

## DETECTION OF QUANTITATIVE LOCI CORRELATION WITH GROWTH TRAITS IN LOCAL QUAIL USING PCR- RFLP TECHNIQUE

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### ABSTRACT

The objective of this study was to investigate the polymorphisms of three loci (*SEMA3E*, *GH* and *TLX*) that related with growth traits in local quail. A total of 720 birds (males and females) from three lines (desert, brown and white) were used. The results revealed that the effects of the line were significant on bird body weight and carcass weight and dressing percentage at 180 days at age. The Best Linear Unbiased Prediction (BLUP) value overall birds for body weight was ranged from -9.2173 to 10.0117, these results showed there were significant differences among high and low BLUP value groups in three quail lines under study. The PCR-RFLP results overall three lines showed that there were three, three and two alleles for *SEMA3E*, *TLX* and *GH* locus, respectively. This alleles gives twelve differences genotypes, the desert male quail with ACABAA genotype and desert female quail with AAABAA genotype (high group) for three loci under study give significantly higher body and carcass weight compared with another groups. In conclusion results showed that there are agreements between BLUP values with PCR-RFLP results to select best birds and the selection process with molecular technique can play a major positive and rapid role to improve and increase growth traits in these lines of local quail in Iraqi Kurdistan region.

Key words: BLUP, growth traits, genes, RFLP, alleles.

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اجاد الارتباط الكمي لصفات النمو في السمان المحلي باستخدام تقنية PCR-RFLP

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المستخلص

ان الهدف من الدراسة الحالية هو التحقيق في تعدد المظاهر لثلاثة مواقع (*SEMA3E* و *GH* و *TLX*) المتعلقة بصفات النمو في السمان المحلي. تم استخدام 720 طائر (ذكور وإناث) من ثلاث خطوط محلية (الصحراء والبني والأبيض). أوضحت النتائج أن تأثير الخط معنوي على وزن جسم الطائر ووزن الذبيحة ونسبة التصافي في 180 يوم. تراوحت قيمة أفضل التنبؤ الخطي غير المتحيز (BLUP) لوزن الجسم من -9.2173 إلى 10.0117 ، وأظهرت النتائج هذه الدراسة بأن يوجد اختلافات معنوية بين قيم العالية والمنخفضة لـ BLUP في ثلاث خطوط السمان. اوضحت النتائج الإجمالية لـ PCR-RFLP في خطوط الثلاثة بأعطاء ثلاث ، ثلاث، وإثنان من الأليلات المختلفة لمواقع *SEMA3E* و *TLX* و *GH* على التوالي. اي ان هذه الأليلات تعطي اثني عشر تراكيب وراثية مختلفة، حيث ان ذكور السمان الصحراوي ذو التركيب الوراثي ACABAA وأنثى السمان الصحراوي ذات التركيب الوراثي AAABAA (مجموعة عالية انتاج ) لثلاثة مواقع في هذه الدراسة لها أعلى قيمة معنوية في وزن الجسم و وزن ذبيحة مقارنة بالمجموعات الأخرى. وعلى ضوء هذه النتائج أوضحت بأن هناك التوافق بين قيم BLUP مع نتائج PCR-RFLP لاختيار أفضل الطيور وأن عملية الانتخاب مع استخدام التقنية الجزيئية يمكن أن تلعب دورًا إيجابيًا وسريعًا لتحسين وزيادة صفات النمو في هذه الخطوط السمان المحلية في إقليم كردستان العراق.

الكلمات المفتاحية: BLUP، الصفات النمو، الجين، RFLP، الأليلات.

