

RESEARCH ARTICLE

Impact of Removal of Incubated Eggs or Replacing them with Dummy Eggs on the Behaviour and Performance of Egyptian Baladi Pigeons

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Abstract

This study aimed to investigate the impacts of egg removal or replacement with dummy eggs on behaviour, performance, and prolactin levels in Egyptian Baladi pigeons. Twenty pairs of mature Egyptian Baladi pigeons (12-18 months) were distributed into four groups (5 replicates each, one pair per replicate). Group 1 (G1): parents incubated two eggs without remove (control), G2: incubated eggs were removed and replaced by dummy faked eggs, G3: included the removal of 1st egg only after laying, and G4: each pair had two eggs removal after 2nd egg lay (incubate no eggs). Using a digital camera for recording and analysing the behaviour. The results showed that the most of maintenance behaviour (kinetic, ingestive, and body comfort behaviour) were higher in G1 and G2 than other groups. While resting behaviour (crouching and perching) increased in more stressed groups (G3 and G4) due to egg removal. Most of the courtship behaviour in breeding males and females increased in G3 and G4 as they began another egg cycle rapidly after egg removal. However, G1 and G2 showed greater reproductive behaviour after egg laying than other groups, because breeding parents in these groups had already incubated either natural or dummy eggs. Meanwhile, egg searching and peeping increased by egg removal in G3 and G4. The incubation period was prolonged in G2 compared with other groups, but egg laying interval increased in G1 compared with others as they had hatched eggs and reared squabs, so prolactin level increased in groups (G1 and G2) with incubated eggs. The current study proved that egg removal and/or replacement affected most of the maintenance and reproductive behaviour of Egyptian Baladi pigeons. In conclusion, egg removal techniques can be used as a method of breeding control for pigeons while egg replacement by faked eggs could help in artificial incubation techniques.

Keywords: Behaviour, Pigeon, Incubated egg, Dummy eggs, Removal eggs.

Introduction

Pigeons are monogamous birds. They reach to sexual maturity and form pairs, a female lays eggs, egg brooding by both parents to ensure the proper development of the embryo, successful hatching and birth of the chick [1]. The breeding season of *Columba livia* is long and lasting all year around but reproductive peaks occur

during the spring and summer, the breeding cycle includes non-breeding, courtship and mating, nest-site selection and nest-building, incubation and feeding the young [2,3].

Egg removal or replacement could influence the period of parent' egg laying cycle by prolonging or shortening it. Removing an egg can result in multiple

attempts (courtship displays and mounting) at breeding so the cycle may be shortened, whereas replacing the removed egg may delay such attempts and so increase reproduction costs for female. Egg quality in the next egg cycles could be affected by removal of incubated eggs [4]. Birds avoid regions where there is a lot of disturbance, they also move quickly in response to human presence, alter their flight patterns, and vary their incubation patterns [5]. Pigeons make several efforts to mate and lay new eggs, in response to the removal of their eggs. Considering that pigeons can quickly recover in the event of a clutch loss or sterility of eggs when freshly laid eggs were taken out and replaced with fake eggs, partners abandoned dummy-egg incubation after the standard incubation period, then proceeded to build their nests and court before laying new eggs, resulting in the 4-week cycle [2]. Egg replacement could help in the artificial incubation procedures to prevent parents from abandoning their nests by using dummy eggs after removing the natural ones [6]. To control pigeon breeding, it is usually preferable to add eggs (shake to destroy viability while leaving the shell intact) rather than remove or shatter the eggs because the birds will continue to incubate, delaying subsequent nesting and prolonging nest site occupation [7]. After around eighteen days of incubation, parents find that their eggs have not hatched, so they abandon the nest and eggs [8]. The second pattern of a delayed increase in prolactin

(PRL) occurs in many birds that lay smaller clutches and hatch altricial young. Elevated PRL is required to stay in contact with the eggs during brooding [9]. During courtship, the prolactin hormone level was marginally around the normal level, then the increase started seven days

after egg laying and continued to climb until hatching. The tactile and visual cues that the eggs and nest provide during egg incubation may be responsible for these variations in the prolactin levels in the blood [10]. It is suggested that prolactin regulates the brain's physiological and behavioural reactions to stress [11]. The aim of this study was to evaluate the effects of incubated egg removal or replacement procedures on breeding pigeon behaviour, performance, and prolactin levels.

Materials and methods

Birds and Management

This study was conducted after the approval of the Institutional Animal Care and Use Committee at the Faculty of Veterinary Medicine, Zagazig University (ZU-IACUC/2/F/381/2022).

Twenty pair of mature Egyptian Baladi pigeons, aged between 12-18 months (20 males and 20 females) were used. The pens in which the birds were housed permitted enrichment and a range of behaviours, including flying when feasible. Each pair was housed in open-style wooden cages that were 150 cm above the floor and externally suspended from walls. Each battery had a barrier at the edge that was 3 cm high to prevent the nest from slipping, and perch sites with a height of 120 cm were hung on the opposite wall of the divided boxes. The partitions within the boxes were limited to 4 cages in height and 4 cages in width. We gave each pair of pigeons two nests to raise their squabs in. To maintain a normal breeding cycle, both nests were used alternately for egg incubation and rearing their hatchlings [13]. The experimental home and all its equipment (feeders, waters, and nest bowl) were fumigated prior to the experiment [14]. Adult pairs were identified by permanent,

non-toxic colours and by tying contrasting colours of cloth at the tips of their wings. To designate the specific nest place for each pair, nests are numbered. According to Xie *et al.*, [15], the lighting system used a 16L: 8D lighting schedule, and the light intensity was 2.5 w/m [16]. Neon Lamb (30w) supplied artificial light. The average daily temperature was $23 \pm 2^{\circ}\text{C}$ with 60–75% relative humidity.

