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TRANSGENIC QUAIL: TOWARDS IMPROVE EGG PRODUCTION AND EGG QUALITY.

By

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Abstract: *The present study was conducted to determine the influence of transgenesis of Japanese quail on the egg production and external and internal egg quality during the first 45 days of lay such as egg production percentage, egg weight, egg mass, egg shape index, egg shell percentage, egg Shell thickness, yolk index, yolk weight percentage, and albumen weight percentage, and transgenic response was measured. The experimental population used in this study based on two generations which has been described in details previously* ***(Rabie et al., 2009)****, which produced by inoculation of the fragmented purified DNA or genes - as part of growth hormone gene - from pituitary gland either from Hubbard broiler breeders chickens (HB) or Muscovy ducks (MD), at two levels of concentration (20 and 40ng) into embryonic cell of two Japanese quail strains (Brown and Golden).*

The obtained results showed that inoculated MD gene$_{40ng}$ increased egg number and egg mass of first generation (G_1) for transgenic Brown quail (33.3 & 436.9) compared to the HB gene at the same level (19.8 & 240.17). The same trend was also, noticed in the second generation (G_2) for Golden quail strain. Golden quail had more variation due to transgenesis than Brown quail during G_1 and G_2. MD DNA source increased shell weight percentage of both Brown and Golden quail strains during G_1 (14.9 & 15.23 %) compared to that the HB source (13.88 & 10.92%), while MD gene$_{40ng}$ increased egg yolk weight percentage of Golden quail (32.64%) compared to MD DNA$_{20ng}$ during G_2 (30.29%). Moreover, Brown quail of HB gene origin showed positive transgenic response in egg number and egg mass, while Golden quail strain had positive transgenic response on egg number with both MD DNA, and MD gene sources.

INTRODUCTION

The Japanese quail (Coturnix coturnix japonica) is gaining widespread favour as a pilot animal among investigators in avian genetics, nutrition, and physiology. Many studies have been published on egg production in Japanese quail (**Bacon *et al.*, 1986; Strong *et al.*, 1978; Nestor *et al.*, 1982, 1983, Minvielle *et al.*, 2000**). Improvement in the genetic potential has led to higher egg production with increased egg size than achieved earlier (**Yamane *et al.*, 1980; Shrivastav *et al.*, 1989**). During the last decade interest in the application of biotechnology in the form of transgenic livestock and poultry has remained strong. Today, the production of transgenic livestock, poultry, and fish is goal for academic as well as commercial groups. In general, the goal of gene transfer in poultry as agricultural setting is to develop genetically better birds for the efficient production of meat and eggs.

The quail has recently been used as a successful transgenic model for the production of a transgenic bird. A transgene was introduced into freshly laid eggs, showing that the new gene had been successfully introduced into the birds (**Shin *et al.*, 2008; Poynter *et al.*, 2009; Rabie *et al.*, 2009**). Moreover, the DNA and/or genes could become integrated into the genome and was shown occasionally to be functional (**Wagner *et al.*, 1981**). Therefore, the quail is a suitable species for creating transgenic birds. It has a hardy embryo, which survives the introduction of the new transgene well. Although meat production was considered more commonly, the egg breeding became more important in some countries such as Egypt. Besides, the productivity and quality of the breeding eggs have an overall significant for the continuity of flocks for an economical breeding (**Sogut *et al.*, 2001**). Moreover, external and internal quality traits of the eggs are significant in the poultry breeding for their influence on the yield features of the future generations, breeding performances, and quality and growth of the chicks (**Altinel *et al.*, 1996; McDaniel *et al.*, 1978**). While several factors influencing the production and quality characteristics of quail eggs have been reported, information about the relationship between external and internal traits of eggs and age of layer is rather limited (**Yannakopolos and Tserventi-Gousi, 1986**).

The aim of this study is to determine the influences of transgenesis of Japanese quail - which produced by introducing foreign DNA and genes represented as part of growth hormone gene from pituitary gland of broiler breeder chickens and/or Muscovy duck into the embryonic cell of Brown and Golden Japanese quails (**Rabie *et al.*, 2009**) - on the egg production and

external and internal egg quality during the first 45 days of lay such as egg production percentage, egg weight, egg mass, egg shape index, egg shell percentage, egg shell thickness, yolk index, yolk weight percentage, and albumen weight percentage, and transgenic response was measured.

MATERIALS AND METHODS

Experimental population:

The experimental population used in this study based on two generations (G_1& G_2), and has been described in detail previously (**Rabie *et al.*, 2009**). Briefly, two different sources of DNA and growth hormone gene from Hubbard Broiler (HB), and Muscovy Duck (MD) at two levels of concentration (20 and 40ng) were inoculated in two lines of Brown and Goden Japanese quail. Five-hundred and eighty-three eggs in one hatch were obtained from laying Japanese quail (brown and golden) reared under normal conditions in the Poultry Research laboratory, Faculty of Agriculture (Saba Bacha), Fac. of Agric. Alexandria University (Figure 5). Eggs were collected and stored up to seven days in 12-15°C temperature. At the third day of incubation, the fertile eggs were randomly assigned into eight experimental groups as follow: The groups of egg injected with 20 and 40 ng/µl for Hubard broiler breeder genomic DNA (HB DNA_{20ng}, HB DNA_{40ng}) were 94 and 51 eggs, respectively, while the corresponding numbers of eggs injected with 20 and 40 ng/µl genes/egg (HB $gene_{20ng}$, HB $gene_{40ng}$) were 39 and 71 eggs, respectively. The groups of eggs injected with 20 and 40 ng/µl for Muscovy ducks genomic DNA (MD DNA_{20ng}, MD DNA_{40ng}) were, 95 and 27 eggs respectively, while the corresponding numbers of eggs injected with 40 ng/µl gene/egg (MD $gene_{40ng}$) was 70 eggs. In addition, 136 Eggs were used as a control without any treatment.

Hatched quail chicks from different families per treatment were wing-banded on the day of hatch and brooded in floor pens. The house temperature was kept at about 35°C during the first 3 days, 32°C during next 4 days and gradually decreased by 2°C weekly until the end of the third maintained at 24°C. Chicks were fed a growing diet of 24.1% protein and 2860 kcal/kg, till 5 wk of age. Pullets were fed a layer diet of 20.06% protein and 2820 kcal/kg, from 6-14 weeks of age. Two lines of quail were reared for both meat and egg production in the consequence two generations (G_1, and G_2). In recent study, the egg production line was used. In the G_1, 155 quails (75 Brown, and 80 Golden), and in G_2, 468 quails (226 Brown, and 242 Golden) were used. Phenotypic data were measured as follows:

Egg production traits:

Hen day egg production percentage was calculated. In addition, an egg number/hen/45 day was calculated by dividing the total number of eggs that were collected by hen number for each treatment. Egg mass was calculated by multiplying the average number of eggs for each hen and the average weight of eggs (during the first 45 days).

Egg Quality Traits:

In recent study, 0.01 g sensitive electronic scale was used for weighing the eggs; a compass sensitive to 0.01 mm was used for measuring the length, width, yolk diameter of the eggs; a table with a flat glass on it was used on which the eggs are broken; and a 3-legged micrometer sensitive to 0.01mm was used for measuring the height of yolk and a micrometer sensitive to 0.01 was used for measuring the shell thickness.

The collected eggs were balanced in order to determine their weights. The width and length of the egg were measured. After this process, the eggs were broken on table with a glass cover in order to measure yolk height, and yolk diameter. The egg shells were washed under slightly flowing water so that albumen remains are removed. The washed eggshells were left to dry in the open air for 24 hours. Then, they were balanced together with the eggshell membrane. Finally, samples taken from sharp, blunt and equatorial parts were measured and the average shell thickness was obtained from the average values of these three parts **(Tyler, 1961)**. Egg shape index was determined according to **Romanoff and Romanoff (1949)**. Albumen weight percentage was also calculated. Yolk index was estimated by dividing the height of yolk on its diameter as reported by **Funk (1948).**

Transgenic Response:

The realized transgenic response was estimated according to the numerator of following equation **(El-Tahawy, 2005)**. $R = (S_2 - S_1) - (C_2 - C_1)$; Where S_1 and S_2 were the mean of transgenic at the first and second generation respectively; C_1 and C_2: were the mean of control at the first and second generation respectively.

