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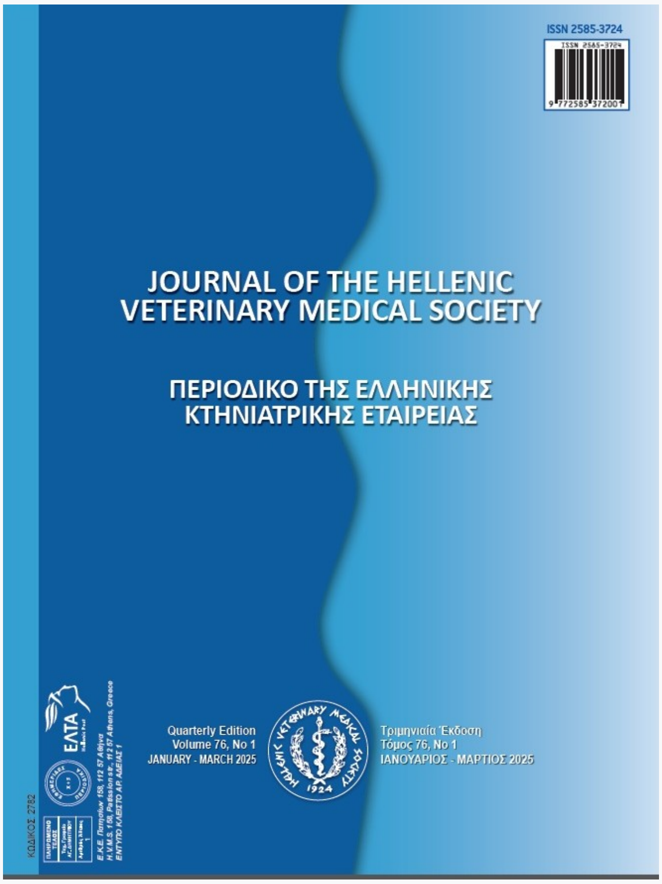


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# Research article
# Ερευνητικό άρθρο

# Determining an optimal forage-to-concentrate ratio on performance traits and blood metabolites of fattening lambs

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**ABSTRACT:** In intensive rearing, a combination of concentrate and forage is used for lamb fattening. Research was conducted to determine the optimal ratio of forage-to-concentrate for fattening male Zel lambs. Twenty 16-week-old lambs were used for 90 days in a completely randomized design with four treatments and five replicates. Treatments included forage-to-concentrate ratios of 80:20, 70:30, 60:40, and 50:50. Various traits were measured during the study. The collected data were analyzed by LSmeans and regression procedures in SAS software. The effect of forage-to-concentrate ratios was significant on all performance traits ($p<0.01$), with Significant linear changes observed in the mentioned traits ($p<0.05$). However, the effect of different forage-to-concentrate ratios was not significant on all blood metabolites ($p>0.05$) except for cholesterol concentration ($p<0.05$). Based on the current findings and considering limitations associated with increasing concentrate consumption, the optimum ratio of forage to concentrate is suggested to be 50:50.

***Keywords:*** blood metabolites; cholesterol; concentrate; forage; lamb; performance.

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## INTRODUCTION

Animal fattening is the best way to supply the animal protein. Fattening involves the optimal rearing of animals to achieve maximum weight gain at minimal cost over a determined period (Hosseini et al., 2019). Fattening through pasture utilization is among the most cost-effective methods. However, some of the feed energy is expended through the animal's movement in this approach. Of course, it may not be suitable for countries with poor pasture conditions or those lacking pasture altogether. Fattening lamb on pastures or using intensive methods relying solely on forage may result in carcasses with a higher ratio of lean meat, but can reduce growth rates and produce lighter carcasses. Conversely, lambs fattened with concentrate-based rations increased growth rates and yield heavier carcasses (Murphy et al., 1994). On the other hand, concentrates could offer greater nutritional benefits compared to forages. Sheep grazing alone typically cannot meet the nutritional needs for optimal performance because pastures are not rich in nutritional materials. However, grazing on pasture can reduce production costs and enhance profitability (Jiang et al., 2022).