Pigeons were fed and drank ad-libitum. Twice a day, fresh water from a clean source was supplied. Two times a day, at 8 a.m. and 6 p.m., the feeders were refilled. For a maximum of seven days following hatching, additional noon feeding was necessary. The commercial pellet diet used to feed the pigeons was purchased from the El-HAYANI factory.

The ingredients of the pellets according to factory sheet were yellow corn, soybeans, sorghums, wheat bran, corn gluten, calcium carbonate lime powder, sodium chloride NaCl, a mixture of minerals, salts, and vitamins, methionine (D-L), and choline chloride. The diet's chemical composition was 17% crude protein, 29.5 % fat, 3.74 % crude fiber and metabolizable energy not less than 2750 k.cal/kg. Day after day, more grit was given. The composition of this mixture includes several safe granite gravels, minerals, trace elements, and vitamins (MN-ZN-FE-CU-I-SE-CO-CA-PH-NA, B1, B2, B6, B12, biotin, pantothenic acid, folic acid, and nicotinic acid).

Pigeons were vaccinated against paramyxovirus (PMV-1) and pigeon pox virus in compliance with Marlier and Vindevogel [17].

Experimental design

Twenty pairs of Baladi Egyptian breed pigeons were tested in the period between the managerial practices and the next egg

cycle for each pair (which took about 2 months), they were divided into four groups (5 replicates each, one pair per replicate). The first group (G1) incubated two eggs without removal (control). The second group (G2) was exposed to removal of their eggs and replaced by dummy faked eggs. The third group (G3) had removal of 1st egg only after laying (1st egg was removed immediately after laying without return and observed the effect of this removal on laying and incubating of 2nd egg by the parent through the egg cycle). The fourth group (G4) had removal of their two eggs which was done immediately after laying of 2nd egg and not returned to the parent, so the breeding pigeon had no incubated eggs.

In G2, the eggs were removed just after 2nd egg was laid, punctured with a hypodermic needle, and shaken vigorously. The resulting sterile eggs were used as dummy eggs [2,18]. We transferred natural eggs and replaced them with artificial dummy eggs of similar size and colour to record the effect of incubating dummy eggs on behaviour, performance, and hormonal level of the parent [6]. The two dummy eggs remained in the nest to be incubated until the parent abandoned the nest, after which, they were removed; if any dummy egg was broken it was replaced by another one through the incubation period.

Data Collection

Behavioural Observation

To record the behaviours of the birds, video cameras (DVR) were placed in pens room and connected to a computer in a different room. The behavioural patterns of each group were recorded using the focal sample technique [19]. Pigeons were expected to adapt to their new environment and food, to form pairs, and

maintain their capacity for reproduction and production during the first month of the trial. Twice a day, between 8 and 10 a.m. and 2 and 4 p.m., observations were recorded. In the period between the managerial practices and the next egg cycle for each pair, each pair was examined for 20 min every day. The duration and frequency of the following behaviours were recorded:

A-Maintenance behaviour:

1-Ingestive Behaviour [20]

- Feeding: The head of pigeon was inside the feeder, pecking at the feeder with short intervals, during which the bird adopted an upright posture, showed swallowing and beak movements, and then resumed pecking again.

- Foraging: birds pecking food from the ground

- Drinking: bird's head in contact with water. Each beak immersion and release from the water reservoir's spout was logged as a drinking bout.

2-Comfort Behaviour [21]

-Feather Preening: gentle rubbing the beak or lower limbs over or between the feathers.

-Other comfort behaviour: include wings and legs stretching, wing flapping, wing ruffling, wing and tail shacking, head shaking and foot pecking.

3-Kinetic Behaviour [22]

-Standing: both feet are in contact with the floor, no other body part is in contact.

-Walking: Bird is taking at least two steps.

-Running: bird in the process of steps without stopping.

4-Resting Behaviour [22]

- Crouching: lying or sitting with

breast on the floor with or without closed eyes.

- Perching (Alert immobility): the bird is in a peaceful upright waking posture, standing on two legs, blinking rapidly, but making no head or body exploring movements [20].

5-Aggressive Behaviour

- Attacking: active agonistic behaviour by pigeons against another one, like pecking, chasing, beating with wings and kicking with feet.

- Fighting: two pigeons actively engage in fighting with each other.

B) Reproductive Behaviour [23, 24]

-Male sexual activity: as head bowing, female driving and pushing.

-Female sexual activity: as pushing male with being driven and lay 2nd egg on another nest.

- Male and female sexual activity: as collection of nest material, nest building, reciprocal preening (mutual preening), nest defence, sitting on the new nest, egg sitting (natural incubating of the eggs), egg turning (turning the eggs on each side by the parent's beak or legs through the incubation period) and egg searching or peeping behaviour (i.e. angular or ballistic, stretching movements of the head and neck directed in all directions, occurring in the absence of locomotion, observed mainly while egg searching).

Reproductive Performance

- Incubation period: days that parent spent sitting on the eggs until hatching.

- Egg laying interval: the duration in days between the first egg laid and the first egg of the subsequent clutch [25].

-Egg weight (g): the egg weighted after its removal or replacement.

