

Evaluating of *Nigella sativa* meal (black cumin meal) in lamb diets and its impact on the growth performance, digestion coefficient of nutrients, nitrogen balance and rumen fermentation

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ABSTRACT

The purpose of this study was to evaluate the effects of adding *Nigella sativa* meal (NSM) at varying levels on the performance of growth, digestibility of nutrients, nitrogen balance and rumen fermentation of growing lamb. Thirty-five Rahmani lambs were divided into five groups at random for 90 days. NSM was added to the diets of lambs at 0, 6, 12, 18 and 24% for G1 (control), G2, G3, G4 and G5, respectively. The parameters measured were weight gain, feed consumption, digestibility of nutrients, nitrogen balance, and rumen fermentation. Lambs fed with NSM diets had significantly higher weight gain, feed intake, and feed efficiency compared with the control group. Nutrient digestibility like dry matter, crude protein, fat, fiber, and carbohydrates were improved with the addition of NSM. Rumen values revealed increased ammonia and volatile fatty acid concentrations, with reduced protozoa and bacterial numbers. It could be concluded that, the inclusion of 18% NSM proved to be the most effective in enhancing final weight, nutrient digestibility, and rumen function.

Introduction

An extreme lack of farm animal feed is encountering in the whole world. So, researchers are looking for better and more innovative feed solutions that could improve animals' access to nutrients and have other positive effects on growth performance (Melody and Amit, 2021; Yadav *et al.*, 2021; Kisku and Singh, 2022). Efforts in livestock farming aim to lower feed costs, increase production efficiency, and maximize the utilization of dietary protein (Schwab and Broderic, 2017). The rising costs and fluctuating supply of traditional feed ingredients (corn grains and soy-bean meal) as source of protein have driven interest in replacing it by alternative feeds may enhance product quality and lower production costs (Al-Khaza'leh *et al.*, 2015; Obeidat, 2020).

Nigella sativa meal (NSM) is one of the alternative feed sources that can be used as a feed supplement for farm animals, including lambs (Obeidat, 2020; 2021). The NSM has been produced from its seeds for extracting *Nigella sativa* oil for human health supplies. The processing of *Nigella sativa* seeds generates a large quantity of waste known as NSM. This waste could be used as a supplement to animal feed (Obeidat, 2020). The herb *Nigella sativa* belongs to the Ranunculaceae family (Ahmad *et al.*, 2021). Numerous nations, including Egypt, Saudi Arabia, India, and Pakistan, cultivate it. Black cumin seed (*Nigella sativa*), known as a panacea, has long been utilized in medicine to treat a wide range of illnesses in several forms, including extract, paste, powder, and essential oil (Hannan *et al.*, 2021; Obeidat and Alqudah, 2023). The process of extraction of oil from the seeds resulted in a large biomass production (e.g., meal), which represent about 60 to 68% of the seeds (Kour and Gani, 2020). The composition of this meal includes 93.18% dry matter, 11.75% ether extract, 31.56% crude protein, 6.41% crude fiber, 7.12% ash, 1.16% lysine, 0.56% DL-methionine, 0.27% Ca and 0.35% P with a reasonable amount of antioxidants (Dabeer *et al.*, 2022; Fathi *et al.*, 2023).

The high protein content (more than 30%) of NSM was attractive

for using it in animal nutrition (Zaky *et al.*, 2021). Lambs can be successfully fed NSM, which is a good source of protein (Ramdani *et al.*, 2024) because of its rich amino acid content. Furthermore, black cumin seed meal is a great substitute for animal feed because it contains natural products and secondary metabolites that are high in nutrients like crude protein (CP), essential amino acids, and vital lipids (Obeidat, 2020). Small ruminants' nutritional intake, nutrient digestibility, growth, immunity, and gastrointestinal health were all enhanced by *Nigella sativa* (Singh *et al.*, 2022). Additionally, earlier research found that feeding NSM as a substitute meal to Awassi growing lambs enhanced their growth performance, feed consumption, and feed efficiency (Obeidat, 2020; 2021). It has been demonstrated that adding 1–35% *Nigella sativa* meal improves animal performance (Abd El-Rahman *et al.*, 2011).

