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Impact of dosed Awassi sheep weanlings with thyme and fenugreek extracts on some cellular and biochemical blood parameters.

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Abstract:

The current study was conducted at the First Agricultural Research and Experiment Station (12 km) southeast of Al-Muthanna Governorate, College of Agriculture, Al-Muthanna University. The study was conducted from October 5, 2024, to February 28, 2025. It aimed to demonstrate the effect of using plant extracts of thyme, fenugreek, and their mixture on cellular and biochemical blood parameters of Awassi sheep weanlings. Twenty Awassi sheep weanlings, 4-6 months old, were used in the study after a 14-day acclimatization process. The actual experiment was then implemented for three months. The results of this study showed that the use of natural plant extracts, including thyme powder, fenugreek, and their mixture, had a positive and significant effect on the productive performance of Awassi sheep weaning. The results of the study showed that the mixture treatment (thyme+ fenugreek) significantly outperformed the final weight of the treated animals.

Keywords: Dosage, thyme, fenugreek, water extracts, cellular, biochemical, blood, Awassi sheep, weanlings.

Introduction:

Sheep have played a major and important role in the economies of countries around the world, both ancient and modern. Sheep are characterized by the ease of establishing and providing pens, low care costs, and low nutritional requirements. They also have a high

feed conversion efficiency, enabling them to utilize crop residues and fodder. Sheep are naturally pastoral animals, tending to gather and graze on natural grasses and weeds (Al-Zubaie, 1999).

The use of medicinal plants has recently attracted the attention of many researchers. They are produced

through several methods without causing dangerous side effects, such as synthetic preparations and medications composed of chemical compounds. These have proven to have negative effects on the human body and health (Hijazi, 2000; Al Salman and Al-Gharawi, 2019).

The use of medicinal plants in Iraq dates back thousands of years. Clay tablets and inscriptions from the Sumerian and Babylonian civilizations demonstrate the importance of using and developing medicinal plants throughout the ages. The first pharmacy in the world was in Baghdad in the late eighth century AD, during the Abbasid era. Herbal medicine is therefore an ancient and well-known practice in Iraqi society. It is a source of healing for many diseases, and many people trust its effectiveness in treatment and recovery (Al Bayaty, 2011; Al-Gharawi and Ebade, 2020).

Numerous studies have demonstrated the beneficial effects of medicinal plant extracts, including thyme, it contains numerous compounds with great potential for use in the treatment of cancer. This is due to its pharmacological properties and its ability to eliminate cancer cells through various molecular mechanisms, including autophagy, necrosis, and apoptosis (Rathod *et al.*, 2021; Bouhtit *et al.*, 2021). Thyme is used in the perfume and cosmetics industry due to its unique aromatic properties, it is also used to treat sore throats, diarrhea, chronic gastritis, and

loss of appetite (Zaruelo and Crespo, 2002).

Recent studies have also shown that fenugreek has antioxidant effects because it is rich in active chemical components, such as flavonoids and polyphenols, which have the ability to reduce hydrogen and displace oxygen atoms. It also prevents the development of breast cancer. The anti-cancer effects of fenugreek are attributed to the flavonoids it contains. In addition, it contains diosgenins, which in turn lower cholesterol levels (Yadav *et al.*, 2010). Studies conducted on mice have shown that fenugreek has mitogenic effects and stimulates the proliferation of bone marrow cells in vertebrates (de La Fuente and Victor, 2002).

The present study aims to demonstrate the effect of dosing Awassi sheep weanlings with thyme and fenugreek extracts and their mixture on some cellular and biochemical blood parameters.

Materials and Methods

The current experiment was conducted in the animal field of the First Agricultural Research and Experiment Station (Umm Al-Akf, 12 km southwest of Al-Muthanna Governorate), College of Agriculture, Al-Muthanna University, from October 5, 2024 to February 28, 2025, the experiment aimed to identify the effect of adding thyme and fenugreek extracts and their mixtures on some

productive traits and body dimensions in Awassi sheep weaners. Twenty-four Awassi sheep weaners, aged four to six months, were used in the study. They were placed in the animal field in pens under natural conditions and were placed for 15-day introductory period to acclimate to the environmental and nutritional conditions of the animal field. The animals were then introduced to the actual experiment, which lasted for three months.

