*Egypt. J. Vet. Sci.* **Vol. 54,** No. 4, pp. 621-629 (2023)

# Egyptian Journal of Veterinary Sciences
https://ejvs.journals.ekb.eg/

## Effect of Adding Omega-3 to Ration on Production and Physiological Performance of Awassi Ewes During Winter Season

**Hanan Waleed Kasim Agwaan**
*Department of Animal Production, College of Agriculture & Forestry, University of Mosul, Iraq.*

OMEGA-3 fatty acids are reported to play an important role for healthy cell membranes, which are necessary for both human and animal health. The aim of this study was to study the role of supplementation of omega-3 in rations of ewes exposed to cold stress during the three winter months on productive performance of ewes. Adding 35ml of omega-3 to Awassi ewes ration led to increasing weight gain during the months of January and February (10-12◦C). At the same time that feed intake rates reached their lowest levels in ewes who dosed 35 ml of Omega in the three months of the study, feed conversion ratio for those ewes improved. All physiological traits represented like body temperature, heart and breath rate stabilized and significantly improved in cold-stressed sheep, especially in those that were add 35 ml of omega-3. All physiological biomarkers corresponded to a clear improvement in the levels of Thyroid-Stimulating Hormone (TSH) and Thyroxin (T4) and in both 25 and 35Omega3 treatment groups, which, in turn, outperformed the control group. It can be concluded that adding omega3 to ewes ration improved productive and physiological traits.

**Keywords:** Omega-3, Production, Sheep, Thyroid, Vital signs.

## Introduction

As alternatives to pharmaceuticals and chemical medications, the most of which had harmful side effects, many nations turned to the use of fish and its oil as well as herbal and medicinal substances, plants, and certain naturally occurring animal products [1]. Alpha linolenic acid (ALA), eicosatetraenoic acid (EPA), and docosahexaenoic acid (DHA) are three omega-3 fatty acids that are crucial for healthy cell membranes, which are necessary for both human and animal health and appropriate physiological function [2]. Since none of these fatty acids are created naturally by the body, they must be obtained from food [3] While EPA and DHA are found in large quantities in marine fish oil and microalgae, ALA is derived from vegetable oil, which is a form of fatty acid that contains polyunsaturated fatty acids [1]. Dietary fat supplements are commonly used to increase the energy density of ration. Docosahexaenoic acid (22: 6, DHA), one of the two types of fatty acids, and eicosatetraenoic acid (20:5, EPA) are two of the crucial necessary fatty acids which found in Omega components that all the body requirements [4]. Omega-3 is currently known through research in several species that the special advantages of it are because of the presence of Omega-3 necessary long chain fatty acids (LCPs) [5]. These long chain fatty acids activate reproductive and health effects while avoiding rumen alterations [6]. The long chain fatty acids EPA and DHA are particularly abundant in salmon oil, according to Coleman et al. [7], feeding salmon oil directly affects a flock's entire production cycle, including fertility, milk production in ewes, which improves lamb viability, and general health and disease resistance. Omega-3 DHA and EPA-rich sheep diet has been showed to enhance critical aspects of the whole sheep reproductive cycle, as well as to provide lambs a better start in life because of the mother's higher-quality colostrum and the greater resilience to obstacles encountered early in life [8].

Corresponding author: Hanan Waleed Kasim, E-mail: hanan_aqwaan@uomosul.edu.iq, Tel.: 009647739050821
ORCID: 0000-0002-1206-0030
(Received 12/03/2023, accepted 01/05/2023)
DOI: 10.21608/EJVS.2023.199515.1461

*Aim of the study:* Because of the limited studies addressing the effect of Omega-3 on general productive and physiological and metabolic vital systems, therefore, this study was conducted to show the effect of Omega-3 at two levels during the cold winter season on the productive and physiological performance and the concentration of thyroid stimulating and thyroxin hormone in Awassi ewes, because dietary fat supplements are commonly used to increase the energy density of rations.

## Material and Methods

*Experimental animals:* This study was conducted in a private field, North of Mosul, Baybukht region from December 2022 to February 2023(10-12◦C). A total of twenty-one Awassi ewes' weight (41.35 ±1.5 kg) and age between 1.5 and 2 years were used. Following 15 days adaptation period, ewes were assigned to three equal groups (7 ewes/ group) and received concentrate 3% of their live weight [9]. The first group(control) fed on concentrate diet with no additive, while T1 and T2, Omega 3 was added to the basal diet at the rate of 25 and 35 ml/ ewe respectively. The ingredient and chemical composition of the basal diet is shown in Table 1. Clean water was available constantly.

