as zinc and iron, selenium, as well as B vitamins, phosphorus, magnesium, and cobalt. Additionally, they can help increase the intake of vitamin E, important minerals like calcium, magnesium, potassium, and omega-3 fatty acids [20]. Integrating inexpensive, non-traditional feed ingredients into rabbit diets helps alleviate feed shortages, reduce feeding costs, and address environmental concerns. In this work, the effect of incorporating mealworm frass in diets of Gabaly-growing rabbits was studied, based on the parameters of nutrient digestibility, productive performance, feeding value, carcass characteristics, and also economic feed efficiency. The sustainability aspect of nutrient recycling and waste management is mentioned but not investigated in depth in this work.

2. Materials and methods

This study was conducted at El-Gemmaiza Animal Production Research Station, El-Gharbia Governorate, which belongs to the Animal Production Research Institute (APRI), Agriculture Research Center (ARC), Ministry of Agriculture, Egypt.

2.1. Frass

The mealworm frass (MWF), obtained in powdered form, was sourced from the AbouAbdo farm in Basos, Qalyubia Governorate, Egypt. This farm specializes in large-scale insect cultivation. The mealworms were only nourished with agricultural by-products or agriwaste, specifically wheat bran. This is an official requirement for feed materials for farmed animals. In this work, the frass was sterilized at a temperature of 70 °C for 60 min and utilized without the addition of any chemical substances.

2.2. Feeding and management

A group of fifty-two-six-week-old Gabaly rabbits in the growth stage were selected after being weaned. Four groups were formed, each consisting of thirteen rabbits based on their original live body weight, averaging 653 ± 15 g. The trial lasted from 6 to 15 weeks of age. 4 diets in pellet form with similar energy, protein, and fiber content were created. Mealworm frass (MWF) was included in the diets for the four groups: G1 (control), G2, G3, and G4, at amounts of 0, 1, 2, and 3 %, respectively. The pelleted diets for raising rabbits complied with the Agriculture Ministry Decree [21]. The rabbits were kept in well-ventilated cages made from galvanized wire with natural ventilation provided through the window. They had unrestricted access to experimental diets and potable water from automated drinkers equipped with individual nipples for each enclosure. The cages were cleaned daily to remove urine and faces that fell onto the floor. All rabbits were maintained under identical management, hygiene, and ambient conditions. The animals had been immunized against common illnesses. The live body weight of each rabbit was determined at the start of the experiment and then weekly until they reached the age suitable for sale (15 weeks). Throughout the trial period, data on live body weight, weight gain, feed intake, and feed conversion (measured in grams of feed per gram of gain) were collected, and the economic feed efficiency was calculated. The ingredients and composition of the 4 experimental diets are displayed in Table 1.

2.3. Digestibility trial

Four digestibility trials (three rabbits per group) were carried out after the feeding trial (15 weeks of age) to determine the feeding values and nutrient digestibility of the tested diets. Separating the rabbits in a metabolic cage made dropping collection easier. Daily monitoring of feed consumption was conducted, and a systematic collection of fecal samples began 24 h after each meal. Each day, the faces of the rabbits were collected in the morning for 5 days. A 2 % boric acid solution was sprayed on the faces to absorb ammonia. The faces were then dried in a 60 °C oven until they reached a constant weight. After drying, the faces were finely crushed and chemically analyzed. Representative diet and faces samples were analyzed according to the A.O.A.C [22]. recommendations. The Abou-Raya [23] approach was used to calculate nutrient digestion coefficients and feeding values.

Table 1
Ingredients and calculated chemical composition of the experimental diets (as fed).