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## INTRODUCTION

Quail is a model animal species due to its small body size, fast growth rate, early sexual maturity, lower cost of care and high reproductive rate (13). Quails have been used to investigate different studies for several purposes since it is closely related to chickens (6). The quail is a sexually dimorphic bird, females have a large body size and require more time to reach sexual maturity than males (25). Producing high-performance poultry lines is the main goal of breeders and farmers, industrially (15). It has been investigated that genetic factors control that both production and reproductive traits (17). Genetic improvement of economic traits of quail procedures for multiple-trait genetic evaluation of livestock requires accurate estimates of genetic and environmental parameters (3). Best Linear Unbiased Prediction (BLUP) is the current method of choice for genetic evaluation of traits. Traditionally, the selection of animals for breeding is based on two types of data, pedigree and phenotypes (2). Therefore, genetic studies of poultry species rely on BLUP in their investigation. The development of molecular biology and particularly DNA base ANDd markers facilitated genetics and animal breeding of livestock studies (11). Molecular genetic markers are important tools to analyze genomes and associate heritable traits with an underlying variation of genomes. In addition, experimental of candidate genes and their effects on the phenotypic traits the basis of marker-associated selection (MAS) (15). The quail has been considered as a model animal in several biological and biomedical studies, the identification of quail genome has been to facilitated various studies and recognize distinct lines of quail (12, 1). The ability of quail to growth and reproductive during certain periods varies greatly due to its diversity among the individuals. It is known that the nature of growth and reproduction are generally influenced by Growth Hormone (GH), steroids and peptides (29). A secreted class 3 semaphorinis encoded *SEMA3E* gene is specific vascular beds to trigger repulsion of endothelial cells, it is also involved to modulate axonal growth and regulate synaptic connectivity for the correct wiring of the

central nervous system (CNS) (8). Moreover, Song *et al.*, (31) demonstrated that T-cell leukemia translocation (*TLX*), also known as Hox11 is considered as an important target in neural development through the progressive regulation of cell cycle in Neural Stem Cells (NSCs). Therefore, the objective of the present study is to identify the genetic polymorphism and best genotypes for quail's growth using PCR-RFLP for three specific genes (*SEMA3E*, *GH* and *TLX*).

## MATERIALS AND METHODS

### Measurement of growth and carcass traits

This study was carried out in Grdarasha researches unit, under the supervision and regulations of ethic committee at college of agricultural engineering sciences, Salahaddin University-Erbil. For this purpose 720 newly hatched quail chicks of three local lines, desert (214), brown (250) and white (256) each line were randomly distributed into ten families; the mating system was in a ratio of one male to three females. The estimated BLUP of %10 (top and bottom) form both male and female in local quails according to high and low body weight. The birds were raised under the same living conditions and feeding. A total 144 quail birds were selected for the study (weight and carcass trait), including 72 quails from the high production (H) and low production (L) line for each sex were isolated according to their differences in feather color and body weight. The quails were individually weighed and slaughtered at 6 months by cutting the jugular vein. Blood from each quail were collected for Genomics analysis according to the procedure of Oeywale,( 22) as described by (33). The birds were then properly bled (about 4 minutes) and feathers removed manually after dipping in hot water for about two minutes. Dressed percentage were calculated according to procedure and formulae of (6).

$$\text{Dressed \% (DP)} = \frac{\text{Carcass yield}}{\text{Live body weight}} \times 100$$

### Sample collection and DNA extraction

Genomic DNA was extracted from the blood of 144 local quails. One mL of blood samples was collected from each bird and then put in a 3mL of anti-coagulant Tris-ethylene di amine tetra acetic acid (EDTA) tube. DNA was then extracted from the blood sample of each bird

using the DNeasy® blood kit (GeNet Bio, Korea) according to the manufacturer's instructions. The quantity and quality of DNA was checked by Nanodrop (1000 UK) spectrophotometer and gel electrophoresis.

### Selection of the loci and genotyping of the samples

The respective loci were selected according to the chromosome map that was previously constructed by (27). The loci and primers designed for this study were shown in Table 1. The primers were designed based on the sequences submitted to the GenBank by (27), using Primer-Blast of NCBI (<http://www.ncbi.nlm.nih.gov>). The total reaction volume of PCR was 25 µL consisting 10 µL of Green Master Mix (25 units/mL Taq DNA polymerase 200 µM of each dNTPs and 1.5 mM of MgCl<sub>2</sub>), 1 µL forward and reverse of primer of each gene, 1 µL of DNA template, and the volume was completed with 12 µL of DNase free water. The thermocycling conditions for gene 1 (CJA1), and 3 (CJA3) included initial denaturation step at 94°C for 5 min, followed by 32 cycles at 94 °C for 1 min, annealing at 60 °C for 1 min, elongation at 72 °C for 1 min and a final extension step at 72 °C for 10 min. For GH gene The thermocycling conditions included an initial denaturation step at 95 °C for 5 min, followed