Statistical Analysis:

Data were analyzed using SAS program **(SAS, Institute, Inc., 1997)**, by the application of the General linear model procedure (GLM). Test of Significance for the differences between treatments or levels were done according to **Duncan (1955)**. The statistical model used was nested (unbalanced) design as follows:

$Y_{ijklm} = \mu + BR_i + BR_i(SO)_j + DG_k(BR*SO)_{ij} + LEV_l(BR*SO*DG)_{ijk} + e_{ijklm}$

Where: Y_{ijklm}= Observations, μ= Overall means, BR_i= The effect of breeds (1,2), $BR_i(SO)_j$= The effect of breeds within source (1,2,3), $DG_k(BR*SO)_{ij}$= The effect of breeds/ sources/ DG (1,2,3), $LEV_l(BR*SO*DG)_{ijk}$= The effect of breeds/ sources/ DG/ levels (1,2,3), and e_{ijklm}= Residual

RESULTS

Egg production traits:

First Generation:

Egg number and laying rate during the first 45 days of lay:

Least squares means of egg number during the first 45 days of lying for Brown and Golden quail hens treated with different sources of DNA or gene and control group are presented in Tables (1 and 2). Inoculated MD $gene_{40ng}$ significantly ($P< 0.05$) increased egg number during the first 45 days in Brown quail (33.30) compared to the HB $gene_{40ng}$ (19.80). No significant differences were shown in egg number during the first 45 days of transgenic Golden quail due to (HB and MD) DNA source and genes within the studied levels (20 or 40ng) (Table 2). Regardless significance, the obtained results demonstrated that egg number and laying rate of Brown quail treated by HB- DNA source more than control by (13.3 and 15.5%), respectively. However, the increase in these traits reached to (7.2 and 7.6%) in Golden quail.

Table (1): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs. Duck) and type (DNA vs. Gene) on egg traits during the 1st generation.

Breeds	Source / Type	Egg number hen/45 day			Rate of laying /45 days			Average egg weight (gram)			Egg mass hen/ 45 day (gram)		
		HB	MD	C	HB	MD	C	HB	MD	C	HB	MD	C
B	DNA	33.75± 3.15^{ax}	27.00± 4.95^{ax}	27.00± 2.25^{ax}	74.55± 6.55^{ax}	59.84± 11.08^{ax}	59.74± 4.94^{ax}	12.39± 0.28^{ax}	11.91± 0.37^{ax}	12.07± 0.35^{ax}	418.16± 38.25^{ax}	321.57±5 3.10^{ax}	325.89± 21.15^{ax}
	Gene	27.45± 0.45^{ax}	33.30± 1.80^{ax}	27.00± 2.25^{ax}	61.02± 9.53^{ax}	74.41± 3.64^{ax}	59.74± 4.94^{ax}	12.64± 0.59^{ax}	13.12± 0.27^{abx}	12.07± 0.35^{acx}	346.97± 59.85^{ax}	436.90± 27.45^{abx}	325.89± 21.15^{acx}
	Average	30.60± 2.25^{aZ}	30.15± 2.70^{aZ}	27.00± 2.25^{aZ}	67.78± 5.43^{aZ}	67.13± 5.54^{aZ}	59.74± 4.94^{aZ}	12.51± 0.27^{aZ}	12.52± 0.30^{aZ}	12.07± 0.35^{aZ}	382.81± 32.40^{aZ}	377.48± 33.75^{aZ}	325.89± 21.15^{aZ}
G	DNA	34.65± 2.25^{ax}	28.35± 5.40^{ax}	31.05± 2.70^{ax}	77.20± 4.98^{ax}	63.17± 12.31^{ax}	69.14± 6.16^{ax}	12.49± 0.19^{ax}	12.45± 0.34^{ax}	12.31± 0.39^{ax}	432.78± 25.20^{ax}	352.96±6 8.85^{ax}	382.23± 37.35^{ax}
	Gene	32.40± 3.15^{ax}	21.60± 4.95^{bx}	31.05± 2.70^{ax}	71.59± 7.39^{ax}	47.80± 10.76^{bx}	69.14± 6.16^{ax}	12.99± 0.44^{ax}	12.11± 0.37^{ax}	12.31± 0.39^{ax}	420.88± 49.50^{ax}	261.58± 63.00^{bx}	382.23± 37.35^{ax}
	Average	33.30± 1.80^{aZ}	24.75± 3.60^{bcZ}	31.05± 2.70^{acZ}	74.39± 4.26^{aZ}	55.49± 8.07^{bcZ}	69.14± 6.16^{acZ}	12.74± 0.23^{aZ}	12.28± 0.26^{aZ}	12.31± 0.39^{aZ}	424.24± 26.55^{aZ}	303.93± 6.35^{bcZ}	382.23± 37.35^{acZ}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{x-y}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

$^{Z-W}$ Means within the same column, between Average, having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (2): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs. Duck), type (DNA vs. Gene) and levels (20 vs. 40) on egg traits during the 1st generation.

Breeds	Type / Source	Egg number hen/ 45 day				Rate of laying /45 days				Average Egg weight (grm)				Egg mass hen/ 45 day			
		DNA		Gene		DNA		Gene		DNA		Gene		DNA		Gene	
		20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl
B	HB	28.35± 3.15^{ab}	38.70± 0.90^{ab}	35.10± 2.25^{ab}	19.80± 3.60^{ab}	62.91± 7.26^{ab}	86.19± 1.75^{ab}	77.7± 5.32^{ab}	44.34± 8.42^{ab}	12.32± 0.24^{ab}	12.46± 1.12^{ab}	13.15± 0.10^{ab}	12.13± 0.97^{ab}	349.27± 42.75^{ab}	482.2± 33.75^{ab}	461.5± 27.90^{ab}	240.17± 45.90^{ab}
	MD	27.00± 4.95^{ab}	-	-	33.30± 1.80^{ab}	59.84± 11.0^{ab}	-	-	74.41± 3.64^{ab}	11.91± 0.37^{ab}	-	-	13.12± 0.27^{ab}	321.57± 53.10^{ab}	-	-	436.90± 27.45^{ab}
	C	27.00± 2.25^{b}	27.00± 2.25^{b}	27.00± 2.25^{b}	27.00± 2.25^{ab}	59.74± 4.94^{b}	59.74± 4.94^{b}	59.74 ±4.94^{b}	59.74± 4.94^{b}	12.07± 0.35^{b}	12.07± 0.35^{b}	12.07± 0.35^{b}	12.07± 0.35^{b}	325.89± 21.15^{b}	325.89 ±21.15^{b}	325.89 ±	325.89± 21.15^{b}
G	HB	30.60±2 .25^{ab}	39.15± 1.80^{ab}	34.20± 1.80^{ab}	30.15± 6.75^{ab}	67.90± 4.62^{ab}	86.51± 3.65^{ab}	75.7± 4.05^{ab}	67.46± 15.4^{ab}	12.85± 0.11^{ab}	12.13± 0.32^{abc}	13.77± 0.18^{abc}	12.21± 0.57^{abc}	393.21± 26.10^{ab}	474.8± 23.40^{ab}	470.93 ±	368.13± 96.30^{ab}
	MD	28.35± 5.40^{ab}	-	-	21.60± 4.95^{ab}	63.17± 12.31^{ab}	-	-	47.80± 10.76^{ab}	12.45± 0.34^{ab}	-	-	12.11± 0.37^{ab}	352.9± 68.85^{ab}	-	-	261.58± 63.00^{ab}
	C	31.05± 2.70^{b}	31.05± 2.70^{b}	31.05± 2.70^{b}	31.05± 2.70^{ab}	69.14± 6.16^{b}	69.14± 6.16^{b}	69.1± 6.16^{b}	69.14±6. 16^{ab}	12.31± 0.39^{b}	12.31± 0.39^{b}	12.31± 0.39^{b}	12.31± 0.39^{b}	382.23± 37.35^{b}	382.2± 37.35^{b}	382.23 ±	382.23± 37.35^{b}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

^{x,y} Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Average egg weight during the first 45 days of laying:

The effect of transfused hens with (HB) or (MD) DNA or gene at different levels (20 or 40ng) and control on average egg weight during the first 45 days of laying Brown or Golden quail layers is given in Tables (1and 2). Inoculated MD $gene_{40ng}$ significantly ($P< 0.05$) increase average egg weight in Brown quail during the first 45 days (13.12 g) compared to the MD DNA_{20ng} (11.90g) and control group (12.07g). HB $gene_{20ng}$ significantly ($P< 0.05$) increase average egg weight of Golden quail during the first 45 days (13.77g) compared to HB $gene_{40ng}$ (12.21g) and HB DNA_{40ng} (12.13 g). Considering the Brown quail. MD $gene_{40ng}$ significantly increase the average egg weight compared to quail that treated by MD DNA by 10.2%, and 8.7% compared to the control group.

