



Should Nest Construction Be Intervened in Domestic Bird Breeding?

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Received: 14.01.2025

Accepted: 13.02.2025

Published: 25.03.2025

How to cite: Özkök, A.O. & Kılınç, G. (2025). Should Nest Construction Be Intervened in Domestic Bird Breeding? *J. Anatol. Env. Anim. Sci.*, 10(2), 126-131. <https://doi.org/10.35229/jaes.1619540>

Atf yapmak için: Özkök, A.O. & Kılınç, G. (2025). Evcil Kuş Yetiştiriciliğinde Yuva Yapımına Müdahale Edilmeli mi? *Anadolu Çev. Hay. Bil. Derg.*, 10(2), 126-131. <https://doi.org/10.35229/jaes.1619540>

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Abstract: It is aimed to get as many offspring as possible from domestic songbirds during the breeding season. However, the insufficiency of the nest material used during nest construction and the individual abilities of the birds in nest construction directly affect the success of the breeding season. This study evaluates the nests built by two groups of female Gloster canaries using the same nest material, as well as the effects on the hatching rates of fertile eggs and the development of offspring. For this purpose, 8 female canaries were involved in this study, 4 in the 1st group (T-1) and 4 in the 2nd group (T-2). After artificially inseminating the female birds showing the signs of estrus, the hatching rates of the eggs were examined. No intervention was performed during the nest construction in Group T-1, whereas the nests of Group T-2 were intervened during the construction process, and the hatching rates of fertile eggs were observed. It was determined that the group T-2 was more successful than the group T-1. It may be possible to obtain more offspring by eliminating the deficiencies observed in the nests of the birds during nest construction to a certain extent. This study aims to determine the effects of interventions made to the nests of female birds inseminated under equal conditions on the incubation process and the hatching rate.

Keywords: Artificial insemination, canary bird, nest material, nest intervention.

Evcil Kuş Yetiştiriciliğinde Yuva Yapımına Müdahale Edilmeli mi?

Öz: Üreme mevsiminde evcil ötücü kuşlardan mümkün olduğunca çok yavru elde edilmesi amaçlanmaktadır. Ancak yuva yapımında kullanılan yuva malzemesinin yetersizliği ve yuva yapımında kuşların bireysel yetenekleri üreme mevsiminin başarısını doğrudan etkilemektedir. Bu çalışmada aynı yuva malzemesini kullanarak iki grup dişi Gloster kanaryasının yaptığı yuvalar değerlendirilmiş, döllenmiş yumurtaların çıkış oranları ve yavruların gelişimi üzerine etkileri değerlendirilmiştir. Bu amaçla 1.grupta (T-1) 4, 2.grupta (T-2) 4 olmak üzere toplam 8 adet dişi kanarya kullanılmıştır. Kızgınlık gösteren dişi kuşlar suni tohumlama ile tohumlanarak yumurta çıkış oranları değerlendirilmiştir. T-1 grubundaki yuvalara yuva yapımı sırasında müdahale edilmemiş, T-2 grubundaki yuvalara ise yuva yapımı sırasında müdahale edilmiş ve döllenmiş yumurtaların çıkış oranları gözlenmiştir. T-2 grubunun T-1 grubuna göre daha başarılı olduğu sonucuna varılmıştır. Yuva yapımı sırasında kuşların yuvalarında gözlenen eksikliklerin belli bir oranda giderilmesiyle daha fazla yavru elde edilmesi mümkün olabilir. Bu çalışmanın amacı eşit şartlarda tohumlanan dişi kuşların yuvalarına yapılan müdahalelerin kuluçka süreci ve yumurtadan çıkma oranına olan etkilerini belirlemektir.

Anahtar kelimeler: Kanarya, yuva materyali, yuva müdahalesi, suni tohumlama.