by 35 cycles of denaturation at 94 °C for 30 s, primer annealing at 56 °C for 45 s, elongation at 72 °C for 1 min, and an extension step at 72 °C for 5 min. The digestion of 10 µL of PCR product was made using the restriction enzyme (27) with some minor modifications and also based on the instructions of the manufacturer (Thermo Scientific) Table 2. Table 3 shows the restriction enzymes and reaction conditions that were used in the current study. Electrophoresis analysis was made for PCR products using 2.5% agarose gel stained with safe day (Cat. No. B-2010, GeNet Bio, Korea). The agarose gel was run at a constant voltage of 100 V/cm for 45 min. The bands were subsequently visualized by UV transilluminator and the gel photographed (Proxima 2500 Isogene Life science, Netherland). In this study a total of 6 PCR amplicons (2 for each genes) were sent to the commercial company for purified and sequencing (Macrogen Inc. South Korea). The sequences similarity analysis with previous sequences published in GenBank were performed using the BLAST programme (<http://www.ncbi.nlm.nih.gov/BLAST>). BLAST analysis approved that all sequences were SEMA3E, TLX and GH gene with 99-100 identities with previous published this genes

**Table 1. Name, Chromosomal location, and Sequence of Primers used in this Study**

Locus	Chromosomal Location (cM)	Accession Number	Primer Sequence (5'-3')
SEMA3E	CJA1 (70.5)	MN542411	Forward-ATACTCCAGCTGAGTGGGGA Reverse-CAGAAGTATGAGGGAGATCAG
TLX	CJA3 (165.4)	MN542412	Forward-ACACTAGGAACATAATGGGCT Reverse-TCACTGTGGCGTTTCAGATT
GH	CJA5 (82.3)	MN542413	Forward-ATCCCCAGGCAAACATCCTCG Reverse-CCTCGACATCCAGCTCACAT

**Table 2. Name and Reaction conditions of Restriction Enzyme used for each Locus**

Locus	Restriction enzyme	Amount(Unit)	Incubation(°C/h)	Buffer
SEMA3E	<i>Hae III</i>	5	37/3	R
TLX	<i>Pst I</i>	5	37/3	O
GH	<i>Msp I</i>	5	37/3	Tango

**Table 3. PCR Product Digestion component for all studied Genes**

Digestion component	Volume
PCR product	10µl
Reaction 10X buffer	2 µl
Nuclease-free water	17 µl
reaction enzyme	1 µl
Final volume	30µl

### Statistical analysis

**Performance (field) data:** To analyze the data for quail's body weight, carcass weight and dressing %, the PROC GLM (General Linear

Model) procedure SAS, (28) was utilized. Fixed effects study was using the following model:

$$Y_{ijkl} = \mu + L_i + S_j + LS_{ij} + \varepsilon_{ijkl}$$

Where:  $Y_{ijkl}$  = body weight, carcass weight and dressing % 1<sup>th</sup> quail, of  $i^{th}$  line ( $L_i$ ,  $i=1$ , brown,  $i=2$ , desert and  $i=3$ , white), of  $j^{th}$  Sex ( $S_j$ ,  $j=1$ , male and  $j=2$ , female), of  $k^{th}$  interaction between line and sex ( $K_{ij}$ ,  $ij=1$ , Desert male, 2= Desert females, 3= Brown male, 4= Brown female, 5= White male and 6= White female),  $\mu$  = Population mean;

$\varepsilon_{ijkl}$  = random error. It was assumed to be independently and normally distributed with

mean zero and variance  $\delta^2 e$ . For genetics evaluation of quail (High and low production) for various performance traits, Best Linear Unbiased Prediction (BLUP) procedure described by (28) was applied. The model used for this purpose was the Mixed Model (Fixed + Random effects) of (28) software. The three quail lines were assembled in three groups; high (10%), medium (80%) and low (10%) production, according to the BLUP values

#### Molecular analysis

Genotypes of polymorphic loci were determined by direct counting of the bands. The gene frequencies for each locus in each sample were calculated using the following equations:

$$p = \frac{2(AA)+AB}{2N} \quad q = \frac{2(BB)+AB}{2N}$$

where  $p$  = the gene frequency of allele A,  $q$  = the gene frequency of allele B and  $N$  = the total number of birds tested and tested to Hardy-Weinberg ratios using was calculated using GENPOP software version, 3.3 (24). The genotypes effects for body weights, carcass weight and dressing% were fitted to following equations:

$$Y_{ijklo} = \mu + A_i + S_j + C_k + P_l + \varepsilon_{ijklo}$$

Where:  $Y_{ijklo}$  = Body weights, carcass weight and dressing% of  $o^{th}$  bird, of  $i^{th}$  GH ( $A_i$ ,  $i=1$ , AC,  $i=2$ , AB and  $i=3$ , CC), of  $j^{th}$  SEMA3E ( $S_j$ ,  $j=1$ , AB,  $j=2$ , BC and  $j=3$ , CC), of  $k^{th}$  TLX ( $C_k$ ,  $k=1$ , AA,  $k=2$ , AB, and  $k=3$ , AC), of  $l^{th}$  all genes combinations ( $P_l$ ,  $l=1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, \text{ and } 12$ ),  $\mu$  =