Egg mass during the first 45 days of laying:

The effect of transfused hens with foreign (HB) or (MD) DNA or gene at different levels (20 or 40ng) and control egg mass at 45 days of laying in both Brown or Golden quail layers is given in Tables (1and 2). Inoculated MD $gene_{40ng}$ significantly ($P< 0.05$) increase egg mass during the first 45 days of Brown quail from (436.90g) compared to the HB $gene_{40ng}$ (240.17g) and control group (325.89g). Moreover, MD gene, on the average of the two levels, improved Brown quail egg mass during the first 45 days compared to the corresponding control group (436.90 Vs. 325.89 g), respectively (Table 1).

Second Generation:

Egg number during the first 45 days of laying:

Wide range of egg number of the different treated groups subjected to DNA or gene inoculation was observed in Tables (3 and 4). The average of the two levels, improved Golden quail egg number during the first 45 days compared to the corresponding HB gene (39.60 vs. 30.15) (Table 3). Inoculated MD DNA_{20ng} significantly ($P< 0.05$) increase egg number in Brown quail during the first 45 days from (40.95) compared to the HB DNA_{20ng} (28.80); the corresponding value for egg production percentage was (90.74%) and (63.56%), respectively. In Golden quail MD $gene_{40ng}$ significantly ($P< 0.05$) increase egg number during the same period from (36.00) compared to the HB $gene_{40ng}$ (22.50). The corresponding value for egg laying rate was (79.68%) and (49.63%), respectively (Table 4).

Average egg weight and egg mass during the first 45 days of laying:

In the present study, The effect of transfused hens with (HB) or (MD) DNA or gene at different levels (20 or 40ng) and control on egg weight, and egg mass during the first 45 days of laying Brown or Golden quail layers is given in Tables (3 and 4). There were no significant differences among hen treated groups were observed on average egg weight.

Egg quality traits:

First Generation:

Proportional (percentages %) weight of various egg components (albumen, yolk, and shell); egg shell thickness, yolk index, and egg shape Index were evaluated as an indication of egg quality traits. Data presented in Tables (5 and 6) showed the average values (least square means ± standard errors) for egg quality traits as affected by inoculation of (HB vs MD) DNA and genes sources and levels.

Albumin weight percentage:

Non Significant variation ($P<0.05$) was observed due to treatment by DNA and gene sources which as a function of albumen and egg weights, but not on proportional egg albumen weight (Tables 5 and 6). The results showed also that MD gene source significantly decreased albumen weight percentage as compared to control group of Golden quail (Table 5). No significant differences were shown in albumen weight percentage of transgenic Brown and Golden quail due to (HB and MD) DNA source and genes within the studied levels (20 or 40ng).

Egg yolk weight percentage and yolk index:

No significant differences were shown in yolk weight percentage of transgenic Brown and Golden quail due to (HB and MD) DNA source and genes within the studied levels (20 or 40ng).

Egg shell weight percentage, egg shape index, and shell thickness:

The mean values of egg shell weight percentage, egg shape Index and shell thickness of different DNA and gene treatments and levels are shown in Tables (5 and 6). Quail strain effect was only cleared ($P < 0.05$) for shell weight percentage and egg shape index on the whole average. In Brown quail, shell weight percentage was insignificantly affected by inoculated HB gene and DNA (20ng). However, it was significantly ($P<0.05$) decreased in the group treated by HB $gene_{40ng}$ (12.50%) as compared to the aforementioned groups. Inoculated HB DNA_{40ng}

significantly (P< 0.05) decreased shell weight percentage as compared to the control group. The decreased amounted to 14.3%. Concerning Golden quail, the results showed that inoculated MD $gene_{40ng}$ significantly (P< 0.05) improved egg shell weight percentage by 9.7 and 13.5% as compared to the group treated by MD DNA_{20ng} and control group, respectively. In addition, inoculated HB DNA_{40ng} increased egg shell weight percentage by 15.3 and 13.3% as compared to the transfused HB DNA_{20ng}, and control group (Table 6). Although, no significant differences were shown in shell thickness (mm) of transgenic Brown quail due to (HB and MD) DNA and genes within the studied levels (20 or 40ng), shell thickness (mm) of transgenic Golden quail HB $gene_{20ng}$ was significantly (P< 0.05) improved by 10.6, 11.7, 8.0 and 9.6 % as compared to the group treated by HB $gene_{40ng}$, HB DNA at levels 20ng and 40ng, and the control group, respectively. In control group of Golden quail egg shape index was significantly (P< 0.05) increased by 4.7% as compared to control group Brown quail (79.43 vs. 75.9). It was observed that transgenic Brown quail by HB $gene_{20ng}$ significantly (P< 0.05) increased egg shape index by 4.7% (Table 6). The other levels of different HB and MD DNA and gene origin did not induce any significant increase in egg shape index of both quail strains (Table 6).

Second Generation:

Proportional weight of various egg components (albumen, yolk, and shell); egg shell thickness, yolk index, and egg shape index were evaluated as an indication of egg quality traits. Data presented in Tables (7 and 8) showed the average values (least square means ± standard errors) for egg quality traits as affected by (HB vs MD) DNA and genes sources and levels.

Albumin weight percentage:

MD Gene source effects were evident (P < 0.05); on the average, improved Brown quail albumen weight percentage compared to the corresponding control group (55.45 vs. 53.91%).

Moreover, MD $gene_{40ng}$ significantly (P< 0.05) improve albumen weight % of Brown quail from (55.45%) compared to the control group (53.91%).

Table (3): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs. Duck), and type (DNA vs. Gene) on egg traits during 2nd generation.

Traits		Egg Number hen/45 day			Rate of laying /45 days			Average egg weight (gram)			Egg mass hen/45day (gram)		
Breeds	Source / Type	HB	MD	C	HB	MD	C	HB	MD	C	HB	MD	C
B	DNA	34.65± 4.05^{ax}	40.95± 4.50^{ax}	34.65± 4.05^{ax}	77.33± 9.31^{ax}	90.74± 10.43^{ax}	77.07± 8.50^{ax}	12.00± 0.59^{ax}	12.14± 0.84^{ax}	13.08± 1.12^{ax}	415.80± 59.85^{ax}	497.10± 21.60^{ax}	453.22± 55.35^{ax}
	Gene	37.35± 2.70^{ax}	34.20± 2.70^{ax}	34.65± 4.05^{ax}	82.96± 5.78^{ax}	75.86± 6.02^{ax}	77.07± 8.50^{ax}	13.20± 0.44^{ax}	13.16± 0.52^{ax}	13.08± 1.12^{ax}	493.02± 39.60^{ax}	450.07± 44.10^{ax}	453.22± 55.35^{ax}
	Average	36.00± 2.25^{aZ}	37.35± 2.25^{aZ}	34.65± 4.05^{aZ}	80.15± 2.20^{aZ}	83.30± 5.32^{aZ}	77.07± 8.50^{aZ}	12.60± 0.35^{aZ}	12.65± 0.44^{aZ}	13.08± 1.12^{aZ}	453.60± 34.65^{aZ}	472.48± 34.65^{aZ}	453.22± 55.35^{aZ}
G	DNA	39.60± 1.80^{ax}	41.85± 0.45^{ax}	40.50± 1.80^{ax}	87.96± 4.44^{ax}	92.96± 0.98^{ax}	90.37± 3.85^{ax}	13.01± 0.24^{ax}	12.70± 0.46^{ax}	13.05± 0.24^{ax}	515.20± 27.90^{ax}	531.50± 24.75^{ax}	528.53± 22.95^{ax}
	Gene	30.15± 4.05^{bx}	36.00± 2.70^{bcx}	40.50± 1.80^{acx}	66.62± 9.02^{bx}	79.68± 5.95^{bcx}	90.37± 3.85^{acx}	13.20± 1.23^{ax}	13.62± 0.28^{ax}	13.05± 0.24^{ax}	397.98± 68.40^{ax}	490.32± 39.60^{ax}	528.53± 22.95^{ax}
	Average	34.65± 2.25^{aZ}	38.70± 2.25^{aZ}	40.50± 1.80^{aZ}	77.29± 5.23^{aZ}	86.32± 4.55^{aZ}	90.37± 3.85^{aZ}	13.11± 0.59^{aZ}	13.16± 0.27^{aZ}	13.05± 0.24^{aZ}	454.26± 37.35^{aZ}	509.29± 22.95^{aZ}	528.53± 28.35^{aZ}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{x-z}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

$^{Z-W}$ Means within the same column, between Average, having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (4): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs Duck), type (DNA vs. Gene) and levels (20 vs. 40) on egg traits during 2nd generation.