-Prolactine level:

Blood samples were taken from breeding females to measure the serum prolactin level. Blood samples were taken at the mid-incubation from females in the control and dummy egg groups, but in the other two groups, the samples were taken 24 h after the egg removal. Blood samples were taken from the wing vein into tubes devoid of anticoagulant in to acquire serum [3]. Precautions and procedures followed the guidelines provided by EL Shoukary *et al.* [24].

Data Analysis

Mean \pm the standard error of means (SEM) was used to express the data of the results. The fourth treatment groups' effects on the various behavioural patterns and performance metrics were evaluated using one-way analysis of variance (ANOVA) by Duncan multiple test as post hoc test and the T-test only was used mainly for prolactin level measurement. The value $p \leq 0.05$ was utilized as the threshold for statistical significance. The Statistical Package for Social Sciences version 24.0 (SPSS, IBM Corp., Armonk, NY) and Graph Pad Prism 8.0.2 (Graph

Pad Software, Inc.) were used for all analyses and charts [26].

Results

Effect of incubated egg removal or replacement on some of maintenance behaviour of breeding pigeons

Table (1) revealed that there was a significant impact of egg removal or replacement on the kinetic behaviour (time and frequency) of breeding pigeons. Standing (duration and frequency), running (duration and frequency) and flying duration were significantly ($p \leq 0.05$) decreased in treatment groups (G2, G3, and G4) in comparison with the control group. Furthermore, a non-significant decrease in walking behaviour (time and frequency) in treatment groups was recorded.

Moreover, resting behaviour was significantly ($p \leq 0.05$) affected by egg removal or replacement procedures. The duration and frequency of perching and crouching behaviour showed the highest values in G4 (303.54 ± 25.93 sec and 10.17 ± 0.98 bouts, 80.08 ± 24.25 sec and 2.42 ± 0.73 bouts; respectively) compared to G1, G2 and G3.

Table (1): Effect of incubated egg removal or egg replacement on duration and frequency of kinetic and resting behaviour of breeding pigeons (Mean/20min \pm SE).

Item	G1	G2	G3	G4	P value
Standing duration (sec)	93.80 ± 10.07^a	53.75 ± 8.79^b	58.58 ± 13.0^b	51.58 ± 7.82^b	0.005
Standing frequency	16.27 ± 1.64^a	7.10 ± 1.28^b	7.92 ± 1.60^b	6.38 ± 0.89^b	0.00
Walking duration (sec)	52.85 ± 8.73	28.02 ± 5.94	43.96 ± 10.81	32.63 ± 8.46	0.11
Walking frequency	9.98 ± 1.52	6.06 ± 1.24	6.96 ± 1.60	5.54 ± 1.189	0.11
Flying duration(sec)	3.37 ± 0.72^a	3.00 ± 1.12^{ab}	0.85 ± 0.26^b	2.54 ± 0.68^{ab}	0.03
Flying frequency	2.82 ± 0.60	2.71 ± 1.10	0.81 ± 0.25	2.42 ± 0.67	0.06

Running duration (sec)	1.85 ± 0.645 ^a	0.23 ± 0.12 ^b	0.13 ± 0.13 ^b	0.00 ± 0.00 ^b	0.02
Running frequency	0.38 ± 0.14 ^a	0.08 ± 0.04 ^{ab}	0.04 ± 0.04 ^{ab}	0.00 ± 0.00 ^b	0.03
Perching duration (sec)	172.43 ± 26.14 ^b	201.33 ± 34.13 ^b	208.52 ± 30.48 ^b	303.54 ± 25.93 ^a	0.04
Perching frequency	5.08 ± 0.79 ^b	6.27 ± 0.93 ^b	6.75 ± 1.16 ^b	10.17 ± 0.98 ^a	0.007
Crouching duration (sec)	63.17 ± 14.82 ^b	28.42 ± 15.3 ^d	74.79 ± 18.1 ^{ab}	80.08 ± 24.25 ^a	0.003
Crouching frequency	2.25 ± 0.52 ^b	1.33 ± 0.21 ^c	2.30 ± 0.55 ^a	2.42 ± 0.73 ^a	0.008

^{abc} Means within the same row carrying different superscripts are significantly different at $P \leq 0.05$ based on Duncan multiple significant difference test. G1: control group, G2: dummy egg group, G3: remove 1st egg only after laying, G4: remove all egg after laying.

The ingestive behaviour mainly feeding and drinking (duration and frequency) were higher in the control group (16.90 ± 5.92 sec, 1.22 ± 0.38 bouts and 3.50 ± 1.30 sec, 0.67 ± 0.27 bouts, respectively) without significance.

Meanwhile, the highest foraging behaviour (duration and frequency) were detected in the dummy egg group (114.56 ± 26.45 sec, 5.92 ± 1.28 bouts) followed by the control group and the lowest values were noted for G3 and G4 (Table 2).

Table (2): Effect of incubated egg removal or replacement on duration and frequency of ingestive behaviour of breeding pigeons (Mean/20min ± SE).

Item	G1	G2	G3	G4	P value
Feeding duration(sec)	16.90±5.92	11.02 ± 4.07	4.42 ± 2.995	6.42 ± 3.37	0.37
Feeding frequency	1.22 ± 0.38	0.67 ± 0.21	0.29 ± 0.18	0.54 ± 0.22	0.22
Drinking duration(sec)	3.50 ± 1.30	3.15 ± 0.95	2.71 ± 1.39	1.75 ± 1.18	0.82
Drinking frequency	0.67 ± 0.27	0.63 ± 0.20	0.50 ± 0.25	0.29 ± 0.21	0.78
Foraging duration(sec)	91.27 ± 16.20	114.56 ± 26.45	56.33 ± 16.51	49.13 ± 16.12	0.17
Foraging frequency	6.90 ± 1.07	5.92 ± 1.28	3.21 ± 0.96	3.67 ± 0.97	0.13

^{abc} Means within the same row carrying different superscripts are sig. different at $P \leq 0.05$ based on Duncan multiple Significant Difference test.

