Vol. 1 No.2 (Mei 2024): 83-89

e-ISSN 3048-4081 p-ISSN 3048-2607

Bestindo of Animal Science

Journal Homepage: https://bestindolestari.id/journal/index.php/bas/

Effect of Combination of Energy Levels in Feed and Duck Egg Yolk as a Semen Diluent on the Quality of Native Rooster Spermatozoa

Binti Khopsoh[1*] and Nining Haryuni[2]

[1,2] *Universitas Nahdlatul Ulama Blitar*
Jl. Masjid No.22 Kauman, Kec.Kepanjenkidul, Kota Blitar, Indonesia
[*]E-mail: khopsohbinti@gmail.com

(received: **Maret 2024** ; revised: **April 2024** ; accepted: **Mei 2024**)

ABSTRACT

This study aims to determine the effect of energy levels in feed and the use of duck egg yolks as a semen diluent on the quality of spermatozoa. The material used is 80 weeks old native rooster, eosin, 3% NaCl, duck egg yolk and aquadest. This research is a biological research using a 2x4 factorial completely randomized design (CRD). The first factor was the energy level in the feed (E) with 2 levels (2750 and 2850kcal/kg) and the second factor was the level of egg yolk as a semen diluent (P) (0, 4 and 8 ml/ml semen). Semen was collected using a massage method from the abdomen to the cloaca. The results of statistical analysis showed that the energy level in the feed had a very significant effect ($p<0.01$) on the movement of spermatozoa and significantly ($p<0.05$) on live spermatozoa. Dilution using egg yolk from duck eggs gave a very significant effect ($p<0.01$) on the pH of semen and the number of live spermatozoa. The average pH of the semen obtained in this study ranged from 6.00 – 8.83; mass movement score 2.25-3.00; spermatozoa motility by 80%; live spermatozoa were 89.50-92.50% and spermatozoa abnormalities were 2.27-2.28%. This study concluded that the quality of semen can be improved by increasing the energy level in the feed and duck egg yolk can be used as a semen diluent. The best energy level to improve semen quality is 2850 kcal/kg and egg yolk as a semen diluent can be used up to 4ml/ml semen.

Key Words: Metabolic energy, semen diluent, egg yolk, spermatozoa quality

INTRODUCTION

Population growth in Indonesia which reaches 3-4 million per year or around 1.49% provides great opportunities for culinary businesses (Haryuni et al. 2019; Haryuni 2014, 2018, 2021). On the other hand, the global economic developments in Indonesia have an impact on the increasing demand for poultry meat due to the wider culinary business. Poultry meat consumption in Indonesia in 2017 was 2,305,000 tons, of which 9% was supplied from native chicken (Haryuni et al. 2022; Haryuni & Fanani, 2017; Haryuni & Muanam,2023). Demand for native chicken meat in 2019 can only be supplied by 3%. This provides a great opportunity for domestic chicken business development (Haryuni et al. 2021). One of the national assets in the livestock sector that supports the socio-economic sector of the community is native chicken. The disadvantages of extensive rearing include difficult health control, low growth and slow development caused by natural mating (Haryuni et al. 2020). One of the efforts to develop native chickens is by using artificial insemination (IB) technology. The advantage of artificial insemination technology is that one male can be used to inseminate 20-30 hens (Prabhakar et al. 2020).

One of the factors that play an important role in the success of artificial insemination is cement diluent. The quality of cement without diluent will decrease when stored for 20-60 minutes at room temperature. Semen stored at room temperature for 20 minutes without the addition of diluent can cause a decrease in fertility of about 10% and semen stored at temperatures above 300 C for one and a half hours has an impact on low fertility to zero fertility (Roiter, Ya and Konopleva, A 2020). One of the diluents that can be used is duck egg yolk. According to (Astuti et al. 2020) duck egg yolks have high nutritional value. Lippoprotein and lecithin high in egg yolks can be used as an energy source for spermatozoa. (Haryuni et al. 2020) reported that increasing the energy level in the feed can improve the quality of spermatozoa. Therefore, it is necessary to research the combination of energy levels in feed and the use of duck egg yolks as a diluent for native rooster semen.