Items	Experimental diets			
	G0	G1	G2	G3
Ingredients, %				
Clover hay, 12 % crude protein	32	32	32	32
Mealworm frass	0	1	2	3
Bearn	22	22	22	22
Yellow corn	8.45	8.49	8.63	8.63
Soybean meal, 44 % crude protein	17.2	17.2	17.15	17.05
Wheat bran	14	13	11.9	11
Molasses	3.5	3.5	3.5	3.5
DL-Methionine	0.12	0.12	0.12	0.12
Vitamins and minerals mixture	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50
Limestone	1.2	1.2	1.2	1.2
Di calcium phosphate	0.50	0.50	0.50	0.50
Total	100	100	100	100
Calculated analysis[1]				
Dry matter. %	86.87	86.73	86.59	86.46
Organic matter, %	80.90	79.90	78.86	77.84
Crude protein, %	17.00	17.02	17.02	17.00
Ether extract, %	2.67	2.85	3.03	3.22
Nitrogen-free extract, %	47.72	46.43	44.96	43.61
Ash, %	5.97	6.82	7.74	8.62
Digestible energy [2], kcal/kg	2508	2512	2503	2501
Crude fiber, %	13.50	13.61	13.83	14.00
Neutral detergent fiber, (NDF)%	37.84	37.74	37.60	37.88
Acid detergent fiber, (ADF)%	21.80	21.66	21.48	21.86
Hemicellulose %	16.04	16.08	16.12	16.02
Calcium, %	1.02	0.885	0.754	0.659
Total phosphorus, %	0.519	0.495	0.467	0.424
Methionine	0.36	0.34	0.33	0.31
Lysine	0.81	0.72	0.64	0.56
Digestible energy: Crude protein	147.5	147.5	147.1	147.1

G0: 0 % MWF, G1:1 % MWF, G2: 2 % MWF, and G3:3 % MWF. 1. According to, 2. Calculated according to Cheeke [24]: DE (Kcal/g) = 4.36–0.0491 (%NDF), NDF % = 28.924 + 0.657 (crude fiber %), ADF % = 9.432 + 0.912 (crude fiber %), ADF = cellulose + lignin, and hemicellulose = NDF % - ADF %.

2.4. Carcass traits

Following the finishing of the feeding experiments, a selection of three rabbits from each dietary group were chosen at random to assess the carcass traits. The rabbits had a fasting period of around 12 h before their pre-slaughter weight was measured separately. Following the process of fully removing blood and skin, the carcass without any internal organs was weighed individually, as by Cheeke's [24] instructions.

2.5. Blood sampling and analysis

Each blood sample was obtained from separate rabbits that had been slaughtered. The samples were collected in centrifuge tubes that were free of moisture and contained a small amount of heparin solutions. The tubes were then centrifuged at a speed of 3000 rpm (revolutions per minute) for 20 min. This process was carried out to separate the blood plasma. The resulting plasma was subsequently stored in a deep freezer at a temperature of approximately −20 °C until it was analyzed to determine the different blood parameters. The levels of total proteins, albumin, glucose, aspartate aminotransferase (AST), alanine aminotransferase (ALT), cholesterol, triglycerides, and urea were measured using calorimetric methods. Chemical commercial kits from Diamond Diagnostics, Egypt, were used, and the procedures followed the instructions provided by the manufacturers. Plasma total proteins were assessed using the method described by Armstrong and Carr [25], albumin was measured following the procedure outlined by Doumas et al. [26], AST and ALT levels were determined based on the method developed by Reitman and Frankel [27], cholesterol was quantified using the Fossati and Principle [28] method, triglycerides were measured according to Richmond [29], and glucose and urea levels were determined following the procedure described by Siest et al. [30].

2.6. Statistical analysis

The data were examined for all variables using the general linear model approach in SAS software version 9.1 (SAS Institute, USA) [31] to determine the differences between means.

The utilized model was: $Y_{ij} = \mu + T_i + e_{ij}$

The equation represents the relationship between the observation Y_{ij}, the general mean μ, the effect of treatment T_i, and the experimental random error e_{ij}. The percentages were subjected to an arc-sin transformation to mimic a normal distribution before analysis. The comparison of variables that showed a significant F-test was conducted using Duncan's multiple-range test [32]. All declarations of statistical significance were determined using a probability level of ($P < 0.05$).

3. Results and discussion

3.1. Chemical composition of feedstuffs and diets

Frass is the excrements and shedded exoskeletons of larvae of a mealworm (*Tenebrio molitor* L.), fed exclusively on agricultural by-products or agri-waste. Also, frass is gaining recognition as a valuable component in animal feed. The chemical constituents of mealworm frass (MWF) and the experimental diets may be found in (Table 2). The findings indicated that MWF has a high nutritional content. The reported values for dry matter (DM), organic matter (OM), crude protein (CP), crude fibre (CF), ether extract (EE), Ash, and nitrogen-free extract (NFE) were 87.08 %, 84.98 %, 22.59 %, 17.62 %, 2.78 %, 17.24 %, and 41.99 %, respectively. Mealworm frass is rich in essential nutrients and bioactive compounds, offering potential benefits for animal nutrition and contributing to more sustainable agricultural practices [2,33]. The MWF boasts a robust profile that can enhance the quality of animal diets. It contains significant amounts of protein, ranging from 5 % to 15 %, which is particularly beneficial for the growth and development of livestock, especially in protein-intensive sectors like poultry and aquaculture [2]. The fiber content in frass, mainly from chitin, a component of mealworm exoskeletons, acts as probiotics, promoting gut health and enhancing immune responses [34].