These animals were raised in four standard pens, divided internally by iron partitions into four sections, each section contained five Awassi sheep weaners, which were randomly assigned to each experimental treatment. The pens contained special iron playpens that allowed the animals freedom of movement. They also contained feeders and drinking water. The animals were fed concentrated feed, provided twice daily in sufficient quantities.

The weaning animals (aged 4-6 months) were divided into four treatments according to the plan, as follows:

T1: The control group.

T2: were given thyme extract only at a rate of 10 ml per animal daily and were considered the thyme group.

T3: were given fenugreek extract only at a rate of 10 ml per animal daily and were considered the fenugreek group.

T4: were given thyme + fenugreek extract at a rate of 10 ml per animal daily and were considered the thyme and fenugreek mixture group.

Wild thyme and fenugreek seeds were obtained from the local markets of Muthanna Governorate and were manually cleaned of any impurities and ground in a home electric grinder to obtain a fine powder.

The German extract was prepared according to the method of Hernandez *et al.* (1994), which involves mixing a quantity of dry thyme leaf powder with a quantity of distilled water at a ratio of 1 gm to 2 ml of distilled water. The mixture was placed in a water bath at 60°C for one hour. The solution was left to stand for 24 hours at room temperature. The resulting mixture was then filtered through several layers of sterile medical gauze. The concentrated liquid was then ready and the extract was used. The fenugreek seed extract was prepared in the same manner. For the (thyme + fenugreek) mixture, 0.5 gm of thyme powder and 0.5 gm of fenugreek seed powder were taken per 2 ml of water. The same preparation method was used. The concentrated liquid was then ready and the extract was used to dose weaned Awassi sheep, at a rate of 10 ml of each extract per animal per day.

Results and Discussion

Table (1) showed a significant effect ($P \leq 0.05$) of the different nutritional treatments (control, thyme,

fenugreek, and mixture) on the hematological characteristics of Awassi sheep weanlings, including: packed cell volume (PCV), hemoglobin concentration (Hb), red blood cell count (RBC), and white blood cell count (WBC).

The mixture treatment (thyme+ fenugreek) showed a significant superiority over the other treatments in the midpoint and end of the experiment in most hematological parameters. It recorded a significant superiority ($P \leq 0.05$) over the other treatments at the midpoint of the experiment on red blood cell counts (RBC) was 6.23. However, no significant differences were observed in white blood cell counts (WBC) in all the treatments. No significant differences were observed in WBC counts in any of the treatments. This treatment also recorded a significant superiority ($P \leq 0.05$) on hemoglobin (Hb) level, with a value of 11.90 compared to the control group (11.33). The same treatment recorded a significant superiority ($P \leq 0.05$) on PCV (31.96) compared to the control treatment (30.43).

At the end of the experiment, this treatment recorded the highest value in red blood cell count (RBC) at the end of the experiment (6.56). This indicates the presence of Significant differences ($P \leq 0.05$) compared to the control group, which recorded the lowest value (5.80), indicating that the mixture treatment supported red blood cell production. This treatment also achieved a significant superiority at the end of the experiment ($P < 0.05$) in white blood cell count (WBC) was 6.73 over the control group (6.53), reflecting an improvement in the immune system of the animals. This treatment also achieved a significant superiority ($P \leq 0.05$) on hemoglobin (Hb) concentration (11.96) compared to the control group (11.50). This indicates a clear improvement in the blood's ability to transport oxygen compared to other treatments. It also achieved a significant superiority ($P \leq 0.05$) in the packed cells volume (32.66) over the control treatment (30.43), which recorded the lowest value.

Table (1) The effect of doses of thyme and fenugreek extracts and their mixture on the cellular blood parameters of blood serum of weaned Awassi sheep (mean \pm standard error).