Traits		Egg number hen/45day				Rate of laying /45 days.				Average egg weight (gram)				Egg mass hen/45day (gram)			
Breeds	Type / Source	DNA		Gene		DNA		Gene		DNA		Gene		DNA		Gene	
		20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl
B	HB	28.80± 4.50 ^{ab}	40.95± 4.95 ^{aA}	34.20± 3.15 ^{aA}	40.50± 4.50 ^{aA}	63.56± 9.92 ^{aA}	91.11± 11.20 ^{aA}	75.93± 6.77 ^{aA}	90.00± 9.10 ^{aA}	12.81± 0.64 ^{aA}	11.19± 0.90 ^{aA}	13.06± 0.54 ^{aA}	13.34± 0.10 ^{aA}	368.93± 71.55 ^{aA}	458.23± 40.05 ^{aA}	446.65± 41.40 ^{aA}	540.20± 100.35 ^{aA}
	MD	40.95± 4.50 ^{aAB}	-	-	34.20± 2.70 ^{aA}	90.74± 10.43 ^{aAB}	-	-	75.86± 6.02 ^{aA}	12.14± 0.84 ^{aA}	-	-	13.16± 0.52 ^{aA}	497.13± 21.60 ^{aA}	-	-	450.07± 44.10 ^{aA}
	C	34.65± 4.05 ^{aAB}	4.65± 4.05 ^{A}	34.65± 4.05 ^{A}	34.65± 4.05 ^{A}	77.07± 8.50 ^{aAB}	77.07± 8.50 ^{A}	77.07± 8.50 ^{A}	77.07± 8.50 ^{A}	13.08± 1.12 ^{A}	13.08± 1.12 ^{A}	13.08± 1.12 ^{A}	13.08± 1.12 ^{A}	453.22± 55.35 ^{A}	453.22± 55.35 ^{A}	453.2± 55.35 ^{A}	453.2± 55.35 ^{A}
G	HB	37.80± 3.15 ^{aA}	40.9± 2.25 ^{aA}	37.80± 1.80 ^{aA}	22.50± 6.30 ^{bA}	84.45± 6.52 ^{aA}	91.48± 5.23 ^{aA}	83.61± 3.94 ^{aA}	49.63± 14.42 ^{bA}	12.71± 0.17 ^{aA}	13.31± 0.57 ^{aA}	13.21± 0.70 ^{aA}	13.19± 0.95 ^{aA}	480.44± 40.05 ^{aA}	545.04± 25.20 ^{aA}	499.3± 29.25 ^{aA}	296.7± 112.05 ^{bA}
	MD	41.85± 0.4 ^{aA}	-	-	36.00± 2.70 ^{aA}	92.96± 0.98 ^{aA}	-	-	79.68± 5.95 ^{aA}	12.70± 0.46 ^{aA}	-	-	13.62± 0.28 ^{aA}	531.50± 24.75 ^{aA}	-	-	490.3± 39.60 ^{aA}
	C	40.50±1 .80 ^{A}	40.5± 1.80 ^{A}	40.50± 1.80 ^{A}	40.50± 1.80 ^{A}	90.37± 3.85 ^{A}	90.37± 3.85 ^{A}	90.37± 3.85 ^{A}	90.37± 3.85 ^{A}	13.05± 0.24 ^{A}	13.05± 0.24 ^{A}	13.05± 0.24 ^{A}	13.05± 0.24 ^{A}	528.53± 22.95 ^{A}	528.53± 22.95 ^{A}	528.5± 22.95 ^{A}	528.5± 22.95 ^{A}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

^{A} Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (5): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs. Duck) and type (DNA vs. Gene) on egg quality traits during the 1st generation.

Traits		Egg weight (grm)			Egg Shape index			Shell thickness (mm)			Shell weight (%)		
Breeds	Source / Type	HB	MD	C	HB	MD	C	HB	MD	C	HB	MD	C
B	DNA	12.83 ± 0.20^{bx}	14.54 ± 0.31^{acx}	13.43 ± 0.40^{bcx}	76.93 ± 0.71^{ax}	78.06 ± 1.77^{ax}	75.90 ± 1.81^{ax}	0.212 ± 0.003^{ax}	0.228 ± 0.004^{ax}	0.218 ± 0.004^{ax}	13.95 ± 0.35^{ax}	15.32 ± 1.03^{ax}	15.12 ± 0.35^{ax}
	Gene	14.85 ± 0.34^{ax}	14.90 ± 0.17^{ax}	13.43 ± 0.40^{bx}	78.34 ± 0.61^{ax}	77.95 ± 0.67^{ax}	78.90 ± 1.81^{ax}	0.215 ± 0.004^{ax}	0.218 ± 0.003^{ax}	0.218 ± 0.004^{ax}	13.82 ± 0.55^{bx}	14.48 ± 0.32^{bcx}	15.12 ± 0.35^{acx}
	Average	13.84 ± 0.32^{bZ}	14.72 ± 0.15^{aZ}	13.43 ± 0.40^{bZ}	77.63 ± 0.47^{aZ}	78.01 ± 0.62^{aZ}	75.90 ± 1.81^{aW}	0.214 ± 0.003^{aZ}	0.223 ± 0.003^{aZ}	0.218 ± 0.004^{aZ}	13.88 ± 0.32^{bW}	14.90 ± 0.33^{aZ}	15.12 ± 0.35^{aZ}
G	DNA	13.64 ± 0.31^{ax}	13.53 ± 0.46^{ax}	13.35 ± 0.35^{ax}	79.75 ± 0.81^{ax}	78.05 ± 1.16^{ax}	79.43 ± 1.67^{ax}	0.208 ± 0.005^{ax}	0.218 ± 0.008^{ax}	0.209 ± 0.003^{ax}	14.87 ± 0.44^{ax}	14.53 ± 0.36^{ax}	14.05 ± 0.33^{ax}
	Gene	14.67 ± 0.22^{ax}	13.93 ± 0.39^{bxz}	13.35 ± 0.35^{bx}	78.87 ± 0.59^{ax}	78.42 ± 0.46^{ax}	79.43 ± 1.67^{ax}	0.218 ± 0.00^{ax}	0.215 ± 0.004^{ax}	0.209 ± 0.003^{ax}	14.97 ± 0.33^{ax}	15.94 ± 0.50^{abx}	14.05 ± 0.33^{acx}
	Average	14.16 ± 0.19^{aZ}	13.73 ± 0.30^{acw}	13.35 ± 0.35^{bcZ}	79.31 ± 0.48^{aZ}	78.23 ± 0.47^{aZ}	79.43 ± 1.67^{aZ}	0.213 ± 0.003^{aZ}	0.216 ± 0.004^{aZ}	0.209 ± 0.004^{aZ}	14.92 ± 0.26^{aZ}	15.23 ± 0.38^{abZ}	14.05 ± 0.33^{acZ}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{x-z}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

$^{Z-W}$ Means within the same column, between Average, having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (5): Continued

Traits		Yolk weight (%)			Yolk index			Albumen weight (%)		
Breeds	Source / Type	HB	MD	C	HB	MD	C	HB	MD	C
B	DNA	33.04± 0.83^{ax}	32.87± 0.08^{ax}	31.16 ± 1.05^{ax}	0.487± 0.007^{ax}	0.487± 0.008^{ax}	0.498± 0.012^{ax}	53.00± 0.97^{ax}	51.81± 0.98^{ax}	53.72± 1.01^{ax}
	Gene	33.30± 0.87^{ax}	32.41± 0.53^{ax}	31.16 ± 1.05^{ax}	0.473± 0.008^{ax}	0.478± 0.010^{ax}	0.498± 0.012^{ax}	52.88± 1.15^{ax}	53.11± 0.56^{ax}	53.72± 1.01^{ax}
	Average	33.17± 0.60^{aZ}	32.64± 0.43^{aZ}	31.16 ± 1.05^{aZ}	0.480± 0.005^{aZ}	0.483± 0.008^{aZ}	0.498± 0.012^{aZ}	52.94± 0.74^{aZ}	52.46± 0.49^{aZ}	53.72± 1.01^{aZ}
G	DNA	33.07± 0.59^{ax}	31.73± 1.03^{ax}	31.96 ± 1.33^{ax}	0.482± 0.012^{ax}	0.468± 0.012^{ax}	0.479± 0.09^{ax}	52.16± 0.77^{ax}	53.74± 1.24^{ax}	54.00± 1.51^{ax}
	Gene	33.54± 0.61^{ax}	32.82± 0.73^{ax}	31.96 ± 1.33^{ax}	0.479± 0.013^{ax}	0.478± 0.010^{ax}	0.479± 0.09^{ax}	51.49± 0.66^{ax}	51.24± 1.01^{acx}	54.00± 1.51^{abx}
	Average	33.31± 0.44^{aZ}	32.27± 0.60^{aZ}	31.96 ± 1.33^{aZ}	0.481± 0.009^{aZ}	0.473± 0.007^{aZ}	0.479± 0.009^{aZ}	51.77± 0.51^{aZ}	52.49± 0.81^{aZ}	54.00± 1.51^{aZ}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{x-y}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

$^{Z-W}$ Means within the same column, between Average, having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (6): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs. Duck), type (DNA vs Gene) and levels (20 vs. 40) on egg quality during the 1st generation.