The chemical composition of the experimental diets (Table 2) exhibited little variation in the percentages of DM, OM, CP, and NFE from G0 to G3. The control diet (G0) had the highest recorded values of DM, OM, and EE at 90.57 %, 89.96 %, and 5.25 %, respectively. In contrast, G3 exhibited the highest percentages of CP, CF, and ash, which were 19.51 %, 12.49 %, and 11.30 %, respectively.

Table 2
Chemical composition of ingredients and experimental diets (% on DM basis).

Item	DM	OM	CP	CF	EE	NFE	Ash
MWF	87.08	84.98	22.59	17.62	2.78	41.99	17.24
Chemical composition of experimental diets							
G0	90.6	90	19.1	12.55	5.3	53.1	10.0
G1	90.7	89.2	19.4	12.16	4.5	53.1	10.9
G2	89.6	88.9	19.0	12.17	4.6	53.1	11.1
G3	89.3	88.7	19.51	12.49	4.3	52.4	11.3

MWF: mealworm frass, G0 (control diet): diet contains 0 % MWF, G1: diet contains 1 % MWF, G2: diet contains 2 % MWF, and G3: diet contains 3 % MWF. DM: dry matter; OM: organic matter; CP: crude protein; CF: crude fibre; EE: ether extract; NFE: nitrogen-free extract.

3.2. Digestion coefficients and feeding values of experimental diets

Table (3) displays the coefficients of digestibility and nutritional values of the experimental diets. The inclusion of MWF in experimental diets led to a significant improvement in the digestibility coefficients, higher than those of the control diet (G0). The diet containing 3 % MWF (G3) had the highest levels of all digestible nutrients. Palatability is considered to be the combined effect of several dietary characteristics, and there exists an important relationship between taste and nutritional value [35]. The utilization of MWF in the feeds increased voluntary feed intake, suggesting an enhancement in the palatability of the frass-based diets for rabbits. Total digestible nutrients (TDN) values for the experimental groups exhibited substantial differences ($P < 0.05$), with G2 and G3 having the greatest TDN values compared to the other groups. The digestible crude protein (DCP) showed an increase ($P < 0.05$) as the level of frass in the experimental diets was rising.

3.3. Productive performance

The effect of different experimental diets on body weight, total weight gain, daily weight gain, daily feed intake, and feed conversion is shown in Table (4). The results indicated that there were no significant differences in body weight across the treatment groups at 6, 10, and 15 weeks of age. However, the group defined as G3 had the greatest body weight of 2023 g at 15 weeks of age. During the periods of 6–10 and 10–15 weeks, G3 had the largest daily weight gain, recording 23.87 g and 25.75 g, respectively. Additionally, G3 had the highest total weight gain of 668.46 g and 1384.62 g during the periods of 6–10 weeks and 6–15 weeks, respectively. The digestibility coefficients of most nutrients and feeding values were enhanced by the examined groups G1 to G3, leading to an increase in body weight, total body weight gain, and daily weight gain. Regarding the daily consumption of food, there were no significant differences identified among the different groups in the trial. Feeding rabbits with diets containing frass resulted in improved feed conversion, especially for G3 (3.14 and 3.64 g feed/g daily gain), compared to the control group (4.12 and 4.64g feed/g daily gain), at 6–10 and 6–15 weeks of age, respectively. These differences among treatment groups were significant ($P < 0.05$). The observed outcomes could be attributed to the enhancement in overall growth during the entire experimental duration when fed a diet of 3 % MWF (G3). The inclusion of MWF in the diets enhanced the growth performance of Gabaly rabbits. High weight growth was seen following 15 weeks of feeding with the 3 % MWF diet. The enhancement in growth was positively associated with an increase in the digestibility of diets containing MWF. The utilization of MWF in the feeds increased voluntary feed intake, suggesting an enhancement in the palatability of the frass-based diets for rabbits. Gasco et al. [36] discovered that there were no apparent differences in growth performance between the control group and the experimental group when soybean oil was replaced with lipids from yellow mealworm larvae. The results of this study are consistent with the findings of Ayaz et al. [9], which showed that adding mealworm frass to the diet substantially improved the growth and weight gain of sheep. Several studies have already assessed the possibility of mealworm larvae meals as a substitute protein source for poultry. The majority of trials suggest replacement rates ranging from 25 % to 30 %. The study conducted by Hu et al. [37] found that substituting 25 % of fish meals with black soldier fly larvae meal in yellow catfish (*Pelteobagrus fulvidraco*) did not result in any significant variation in the growth index when compared to the control group. The inclusion of *T. molitor* larvae at a rate of 6 % in pig diets resulted in increased body weight , average daily gain, acid detergent fiber intake, and gain-to-feed ratio (G:F ratio) in weaning pigs (0–5 weeks after weaning) [38]. In their study, Chen et al. [39] found that increasing the concentration of *T. molitor* protein up to 6 % in the diets of weaned pigs resulted in a linear improvement in both body weight and body weight gain. Strychalski et al. [40] discovered that rabbits fed mealworm larvae had significantly higher final body weight and daily body weight gains compared to the control group. In addition, they were observed to have superior apparent total tract digestibility of either extract, acid detergent fiber, and acid detergent lignin.