Treatments	Mid-experiment				End-experiment			
	RBC	WBC	Hb	PCV	RBC	WBC	Hb	PCV
Control	5.56 \pm 0.12 B	6.40 \pm 0.11 A	11.33 \pm 0.08 C	30.43 \pm 0.12 C	5.80 \pm 0.05 B	6.53 \pm 0.08 A	11.50 \pm 0.05 C	11.50 \pm 0.05 C
Thyme		6.56	11.50	30.73	5.86	6.63	11.70	11.70

	5.80 ± 0.05 B	± 0.12 A	± 0.05 CB	± 0.12 CB	± 0.08 B	± 0.08 A	± 0.05 B	± 0.05 B
Fenugreek	5.93 ± 0.17 BA	6.50 ± 0.11 A	11.66 ± 0.08 B	30.83 ± 0.08 B	6.00 ± 0.15 B	6.20 ± 0.05 B	11.50 ± 0.05 C	11.50 ± 0.05 C
Mixture	6.23 ± 0.06 A	6.33 ± 0.06 A	11.90 ± 0 A	31.96 ± 0.03 A	6.56 ± 0.03 A	6.73 ± 0.06 A	11.96 ± 0.03 A	11.96 ± 0.03 A
Sig.	*	N.S	*	*	*	*	*	*

This indicates improved oxygen transport efficiency, these results indicate an improvement in physiological and hematologic status due to the positive effect of the active ingredients found in both thyme and fenugreek, such as volatile oils (such as thymol and thymol in thyme) and saponins and flavonoids in fenugreek, which contribute to enhancing the efficiency of the immune system and improving digestion and metabolism (Al-Kassie, 2010; Abdulrahman, 2017).

The thyme treatment ranked second in most traits, indicating a unique positive effect of thyme on stimulating hematopoiesis. This was indicated by Hashem *et al.* (2013), who demonstrated that adding thyme to lamb rations led to a significant improvement in blood parameters due to its antioxidant and digestive enzyme-stimulating properties. The fenugreek treatment showed intermediate results between the control and mixed treatments, with significant differences in some traits at the midpoint and end of the experiment, such as red blood cell

(RBC) counts, which recorded a significant effect ($P \leq 0.05$) at the midpoint and end of the experiment (5.93 and 6.00) compared to the other treatments (control and thyme), agreed with Al-Mutioti (2018), who observed a significant increase in the red blood cell count in Awassi rams treated with fenugreek. This treatment showed a significant ($P \leq 0.05$) increase on Hb concentration (11.66) over the control treatment (11.33) at the midpoint of the experiment. However, at the end of the experiment, no significant differences were observed in this treatment. This was consistent with Al-Mutioti (2018) observed, who noted a significant increase ($P \leq 0.05$) in the concentration of hemoglobin (Hb) in the blood of Awassi rams with the fenugreek treatment. These results were consistent with Al-Janabi (2010) observed, who observed significant differences in RBC count and Hb in the pre-pregnancy, early pregnancy, and late pregnancy stages. The increased hemoglobin concentration and red blood cell count may be due to

fenugreek containing fatty acids, phospholipids, and trace minerals such as iron and vitamin C (Samara *et al.*, 2009). This is consistent with what was reported by (Nefzaoui, 2001), who stated that fenugreek contributes to improving growth and blood function by stimulating red blood cell production and increasing iron absorption. It was also noted that the white blood cell count (WBC) decreased significantly at the end of the experiment ($P < 0.05$) (6.20) in weaned Awassi sheep given fenugreek seed extract (the third treatment) compared to the three other treatments (control, thyme, and the mixture) (6.53, 6.63, and 6.73), respectively.