*Physiological vital indicators:* The bio physiological indicators (respiratory rate, rectal temperature and pulse rate) of each of the experimental animals were measured when animals in rest state in single cage at the end of each month of the experiment, as the measurement process for the three groups took from one to one and management strategies.

*Blood measurements:* Blood samples were collected from all the ewes monthly, blood samples were taken from the jugular vein using evacuated blood collection tubes ,serum was separated by leaving the sample in an upright position for 2 h in a cooling box followed by centrifugation at 3000 rpm/ min .Quantitative determinations of ewes thyroid stimulating hormone (TSH) and T4 were estimated in undiluted serum samples using an ELISA Kit (MyBiosource, Mumbai, Maharashatra, India).

*Statistical analysis:* Statistical analysis: Experimental data were statistically analyzed using completely randomized design (CRD), and significant differences among means were tested according to Duncan [9].

## Results and Discussion

*Productive traits* The results of the statistical analysis of Tables (2) indicated that there was no significant increase in the body weights of ewes and in their daily weight gain in December, While dry matter intake in the T2 group decreased (P≤0.05) compared to the rest of the transactions, the same group recorded the lowest feed conversion ratio (P≤0.05) compared to the control group. In January, the T2 group recorded a significant increase (P≤0.05) in the body weights of the ewes compared to the control group. The daily weight gain in T2 was also at its highest level (P≤0.05) compared to T1 and control groups, as was the case in February. While the dry matter intake and the feed conversion ratio for group T2 were at their lowest level in January (P≤0.05) compared to other experimental groups. While the body weights of the ewes of the T2 group recorded their highest levels (P≤0.05), so did the daily weight gain in February, Results of the present study revealed that body weight of Awassi ewes were significantly increased by adding Omega 3 during the second and third month of experiment because fat supplementation balances energy level and promotes follicular growth and development [12,13] While in the last month of the experiment (February), dry matter intake in T2 was at its lowest (P≤0.05) compared to other experimental groups. The T1 and T2 groups also recorded the lowest feed conversion ratio (P≤0.05) in February compared to the control group.

According to the present study, body Weight and daily weight gain increased significantly as shown in Table (2). Dietary Omega-3 fatty acids are also essential for normal growth and metabolism. The results of current study agreed with Okukpe et al. [14],which found that Omega-3 fatty acids enhance feed intake and feed efficiency when given to dwarf goats at various dosages over eight weeks. Supplemental Omega-3 in the diet renders the fat in the rumen somewhat inactive and may occasionally prevent a decline in the intake of dry matter [15]. Certain amino acids and fatty acids may be able to avoid ruminal bio hydrogenation and then become accessible for absorption and use in the small intestine, which thus leads to the maximum use of structural as well as fatty amino acids, thus supporting the process of protein synthesis, activating metabolic activity in the body and increasing muscle mass in the animal, leading to an increase in its weight and improve the energy balance and increase body weight gain

[16]. The addition of Omega-3 fatty acids reduced the amount of dry matter intake but considerably increased daily weight gain, showing improved feed conversion ratio [17].

The results of the current study agree that the decrease in the coefficient of nutritional conversion in Table (2) for the coefficients T1 and T2 with the findings of [18] which found that when lambs fed an Omega-3 diet put on the maximum weight, gaining 230 g/head/day, compared to 224 g/head/day for lambs fed whole grain pellets (MSM) and 194 g/head/day for controls ($p \leq 0.0390$). As a result of Omega-3 intake to livestock, the higher average daily gain with lower feed intake suggests better nutrient utilization from the abomasum, either as a result of altered rumen ecology and environment that favors less bio hydrogenation and more by-pass proteins from the rumen or as a result of a higher rate of turnover of volatile fatty acid absorption [3]. Economically speaking, the somewhat higher feed cost of the Omega-3 fortified diet was offset by the best feed efficiency, which involved eating less feed but increasing average daily gains the quickest [19]. This resulted in increased profitability. Therefore, adding Omega-3 to diets improved feed conversion ratio, which may help lot-fed lambs reach finishing weight earlier and save expenses while increasing profitability for sheep producers [20].