Population mean,  $\varepsilon_{ijklo}$  = random error. It was assumed to be normally and

independently distributed with mean zero and variance  $\delta^2 e$ .

## RESULTS AND DISCUSSION

### Phenotypic evaluation

**Body weight:** Data presented in Table 4, shows that lines had a significant ( $p \leq 0.01$ ) effects on body weight were ( $241.30 \pm 9.43$ ), ( $232.65 \pm 7.72$ ) and ( $225.70 \pm 8.02$ ) for desert, brown and white respectively. On the other hand, the statistical analysis for this trait revealed that the differences between males and females were highly significant ( $p \leq 0.01$ ) on body weight ( $205.77 \pm 5.18$ ) and ( $260.67 \pm 4.12$ ) respectively. While regarding the effect of interaction (sex  $\times$  line); the findings in Tables 4 showed the significant effects of interaction (sex  $\times$  line) on body weight and recorded higher interaction between female and lines. Our results were in agreement with a recent study that observed by Al-Kafajy et al., (5) who stated that the desert line had higher significant values of body weight compared with the black and white lines. While sex of the chicks had significant effect on body weight, the higher weights in females could be due to higher weight of reproductive organs such as ovaries and oviducts, could be due to higher weight in females than males (9). Similar findings were also made by (23) who showed the higher significant effect of Sex on body weight ( $P < 0.01$ ) in two strains of Japanese quail.

### Carcass traits

Carcass traits like any other quantitative traits are largely affected by the interaction between genetic and environmental factors (30). Japanese quail (carcass weight/live body weight) was ranged from 60 to 70 - 75 % depended on slaughter age, line and sex (6). The results of Carcass weight and Dressing percentage of three different line plumage colors are presented in Table 4. In the current study significant differences in Carcass weight among lines were showed. That could be due to genetic variance among studied lines. Nonetheless, no significant differences were observed in dressing percentage. The overall least squares means for Carcass weight and Dressing percentage at 6th month of age were ( $166.60 \pm 5.26$ ,  $161.70 \pm 4.46$  and  $156.90 \pm$

4.09) g and (70.04± 1.34, 70.02± 1.35 and 70.14± 1.34) % respectively in desert, brown and white lines. While the sex effect for this trait was significantly higher ( $p \leq 0.01$ ) between male and female. The female (168.63±3.28) was higher than male (154.87±3.90) in Carcass weight but in Dressing percentage trait male (75.55±0.36) was higher than female (64.58±0.37). The effect of interaction (Line× Sex) on Carcass weight and Dressing percentage was significantly ( $p \leq 0.01$ ). Nasr *et al* (21) was reported that slaughter and carcass characteristics were significantly affected by

quail sex. In addition Lotfi *et al.*, (16) was found that Carcass weight was higher in females than males, and also suggested that there are a correlation in between reproductive organs and body weight, therefore the mature female quail weight was higher than in broiler chickens at 42 days of age and consequently, the carcass yield will be lower in Japanese quail. Also, Sabow, (26) indicated significant differences among Japanese quails with different feather colors on slaughter weight and carcass yields at 10 weeks of age.

**Table 4. Least Squares Means and Standard Error for different Traits**

Traits	N.	Body weight(g)	Carcass weight(g)	Dressing percentage (%)
Overall	720	233.22± 7.72	161.75±4.46	70.06±1.35
Lines		**	**	NS
Desert	214	241.30± 9.43a	166.60± 5.26a	70.04± 1.34a
Brown	250	232.65±7.72ab	161.70± 4.46ab	70.02± 1.35a
White	256	225.70± 8.02b	156.90± 4.09b	70.14± 1.34a
Sex		**	**	**
Male	356	205.77±5.18b	154.87±3.90b	75.55±0.36b
Female	363	260.67±4.12a	168.63±3.28a	64.58±0.37a
Line* Sex		**	**	**
Desert Male	103	210.60±10.43b	157.10±7.70ab	75.43±0.86a
Desert Female	111	272.00±7.59a	176.10±6.11a	64.65±0.57b
Brown Male	127	208.10±9.15b	157.00±7.06ab	75.44±0.67a
Brown Female	123	257.20±5.82a	166.40±5.43ab	64.60±0.84b
White Male	131	198.60±7.61b	150.40±5.84b	75.79±0.29a
White Female	125	252.80±7.09a	163.40±5.22ab	64.50±0.55b

a,b,c Column means within parameter with common superscripts do not differ (\*\*  $P < 0.01$ ), NS - not significant.