MATERIALS AND METHODS

Materials and Experiemental Design

The materials used in this study were 24 native rooster aged 80 weeks, 70% alkohol, eosin citrate, 3% NaCl, duck egg yolk, aquadest and semen. The diluent is made by mixing duck egg yolk with NaCl with a ratio of 1:1. This research is biological and laboratory research. This study used a 2x4 factorial Completely Randomized Design (CRD). The first factor was the energy level in the feed (E) and the second factor was the dilution of semen using duck egg yolk (P). This study used 2 energy levels (2750 and 2850 kcal/kg) and 3 dilution concentrations (0, 4 and 8 ml/ml semen) with each treatment repeated 4 times.

Bird Managemen and Feed

Biological research used 24 native rooster aged 80 weeks with body weights ranging from 2.10-2.30 kg and in good health. Males were placed in individual cages with a size of 70 x 50 x 100 cm.

Table 1. Composition of diets

Ingredients	Experimental feed					
	E1P0	**E1P1**	**E1P2**	**E2P0**	**E2P1**	**E2P2**
Corn (%)	52.33	52.33	52.33	50.33	50.33	50.33
Soy bean meal (%)	22.20	22.20	22.20	22.40	22.40	22.40
Meat bone meal (%)	8.20	8.20	8.20	8.00	8.00	8.00
Rice bran (%)	8.00	8.00	8.00	7.90	7.90	7.90
Grit (%)	5.00	5.00	5.00	5.00	5.00	5.00
Limestone (%)	3.10	3.10	3.10	3.10	3.10	3.10
Oil (%)	0.20	0.20	0.20	2.30	2.30	2.30
Complete premix (%)	0.50	0.50	0.50	0.50	0.50	0.50
DCP (%)	0.30	0.30	0.30	0.30	0.30	0.30
Salt (%)	0.10	0.10	0.10	0.10	0.10	0.10
Sodium Bicarbonat (%)	0.07	0.07	0.07	0.07	0.07	0.07

Calculations using Brill Formulation software

Table 2. Nutrient of diets

Ingredients	Experimental feed					
	E1P0	**E1P1**	**E1P2**	**E2P0**	**E2P1**	**E2P2**
ME (kcal/kg)	2751.40	2751.40	2751.40	2851.60	2851.60	2851.60
Crude protein (%)	19.12	19.12	19.12	19.01	19.01	19.01
Crude fat (%)	4.23	4.23	4.23	6.20	6.20	6.20
Crude fiber (%)	3.25	3.25	3.25	3.17	3.17	3.17
Calsium (%)	3.91	3.91	3.91	3.91	3.91	3.91
Total phosphorus (%)	0.78	0.78	0.78	0.77	0.77	0.77
Phospor avail (%)	0.49	0.49	0.49	0.49	0.49	0.49
Sodium (%)	0.13	0.13	0.13	0.13	0.13	0.13
Chloride (%)	0.15	0.15	0.15	0.15	0.15	0.15
Ash (%)	3.65	3.65	3.65	3.67	3.67	3.67

Calculations using Brill Formulation software

Feeding twice a day is 40% given in the morning at 7.00 AM and 60% in the afternoon at 2.00 PM (Ridwan et al., 2022; Haryuni et al., 2024; Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020). Drinking water was given ad libitum.

