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ESTIMATION OF GENETIC VARIATION AND BREEDING VALUE AND THE EFFECT OF ALLELE SUBSTITUTION OF GROWTH TRAIT IN IRAQI AWASSI SHEEP DEPENDING ON GROWTH HORMONE GENE POLYMORPHISM

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ABSTRACT

This study was conducted on 27 awassi ewes for detecting the breeding value, genetic variation components and the effect of substitute allele for weight trait at birth and weaning age to their offspring depending on the genetic information for growth hormone gene (second coding region - exon2 A718G) using PCR-RFLP (HaeIII) technique, there was three genotypes (AA, Aa, aa) with genetic distribution 0.44, 0.49 and 0.07 respectively. The AA genotype superior to other genotypes in breeding value and dominant deviation also the dominant variance superior to additive variance in weight trait. the average allele substitute and average effect of allele of A allele was higher than a allele in weaning weight. This result shows the priority of A allele in selection program of weaning weight for the trait of weight at weaning.

Keywords: Awassi sheep, gene, growth hormone, genetic variation, breeding value, allele substitution.

Introduction

The growth hormone gene located on the Third chromosome in the sheep It's about (1.8 kb) (Hajihosseini *et al.*, 2013) it encodes for growth hormone which play an important role in body growth and development of immunity system and its component's (Cobra *et al.*, 2013) The nucleotide sequence of a growth hormone gene is similar to that found in small ruminants and is less similar in cows (Cobra *et al.*, 2013) a lot of searchers used the development of biotechnology for studying genetic variation of this gene to the prediction for the future production or selection of the individual which will be parents for the next generation after knowing the individual's different In a DNA level (Sodhi, 2014; Tamer *et al.*, 2016; Abd AL-Muhsen *et al.*, 2019), in Iraqi (Mahdi *et al.*, 2018) studied the growth hormone gene in three Iraq sheep's breeds to detecting the amount of the Genotype and the alleles for tow genetic region of the growth hormone gene, estimating the breeding value is one of the best ways to evaluate the animal for selection the best depending on the genetic information and detecting the component of variance and the interaction between alleles which depend on knowing additive effect of genes information and the effect of dominant interaction between allelic gene and the epistasis effect between the not allelic gene (Falconer and Mackay, 1996) a lot of studies that depend on the genetic information in selection programs doesn't depend only on the additive effect value and but also

interest with dominate effect which represent the interaction between the alleles and the Interaction with the environment also (Braubar *et al.*, 2012).

This study aims to estimate the genetic variation, breeding value and the effect of allele substitution of growth hormone gene for birth and weaning weight traits in sheep.

Materials and Methods

This study was conducted on 27 lamps (single birth) belong to Awassi ewes (2-5 ages) in a sheep breeding station in Bagdad governorate for the five months/2019. The birth weight was taken at the birth and the weaning was taken after 15 week.

Blood samples from each ewe were collected at the same time from jugular vein after giving birth, the blood samples was reduced in a tube containing anticoagulant (0.5 ml of EDTA) transferred by a cool box then stored in freezing at -20C temperature till transferred to the lab to extracting DNA (in molecular bio and medical technology department/ AL-Nahrain university), the polymorphism of the studied region (422bp) of growth hormone gene (exon2/A781G) was conducted by using PCR-RFLP with HaeIII restriction enzyme.

The primers (f-TCTGCCTGCCCTGGACT, R-GGAGAAGCAGAAGGCAAC) and PCR procedure (table1) was down according to Hue *et al.*, 2009.

Table 1 : the amplification program of studied region of growth hormone gene (exon2 -422bp).

	Steps	Temperature C°	Time	Number of Cycles
1	start Denaturation	94	5 min.	cycles 1
2	Denaturation	95	30 sec	13 cycles and decrease the temperature 1C° per each cycle
	Annealing	65	30 sec.	
	Extension	72	45 sec.	
3	Denaturation	95	30 sec.	cycles 35
	Annealing	52	30 sec.	
	Extension	72	45 sec.	
4	end Extension	72	7 min.	
5	Finish	4	Unlimited	

The equations for each calculated value were applied

As follows ((Falconer & Mackay ,1996)):

1. The average effect of allele A = $q[a+d(q-p)]=\alpha A$
2. The average effect of allele a = $-p[a+d(q-p)]=\alpha a$
3. The effect of substitution of alleles = $\alpha A - \alpha a$
4. The breeding values: $AA=2 \alpha 1$, $Aa= \alpha 1+\alpha 2$, $aa=2\alpha 2$
5. The dominant deviations: $AA=2 q^2 d$, $Aa= 2pqd$, $aa=2 p^2 d$

Result and Dissection

According to PCR-RFLP there was three genotypes in the studied region of growth hormone gene the AA(422bp), Aa (422bp, 366bp, 56bp) and aa (366,56) as a dominant homo, hetro and recessive genotypes respectively.