A significant increase in the concentration of packed cell volume (PCV) was also observed in the middle and end of the experiment. This does not agree with what was reported by (Al-Janabi, 2010), which indicated no significant effect on the size of packed cells in female goats. This may be due to the saponin found in fenugreek seeds, which lyses red blood cells by increasing the permeability of their membranes. This allows large, immature red blood cells to quickly enter the circulation, causing a decrease in hemoglobin concentration and the number of packed red blood cells (Robert, 1977; Saad El-Din, 1986; Al-Naimi, 1999).

These results indicate that the use of natural herbal supplements such as thyme and fenugreek, especially when

combined, can significantly improve blood parameters in weaned Awassi sheep, which may positively impact the animals' productive and immune performance. It is recommended that the mixed feed be used as an effective nutritional option in weaning programs.

The control group (which did not receive any herbal supplements) recorded the lowest values across all traits, indicating that herbal treatments played an effective role in improving blood parameters, reflecting improvements in overall health and physiological functions.

Tables (2) and (3) indicate that feeding Awassi sheep weanlings with extracts of thyme, fenugreek, and the mixture led to positive effects on some blood biochemical indicators, reflecting an improvement in the physiological performance of the animals.

At the midpoint of the experiment, the study showed an increase in total cholesterol and LDL concentrations in the thyme cholesterol (52.30), LDL (26.06), and fenugreek cholesterol (52.66), LDL (25.66) groups compared to the control cholesterol (54.30), LDL (30.06), and the mixture cholesterol (49.66), LDL (21.33). This may be attributed to the fact that these plants contain volatile oils and active compounds such as thymol and saponins, which may affect lipid metabolism by increasing cholesterol absorption or synthesis (Al-Mamary, 2002). The significant improvement in

HDL levels with fenugreek treatment is attributed to the flavonoids and saponins found in fenugreek seeds, which raise HDL cholesterol and lower LDL cholesterol, thus supporting cardiovascular health (Sharma, 2018).

The mixed treatment showed a significant ($P \leq 0.05$) increase in total protein (7.30) and albumin (3.33), indicating improved nutritional status and protein synthesis. This can be explained by the effect of natural antioxidants in herbs on liver function and increased protein synthesis efficiency (Craig, 1999). Fennel also significantly affected total protein (6.26) and albumin (2.80).

A decrease in GOT and GPT levels was also recorded in the thyme, fenugreek, and mixture treatments compared to the control, reflecting improved liver health and reduced cellular damage. This indicates the protective effect of these additives against oxidative stress, as indicated by Hashemipour *et al.* (2013) in their study on the effect of medicinal herbs on liver enzymes.

The significant decrease in triglycerides with fenugreek and the mixture, could be attributed to the stimulation of energy expenditure and reduction of lipid synthesis in the liver thanks to the active compounds in the plants, as demonstrated in their study on the effect of fenugreek on blood lipids (Gupta *et al.*, 2001).

The significant decrease in glucose concentration in the thyme, fenugreek, and mixture treatments compared to the control group may be explained by the ability of these plants to enhance insulin secretion and improve cell sensitivity, which contributes to lowering blood sugar levels. Finally, the increase in globulin in the thyme and mixture treatments indicates an improved immune response in the animals, as the phenolic compounds and flavonoids found in the herbs can enhance the formation of antibodies, thus enhancing the animal's overall immunity (Abu-Darwish *et al.*, 2010).

Table (2) The effect of doses of thyme and fenugreek extracts and their mixture on the biochemical properties of blood at Mid-experiment of blood serum of weaned Awassi sheep (mean \pm standard error).