As depicted in Table (3), incubated egg removal or replacement had a prominent effect on the comfort behaviour of breeding pigeons. The preening frequency was significantly higher in the control

group (14.12 ± 0.99 bouts) compared to G2 (10.23 ± 0.86 bouts), G3 (8.33 ± 0.97 bouts), and G4 (8.04 ± 1.07 bouts). Additionally, the preening duration was higher in G1 than the other without

significance level. A decrease of other comfort behaviour (time and frequency) with significance ($p \leq 0.05$) was observed in treated groups more than in the control

group. Aggressive behaviour (fighting and attacking) was increased without significance in G2, G3, and G4 than the control group.

Table (3): Effect of incubated egg removal or replacement on duration and frequency of body comfort and aggressive behaviour of breeding pigeons (Mean/20min \pm SE)

Item	G1	G2	G3	G4	P value
Preening duration(sec)	213.06 \pm 21.88	202.58 \pm 19.99	159.58 \pm 22.41	173.04 \pm 23.70	0.39
Preening frequency	14.12 \pm 0.99 ^a	10.23 \pm 0.86 ^b	8.33 \pm 0.97 ^b	8.04 \pm 1.07 ^b	0.00
Other body* Comfort duration (sec)	21.03 \pm 1.60 ^a	8.88 \pm 0.95 ^b	7.29 \pm 1.58 ^b	7.13 \pm 1.06 ^b	0.00
Other body* Comfort frequency	10.45 \pm 0.76 ^a	3.94 \pm 0.40 ^b	2.38 \pm 0.37 ^b	2.67 \pm 0.29 ^b	0.00
Attacking duration(sec)	0.92 \pm 0.39	1.30 \pm 0.72	2.42 \pm 0.92	1.54 \pm 0.89	0.61
Attacking frequency	0.29 \pm 0.12	0.62 \pm 0.36	0.50 \pm 0.20	0.33 \pm 0.17	0.81
Fighting duration(sec)	0.00 \pm 0.00	0.33 \pm 0.33	0.79 \pm 0.61	0.52 \pm 0.19	0.39
Fighting frequency	0.00 \pm 0.00	0.03 \pm 0.03	0.13 \pm 0.09	0.29 \pm 0.12	0.18

^{abc} Means within the same row carrying different superscripts are sig. different at $P \leq 0.05$ based on Duncan multiple Significant Difference test, (*) other body comfort including: wings and legs stretching, Wing flapping, Wing ruffling, Wing and tail shacking, Head shaking and Foot pecking.

Table (4) revealed that the courtship or foreplay behaviour of males (including head bowing and pushing female) was significantly higher in groups of egg removal without returning eggs (G3 and G4) than in groups of egg incubated either true or faked eggs (G1 and G2, respectively), while the difference of driving and circling of male to female was not significant among the groups.

A non-significant increase of pushing males by females was demonstrated in G3 and G4 compared to G1 and G2. Furthermore, egg laying outside the nest was markedly observed in G3 (0.50 \pm 0.35 sec, 0.08 \pm 0.06bout) rather than the other groups.

An obvious impact of incubated egg removal or replacement on the most of male and female sexual activities studied is shown in Table (4). Nest material collection and nest building were higher in G1 and G2 than in G3 and G4. The highest duration and frequency of nest defence (3.69 \pm 1.27 sec, 0.94 \pm 0.32 bouts), egg sitting (372.27 \pm 61.17 sec, 12.44 \pm 2.10 bouts) and egg turning (5.44 \pm 1.42 sec, 10.54 \pm 9.52 bouts) were observed in G2 in comparison with the other groups.

Regarding mutual preening behaviour, it was increased significantly in males and females of losing egg groups (G3 and G4) while G4 demonstrated the highest values of time and frequency (51.21 \pm 8.04 sec and 4.08 \pm 0.62 bout, respectively)

followed by G3 (22.00 ± 7.01 and 1.46 ± 0.43 bout, respectively).

Moreover, sitting of parents on the new nest (time and frequency) was significantly greater in G4 (145.46 ± 40.83 sec, 12.25 ± 7.15 bout) followed by

G3 (133.92 ± 39.13 sec, 4.46 ± 1.22 bout) than other groups. Additionally, searching for eggs or peeping behaviour by males and females was significantly greater in G3 and G4 than in G2, while in G1, parents did not record a search for eggs.