INTRODUCTION

It is well known that nest construction is crucial for the successful reproduction of many bird species (Healy et al., 2023). Birds inhabiting different environments have developed a wide range of architectural nest designs in line

with the conditions of their habitats (Jessel et al., 2019). Previous observations suggest that the quality of nest construction is shaped by the bird's habitat, life experiences, and individual behavior (Healy et al., 2023). The thermal insulation and temperature variations within the nest may vary depending on the properties of the materials used in nest

construction and the individual skill of the bird during the building process (Lamprecht & Schmolz, 2004). Even though nests vary in shape and size among different bird species, they must be structured in a way that ensures the survival of the offspring despite environmental challenges, thereby maximizing hatching success and the healthy growth of the hatchlings (Biddle et al., 2019). It was reported that some bird species prioritize not only thermal insulation but also the color of the nest materials as a means of camouflage against predators, in harmony with their environmental conditions (Mayer et al., 2009). Birds may use different nesting materials based on the climate and habitat in which they reside. In addition to serving as structural components, these materials may provide various benefits, such as antibacterial or antiparasitic properties (Bach et al., 2022). The type and quantity of materials used in nest construction are closely related with the nest's functionality, particularly its role in ensuring a successful incubation period (Deeming, 2023). The materials selected for nest construction vary based on geographical location, ambient temperature, and the availability of materials in the environment. Even though this variation differs across bird species, it still is an important factor in supporting the development of offspring throughout the incubation period (Biddle et al., 2018). The selection and physical properties of nesting materials influence nest construction, which in turn affects hatching success and chick development indirectly. Therefore, the characteristics of the materials used in building the nests are very important (Breen et al., 2021). Furthermore, the permeability of the nest walls to water vapor in the environment was determined to be directly related to eggshell conductivity, egg mass, and incubation duration (Deeming, 2011).

Even though it is not precisely known which criteria birds use when constructing nests, it is thought that, in addition to instinctive knowledge, learning also plays a role in nest-building behavior (Walsh et al., 2013). Bird species exhibit not only individual skills but also the capacity to learn while building nests. Moreover, they have specialized in selecting materials in line with the physiological needs of the nest (Tello-Ramos et al., 2022). Birds choose the necessary materials for nest construction from those available in their environment, because material availability varies across different regions. This behavioral pattern may be acquired either individually or through social learning from other bird communities (Mennerat et al., 2009). In a study conducted on a songbird species, no significant temperature differences were observed depending on the size of the nest. Consequently, it was suggested that the size of the nest is determined entirely by the bird's individual preference (Sonnenberg et al., 2020).

This study aims to determine the effects of interventions made to the nests of female birds inseminated

under equal conditions on the incubation process and the hatching rate.

MATERIAL AND METHOD

Ethics Committee Permission: This study was approved by the Ondokuz Mayıs University Animal Experiments Local Ethics Committee (2024-49).

Experimental Plan: Eight female Gloster Canaries were used in this study. Each canary was housed individually in breeding cages (60×40×50 cm). A photoperiodic light cycle of 16 hours of light and 8 hours of darkness was applied to induce sexual stimulation (Chiver et al., 2022). Since some male birds may disrupt the nest, no male canary was housed alongside the females.

Semen Collection and Evaluation of Spermatological Parameters: Sperm collection was performed using the cloacal massage method (Cramer et al., 2021). These semen samples were immediately diluted at a 1:1 ratio with Dulbecco's Modified Eagle Medium (DMEM) and then examined for relevant sperm parameters. Freshly diluted semen from each bird was evaluated under a microscope heated to 38°C at 20× magnification. The sperm samples collected from male birds were evaluated in terms of total motility, abnormal spermatozoa rate, and sperm concentration. Birds that did not meet the criteria of >70% motility, <10% abnormal spermatozoa, and >80% viability were excluded from the study. The average sperm concentration of the sampled birds was determined to be in the range of 85×10^6 to 90×10^6 .