E1P0 = Energy 2750 kcal/kg + 0 ml diluent

E1P1 = Energy 2750 kcal/kg + 4 ml diluent

E1P2 = Energy 2750 kcal/kg + 8 ml diluent

E2P0 = Energy 2850 kcal/kg + 0 ml diluent

E2P1 = Energy 2850 kcal/kg + 4 ml diluent

E2P2 = Energy 2850 kcal/kg + 8 ml diluent

Semen collection

Semen used in laboratory research was collected using the massage method. Before doing the massage the area between the abdomen and the cloaca was cleaned using a tissue that had been moistened with 70% alcohol. Massage is done slowly in the area between the abdomen and the cloaca (Haryuni et al., 2024; Haryuni et al., 2022). The massage was stopped when the tail of the male began to lift and then a scale tube was prepared to accommodate the semen that came out (Ridwan et al., 2022; Haryuni et al., 2024; Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020).

Parameters Measure

Semen pH

The steps taken to determine the pH of the semen produced are as follows: litmus paper along with a standard to see the pH value is prepared, the tip of the litmus paper is dipped in semen, then waited for 60 seconds, the color changes that occur on the litmus paper are observed, the colors observed are matched with standard litmus paper to determine the pH and recorded (Haryuni et al., 2024; Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020)..

Spermatozoa mass movement

The mass movement of spermatozoa can be observed using a microscope at 400x or 100x magnification (Haryuni et al., 2024; Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020; Ridwan et al., 2022). The indicators for assessing the mass movement of spermatozoa according to (Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020) are:

a. Very good (+ + +), it looks like a big thick wave, many, dark and moving actively to form a thick black cloud.

b. Good (++), it looks like the form of small waves that are thin, rare, moving slowly and seem less clear.

c. Not good (+), only the progressive motion of the individual appears and there is no visible mass collection that forms like a wave.

d. Bad (0), the only visible movement of individuals in small numbers and looks sluggish.

Spermatozoa motility

Spermatozoa motility is calculated by looking at the movement of spermatozoa that move actively and progressively forward (Ridwan et al., 2022; Haryuni et al., 2024; Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020).

Living spermatozoa

Measurement of live spermatozoa was carried out by dripping semen on an object glass, then 2 drops of eosin citrate dye and observed under a microscope with a magnification of 400x. Indications of live spermatozoa are colorless (transparent), dead sperm will be red. The percentage of live spermatozoa was calculated using the following formula (Ridwan et al., 2022; Haryuni et al., 2024; Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020).

Spermatozoa abnormalities

Spermatozoa abnormalities in general can be observed in terms of shape (head shape, head size, tail shape, tail size etc.). The percentage of spermatozoa abnormalities can be calculated as follows (Ridwan et al., 2022; Haryuni et al., 2024; Haryuni et al., 2022; Haryuni et al., 2021; Haryuni et al., 2020).

RESULTS AND DISCUSSION

The average quality of the spermatozoa from the research is presented in Table 3.

Semen pH

Statistical analysis showed that the energy level in the feed of native rooster had no significant effect (P>0.05) on the semen pH. The average pH obtained in this study ranged from 6.83-6.87. This result is almost the same as the study of (Astuti et al. 2020) who got a cement pH value of 6.00 and lower than the results of the (Getachew 2016) study which found the pH of chicken semen was 7.20-7.60.

Spermatozoa metabolism plays an important role in the magnitude of the pH value of semen. Spermatozoa metabolism is carried out anaerobically when they are outside the male's body. Anaerobic metabolism runs without oxygen where the product of this anaerobic metabolism is lactic acid. The high lactic acid content in cement causes the cement pH to become lower (Hakim and Humaidah 2019). (Usman, Tijjani et al. 2021) reported that the pH of chicken semen was influenced by several factors including race, age, nutritional status, season and frequency of semen collection. (Widiastuti et al. 2018) reported that duck egg yolk phosphate diluent gave the most optimal results in maintaining progressive motility and viability of pelung chicken spermatozoa stored at 4ºC for 48 hours.