		Egg weight (grm)				Egg shape index				Shell thickness (mm)				Shell weight (%)			
Breeds	DG	DNA		Gene		DNA		Gene		DNA		Gene		DNA		Gene	
	Type	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl
B	HB	13.62± 0.18^{bA}	12.03± 0.15^{cA}	14.71± 0.20^{aA}	14.99± 0.07^{aA}	78.43± 0.76^{aB}	75.44± 1.45^{aA}	79.46± 0.67^{aA}	77.21± 0.91^{aA}	0.214± 0.003^{aA}	0.210± 0.015^{aA}	0.222± 0.005^{aA}	0.208± 0.007^{aA}	14.87± 0.35^{aA}	13.04± 0.89^{bcA}	15.12± 0.78^{aA}	12.51± 0.50^{bcA}
	MD	14.54± 0.31^{aA}	-	-	14.91± 0.17^{aA}	78.06± 1.77^{aB}	-	-	77.95± 0.67^{aA}	0.228± 0.004^{aA}	-	-	0.216± 0.003^{aA}	15.32± 1.03^{aA}	-	-	14.48± 0.33^{aA}
	C	13.43± 0.40^{A}	13.43± 0.40^{A}	13.43± 0.40^{A}	13.43± 0.40^{A}	75.90± 1.81^{A}	75.90± 1.81^{A}	75.90± 1.81^{A}	75.90± 1.81^{A}	0.218± 0.004^{A}	0.218± 0.004^{A}	0.218± 0.004^{A}	0.218± 0.004^{A}	15.12± 0.35^{A}	15.12± 0.35^{A}	15.12± 0.35^{A}	15.12± 0.35^{A}
G	HB	14.60± 0.24^{aA}	12.68± 0.18^{bA}	15.34± 0.22^{aA}	14.00± 0.28^{abA}	80.12± 0.86^{aA}	79.37± 1.73^{aA}	77.70± 0.67^{aA}	80.05± 0.98^{aA}	0.205± 0.005^{bA}	0.212± 0.010^{bA}	0.229± 0.005^{aA}	0.207± 0.004^{bA}	13.82± 0.48^{bA}	15.93± 0.46^{aA}	14.98± 0.49^{bcA}	14.96± 0.36^{bcA}
	MD	13.53± 0.46^{aA}	-	-	13.93± 0.39^{aA}	78.05± 1.16^{aA}	-	-	78.42± 0.46^{aA}	0.218± 0.008^{aA}	-	-	0.215± 0.004^{aA}	14.53± 0.36^{bA}	-	-	15.94± 0.50^{aAB}
	C	13.35± 0.35^{A}	13.35± 0.35^{A}	13.35± 0.35^{A}	13.35± 0.35^{A}	79.43± 1.67^{A}	79.43± 1.67^{A}	79.43± 1.67^{A}	79.43± 1.67^{A}	0.209± 0.004^{A}	0.209± 0.004^{A}	0.209± 0.004^{A}	0.209± 0.004^{A}	14.05± 0.33^{A}	14.05± 0.33^{A}	14.05± 0.33^{A}	14.05± 0.33^{A}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

^{A,B} Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers. MD= Muscovy Ducks. C= control. B =Brown, and G= Golden.

Table (6): Continued.

Traits		Yolk weight (%)				Yolk index				Albumen weight (%)			
Breeds	Type / Source	DNA		Gene		DNA		Gene		DNA		Gene	
		20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl
B	HB	32.01 ± 0.96 $^{a\lambda}$	34.08 ± 0.49 $^{a\lambda}$	33.57 ± 1.42 $^{a\lambda}$	33.03 ± 1.10 $^{a\lambda}$	0.467 ± 0.007 $^{a\lambda}$	0.507 ± 0.018 $^{a\lambda}$	0.485 ± 0.010 $^{a\lambda}$	0.460 ± 0.012 $^{a\lambda}$	53.12 ± 1.15 $^{a\lambda}$	52.88 ± 1.20 $^{a\lambda}$	51.31 ± 1.91 $^{a\lambda}$	54.45 ± 1.18 $^{a\lambda}$
	MD	32.87 ± 0.08 $^{a\lambda}$	-	-	32.41 ± 0.53 $^{a\lambda}$	0.487 ± 0.008 $^{a\lambda\lambda}$	-	-	0.478 ± 0.010 $^{a\lambda\lambda}$	51.81 ± 0.98 $^{a\lambda}$	-	-	53.11 ± 0.56 $^{a\lambda}$
	C	31.16 ± 1.05 $^{\lambda}$	31.16 ± 1.05 $^{\lambda}$	31.16 ± 1.05 $^{\lambda}$	31.16 ± 1.05 $^{\lambda}$	0.498 ± 0.012 $^{\lambda\lambda}$	0.498 ± 0.012 x	0.498 ± 0.012 $^{\lambda}$	0.498 ± 0.012 $^{\lambda\lambda}$	53.72 ± 1.01 $^{\lambda}$	53.72 ± 1.01 $^{\lambda}$	53.72 ± 1.01 $^{\lambda}$	53.72 ± 1.01 $^{\lambda}$
G	HB	32.41 ± 0.83 $^{a\lambda}$	33.74 ± 0.72 $^{a\lambda}$	32.98 ± 0.67 $^{a\lambda}$	34.11 ± 1.19 $^{a\lambda}$	0.462 ± 0.008 $^{a\lambda}$	0.501 ± 0.026 $^{a\lambda}$	0.491 ± 0.021 $^{a\lambda}$	0.468 ± 0.007 $^{a\lambda}$	53.78 ± 0.77 $^{a\lambda}$	50.34 ± 1.07 $^{a\lambda}$	52.04 ± 0.70 $^{a\lambda}$	50.93 ± 1.37 $^{a\lambda}$
	MD	31.73 ± 1.03 $^{a\lambda}$	-	-	32.82 ± 0.73 $^{a\lambda}$	0.468 ± 0.012 $^{a\lambda}$	-	-	0.478 ± 0.010 $^{a\lambda}$	53.74 ± 1.24 $^{a\lambda}$	-	-	51.24 ± 1.01 $^{a\lambda}$
	C	31.96 ± 1.33 $^{\lambda}$	31.96 ± 1.33 $^{\lambda}$	31.96 ± 1.33 $^{\lambda}$	31.96 ± 1.33 $^{\lambda}$	0.479 ± 0.009 $^{\lambda}$	0.479 ± 0.009 $^{\lambda}$	0.479 ± 0.009 $^{\lambda}$	0.479 ± 0.009 $^{\lambda}$	54.00 ± 1.51 $^{\lambda}$	54.00 ± 1.51 $^{\lambda}$	54.00 ± 1.51 $^{\lambda}$	54.00 ± 1.51 $^{\lambda}$

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{\lambda\lambda}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers. MD= Muscovy Ducks. C= control. B =Brown. and G= Golden.

The inoculation MD DNA_{20ng} significantly ($P< 0.05$) improved albumen weight % of Golden quail from (54.97%) compared to MD $gene_{40ng}$ (52.96%); HB DNA_{40ng} and HB $gene_{20ng}$ significantly ($P< 0.05$) difference (55.01 &54.96%) compared to HB DNA_{20ng} (52.81%), respectively (Table 8).