In intensive rearing methods, a ration containing both concentrate and forage is used for lamb fattening. Considering that forage is fibrous, it alone cannot supply the necessary energy and protein for the rapid fattening of lambs within a short period. Therefore, to achieve the desired fattening weight in a specific and short time, it is essential to incorporate ration rich in both energy and protein (concentrate). A suitable ration containing a balanced mixture of concentrates and forages typically yields the best performance in lamb fattening. Shi et al., 2018 reported that the supplementing concentrate to forage-based rations increases the heat produced by visceral organs while reducing the energy content of fecal dry matter and urinary nitrogen, thereby increasing production efficiency. In rations consisting concentrate and forage, the efficiency of nutrient utilization for animal tissue production is increased (Nie et al., 2020). Determining the appropriate ratio of forage-to-concentrate in the ration is important for lambs fattening. This ratio significantly affects digestibility and ruminal fermentation properties. Traits related to lamb fattening in the intensive methods are closely linked to the energy and protein levels of the ration. In other words, reducing the time required for lamb fattening hinges on determining the optimal concentrate to forage ratio (Borton et al., 2005; Nie et al., 2020).

The Zel lambs are the only breed in Iran without a fat-tail characteristic. They have a narrow tail with 7 tail nuts, typically measuring to 10 to 15 cm in length. The absence of tail fat in Zel sheep is attributed to fat accumulation and storage between tissues. This unique characteristic has increased the quality and marketability of Zel lamb meat (Hosseini et al., 2019), making lambs of this breed an attractive option for fattening.

The research was conducted to determine the optimal forage-to-concentrate ratio on performance traits and blood metabolites of male Zel breed lambs.

## MATERIALS AND METHODS

### Lambs, treatments, and the experimental conditions

The study involved 20 male Zel breed lambs at the research farm of the Islamic Azad University of Qaemshahr. These lambs were 16 weeks olds with a mean body weight of 27.8±2.1 kg. Experimental treatments included rations with varying ratios of forage-to-concentrate (80:20, 70:30, 60:40, and 50:50). The metabolizable energy and crude protein content of the rations were 2.10, 2.15, 2.40, and 2.55 mcal/kg of dry matter and 14.0, 14.3, 15.2, and 16.3%, respectively (Table 1). The rations' Neutral Detergent Fiber (NDF) were 32.7, 32.2, 30.7, and 28.9 % respectively, while the Acid Detergent Fiber (ADF) content of the rations was 24.8, 22.7, 20.6, and 18.5 %, respectively (Table 1). These rations were formulated based on the tables of nutritional needs and requirements of animals and were presented in Table 1 (AFRC, 1992).

Each lamb was allocated a designated box for the experiment. Experimental feeds were randomly assigned to lambs, and both water and feed were provided in their respective boxes. Before the beginning of the experimental period, each lamb received albendazole anti-parasitic tablets at a dosage of 7.5 mg per kilogram of body weight. Thus, individual feed intake was measured. The duration of the experiment spanned 90 days. Lambs were weighed weekly after a 10 hours feed abstinence, with weighing conducted at 6 am. Feeding occurred three times daily at 6 am, 2 pm, and 8 pm, providing feed until the lambs reached appetite. Additionally, clean water was constantly available to the lambs throughout the experiment.