Table (4): Effect of incubated egg removal or replacement on duration and frequency of reproductive behaviour of breeding pigeons (Mean/20min \pm SE)

Item		G1	G2	G3	G4	P value
Male courtship behaviour						
Head bowing	Duration	0.23 ± 0.17^b	0.25 ± 0.25^b	6.46 ± 3.70^a	5.63 ± 3.59^a	0.01
	Frequency	0.05 ± 0.04^b	0.02 ± 0.02^b	0.58 ± 0.31^a	0.46 ± 0.27^a	0.008
Driving female	Duration	1.95 ± 1.95	0.40 ± 0.29	0.71 ± 0.71	2.96 ± 1.30	0.71
	Frequency	0.18 ± 0.18	0.06 ± 0.05	0.08 ± 0.08	0.25 ± 0.12	0.84
pushing female	Duration	0.00 ± 0.00^b	0.00 ± 0.00^b	0.33 ± 0.33^{ab}	0.50 ± 0.35^a	0.05
	Frequency	0.00 ± 0.00^b	0.00 ± 0.00^b	0.04 ± 0.04^{ab}	0.08 ± 0.06^a	0.04
Female courtship behaviour						
Pushing male	Duration	0.20 ± 0.14	0.42 ± 0.30	0.71 ± 0.42	0.96 ± 0.74	0.56
	Frequency	0.03 ± 0.02	0.06 ± 0.05	0.38 ± 0.33	0.21 ± 0.15	0.22
Being driven	Duration	0.43 ± 0.28	0.25 ± 0.25	0.00 ± 0.00	0.20 ± 0.20	0.72
	Frequency	0.08 ± 0.05	0.04 ± 0.04	0.00 ± 0.00	0.13 ± 0.13	0.64
Lay 2nd egg on another nest	Duration	0.00 ± 0.00^b	0.00 ± 0.00^b	0.50 ± 0.35^a	0.00 ± 0.00^b	0.01
	Frequency	0.00 ± 0.00^b	0.00 ± 0.00^b	0.08 ± 0.06^a	0.00 ± 0.00^b	0.01
Male and female reproductive behaviour						
Collecting nest materials	Duration	5.96 ± 2.81	3.55 ± 2.10	3.02 ± 1.96	2.25 ± 1.24	0.80
	Frequency	0.48 ± 0.27	0.29 ± 0.18	0.20 ± 0.14	0.13 ± 0.07	0.77
Nest building	Duration	25.93 ± 5.27^a	8.79 ± 3.59^b	5.17 ± 1.59^c	3.92 ± 1.93^d	0.001
	Frequency	3.83 ± 0.77	2.4 ± 1.49	0.58 ± 0.17	0.46 ± 0.23	0.10
Mutual preening duration	Duration	8.95 ± 3.81^b	9.25 ± 2.99^b	22.00 ± 7.01^b	51.21 ± 8.04^a	0.00
	Frequency	0.88 ± 0.35^b	0.60 ± 0.19^b	1.46 ± 0.43^b	4.08 ± 0.62^a	0.00
Nest Defending	Duration	0.73 ± 0.44^b	3.69 ± 1.27^a	0.33 ± 0.33^b	0.21 ± 0.21^b	0.01
	Frequency	0.25 ± 0.13^b	0.94 ± 0.32^a	0.04 ± 0.04^b	0.04 ± 0.04^b	0.01
Egg Sitting	Duration	258.35 ± 42.43^{ab}	372.27 ± 61.17^a	93.96 ± 36.69^b	112.33 ± 41.31^b	0.001
	Frequency	9.45 ± 1.68^a	12.44 ± 2.10^a	0.00 ± 0.00^b	$2.58 \pm .998^b$	0.001
Egg turning and checking	Duration	$3.60 \pm .996^{ab}$	5.44 ± 1.42^a	0.38 ± 0.38^b	0.43 ± 0.43^b	0.006
	Frequency	0.58 ± 0.16	10.54 ± 9.52	0.04 ± 0.04	0.05 ± 0.05	0.464
Sitting on new nest	Duration	50.45 ± 18.12^c	60.50 ± 20.08^{bc}	133.92 ± 39.13^{ab}	145.46 ± 40.83^a	0.03
	Frequency	1.92 ± 0.67^b	1.85 ± 0.62^b	4.46 ± 1.22^b	12.25 ± 7.15^a	0.02
Egg searching	Duration	0.00 ± 0.00^b	0.25 ± 0.25^b	2.67 ± 1.12^b	5.50 ± 2.48^a	0.00
	Frequency	0.00 ± 0.00^b	0.08 ± 0.08^b	0.46 ± 0.19^a	0.63 ± 0.29^a	0.001

^{abc} Means within the same row carrying different superscripts are sig. different at $P \leq 0.05$ based on Duncan multiple Significant Difference test.

Effect of incubated egg removal or replacement on reproductive performance of breeding pigeons

Table (5) showed the incubation period in the first egg cycle during egg removal or replacement. Pigeons in G1 normally brooded eggs for (17.33 days) and parents in G2 incubated the eggs for (20.33 days), so the incubation period in G1 had its normal cycle and eggs hatched after (17.33 ± 0.21) days, while in G2, the incubation period (20.33 days) was prolonged than the known period with a

significant level. On the opposite, G3 recorded no incubation for the eggs. Egg laying intervals were increased significantly ($p \leq 0.05$) in G1 (33.67 days) followed by G2 (28.67 days) while G3 and G4 recorded the lowest egg laying interval (10.33 and 12.67 days, respectively).

Egg weight didn't differ significantly between control and egg removal or replacement groups but it recorded the lowest weight in G3.

Table (5): Effect of incubated egg removal or replacement on the performance of breeding pigeons (Mean ± SE).