*Physiological vital indicators*

Throughout the three months of the experiment, results of the statistical analysis of Table (3) indicated that the body temperature, pulse and breath rates of the ewes in T2 showed a significant decrease ($P \leq 0.05$) compared to the T1 and control group. Although there were computational differences in T1 compared to the control group, they were insignificant. Despite the significant decrease in the T2 group compared to the control, the T2 treatment had the lowest rates for their body temperature, heart rate, as well as respiratory rate compared to both the T1 and control groups

The significant effect of Omega-3 recorded rectal temperature is attributed to the positive effect of Omega-3 - in reducing the effect of stress in periods of low temperatures in winter, especially in the concentration of 35 ml/ewe (T2), which was given Omega-3 compared to the control group, which led to a decrease in overall body temperature and rectal temperature within normal range. During the winter, Omega 3 PUFAs act as mediators of thermogenesis [5]. Omega-3 fatty acids help maintain body temperature under environmental stress condition [18]. It is well known that dietary Omega-3 has an important role in producing eicosanoids, which regulate body temperature. In general, giving animals Omega-3 fatty acids may improve their health for three months[21] .The parent components of several end cannabinoids and end cannabinoid-like chemicals, including Omega-3, are known to influence the immune system, neurotransmission, metabolism, thermogenesis, circadian rhythms, and thermogenesis[22] .In addition, studies have demonstrated that Omega-3 fatty acid treatment maintain breathing [23] .Omega-3 fatty acid supplementation can actually improve lung function, and may improve lung health and lower the incidence of exercise-induced asthma[24] .So, Fish oil reduces heart rate and oxygen consumption during exercise[25]. Supplementing with Omega-3 fatty acids increases maximum oxygen absorption and endothelial function under cold stress conditions [26].

**TABLE 1. Ingredient and chemical composition of basal ration.**

| *Ingredient | % | **Chemical composition | % |
|---|---|---|---|
| Crushed barley | 23 | Dry matter | 92.77 |
| Wheat bran | 22 | Organic matter | 95.31 |
| Soybean | 10 | Crud protein | 13.80 |
| Yellow corn | 18 | Crud fiber | 6.23 |
| Wheat | 26 | Ether extract | 1.41 |
| Salt | 0.5 | Metabolizable energy Kcal/Kg | 2585 |
| Limestone | 0.5 | | |

*Ingredient of ration according to NRC [10]
** Chemical composition according to AOAC(2002) [11]

TABLE 2. The effect of adding Omega-3 concentrations on some productive traits in Awassi ewes (means±SE) .

| Parameters | Months | Control No additive | T1 Added 25ml Omega3 /ewe | T2 Added 35ml Omega3 /ewe |
|---|---|---|---|---|
| Body weight (Kg) | December | 41.19 ± 1.27[a] | 46.26 ± 1.4[a] | 47.35 ± 1.57[a] |
| | January | 48.8 ± 1.53[b] | 51.23 ± 1.71[ab] | 56.82 ± 1.77 [a] |
| | February | 52.47 ± 1.55[b] | 57.48 ± 1.91[b] | 68.51 ± 1.28[a] |
| Daily weight gain(gm) | December | 3.19±0.72[a] | 5.26±1.07[a] | 6.35±1.21[a] |
| | January | 4.61±0.46[b] | 4.97±0.84[b] | 9.47±1.19[a] |
| | February | 3.67±0.84[b] | 6.25±1.45[b] | 11.69±0.94[a] |
| Dry matter intake | December | 25.17±1.06[a] | 22.51±1.27[a] | 17.34±1.41[b] |
| | January | 24.52±1.31[a] | 22.82±0.94[a] | 16.48±1.28[b] |
| | February | 28.43±1.67[a] | 20.74±1.35[b] | 12.89±1.17 [c] |
| Feed conversion ratio kg/kg | December | 7.9±1.82[a] | 4.3±1.08[ab] | 2.7±1.1[b] |
| | January | 5.3±1.2[a] | 5.6±1.17[a] | 1.7±0.87[b] |
| | February | 7.7±1.37[a] | 3.3±1.3[b] | 1.1±0.94[b] |

Data expressed as (Mean ± Stander error).
Different letters in a row to every trait indicate significant differences at (P≤ 0.05).

TABLE 3. The effect of Omega-3 concentrations dosing on some Physiological vital indicators in Awassi ewes (means±SE).

| Parameters | Months | Control No additive | T1 Added 25ml Omega3 /ewe | T2 Added 35ml Omega3 /ewe |
|---|---|---|---|---|
| (Body Temperature (˚C | December | 39.06 ± 0.35[a] | 38.95 ± 0.11[a] | 37.68 ± 0.31[b] |
| | January | 39.67 ± 0.17[a] | 39.4 ± 0.29[a] | 37.72 ± 0.28[b] |
| | February | 39.51 ± 0.16[a] | 39.12 ± 0.12[a] | 37.52 ± 0.46[b] |
| Pulse Rate (Pulse/ minute) | December | 75.0 ± 1.09[a] | 1.26[ab] ± 73.31 | 71.1 ± 4.20[b] |
| | January | 75.62 ± 2.98[a] | 1.15[a] ± 74.07 | 1.48[b] ± 70.38 |
| | February | 75.05 ± 1.88[a] | 1.06[a] ± 73.12 | 68.14 ± 4.20[b] |
| Breath Rate (Respiratory cycle / minute) | December | 53.24 ± 1.09[a] | 1.18[a] ± 50.7 | 44.37 ± 1.1[b] |
| | January | 56.19 ± 1.24[a] | 52.44 ± 1.52[a] | 42.48 ± 1.71[b] |
| | February | 54.67 ± 1.13[a] | 50.31 ± 1.3[a] | 1.52[b] ± 38.25 |