### Traits measurement

Feeding was done twice a day and any remaining

**Table 1** Experimental rations used and their compounds (%)

| concentrate ratio) | (Forage to | Treatments | | |
|---|---|---|---|---|
| 50:50 | 60:40 | 70:30 | 80:20 | Ration components |
| 10 | 7 | 5 | 3 | Soybean meal |
| 12 | 9 | 6 | 3 | Corn |
| 10 | 7 | 4 | 2 | Barley |
| 8 | 7 | 5 | 2 | Wheat bran |
| 50 | 60 | 70 | 80 | Alfalfa |
| 3.5 | 3.5 | 3.5 | 3.5 | Molasses sugarcane |
| 3 | 3 | 3 | 3 | Di-calcium phosphate |
| 0.5 | 0.5 | 0.5 | 0.5 | Supplement[1] |
| 0.5 | 0.5 | 0.5 | 0.5 | Salt |
| 0.5 | 0.5 | 0.5 | 0.5 | Bicarbonate Sodium |
| 2 | 2 | 2 | 2 | Calcium carbonate |
| | | estimated | compounds | Chemical |
| 2.55 | 2.40 | 2.15 | 2.10 | Metabolizable energy (Mcal/Kg) |
| 16.3 | 15.2 | 14.3 | 14.0 | Crude protein (%) |
| 10.9 | 10.7 | 10.4 | 10.1 | Calcium (g/Kgr) |
| 6.3 | 6.2 | 6.1 | 6.0 | Phosphorus (g/Kgr) |
| 28.9 | 30.7 | 32.2 | 32.7 | NDF[2] (%) |
| 18.5 | 20.6 | 22.7 | 24.8 | ADF[3] (%) |

1- Each kilogram of supplement includes 2400 international units of Vitamin A, 2000 international units of Vitamin $D_3$, 1900 international units of Vitamin E, 1.15 grams of Manganese, 1.19 grams of Zinc, 51.2 grams of Magnesium, 0.52 grams of Iron, 3 g of Sulfur, 0.52 g of Copper, 14 mg of Cobalt, 26 mg of Iodine and 10 mg of Selenium. 2- Neutral detergent fiber. 3- Acid detergent fiber.

feed was collected and weighed daily. Lambs were weighed every 15 days, with individual weights recorded to monitor weight changes. The average daily weight gains during the fattening period (in grams) relative to the metabolic body weight (in kilograms) at the end of the experiment served as an indirect measure of feed efficiency (metabolic conversion coefficient). The Kleiber coefficient, which reflects the relationship between the metabolic weight of the animal and its maintenance and production requirements, was also considered. Additionally, the feed conversion ratio was calculated by dividing the average dry matter consumed by the average live weight gain of the lambs within each treatment group.

A day before finishing the experiment, blood samples were collected from the left jugular vein of the lambs two hours after feed consumption. Venoject tubes containing heparin under vacuum and a #20 needle head were used for this purpose. Blood serum separation was achieved through centrifugation for 15 minutes at 3500 rpm, after which the serum samples were stored in a freezer at -20 °C. Serum albumin and total protein levels were determined using the Biuret and Bromocresol green dye-binding method, respectively (Gharahveysi, 2018). Serum globulin levels were calculated by subtracting serum albumin from total protein. Serum glucose concentration was determined using the Iranian Pars Azmoun kit through the photometric method with an Autoanalyzer (BT-1500). Blood cholesterol concentration was measured using the Iranian Zist-Shimi kit and a spectrophotometer (Varian 220-Australia). Additionally, the concentrations of thyroxine and tri-iodothyronine were measured using the Iranian Pars Azmoun kit and an ELISA device.

### Statistical analysis

A completely randomized experimental design with four treatments and five replicates was performed. Data for the studied traits were recorded on a computer. Statistical analysis of the data was conducted using LSmeans and regression procedures in SAS software (2000). The statistical model used was as follows:

$$y_{ij} = \mu + T_i + e_{ij}$$

That; $y_{ij}$, the amount of each observation; μ, the mean effect; $T_i$, the treatment effect, and $e_{ij}$ is the residual effect.

