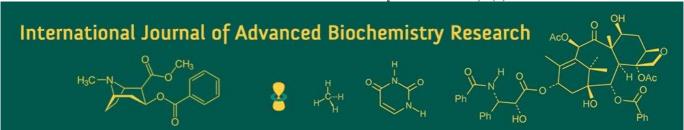
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The effect of two levels of AD3EC on the physiological characteristics of local lambs

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Abstract

This study aimed to investigate the effect of two levels of AD3EC on the physiological characteristics of local sheep lambs. The study period extended over three months, from, $2\1\2025$ to $1\4\2025$. Twelve weaned-age local sheep lambs were used in this experiment, with four lambs per treatment. The first treatment was the control and received no AD3EC. The second treatment received 4 ml of AD3EC, and the third treatment received 3 ml of AD3EC. The AD3EC vitamins improved some physiological characteristics in this study. The results showed a significant increase in packed cell volume, hemoglobin, and white blood cells in the third month for the third treatment compared to the first and second treatments.

Keywords: AD3EC, physiological characteristics, local lambs, hematological parameters

Introduction

After weaning, sheep require nutritional supplements to improve blood parameters. These supplements include the AD3EC complex vitamins, which support growth, improve the immune system, and aid in nutrient absorption. For example, vitamin A is important for maintaining bone growth, vision, and immune function Tanumihardjo (2011) [8]. A study by Soliman (2002) [7] demonstrated the effect of vitamin A supplementation on the physiological responses of ewes and their lambs. Physiological blood parameters improved in the groups that received vitamin A. Vitamin D3 also contributes to the absorption of calcium and phosphorus. Any deficiency in vitamin D3 causes clinical diseases in lambs and improper skeletal formation, as demonstrated in a study by Saleh et al. (2023) [6]. This study demonstrated the effect of vitamin D3 deficiency on Najdi lambs, where the deficiency led to clinical symptoms in temperature, respiration, and heart rate, as well as a decrease in blood components and the occurrence of clinical and blood diseases. Vitamin E is considered an important antioxidant for improving blood parameters. A study by Ali (2023) [1] demonstrated the effect of vitamin E injections on Awassi ewes on some physiological characteristics of their offspring; lambs born to ewes treated with vitamin E showed an increase in blood indicators of hemoglobin, packed cell volume, and white blood cells compared to the control group. As for vitamin C, it is a powerful antioxidant that enhances the health of the immune system of lambs and protects them from infection by enhancing the activity of phagocytic white blood cells to resist bacterial and viral diseases, as shown in a study by Massoudi (2025) [4], where vitamin C made lambs more resistant to pneumonia by improving their blood physiological characteristics. This study aimed to determine the effect of two levels of AD3EC on some blood physiological characteristics-PCV, BH, and WBC—in local sheep lambs.

Materials and Methods

The experiment was conducted from \$\lambda\$1\2025, to \$1\4\2025\$, for a period of three months. It was conducted at the experimental farm of the College of Agriculture and Marshlands, University of Thi Qar. Twelve lambs were divided into three groups, each containing four lambs. The first group, the control group, did not receive vitamins. The second group received 3 ml of AD3EC after diluting it with distilled water. The third group received 4 ml of AD3EC after diluting it with distilled water every five days, in a single dose, according to medical instructions throughout the study period. All lambs were fed dry and green

Corresponding Author: Huda Y Shtib Al- Khafaji College of Agriculture and Marshlands, University of Thi-Qar, Iraq concentrated feed. The concentrated feed mixture consisted of 40% crushed barley, 25% wheat bran, 12% crushed yellow corn, 7% flour, 13% soybeans, and 3% salts. All medical care requirements were provided, including vaccinations for the lambs from the first day. During the period of raising them, they were vaccinated against diseases such as foot-and-mouth disease, lymphadenitis, septicemia, clostridium difficile, and smallpox. As for taking blood samples for laboratory tests, blood samples were taken monthly at the end of each month of the experiment. Blood was drawn from the jugular vein in the neck of the lambs and stored in gel tubes to prevent clotting. These samples were later used to measure packed cell volume, hemoglobin, and white blood cells, as follows:

Measuring the Volume of Packed Cells

The microhematocrit method was used. Blood samples were drawn from gel tubes using a special hematocrit capillary tube placed inside them to draw blood using the capillary action, filling the capillary tube to 80%. The end of the capillary tube, marked with wax, was then sealed, and the microcentrifuge was placed. The device was then covered and operated for 5 minutes at a speed ranging from 10,000 to 15,000 rpm. After the device stopped, it was removed. The tubes were placed on a ruler used to measure packed blood cells. The measurements were read. The blood height in the tube was calculated and recorded.