Data expressed as (Mean ± Stander error).
Different letters in a row to every trait indicate significant differences at (P≤ 0.05).

The potential advantages of the Omega-3 fatty acids EPA and DHA for heart health are well demonstrated in several research. Omega-3 fatty acids may lower heart rate through a variety of processes, but a recent study found that they also directly affect the electrical excitability of cardiac cell membranes, which lowers heart rate [27]. Omega 3 polyunsaturated fatty acids have many biological functions, including the regulation of immune function and improvement of the antioxidant capacity which can help in the treatment of diarrhea, also EPA is seen to have an anti-inflammatory effect particularly in cases of cardiovascular disease. Omega-3 fatty acids had a similar impact on cell excitability to the class I antiarrhythmic medication lidocaine [28] .By perusing the cells with fatty acid-free bovine serum albumin, the omega-3 fatty acid-induced reduction in the beating rate could be easily reversed [17] .This suggests that Omega-3 fatty acids probably do not need to be incorporated into the membrane phospholipid or covalently linked to membrane components in order to be effective. These findings imply that Omega-3 fatty acids have the ability to reduce heart rate by inhibiting the automaticity of ventricular contraction when present in their free form [29].

While branched-chain amino acids and omega-3 fatty acids can boost maximal oxygen intake Vo2max, some dietary supplements, such as omega-3, can increase oxygenation through improved blood flow [23].

*Thyroid Stimulating Hormone (TSH) &Thyroxin Hormone (T4)*

Blood samples were collected via jugular venipuncture from each ewe after disinfecting area with ethyl alcohol 70%.The samples were placed into a vacutainer tube without anticoagulant for serum separation. Serum was separated by leaving the sample in an upright position for 2 h in a cooling box followed by centrifugation at 3000 rpm/ min. Quantitative determinations of ewes thyroid stimulating hormone (TSH) and T4 were estimated in undiluted serum samples using an ELISA Kit (MyBiosource, Mumbai, Maharashatra, India). The detection assay procedures and calculations were undertaken according to the manufacturer's guidelines and protocol, throughout the three months of the experiment, the ewes of T2 group recorded the highest significant level ($P \leq 0.05$) of thyroxin and thyroid stimulating hormone compared to the T1 and control group. Also, the ewes of T1 group outperformed the ewes of the control group as shown in Table (4).

As the temperature drops in winter season as cold weather, thyroid gland of livestock has to activate more than other seasons to keep animals' body warm [30]. The secretion of thyroxin from the thyroid gland is affected when the temperature decreases, which leads to an improvement in vital functions in the body [31]. The secretion of thyroxin from the thyroid gland is affected when the temperature decreases, which leads to an improvement in the vital functions of the sheep's bodies, but their continued exposure to constant cold stress may make the thyroid gland unable to fill the body's need for metabolites and the represented energy necessary for the continuity of vital body activities [32]. Omega-3 fatty acids namely alpha linoleic acid (ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are important components of cell membranes essential for health and normal physiological functioning of livestock [33], Omega-3 fatty acids modify gene transcription in addition to altering signal transmission through altering the lipid content of cell membranes [34]. Omega-3 fatty acids may change thyroid hormone production and release, TSH receptor function, signal transduction, and thyroid follicular cell proliferation and differentiation through these pathways [35]. Studies examining the impact of Omega-3 dietary supplements, plasma concentrations, or erythrocyte cell membrane concentration on the initiation of cold stress on ewes and their lambs [7]. Nutritional supplementation of omega-3 fatty acids could be related to moderate modulations in lipid metabolism in the adipose and inflammation in liver of per partum dairy cows [36] .Numerous variables, including dietary supplements like Omega-3, have an impact on thyroid hormones, which are primarily responsible for controlling tissue development and metabolism. This is corroborated by the findings of Vanderpas [37], which came to the conclusion that an increase in basal metabolic rate causes an increase in body weight growth. These findings showed that the enzymes boosted deiodinase activity in the liver and kidney tissues, converting T4 into the physiologically active hormone T3, either directly or indirectly [38]. The youngsters' rapid development and higher metabolic rates were caused by this in turn. Furthermore, [39] supported the current findings, indicating that T3 levels were reliable predictors of growth variations. Exogenous enzymes seem to have altered the concentrations of T4 and T3, increasing the levels of T3 in the blood.