Table 3. Average quality of spermatozoa

Treatment	Variabel				
	Semen pH	**Spermatozoa mass movement**	**Spermatozoa motility (%)**	**Living spermatozoa (%)**	**Spermatozoa abnormalities (%)**
Metabolic energy level					
E1	6,83 ± 0,39	2,25^{a} ± 0,39	80 ± 0,00	90,17^{a} ± 0,21	2,28 ± 0,001
E2	6,87 ± 0,34	3,00^{b} ± 0,34	80 ± 0,00	91,08^{b} ± 0,37	2,28 ± 0,001
Level of duck egg yolk as a diluent					
P0	8,40^{b} ± 0,05	2,63 ± 0,19	80 ± 0,00	91,75^{b} ± 0,38	2,27 ± 0,001
P1	6,05^{a} ± 0,02	2,63 ± 0,19	80 ± 0,00	90,50^{ab} ± 0,25	2,28 ± 0,001
P2	6,10^{a} ± 0,05	2,63 ± 0,19	80 ± 0,00	89,63^{a} ± 0,06	2,28 ± 0,002
The interaction between the level of metabolic energy and the level of duck egg yolk as a diluent					
E1P0	8,50 ± 0,35	2,25 ± 0,43	80 ± 0,00	91,00 ± 0,71	2,28 ± 0,23
E1P1	6,00 ± 0,00	2,25 ± 0,43	80 ± 0,00	90,00 ± 1,06	2,28 ± 0,22
E1P2	6,00 ± 0,00	2,25 ± 0,43	80 ± 0,00	89,50 ± 1,00	2,28 ± 0,24
E2P0	8,30 ±0,10	3,00 ± 0,00	80 ± 0,00	92,50 ± 1,12	2,27 ± 0,17
E2P1	6,10 ± 0,17	3,00 ± 0,00	80 ± 0,00	91,00 ± 0,71	2,28 ± 0,16
E2P2	6,20 ± 0,20	3,00 ± 0,00	80 ± 0,00	89,75 ± 0,83	2,28 ± 0.18

The difference in superscripts in the same line showed that the energy level in the feed had a very significant effect (P<0.01) on the mass movement of spermatozoa and significantly on live spermatozoa. Egg yolk content in semen dilution has a very significant effect (p<0.01) on the pH of semen and the number of live spermatozoa.

Spermatozoa Mass Movement

Statistical analysis showed that the energy level in the feed of native rooster had a very significant effect (P<0.01) on the mass motility score of spermatozoa. The average mass movement score is 2.25-3.00. This result was influenced by a single factor of energy level in the feed. (Haryuni et al. 2020) reported that energy in feed is the main source to form feed reserves for spermatozoa. Feeds with high metabolic energy can supply nutrients to the spermatozoa in sufficient quantities to be used for their movement to the ovum. Feeds with high metabolic energy have an effect on the high mass movement of spermatozoa. (Dadang et al. 2011) state that energy in the form of ATP generated from metabolic processes is used for the movement of spermatozoa. The higher the energy produced, the higher the movement activity of the spermatozoa.

Spermatozoa Motility

Statistical analysis showed that the energy level in the feed of native roosters had no significant effect (P>0.05) on the motility of spermatozoa. The average motility of spermatozoa is 80%. The results of this study found that the motility of spermatozoa for all treatments was 80%. This result is higher than the research conducted by (Widiastuti et al. 2018) regarding the use of various egg yolks as a diluent on motility and vitality of pelung chicken spermatozoa which obtained motility values ranging from 64-69%. Spermatozoa motility according to (Haryuni et al. 2020) is influenced by the nutritional quality of males.

Living Spermatozoa

Statistical analysis showed that the energy level in the feed of free-range males had a very significant effect (P<0.01) on live spermatozoa. The average percentage of spermatozoa ranged from 89.75-91.75%. This result is the same as the study (Haryuni et al. 2020) which is 92.50% and higher than the study (Widiastuti et al. 2018) which is 72%-78%. (Bathgate et al. 2006) reported that the nutritional content of duck egg yolks when compared with chicken and quail egg yolks turned out to be duck eggs have better nutrients. The use of duck egg yolk as a diluent has more complete nutrition so that it can maintain the life of spermatozoa longer.