Table 3
Digestion coefficients and feeding values of experimental diets.

Item	Experimental diets				SEM
	G0	G1	G2	G3	
Digestibility (%)					
DM	67.3^{b}	70.5^{b}	74.8^{a}	76.2^{a}	1.18
OM	69.4^{b}	70.2^{b}	76.3^{a}	77.6^{a}	1.18
CP	69.8^{b}	74.9^{ab}	76.8^{a}	78.7^{a}	1.97
CF	28.3^{b}	32.6^{b}	47.9^{a}	43.0^{a}	2.82
EE	76.0^{b}	76.8^{b}	85.0^{a}	80.1^{ab}	1.78
NFE	78.3^{b}	77^{b}	81.7^{ab}	85.2^{a}	1.65
Feeding values (%)					
TDN	67.5^{b}	64.1^{b}	74.8^{a}	73.14^{a}	1.07
DCP	13.4^{b}	14.5^{ab}	14.6^{ab}	15.35^{a}	0.38
DE kcal/kg	2988^{b}	2840^{b}	3313^{a}	3240^{a}	47.40

G0 (control diet): diet contains 0 % MWF, G1: diet contains 1 % MWF, G2: diet contains 2 % MWF, and G3: diet contains 3 % MWF. a and b means in the same row for each parameter with different superscripts are significantly different ($P < 0.05$). SEM = standard error of mean.
DM: dry matter; OM: organic matter; CP: crude protein; CF: crude fibre; EE: ether extract; NFE: nitrogen-free extract; TDN: total digestible nutrients; DCP: digestible crude protein; DE: digestible energy.

Table 4
Effect of different experimental diets on growth performance for growing Gabaly rabbits through different ages.

Item	Experimental groups				SEM	P-value
	G0	G1	G2	G3		
Live body weight (g)						
Initial (6 weeks)	640^{a}	678^{a}	659^{a}	638^{a}	15.85	0.336
10 Weeks	1249^{a}	1227^{a}	1316^{a}	1307^{a}	41.32	0.572
15 Weeks	1874^{a}	1946^{a}	1971^{a}	2030^{a}	59.39	0.143
Total weight gain (g)						
6–10 weeks	609^{a}	591^{a}	641^{a}	656^{a}	38.50	0.296
10–15 weeks	625^{a}	7355	675^{a}	7136^{a}	45.50	0.154
6–15 weeks	1234^{a}	1325^{a}	1316^{a}	1392^{a}	54.92	0.102
Average daily gain (g)						
6–10 weeks	20.3^{a}	19.7^{a}	21.4^{a}	21.9^{a}	1.37	0.296
10–15 weeks	20.8^{a}	24.5^{a}	22.5^{a}	24.5^{a}	1.63	0.154
6–15 weeks	20.6^{a}	22.1^{a}	21.9^{a}	23.2^{a}	0.87	0.102
Daily feed intake (g)						
6–10 weeks	81.3	77	77.8	73.7		
10–15 weeks	94.3	99	89.4	83.8		
6–15 weeks	88.5	89.3	84.2	79.3		
Feed conversion (g feed/g gain)						
6–10 weeks	4.1^{a}	4.3^{a}	3.4^{ab}	3.1^{b}	0.32	0.035
10–15 weeks	4.3^{a}	4.2^{a}	3.8^{a}	3.4^{a}	0.32	0.212
6–15 weeks	4.6^{a}	4.5^{ab}	4^{bc}	3.6^{c}	0.20	0.023