**TABLE 4. The effect of Omega-3 concentrations dosing on TSH and T4 in Awassi ewes (means±SE).**

| Parameters | Months | Control No additive | T1 Added 25ml Omega3/ewe | T2 Added 35ml Omega3 /ewe |
|---|---|---|---|---|
| TSH (ng/ml) | December | 2.42 ± 0.36[c] | 2.87 ± 0.31[b] | 4.13 ± 0.19[a] |
| | January | 2.35 ± 0.24[c] | 2.93 ± 0.22[b] | 4.21 ± 0.21[a] |
| | February | 2.51± 0.18[c] | 3.36 ± 0.28[b] | 4.43 ± 0.33[a] |
| T4 (ng/ml) | December | 2.37 ± 0.13[c] | 2.78 ± 0.31[b] | 3.47 ± 0.23[a] |
| | January | 2.25 ± 0.11[c] | 2.91 ± 0.22[b] | 3.53 ± 0.14[a] |
| | February | 2.43 ± 0.1[c] | 3.02 ± 0.28[b] | 3.89 ± 0.15[a] |

Data expressed as (Mean ± Stander error).
Different letters in a row to every trait indicate significant differences at ($P \leq 0.05$).

## Conclusion

According to the results obtained from the study, we concluded:

1. Using of Omega-3 in lamb, ages differences and compared its effects, adult ewes and during the cold stress period in the winter months has led to a noticeable improvement in their weight, monthly weight gain and nutritional conversion efficiency.
2. During the research, the physiological measurements of vital signs such as body temperature, heart rate and breathing in Omega-3 treated ewes during the winter months and cold stress were found to be stable and keep within normal range in the sheep.
3. The treatment of cold-stressed ewes with Omega-3 led to an improvement in Thyroid-Stimulating Hormone as well as an improvement in thyroxin levels, which is responsible for regulating all metabolic processes in the body required for energy production and its balance in the conditions of the environment of ewes raised in the cold winter months.

*Acknowledgments*

The author is very grateful to the University of Mosul/ College of College of Agriculture & Forestry provided facilities, which helped improve the quality of this work. Also thank Dr. Dhyaa Mohammed Taher Jwher , Department of veterinary Public health College of Veterinary Medicine, University of Mosul for his assistant work.

*Conflicts of interest*

The researcher declare that there is no conflict of interest of this work

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## تاثير اضافه اوميجا-٣ الى العليقة في الاداء الانتاجي والفسلجي للنعاج العواسيه خلال موسم الشتاء

**حنان وليد قاسم أغوان**
قسم الانتاج الحيواني - كليه الزراعة والغابات - جامعه الموصل - العراق.

أدى استخدام أوميجا ٣ بتركيزين مختلفين إلى تعرض النعاج البالغة للإجهاد البارد خلال أشهر الشتاء الثلاثة. اضافه ٣٥ مل من أوميجا ٣ إلى عليقه النعاج العواسيه ادى زيادة الوزن خلال شهري كانون الثاني وشباط (١٠-١٢) م°. في نفس الوقت الذي وصلت فيه معدلات استهلاك العلف إلى أدنى مستوياتها في النعاج التي تناولت ٣٥ مل من أوميجا٣ في الأشهر الثلاثة من الدراسة ، تحسنت كفاءة معدلات التحويل الغذائي لتلك النعاج. استقرت جميع المؤشرات الحيوية الفسيولوجية المتمثلة في درجة حرارة الجسم ومعدل ضربات القلب والتنفس وتحسنت بشكل ملحوظ في الأغنام المجهدة بالبرد ، خاصة في تلك التي تم تناولها بجرعة ٣٥ مل من أوميجا ٣. تتوافق جميع قياسات الإنتاجية والعلامات الحيوية الفسيولوجية مع تحسن واضح في مستويات هرمون الغدة الدرقية والثيروكسين وفي كل من مجموعتي المعاملة ب ٢٥ و ٣٥ مل من الاوميجا ٣ ، والتي بدورها تفوقت على مجموعة السيطرة .نستنتج من ذلك بان اضافه اوميجا ٣ لعليقه النعاج حسنت من القياسات الانتاجيه والفسلجيه .

**الكلمات المفتاحية:** أوميجا ٣ ، الإنتاج ، المؤشرات الحيوية ، الأغنام ، الغدة الدرقية.