#### Genetic merit for single trait (BLUP):

===== The estimated Best Linear Unbiased Prediction (BLUP) of local quail for the weight. BLUP values for quail's weight ranged from (0.6527 to 10.0117g, 1.4467 to 8.9887g and 4.2467 to 7.0667g) males and (-8.6613 to -1.6933g, -9.2173 to -2.4293g and -4.3113 to 0.3827) females of desert, brown and white, respectively at six month. These results indicated that there are significant differences of weight trait in between the quails. It means that selection play a crucial role in enhancing weight trait.

#### Molecular characterization of local quail:

#### Allele and genotype frequency in local quail

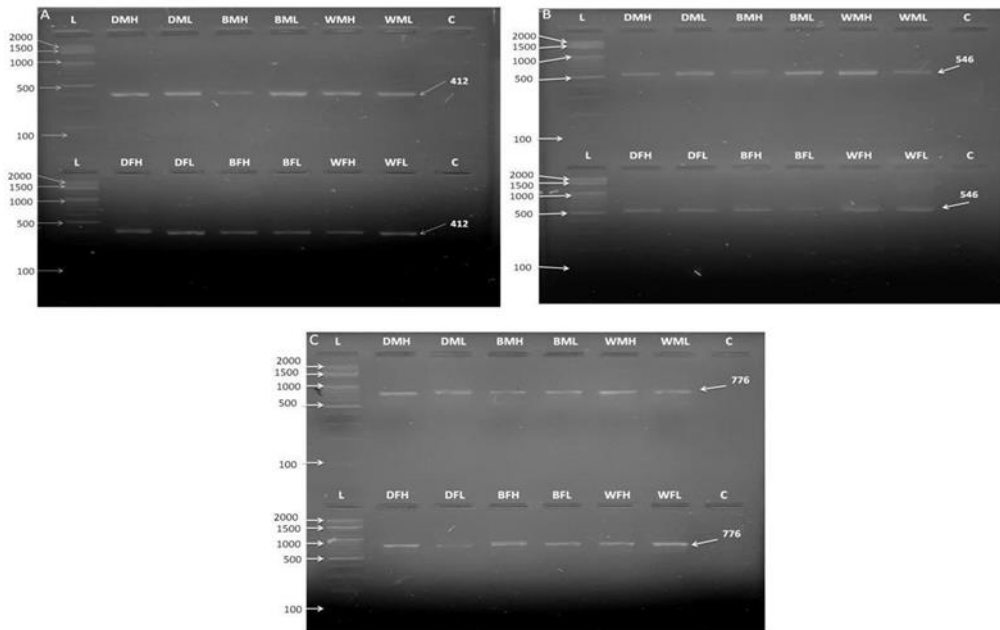
**populations:** The respective loci were successfully amplified using stated primer pairs. PCR product sizes were differed from 412 to 776 bps. After cutting with the appropriate enzymes, one to four different fragments for each locus were observed Table 5. Figure 1A, B and C shows restriction products of particular samples for each locus. Polymorphism was observed for each primer (*SEMA3E*, *GH* and *TLX*) loci. One sequences for each genes obtained were deposited in the GenBank under accession numbers (MN542411, MN542412 and MN542413) for *SEMA3E*, *TLX* and *GH* respectively.