G0 (control diet): diet contains 0 % MWF, G1: diet contains 1 % MWF, G2: diet contains 2 % MWF, and G3: diet contains 3 % MWF. a, b, and c mean in the same row for each parameter with different superscripts are significantly different ($P < 0.05$). SEM = standard error of mean.

3.4. Carcass traits

The data displayed in (Table 5) demonstrated that feeding different experimental diets affected the carcass characteristics of growing Gabaly rabbits. The findings showed that, except for dressing percentage, total edible giblets, and non-edible giblets, there were no significant differences between the experimental groups in all carcass traits studied. The rabbit-fed control diet (G0) had a higher ($P < 0.05$) dressing percentage and total edible giblets values (60.3 and 64.4 %) respectively, compared with G1, which had the lowest values (57.9 and 62 %, respectively), but with insignificant differences with from other groups. On the other

Table 5
Effect of feeding different experimental diets on carcass traits for growing Gabaly rabbits.

Item	Experimental diets				SEM	P-value
	G0	G1	G2	G3		
Pre-slaughter weight, g	1900[a]	1980[a]	1933[a]	1866[a]	67.0	0.292
Fur weight, g	266.8[a]	217.4[a]	225.7[a]	241.3[a]	18.55	0.111
Head weight, g	121.0[a]	131.7[a]	121.7[a]	109.3[a]	7.07	0.067
Liver weight, g	57.7[a]	59.7[a]	53.7[a]	58.7[a]	3.30	0.305
Liver %	3.03[a]	3.01[a]	2.78[a]	3.14[a]	0.11	0.305
Kidney weight, g	13.1[a]	13.8[a]	12.8[a]	13.6[a]	1.28	0.594
Kidney %	0.69[a]	0.69[a]	0.66[a]	0.73[a]	0.05	0.594
Heart weight, g	6.80[a]	6.80[a]	5.23[a]	6.17[a]	0.56	0.102
Heart, %	0.36[a]	0.34[a]	0.27[a]	0.33[a]	0.03	0.102
Carcass weight- without head, g	1025[a]	1016[a]	1023[a]	993[a]	40.12	0.612
Dressing, %	60.3[a]	57.9[a]	59.2[a]	59[a]	0.64	0.059
Edible giblets, %	4.08[a]	4.05[a]	3.7[a]	4.2[a]	0.15	0.086
Total edible, %	64.4[a]	62[a]	62.9[a]	63.2[a]	0.64	0.110
Non-edible, %	33.1[a]	33[a]	31.2[a]	33.9[a]	0.64	0.237

G0 (control diet): diet contains 0 % MWF, G1: diet contains 1 % MWF, G2: diet contains 2 % MWF, and G3: diet contains 3 % MWF.
Total edible parts wt. = carcass wt.(with head) + edible giblets wt.; Edible giblets % = (Liver wt. + kidney wt. + heart wt.)/slaughter wt.*100; Total edible parts % = Total edible parts wt./slaughter wt. x100; Non-edible % = (Skin wt. + legs wt. + Gut truck wt. + lung wt.)/slaughter wt.*100.
a, b means in the same row for each parameter with different superscripts are significantly different (P < 0.05).
SEM=Standard error of the mean.

