

Production Performance and Sperm Characteristics of Quail (*Coturnix-coturnix japonica*) with Different Concentrations of Yolk Immunoglobulin

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(received 03-12-2022; revised 12-09-2023; accepted 27-02-2023)

ABSTRAK

Agasi SY, Ulupi N, Afnan R, Arifiantini RI. 2023. Performa Produksi dan Karakteristik Sperma Puyuh Jantan (*Coturnix-coturnix japonica*) dengan Konsentrasi Immunoglobulin Yolk Berbeda. JITV 28(2):129-135. DOI:<http://dx.doi.org/10.14334/jitv.v28.i2.3173>.

Puyuh diklasifikasikan menurut kekebalannya terhadap konsentrasi IgY yang berbeda. Konsentrasi IgY puyuh berkisar antara 0 sampai 1200 $\mu\text{g ml}^{-1}$ dan konsentrasi IgY ini dapat memengaruhi produksi dan kinerja reproduksinya. Penelitian ini bertujuan untuk menganalisis konsentrasi IgY pada puyuh jantan dan membandingkan performa produksi dan reproduksi pada konsentrasi IgY yang berbeda. Empat puluh dua ekor puyuh jantan usia lima minggu digunakan dalam penelitian ini, 29 puyuh dengan konsentrasi IgY rendah (210-393 $\mu\text{g ml}^{-1}$) dan 13 puyuh dengan konsentrasi IgY sedang (403-564 $\mu\text{g ml}^{-1}$). Performa produksi yang diamati adalah konsumsi pakan, bobot badan awal, bobot akhir, pertambahan bobot badan, konversi pakan, morbiditas, dan mortalitas. Bobot testis dan kualitas semen secara makroskopis dan mikroskopis diuji untuk sifat-sifat reproduksi. Perbedaan performa produksi dan reproduksi masing-masing kelompok dianalisis menggunakan *independent-sample t-test*. Hasil penelitian menunjukkan puyuh jantan dengan konsentrasi IgY berbeda memiliki performa produksi yang sama ($P>0,05$). Hal tersebut dapat diartikan puyuh jantan dengan konsentrasi IgY rendah dan IgY sedang sama baiknya. Berat testis, warna dan pH semen puyuh sama, pada kedua kelompok konsentrasi IgY yang berbeda. Konsistensi semen IgY rendah lebih kental dibandingkan dengan konsentrasi IgY rendah. Puyuh dengan konsentrasi IgY rendah memiliki konsistensi semen yang lebih baik, hal ini berhubungan dengan ukuran testis dari puyuh dengan konsentrasi IgY rendah, lebih berat dibandingkan puyuh dengan IgY sedang. Konsistensi ini berhubungan dengan konsentrasi sperma di dalam semen tersebut. Hasil penelitian menyimpulkan konsentrasi IgY pada puyuh jantan tidak memengaruhi performa produksi. Puyuh dengan konsentrasi IgY rendah memiliki konsistensi sperma yang lebih kental dan konsentrasi sperma yang lebih tinggi.

Kata Kunci: Konsentrasi IgY, Performa, Puyuh, Karakteristik Semen

ABSTRACT

Agasi SY, Ulupi N, Afnan R, Arifiantini RI. 2023. Production Performance and Sperm Characteristics of Quail (*Coturnix-coturnix japonica*) with Different Concentrations of Yolk Immunoglobulin. JITV 28(2): 129-135. DOI:<http://dx.doi.org/10.14334/jitv.v28.i2.3173>.

Quails are classified according to their immunity to different IgY concentrations. Quail IgY concentrations range from 0 to 1200 $\mu\text{g ml}^{-1}$, and these IgY concentrations may affect production and reproductive performance. This study aimed to analyze IgY concentrations in male quail and to compare production and reproductive performance at different IgY concentrations. Forty-two male quail at five weeks of age participated in the study, including 29 quail with low IgY concentration (210-393 $\mu\text{g ml}^{-1}$) and, 13 with intermediate IgY concentration (403-564 $\mu\text{g ml}^{-1}$). The observed productive performances were feed intake, initial body weight, final body weight, weight gain, feed conversion, morbidity, and mortality. In addition, this study observed testicular weight and macroscopic and microscopic semen quality for reproductive traits. Differences in production and reproductive performance of each group were analyzed using an independent-sample t-test. The result showed that male quail with different IgY concentrations were equal in all productive traits ($P>0.05$). This means that male quail with low IgY and medium IgY concentrations are equally good. Testicular weight, semen color and pH were identical at different IgY concentrations. Quails with low IgY concentrations have better sperm consistency, which is related to the weight of the testes, which are heavier in quails with low IgY concentrations than in those with moderate IgY concentrations. This consistency is related to the concentration of sperm in the semen. The result concludes IgY concentrations in male quail did not affect production performance. Quails with low IgY concentration had thicker sperm consistency and higher sperm concentration.

