Different Haematological Condition, Immune System and Comfort of Muscovy Duck and Local Duck Reared in Dry and Wet Seasons

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Abstract. Muscovy and local duck belong to Indonesian local waterfowl for meat and egg production purpose, whose physiological condition is strongly affected by genetic and environmental factors in which physiological condition determines duck productivity. Due to the global climate change including in Indonesia this study is important. This research aimed to study the different haematological condition, immune system and comfort of muscovy and local duck reared in dry and wet seasons. 128 muscovy and local ducks of one-day old of male and female were involved in this study with Completely Randomized Factorial Design (2x2x2). Two factors : breed of ducks (muscovy and local), sex (male and female) and season (dry and wet). The variables included the amount of erythrocyte, leukocyte, differential leukocyte, heterophile-lymphocyte ratio, hemoglobin level, hematocrit value, and total plasma protein. Data were analyzed using Analysis of Variance followed by Duncan test for any different treatment shown in the variables. The result showed that interaction among the duck order, sex and seasons significantly influenced the heamatological condition and welfare of the ducks. Muscovy duck and local duck reared in wet season had a higher haematological status than those of dry season. Leukocyte amount was higher in Muscovy duck than local duck, while Muscovy duck had a lower heterophile-lymphocyte ratio than that of local duck. In conclusion, haematological condition in wet season and dry season was different, the most influential immune system was heterophile and Muscovy duck and local duck were more in comfort in wet season.

Keywords: muscovy duck, haematological, immune system, comfort, dry season, wet season

Abstrak. Entok dan itik termasuk unggas air tradisional Indonesia untuk tujuan produksi daging dan telur yang kondisi fisiologisnya sangat dipengaruhi oleh faktor genetik dan lingkungan dimana kondisi fisiologi menentukan produktivitasnya. Penelitian ini penting dilakukan karena perubahan iklim global juga terjadi di Indonesia. Penelitian ini bertujuan menganalisa kondisi haematologis, sistem kekebalan tubuh dan kenyamanan entok dan itik yang dipelihara selama musim hujan dan kemarau. 128 bibit entok dan itik jantan dan betina berusia 1 hari digunakan dalam penelitian ini dengan Rancangan Acak Lengkap Faktorial (2x2x2). Dua facktor: bangsa (entok dan itik), jenis kelamin (jantan dan betina) dan musim (penghujan dan kemarau). Peubahnya mencakup jumlah eritrosit, leukosit, perbedaan leukosit, rasio heterofil-limfosit, level hemoglobin, nilai hematokrit, dan protein plasma total. Data dianalisa menggunakan analisis variansi dilanjutkan dengan uji Duncan apabila ada perbedaan perlakuan yang muncul dalam peubah. Hasil penelitian menunjukkan bahwa interaksi antar bangsa itik, jenis kelamin dan musim berpengaruh nyata terhadap kondisi haematologis dan kenyamanan itik. Entok dan itik yang dipelihara saat musim penghujan memiliki status haematologi yang lebih tinggi daripada yang dipelihara di musim kemarau. Jumlah leukosit lebih tinggi pada entok daripada itik, sedangkan entok memiliki rasio heterofil-limfosit yang lebih tinggi daripada itik. Disimpulkan bahwa kondisi haematologi berbeda di musim penghujan dan kemarau, sistem kekebalan tubuh yang paling berpengaruh adalah heterofil serta entok dan itik lebih merasa nyaman saat musim penghujan.

Kata kunci: entok, haematologi, sistem kekebalan tubuh, kenyamanan, musim kemarau, musim penghujan

Introduction

Muscovy duck and local duck are prominent than other poultry because of the higher immunity than chicken. Body immune against disease is one criterion in poultry breeding in forming poultry strain for industry. The indicator used to know the body immune

against disease of immunological function is haematological parameter (Talebi et al., 2005) and haematological status highly influences the poultry production level (Ismoyowati et al., 2006).