Parameters	G1	G2	G3	G4	P value
Incubation period(d)	17.33 ± 0.21 ^b	20.33 ± 0.21 ^a	0.00 ± 0.00 ^d	3.00 ± 0.00 ^c	0.001
Egg weight(gm)	17.67 ± 0.33	16.33 ± 0.21	13.00 ± 2.61	16.50 ± 0.22	0.11
Egg laying interval(d)	33.67 ± 0.33 ^a	28.67 ± 0.33 ^b	10.33 ± 0.33 ^d	12.67 ± 0.33 ^c	0.03

^{abcd} Means within the same row carrying different superscripts are sig. different at $P \leq 0.05$ based on Duncan multiple Significant Difference test. G1: control group, G2: dummy egg group, G3: remove 1st egg only after laying, G4: remove all egg after laying, d (day), gm (gram).

Effect of incubated egg removal or replacement on prolactin level of breeding pigeons

As presented in Figure 1, prolactin levels could be affected by egg removal

more than egg replacement, as the prolactin levels in G1 and G2 were nearly the same (0.32 and 0.26 ng/mL, respectively). However, G3 and G4 had lower prolactin levels (0.03 and 0.14 ng/mL, respectively) than G1 and G2.

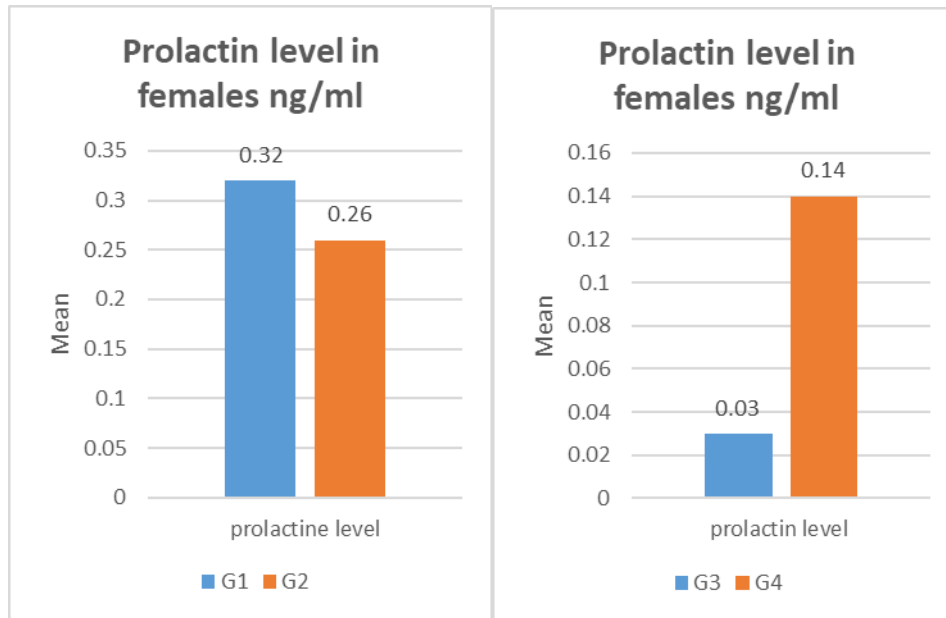


Figure (1): Effect of incubated egg removal or replacement on the level of prolactin in female. G1: control group, G2: dummy egg group, G3: remove 1st egg only after laying, and G4: remove all egg after laying.

Discussion

Incubated egg removal or replacement is considered one of the procedures that influence the behaviour (either maintenance or reproductive), performance, and prolactin level of breeding pigeons. The changes in kinetic behaviour observed in treated groups may be related to increased resting behaviour due to more fear and fear for safety, as the parents had no eggs to incubate, so they spent their days resting and searching for a safe nest site to lay the next eggs. This is inconsistent with Rasmussen *et al.* [27] which investigated that walking impairment and space limitation decreased in fear of broiler chickens.

Concerning the ingestive behaviour, it was observed that it declined with the removal of one incubated egg or both. This may be because G1 and G2 spent more time in egg incubation, so during their leaving nest time, they competed for the feeding and drinking sites more than others. Our results agreed with Hassanien [28] who found that one of the parents left

the nest to eat and drink while the other incubated the eggs to supply them with warmth and turning. Foraging was affected by spending more time on the nest, as in G2. This may be due to the parents spending more time inside the nest at the beginning of incubation to save and protect their eggs from losing again after replacement and at the end of incubation as eggs did not hatch on their own, so the parents foraged in the area around the nest site during their sitting period.

Preening and other comfort behaviours are signs of the welfare of birds. The decrease in comfort behaviour in treated groups compared to the control group indicated less welfare and increased stress in these groups than in the control group. In G1, the parents frequently left the nest when they felt safe about the eggs, followed by G2. In normal circumstances, one parent leaves the nest to preen and spread their wings while the other incubates the eggs to provide warmth [28]. In our study, the groups exposed to changing conditions by removing or

replacing eggs, became more violent (fighting and attacking) against others. This agreed with Wang *et al.* [8] who said that the parents show aggressive behaviours if they feel that their eggs are in danger.

Breeding parents (males and females) perform several attempts of breeding to start a new egg laying cycle mainly if they lose their eggs by removing without replacement [2, 29], So males and females' courtship behaviour increased by breaking the cycle, as the nesting cycle can be interrupted at any time after laying by removing the eggs or young, at this point courtship and nest construction started again [30].