Egg yolk weight percentage and yolk index:

Wide range of egg yolk weight percentage and yolk index of the different treated groups subjected to DNA or gene inoculation was observed in Tables (7 and 8). Quail strain effect was only cleared ($P < 0.05$) for egg yolk weight percentage on the average, where transgenic Golden quails (31.47%) super passed Brown ones (30.15%). Moreover, MD $gene_{40ng}$ significantly ($P< 0.05$) difference egg yolk weight % of Golden quails from (32.64%) compared to the MD DNA_{20ng} and control group (30.29%) and (30.73%), respectively. HB $gene_{40ng}$ significantly ($P< 0.05$) difference egg yolk weight % of Golden quails (32.03%) compared to the control (30.73%), respectively. In Golden quail, MD_{40ng} gene has increased the egg yolk index compared to the control group (0.516 & 0.508), respectively.

Egg shell weight percentage, egg shape index and shell thickness:

The mean values of egg shell weight percentage, egg shape index and shell thickness of different DNA and gene treatments are shown in Tables (7 and 8). Inoculated HB gene at levels (40 or 20ng), MD $gene_{40ng}$ or HB DNA_{20ng} and control group significantly ($P< 0.05$) increased egg shape index of Brown quail (79.47, 77.52, 78.63 and 76.81) compared to the HB DNA_{40ng} (70.78), respectively. In addition, in Golden quail, inoculated HB DNA_{20ng} significantly increased egg shell weight percentage compared to HB DNA_{40ng} and HB gene (20 or 40ng) (15.65 vs. 13.24, and 14.25, 14.12), respectively.

Transgenic Response:

Responses for egg production trait due to transgenesis for two generations with (HB) or (MD) DNA or gene sources are listed in (Table 9). Positive response was detected in egg number, of (HB gene) and (MD) DNA for transgenic Brown quail (2.25 and 6.30, respectively). On the other hand, Positive response was detected in egg mass of HB gene and MD DNA for transgenic Brown quail (18.72 and 48.23, respectively). Moreover, Positive response was detected in egg number of (MD) DNA and gene for transgenic Golden quail (32.24 & 28.44). The corresponding value for rate of laying percentage was 8.56 and 10.65 of both (MD) DNA, and gene respectively for transgenic Golden quail. Positive response in the average egg weight for MD gene (0.77) on Golden quail was observed as well. Negative response was detected in egg number, egg laying rate, egg mass and average egg weight for all Brown and Golden treated quails (Table 9).

Table (7): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs. Duck) and type (DNA vs. Gene) on egg quality during 2nd generation.

Traits		Egg weight (grm)			Egg shape index			Shell thickness (mm)			Shell weight (%)		
Breeds	Source / Type	HB	MD	C	HB	MD	C	HB	MD	C	HB	MD	C
B	DNA	12.75 ± 0.24 ^{bx}	13.25 ± 0.21 ^{bcx}	13.87 ± 0.20 ^{acx}	73.80 ± 0.86 ^{bx}	78.69 ± 0.97 ^{ax}	78.21 ± 0.70 ^{ax}	0.189 ± 0.004 ^{ax}	0.201 ± 0.003 ^{ax}	0.195 ± 0.002 ^{ax}	14.47 ± 0.45 ^{ax}	14.83 ± 0.36 ^{ax}	14.35 ± 0.25 ^{ax}
	Gene	13.62 ± 0.26 ^{ax}	13.96 ± 0.24 ^{ax}	13.87 ± 0.20 ^{ax}	78.50 ± 0.84 ^{ax}	78.63 ± 0.50 ^{ax}	78.21 ± 0.70 ^{ax}	0.193 ± 0.003 ^{ax}	0.200 ± 0.002 ^{ax}	0.195 ± 0.002 ^{ax}	14.58 ± 0.48 ^{ax}	15.05 ± 0.32 ^{ax}	14.35 ± 0.25 ^{ax}
	Average	13.18 ± 0.18 ^{bcZ}	13.60 ± 0.19 ^{acZ}	13.87 ± 0.11 ^{aZ}	76.15 ± 0.62 ^{bZ}	78.66 ± 0.44 ^{aZ}	78.21 ± 0.70 ^{aZ}	0.191 ± 0.003 ^{bZ}	0.201 ± 0.002 ^{acZ}	0.195 ± 0.002	14.53 ± 0.33 ^{aZ}	14.94 ± 0.25 ^{aZ}	14.36 ± 0.25 ^{aZ}
G	DNA	13.64 ± 0.18 ^{ax}	13.73 ± 0.24 ^{ax}	13.76 ± 0.20 ^{ax}	78.90 ± 0.48 ^{ax}	78.13 ± 0.62 ^{ax}	80.30 ± 0.97 ^{ax}	0.195 ± 0.003 ^{ax}	0.199 ± 0.003 ^{ax}	0.194 ± 0.003 ^{ax}	14.44 ± 0.41 ^{ax}	14.74 ± 0.39 ^{ax}	14.97 ± 0.31 ^{ax}
	Gene	13.89 ± 0.26 ^{ax}	14.16 ± 0.17 ^{ax}	13.76 ± 0.20 ^{ax}	79.06 ± 0.72 ^{ax}	78.78 ± 0.64 ^{ax}	80.30 ± 0.97 ^{ax}	0.193 ± 0.003 ^{ax}	0.192 ± 0.002 ^{ax}	0.194 ± 0.003 ^{ax}	14.19 ± 0.21 ^{ax}	14.40 ± 0.30 ^{ax}	14.97 ± 0.31 ^{ax}
	Average	13.76 ± 0.15 ^{aW}	13.94 ± 0.14 ^{aZ}	13.76 ± 0.11 ^{aZ}	78.80 ± 0.41 ^{aW}	78.46 ± 0.47 ^{acZ}	80.31 ± 0.97 ^{abW}	0.194 ± 0.002 ^{aZ}	0.195 ± 0.002 ^{aZ}	0.194 ± 0.003 ^{aZ}	14.31 ± 0.25 ^{aZ}	14.57 ± 0.24 ^{aZ}	14.97 ± 0.31 ^{aZ}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{x-y}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

$^{Z-W}$ Means within the same column, between Average, having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden

Table (7): Continued

Traits		Yolk weight (%)			Yolk index			Albumen weight (%)		
Breeds	Source / Type	HB	MD	C	HB	MD	C	HB	MD	C
B	DNA	31.54 ± 0.55 ^{aX}	30.80 ± 0.70 ^{aX}	31.74 ± 0.42 ^{aX}	0.517 ± 0.011 ^{aX}	0.503 ± 0.08 ^{aX}	0.495 ± 0.005 ^{aX}	54.00 ± 0.67 ^{aX}	54.37 ± 0.63 ^{aX}	53.91 ± 0.43 ^{aX}
	Gene	31.15 ± 0.50 ^{aX}	29.49 ± 0.43 ^{bX}	31.74 ± 0.42 ^{aX}	0.507 ± 0.008 ^{aX}	0.511 ± 0.006 ^{aX}	0.495 ± 0.005 ^{aX}	54.27 ± 0.48 ^{aX}	55.45 ± 0.58 ^{abX}	53.91 ± 0.43 ^{acX}
	Average	31.35 ± 0.36 ^{aZ}	30.15 ± 0.37 ^{aW}	31.74 ± 0.42 ^{aZ}	0.512 ± 0.007 ^{aZ}	0.507 ± 0.005 ^{aZ}	0.495 ± 0.005 ^{aZ}	54.13 ± 0.40 ^{aZ}	54.91 ± 0.45 ^{aZ}	53.91 ± 0.43 ^{aZ}
G	DNA	31.65 ± 0.46 ^{aX}	30.29 ± 0.39 ^{aX}	30.73 ± 0.48 ^{aX}	0.504 ± 0.005 ^{aX}	0.500 ± 0.007 ^{aX}	0.508 ± 0.007 ^{aX}	53.91 ± 0.62 ^{aX}	54.97 ± 0.63 ^{aX}	54.29 ± 0.48 ^{aX}
	Gene	31.41 ± 0.52 ^{aX}	32.64 ± 0.49 ^{abX}	30.73 ± 0.48	0.517 ± 0.011 ^{aX}	0.516 ± 0.007 ^{bcX}	0.508 ± 0.007 ^{acX}	54.41 ± 0.51 ^{aX}	52.96 ± 0.59 ^{bX}	54.29 ± 0.48 ^{aX}
	Average	31.53 ± 0.34 ^{aZ}	31.47 ± 0.39 ^{aZ}	30.73 ± 0.48 ^{aZ}	0.501 ± 0.004 ^{aZ}	0.508 ± 0.005 ^{aZ}	0.508 ± 0.007 ^{aZ}	54.91 ± 0.42 ^{aZ}	53.96 ± 0.47 ^{aZ}	54.29 ± 0.48 ^{aZ}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{X-Y}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

$^{Z-W}$ Means within the same column, between Average, having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (8): Least square means ± standard errors for the effect of Japanese quail breed, nucleic acid source (Broiler vs. Duck), type (DNA vs. Gene) and levels (20 vs. 40) on egg quality during 2nd generation.

