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Article

Relationship of the CYP17 gene (T2231A) to milk production and its components in goats

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ABSTRACT

The experiment was conducted at the Ruminant Research Station of the General Authority for Agricultural Research / Ministry of Agriculture on a sample of 53 goats of two breeds (26 Shami goats and 27 local goats), as well as the Scientific Progress Laboratory specialized in biotechnology and genetic material analysis. In order to determine the genotypes and allelic repeats of the CYP17 gene in goats to determine its relationship to milk production and its components, after the completion of the PCR polymerase chain reaction, the results were sent to the Humanizing Genomics Macrogen Company in South Korea to detect the genotypes of the studied area using the sequencing technology, Two genotypes TT and TA were obtained at the site (T2231A), and the percentage of TT genotype was 26.92% for Shami goats. The percentage of TA genotype was 73.08, and the relationship between them was significant (P≥0.05). The allelic frequency of the T allele was And A 0.63 and 0.37, respectively, where the results showed a significant relationship (P≥0.05) between the genotypes and the amount of total milk production and the percentage of solids that are not fatty. The animals carrying the TT genotype recorded the highest amount of production than those carrying the TA genotype, 571.08 ± 48.69 and 432.24 ± 50.56, respectively. Also, the animals carrying the TT genotype outperformed the animals carrying the TA genotype in the percentage of solid non-fat components, which scored 12.79 ± 1.66, and they scored 9.89 ± 1.63. The percentage of TT genotype for domestic goats was 40.74%, and the percentage of genotype TA was 59.26. the relationship between them was significant (P≥0.05), and the allelic frequency of the T and A alleles was 0.70 and 0.30, respectively. The results also showed a significant relationship (P≥0.05) between the genotypes and the amount of total milk production of the local goats, where the animals carrying the TT genotype recorded the highest amount of production than the animals carrying the TA genotype, and it scored 548.15 ± 72.06 and 350.10 ± 47.94, respectively. Thus, the CYP17 gene can be used in genetic improvement programs.

Keywords: Goats; CYP17 Gene; Genotype; Milk Production.

INTRODUCTION

Goats are among the first animals that humans domesticated more than 10,500 years ago. [1]. Goats are among the most adapted animals to tropical and desert environments. The interest in improving the production of farm animals, including goats, is one of the important aspects to increase the economic return, where animal breeders follow improvement programs that increase the animal's productive capacity by increasing the frequency of distinct alleles in herds and selection for them (The interest in raising and improving goats has recently begun in order to benefit from its meat as well as the milk it produces [2] Goat milk is an important source of nutrients such as protein, fats, sugars and vitamins. The development of molecular biology during the last three decades has opened the methods to study the genes of farm animals and improve them genetically, as selection on the basis of genetic makeup has become an important tool in the process of genetic improvement of farm animals, and for the economic importance of ruminants studies focused on finding ways to improve genotypes through attention In molecular genetics as well as interest in quantitative genetics, selection strategies and programs [3] However, with the increase in population growth and the increase in the demand for animal products such as milk and meat, and with the advancement of modern scientific methods and the availability of information on the work of the genome, it has helped in developing more accurate, less time and costly selection programs As the productive traits are controlled by a number of genetic loci and they are known as quantitative trait sites, and in light of identifying these sites and identifying the associated markers, it is possible to predict the phenotypic variance of the traits to be improved at an early date and to develop plans for selection programs on the basis of these markers, which may be functional mutations, in effector [4]. Sequencing technology has recently been used for genes or parts of a gene. This technology determines the genetic structure of each animal and detects the presence of mutations and their effect on different traits. Among the polymorphic genes, CYP17 is unique because of its ability to catalyze two different types of α17-hydroxylase and 17.20-lyase reactions at one site. Moreover, the ratio of these interactions is physiologically important and may direct the biosynthesis of hormone Steroids towards the production of corticoids. Thus, CYP17 is a very promising target in inhibiting androgen biosynthesis. This gene spans more than 10 kb and is typical of the genomes of all chordates [5,6,7].

MATERIALS AND METHODS

Draw Blood Samples

Blood samples were drawn from the jugular veins of 53 goats using a 10ml medical syringe. The primer was designed for the studied CYP17 gene segment. The CYP17 gene in goats is located on chromosome 26 and consists of seven exons and six introns and has a size of 804 kbp in goats. One region consisting of exon 2, intron 2 and exon 3 has been identified.

"Front starter" TGTAAAACGACGGCCAGTAAGCAGGGAGCTCTACAA

"Rear starter" CAGGAAACAGCTATGACGGAGTGAACTGTAAGAG-GAAAG.