Hemoglobin Measurement

Hemoglobin measurement was performed using the Drabkin method. Twenty microliters of EDTA blood samples were taken and added to 5 ml of Drabkin's solution. The samples were mixed well and placed at room temperature for 10 to 15 minutes until they turned brown. They were then measured using a spectrophotometer at a wavelength of 540 nanometers. The measurements were then read and recorded.

Calculating the Number of White Blood Cells

First, a solution was prepared for counting white blood cells. It was made from 2% glacial acetic acid and 2-3 drops of methylene blue or gentian violet, prepared in 98 ml of distilled water. This solution lysed the red blood cells. Three

hundred eighty microliters of this solution were taken using a micropipette and placed in a test tube. Then, 20 microliters of EDTA blood samples were added. The mixture was mixed well and left for three minutes until all the RBCs were lysed and the WBCs were stained. A chamber slide, called a hemocytometer, was prepared with a cover glass. The space between them was filled with the diluted sample solution, ensuring that no air bubbles were present. The chamber slide was left for three minutes to settle on the slide. Using the objective lens of a microscope with a magnification of $10\times$ or $40\times$, the cells in the four squares designated for counting the WBCs were counted and the results were recorded. The number of cells counted in each square was calculated. The total number of white blood cells was determined using the formula:

Number of cells counted in each square = Dilution ratio \times 4 (for the four squares).

Results and Discussion Packed cell volume

The experimental results showed a significant difference (P<0.05) in the second and third months of the experiment. In the second month, treatment three (4 mL of AD3EC), with a mean of 49.85%, significantly outperformed treatments one and two, with a mean of 43.66% and 40.00%, respectively. In the third month, treatment three (4 mL of AD3EC) was again significantly higher, with a mean of 55.23%, for treatment two 48.25%, compared to, and for treatment the control 41.10%. In the first month, no significant differences were observed; however, treatment three (4 mL of AD3EC) had the highest mean (39.43%), followed by treatment two (3 mL of AD3EC) with a mean of 36.23%, and the control treatment with a mean of 33.30%, as shown in Table 1.

Vitamins AD3EC protected platelets and red blood cells in the goats, leading to improved red blood cell counts in the AD3EC-treated groups. This improvement was also reported in other studies, such as the study by Likttrakulwong (2022), who injected dairy cows intramuscularly with AD3EC vitamins and observed improved hematological parameters during pregnancy.

Table 1: Shows the packed blood cell volume of lambs in the control group T1 and the two treatment groups with two levels of AD3EC during the months of the experiment

Experimental treatment	Packed cell volume			
	The first month	The second month	The third month	
Control treatment	33.30 ± 4.02	40.00±1.82 b	41.10± 1.80c	
Second treatment	36.23±4.14	43.66±2.21 b	48.25±0.96b	
Third treatment	39.43±4.52	49.85± 2.21 a	55.23±2.6a	
Significant	P<0.05	P<0.05	P<0.05	

^{*}Different letters indicate significant differences between treatments.* The first treatment, the control, was not given AD3EC. The second treatment was given 3 ml of AD3EC. The third treatment was given 4 ml of AD3EC.