Physiological condition of poultry is strongly affected by environment namely feed nutrient and climate. Recent climate change happens in Indonesia as in all over the world as a result of global warming that causes a lot of failure in animal breeding due to disease or stress. Accordingly, it is essential to conduct a research on haematological condition, immune system and welfare of the duck reared in wet season and dry season. Environment (Vecerek et al., 2002; Graczyk et al., 2003) and physiological condition affect blood profile which shows the poultry's health like feed nutrient content (Kurtoglu et al., 2005), feed and water limit (Al-Rawashdeh et al., 2000; Iheukwumere and Herbert, 2003), fulfillment (Lamosova et al., 2004), and age (Talebi et al., 2005). Previous publication reported a different blood profile on chicken with different age and strain (Hauptmanova et al., 2002). Haematological characteristic can be used as indicator of poultry's adapting ability towards certain environmental condition. The development of poultry embryo is significantly affected by incubation temperature in which response to the environment can be measured physiologically using haematological parameter (Zhang et al., 2007). Environment to raise poultry determines the poultry's comfort and a continual farming should be considered the comfort factor of the animal (Esteves-Abe, 2008).

Poultry's welfare is a complex condition of a fowl including many aspects namely biological (physical and physiological), psychological, and behavior. Welfare level can be measured from the heterophile/lymphocyte ratio (H/L ratio) (Huff et al., 2005). Gross and Siegel (1983) also stated that H/L ratio can serve as stress indicator in chicken, even Dietert et al. (1996) stated that that indicator is applicable as the biomarker mostly proper with immunology function. Accordingly, this research studied the different haematological condition, immune system and welfare in Muscovy duck and local duck reared in dry season and wet season.

Materials and Methods

This experimental research used a total of 128 waterfowls consisted of one-week old local duck and Muscovy duck, each comprised 16 male and 48 female. Feed given during 1-14 day old was BR1 of 19% PK and 3000 kcal/kg ME, at 2-8 week old feed was given on mixed 35% ground corn, 45% rice bran, and 20% duck concentrate with nutrient feed of 15% PK, 2800 kcal/kg ME, and 1.604 % P.

The instrument used was 32 battery cages measuring 75x60x60 cm. Cage utility, sanitation tools and equipment for haematological examination consisted of standard erythrocyte pipette (101), tube, microscope, leukocyte pipette, glass object, check counter, microhematocrit, cuvette and spectrophotometer.

Research design applied Completely Randomized Factorial Design (2x2x2). Treatments consisted of three factors, factor one was waterfowl species or Muscovy duck and local duck, factor two was sex or male and female, and factor three was seasons or dry season and wet season, comprising a total of eight treatment combinations. Each treatment unit of male and female duck consisted of two and six ducks, respectively, and was repeated four times. The combinations of treatment variables are as follows (1) male local duck reared in dry season, (2) female local duck reared in dry season, (3) male muscovy duck reared in dry season, (4) female muscovy duck reared in dry season, (5) male local duck reared in wet season, (6) female local duck reared in wet season, (7) male muscovy duck reared in

wet season, (8) female muscovy duck reared in wet season.

The measured variables included the amount of erythrocyte, hemoglobin, haematocryte, total plasma protein, the amount of leukocyte, differential leukocyte and heterophile/lymphocyte ratio. Data were analyzed using Analysis of Variance followed by Duncan test for any different treatment shown in the variables.

Research procedure was initiated by raising one day old tagged ducks for 16 weeks, feed was given twice a day as needed and feed intake reduction was administered. Water was given ad libitum.

One blood sample was taken from one duck in each unit at the age of 8 week in early September for dry season blood sample, and at the age of 14 week in early November. 3 ml of blood sample was taken from *vena axillaries,* put in EDTA-filled tube, kept in ice_filled flask, the carried to the laboratory for further examination. The amount of erythrocyte of RBC (mil/ml) was counted using hemocytometer with Hagem dilution (Zhang et al., 2007). The amount of leukocyte (WBC) and differential leukocyte was measured using blood swap method colored with Wright reagent using Hematek Stain Pak. 100 WBC per blood sample was examined using Nikon microscope with 400x magnification, and so were the identification of heterophile, lymphocyte, monocyte, eosinophile and basophile. Total heterophile and eosinophile amount was counted using *Neubauer Hemacytometer* and Nikon microspcope with 100× magnification. Cell coloring used *phloxine B propylene air glycol*. The total amount of lymphocyte, monocyte, and basophile was indirectly determined from the percentage calculation of cell and total heterophile and eosinophile (Chowdhury et al., 2005).

