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Relationship of LMO4 Gene Polymorphism with Milk Production in Iraqi Sheep

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Abstract: The present experiment was conducted in the middle region of Iraq in 2023 on 120 Awassi unimproved breed ewes to determine the effect of LMO4 gene polymorphism on lactation period and milk yield. The LMO4 gene position 20782 consisted of two co-dominant alleles with three genotypes. The results showed a notable difference ($P \leq 0.01$) at the distribution of the samples according to the alleles that were brought about by single nucleotide polymorphism at both places. The LMO4 genotype had a major impact ($P \leq 0.05$) on the total milk yield, the results showed, since ewes carrying the wild type (GG) yielded the largest amount of milk (80.13 kg), while ewes carrying the hetero type (GA) yielded the smallest amount (71.9 kg). The findings showed that the LMO4 genotype had a significant ($P < 0.05$) impact on lactation length; wild genotype (GG) carrying ewes had the longest milk season or lactation length of 12.10 days, and mutant genotype (AA) carrying ewes had the shortest lactation length of 113.70 days.

Keywords: *LMO4 Gene, Awassi Sheep, Milk Production.*

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I. INTRODUCTION

Since the majority of Iraqis prefer mouton, Iraqi sheep breeds are very important, extensively found throughout the entire country, and highly resilient. They are also regarded as the primary source of income for most families in desert nations or regions. Because of its favorable traits and vulnerability to harsh agricultural conditions like low pastures and heat stress in semi-tropical regions, the Awassi sheep breed is among the most significant and famous in Iraq [1].

According to a number of previous studies, a number of genetic markers may increase or define a breed's performance and prove useful in selection. An example of such a marker is the LMO4 gene, which is found on sheep chromosome 21 and consists of two bases. It is regarded as an essential member of the family of genes coding cystein-rich proteins [2].

The LMO4 gene, as per the recent studies, is one of a series of genes involved in muscular functions or activities that sustain body growth and development. It also has a function in histon modification during mitotic cell division [3].

[4] showed that the LMO4 gene is involved in the smooth cell activity and protein control in all mammalian bodies. According to other studies like [5,6], the LMO4 gene is the primary gene coding for the special and unique kinds of proteins responsible for the adaptation of the muscle fibers of mammalian species to provide various activities.

Investigating the LMO4 gene single nucleotide polymorphism, determining its impact on sheep milk yield, and using genotypes to choose the optimum individuals through indirect selection are the main goals of the current study.

II. MATERIALS AND METHODS

120 ewes from the indigenous, unimproved Awassi breed were used in the current investigation, which was carried out in 2023 at a private sheep breeding facility in the center of Iraq.

During that period, genetic analysis was carried out at the ASCO lab in Baghdad (AL-Harithya) to isolate DNA and determine the genotypes of the cystein rich protein gene (LMO4) at position 20782.

Blood samples were taken from the jugular vein in compliance with the technique of [7], and a Promega company supplement was used to extract DNA. The target area, which is found in the second exon, was amplified and detected using the primer:

- *GCCAGATTATGGGACTCAAG and TGCAAAGCCTGTGGTAAG*

The product size was 908 bp, and for both forward and reverse procedures, the annealing temperature was around 60 c°.

The milko-scan gadget was used to evaluate the protein, fat, and lactose levels of the milk right after milking, and each ewe's milk supply was monitored in the station.

A Quantus Fluorometer was used to evaluate the concentration of extracted DNA in order to assess the quality of samples for use in later procedures. One microliter of DNA was mixed with 200 microliters of diluted Quantifluor Dye. After five minutes of room temperature incubation, DNA concentration values were obtained.

PCR products were sent to Macrogen Corporation in Korea so they could be Sanger sequenced using the ABI3730XL, an automated DNA sequencer. Geneious software was utilized for analysis when the findings were received over email.

- *Statistical Analysis:*

Data were analyzed using SAS [8] computer and Chi-square test was used to determine the significant differences among phenotypes:

$$X^2 = \sum \frac{(\text{Observed No.} - \text{Expected No.})^2}{\text{Expected No.}}.$$

According to the linear model below, the significant difference between groups that differed with genotypes (mutant, hetero, and wild) was ascertained using the Duncan multiple range test [9] and the analyses of variance technique in accordance with a totally randomized design:

following model:

$Yijk = \mu + Ti + eijk$

Where: μ: the overall mean
Ti: effect of single nucleotide change
eijk : is a random error.

III. RESULTS AND DISCUSSION

According to alleles resulting from single nucleotide polymorphism in both positions, the results indicated a significant difference ($P \leq 0.01$) in the sample distribution (Table 1). Three genotypes (GG, GA, and AA) and two co-dominant alleles were found at position 20782 of the LMO4 gene. These were caused by point mutations that changed the nitrogen base from guanine (wild type) to adenine (mutant type).

The genotype rates for GG, GA, and AA were 54.17, 30.00, and 15.83%, respectively, whereas the allele frequencies for the G and A alleles were 0.704 and 0.296.

Table -1: Distribution of Genotypes and Allele Frequency of LMO4 Gene (20782 Position).