Replacing the eggs with other dummy eggs allows the female to suspect eggs and stand for a period to check eggs before starting to incubate. Also, removing the first egg in G3 affected the laying of the second egg because the presence of the first egg on the nest stimulated the female and showed that the nest is safe to lay the second egg on it; hence, removing the first egg from the nest, as in G3, let the female suspect about the nest safety and lay the second egg forcedly in any other places, like other nests that were not prepared, on ground, or dropped while standing and damaged. The female of Rock pigeon (*Columba livia*) and Ring Dove will only lay two eggs, regardless of any external cues after egg removal trials. These birds don't change their clutch size in response to egg removal but exhibit behavioural alterations during nesting and incubation [31]. As breeding and nesting behaviour shaped the relationship between adults during incubation and adults with young during the rearing period [32]. Collecting nest material, nest building, and nest arranging are necessary for G1 and G2 as they needed to keep eggs warmer during

incubation and to protect the squabs from cold in G1 after egg hatching. As mentioned before, courtship between breeding parents increased by losing the eggs. Also, mutual preening by both males and females was observed in egg-losing groups for initiating the next breeding cycle.

The management practices have an impact on the breeding phenology of pigeons as the parents prolonged the period of egg sitting, egg turning, and nest defence. In G2 the period of eggs incubating was prolonged until they ensured that eggs would not hatch, so they incubated the eggs and defended the nest more than other groups. This may be attained when the eggs are replaced by dummy eggs; females may interrupt incubation and start a new cycle soon after the end of normal incubation. One possible scenario is that pairs abandon dummy egg incubation after the normal incubation duration (18 days) and then take an additional 2 weeks of nest building and courtship before laying new eggs [2].

Sitting on the new nest, nest exploration or demonstration, and additionally searching for eggs or peeping behaviour by male and female were significantly greater in the egg removal groups than the control. Generally, nest demonstration is more frequent in males and females at the beginning of the egg cycle [29]. Our results agreed with de Souza *et al* [33] who found that pigeon peeping behaviour could be expressed in potentially threatening or harmful situations. The parents searched for their eggs in the surrounding nest sites, but when they didn't find their eggs, they considered those nests unsafe for egg laying, and they took other nests for the next egg laying. In G2, the parents accepted the eggs (dummy) and incubated

them; when they did not hatch, they abandoned them, left the nest, and laid the next eggs in another new nest. Both parents shared in rearing the young (sitting on the young) and feeding squabs, mainly in groups that had fertile incubated eggs that hatched into squabs after the end of incubation [8].

The incubation period of the first egg cycle was significantly affected by egg removal or replacement by dummy eggs. The incubation period in G1 had its normal cycle and eggs hatched after the normal incubation period (17-18 days), while in G2, the incubation period was prolonged than the known period with a significant level. This may be due to the parents waiting the eggs to hatch. In accordance with these findings, sitting breeding pigeons on dummy eggs without recognition that they were empty of embryos, will help in applying artificial incubation to pigeons' eggs and saving the abandoned fertilized eggs by parents, also artificial incubation of the pigeon eggs allows the egg hatching to be faster than that in natural one. The embryonic development of pigeon *Columba livia* domesticus was shorter (17 days) by about 4 days in artificial incubation [34]. Due to total dependent feeding of newly hatched squabs on their parent's milk [35], they should be transferred to parents after removing the dummy incubated eggs, in addition to leaving some broken eggshells in the nests to make the parent sure that they were naturally hatched or deceived the foster parents to feed them.

In G3, breeding pigeons were abandoned or damaged the second egg after laid and moreover in G4, there were no eggs to be incubated after both eggs were removed but the parents still sat on the nest for about 24 h after the second egg laid before removal.

If the eggs were not removed and hatched, the parents reared the squabs for a period before the next egg laying; this will lead to prolonged egg laying intervals as in G1, while if eggs were replaced by dummy eggs, they took the normal period of incubation and may be extended. When the eggs were not hatched the parents abandoned them and started to prepare for the next egg cycle, so the length of the egg laying interval in G2 followed that of G1, but egg removal or losing without replacing them lead to several reproductive attempts from parents for the next egg laying cycle and a shortening of egg-laying cycles. Our finding is supported by those obtained in a previous study that egg-laying cycles were dependent on management practices and either egg removal or replacement [2].

The egg weight was not affected by egg removal or replacement procedures in G2 and G4, but some changes could be observed by the removal of the first eggs in G3, where the weight of the second egg decreased more than normal because the female was more stressed as the presence of the first egg on the nest stimulated her to normally lay the second one. Pigeon intraspecific variation in egg shape and size can result from many factors, for example differences in breeding habits [36].

Egg removal in G3 and G4 allowed the egg laying cycle to take its maximum range (about 10 – 12 days) as they didn't take any rest time from the first cycle, while in the dummy egg group, eggs did not hatch, and the parents left the nest and put eggs only after 8–9 days. This agreed with Cuthbert [30] who recorded that allowing the birds to incubate infertile eggs (dummy eggs) past the typical hatching time is another way to break the breeding cycle. Laying usually happens a few days after the desertion of the

nest rather than the 6–10 days period that is necessary if a removal is undertaken.

Moreover, egg quality in the next cycles had been affected by egg removal in the previous egg cycle as eggs became smaller, with some deformities at the top that were more narrow or infertile. This finding is similar to that obtained by Jacquin *et al.* [2] who declared that the multiple relaying caused by egg removal entails some energetic costs for females, leading to a decrease in individual condition and consequently egg quality. Also, egg harvesting led to a decline in the egg quality and hatching success of the replacement clutches in black-headed gulls [37]. So, egg removal techniques are used as a control of invasive pigeons in some places, which are heavily manifested. Egg removal acts as a method to control birth through regulating populations, as egg removal increases the reproduction cost of females and decreases hatching percentages in the next cycles [7, 38, 39].