The degree to which *Nigella sativa* meal can be included in the lamb diets remains unclear. Consequently, this study aimed to examine the effect of *Nigella sativa* meal in the diets of growing lambs on their growth performance, nutrient digestibility, nitrogen balance, and rumen fermentation for can determine the optimum level of NSM which can be used.

Materials and methods

Ethical approval

The Ethics Committee of the Faculty of Veterinary Medicine, Assiut University approved the animal care and use procedures during running this experiment in compliance with Egyptian laws and regulations (Ethical Approval No. 06/2025/0371).

Animals and experimental diets

This research involved 35 healthy Rahmani ram lambs at Faculty of Veterinary Medicine, Assiut University, Assiut city, Egypt (The coor-

dinates for latitude and longitude are 27.180134 and 31.189283). They were 10.0 ± 1.3 months old on average and weighed roughly 30 ± 1.5 kg. Lambs were in good health and exhibited typical signs of health prior to and throughout the experiment. They were vaccinated and dewormed based on a veterinarian's guidance before the experiment. Each lamb was housed in its own space with separate feeding and drinking arrangements. Natural light cycles and temperatures were maintained, with constant access to fresh water. The lambs' daily diets, which were delivered at 4% of body weight per day and divided into two feedings at 8:00 a.m. and 5:00 p.m. during a 90-day period, were established in accordance with the NRC (2007) guidelines for sheep (Table 1). Feed remnants were collected and weighed consistently. At the start and finish of the trial, lamb weights were recorded, and feed consumption was continuously monitored.

Table 1. Ingredients and composition (%) of experimental diets.

	Experimental diets					NSM
	1	2	3	4	5	
Corn, ground	48.24	47.37	46.5	45.55	44.61	
Soybean meal	20	15	10	5	0	
<i>Nigella sativa</i> meal	0	6	12	18	24	
Wheat straw	30	30	30	30	30	
Limestone, ground	0.96	0.83	0.7	0.59	0.46	
Monosodium phosphate	0	0	0	0.06	0.13	
Common salt	0.5	0.5	0.5	0.5	0.5	
Premix*	0.3	0.3	0.3	0.3	0.3	
Total	100	100	100	100	100	
Chemical composition						
Dry matter	87.43	87.91	88.39	88.79	89.21	95
Crude protein	14.7	14.7	14.7	14.71	14.72	37.5
Ether extract	2.62	3.12	3.63	4.14	4.65	10.26
Crude fiber	13.54	13.95	14.36	14.77	15.18	12.2
Nitrogen free extract	65.04	63.98	62.97	61.93	60.9	32.93
Ash	4.14	4.25	4.35	4.45	4.55	7.11
Calcium	0.51	0.51	0.51	0.51	0.51	1.13
Phosphorus	0.28	0.27	0.25	0.24	0.24	0.35
ME (Mcal/Kg)	2.45	2.47	2.49	2.5	2.52	3.19

*Premix consist of (per 3kg): 20,000,000 IU vitamin A, 10,000 mg vitamin E, vitamin 200,000 IU D3, 10,000 mg Fe, 2500 mg Cu, 100 mg Mo, 20,000 mg Mn, 100 mg Co, 800 mg I, 20,000 mg Zn and 100 mg Se.

Experimental design

Before the experiment began, the lambs were given two weeks to adjust and were then randomly distributed to one of five groups (7 lambs per each). Specifically, group 1 received a basal diet without *Nigella sativa* meal and considered as control. Group 2 diet included 6% NSM, group 3 diet contained 12% NSM, group 4 diet had 18% NSM, and group 5 diet had 24 % NSM, respectively.