hand, the rabbits fed a control diet recorded a significant reduction in non-edibles compared to those fed G1. It could be concluded that MWF levels had no significant effect on most carcass traits, regardless of the dressing percentage, total edible giblet percentage, and non-edible giblets. Dietary inclusion of MWF did not influence the carcass traits of the rabbits. This agrees with Bovera et al. and Biasato et al. [41,42], who did not find any differences in carcass traits of chickens. A similar trend was given by Hwangboet et al. and Cullere et al. [43,44], who did not observe any effects after house-fly maggots and black soldier fly meal inclusion, respectively, in the diet of broiler chickens and quails. On the contrary, Ballitoc and Sun [45] found improved slaughter, dressed carcasses, and eviscerated weights in broiler chickens fed *T. molitor* diets. A similar consideration can also be applied to explaining the growth performance observed in the current study. Indeed, the overall feed conversion ratio increased linearly with increased levels of *T. molitor* meal, reaching its maximum with the inclusion of 15 % of mealworms. The live weight in the finisher period also increased quadratic ally up to a 5 % level of *T. molitor* meal inclusion, then again decreasing up to the inclusion of 15 % of insects. Furthermore, the alterations in intestinal morphology can also justify the observed growth performance. Indeed, since it is well known that the rapid growth of chickens directly depends on the morphological and functional integrity of the digestive tract [46], the relationship between the altered gut morph metric indices and the worsening of the growth performance seems reasonable.

3.5. Blood parameters

Blood analysis is an efficient and fast way to assess the animal's clinical and nutritional condition during a feeding study as outlined by Olabanji et al. [47]. The hematological and serum profiles can assess rabbit health and reveal stress-related metabolites [48]. Table 6 displays the blood parameters. The blood parameters exhibited levels that fell within the usual range [49]. There were no significant differences among the groups evaluated for the majority of the estimated blood parameters, except globulin, ALT, and glucose. These three parameters were significantly decreased (P <0.05) in G3 compared to the control group (G0). The ALT values in MWF rabbits twere similar to the control group (G0), and it is important to note that external factors like the environment, management, and feeding might impact metabolism and have an influence on blood parameters. Protein fractions are used to evaluate the impact of MWF on the immunological function of animals. Bovera et al. and Biasato et al. [42,50] determined that *T. molitor* is safe in poultry diets. Insect protein, apart from unicellular protein (single--cell protein) from bacteria, algae, and yeasts, might become an important feed ingredient in the near future, replacing unsustainable fish meal, not only in aquaculture and avarian species, but also for other animals like rabbits.

Table 6
Blood plasma parameters as affected by different experimental diets.

Item	G0	G1	G2	G3	SEM	P-value
Total protein, g/dL	4.58[a]	4.55[a]	4.75[a]	4.65[a]	0.1	0.09
Albumin (A), g/dL	3.05[a]	3.0[a]	3.08[a]	3.46[a]	0.2	0.16
Globulin (G), g/dL	1.49[a]	1.66[a]	1.88[a]	1.42[a]	0.1	0.09
A/G	1.99	1.93	1.84	2.91	–	–
Urea, mg/dL	3.48[a]	4.05[a]	4.00[a]	3.26[a]	0.3	0.18
AST, IU/L	60.8[a]	58.7[a]	64.0[a]	66.6[a]	4.1	0.557
ALT. IU/L	86.5[a]	71.5[ab]	53.5[b]	50.0[b]	8.4	0.04
Glucose, mg/dL	107[a]	106[a]	103[a]	91[b]	2.8	0.01
Cholesterol, mg/dL	254[a]	251[a]	226[a]	207[a]	17.7	0.269
Triglyceride, mg/dL	210[a]	217[a]	214[a]	215[a]	3.1	0.399

G0 (control diet): diet contains 0 % MWF, G1: diet contains 1 % MWF, G2: diet contains 2 % MWF, and G3: diet contains 3 % MWF. a and b means in the same row with different superscripts are significantly ($P \leq 0.05$) different.SEM = standard error of mean. A/G: Albumin/Globulin ratio.AST: aminotransferase; ALT: alanine aminotransferase.

3.6. Economical evaluation

Table 7 displays the economic aspects of offering experimental diets. The economics of incorporating MWF into rabbits' meals is contingent upon the cost of the studied diets and the resulting growth performance. The cost of 1 kg of feed was 5.62, 5.30, and 4.99 Egyptian pounds (L.E.) for G1, G2, and G3, respectively, in comparison to the control group G0, which had a cost of 5.57 Egyptian pounds. The inclusion of MWF at a maximum of 3 % in the diet of Gabaly rabbits resulted in a higher cost per kilogram of diet. However, the rabbits that were fed on frass diets showed improved weight gain, leading to a decrease in the cost per kilogram of weight gained. Total revenue and net revenue were increased by increasing frass levels in rabbit diets, particularly in G3 (62.3 and 39.3 LE./head, respectively) compared to the control group (56 and 30.8

Table 7
Effect of feeding different experimental diets on economic efficiency for growing Gabaly rabbits.