Spermatozoa Abnormalities

Statistical analysis showed that the energy level in the feed of free-range males had no significant effect (P>0.05) on spermatozoa abnormalities. The average percentage of live spermatozoa ranged from 2.27-2.28%. This result is lower than (Sholihat et al. 2021), which is 4.40-6.50%. Factors that influence spermatozoa abnormalities include disease, heat stress, cryopreservation process, chicken strain, season and post-collection preservation and staining at the time of analysis (Haryuni et al. 2020).

CONCLUSIONS

This research can be concluded that there is no interaction between the energy level in the feed and the level of dilution of semen using duck egg yolk. Improving the quality of semen can be done by increasing the energy level in the feed and using duck egg yolk as a semen diluent. The best energy level to improve semen quality is 2850 kcal/kg and duck egg yolk as a semen diluent can be used up to 4 ml/ml semen.

REFERENCES

Agung, F., Haryuni, N., & Lestariningsih, L. (2024). Pengaruh Tipe Kandang (Closed House dan Open House) dan Umur Ayam terhadap Tingkat Keuntungan Usaha Ayam Petelur. Jurnal Ilmiah Fillia Cendekia, 9(1), 58-65. doi:10.32503/fillia.v9i1.2755

Astuti, T., Sari, R.M., Akbar, S.A., and Cornelio, D., 2020. Dilution Effect of Fresh of Pesisir Cattle ' s Semen with Coconut Water and Duck ' s Egg Yolk on Spermatozoa Quality. International Journal of Agriculture Innovations and Research, 8 (4), 378–385.

Bathgate, R., Maxwell, W.M.C., and Evans, G., 2006. Studies on the Effect of Supplementing Boar Semen Cryopreservation Media with Different Avian Egg Yolk Types on in Vitro Post-thaw Sperm Quality. Reproduction in Domestic Animals, 41 (1), 68–73.

Bhagat, V., V. Khune, S.K. Chourasia, M.K. Gendley, K. Mukherjee. 2016. Linear regression equations for estimation of body weights in Sahiwal calves. Journal of Animal Research, 6: 161-163.

Dadang, O., Saleh, M., and Isyanto, Y., 2011. Dan Fertilitas Spermatozoa Ayam Kate Lokal. Cakrawala Galuh, I (6), 1–6.

Edi, D. N., & Haryuni, N. (2023). Bahan Pakan Ternak Sumber Serat. AE Publishing

EDI, D. N., & HARYUNI, N. (2023). Estimation of Greenhouse Gas Emission Burden of Livestock Sector in East Java Province, Indonesia: Estimasi Beban Emisi Gas Rumah Kaca dari Sektor Peternakan di Provinsi Jawa Timur, Indonesia. Jurnal Teknologi Lingkungan, 24(2), 157-165. https://doi.org/https://doi.org/10.55981/jtl.2023.1004

Getachew, T., 2016. A Review Article of Artificial Insemination in Poultry. World s Veterinary Journal, 6 (1), 25.

Hakim, L. and Humaidah, N., 2019. Pengaruh Lama Penyimpanan semen dengan Pengencer Sitrat dan Kuning Telur terhadap Kualitas Semen Ayam Petarung Magon. Jurnal Rekasatwa Peternakan, 1 (1), 84–89.

Haryuni, N. (2014). Efek Penambahan Jus Daun Sirih (Piper Bettle Linn) Sebagai Aditif Pakan Terhadap Sifat Antibakteri Dan Performans Ayam Petelur (Doctoral dissertation, Universitas Brawijaya).