Performance parameters

The body's starting weight, final weight, and daily & total feed intake were recorded during the experiment. The average daily gain and cumulative body weight gain and the feed conversion ratio (FCR) were also calculated.

Digestibility of nutrients and nitrogen balance

At the end of the growth period, five lambs from each treatment group were chosen to assess nutrient digestibility and nitrogen balance. The selected lambs were housed in individual metabolic cages, each equipped with separate containers for urine and feces collection. Before

data collection, lambs underwent a two-week adaptation period in the pens to acclimate to the experimental conditions.

Throughout the daily collection phase, fecal samples were collected in plastic bags, with each sample weighing approximately 50g. Immediately after collection, 5 % of the sample was frozen at -20°C for later nitrogen content analysis. Additionally, 10 % of the feces were air-dried and stored in plastic buckets until the collection period ended, with daily measurements recorded for accuracy.

For urine collection, a unique setup was employed. Urine was directed through a curved plastic plate, positioned beneath the crate floor, into a 2.5-liter wide-mouth bottle (equipped with a plastic funnel) containing 5 ml of sulfuric acid to prevent nitrogen loss. The collected urine was filtered using glass wool to remove any hair or solid contaminants, and the volume was recorded using a calibrated cylinder. From each day's collection, 20% of the urine volume was sampled and stored at -20°C , with all daily samples from each lamb combined into one container for later nitrogen analysis. The dry matter and other nutrient digestibility coefficients were calculated following the methods outlined by Maynard *et al.* (1979). Feed, urine, fecal, and refusal samples from the digestibility trial were analyzed for nitrogen content using the standard procedures according to the AOAC guidelines (2023).

Chemical analysis

Both feed and feces were proximally analyzed for crude fiber, dry matter, crude protein, ether extract, and ash in accordance with AOAC guidelines (2023).

Rumen parameters

At the conclusion of the digestibility test, three to four hours following the morning meal, rumen content samples were taken from each lamb using a stomach tube. The samples' pH was promptly determined using a digital pH meter (Mettler-Toledo Ltd., England). The samples were then filtered through sterile gauze and separated for various analyses. Following the procedure described by Clergue *et al.* (2023), about 2 ml was used to test the concentrations of volatile fatty acids, another 2 ml for the concentration of ammonia, and 2 ml was fixed and stained for microscopic counting of total protozoa. The total bacterial count was evaluated using plate count agar after a two-day incubation at 30°C , as described by Wang *et al.* (2012). For biochemical analysis, total volatile fatty acids (TVFAs) were measured using the Macro Kjeldahl steam distillation method from Hall *et al.* (2015), while ruminal ammonia nitrogen ($\text{NH}_3\text{-N}$) concentrations were determined using the NH_3 diffusion technique with Kjeldahl distillation, according to AOAC (2023) guidelines.

Statistical analysis

SPSS version 16 was used to conduct statistical analysis (SPSS Inc., Chicago, IL, USA). All measurements are regularly distributed according to the data's normality assessment before analysis. The data was analyzed using a one-way analysis of variance (ANOVA), and any significant differences between treatments were then determined using Tukey's test. The results are shown as mean \pm standard error (SE), and $P < 0.05$ is considered statistically significant.

Results

Performance parameters

Supplementations of NSM significantly affect the performance parameters, such as body weight, weight gain, feed intake, and feed conversion rates (Table 2). The inclusion of 18% NSM exhibited the most favorable outcomes in terms of body weight, weight gain, and feed conversion

efficiency, in contrast to the control group, which registered the least favorable results. All the previous parameters were decreased significantly with high level of NSM (24%).

Nutrient digestibility

Compared to the control group, the addition of NSM resulted in a statistically significant improvement ($P < 0.05$) in the nutrient digestion coefficients (Table 3). The digestibility of ether extract and dry matter varied significantly ($P < 0.05$) between treatments, with the highest digestibility found in the 18% NSM group and the lowest in the control group. In addition, there were notable differences ($P < 0.05$) in the digestion coefficients for crude protein, crude fiber, and nitrogen-free extract, with the control group showing the lowest values and the 24% NSM group the highest. However, when compared to the other treatment groups, the 24% NSM group's digestion coefficients for ether extract and dry matter were lower.