Milk samples were taken every two weeks from each animal, and these samples were analyzed to find out the percentages of milk components (protein, fat, lactose, non-fat solids) using a device (Milk Analazyer milkoscope Julie z7), the total milk production was also calculated from the following equation:

$$\text{Total milk production (kg)} = \text{daily milk production rate x number of days of milking} \quad (1)$$

Statistical Analysis

The data were statistically analyzed using the program Statistical Analysis System–SAS (2012) to study the effect of the genetic phenotypes of the CYP17 gene on the quantity of milk production and components of Shami goats. The significant differences between the means were compared using the [6] polynomial test using the Least square means method. Mathematical model: the relationship of genetic manifestations of the CYP17 gene to the studied traits:

$$Yijkm = \mu + Gi + Aj + Sk + Tl + eijklm \quad (2)$$

Allelic frequency was calculated using the Falconer and Mackay (1996) equation.

$$PA = \frac{2 * \text{No. of Homozygous} + 1 * \text{No. of Heterozygous}}{2 * \text{Total number of samples}} \quad (3)$$

Repeat of the first allele: PA

Since: P + q = 1 then the frequency of the second allele is: qB = 1 - PA

The Chi-square-$\chi 2$ test was also used to compare the percentages of the genotype distribution for each SNP in the CYP17 gene for the studied sample and for both Shami and local goats.

$$X2 = \sum \frac{(Oi - Ei)2}{Ei} \quad (4)$$

RESULTS

The DNA was extracted, and the studied CYP17 gene was duplicated in the coding regions (Exon2, Intron2, Exon3) using PCR technology. The DNA concentration was estimated using a Quantus Fluorometer to ensure the success of the extraction process, and the DNA concentration was between 20-32 ng/µl, where the required CYP17 gene segment with a size of pb 804 was obtained as shown in the figure (1).

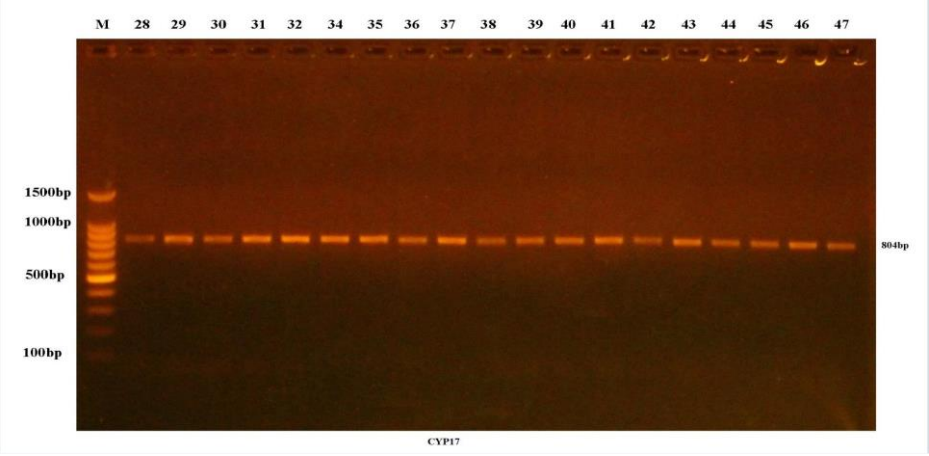

Figure 1. The fragment was extracted from the CYP17 gene (pb 804) by PCR technology.

Changing histidine thymine to adenine at site 2231 of the CYP17 sequence was secreted according to the cDNA strand sequence according to the online vertebrate genome browser Ensempl, and two genotypes were obtained, as shown in figure (2).

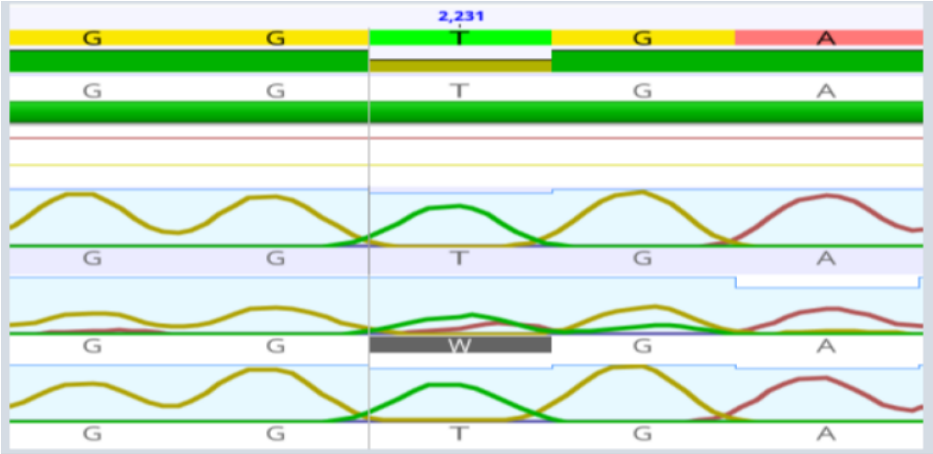

Figure 2. The result of the sequencing technology for the first change in the studied segment of a gene.