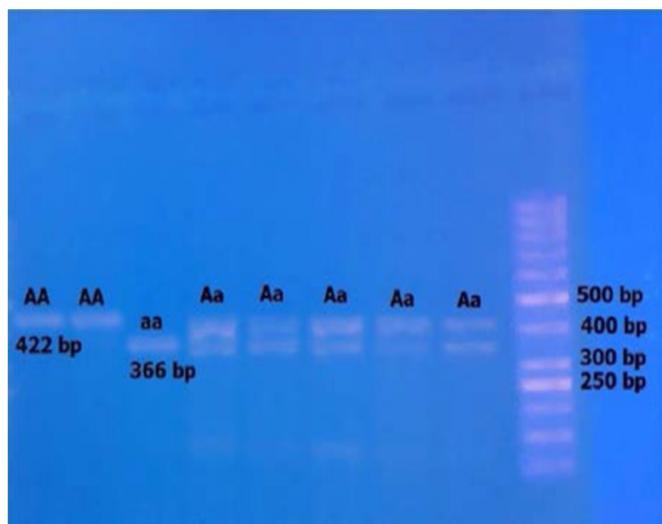


Fig. 1: Enzymatic digestion of PCR products of the Awassi pregnant ewes, 422 bp & 366 bp fragments were appeared, the fragments of 56 bp was not appeared.

A allele surpassed a allele in allele frequency (0.615 and 0.315 respectively) table (2), the distribution of AA and Aa genotypes was close and they surpassed on the a genotype (table 2).

Table 2 : The number, genotype distribution and allele's frequency of growth hormone gene.

Alleles frequency	Alleles	Genotypes distribution	n	Genotype
0.615	A	0.44	12	AA
		0.49	13	Aa
0.315	a	0.07	2	aa

As shown in table (3) that aa genotype has the superiority on the other genotypes (Aa and AA), significantly on Aa genotype and statistically on AA so there is a priority of a allele. The sequence of the breeding value of genotypes was corresponding with weight means. The aa genotype came first and the other genotypes (AA, Aa) respectively (Table 3), this result gives preference to the aa genotype for selection of birth weight because of the appositive correlation between birth and weaning weight (Everett-Hincks *et al.*, 2014). The dominant divisions were higher than the breeding value corresponding with the fact that trait weight is a quantitative trait that has greatly affected by environment (Notter *et al.*, 2005). Dominant division of the three genotype converged greatly (table 3) for the trait of weaning weight because of the lambs in this age is effected by level the care and the management of the heard that reduce the difference between individuals that lead to reducing the genetic variance but doesn't cancel it (Van De Stroet, 2016). The dominant variation had the higher value compared with the additive variance (Table 3) it's a value from an interaction within alleles in the same locus and its interaction with environment (visscher *et al.*, 2000) this result agrees with the fact that weight is effected with a large number of genes (Al-Anbari *et al.* 2018) and it's the real value that inheritants to off- spring (jicai, 2017).

Table 3: Breeding value, dominance deviation and genetic variations of genotypes.

VG	VD	VA	D.D	BV	Means	Trait	Genotype
0.5645	0.379	0.1855	6.061	5.972	6.25	Birth Wight	AA
			5.438	5.613	4.94		Aa
			6.948	5.254	6.82		aa
VG	VD	VA	D.D	BV	means	Trait	Genotype
10.78	10.15	0.63	21.772	21.133	21.71	Weaning weight	AA
			20.061	21.1952	19.65		Aa
			23.403	21.203	23.50		aa

BV: Breeding value, DD: dominance deviation, VA: Additive variance, VD: Dominance variance, VG: Genetic variance.

A allele was the superior in the average allele effect and the Average effect of allele Substitution in birth weight but the result was inverted for the value of weaning weight (table4) which represent the added or the deleted resulting from selection of one alleles (Falconer,1992) when comparing to value shows the preference of mutata genotype because aim of selection is reaching to the best weaning weight.

Table 4: The average allele effect and the Average effect of allele substitution

Average allele effect	Average effect of allele Substitution	birth weight / allele
0.113	0.36	A
- 0.246	-0.36	a
average allele effect	Average effect of allele Substitution	weaning / weight allele
- 0.0028	- 0.009	A
0.005	0.009	a

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