Key Words: IgY Concentration, Performance, Quail, Semen Characteristics

INTRODUCTION

Quail has rapid growth, earlier sexual maturity, high egg production rate, short generation interval (3-4 generations per year), low space requirements, lower feed requirements, short incubation period of hatching eggs, lower feed costs, and lower susceptibility to common chicken diseases (Rahman et al. 2016). Quails were once small birds for toys, but are now used for commercial egg and meat production (Khairani et al. 2016). High quail productivity depends on several factors, including seed, health, feed, environment, and management. Quail productivity can be evaluated by body weight gain, feed efficiency, egg production, disease resistance, and stress. Immunity is influenced by the immune system, which is supported by the function of immune cells to maintain immunity against pathogens that can infect the body. The immune system plays an important role in fighting various diseases (van Seventer & Hochberg 2017).

According to van Seventer & Hochberg (2017), body resistance is the body's response to bacteria, viruses, fungi and parasites, which is influenced by many factors such as feed, husbandry management and genetics. According to Spillner et al. (2012), some poultry species are very sensitive to contact with foreign antigens, which affects the immune system and the production of immunoglobulins (IgY). Immunoglobulin (Ig) is the first substance identified as a molecule in serum that can neutralize various foreign bodies or microorganisms that cause infections. Poultry immunoglobulin consists of IgA, IgM, and IgY. Yolk immunoglobulin plays a role in the poultry's main system, which can inhibit pathogenic bacteria (Zhang et al. 2019). The higher the IgY concentration, the better the immune system (Setyawati 2018). One indicator to measure the immune system of quail is to examine the IgY content in the animals' serum. Immunoglobulins found in blood serum and egg yolk (Pereira et al. 2019). IgY concentration in blood serum ranges from 5-15 mg mL⁻¹ (Gaetani et al. 2017).

High resistance of the body improves production performance (Regar et al. 2013). Reproductive performance depends on the quality of both female and male animals. Males for breeding purposes must have high reproductive performance, including good semen quality. Semen quality plays an important role in fertility (Modupe et al. 2012). Semen quality of poultry can be assessed by macroscopic and microscopic assessments of semen. Macroscopic assessments include semen volume, color, consistency, and pH, while microscopic assessments include mass movement, motility, viability, concentration, and sperm abnormalities (Malik et al. 2013; Elagib et al. 2012). Several researchers have studied the relationship between IgY concentration and cock performance, including Ariyanti et al. (2019) and

Setiawan et al. (2021). Ariyanti et al. (2019) reported that Sentul chickens with high IgY concentrations ($> 9.30 \pm 0.45$ mg mL⁻¹) potentially fertilize more hens. Setiawan et al. (2021) reported that IPB-D1 chickens with high IgY concentrations ($> 9.36 \pm 2.88$ mg mL⁻¹) had better sperm motility and viability compared to low IgY concentrations. Both researchers found that IgY concentration did not affect performance, but high IgY concentrations in chickens showed the potential to fertilize more hens and better sperm motility and viability compared to chickens with low IgY. The aim of this study was to find out whether IgY concentrations in quail also lead to the same results. Therefore, the performance and semen quality as well as the fertilization potential of female quail were investigated in this study.

MATERIALS AND METHODS

Approval of the animal ethics committee

This study was approved by the Animal Ethics Committee of the Faculty of Veterinary Medicine and Biomedical science, Bogor Agricultural University (IPB), under approval number 046/KEH/SKE/XI/2022.

Time and place

The study was conducted from October to December 2021. Quail rearing was conducted in the field laboratory of the Department of Breeding and Genetics, Faculty of Animal Husbandry, Bogor Agricultural University. IgY concentration tests were performed in the integrated laboratory of the Division of Animal Diseases and Public Veterinary Medicine (IPHK), and semen quality tests were performed in the laboratory of the Reproductive Rehabilitation Unit (URR), Division of Reproduction and Obstetrics, School of Veterinary Medicine and Biosciences, IPB University.

