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Study the Effect of Genetic Polymorphism in the Estrogen Receptor Gene (ESR) On the Meat Characteristics and Some Production Traits of Japanese Quail

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Abstract

Study was Procedure in the poultry field of the Agricultural Research Station / College of Agriculture and marshes / Thi- Qar University for the period from 10/10/2023 to 3/15/2024, the period of field and laboratory work, as 150 Japanese quail birds were raised from the age of one day to 35 days.

Birds were numbered with plastic numbers, and the weights were taken weekly to calculate the weekly weight gain and feed conversion efficiency. After the end of the breeding period, the birds were slaughtered and the relative weight of the edible internal organs (heart, liver, gizzard), tenderness, and juiciness were calculated. Genetic analyzes were conducted in the laboratory of the Marshes Research Center/ Thi- Qar University. With the aim of extracting the genetic material and determining the phenotypic structures as well as performing electrophoresis of the studied samples, the amplification product was then sent to the KCM, for the purpose of determining the sequence of the nitrogenous bases of the studied part of the gene, determining the genotype of the ESR gene and studying its relationship with some productive and sensory traits. Which included (the rate of weekly weight gain, the efficiency of food conversion, the relative weight of the eaten parts, freshness and juiciness), where the location of the variation in the nitrogenous bases of the studied gene was diagnosed, and the genotypes resulting from this variation were determined, with the studied piece registered in the Gene Bank LC816736, and the results were as follows:

1- The possibility of amplifying the studied segment of the ESR gene, which is 301 base pairs, and confirming it through electrophoresis examination.

2- Knowing the location of the mutation in segment 100.T>C, as it did not change the amino acids of the ESR gene.
3- Determine Three genotypes as : CC, CT, and TT. The percentages were 12%, 74%, and 14%, and the frequency of the C allele was 0.49, while the frequency of the T allele was 0.51.
4-The results did not show significant differences in the weekly weight gain and food conversion efficiency for the resulting three genotypes.
5- There were no significant differences between the three genotypes of the studied gene for the weights of each of the eaten offal (heart, liver, and gizzard).
6-The results did not show significant differences between the three genotypes of the studied gene for tenderness and juiciness.

Introduction

Most studies and research related to quails have been conducted on Japanese and European quails, while the local quails were not highlighted despite their importance as a source of animal protein and their good quality meat, the quails are small in size, gray in color, striped with black on the back and wings. There are also other colors such as white. The male has black collars around the neck, which distinguish him from the female, and his tail is short. Quail breeding has spread widely and intensively

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in the second half of the last century, especially in Japan, France, Italy and Germany, in order to benefit from their meat and eggs. (Hosni et al., 2016)

Recently, technology in genetics and molecular genetics has provided a number of genetic markers that have aided in genetic analysis and assessment of genetic diversity among different types of breeds and species in order to preserve them as a reliable source of genetic diversity (Abdulkareem, Azhar, 2022).

The productive performance of Japanese quail can be improved by improving its genetic characteristics and paying attention to potential environmental conditions. Quail meat is preferred over chicken meat due to its low fat content (low calories), taste and appetite. Therefore, the demand for quail meat is increasing, as is the search for how to produce such meat in a shorter period. This can be achieved through a specific selection program for high body weight at a certain age (Gnana et al., 2010). The purpose of studying carcass characteristics is to evaluate objective factors related to qualitative and quantitative aspects (Abdullah, Abdulkareem, (2019).

For birds that are genetically superior ,growth rate and production are an important aspect in commercial quail production, and it is impossible to accept that all economic traits are independent in their biology because these traits show phenotypic and genetic correlations resulting from linkage or polymorphism. The overall production performance of Japanese quail can be improved by knowing the selection indicators for several traits. Although reports are available on many types of selection indicators for chickens, this information is scant for Japanese quail and this leads to the search for good selection indicators for genetic improvement and the development of selection criteria that can be used in a future breeding program (Khairy et al., 2016).

The role of ESR in poultry production performance has been increasingly studied in recent years. The ESR gene is one of the candidate genes for discovering polymorphisms associated with production and egg traits in chickens, animals, and others (Wu et al., 2015).

I. Materials and Methods

This study was conducted in the animal production field of the Agricultural Research Station/College of Agriculture and Marshes / Thi- Qar University for the period from (10/10 2023) until the study period included two stages:

Field work stage: 150 unsexed Japanese quail chicks were raised from one day old to 35 days old using a cage-rearing system. The birds were sexed at three weeks of age. The cages were prepared to house the birds, made of wood and surrounded by plastic BRC. Dimensions of the cage (height: 150 cm, depth: 90 cm, width: 90 cm). Lighting was continuous for 24 hours, and water and feed were given freely. The preventive and health program recommended by the Agricultural Research Office was recommended, where some of the bird's characteristics were studied. Such as weight gain and feed conversion efficiency, and to ensure obtaining the required heat according to the age of the birds during the study period, as stated in (Abdel Majeed and Mahrous, 2001).