Determination of Motility: Sperm motility assessment was conducted twice at different time points for each bird. Following the sperm collection, sperm motility and kinematic parameters were analyzed using a Computer-Assisted Sperm Analysis (CASA) system (SCA®, Microptic, Barcelona, Spain). The CASA system was supported by a negative phase contrast microscope with a heated stage (Nikon Eclipse, Tokyo, Japan). Total sperm motility (%) and motile sperm concentration parameters were evaluated by analyzing at least five microscopic fields using the software system (Yang et al., 2019).

Evaluation of Spermatozoa Morphology: The Gloster canary produces an average of 2 µL of semen. After the semen sample was transferred to 20 µL of Phosphate Buffered Saline (PBS) solution containing 5% formaldehyde, 10 µL of sperm sample was spread on the slide and left to dry at a 45-degree angle. After drying, the slide was immersed in a jar containing SpermBlue dye and left for 2 minutes. After the slide was stained with SpermBlue, it was left to dry. After the drying process, the slide was washed superficially with distilled water, and morphological abnormalities were evaluated (Esin et al., 2023). The abnormal spermatozoa rate was evaluated in

terms of bent or missing flagellum, multiple flagellum, and abnormal head formations at $40 \times$ magnification (Cramer et al., 2019).

Determination of Sperm Vitality Rate: Semen samples were thawed in a water bath at 37°C and gently mixed with eosin-nigrosin stain at a 1:1 ratio. The sample was then spread onto a slide and immediately air-dried. At least 200 spermatozoa were counted at $400\times$ magnification. Partially or fully stained spermatozoa were classified as dead, whereas unstained spermatozoa were considered viable (Bakst & Cecil, 1997).

Determination of Sperm Density: The hemocytometric method was employed to determine sperm concentration in this study. The Gloster canary is a small-sized bird, producing an average semen volume of approximately $2 \mu\text{L}$. The sperm density per mL was calculated using the standard avian sperm concentration assessment method (Bakst & Cecil, 1997). The average number of spermatozoa (5 squares were counted in each section in the hemocytometer, and the number found was divided by the number of squares counted) was x Dilution rate of $\times 10^4$.

Artificial Insemination: Semen obtained from canaries that met the required sperm quality parameters was used for artificial insemination without dilution. No contaminated semen was used for insemination (Özkök, 2022). When the female canary started building a nest, insemination was performed every two days until the first egg was laid. The nest-building process was observed to be completed within 3-6 days, while egg-laying occurred 1-3 days after the completion of the nest. The room temperature was maintained at $22\text{-}25^{\circ}\text{C}$, and relative humidity was kept at 60-65%.

Nest material: Three types of nesting materials were used in this study: flax fiber, sisal fiber, and hemp. To standardize all nests, flax fiber was provided first. After the primary nest structure was completed, sisal fiber was introduced, followed by hemp. No intervention was performed on the nests until the egg-laying process was completed. The nests were left undisturbed in T-1, whereas modifications were made after nest construction was completed in T-2. These modifications included compressing the nest, reinforcing incomplete nest walls with flax fiber, and adjusting the nest base.

Collecting eggs: The eggs were collected every morning starting from the first laid egg and replaced with artificial eggs. The collected eggs were numbered and stored. Once all eggs had been collected, the real eggs were placed back into the nests of the birds in the first group without any intervention. In the second group, the real eggs were placed in the nests after the nesting structure had been adjusted. On the 8th day, the eggs were examined using a light source to determine fertilization status, and the

fertilized eggs were recorded. This study focused solely on the hatching rates of fertilized eggs. Employing the data obtained, the following formula was used to calculate the hatching rate for each group:

$$\text{Egg hatching rate (\%)} = \frac{\text{Number of Hatching}}{\text{Number of Fertile Eggs}}$$



Figure.1 Different nest materials (fiber yarn, hemp, linen rope).

Statistical Analysis: Differences between groups were made with the independent t-test. SPSS 20.0 package program was used for this purpose. The effects (significance) of the groups were evaluated at $p < 0.05$ level (IBM., Corp., 2011).