Haryuni, N. (2018). Analisis Kinerja Finansial Kenaikan Harga Dedak Padi Terhadap Tingkat Pendapatan Peternak Ayam Petelur Di Kabupaten Blitar Jawa Timur. Jurnal Ilmiah Fillia Cendekia, 3(1), 10-15.

Haryuni, N. (2018). Methane Mitigation Technology In Ruminants To Reduce The Negative Impacts Of Global Warming. Journal Of Development Research, 2(2), 55-58.

Haryuni, N. (2021). Pengaruh Tingkat Energi dan Dosis Vitamin E-Selenium dalam Pakan Terhadap Penampilan Produksi dan Reproduksi Induk Pembibit Joper [Disertasi]. Universitas Brawijaya.

Haryuni, N. (2023). Pedoman Praktikum Nutrisi Unggas. PT. Bestindo Berkah Lestari.

Haryuni, N. (2024). Study of the Quality and Strategic Role of Corn for Poultry Industry Development. Bestindo Journal of Animal Nutrition and Forage, 1(1), 41-50.

Haryuni, N., & Fanani, Z. (2017). Study of feasibility on broiler business development. Journal of Development Research, 1(2), 63-67.

Haryuni, N., & Muanam, M. K. (2023). Potret BUMDES Sido Makmur Sejahtera. PT. Bestindo Berkah Lestari.

Haryuni, N., Anam, M. K., Mitra, I. K., Yaqin, M. A., Yeiputa, G. C., Lestari, I. D., ... & Ma'mun, M. B. (2023). Strategi Cerdas Pemeliharaan Ayam Petelur. PT. Bestindo Berkah Lestari.

Haryuni, N., Arif Tribudi, Y., Hasanah, N., & Angga Prastya, R. (2024). Improving the productivity of Joper chickens with fermented soy milk waste (SMW). BIO Web of Conferences, 88, 00043. https://doi.org/10.1051/bioconf/20248800043

Haryuni, N., Fajar, M. S. R., Kasanah, R. D., Rahayu, P., Nafi'uddin, M. A., Akbar, M. A. R., & Rif'at, F. (2023). Strategi Memilih Bibit Ayam Petelur. PT. Bestindo Berkah Lestari.

Haryuni, N., Harliana, & Alam, Y. (2024). Basic Knowledge of Animal Feed Formulation. Bestindo Journal of Animal Nutrition and Forage, 1(1), 25-33.

Haryuni, N., Hartutik, H., Widodo, E., & Wahjuningsih, S. (2021). Interaction effect of vitamin E-selenium supplementation and metabolic energy on reproductive performance of Joper Breeders.

Haryuni, N., Hartutik, Widodo, E., and Wahjuningsih, S., 2021. Interaction effect of vitamin E-selenium supplementation and metabolic energy on reproductive performance of Joper Breeders. Indonesian Journal of Animal and Veterinary Science, 26 (3), 124–131.

Haryuni, N., Hartutik, Widodo, E., and Wahjuningsih, S., 2022. Effect of energy and dose of vitamin E selenium on improving the reproduction performance of Joper brood stock. E3S Web of Conferences, 335, 00036.

Haryuni, N., Khopsoh, B., Hasanah, N & Sikone, H.Y. (2024). Potensi soy milk waste untuk optimalisasi reproduksi pejantan ayam buras. PT. Bestindo Berkah Lestari.

Haryuni, N., Khopsoh, B., Yeiputa, G. C., Lestari, I. D., Anam, M. K., Mitra, I. K., Yaqin, JM. A., Purnama, J., Atallah, M. T., Saputra, Y. E., Muchlisin, M. I., Rastika, R. (2024). Biosekuriti dan Pengendalian Penyakit Pada Ayam Petelur. PT. Bestindo Berkah Lestari.

Haryuni, N., Lestariningsih & Y. A. Tribudi. (2022). Pemanfaatan Soy Milk Waste sebagai Bahan Pakan Unggas. CV. Haura Utama.