Haematocrit was determined using Micro-capillary Reader. Blood was centrifuged using *Hettich Haematokrit centrifuge* for five minutes to have the haematocrit value stated on the Micro-capillary Reader (Chowdhury et al., 2005). Hemoglobin concentration (g/100ml)was measured using spectrophotometri. The concentration of plasma protein and blood albumin was measured using spectrophotometer, with standard protein and protein albumin solution (Pierson, 2000).

Results and Discussion

The different haematology in Muscovy duck and local duck reared in dry season and wet season

Haematological status indicated that blood profile in optimal balance or around normal parameter made the physiological process run well, leading to an optimum production. Haematological parameters observed in this research include the amount of erythrocyte, hemoglobin level, hematocrit, erythrocyte index and total plasma protein.

Analysis result showed that interaction between season, class and sex of the duck had significant influence on the amount of erythrocyte, PCV and total plasma protein level. Interaction of season and duck class significantly affected hemoglobin level. Haematological condition in dry season was lower than that in wet season. This indicated that environmental factors namely temperature and humidity strongly influenced the duck's physiology. Wet season, on the other hand had lower temperature and higher humidity that caused the ducks to consume more feed, haematological value was therefore higher than that in dry season (Olayemi and Arowolo, 2009). The highest average was 2.46±0.34 mil/µ in male Muscovy duck and the lowest was 1.90±0.16 mil/µ (Tabel 1) in female Muscovy duck. This is in line with Sturkie (1976) that erythrocyte is a blood-building component which in fowl is elliptical with core, measuring 12.8 µm and 6.6 µm in length and width diameter, respectively. While in chicken, it is

10.7 µm and 7.06 µm in length and width diameter, respectively. The amount of erythrocyte and blood cell volume was influenced by age, sex, hormone, hypoxia and other factors. Furthermore, it is stated that the amount of erythrocyte is 2 mil/mm^3 in ducks, 3.8 mil/mm^3 in rooster, 3 mil/mm^3 in hen. Olayemi and Arowolo (2009) reported that the average amount of erythrocyte in Nigerian duck (*Anas plathirynchos*) in wet season and dry season is 2.46±0.45 mil/µl and 2.02±0.56 mil/µl, respectively. Hemoglobin and hematocrit level was relatively equal in Muscovy duck and local duck, either male or female. Hemoglobin is red blood cell pigment that gives red color with complex structure containing iron, protein (globin), and pigment (heme) to bound and circulate oxygen to the tissues. One milliliter of erythrocyte contains 1.1 mg iron whose amount depends totally on hematocrit value and blood volume. This research result is lower to that of Ismoyowati et al., (2006), reporting that hemoglobin in Tegal duck of production period is 10.96-12.17 g/dl. This difference was due to the different age of ducks on sampling.

Hematocrit in Muscovy duck and local duck is relatively equal in both sexes. Hematocrit is the comparison of erythrocyte and blood plasma in percent volume, in which hematocrit level is positively correlated with the amount of erythrocyte and hemoglobin level. Hematocrit value is related to the need of oxygen dealing with metabolism product. The average value of hematocrit in male Muscovy duck is biologically higher than that of female Muscovy duck and local duck, owing to the higher body weight of male Muscovy duck than other waterfowls. This result is contradictory with that of Elagib and Ahmed (2011) that Sudan rooster had higher hemoglobin and hematocrit than those of hen. The highest average of MCV and MCH is in female Muscovy duck, demonstrating different erythrocyte volume of Muscovy duck and local duck in both sexes, because of water content in the blood and blood viscosity (Olayemi and Arowolo, 2009).