RESULTS

The collected data were analyzed based solely on fertilized eggs. The nests were left untouched in the T-1 group, whereas the nests were inspected during the nest-building process, and observed deficiencies were corrected in the T-2 group. The main interventions involved compressing the nest base, completing the sidewalls, and adjusting the nest height. The eggs were removed from the nests and replaced with artificial eggs until egg-laying was complete. Once the laying process was finished, the original eggs were returned to the nests. On the 8th day, the eggs were checked with a light source, and fertilized eggs were recorded. The hatching rates of healthy chicks were compared between the two groups. There was a significant difference in hatching rates between the intervention and non-intervention groups ($p < 0.05$). The hatching rate of fertilized eggs was higher in the intervention group (0.68) (Table 1). In the T-1 group, the primary factors affecting hatching success were disruptions in egg rotation, which led to hatching failures, and early embryonic mortality due to irregular heating.

Table.1 Hatching rates in uninterrupted and intervened nests.

Groups	Hatching Rate (%)
T-1	$\%41 \pm 0.060$
T-2	$\%68 \pm 0.105$
P	0.047
T-1. No intervention	T-2. Intervention



Figure. 2 Examples of intervened nests.



Figure.3 Uninterrupted nests

DISCUSSION

Previous studies demonstrated that nest structure directly influences reproductive success in birds. Moreover, the ability to construct a nest is influenced by a bird's individual skills, life experiences, and environmental conditions.

Even though the specific criteria birds use for nest-building are not known, it is thought that in addition to instinctive knowledge, birds can also learn certain aspects of nest construction (Walsh et al., 2013). Bird species specialize in selecting nesting materials that meet their physiological needs (Tello-Ramos et al., 2022). Since nesting materials vary by region (Mennerat et al., 2009), nest-building appears to be shaped by individual abilities, life experiences, and environmental factors. The present study revealed that each bird exhibited different nesting capabilities, which in turn affected incubation success. A long-term study on nonett birds examined over a thousand nests over a decade, analyzing nest height, width, and

materials used. The study concluded that genetic inheritance plays a minimal role in nest-building behavior among nonetts (Järvinen et al., 2017). There is no available data on the nesting success of the parents of the birds used in the present study or on how nest-building traits may have been genetically transmitted. However, it was observed that birds provided with the same nesting materials tended to build similar nests. Furthermore, birds whose nests were adjusted during this study built subsequent nests with similar structural features. Another key finding was that as the duration of the nesting process increased, the amount of nesting material also increased, leading to greater nest height, inner width, and nest wall thickness. A higher accumulation of soft nesting material at the nest base disrupted the routine rotation of eggs during incubation, leading to embryo mortality due to embryos adhering to the eggshell. On the other hand, when the nesting process was shortened, inadequate nest construction led to heat loss, potentially increasing embryo mortality. The most common deficiency in the nests was related to nest base

construction. Since birds do not live in social groups and build nests individually, they do not have the opportunity to learn nest-building techniques. Therefore, this suggests that nest-building behavior is largely instinctual and genetically inherited.

CONCLUSION

This study demonstrated that reducing nest deficiencies through simple interventions significantly lowered chick mortality. Therefore, addressing structural issues in nests during construction could help mitigate chick losses that negatively impact reproductive success.

Contributions: Arda Onur Özkök and Gözde Kılınç contributed to the study conception and design. Material preparation and data collection were performed by Arda Onur Özkök. All authors (Arda Onur Özkök and Gözde Kılınç) contributed data analysis, writing, and editing the last draft of the manuscript. The manuscript has been read and approved by all named authors and that there are no other persons who satisfied the criteria for authorship but are not listed.

Conflict of interest: The authors declare no competing interests.

Ethics Committee Approval: This study was approved by the Ondokuz Mayıs University Animal Experiments Local Ethics Committee (2024-49).

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