Haryuni, N., Lestariningsih, L., & Khopsoh, B. (2022). Perbaikan Kualitas Semen Ayam Kampung Melalui Peningkatan Energi Metabolisme Pakan. Journal of Science Nusantara, 2(3), 123-129.

Haryuni, N., Lidyawati, A., and Khopsoh, B., 2019. The Effect of Vitamin E - Selenium Addition Level in Feed Against Fertility and Hatching Eggs of Sentul Chicken Crosses With Laying Hens. Jurnal Ilmiah Peternakan Terpadu, 7 (3), 287–292.

Haryuni, N., Lidyawati, A., Khopsoh, B., & Hasanah, N. (2020). Pengaruh level energi dalam pakan terhadap kualitas spermatozoa Ayam Kampung secara mikroskopis. Jurnal Ilmu Peternakan Terapan, 4(1), 7-13.

Haryuni, N., Lidyawati, A., Khopsoh, B., & Hasanah, N. (2020). Pengaruh level energi dalam pakan terhadap kualitas spermatozoa Ayam Kampung secara mikroskopis. Jurnal Ilmu Peternakan Terapan, 4(1), 7-13.

Haryuni, N., Lidyawati, A., Khopsoh, B., and Hasanah, N., 2020. Pengaruh Level Energi Dalam Pakan Terhadap Kualitas Spermatozoa Ayam Kampung Secara Mikroskopis. Jurnal Ilmu Peternakan Terapan, 4 (1), 7–13.

Haryuni, N., Pt, S., Pt, M., & Prastiya, R. A. (2023). Pedoman Evaluasi Fisik Kualitas Jagung. PT. Bestindo Berkah Lestari.

Haryuni, N., Utama, R. W. P., Santika, W. N., Hidayah, A. K., Almi, S. F., Zulfa, P. M. M., & Saifudin, M. (2023). Prospek Bisnis Ayam Petelur. PT. Bestindo Berkah Lestari.

Haryuni, N., Widodo, E., & Sudjarwo, E. (2015). Aktivitas antibakteri jus daun sirih (Piper bettle linn) terhadap bakteri patogen dan kualitas telur selama penyimpanan. TERNAK TROPIKA Journal of Tropical Animal Production, 16(1), 48-54.

Haryuni, N., Widodo, E., & Sudjarwo, E. (2017). Efek Penambahan Jus dan Daun Sirih (Piper bettle linn) Sebagai Aditif Pakan Terhadap Peforma Ayam Petelur. Sumber, 4(04), 13-16.

Haryuni, N., Widodo, E., Tribudi, Y. A., & Wahjuningsih, S. (2022). Impact of Aging on Sperm Quality of Sentul Roosters. Indonesian Journal of Animal & Veterinary Sciences, 27(4).

Hasan, M. N., Haryuni, N., & Lestariningsih, L. (2023). The Effect of Unproductive and Productive Chicken Ratio on the Feasibility of Laying Hens Business. Jurnal Ternak, 14(2), 59-66.

Hasanah, N., Haryuni, N., & Wahyono, N. D. (2024, May). The effect of EM-4 dosage in fermentation on the quality of soy milk waste (SMW) as an alternative feed ingredient to increase production cost efficiency in the poultry business. In IOP Conference Series: Earth and Environmental Science (Vol. 1338, No. 1, p. 012020). IOP Publishing.

Hasanah, N., Kustiawan, E., Nurkholis, N., Prasetyo, B., Amalia, R., Bahri, A., & Haryuni, N. (2023). Evaluasi Performa Produksi Ayam Petelur Sistem Closed House di UD. Supermama Farm Banyuwangi. Jurnal Ilmiah Fillia Cendekia, 8(2), 64-71. doi:10.32503/fillia.v8i2.3791

Hasanah, N., Wahyono, N. D., Subagja, H., & Haryuni, N. (2024, May). Optimization model: Broiler chicken farmers plasma members from the core partnership of broiler chicken farming in Indonesia for recruitment and welfare. In IOP Conference Series: Earth and Environmental Science (Vol. 1338, No. 1, p. 012069). IOP Publishing.