Nitrogen Balance

Nigella sativa meal (NSM) supplementation had a significant ($P < 0.05$) impact on nitrogen intake and retention (Table 4). Compared to the control group, lambs fed NSM diets had significantly lower urinary nitrogen levels and greater fecal nitrogen levels ($P < 0.05$). The highest nitrogen

retention was seen in lambs fed on the 18%NSM diet, while the control and high level of NSM (24%) groups had the lowest retention.

Rumen Parameters

The rumen pH values ranged from 6.35 to 6.40, within the normal range, with no significant differences ($P > 0.05$) (Table 5). Supplementation of diets with NSM significantly improved ruminal ammonia (NH_3) concentrations ($P < 0.05$), ranging from 20.10 to 22.81 mg/100ml. The 24% NSM group showed the highest NH_3 concentration, while the control group had the lowest. Bacterial and total protozoa count dropped significantly ($P < 0.05$) as NSM levels increased, with the control group having the highest counts and the 24% NSM group having the lowest. The concentrations of total and partial volatile fatty acids (VFA), such as acetate, propionate, and butyrate, were also found to differ significantly ($P < 0.05$). The 18% NSM group had the highest VFA concentrations, while the control and high level of NSM (24%) groups had the lowest values.

Discussion

Dietary inclusion of *Nigella sativa* meal (NSM) to lamb diets significantly improved body weight, weight gain, feed intake, and feed conversion efficiency ($P < 0.05$). In our study *Nigella sativa* meal increased weight gain which agreed with the previous studies in lamb (Obeidat,

Table 2. *Nigella sativa* meal supplementation's impact on lambs' growth performance.

Items	Experimental groups					SEM**	P Value
	1	2	3	4	5		
Initial body weight (Kg)	30.05	30.46	30.4	30.25	30.33	0.22	0.72
Final body weight (Kg)	40.20 ^{d*}	40.51 ^d	42.57 ^b	44.61 ^a	41.25 ^c	0.23	0.00
Total weight gain (Kg)	10.15 ^d	10.05 ^e	12.17 ^b	14.36 ^a	10.92 ^c	0.02	0.00
Average daily gain (g)	112.78 ^d	111.67 ^c	135.22 ^b	159.56 ^a	121.33 ^c	0.17	0.00
Total feed intake (Kg)	84.45 ^d	84.22 ^d	97.78 ^b	105.25 ^a	91.98 ^c	0.16	0.00
Daily feed intake (g)	938.33 ^d	935.74 ^d	1086.44 ^b	1169.41 ^a	1022 ^c	1.83	0.00
Feed conversion ratio	8.32 ^c	8.38 ^b	8.03 ^d	7.33 ^e	8.42 ^a	0.01	0.00

*Means in the same row with different superscripts are significantly different ($P < 0.05$). **SEM, pooled standard errors of means.

Table 3. Effect of *Nigella sativa* meal supplementation on the nutrient digestibility.

Items	Experimental groups					SEM	P-Value
	1	2	3	4	5		
Dry matter	72.69 ^{e*}	75.35 ^d	79.25 ^b	82.34 ^a	77.56 ^c	0.13	0.00
Crude protein	64.29 ^e	68.63 ^d	72.85 ^c	75.79 ^b	78.46 ^a	0.14	0.00
Ether extract	68.38 ^e	71.81 ^d	74.39 ^b	77.45 ^a	72.75 ^c	0.17	0.00
Crude fiber	58.54 ^d	60.63 ^c	65.69 ^b	66.41 ^b	70.65 ^a	0.16	0.00
Nitrogen free extract	74.96 ^d	78.81 ^c	83.58 ^b	84.97 ^a	85.59 ^a	0.19	0.00

*Means in the same row with different superscripts are significantly different ($P < 0.05$).