The level of prolactin had been affected by egg removal or replacement mainly in the female pigeon. The prolactin level was near level points in control and dummy egg groups as females of both groups incubating the eggs (either true or fake). While in G3 and G4 females, the levels of prolactin decrease to a minimum level due to the absence or abundance of eggs. Similarly, a higher level of prolactin was in incubated females than in non-incubated or egg laying only ones [3]. Also, prolactin levels start to increase in pigeons after the full egg clutch is complete and starts incubation [9].

The lowest level of incubated female prolactin may be attributed to the loss of both eggs and leaving the nest without any eggs to be incubated. This is

supported with the findings of Jin *et al.* [40] that the level of prolactin decreased with nest deprivation.

Conclusions

Egg removal without replacement affected the maintenance behaviour of birds as it affected the bird's activity and viability, and most of the reproductive behaviour of males and females was directed to start a new egg cycle after breaking the previous one. Incubation period, egg laying interval, and prolactin level were decreased to the lowest level, but egg replacement by dummy eggs had maintenance and reproductive records similar to those of the controlled groups except that they had a prolonged incubation period.

Conflict of interest

The authors declare they have no conflict of interest.

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الملخص العربي

تأثير إزالة أو استبدال البيض المحضن على سلوك وأداء الحمام البلدي المصري

هيه سعيد عبد الرحمن غريب، إسراء هشام محمد سعد داؤد*، محمد يوسف ابراهيم ، الصادق خليل يوسف، وأسماء ابراهيم عبد العاطي قسم سلوكيات ورعايه الحيوان والدواجن والاحياء المائية – كلية الطب البيطري – جامعة الزقازيق – الزقازيق – 44511 – مصر. الهدف من هذه الدراسة هو تقييم إجراءات إزالة أو استبدال البيض المحضن فيما يتعلق بسلوك الحمام البلدي المصري اثناء التحضين، وأدائه ، ومستوى هرمون البرولاكتين. قد تم توزيع عشرين زوجا من الحمام البلدي المصري الناضج (12-18 شهرا) على أربع مجموعات (كل مجموعه تحتوي على خمس ازواج متماثله) ، المجموعه الاولى: يرقد الحمام البلدي المصري علي بيضتين دون إزالة (المجموعه الحاكمه)، والمجموعه الثانيه: أزيل البيض المحضن واستُعيض عنه ببيض مزيف وهمي، والمجموعه الثالثه: شمل ذلك إزالة البيضة الأولى فقط لكل زوج بعد وضعها، والمجموعه الرابعه : كل زوج تمت إزالة كلا من البيضتين بعد وضع البيضة الثانيه، وقد سُجِّل بعض السلوكيات المتعلقة بالبقاء والإنجاب لكل زوج من الوالدين باستخدام آلة تصوير رقمية تم تحليلها. وبالإضافة إلى ذلك، تم تقييم فترة الحضانه، وفترة وضع البيض، ومستوى البرولاكتين . أظهرت النتائج أن معظم السلوكيات المسئوله عن البقاء (السلوك الحركي والغذائي وسلوك الاراحة) ازدادت في المجموعه التي تحتضن بيض طبيعى (المجموعه الاولى) والمجموعه التي احتضنت بيض مزيف (المجموعه الثانيه) عن المجموعات الاخرى. في حين ازداد سلوك الجلوس والوقوف على الاعمدة جانبيه لقفص التحضين فى المجموعات التي تعاني مزيد من الخوف والتوتر بإزاله البيض (المجموعه الثالثه والرابعه). ازداد معظم سلوك المغازلة والمداعبة في الازواج التابعه للمجموعات الثالثه والرابعه حيث بدأوا بسرعة دورة بيض أخرى بعد إزالة البيض من العش، لكن معظم السلوك الإنجابي بعد وضع البيض ازداد في المجموعتين الاولى والثانيه أكثر من المجموعات الأخرى حيث أن الوالدين في هاتين المجموعتين كانا بالفعل قد احتضانا بيضاً سواء طبيعياً أو زائفاً فبالتالى قاما باحتضانه والاعتناء به. وفي الوقت نفسه، ازداد البحث والتنبيش عن البيض في مجموعات الحمام البلدي المصري بسبب إزالته في المجموعتين الثالثه والرابعه. كانت فترة حضانه البيض ممتده في المجموعه الثانيه اكثر من غيرها ويرجع ذلك الى ان الازواج كانت ترقد على بيض زائف لذلك كانوا فى انتظار فقس البيض ولكن فترة وضع البيض بدايه من الدوره السابقه ازدادت فى المجموعه الاولى حيث أن البيض المحضن من الازواج قد فقس ونتج عنه زغاليل صغار تحتاج رعايه وتربيه من الابوين قبل وضع بيض الدوره القادمه وبالتالي ازداد مستوى البرولاكتين في المجموعات التي قامت بأحتضان البيض (المجموعه الاولى والثانيه) وانخفض بشكل حاد الى ادنى مستوياته بإزاله البيض (المجموعه الثالثه والرابعه). وقد خلصت نتائج البحث الى انه استبدال البيض ببيض اخر مزيف يطيل من فتره حضانه البيض الطبيعى لكن سلوك الابوين يظل مشابه لنظيرهما اللذان يحتضنوا بيض طبيعى ، وايضا يجب الحفاظ علي البيضة الأولى أمانة أثناء تربية الحمام حتى يتم وضع الثانيه بنجاح لان عدم وجود البيضة الاولى فى العش يؤدي الى اضطراب الام وفقدان البيضة الثانيه بسبب هجر العش .