Item	Experimental diets			
	G0	G1	G2	G3
Price/kg diet, (L.E	4.5	4.5	4.5	4.6
Total feed intake/rabbit, g	5576	5624	5307	4996
Total feed cost/rabbit, L.E	25.2	25.6	24.0	23.0
Total weight gain/rabbit, g	1244	1295	1341	1385
Feed cost/kg gain, L.E	20.2	19.7	18.1	16.6
Total revenue/rabbit, L.E	56.0	58.3	60.3	62.3
Net revenue/rabbit, L.E	30.8	32.7	36.1	39.3
Economic efficiency	1.22	1.28	1.49	1.71
Relative economic efficiency, %	100	105	122	140

G0 (control diet): diet contains 0 % MWF, G1: diet contains 1 % MWF, G2: diet contains 2 % MWF, and G3: diet contains 3 % MWF. Based on the prices of the Egyptian market during the experimental period (2022). Net revenue (L.E)= (Total revenue/rabbit (L.E))-(Total feed cost/rabbit (L.E.)).Economic efficiency = (Net revenue/rabbit(L.E))/(Total feed cost/rabbit (L.E)).Feed cost/kg gain = Total feed cost (L.E)/Total weight gain/rabbit (kg), the prices of live body weight of rabbits = 45 LE/kg.

LE./head, respectively). This increased the overall revenue per rabbit fed on frass diets, indicating an improvement in economic efficiency ranging from 5 to 40 % compared to rabbits fed the control diet.

These findings are consistent with previous studies by Van Huis et al. and Mertenat et al. [2,51]. Production using MWF is often more cost-effective compared to traditional feed crops such as soy or fish meal [51]. In addition, Van Huis et al. [2] noted that MWF's nutrient-rich composition can reduce the need for additional supplements, thereby lowering feed formulation costs. Moreover, incorporating frass improved feed conversion ratios, meaning animals required less feed to gain weight, thus enhancing production efficiency and profitability for farmers [34].

3.7. Minerals in rabbits' meat

Table 8 outlines the concentrations of various elements found in rabbit meat across different groups (G0, G1, G2, and G3). These elements are classified into essential elements, potentially toxic (heavy) metals, and others. None thereof were significantly influenced by the different diets, except for copper, which was notably higher in the control group (G0) at 87.9 mg/kg (P <0.001), though still lower than the concentration found in MWF (147.9 mg/kg).

Essential elements detected in the samples include calcium, copper, iron, magnesium, manganese, molybdenum, zinc) and cobalt). These minerals play vital roles in various physiological functions [52,53]. Conversely, potentially toxic elements, such as cadmium, lead, nickel, vanadium, aluminum, and barium, pose significant health risks when present in meat at elevated levels [54].

The analysis of these heavy metals in the meat samples raises considerable concerns about potential contamination and associated health risks [55]. Noteworthy heavy metals, including cadmium (Cd), lead (Pb), nickel (Ni), chromium (Cr), and cobalt (Co), were evaluated across the different treatment groups (G0, G1, G2, and G3), revealing varying concentrations of these elements.

Cadmium levels in the groups ranged from less than 0.003–6.93 mg/kg. Notably, the G2, G3, and G0 groups exhibited concentrations that significantly exceeded the EU and FAO/WHO safety limits of 0.05 mg/kg for meat, highlighting a severe contamination issue. Cadmium is recognized for its toxic effects, particularly on the kidneys and bones, making the elevated levels found in these samples a serious health risk. In contrast, the MWF frass contained less than 0.003 mg/kg.

Lead contamination was another major concern, with levels in all groups far exceeding the permissible limit of 0.1 mg/kg for muscle meat set by the EU and FDA. The observed concentrations (ranging from 127 to 168.3 mg/kg) suggest significant lead presence, which could lead to adverse health effects including neurological damage and developmental issues. Nevertheless, MWF frass had the lowest level of lead (16.8 mg/kg).