Treatments	Chol.	LDL	HDL	TP	Alb.	GOT	GPT	Tri.	Glo.	Glob.
Control	0.30 \pm 5 4.30 A	0.06 \pm 3 0.06 A	0.03 \pm 1 8.03 A	0.06 \pm 6 .06 B	0.06 \pm 2 .33 C	0.33 \pm 6 5.33 B	0.16 \pm 1 9.83 B	0.16 \pm 3 7.83 A	0.33 \pm 5 5.33 A	0.13 \pm 3. 73 A
Thyme	1.30 \pm 5 2.30 BA	2.06 \pm 2 6.06 BA	0.36 \pm 1 7.36 A	0.16 \pm 5 .86 B	0.13 \pm 2 .46 C	0.66 \pm 6 4.66 B	1.50 \pm 2 2.50 BA	1.83 \pm 3 3.83 B	1.33 \pm 5 3.33 BA	0.30 \pm 3. 40 A
Fenugreek	1.20 \pm 5 2.66 BA	1.20 \pm 2 5.66 B	1.20 \pm 1 9.33 A	0.42 \pm 6 .26 B	0.11 \pm 2 .80 B	1.00 \pm 6 5.00 B	1.45 \pm 2 4.33 A	1.20 \pm 3 2.66 CB	1.52 \pm 5 3.00 BA	0.34 \pm 3. 70 A
Mixture	0.33 \pm 4 9.66 B	0.66 \pm 2 1.33 C	0.33 \pm 1 2.66 B	0.10 \pm 7 .30 A	0.06 \pm 3 .33 A	0.33 \pm 6 8.66 A	1.00 \pm 2 2.00 B	0.33 \pm 2 9.33 C	0.33 \pm 5 0.66 B	12.73 \pm 1 6.53 A

Sig.	*	*	*	*	*	*	*	*	*	N.S
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At the end of the experiment, the study results showed that adding thyme, fenugreek, and their mixture to the diets of Awassi sheep weaners had a significant effect ($P < 0.05$) on most biochemical characteristics compared to the control group. A significant decrease in total cholesterol concentration was observed in the groups treated with thyme, fenugreek, and their mixture compared to the control group, with the lowest values recorded in the thyme-only group. This result indicates that thyme possesses cholesterol-lowering properties, possibly due to its content of phenolic compounds and volatile oils that contribute to improving lipid metabolism (Hashemipour *et al.*, 2013). While Dugar (1980) explained that the polyhydroxylated phenolic compounds found in plants have the ability to inhibit free radical activity and limit lipid oxidation, these phenolic compounds cause slow oxidation of

cholesterol products. The reason for this decrease may be due to the presence of diosgenin and steroidal saponins in fenugreek seeds, which are the main components involved with cholesterol in the formation of sex hormones (Al-Obaidi, 2010).

Fenugreek seeds also contain saponins, which reduce cholesterol synthesis (Hamoudi *et al.*, 2006). Fenugreek seeds also contain steroidal compounds that work to absorb bile salts and associated cholesterol, thus reducing their absorption and excretion in the feces. These results are consistent with what Al-Janabi (2010) reported, regarding a decrease in cholesterol concentrations during late pregnancy and after birth. This study indicates that fenugreek seeds contain a high percentage of fiber. Or it may increase the conversion of cholesterol in the liver into new bile salts, thereby lowering its level in the blood (Eastwood, 1975; Tsai *et al.*, 1976; Anderson, 1990).

Table (3) The effect of doses of thyme and fenugreek extracts and their mixture on the biochemical properties of blood at End-experiment of blood serum of weaned Awassi sheep (mean \pm standard error).

Treatments	Chol.	LDL	HDL	TP	Alb.	GOT	GPT	Tri.	Glo.	Glob.
Control	0.05 \pm 5 5.20 A	0.05 \pm 30 .30 A	0.05 \pm 1 8.30 B	0.05 \pm 6 .10 B	0.05 \pm 2 .40 C	0.44 \pm 6 4.66 B	0.15 \pm 2 0.70 B	0.05 \pm 3 8.60 A	0.20 \pm 5 5.90 A	1.20 \pm 5 2.66 B
Thyme	0.65 \pm 5 2.20 B	0.14 \pm 25 .76 B	0.08 \pm 1 8.33 B	0.21 \pm 6 .06 B	0.06 \pm 2 .83 B	0.57 \pm 6 6.00 B	0.76 \pm 2 4.50 A	0.57 \pm 3 3.50 B	0.28 \pm 5 3.50 B	0.17 \pm 3. 33 B
Fenugreek	0.28 \pm 5 2.50 B	0.33 \pm 25 .56B	1.25 \pm 2 1.00 A	0.44 \pm 6 .76 BA	0.03 \pm 2 .93 A	0.60 \pm 6 6.16 B	0.96 \pm 2 3.70 A	0.36 \pm 3 2.70 B	0.26 \pm 5 3.06 B	0.28 \pm 4. 00 BA
Mixture	0.16 \pm 5	0.66 \pm 22	0.33 \pm 1	0.06 \pm 7	0.06 \pm 3	0.16 \pm 6	0.50 \pm 2	0.16 \pm 3	0.16 \pm 5	2.26 \pm 4.