Hemoglobin

The results showed a significant increase in hemoglobin levels in the third month of the experiment. The third treatment, which received 4 ml of AD3EC, with an average of, 19.75 g/cm3, outperformed the second treatment, which received 3 ml of AD3EC, with an average of 17.74 g/cm3, compared to the control treatment, which averaged 13.53 g/cm3. the control treatment Similarly, in the second month, the third treatment, which received 4 ml of AD3EC, with an average of 18.00 g/cm3.significantly outperformed, which

received 3 ml of AD3EC,.This was followed by the second treatment, which received 3 ml of AD3EC, with an average of 16.25 g/cm3. Compared to the control group, the lowest percentage was 14.51 g/cm3. However, in the first month of the experiment, there were no significant differences, with an increase in hemoglobin values for the third treatment, which received 4 ml of AD3EC, with an average of 11.42 g/cm3, followed by the second treatment, which received 3 ml of AD3EC. The average AD3EC level was 10.23 g/cm3, and finally, the control treatment averaged 9.47 g/cm3, as

shown in Table 2. Vitamin A increases the activity of enzymes and protects blood cells from breakdown, which increases hemoglobin levels (Nadide and Ebru, 2005) ^[5]. This study is consistent with a study by Likttrakulwong (2022) in which he conducted an experiment on dairy cows.

Injecting AD3EC vitamins intramuscularly improved the blood parameters of cows during pregnancy. It is also consistent with a study by Daghash (1993) [2] to evaluate the effect of AD3EC injections on buffalo calves, as the vitamins improved physiological blood parameters.

Table 2: Hemoglobin g/cm3 of lambs in the control group and the two treatment groups with two levels of AD3EC during the months of the experiment

Experimental treatment	hemoglobin			
	The first month	The second month	The third month	
Control treatment	9. 47± 1.18	14.51 ± 0.47 c	$13.53 \pm 0.52c$	
Second treatment	10.23±1.50	16.25±0.97b	17.74±0.58b	
Third treatment	11.42±1.54	18.00± 0.89a	19.75±0.96a	
Significant	Non-significant	P<0.05	P<0.05	

^{*}Different letters indicate significant differences between treatments.* The first treatment, the control, was not given AD3EC. The second treatment was given 3 ml of AD3EC. The third treatment was given 4 ml of AD3EC.

White Blood Cell Counts

The results of the experiment showed a significant increase in white blood cell counts in the third and second months compared to the results of the first month. In the third month, the significant increase was for the third treatment, which received 4 ml of AD3EC, with an average of 9.87 x 10^3 cells/cm3. This was followed by the second treatment, which received 3 ml of AD3EC, with an average of 7.97 x 10^3 cells/cm3. The lowest was the control treatment, which had an average of 3.05 x 10^3 cells/cm. In the second month of the experiment, the third treatment, which received 4 ml of AD3EC, had the highest white blood cell count, with an average of 9.78 x 10^3 cells/cm. This was followed by the second treatment, which received 3 ml of AD3EC, with an average of 6.74 x 10^3 cells/cm, compared to the average of the first control treatment, which reached 3.61 x

 $10^3 cells/cm.No$ significant differences were observed during the first month of the experiment, with the third treatment that received 4 ml of AD3EC, having a superior value, with an average of 3.66 x 10^3 cells/cm3, followed by the second treatment that received 3 ml of AD3EC with an average of 3.59 x 10^3 cells/cm3, compared to the control treatment that averaged 3.58 x 10^3 cells/cm3.

AD3EC complex vitamins work to overcome stress factors affecting lamb health and enhance the immune system's ability to resist all diseases by improving the level of white blood cell count within physiological parameters, as in the study by Likttrakulwong (2022) in his experiment on dairy cows, where they were injected intramuscularly with AD3EC vitamins. This also agrees with a study conducted by Daghash (1993) [2] to evaluate the effect of AD3EC injections on buffalo calves.

Table 3: Shows the number of white blood cells x 10³ cells/mm3 for the sheep of the control group T1 and the two treatment groups with two levels of AD3EC during the months of the experiment

Experimental treatment	White blood cells		
	The first month	The second month	The third month
Control treatment	3.58± 0.65	3.61± 0.26b	$3.05 \pm 0.26c$
Second treatment	3.59±0.86	6.74±0.55a	7.97±0.54b
Third treatment	3.66 ±0.87	$78.9 \pm 0.58a$	9.87 ±0.81a
Significant	Non-significant	P<0.05	P<0.05

^{*}Different letters indicate significant differences between treatments.* The first treatment, the control, was not given AD3EC. The second treatment was given 3 ml of AD3EC. The third treatment was given 4 ml of AD3EC.

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