Nickel was consistently detected at high levels across all treatments, with concentrations varying between 343 and 362 mg/kg, including MWF frass within this range. While there are no established regulatory limits for nickel in meat, these elevated levels indicate a persistent contamination issue. Chronic exposure to high nickel concentrations may lead to allergic reactions and potentially toxic effects, underscoring the necessity for further investigation into its sources. Additionally, vanadium concentrations in the meat ranged from 3.77 to 31.07 mg/kg, which is relatively elevated compared to typical dietary exposure levels.

Chromium levels showed significant variation across different groups, with concentrations ranging from 5 to 42 mg/kg. In contrast, the MWF frass exhibited the lowest concentration at 2 mg/kg.

Cobalt is essential in trace amounts for the synthesis of vitamin B12; however, excessive intake can result in health problems, including thyroid dysfunction and cardiotoxicity. The low levels of cobalt

Table 8
Concentration of minerals (mg/kg) in rabbit meat across different experimental diets and mealworm frass (MWF).

Item	G0	G1	G2	G3	MWF	SEM	P value
Essential elements, mg/kg							
Calcium	3311	2258	2432	4319	134	35.6	0.128
Copper	87.9[a]	24.7[b]	25.0[b]	21.7[b]	147.9	4.61	0.001
Iron	182	204	278	218	1050	78.97	0.751
Magnesium	587	607	642	628	3680	53.79	0.141
Manganese	24.6	18.7	19.4	8.5	715.4	9.80	0.496
Molybdenum	36.8	9.6	11.6	25.7	51.8	25.71	0.560
Zinc	104	117	124	120	285	15.07	0.542
Cobalt	<0.002	<0.002	<0.002	<0.002	<0.002	–	–
Chromium	7.9	9.5	1.7	27.8	2.4	21.20	0.619
Heavy metals, mg/kg							
Aluminum	115.9	143.2	174.9	118.3	500.9	29.2	0.230
Barium	14.1	5.5	7.6	6.8	39.6	4.2	0.250
Lead	126.9	129.3	168.3	167.4	16.8	101.1	0.066
Cadmium	5	<0.003	4.5	6.9	<0.003	5.94	0.560
Nickel	354	343	355	362	342	34.68	0.909
Vanadium	18.3	28.9	3.8	31.1	35.10	26.84	0.679
Other elements, mg/kg							
Boron	36.6	48.6	21.9	33.3	95.5	10.7	0.178
Strontium	11.7	8.2	5.4	12.6	224.6	2.38	0.173

G0 (control diet): diet contains 0 % MWF, G1: diet contains 1 % MWF, G2: diet contains 2 % MWF, and G3: diet contains 3 % MWF. a and b means in the same row for each parameter with different superscripts are significantly different (P < 0.05). SEM = standard error of mean.

observed across treatments indicate a minimal risk associated with this metal. Generally, cobalt was found at low concentrations in all groups, including MWF frass, measuring less than 0.002 mg/kg.

Previous research suggests that MWF frass improves growth performance and economic efficiency without negatively impacting animal health, as evidenced by blood parameters (see Table 6). Additionally, frass is abundant in essential minerals such as calcium, phosphorus, and magnesium, along with trace elements like iron, zinc, and manganese, all of which are vital for maintaining various physiological functions in animals [56].

However, despite these advantages, challenges remain regarding the widespread adoption of frass.

4. Conclusions

This study indicates that incorporating mealworm frass (MWF) at a maximum level of 3 % into the diets of growing Gabaly rabbits can improve performance without adversely affecting the rabbits' health.

CRediT authorship contribution statement

The formulation of abstract ideas and concepts was conducted by H. A.M.H., M.H.A., and A.M.H. The work involved data curation, formal analysis, and various techniques. The contributors to this research include N.E.M.E, Y.L.P, A.A, M.A.Z, and A.S.E., all of whom played a significant role in the investigation and the writing of the original draft. H.A.M.H., Y.L.P., M.A.R., G.T.E., M.L., and A.Z.M.S. were responsible for writing, reviewing, and editing the manuscript. All authors have reviewed and approved the final version of the manuscript that has been published.

Informed consent statement

Not applicable.

Ethics statement

The Animal Production Research Institute, Agricultural Research Center, Egypt ethical committee has approved the animal investigations. The code is ARC-AP-22-26. Rabbit.

Data availability statement

The data presented in this study are available on request from the corresponding author.

Funding

This study did not get any financial support from other sources.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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