Table (1) shows the temperatures during the study period

Week	Temperature
1	35
2	32
3	30
4	27
5	24

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Laboratory work stage: The blood collection process took place when the birds were slaughtered, where 5 ml of blood was collected for each test tube from each bird and stored in test tubes containing the anticoagulant EDTA. The samples were transferred in a refrigerated box and kept frozen at a temperature of (-20). Celsius until the time of extraction of genetic material.

Sensory Tests:

The samples were evaluated according to the method of Tahir (1979), where cooked meat samples were presented for the purpose of sensory evaluation by a number of experienced arbitrators in the Department of Animal Production at the College of Agriculture and Marshes to evaluate the samples in terms of freshness and juiciness.

Table (2) shows Sensory evaluation form

Sensory Evaluation Form			
Degree	**Evolution**	**Degree**	**Evolution**
9	**Excellent**	**5-6**	**Middle**
8	**Very good**	**4-3**	**Acceptable**
7	**Good**	**2-1**	**Unacceptable**

DNA Extraction

DNA was extracted from blood samples by method included in the diagnostic kit (the kit supplied by the Korean company Geneaid)

Primers Preparation

The initials for ESR gene were prepared by the KCM in the form of a dried powder of two primers separated from each other, each primer placed in a special tube with a label showing the sequence of nitrogenous bases.

Table (3) shows the primers used for ESR gene

Gene	Primers	Widget size	Source
ESR 4	**F 5'- CGGGCGAATGATGAAACA- 3'** **R 5'- CCCAGTTGATCATGTGCA– 3'**	**Base pair 301**	**Dong 2020 , others**

Polymerase Chain Reaction (PCR):

To amplify the DNA of the ESR gene, Tables (4) and (5) show the materials used in molecular detection using polymerase chain reaction for the studied ESR gene. The samples are placed in the polymerase chain reaction device.

Table (4) shows Materials used in the PCR technique and their quantities

Chemical	Master Mix	DNA template	Data		Distilled water	Final size
			Reverse	Forward		
Volume (micro liters)	**13**	**4**	**1**	**1**	**6**	**25**

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Table (5) shows The program for PCR technology for gene ESR

Gene	Stages	Temperature	Time (minutes)	Courses Number
ESR Gene	Primary Metamorphosis	95C	5 m.	1
	Metamorphosis	95C	30 S.	35
	Adhesion	58C	30 S.	
	Elongation	72C	45 S.	1
	Final elongation	72C	10 m.	

Statistical Analysis

Statistical analysis of the study data was conducted using the ready-made statistical program (SAS Statistical analysis system (SAS, 2012) and included studying the effect of the studied gene (ESR) and by week on the traits under study (growth and carcass traits in quail) and according to the mathematical model below, and the significant differences were compared between Means were calculated using Duncan's multinomial test (Duncan, 1955) and using a completely randomized design (CRD).

Mathematical model: $Yijk=\mu+GI+Eijk$

Results and Discussion

Weekly Weight Gain:

The results in Table 6 indicate that there are no significant differences between the genotypes in the rate of weekly weight gain during the breeding period of five weeks, as the weight gain at the end of the experiment (fifth week) for the genotypes TT, CT, CC was (27.428, 27.783, 30.166 grams). Respectively.

Table(6) shows the weekly body weight ± standard error between the different genotypes in ESR G. for the weeks from the first to the fifth week (Japanese quail) respectively.

Traits	Genetic structure	No.	Mean (g) ± standard error	Significant
First week	CC	6	±1.668021.50	N.S
	CT	37	±0.551024.54	
	TT	7	21.857 ±1.335	
Total		**50**	**22.632 ±1.558**	
Second week	CC	6	22.486 ±0578	N.S
	CT	37	52.428 ±0.961	
	TT	7	57.428 ±0.578	
Total		**50**	**23.759 ±1.221**	
Third week	CC	6	52.1667 ±1.796	N.S
	CT	37	53.162 ±0.834	
	TT	7	53.142 ±1.56	
Total		**50**	**52.490±1.228**	

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Fourth week	CC	6	45.166 ±3.637	N.S
	CT	37	42.891 ±1.082	
	TT	7	43.142 ±3.367	
Total		**50**	**43.733 ±2.698**	
Fifth week	CC	6	30.166 ±3.070	N.S
	CT	37	27.783 ±1.246	
	TT	7	27.428 ±3.293	
Total		**50**	**28.459±2.536**	

Feed Conversion Efficiency

The results of Table 7 indicate that there are no significant differences between the genotypes in the feed conversion efficiency during the rearing period of five weeks. The feed conversion efficiency at the end of the experiment (fifth week) for the genotypes CC and CT was 5.504, 5.504, and 4.713 grams of TT, respectively, and was not available. Previous studies on the relationship of the gene under study with feed conversion efficiency.