Genotype	**No.**	**%**	**Allele frequency**	
GG	65	54.17	G	A
CA	39	30.0	0.704	0.296
AA	16	15.83		
Chi-square	54.4667**			

****(P≤0.01)**

The LMO4 genotype had a substantial ($P \leq 0.05$) impact on the overall milk output, according to the results (Fig.1). Ewes with the wild genotype (GG) had the highest milk yield, at 80.13 kg, while ewes with the hetero genotype (GA) had the lowest, at 71.9 kg.

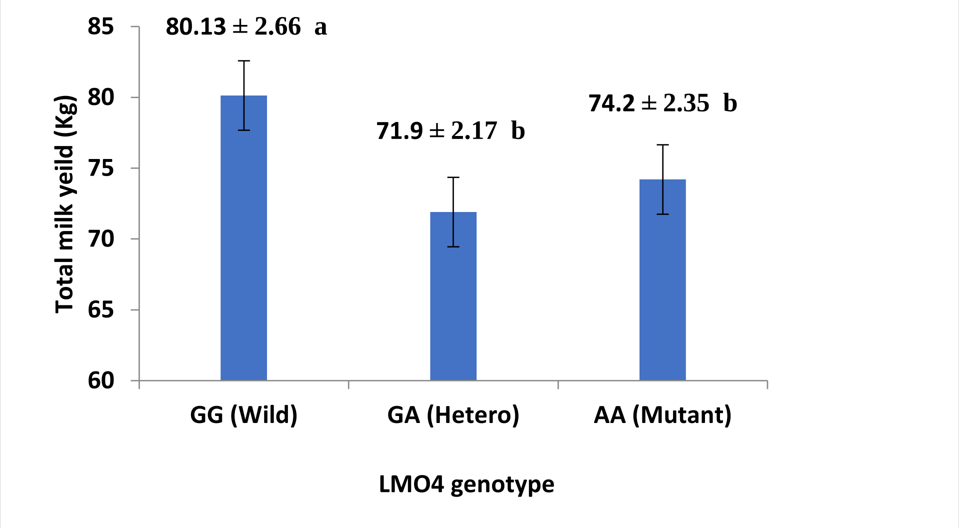

Fig. 1 : Effect of LMO4 Genotype on Total Milk Yield.

The findings indicated that the LMO4 genotype had a substantial ($P < 0.05$) impact on lactation time (Fig.2). Ewes with the wild genotype (GG) had the longest milk season or lactation period, measuring 12.10 days, whereas ewes with the mutant genotype (AA) had the shortest lactation period, measuring 113.70 days.

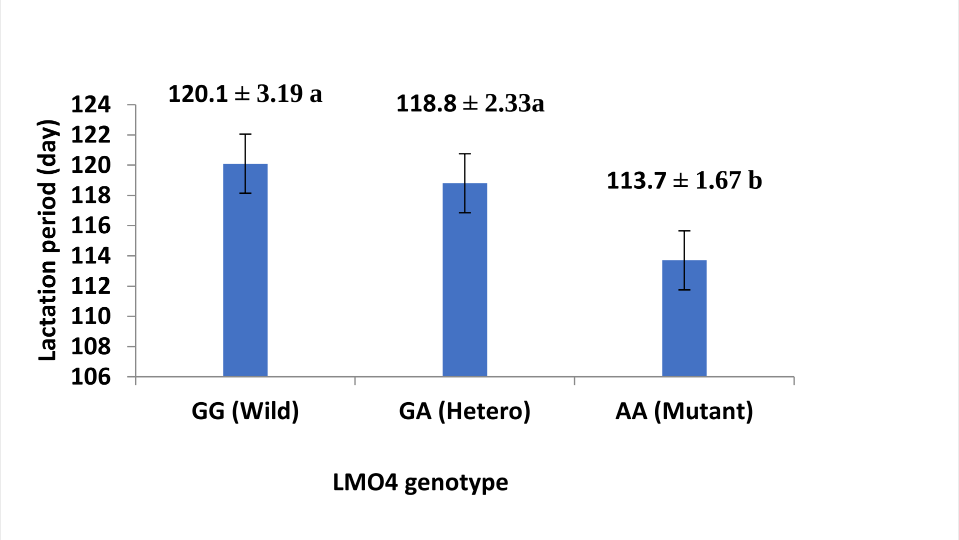

Fig. 2 : Effect of LMO4 Genotype on Lactation Period.

The present study's findings were consistent with other earlier investigations that suggested this gene or related genes had a major impact on sheep performance in numerous regions throughout the globe. According to [10,11,12,13], genetic variables impact milk yield or composition to varying degrees. A few genes that affect sheep milk performance will be examined for their important roles, including those in the gene family.

Numerous investigations have demonstrated that the LMO4 gene is essential for the development of the body in many mammalian species [14], and its activity is linked to numerous genes that have combined effects on a variety of physiological pathways within the body and have a positive or negative reflex in metabolism and development. In order to regulate the antiapoptotic signaling in cisplatin ototoxicity, STAT3, a major downstream target of LMO4, is essential. Additionally, LMO4 connects with several transcription factors and co-regulators that promote the transcription of anti-apoptotic genes and serves as a scaffold for protein complexes. [15,16] [17].

In conclusion, the LMO4 gene is a significant gene that has a wide range of effects on sheep's body growth and dairy production. We can use this gene in selection programs to select for good individuals with stable genotypes and use indirect selection to improve sheep performance.

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