Traits		Egg weight (grm)				Egg shape index				Albumen weight (%)			
Breeds	Type	DNA		Gene		DNA		Gene		DNA		Gene	
	Source	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl
B	HB	13.74 ± 0.21 $^{a\chi}$	11.75 ± 0.05 $^{b\chi}$	13.52 ± 0.41 $^{a\chi}$	13.72 ± 0.31 $^{a\chi}$	76.81 ± 0.76 $^{a\chi}$	70.78 ± 2.75 $^{b\chi}$	77.52 ± 0.94 $^{a\chi}$	79.47 ± 1.45 $^{a\chi}$	53.95 ± 0.75 $^{a\chi}$	54.04 ± 1.05 $^{a\chi}$	53.56 ± 0.46 $^{a\chi}$	54.98 ± 0.86 $^{a\chi}$
	MD	13.25 ± 0.20 $^{b\chi}$	-	-	13.96 ± 0.24 $^{a\chi}$	78.69 ± 0.97 $^{a\chi}$	-	-	78.63 ± 0.50 $^{a\chi}$	54.37 ± 0.63 $^{a\chi}$	-	-	55.45 ± 0.58 $^{a\chi}$
	C	13.87 ± 0.20 $^{\chi}$	13.87 ± 0.20 $^{\chi}$	13.87 ± 0.20 $^{\chi}$	13.87 ± 0.20 $^{\chi}$	78.21 ± 0.70 $^{\chi}$	78.21 ± 0.70 $^{\chi}$	78.21 ± 0.70 $^{\chi}$	78.21 ± 0.70 $^{\chi}$	53.91 ± 0.43 $^{\chi}$	53.91 ± 0.43 $^{\chi}$	53.91 ± 0.43 $^{\chi}$	53.91 ± 0.43 $^{\chi}$
G	HB	13.37 ± 0.20 $^{a\chi}$	13.91 ± 0.38 $^{a\chi}$	13.95 ± 0.47 $^{a\chi}$	13.82 ± 0.20 $^{a\chi}$	79.44 ± 0.53 $^{a\chi}$	78.36 ± 1.00 $^{a\chi}$	78.97 ± 1.01 $^{a\chi}$	79.15 ± 1.08 $^{a\chi}$	52.81 ± 0.82 $^{b\chi}$	55.01 ± 0.69 $^{a\chi}$	54.96 ± 0.72 $^{a\chi}$	53.85 ± 0.71 $^{b\chi}$
	MD	13.73 ± 0.24 $^{a\chi}$	-	-	14.16 ± 0.17 $^{a\chi}$	78.14 ± 0.62 $^{a\chi}$	-	-	78.78 ± 0.64 $^{a\chi}$	54.97 ± 0.63 $^{a\chi}$	-	-	52.96 ± 0.59 $^{b\chi}$
	C	13.76 ± 0.20 $^{\chi}$	13.76 ± 0.20 $^{\chi}$	13.76 ± 0.20 $^{\chi}$	13.76 ± 0.20 $^{\chi}$	80.31 ± 0.97 $^{\chi}$	80.31 ± 0.97 $^{\chi}$	80.31 ± 0.97 $^{\chi}$	80.31 ± 0.97 $^{\chi}$	54.29 ± 0.48 $^{\chi}$	54.29 ± 0.48 $^{\chi}$	54.29 ± 0.48 $^{\chi}$	54.29 ± 0.48 $^{\chi}$

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{\chi\gamma}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (8): Continued.

Traits		Shell thickness (mm)				Shell weight (%)				Yolk weight (%)				Yolk index			
Breeds	Type	DNA		Gene		DNA		Gene		DNA		Gene		DNA		Gene	
	Source	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl	20 ng/µl	40 ng/µl
B	HB	0.197± 0.005^{a}	0.180± 0.001^{a}	0.196± 0.005^{a}	0.191± 0.003^{a}	14.89± 0.50^{a}	14.05± 0.36^{a}	14.87± 0.73^{a}	14.29± 0.61^{a}	31.16± 0.61^{a}	31.92± 0.41^{a}	31.57± 0.68^{a}	30.72± 0.74^{a}	0.506± 0.012^{a}	0.529± 0.7^{a}	0.516± 0.012^{a}	0.497± 0.010^{a}
	MD	0.201± 0.003^{a}	-	-	0.200± 0.002^{a}	14.83± 0.36^{a}	-	-	15.06± 0.32^{a}	30.80± 0.70^{a}	-	-	29.49± 0.43^{a}	0.503± 0.008^{a}	-	-	0.511± 0.006^{a}
	C	0.195± 0.002^{a}	0.195± 0.002^{a}	0.195± 0.002^{a}	0.195± 0.002^{a}	14.35± 0.25^{a}	14.35± 0.25^{a}	14.35± 0.25^{a}	14.35± 0.25^{a}	31.74± 0.42^{a}	31.74± 0.42^{a}	31.74± 0.42^{a}	31.74± 0.42^{a}	0.495± 0.005^{a}	0.495± 0.005^{a}	0.495± 0.005^{a}	0.495± 0.005^{a}
G	HB	0.192± 0.003^{a}	0.197± 0.003^{a}	0.188± 0.004^{a}	0.197± 0.003^{a}	15.65± 0.49^{a}	13.24± 0.36^{b}	14.25± 0.37^{b}	14.12± 0.20^{b}	31.55± 0.58^{a}	31.76± 0.80^{a}	30.78± 0.69^{a}	32.03± 0.78^{a}	0.507± 0.006^{a}	0.501± 0.009^{a}	0.500± 0.008^{a}	0.496± 0.008^{a}
	MD	0.198± 0.003^{a}	-	-	0.192± 0.002^{a}	14.74± 0.39^{a}	-	-	14.40± 0.30^{a}	30.29± 0.38^{b}	-	-	32.64± 0.49^{a}	0.500± 0.007^{a}	-	-	0.516± 0.007^{a}
	C	0.194± 0.003^{A}	0.194± 0.003^{A}	0.194± 0.003^{A}	0.194± 0.003^{A}	14.97± 0.31^{A}	14.97± 0.31^{A}	14.97± 0.31^{A}	14.97± 0.31^{A}	30.73± 0.48^{A}	30.73± 0.48^{A}	30.73± 0.48^{A}	30.73± 0.48^{A}	0.508± 0.007^{A}	0.508± 0.007^{A}	0.508± 0.007^{a}	0.508± 0.007^{A}

$^{a-c}$ Means within the same row having different letters are significant different ($p \leq 0.05$).

$^{A-C}$ Means within the same column (Breed) having different letters are significant different ($p \leq 0.05$).

HB = Hubbard Broilers, MD= Muscovy Ducks, C= control, B =Brown, and G= Golden.

Table (9): Transgenic responses due to transgenesis at the 2 [nd] generation compared to the 1 [st] generation on egg trait.

Traits		Egg trait							
		Egg number		Rate of laying /45 days		Average weight		Egg mass(gram)	
Breed	source / Type	HB	MD	HB	MD	HB	MD	HB	MD
B	DNA	-6.75	+6.30	-14.55	+13.57	-1.40	-0.78	-129.69	+48.23
	GENE	+2.25	-6.75	+4.61	-15.88	-0.45	-0.97	+18.72	-114.16
	Average	-2.25	-0.45	-4.96	-1.16	-0.92	-0.88	-56.54	-32.33
G	DNA	-4.50	+4.05	-10.47	+8.56	-0.22	-0.49	-63.88	+32.24
	GENE	-11.70	+4.95	-26.20	+10.65	-0.53	+0.77	-187.20	+28.44
	Average	-8.10	+4.50	-18.33	+9.60	-0.37	+0.14	-116.28	+59.06

HB = Hubbard Broilers. MD= Muscovy Ducks. C= control. B =Brown, and G= Golden

.DISCUSSION

Although **Salter *et al.* (1999)** concluded that Japanese quail will not be useful in avian transgenic studies involving vertical transmission of avian leukosis viruses (ALV) retroviral vectors, methods such as transfection of the primordial germ cells (PGC), blastodermal cells, embryonic stem cells, and injection of cloned DNA into fertilized ova, now routine for mice with eventual application to large animals, will eventually prove to be useful in constructing transgenic birds **(Petitte *et al.*, 1993; Salter *et al.*, 1993; Sang *et al.*, 1993)**, perhaps using Japanese quail as a model system, confirmed by **Rabie *et al.* (2009)**. In this study, the transgenic quail population constructed by **Rabie *et al.* (2009)** has used as a model system to examine the egg production, and internal and external egg quality of this transgenic population.