## RESULTS

### Performance traits:

As presented in Table 2, the effect of different ratios of forage-to-concentrate was found to be significant for all performance traits ($p<0.01$). Treatment with an equal ratio of forage-to-concentrate caused significant changes in these traits as well ($p<0.01$). The treatment with a forage to concentrate ratio of 50:50 exhibited the highest slaughter weight (58.9) and weight gain during the fattening period (31.2), as well as the highest daily weight gain (347), Kleiber coefficient (16.32), and feed intake (2280). Conversely, the lowest values for these traits were observed in the treatment with a forage to concentrate ratio of 80:20 (slaughter weight: 50.6; weight gain: 22.8; daily weight gain: 253; Kleiber coefficient: 13.34; feed intake: 2109). Also, the treatment with a forage to concentrate ratio of 80:20 had the highest feed conversion ratio (FCR) of 8.34, while the treatment with a ratio of 50:50 had the lowest FCR of 6.57. Moreover, a linear increase was observed in traits such as slaughter weight ($p<0.05$), weight gain in fattening period ($p<0.05$), daily weight gain ($p<0.05$), feed intake ($p<0.01$), feed conversion ratio ($p<0.01$), and Kleiber coefficient ($p<0.01$) with an increase in concentrate to forage ratio. This increase followed a constant linear coefficient, indicating a consistent trend. It should be noted that no significant nonlinear relationships were observed ($p>0.05$). The significant change observed in treatments with an equal ratio of forage-to-concentrate ($p<0.01$) can be attributed to the presence of dense substances containing energy and protein in the feed.

### Blood metabolites

On Tables 3 and 4, it can be seen that the effect of different forage-to-concentrate ratios on cholesterol concentration was indeed significant ($p<0.05$). However, no significant effect was observed for other blood metabolites ($p>0.05$). The treatment with a forage to concentrate ratio of 80:20 exhibited the highest cholesterol concentration (56.32), while the treatment with a ratio of 50:50 had the lowest concentration (40.70). With an increasing concentrate-to-forage ratio, there was a linear increase in cholesterol concentration ($p<0.01$), following a constant linear coefficient.

## DISCUSSION

### Performance traits

In the current study, the optimal ratio of forage to concentrate was found to be 50:50 for performance traits, aligning with previous research in this area (Glimp et al., 1989; Hatfield et al., 1997; Mahgoub et al., 2000). In research done on Turkish Merino lambs, in rations with or without rumen-protected fat, the effect of forage and concentrate consumption on live weight, average daily gain, and FCR was not

**Table 2** The effect of different forage-to-concentrate ratios on the feed intake, body weight traits of Zel fattening lambs

| Kleiber coefficient | Feed conversion ratio[1] | Feed intake (g/d) | Daily weight gain (g/d) | Weight gain in fattening (kg) | Slaughter weight (kg) | Initial weight (kg) | Treatments(forage-to-concentrate ratio) |
|---|---|---|---|---|---|---|---|
| 13.34$^{a}$ | 8.34$^{a}$ | 2109$^{a}$ | 253$^{a}$ | 22.8$^{a}$ | 50.6$^{a}$ | 27.8 | 80:20 |
| 14.29$^{a}$ | 7.84$^{a}$ | 2196$^{a}$ | 280$^{a}$ | 25.2$^{a}$ | 52.8$^{a}$ | 27.6 | 70:30 |
| 14.65$^{a}$ | 7.51$^{a}$ | 2201$^{a}$ | 293$^{a}$ | 26.4$^{a}$ | 54.3$^{a}$ | 27.9 | 60:40 |
| 16.32$^{b}$ | 6.57$^{b}$ | 2280$^{b}$ | 347$^{b}$ | 31.2$^{b}$ | 58.9$^{b}$ | 27.7 | 50:50 |
| 0.01 | 0.01 | 0.01 | 0.01 | 0.01 | 0.01 | 0.46 | P-value[2] |
| 0.28 | 0.19 | 82.8 | 9.12 | 0.78 | 0.91 | 0.89 | SEM[3] |
| 0.00 | 0.00 | 0.00 | 0.03 | 0.03 | 0.03 | 0.89 | Linear[4] |
| 0.35 | 0.27 | 0.08 | 0.21 | 0.13 | 0.10 | 0.96 | Nonlinear |

1- Feed conversion ratio is feed intake divided by daily weight gain. 2- Probability value. 3- Standard error of mean. 4- P-values of regression coefficients are presented. Common letters in each column indicate no statistically significant difference ($p>0.05$).