	0.83	.33	3.16	.46	.63	9.83	3.00	0.66	1.66	63
	C	C	C	A	A	A	A	C	C	A
Sig.	*	*	*	*	*	*	*	*	*	*

Regarding low-density lipoprotein (LDL) cholesterol, the treatments (thyme, fenugreek, and mixture) showed a significant decrease compared to the control (thyme: 25.76, fenugreek: 25.56, and mixture: 22.33). This supports the positive effect of plant additives on cardiovascular health in animals (Abd El-Hack, 2016). These results are consistent with (Al-Janabi, 2010) regarding the reduction in LDL in the fenugreek treatment. The thyme treatment, on the other hand, was consistent with the results of (Al-Rubaie *et al.*, 2009).

The study results showed significant superiority of the mixture and fenugreek treatments in total protein concentration (7.46, 6.76) and albumin (3.63, 2.93), respectively, over the thyme and control treatments (6.06, 6.10) and albumin (2.83, 2.40), respectively. This indicates improved metabolic performance and liver function as a result of the use of plant supplements (Alagawany *et al.*, 2015). This improvement is attributed to the stimulating effects of the active compounds in thyme and fenugreek, which may enhance protein synthesis in the liver. This study is consistent with (Al-Janabi, 2010), who observed a significant increase in the addition of fenugreek seeds and its effect on the total protein and albumin levels of the

first and second groups of Shami goats, both during early and late pregnancy, and after parturition. The results of this study are consistent with several studies on the increase in total protein and albumin levels, were consistent with Nazar (1994) and Al-Saiady *et al.* (2007). However, they were inconsistent with Al-Shaikh *et al.* (1999), Boban *et al.* (2006), and Abo-El-Nor *et al.* (2007).

As for the GOT and GPT enzymes, the mixture treatment showed a significant ($P < 0.05$) superiority (69.83 GOT, 23.00 GPT) compared to the control (64.66 GOT, 20.70 GPT), indicating the role of plant additives in protecting the liver and reducing cellular damage (Abou-Elkhair *et al.*, 2014).

The results also showed a significant decrease in triglyceride concentrations across the different treatments, with levels reaching 38.60 in the control treatment, 33.50 in the thyme treatment, 32.70 in the fenugreek treatment, and 30.66 in the mixed treatment. This reflects an improvement in lipid metabolism in the treated weanlings. Previous studies have indicated that plant extracts can reduce triglyceride synthesis by inhibiting lipid-synthesizing enzymes (Saleh, 2018). The decrease in triglycerides is due to the fact that fenugreek seeds are a

rich source of fiber and pectin, which absorbs bile acids (Mitra and Bhattacharya, 2006). This reduces the level of fat absorbed from the gastrointestinal tract, as well as increases the biosynthesis of steroid hormones, which rely on these fats as a building block (Al-Janabi, 2010). Increased insulin secretion also reduces the effectiveness of lipase in fat cells, resulting in reduced breakdown of stored triglycerides and a lower blood level. Glucose concentrations were significantly reduced in the treated groups compared to the control group, indicating improved glucose utilization due to the metabolic-stimulating properties of thyme and fenugreek (Kandil, 2012). As for globulin, the results showed a significant increase in some treatments, indicating an improved immune response, which is linked to increased antioxidant activity and active compounds in the plants used (Dhama, 2015).

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