In Table (1), the TA genotype showed a clear superiority over the TT genotype, which amounted to 73.08 and 26.92, respectively, with the absence of the AA mutated genotype. We also note that the T allele, whose frequency was 0.63, was superior to the allele A, whose frequency was 0.37, in the sample of the studied Shami goats. The Table results for the local goat sample studied showed the superiority of the TA genotype over the TT genotype. The percentage of the genotype for TA and TT was 59.26 and 40.74, respectively, with the absence of the AA mutated genotype, and the frequency of the T and A alleles was 0.70 and 0.30, respectively, where we notice that the T allele is superior to A.[8,9]

Strain	(Genotype)	Numbers	Percentage (%)	Allele	Repetition
Al-Shami	TT	7	26.92	T	0.63
	TA	19	73.08		
	AA	0	0.00	A	0.37
	Average	26	100%		
	Chi-square value ($\chi 2$)	--	** 9.278	--	--
Local	TT	11	40.74	T	0.70
	TA	16	59.26		
	AA	0	0.00	A	0.30
	Total	27	100%		
	Chi-square value ($\chi 2$)	--	** 9.865	--	--
****P<0.01**					

Table 1. Number and percentages of genotypes and allelic repeats in the CYP17 gene (SNP3 2231) in a sample of Shami and local goats

The results in Table (2) showed that there were significant differences ($P \leq 0.05$) in the total milk production rate and the percentage of non-fat solids between the TT and TA genotypes for Shami goats. The total milk yield was 571.08 ± 48.69 and 432.24 ± 50.56 kg, and the percentage of non-fat solids was 12.79 ± 1.66 and 9.89 ± 1.63% for both genotypes TT and TA, respectively. While there were no significant differences in the other milk components, the percentage of fat was 3.11 ± 0.31 and 4.46 ± 0.66%, the percentage of protein was 3.03 ± 0.14, 3.02 ± 0.10%, and the percentage of lactose was 4.22 ± 0.25 and 4.33 ± 0.09% for both genotypes.

TT and TA for Shami goats, respectively. While there were significant differences * (P≤0.05) in the rate of milk production 548.15 ± 72.06 and 350.10 ± 47.94 liters for both genotypes TT and TA, respectively, for local goats, It was noted that there were no significant differences in the other milk components, where the percentage of fat was 2.58 ± 0.46, 2.01 ± 0.40%, the percentage of protein was 3.07 ± 0.02, 2.87 ± 0.09%, the percentage of lactose was 4.57 ± 0.04, 4.75 ± 1.05%, and the percentage of non-fat solids was 8.35 ±0.06 and 9.85 ±1.74% for both TT and TA genotypes, respectively.

Strain	(Genotype)	mean ± standard error				
		Total milk production rate/kg	%fat	Protein%	%lactose	%non-fat solids
Al-Shami	TT	±48.69 571.08	3.11 ±0.31	3.03 ±0.14	4.22 ±0.25	±1.66 12.79
	TA	432.24±50.56	4.46 ±0.66	3.02 ±0.10	4.33 ±0.09	±1.63 9.89
	significant level	*	NS	NS	NS	*
	(P≤0.05), NS: not significant.					
Local	TT	±72.06 548.15	2.58 ±0.46	3.07 ±0.02	4.57 ±0.04	±0.06 8.35
	TA	350.10±47.94	2.01 ±0.40	2.87 ±0.09	4.75 ±1.05	±1.74 9.85
	significant level	*	NS	NS	NS	NS
	(P≤0.05), NS: not significant.					

Table 2. Relationship of the CYP17 gene (SNP: 2231) genotypes with total milk production and milk composition for Shami and local goats.

DISCUSSION

The CYP17 gene is an important gene in livestock genetic improvement programs, as this enzyme encodes a subgroup of enzymes belonging to the major cytochrome p450 family. Its expression is restricted to the endoplasmic reticulum and plays an important role in steroid hormone synthesis reactions. The genetic variation detected in the studied region (T2231A) clearly affects the total milk production and the proportion of non-fat solids in goats. Due to the small size of the sample available during our study, there was no clear effect of the gene on the other milk components for the percentage of fat, protein and lactose, so it is recommended to analyze a larger number of goat samples.

CONCLUSIONS

The study concludes the relationship of CYP17 gene polymorphism to the productive performance of milk and its components in goats.

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