**Table 3** The effect of different forage-to-concentrate ratios on the blood metabolites of Zel fattening lambs

| Albumin/ Globulin | Globulin (g/dl) | Albumin (g/dl) | Total protein (g/dl) | Glucose (mg/dl) | Treatments(forage-to-concentrate ratio) |
|---|---|---|---|---|---|
| 2.54 | 1898.72 | 4831.17 | 6102.80 | 86.84 | 80:20 |
| 2.57 | 1885.53 | 4852.78 | 6153.87 | 86.70 | 70:30 |
| 2.60 | 1879.42 | 4883.41 | 6170.36 | 86.45 | 60:40 |
| 2.66 | 1840.10 | 4892.53 | 6181.15 | 86.28 | 50:50 |
| 0.39 | 0.25 | 0.32 | 0.41 | 0.51 | P-value[1] |
| 0.29 | 46.80 | 25.19 | 68.48 | 0.56 | SEM[2] |
| 0.49 | 0.38 | 0.47 | 0.25 | 0.63 | Linear[3] |
| 0.38 | 0.23 | 0.36 | 0.27 | 0.45 | Nonlinear |

1- Probability value. 2- Standard error of mean. 3- P-values of regression coefficients are presented. Common letters in each column indicate no statistically significant difference ($p>0.05$).

**Table 4** The effect of different forage-to-concentrate ratios on the serum concentration of urea, cholesterol, triiodothyronine, and thyroxine of lambs of Zel fattening lambs

| Thyroxine (µg/dl) | Tri-iodothyronine (ng/dl) | Cholesterol (mg/dl) | Urea (mg/dl) | Treatments(forage-to-concentrate ratio) |
|---|---|---|---|---|
| 3.61 | 1.18 | 56.32[a] | 35.80 | 80:20 |
| 3.68 | 1.19 | 51.12[a] | 35.98 | 70:30 |
| 3.71 | 1.21 | 46.92[ab] | 36.19 | 60:40 |
| 3.70 | 1.23 | 40.70[b] | 36.29 | 50:50 |
| 0.27 | 0.35 | 0.04 | 0.31 | P-value[1] |
| 0.15 | 0.09 | 0.48 | 1.59 | SEM[2] |
| 0.21 | 0.32 | 0.04 | 0.28 | Linear[3] |
| 0.34 | 0.51 | 0.29 | 0.35 | Nonlinear |

1- Probability value. 2- Standard error of mean. 3- P-values of regression coefficients are presented. Common letters in each column indicate no statistically significant difference ($p>0.05$).