Table 4. Effect of *Nigella sativa* meal supplementation on the nitrogen utilization.

Items	Experimental groups					SEM	P-Value
	1	2	3	4	5		
Nitrogen intake (g/h/d)	22.01 ^{d*}	22.01 ^d	25.29 ^b	27.52 ^a	24.07 ^c	0.13	0.00
Fecal nitrogen (g/h/d)	3.50 ^d	3.70 ^d	4.15 ^c	4.65 ^b	5.10 ^a	0.09	0.00
Digested nitrogen(g/h/d)	18.51 ^{cd}	18.31 ^d	21.14 ^b	22.87 ^a	18.97 ^c	0.13	0.00
Urinary nitrogen (g/h/d)	6.85 ^a	6.50 ^b	6.18 ^b	5.23 ^c	5.11 ^c	0.08	0.00
Nitrogen balance (g/h/d)	11.66 ^d	11.81 ^d	14.96 ^b	17.64 ^a	13.86 ^c	0.18	0.00
NB of intake**	52.98 ^c	53.66 ^c	59.14 ^b	64.10 ^a	57.59 ^b	0.62	0.00
NB of absorbed N	62.99 ^d	64.50 ^d	70.75 ^c	77.13 ^a	73.06 ^b	0.48	0.00

*Means in the same row with different superscripts are significantly different ($P < 0.05$). ** NB: Nitrogen balance. **SEM, pooled standard errors of means.

Table 5. Effect of *Nigella sativa* meal supplementation on the ruminal parameters.

Items	Experimental groups					SEM	P-Value
	1	2	3	4	5		
pH	6.35	6.37	6.36	6.4	6.38	0.02	0.29
NH ₃ -N (mg/100ml)	20.10 ^c	20.72 ^{bc}	21.85 ^{ab}	22.81 ^a	21.13 ^{bc}	0.26	0.00
Protozoa (x10 ⁵ cell/ml)	4.53 ^a	4.36 ^a	4.12 ^{ab}	3.81 ^{ab}	3.25 ^b	0.23	0.02
Total bact. count (x10 ¹¹ /ml)	5.87 ^a	5.45 ^{ab}	4.85 ^{abc}	4.30 ^{bc}	3.75 ^c	0.27	0.00
Short-chain fatty acids (mEq/100ml)							
Acetic Acid	4.15 ^c	4.40 ^{bc}	4.75 ^{ab}	5.12 ^a	4.65 ^{abc}	0.12	0.00
Propionic Acid	1.86 ^c	2.15 ^{bc}	2.52 ^{abc}	3.21 ^a	3.02 ^{ab}	0.22	0.01
Acetic/ Propionic Acid	2.23 ^a	2.06 ^{ab}	1.88 ^{ab}	1.64 ^b	1.71 ^{ab}	0.12	0.03
Butyric Acid	1.10 ^b	1.23 ^b	1.51 ^b	2.29 ^a	2.15 ^a	0.12	0.00
Isobutyric Acid	0.06 ^b	0.08 ^{ab}	0.14 ^{ab}	0.18 ^a	0.16 ^{ab}	0.02	0.01
Valeric Acid	0.28 ^c	0.35 ^{bc}	0.82 ^{ab}	1.10 ^a	0.94 ^a	0.10	0.00
Isovaleric Acid	0.15 ^a	0.17 ^{ab}	0.22 ^{ab}	0.27 ^a	0.25 ^{ab}	0.02	0.01
Total SCFA	7.60 ^c	10.44 ^b	11.84 ^{ab}	13.81 ^a	12.88 ^{ab}	0.55	0.00
Profile (% of total SCFA)							
Acetic Acid	54.86 ^a	42.22 ^b	40.16 ^{bc}	37.15 ^{bc}	36.19 ^c	1.11	0.00
Propionic Acid	24.42	20.56	21.31	23.07	23.43	0.93	0.08
Butyric Acid	14.49 ^{ab}	11.73 ^c	12.70 ^{bc}	16.57 ^a	16.69 ^a	0.48	0.00

*Means in the same row with different superscripts are significantly different (P<0.05).