Examination of IgY concentration of male quails

The 42 male quails used in this study were five weeks old. The initial body weight of the quails ranged from 100-137 g head⁻¹. The determination of total IgY concentration in blood serum was performed using the indirect enzyme-linked immunosorbent assay (ELISA) method according to Murai et al. (2016). Blood was collected from the brachial vein of the wing using a 0.5-mL syringe. The total IgY values of all samples were averaged. Quails with IgY concentration above the average are classified as quails with high IgY concentration. Quails with IgY concentration equal to or below the average are classified as quails with low IgY concentration.

Quail rearing

The quails were housed in wire mesh cages measuring 15 x 30 x 25 cm³ and were provided with food and water in each cage. The cages were equipped with a 7-watt lamp for illumination at night. New Hope P-100 laying quail feed was used in this study. New Hope P-100 contains a maximum moisture content of 13%, minimum protein of 20%, minimum fat of 7%, maximum ash of 7%. Quails were fed at 7:00 am and 4:00 pm and received drinking water ad libitum. Quails and feed residues were weighed weekly to determine body weight gain and feed consumption.

Evaluation of quail performance.

Quail performance at different IgY concentrations was evaluated. Feed consumption (g) was calculated from the number of feedings minus feed leftovers. Final body weight (g) is the body weight of the quail at the end of the study. Body weight gain (g) was determined by calculating the final and initial body weight. The feed conversion ratio was determined from the amount of feed consumed divided by the amount of weight gain. Morbidity (%) was determined by dividing the number of sick quail by the total number of quail and multiplying by 100%. Mortality (%) was determined by dividing the number of quail that died by the total number of quail and multiplying by 100%.

Testing of quail semen properties

Semen collection

Quail sperm collection was performed on 13-week-old quails. Sperm collection was performed using the epididymal collection method. Quails were slaughtered and dissected, and both testes were freed from other tissues and weighed with a digital scale. Sperm was collected from the right and left epididymis (Ouennes et al. 2019).

Evaluation of the semen

The semen was examined macroscopically and microscopically. The macroscopic evaluation included color, consistency and pH. Due to the technique of sperm collection from the epididymis, sperm volume was not determined in this study. The pH of the semen was measured using a special pH indicator paper (Merck scale 6.4-8). Five µl of semen was dropped onto the pH paper and allowed to stand for 15-30 seconds. The consistency of the semen was classified as thin, medium,

and thick, and the color of the semen was observed visually.

Microscopic examination of semen, including motility, viability, concentration, and sperm abnormalities, was performed using a binocular microscope (Olympus CX23). Sperm motility was examined by mixing 2 µl of semen with 20 µl of physiological saline. The solution was homogenized and covered with a coverslip. The slides were viewed under a microscope at 400× magnification. Sperm motility was assessed by estimation from 5 fields of view by comparing the number of sperm moving forward with other sperm movements. Motility values were expressed as percentages. Sperm viability was determined by mixing 5 µl of semen with 50 µl eosin-nigrosin solution. The mixture was homogenized, then a test preparation was made and dried on a heating table for 10-15 seconds. The preparations were viewed under a microscope at 400× magnification. The percentage of live sperm was counted in 10 fields of view, and the minimum number of cells had to be >200. Live sperm do not absorb color, while dead sperm stain purple. The percentage of live sperm is calculated according to the formula: Number of live sperm divided by the total number of sperm multiplied by 100%.

When observing sperm abnormalities, the same staining is used to examine live sperm. The percentage of abnormal and normal sperm is determined in 10 fields of view with a minimum cell count of 200 cells (Hambu et al. 2016). The percentage of abnormal sperm is calculated using the formula: number of abnormal sperm divided by the total number of sperm and then multiplied by 100%. Sperm concentration was calculated using a Neubauer chamber. The semen was diluted 500 times with formol saline (2 µL semen in 998 µL formol saline). The mixed solution was placed in the counting chamber and viewed under a microscope at 400× magnification. Sperm counts were performed on five counting boxes. The sperm concentration was calculated using the formula: Number of sperm counted x 25 x 10⁶.

Data analysis

Data on production performance (feed consumption, initial body weight, final body weight gain, feed conversion), testicular weight, and semen quality (color, pH, sperm consistency, motility abnormalities, and sperm concentration) were analyzed by independent-sample t-test (Mishra et al. 2019) using the SAS program. Data on overall morbidity and mortality were analyzed descriptively.