**Table 5. Fragment and Restriction products size for all Genes**

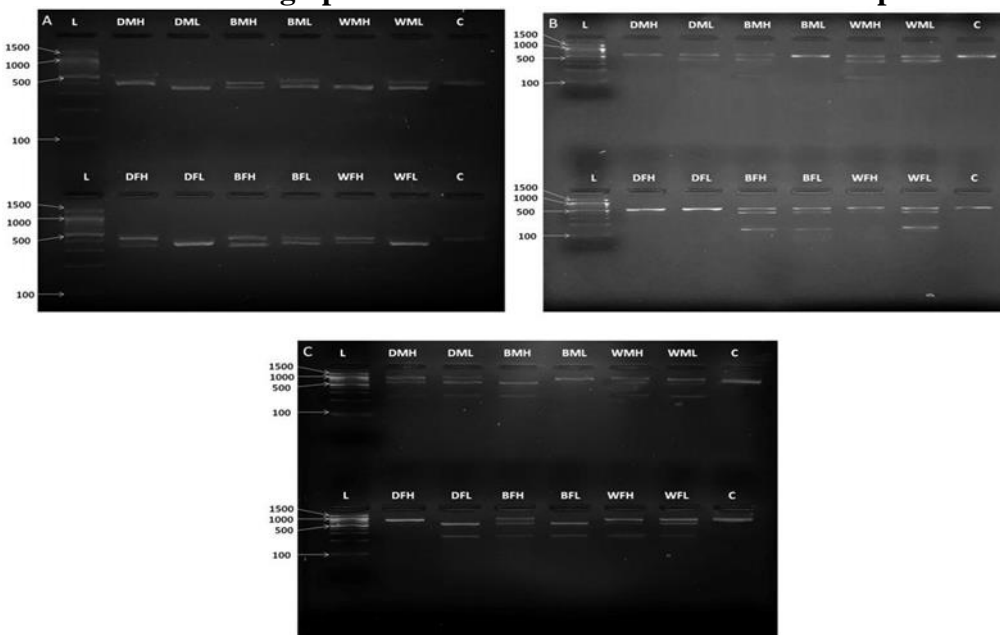
Locus	Size of PCR Product (bp)	Enzyme	Cut	Lengths of Restriction Fragments (bp)
SEMA3E	412	Hae III	+	412/ 362+50/ 335+77
TLX	546	PstI	+	546/ 404+142
GH	776	Msp 1	+	776/ 529+241

Genotype and number of bands for each loci was shown in Table 6. For the *SEMA3E* locus three different alleles (A, B and C), three genotypes (AB, BC and CC) with an approximate size of 412 *bps* as dominant PCR product were observed in Figure 2A, while three different alleles such as A, B and C and also three genotypes namely AA, AB and AC were found for the *TLX* locus, a single amplification product of approximately 546 *bp* was obtained. Figure 2B. Results obtained from the digestion showed the presence of polymorphism in *GH* gene fragment at 776 *bp*-sized quail research was to produce two types of alleles (A and C) with three genotypes (AA, AC and CC) Figure 2C, The allele frequencies of *SEMA3E*, *GH* and *TLX* loci for males and females in three local lines ranged from A: 0.083-0.750, B: 0.000-0.333 and C: 0.208-0.583 respectively. The allele frequency for A; males and females in *TLX* loci had the highest frequencies (0.667 and 0.750) and the least was of *SEMA3E* loci (0.000 and 0.083). For allele B; males and females (0.333) had the highest in *SEMA3E* loci and the least frequencies in *TLX* was (0.000-0.167). Frequency of C; 0.583 and 0.500 of males and females was recorded in *SEMA3E* and *GH* loci respectively and the least was 0.167-0.250 in *TLX* Loci. The genotypic frequencies of males and females were higher for AA in *TLX* (0.562 and 0.445), AC (0.50 and 0.486) *GH* and BC (0.523) *SEMA3E* genes compared to the (AB and CC) genotypes. While, genotypic frequency in *TLX* was higher AA (0.562 and 0.445) in males and females compared to AB and AC genotypes as given in Table 7. These results agree with Bozkaya *et al.* (7) that detected the possibility of using each *SEMA3E*