2021; Sadarman *et al.*, 2021; Obeidat *et al.*, 2023; Le Graverand *et al.*, 2023; Juandita *et al.*, 2024; Ramdani *et al.*, 2024; Gaafar *et al.*, 2025), goats (Zaher *et al.*, 2020; Ahmed *et al.*, 2022) and calves (El-Nagar *et al.*, 2023; Elsayed *et al.*, 2025). Furthermore, the addition of NSM increased the Awassi lambs' dry matter consumption and weight gain. This improvement can result from consuming more dry matter, particularly energy, which is one of the key elements influencing growth performance. Because fat provides 2.25 times more energy than CP or carbohydrates, a higher fat intake and digestion in the supplement results in more energy (Obeidat, 2020).

NSM increased body length gain along with the increased ADG of Garut sheep. It could be evidence that high protein amount of NSM could increase protein intake for muscle and bone formation (Wahyudi *et al.*, 2023). Increased amino acid absorption due to protein availability of NSM resulted in enhanced growth performance (Wang *et al.*, 2020). Furthermore, Elsayed *et al.* (2025) found that NS supplementation greatly increased the calves' food intake and weight gain.

High performance in calves due to NS addition could be related with higher content of protein, essential amino acids and fatty acids in NS increased availability of nutrients for growth. These beneficial effects can be attributed to the high levels of essential fatty acids in *N. sativa*, such as linolenic, oleic, and linoleic acids, which play a crucial role in growth (Sadarman *et al.*, 2021). Additionally, important chemical compounds like alkaloids, flavonoids, essential oils, trace minerals, and amino acids contribute to both production and health in small ruminants (Ahmed *et al.*, 2021). However, 24% NSM diet led to reduced weight gain, likely due to amino acid imbalances (Zeweil *et al.*, 2008).

Nigella sativa meal is a palatable feed ingredient, which is consistent with earlier research in lamb (Cherif *et al.*, 2018; Le Graverand *et al.*, 2023; Juandita *et al.*, 2024; Ramdani *et al.*, 2024) and goats (Ahmed *et al.*, 2022). This may explain the increased feed intake observed in the groups fed on *Nigella sativa* meal. The aroma from NSM diet due to flavonoids, alkaloids, phenolics, and tannins plays a role in stimulating livestock to choose the diet quickly and increase palatability (Adiwimarta, 2021). Lambs' dry matter intake and daily growth increased when BCS was added to their small ruminant diets. One possible explanation is the phytochemical content and protein degradability of the seeds (Abd El-Hakim *et al.*, 2021; Sadarman *et al.*, 2021). High level of NSM (24%) decreased significantly feed intake which affects linearly on the body weight gain. Because of its

alkaloids, NS has a bitter and spicy taste, which may be the cause of its detrimental effects on feed intake and consequent growth performance (Ershadi *et al.*, 2022). The effectiveness of feed, which is related to various issues such as economic, environmental, and social, is a crucial factor in the farming industry.

Feed conversion improved with *Nigella sativa* meal supplementation which in harmony with other results (Ahmed *et al.*, 2022; Le Graverand *et al.*, 2023; Juandita *et al.*, 2024). Furthermore, the improvement of digestive enzyme activity with stimulation of intestinal motility by NS could be led to increased feed efficiency and conversion (Wei *et al.*, 2022). In addition, Hassan *et al.* (2024) concluded that NSM supplementation increased significantly total and daily weight gain with lower feed conversion values in growing lambs.