In recent study, the egg production percentage in Brown quail which treated with MD DNA_{20ng} (90.74%) is superpassed the previous study **(Bamgartner 1994)** who stated that average annual laying intensity (per bird housed) in Estonian quail 86% with average mortality rate 7.4% for 0-412 days. Despite the fact that **Vali (2007), and Nestor and Bacon (1982)** reported that change in egg weight in quail paralleled those of increase ages, **Vali (2007)** found that egg weight at 175 days of age were significantly different from the other ages, but there were no any significant differencs at 70 and 115 days of ages. Also he found that the highest egg wegiht of Japanese quail 11.16g, that lower than the results of this study for the average of egg weight (13.12g) in Brown quail that inoculated with MD $gene_{40ng}$. In addition, **Minvielle *et al.*, (2000)** also reported egg weight after six months in four purebred and their reciprocal crosses, which there were significantly different among groups. Meanwhile, the mean egg weight observed in this study was higher than those reported by **Wilson *et al.*, (1961)** (9.81g); **Mohanty *et al.*, (1987)** (6.4g at 15 months of age; **Sachdev *et al.*, (1989)** (9.28g) and **Narayanankutty *et al.*, (1989)** (8.56-9.93g at 12 and 24 weeks of age. Such a wide variation in egg weight could be not only due to different lines of quails, their age, feeding and managemental practices, but due to the expression of the GH gene introduced from Muscovy duck. The results were not surprising; the Muscovy duck egg production caracteristics is famed by the high egg weight average (72±8g) **(Banga-Mboko *et al.*, (2007)**. Moreover, a gradual increase in egg weight with age was also reported by **Tiwari and Panda (1978), Yannakopoulos and Tserveni-Gousi (1986) and Nagarajan *et al.*, (1991).** Athough there were no effect of trangenisis on first generation on yolk weight, youlk weight percentage, and Albumen wight percentage, The effect of transgenisis was appeared in the G_2 brown quail treated by Md $gene_{40ng}$ which improved the albumen weight percentage (55.45%), and the same effect on golden quail on yolk weight percentage, egg yolk index. These results in contrast with **Kul and Seker, (2004)** who found that almost all internal quality traits of the egg were changed at the significant levels

depending on the change occurred in the egg weight with respect to the external quality traits of the egg. However, the yolk and shell rates changed opposite to the albumen ratio and albumen index.

At the first generation, the egg shape index hs been improved by 4.7% at the Brown quail that treated with HB $gene_{20ng}$. This stand out against the shell weight percentage that has decreased when the Brown quail treated with HB gene and HB DNA at the level of 40ng by 12.5, 14.3% respectively. Also, MD $gene_{40ng}$ by 13.5% compared to control group. In contrast, at the G_2, HB DNA_{20ng} increased shell weight percentage (15.65). these results draw a distinction with **González (1995)** who found that egg weight, egg length, youlk height and youlk index increased as quail aged while the egg shape index, and albumen height decreased at the end of experiment (39 weeks of age). Although there is no significant differences on shell thickness in brown quail due to treatments at the first generation, at the 2nd generation, the shell thiskness has been improved in Golden quail (0.229) by treated with HB $gene_{20ng}$. This results were superpassed the results obtained by **González (1995)** who found that highest values of shell thickness (0.201 mm) obtained at 17 weeks of age, indicated that a better quaity of egg was attained at the beginning of lay. **Nazligül *et al.*, (2001)** found that egg production, egg weight, and egg quality characteristics were affected by the age of the quail. As the quail age increased, egg weight, yolk and albumen weight and shell weight increased, while shell thickness decreased.

In general, the obtained results, in recent study, was in concurrence with **Sezer (2008)** who found that high positive genetic and phenotypic correlation among egg, yolk and albumen mass and negative correlations between yolk and albumen ratio were detected. This result indicates that selection for increased egg weight will increase both yolk and albumen weight, but an increase in the proportion of yolk will reduce the albumen part of egg.

Few research laboratories have access to facilities for raising and breeding birds that will allow them to exploit transgenic technologies fully. Establishment of centers where transgenic birds can be generated and bred could be of benefit to the research community. In conclusion, the results of this study suggested that the use of inoculation method which has been used to produce the transgenic quail expressed Muscovy Duck growth hormone gene **(Rabie *et al.*, 2009)** may be an effective method for production of transgenic birds, these findings have successfully confirmed in this study. Moreover, it is likely in the next few years that this method will be developed to a stage where it is feasible to apply it to answering many questions, not only in studies of development but in applied areas, for example the study of disease, investigation of quantitative traits and exploiting the potential for production of pharmaceutical proteins in eggs. It is also possible that this method may be successfully combined or

that a fresh approach may be taken to develop a technically feasible and efficient method for transgenesis in the chick.

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الملخص العربي

السمان النقلجيني : نحو تحسين انتاج البيض و جودة البيض

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أجريت هذه الدراسة لتقدير تأثير نقلجينية السمان الياباني على انتاج البيض و جودة البيض الخارجية و الداخلية خلال ٤٥ يوم الأولى من وضع البيض مثل النسبة المئوية لإنتاج البيض، وزن البيض، كتلة البيض،دليل شكل البيضة، النسبة المئوية لوزن قشرة البيض، سمك قشرة البيض، دليل الصفار، النسبة المئوية لوزن الصفار، النسبة المئوية لوزن البياض و كذلك قد قدرت درجة الإستجابة للنقلجينية.

العشيرة التجريبية التي استخدمت في هذه الدراسة بنيت على جيلين و التي وضحت بالتفصيل سابقا (ربيع وأخرون، ٢٠٠٩) و التي أنتجت من حقن أجزاء منقاه من الحامض النووي الديوكسي ريبوزي (الدنا) أو الجين - كجزء من جين هورمون النمو- من الغدة النخامية سواء كان مصدره من أمهات دجاج اللحم هابارد (HB) أو من البط المسكوفي (MD) عند مستويان من التركيز هما ٢٠، ٤٠ نانوجرام داخل الخلايا الجنينية لسلالتين من السمان الياباني (البني و الذهبي).

أوضحت النتائج أن الحقن بالجين الذي مصدره البط المسكوفي (٤٠ نانوجرام) ادى الى زيادة في عدد وكتلة البيض المحور (٣٣.٣ بيضه & ٤٣٦.٩ جم) في الجيل الأول بالمقارنه بالجين الذي مصدره أمهات دجاج اللحم هابارد (١٩.٨ بيضه &٢٤٠.١٧ جم). لوحظ هذا الإتجاه أيضا في الجيل الثاني لسلالة السمان الذهبي. وجد تباين أكثر بالسمان الذهبي نتيجه لعملية النقلجينية مقارنه بالسمان البني اثناء الجيلين الاول و الثاني. أدى الحقن بالدنا المأخوذ من البط المسكوفي الى زيادة الوزن النسبي لقشرة البيض لكلا من سلالتي السمان البني والذهبي خلال الجيل الاول (١٤.٩ & ١٥.٢٣ %) مقارنة بالدنا من امهات دجاج اللحم (١٣.٨٨ & ١٤.٩٢ %) على التوالي. بينما حقن الجين الماخوذ من البط المسكوفي بتركيز ٤٠ نانوجرام ادي الى زياده في الوزن النسبي لصفار البيض بالسمان الذهبي (٣٢.٦٤ %)، حقن الدنا من البط المسكوفي بتركيز ٢٠ نانوجرام زاد نسبة وزن الصفار (٣٠.٢٩ %) خلال الجيل الثاني.

إضافة الى أن السمان البني المعامل بالجين من أصل امهات دجاج اللحم هابارد أظهر استجابة موجبة للنقلجينات في عدد البيض و كتلة البيض بينما السمان الذهبي أظهر استجابة موجبة في عدد البيض لكلا من الدنا و الجين الذي مصدره البط المسكوفي.