Khopsoh, B., Diyaningsih, M. V., & Haryuni, N. (2022). Penggunaan H2O2 (Hidrogen Peroksida) untuk Mengurangi Kadar Coliform Air Pada Peternakan Ayam Petelur di Kabupaten Blitar. Jurnal Riset dan Konseptual, 7(1), 187-196.

Nahroni, A. T., Haryuni, N., & Alam, Y. (2023). PENGARUH WAKTU SANGRAI TERHADAP KADAR AIR, KONSENTRASI AFLATOKSIN DAN KUALITAS FISIK JAGUNG UNTUK PAKAN TERNAK. Journal of Science Nusantara, 3(3), 91-97.

Prabhakar, R., Priyadharshini, M., Moorthy, M., and Edwin, S, C., 2020. Promoting Fertility in Crossbred Chicken by Artificial Insemination Promoting Fertility in Crossbred Chicken by Artificial Insemination. Veterinary Research International, 08 (02), 140–141.

Ramadani, A., & Haryuni, N. (2023). Use of Local Feed Ingredients as an Alternative to Support Productivity of Native Chickens. JURNAL PETERNAKAN (JURNAL OF ANIMAL SCIENCE), 8(1), 1–6.

Roiter, Ya, S. and Konopleva, A, P., 2020. Universal biotechnological medium for sperm dilution during poultry artificial insemination Universal biotechnological medium for sperm dilution during poultry artificial insemination. In: AGRITECH IOP Conf. Series: Earth and Environmental Science 315 (2019) 042020. 1–6.

Rozaqi, M. R., Haryuni, N., & Alam, Y. (2023). PENGARUH SUHU PEMANASAN METODE SANGRAI TERHADAP PENINGKATAN KUALITAS FISIK DAN PENURUNAN KONSENTRASI AFLATOKSIN PADA JAGUNG. Journal of Science Nusantara, 3(3), 114-121.

Sholihat, A., Wibisana, G., Wibowo, I.L., Muchtar, K., Guru, P., Ibtidaiah, M., Tarbiyah, F., Islam, U., Sunan, N., and Djati Bandung, G., 2021. No Title. In: PROCEEDINGS UIN SUNAN GUNUNG DJATI BANDUNG. 17–27.

Sikone, H.Y., Haryuni, N & Dos Santos, E.P. (2024). Kapita Selekta Sistem Produksi Ternak di Nusa Tenggara Timur. PT. Bestindo Berkah Lestari.

Tribudi, Y. A., Tohardi, A., Haryuni, N., & Lesmana, V. (2022). Pemanfaatan tepung larva black soldier fly (hermetia illucens) sebagai subtitusi tepung ikan terhadap performa ayam joper periode stater. Jurnal Nutrisi Ternak Tropis, 5(1), 45-51.

Tribudi, Y. A., Tohardi, A., Haryuni, N., & Lesmana, V. (2022). Pemanfaatan tepung larva black soldier fly (hermetia illucens) sebagai subtitusi tepung ikan terhadap performa ayam joper periode stater. Jurnal Nutrisi Ternak Tropis, 5(1), 45-51.

Usman, Tijjani, H., Mohammed, S., Haladu, S., and Kari, A., 2021. Assessment of semen characteristics among three phenotypes of chicken raised in Akko , Gombe State of Nigeria. Journal of Agrobiotechnology, 12 (1), 40–48.

Widiastuti, W.A., Bebas, W., and Trilaksana, I.G.N.B., 2018. Penggunaan Berbagai Kuning Telur Sebagai Bahan Pengencer Terhadap Motilitas dan Daya Hidup Spermatozoa Ayam Pelung. Indonesia Medicus Veterinus, 7 (3), 251.