From previous studies, there are several modes of action for effect of NS as growth promoters including important role in enhance enzyme secretion and bile acids, nutrients digestion and absorption, modulate rumen fermentation and microbiota as well as improve immunity (Griss *et al.*, 2020; Costa *et al.*, 2021; Irawan *et al.*, 2020; Sadarman *et al.*, 2021; El-Nagaret *et al.*, 2023).

Incorporating *Nigella sativa* meal (NSM) in the lamb diets has shown remarkable benefits, significantly enhanced nutrient digestibility (P<0.05). This enhancement is largely attributed to increased rumen microbial activity and elevated levels of total volatile fatty acids (TVFAs), which contribute to improved growth and health in these animals (Antonius *et al.*, 2024). Its rich degradable proteins and bioactive compounds, including saponins and flavonoids, promote beneficial microbes, boosting nutrient utilization (Oluduro, 2012). By increasing dietary inclusion of NSM induced positive effects on digestibility not only stimulates digestive enzymes but also enhanced microbial activity and optimizes nutrient absorption (Obeidat, 2020; Sadarman *et al.*, 2021; Rahmy *et al.*, 2024).

Prior research has demonstrated that NSM supplementation improves nutrient utilization efficiency by increasing feed intake and nutrient digestibility (Sadarman *et al.*, 2021). NSM supplementation increased DM, OM and CP digestibility in lambs (Abdullah and Farghaly, 2019; Ramdani *et al.*, 2024). Additionally, feeding NSM to Awassi ewes enhanced the lambs' daily gain and the digestibility of CP and CF while also increasing N retention (Obeidat, 2020; Obeidat *et al.*, 2023). Goats fed on the diets supplemented with NS had greater DM, CP, and CF digestibility compared to control one (Ahmed *et al.*, 2022). Obeidat (2020) found that, the lower

dustiness of NSM due to role of fat in the NSM or improvement of DM, CP and EE digestibility, N retention resulted in enhancement of dry matter intake in the lamb fed on the diets supplemented with NSM. Another explanation could be the antibacterial action on the harmful microbes in the digestive tract, which improved gut health and raised palatability.

High-quality crude protein that is easily digested is present in *Nigella sativa* meal. For rumen microbial activity, lambs' diets with higher protein and NSM content may be beneficial (Retnani et al., 2019). Furthermore, Barkah et al. (2019) found that NSM was a protein source in ruminant diets that had a major impact on the generation of rumen ammonia (NH₃), which was utilized by rumen microbes for the synthesis of microbial protein and improved digestibility. Tannin compound in NSM was help to be directly digested protein content without fermentation by microbes in the rumen of sheep. Tannin was a bypass protein would be beneficial, which would escape rumen microbial degradation, which was used by the sheep's body through the post-rumen tract (small intestine) (Sagito et al., 2022).

High amounts of NH₃ generated by the fermentation of sheep's protein led to the growth of cellulolytic bacteria, which improved the digestibility of fiber and the capacity to break down crude fiber (cellulose) (Zeng et al., 2023). Numerous studies found that adding NSM to the diet or replacing protein with it enhanced nutrient digestibility, particularly for CP. The polyphenols when present in NSM at higher concentrations would increase the digestibility of CP and CF due to antimicrobial effect of NSM which enhanced microorganisms of rumen and their digestion capabilities (Kholifand Olafadehan, 2021; Sadarman et al., 2021; Kholif, 2023; Obeidat et al., 2023).

In this study, *Nigella sativa* meal increased significantly nitrogen intake, nitrogen digested and nitrogen retention up to 18% level then decreased with high level of NSM (24%). With increased rates of NS, Nitrogen (N) intake, and N digested were found to be significantly ($p < 0.01$) increased (Sadarman et al., 2021). N retention in growing Awassi lambs fed on the NSM supplementation was better compared with control group. The positive N retention, which reflected on the growth performance of lambs and explained by increased CP digestibility (Obeidat, 2020; Obeidat et al., 2023). In addition, Retnani et al. (2019) found more N retention in lambs fed NSM, which indicated that the NSM have high quality of CP compared with other protein sources. Compounds found in *N. sativa*, such as thymoquinone and essential amino acids, likely boosted protein synthesis and thyroid hormone secretion (Amin and Hosseinzadeh, 2016).