and *TLX* loci to study recombination frequencies in the Japanese quails populations out of the eight loci studied (*SEMA3E*, *IFR1*, *HAL*, *LOC396025*, *UGP2*, *LOC396192*, *TLX* and *BMP5*), polymorphism was detected in the *SEMA3E* and *TLX* loci; five of their loci were regarded as monomorphic and one locus (*HAL*) wasn't amplifiable by PCR. Deef *et al.* (10) indicated that the PCR-RFLP was performed in terms of revealing the genetic characterization and also genetic relationship of the five species of quails. The Common quails are found to be *Coturnix coturnix*, bobwhite quail *Colinus virginianus*, and three quails belong to *Coturnix japonica* including panda quail, Japanese quail, dotted white quail. Highly polymorphic restriction profiles were recorded from the analysis of fragments that were generated by digestion of PCR products with the restriction enzyme *NlaIII*. A wide variability in intra specific *SEMA3E* and *TLX* genes was obtained among the respective quails. Previous studies on allele frequencies have been used the same method as in the current study, their results have varied among populations from 0.000 to 1.000 (19). Allele and genotype frequencies were observed in the analyzed samples are shown in Table 5. To our knowledge, there is no previous study examining the polymorphism of the respective loci in other populations of Japanese quails. Despite Sasazaki *et al.*, (27) mapped out the loci on the *CJA1* and *CJA3*, no data on allele frequencies or heterozygosity were reported by those researchers. Therefore, results from the current study were extensively compared with those that have been reported on other loci or species and *GH*.



**Figure 1. Polymerase chain reaction based restriction fragment length polymorphism profiles of A) SEMA3E; B) TLX and C) GH of pulled samples in three local lines. L: DNA marker, DMH: desert male high production, DML: desert male low production, BMH: brown male high production, BML: brown male low production, WMH: white male high production, WFL: white male low production, DFH: desert female high production, DFL: desert female low production, BFH: brown female high production, BFL: brown female low production, WFH: white female high production and WFL: white female low production**



**Figure 2. Digestion of PCR products of A) SEMA3E; B) TLX and C) GH of pulled samples in three local lines. L: DNA marker, DMH: desert male high production, DML: desert male low production, BMH: brown male high production, BML: brown male low production, WMH: white male high production, WML: white male low production, DFH: desert female high production, DFL: desert female low production, BFH: brown female high production, BFL: brown female low production, WFH: white female high production and WFL: white female low production**

**Table 6. Band number and Fragments size (Bp) for GH, SEMA3E and TLX Genes in Local Quails**

Populatio n/group	GH		SEMA3E		TLX	
	Genotype and No. of band	band Size bp	Genotype and No. of band	band Size bp	Genotype and No. of band	band Size bp
DMH	AC 3	776+539+237	AB 2	336+412	AA 1	546
DML	AC 3	776+539+237	CC 2	335+77	AB 2	546+404
BMH	CC 2	539+237	BC 2	362+50	AC 3	546+404+142
BML	AA 1	776	BC 2	362+50	AC 1	546
WMH	AB 3	776+539+237	BC 2	335+77	AA 3	546+404+142
WML	AC 3	776+539+237	BC 2	335+77	AB 2	546+404
DFH	AA 1	776	AB 2	336+412	AA 1	546
DFL	CC 2	539+237	CC 2	362+50	AA 1	546
BFH	AC 3	776+539+237	BC 2	362+50	AC 3	546+404+142
BFL	CC 2	539+237	BC 2	362+50	AC 3	546+404+142
WFH	AC 3	776+539+237	BC 2	362+50	AA 1	546
WFL	AC 3	776+539+237	CC 2	335+77	AC 3	546+404+142

**Table 7. Allele and Genotype Frequency of SEMA3E, GH and TLX Genes in male and female of Local Quail**

Locus	Sex	Local quail					
		Allelic frequency			Genotype frequency		
SEMA3E	n=144	A	B	C	AB	CC	BC
	Male	0.083	0.333	0.583	0.113	0.523	0.364
	female	0.083	0.333	0.583	0.113	0.523	0.364
	overall	0.083	0.333	0.583	0.113	0.523	0.364
TLX	n=144	A	B	C	AA	AB	AC
	Male	0.666	0.167	0.167	0.444	0.278	0.278
	female	0.750	0.000	0.25	0.562	0.000	0.438
	overall	0.708	0.083	0.208	0.501	0.143	0.356
GH	n=144	A	C	AA	AC	CC	
	Male	0.500	0.500	0.25	0.500	0.25	
	female	0.417	0.583	0.174	0.486	0.340	
	overall	0.458	0.542	0.210	0.496	0.294	

**Table 8. The mean of high and low level on Body weight, Carcass weight, and Dressing percentage traits in Local Quails as influenced by the different Genotypes**