RESULTS AND DISCUSSION

The results of indirect Elisa IgY measurement in quail ranged from 210 to 564 µg ml⁻¹. Quail IgY

according to Murai et al. (2016) ranged from 0 to 1200 µg ml⁻¹. The IgY results have only "low IgY" and "moderate IgY" categories. Quail with low IgY had a value of 210-393 µg ml⁻¹ with a total of 29 quail and quail with moderate IgY had a value of 403-564 µg ml⁻¹ with a total of 13 quail.

Male quail performance

The results of the analysis of male quail performance are presented in Table 1. The performance of male quail with different IgY concentrations showed no differences (P>0.05) in feed consumption, initial body weight, final body weight, weight gain, and feed conversion. This result means that male quails with low and medium IgY concentrations performed equally well. The feed conversion ratio of male quail with low IgY concentration and male quail with medium IgY concentration has a value of 5.98 ± 2.49 and 5.37 ± 2.08, respectively. This feed conversion ratio is in the high category. This is due to the fact that quail enter the production phase at nine weeks of age, so the digested feed nutrients are used for reproductive organ development rather than growth (Panjaitan et al. 2012).

The results of this study are better than those of Dewi et al. (2016), who put the feed conversion value of male quail at 6.31. Khalil (2015) indicated that the feed conversion value of quail is 3.3-4.9. The lower the feed conversion value, the more efficient the feed conversion and vice versa. Mortality of quail with low IgY concentration was 3.45% (1 of 29 birds) higher than quail with medium concentration. The mortality was not due to quail disease, but was due to cage wire entrapment.

Morbidity of male quail with different IgY concentrations was 0%. During the study, no quail showed disease symptoms such as watery eyes, matted and dull plumage, and lethargy. Quail are known to be highly resistant to disease (Rabiu-Mohammed & Ejiofor 2015). The results of this study confirm reports by Ariyanti et al. (2019) and Setiawan et al. (2021) that low or high concentrations of IgY do not affect production performance in chicken and quail.

Testicular weight

The results of the analysis of testicular weights of quails are shown in Table 2. The testicles are the organs that produce sperm. Testicular weight provides information about sperm productivity. The results showed that testicular weights did not differ between quails with low and medium IgY concentration (P>0.05), nor between right and left testicular weights. Testicular size is used as a supportive indicator of sperm production. A positive correlation between testis, body weight, and sperm production has been found in some animals (Indriastuti al. 2020; Perumal 2014). Testicular weight is dependent on age, breed, season, and diet. The male quail used in this study were of the same age, breed, and diet. As quail age, body weight increases and so does the weight of the organs in the body, which include the testes. The testes consist internally of testicular tubules (85%-95% of the testicular volume), where spermatogenesis takes place. The larger the volume of testicular tubules in the testis, the more sperm are produced. The walls of the testicular tubules are

Table 1. Production performance of 9-week-old male quails with different IgY concentrations

Variables	Low IgY	Moderate IgY
Feed consumption (g head ⁻¹ week ⁻¹)	160.78±27.17	165.79±7.52
Initial body weight (g head ⁻¹)	117.14±9.39	114.69±12.91
Final body weight (g head ⁻¹)	144.82±12.75	145.54±15.15
Body weight gain (g head ⁻¹ week ⁻¹)	26.90±10.29	30.85±10.38
Feed conversion	5.98±2.49	5.37±2.08
Morbidity (%)	0	0
Mortality (%)	3.45	0

Table 2. Testicular weight of quail with different IgY concentrations.

Testis	Low IgY (n=6)	Moderate IgY (n=6)
Right (g)	2.16±0.63	1.94±0.37
Left (g)	2.52±0.37	1.92±0.73
Mean (g)	2.34	1.93

composed of connective tissue and germinal epithelial tissue, which plays a role in spermatogenesis. Testicular weight is closely related to sperm concentration. The results showed that male quail with low IgY concentrations tended to have higher testicular weight. This fact is expected to influence the amount of sperm produced.

Macroscopic semen quality

The results of the macroscopic analysis of quail sperm quality are shown in Table 3. The color and pH of quail semen with different IgY concentrations did not differ ($P>0.05$). Quail with low IgY concentration had thicker sperm consistency than quail with medium IgY concentration ($P<0.05$) (Table 3). Semen consistency and color in poultry, including chickens, can describe sperm concentration (Junaedi et al. 2016). Semen that is thick and has a milky white color has a high sperm concentration and vice versa. The higher the sperm concentration, the more intense the color (Sujoko et al. 2009).