The improvement in previous growth performance data included feed intake, nutrient digestibility and N balance data, explained that *Nigella sativa* meal is ingredient rich in nutrients and could be used in growing lambs as part of the diets.

In contrast to rumen parameters, pH was not significantly different between different experimental groups. This outcome is consistent with that of Abdullah and Farghaly (2019), who observed that when NSM was used in place of cottonseed meal in growing lambs, the pH of the rumen remained constant at 6.2 to 6.4 across all dietary treatment groups. In addition, Thayalini et al. (2018) stated no significant differences in the rumen pH of goats supplemented with *Nigella sativa* seed meal.

As NSM levels rose to 18%, the ruminal concentrations of NH₃-N and total VFAs increased; however, at high levels (24%), they dropped. These results in harmony with that found by Abdullah and Farghaly (2019) who reported that NH₃-N and total VFAs were increased with increasing level of *N. sativa* meal in the diet of growing lambs. Growing lambs fed *N. sativa* meal may have higher rumen NH₃-N concentrations, which could lead to improved digestibility and CP uptake. In the same setting, NS treatment raised NH₃-N concentrations ($P < 0.01$) (Cherif et al., 2018).

When compared to the control group, the higher concentration of total volatile fatty acids for animals fed *N. sativa* meal supplementation is consistent with earlier findings that total volatile fatty acids increased ($P < 0.05$) as the amount of *N. sativa* in lamb diets increased (Klevenhusen

et al., 2014; El-Naggar et al., 2018; Abdullah and Farghaly, 2019).

In this study, there were increased in acetic, propionic, butyric, isobutyric, valeric and isovaleric acids with increased levels of NSM up to 18% then decreased with high level of NSM (24%), while acetic/propionic ratio was decreased. These findings concurred with those of Medjekal et al. (2017), who found that the acetic/propionic ratio fell while propionic acid and total VFA rose. Rumen parameters showed stable pH levels, with increased NH₃-N and total volatile fatty acid (VFA) concentrations, indicating enhanced ruminal fermentation and microbial activity (Antonius et al., 2024).

Our investigation found a significant decrease ($P < 0.05$) in the overall protozoal count in lamb rumen fluids. This finding was consistent with other research showing that higher amounts of NSM in lamb diets reduced the number of protozoa (Cherif et al., 2018; Abdullah and Farghaly, 2019). Thymoquinone and thymohydroquinone, which are found in *N. sativa* and have antibacterial qualities against a variety of ruminal microorganisms, may be the cause of these results (Niu et al., 2020; Singh et al., 2022). Furthermore, black cumin seed (*Nigella sativa*) exhibited antibacterial activity against a wide range of pathogenic bacteria, fungi, and bacteria. Its antibacterial properties, which include thymoquinone and paracymene, could be the cause of this (Khan, 2018; Balahbib et al., 2021). Another study found that *Nigella sativa* meal supplementation dramatically reduced ($P < 0.05$) the population of protozoa and enhanced microbial protein but had no effect ($P > 0.05$) on pH, ammonia concentration, or the formation of volatile fatty acids (VFA) (Widyarini et al., 2021).

Conclusion

Based on the results from the current study, *Nigella sativa* meal (NSM) can effectively supplement up to 18% in lamb diets without compromising growth, nutrient utilization, or overall health. Notably, lambs fed on the 18% NSM diet achieved superior final weight, improved feed efficiency, enhanced nutrient digestibility, and optimal rumen fermentation. Thus, replacing up to 18% of NSM is recommended as an optimal strategy to maximize growth performance, health, and profitability in growing Rahmani lambs, making it a promising alternative feed option.

Conflict of interest

The authors have no conflict of interest to declare.

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