Population/group	Genotype for all genes	Body weight (g)	Carcass weight(g)	Dressing percentage (%)
DMH	ACABAA	251.00± 2.65c	184.33± 8.99ab	75.84± 1.79a
DML	ACCCAB	173.00± 3.61d	129.67± 2.03d	74.97± 0.62a
BMH	CCBCAC	243.33± 2.73c	182.33± 3.79ab	76.26± 0.44a
BML	AABCAC	176.67± 10.48d	131.67± 7.33d	74.57± 0.72a
WMH	ABBCAA	230.33± 1.45c	174.33± 2.03b	75.90± 0.41a
WML	ACBCAB	173.67± 4.09d	130.33± 2.69d	75.36± 0.76a
DFH	AAABAA	299.33± 10.41a	197.67± 10.14a	65.96± 1.08bc
DFL	CCCCAA	244.67± 1.20c	155.33± 1.45c	63.49± 0.29cd
BFH	ACBCAC	281.00± 6.56ab	192.33± 6.74ab	68.41± 0.91b
BFL	CCBCAC	240.00± 3.51c	150.00± 1.00c	62.53± 1.17d
WFH	ACBCAA	277.67± 6.17b	184.00± 4.51ab	66.26± 0.15bc
WFL	ACCCAC	235.33± 2.52c	151.33± 1.86c	61.749± 0.82d

<sup>a,b,c</sup> Column means within parameter with common superscripts do not differ (\*\* P < 0.01). DMH: desert male high production, DML: desert male low production, BMH: brown male high production, BML: brown male low production, WMH: white male high production, WML: white male low production, DFH: desert female high production, DFL: desert female low production, BFH: brown female high production, BFL: brown female low production, WFH: white female high production and WFL: white female low production.



### Genotypes associations with body weight traits in local quails

The effects of genetic (RFLP) markers on studied economic traits in local quail are illustrated in Table 8. There was a significant association between RFLP patterns and live weight at 6<sup>th</sup> month of age, carcass weight, and dressing percentage. The best genotype for local quail at three loci in the present study was AAABAA in body weight (299.333 g), carcass weight (197.667 g) in the DFH population. While, higher the dressing percentage was (76.264%) was recorded in CCBCAC genotype in the BMH population. This result showed that the selection play a major role to increase body weight, carcass weight and dressing percentage of local quail. Also, the desert quail had the highest in body weight and carcass weight when it compared to brown and white lines and females are highest than males, while in carcass weight the males was higher than females. Furthermore, The ACCCAB genotype had the lowest body weight (173) g and Carcass weight (129.667 g) of and DML population and ACCCAC genotypes had the lowest dressing percentage (61.749%) of the WFL population. The effect of selection for quail of high and low body weight, Carcass weight and dressing percentage revealed the variance in between males and females within and between three lines of local quails. The variation in quantitative trait is controlled by several genetic loci called quantitative trait loci (QTL). While to select animals for selective breeding programmers, genetic markers for QTL that are linked to the trait gene (34). Data collected from PCR-RFLP and the use of *MspI* enzyme observes the effect of *GH* gene polymorphic of quail on weight. Polymorphism of this gene showed different effects on the patterns of body weight in which the average weight of male quail is relatively lower than the female quail (29). the results by Nasirifar *et al.* (20) detected four distinctive alleles (A, B, C, and D) and four different genotypes (AA, AB, AC, and AD) in population of Japanese quails in *GH* marker site, which could be detected by *MspI* restriction enzyme showed a significant ( $P < 0.05$ ) association between RFLP patterns with live weight and carcass weight. Carcass

weight of the AB birds was lower than that of other genotypes. Through employing PCR-RFLP technique researchers have been able to notice a difference in poultry species as chickens, ducks, quail, rabbits, and turkeys (4). while Zhu *et al.*, (37) detected a significant correlation between *GH* gene and quality of meat in Anka and Rugao hens Furthermore, Zhang *et al.* (36) observed a positive link between intron 1 of *GH* gene, and composition of body and fatness in duck. In contrast, polymorphism of *GH* and productive traits in Mazandaranian native poultry were found to be not correlated (14). Results from the current study indicated that there was a significant association between three genes and the composition of carcasses in local quail. These results can also be a valuable index to properly select and to genetically improve local quail. Finally, further researches and experiments are recommended in order to clarify direct interaction of polymorphism of three genes with growth and carcasses composition traits in local quail.

### CONCLUSION

It is concluded that a great genetic variation exist among the three lines studied. Candidate genes *SEMA3E*, *TLX* and *GH* genes were successfully amplified in local quail, indicating that these regions are conserved and some genotypic profiles of the above candidate genes were found to be associated with growth trait in local quail and could be used as a potential marker for selection for body weight in the production of local quail lines.

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