significant (p>0.05) (Gumus et al., 2022). In another research conducted on Spanish Lacaune male lambs, the effect of high-cereal concentrate on body weight, feed intake, FCR, and carcass traits was not significant (p>0.05) (De evan et al., 2020). In a research conducted on Chinese Small-Tailed crossbred rams, rations containing corn straw and millet straw were used. The ration containing an equal ratio of corn straw and millet straw had a significant effect on body weight, and average daily gain (p<0.05) (Chen et al., 2020). In the studies mentioned, the treatments and objectives differ from the present study, which could explain the discrepancy between their results and recent findings. The better response of lambs to higher growth rates can be related to the optimal concentration of energy and protein in the experimental rations. Several researchers have reported that rations containing 70% concentrate stimulate proper growth performance in lambs and fattening calves (Glimp et al., 1989; Mahgoub et al., 2000; Langlie, 2020; Fimbres et al., 2002). Furthermore, the effects of a high percentage of concentrate in the ration are reflected in the dry matter consumed by various animals such as lambs (Hatfield et al., 1997; McLeod and Baldwin, 2000), goats (Lu and Potchoiba, 1990), and fattening calves (Lardy et al., 2004). As animal consume feed to meet their energy needs, their appetite decreases once these needs are fulfilled. The improved conversion ratio of rations with an equal forage-to-concentrate ratio may result from the adequate supply of energy and protein, leading to a better response from lambs. However, feeding high-concentrate rations can lead to increased production of volatile fatty acids and lactic acid, decreased ruminal pH, and finally the occurrence of sub-acute ruminal acidosis. This condition may be emerged by feeding barley at high levels due to its higher ruminal fermentability compared to corn, potentially increasing the incidence of sub-acute ruminal acidosis. The primary damage of sub-acute ruminal acidosis is chronic inflammation in the rumen wall, which can facilitate the transfer of bacterial endotoxins from the rumen environment to the bloodstream. Decreased rumen pH and increased death of Gram-negative bacteria can stimulate systemic inflammation by transferring lipopolysaccharide wall fragments from these bacteria to the bloodstream (Jiang et al., 2022).

The Kleiber coefficient shows that metabolic rate is directly linked to body mass, indicating that larger organisms have proportionally higher metabolic rates (Ramos et al., 2020). In the current study, the highest Kleiber coefficient was found in the treatment with an equal ratio of forage to concentrate, confirming the earlier statement. This coefficient holds significant implications for understanding energy requirements and the ecological roles of different organisms. It helps clarify why larger animals generally exhibit lower metabolic rates per unit of body mass compared to smaller animals, and why larger animals can thrive on relatively less food per unit of body mass (Preziuso et al., 1999).

**Blood metabolites**

According to the current findings, in research conducted on Turkish Merino lambs, the effect of forage and concentrate consumption on cholesterol and LDL was significant in rations with or without rumen-protected fat (p<0.05) (Gumus et al., 2022). In another research done on Spanish Lacaune male lambs, the effect of high-cereal concentrate on total proteins, and cholesterol was significant (p<0.05) (De evan et al., 2020). Also, in a research conducted on Chinese Small-Tailed crossbred rams, rations containing corn straw and millet straw were used. The ration containing an equal ratio of corn straw and millet straw had a significant effect on cholesterol, blood glucose, and total protein

($p<0.05$) (Chen et al., 2020). Increasing the amount of concentrate in the ration, has been linked to elevated cholesterol production by liver cells and increased absorption from the small intestine (Wang et al., 2023). This increase is attributed to the increased activity of 3-Hydroxy-3-MethylGlutaryl-Coenzyme A (HMG-CoA) reductase, which is the primary enzyme in cholesterol biosynthesis (Kedar and Chakrabarti, 1982). Also, the availability of Nicotinamide Adenine Dinucleotide Phosphate (NADPH), an important substrate in cholesterol biosynthesis, tends to rise with increased concentrate intake (Sharma and Kataria, 2011). Cholesterol synthesis in ruminants occurs primarily in the liver and is distributed throughout the bloodstream and tissues. Ruminants, like sheep, typically exhibit lower blood cholesterol levels compared to non-ruminant animals due to their unique digestive system, which efficiently processes and utilizes the cholesterol of ration. The rumen fermentation process also plays a role in cholesterol metabolism.

## CONCLUSIONS

The concentrate feed is palatable and rich in energy and nutrients. Consequently, consuming the optimal amount of concentrate (if the consumption limit is considered) causes desired weight gain. In the present study, the 50:50 forage-to-concentrate ratio was found to be effective in the production traits and blood metabolites of fattening lambs.

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## CONFLICT OF INTEREST

The authors do not have any potential conflicts of interest to declare.

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