Table (7): shows efficiency of food conversion ± the standard error among the different genotypes in ESR g from the first week to the fifth week for the Japanese quail.

Traits	Genetic structure	No.	Mean (g) ± standard error	Significant
First week	CC	6	3.833 ±0.295	N.S
	CT	37	3.326 ±0.084	
	TT	7	3.739 ±0.218	
Total		**50**	**3.632 ±0.199**	
Second week	CC	6	5.90 ±0.543	N.S
	CT	37	5.179 ±0.153	
	TT	7	4.503 ±0.192	
Total		**50**	**4.924 ±0.888**	
Third week	CC	6	3.6020 ±0.438	N.S
	CT	37	3.162 ±0.049	
	TT	7	3.205 ±0.064	
Total		**50**	**3.323 ±0.183**	
Fourth week	CC	6	2.746 ±0.230	N.S
	CT	37	2.934 ±0.106	
	TT	7	2.911 ±0.283	
Total		**50**	**2.863 ±0.283**	
Fifth week	CC	6	4.713±0.593	N.S
	CT	37	5.504 ±0476	
	TT	7	5.507 ±0.973	
Total		**50**	**5.241±0.680**	

Eaten Entrails

The results shown in Table 8 indicate that there are no significant differences in the relative weight of the heart muscle, and the proportions of the genotypes CC, CT, and TT were (1.899, 2.021, 1.914), respectively. These results agreed with Jun et al. (2021) for a study on a bird. Korean quail, where the genotypes CC, CT, TT and their ratios were (1.180, 1.122, 1.006), respectively. At the same time, this result did not agree with Jun et al. (2020), who indicated that there were significant

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differences at the level (P≤0.05) when he conducted a study. On the Chinese yellow quail, it was found that there was a significant superiority of the CC genotype over (TT1.067, 0.80), respectively.

As for the results of the relative weight of the liver and gizzard, they indicated that there were no significant differences for the genotypes CC, CT, and TT for the experimental samples, and this is what Jun and others found in (2020) who showed that there were no significant differences for the Korean quail in the genotypes TT, CC (5.773). 7,600, respectively, as well as what was confirmed by Jun and others in (2021) in a study of Chinese yellow quail with genotypes CC, CT, TT (3,700, 3,500,833), respectively.

Table (8): Shows the Relative weights of internal organs (g) ± the standard error between the different genotypes in ESR g. of the Japanese quail.

Traits	Genetic structure	No.	Mean (g) ± standard error	Significant
Heart	CC	6	1.899 ±0.106	N.S
	CT	37	2.021 ±0.041	
	TT	7	1.914 ±0.121	
Total		**50**	**1.499 ±0.089**	
Liver	CC	6	3.0313 ±0.297	N.S
	CT	37	3.3587 ±0.058	
	TT	7	3.082 ±0.039	
Total		**50**	**3.157 ±0.131**	
Gizzard	CC	6	2.703 ±0.142	N.S
	CT	37	2.855 ±0.055	
	TT	7	2.608±0.057	
Total		**50**	**2.722 ±0.084**	

Tender and Juiciness

The results of Table 9 indicate that there are no significant differences in freshness between the genotypes CC, CT, and TT (7.402, 7.325, 6.561), and also no significant differences were observed in juiciness between the genotypes CC, CT, and TT (7.217, 7.110, 6.65) as shown below:

Table (9): Shows Meat sensory Traits, % ± standard error between the genotypes of the ESR G. for Japanese quail.

Traits	Genetic structure	No.	Mean (g) ± standard error	Significant
Tender	CC	6	6.561 ±0.602	N.S
	CT	37	7.325 ±0.170	
	TT	7	7.402±0.329	
Total		**50**	**7.069 ±0.367**	
Juiciness	CC	6	6.65±0.523	N.S
	CT	37	7.110±0.154	
	TT	7	7.217±0.267	
Total		**50**	**6.992 ±0.944**	

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