The acidity (pH) of the semen determines the life status of the sperm within it. A high or low semen pH leads to a faster sperm death (Sujoko et al. 2009). Semen pH is the same for low IgY and medium IgY, namely 7.3. Fresh semen from poultry is usually slightly alkaline, with an average pH between 7.0 and 7.6. Sperm pH is influenced by several factors, including sperm activity in the breakdown of fructose, which lowers pH. The high activity of sperm in decomposing energy sources from fructose increases the production of lactic acid in sperm, making the pH more acidic. Table 3 shows a significant difference in sperm consistency. Quail with low IgY

concentrations have better sperm consistency, which is related to the fact that the testes of quail with low IgY concentrations are heavier than those of quail with medium IgY concentrations. This consistency is related to the concentration of sperm in the semen.

Motility, viability, and abnormalities of quail sperm did not differ between quails with different IgY concentrations ($P>0.05$). This result is due to the fact that all quail were in good health at the time of semen collection. Relatively healthy quails produce the same production. The sperm motility of quail in this study ranged from 46.67 ± 15.06 to 51.67 ± 24.63 , which is lower than the study of Lesmono et al. (2017) on six-week-old quail, which ranged from 80% to 88%. Sperm viability ranged from $58.71 \pm 12.31\%$ to $66.35 \pm 9.54\%$. Sperm viability is a crucial indicator for assessing good or poor sperm quality. In this study, quail sperm abnormality was very high and ranged from 38.97 ± 13.65 to $52.57 \pm 26.52\%$. According to Lesmono et al. (2017), sperm abnormality in quail is $\pm 7.4\%$. Abnormal sperm correlate with motility, fertility and hatchability in poultry (Feyisa et al. 2018).

The sperm concentration of quail with low IgY concentration ($1521.88 \pm 18.92 \times 10^6$) was higher than quail with medium IgY concentration ($831.25 \pm 388.00 \times 10^6$). The sperm concentration in this study was higher than that reported by Chelmonska et al. (2008), which was $120-312 \times 10^6 \text{ ml}^{-1}$. Chelmonska et al. (2008) reported that quail sperm concentration can reach $2240-2640 \times 10^6 \text{ ml}^{-1}$ with appropriate male selection. High sperm concentration increases the potential of females to be inseminated. The amount of quail sperm required for artificial insemination is not known with certainty. Thélie et al. (2019) indicated that using $15-60 \times 10^6$ sperm per insemination results in a

Table 3. Macroscopic quality of quail semen with different IgY concentrations

Variables	Low IgY	Moderate IgY
Semen color	2±0.0	1.5±0.55
Semen pH	7.3±0.29	7.3±0.38
Semen consistency	2±0.0 ^b	1.17±0.41 ^a

Different letters in the same row indicate significant differences ($P<0.05$). Color 1= cloudy white, 2= milky white; Consistency 1= medium, 2= thick

Table 4. Microscopic quality of quail semen with different IgY concentrations

Variables	Low IgY	Moderate IgY
Sperm motility (%)	51.67 ± 24.63	46.67 ± 15.06
Sperm viability (%)	66.35 ± 9.54	58.71 ± 12.31
Sperm abnormalities (%)	38.97 ± 13.65	52.57 ± 26.52
Sperm concentration ($\times 10^6 \text{ mL}^{-1}$)	1521.88 ± 18.92^b	831.25 ± 388.00^a

Different letters following numbers in the same row indicate significant differences ($P<0.05$)

high fertility rate of up to 80%. The high sperm concentration in quail with low IgY concentration is influenced by testicular weight and sperm consistency.

Microscopic semen quality in quail with different IgY concentrations

The results of the microscopic quality analysis of quail semen are shown in Table 4. Quail sperm quality was varied in this study. These differences may be explained by individual variations as reported by Indriastuti et al. (2020) in Bali cattle. When collected by abdominal massage, semen quality was also varied. Breed, location, diet, age, and climate differ as sperm parameters may varied depending on these factors (das et al. 2016; Kuzlu & Taskin 2017; Mavi et al. 2019). This is also due to the uneven distribution of data in each group. Testicular weights from this study were positively correlated with better sperm concentration in quail. This fact was evident in quail with low IgY concentrations. Quails with low IgY concentrations have a weaker immune system. Therefore, to maintain the immune system of quail, better maintenance management, feeding management, and biosecurity for quail with low IgY concentrations are needed to maintain their immune system and use them for breeding.

CONCLUSION

There is no difference in the production performance of male quail with low and medium IgY concentrations. Quails with low and medium IgY had the same testicular weight, semen color and pH. Semen consistency and sperm concentration of quails with low IgY were better than quails with medium IgY.

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