Variance analysis also showed significant difference of total protein plasma and albumin in Muscovy duck and local duck as well as in male and female local ducks. Plasma protein consists of three protein fractions namely albumin, globulin, and fibrinogen. According to Harper et al., (1984), total plasma protein level is 3.2-5.6 g/dl, albumin is 52-56% and globulin is 29.4-54%. Plasma protein functions as body protein reserve, circulates non-statically, exchanges continuously with unstable tissue reserve whose amount is equal with circulating protein to make a dynamic balance. When body lacks of protein, it takes protein and plasma protein reserves from tissues for metabolism as happens in Muscovy duck with the highest body weight rather than in local duck due to a high metabolism for meat synthesis so body takes

Tabel 1. Average amount of erythrocyte, hemoglobin, PCV value, and total blood plasma protein of Muscovy duck and local duck in dry season and wet season

Season	Water fowl	Erythrocyte (mil/ul)	Hb (g/dl)	PCV (%)	TPP (g/dl)
Dry season	MMD	2.46±0.34^{bc}	7.81±1.85^{a}	38.38±7.54^{ab}	4.38±2.37^{a}
	FMD	1.90±0.16^{a}	8.19±1.07^{a}	34.00±6.02^{a}	3.85±1.92^{a}
	MLD	2.08±0.35^{a}	8.40±0.91^{a}	35.63±5.21^{ab}	5.80±1.39^{b}
	FLD	2.31±0.38^{b}	7.76±0.76^{a}	34.63±7.27^{a}	7.13±1.45^{c}
Wet season	MMD	2.88±0.24^{cd}	8.02±0.95^{a}	38.70±1.89^{ab}	4.06±0.46^{a}
	FMD	3.01±0.26^{d}	8.31±0.55^{a}	40.50±3.84^{b}	3.40±0.35^{a}
	MLD	3.14±0.25^{d}	9.09±0.66^{b}	41.80±3.46^{b}	5.06±0.91^{ab}
	FLD	2.70±0.45^{c}	8.85±0.41^{b}	40.80±1.23^{b}	3.86±0.46^{a}

MMD : male muscovy duck, FMD : female muscovy duck, MLD : male local duck, FLD : female local duck
Values bearing different superscript at the same column differ significantly in Tukey test

plasma protein to meet the need of protein.

The different immune system and comfort level of Muscovy duck and local duck reared in dry season and wet season

Measuring poultry comfort level can be done using several parameters such as physiology parameter including hormonal level, heartbeat (Craig et al*.,* 1986), and immunological status (Gross and Siegel, 1983). As stated by Eicher (2003) that multi-disiplinary approach towards poultry comfort has to include immunological status. Moreover, immunologically the first response to stress is neutrophile circulation increase, phenotypic lymphocyte change, cell activity change and activation against acute stress response. Ismoyowati (2007) stated haematological status indicated that blood profile with an optimum balance or around normal parameter made the physiological process in the fowl run well, leading to an optimum production as well.

The measurement of fowl comfort can be seen from measuring the heterophile/ lymphocyte ratio (H/L ratio) (Huff et al*.,* 2005). Gross and Siegel (1983) also stated thay H/L ratio can be used as stress indicator in chicken haematologically, even Dietert et al., (1996) mentioned that this indicator can be used as the most appropriate biomarker with immunological function. Pingel (1999) stated that heat stress increased heterophile/ lymphocyte ratio which is more liable as fowl comfort than blood corticosterone level. Fujita et al., (1998) stated that fowl in welfare or in normal physiological condition and in environment with H/L value lower than distressed or under pressure fowls. While research result by Ismoyowati (2007) reported that highly productive local duck has average H/L ratio of 0.417± 0.125 lower than H/L in population.

Variance analysis shows interactions of seasons, duck class, and sex variables has significant effect towards ($p<0.05$) the amount of leukocyte, differential leukocyte and heterophile/lymphocyte ratio as the stress indicator in fowls. The highest amount of leukocyte was 9110,00±2845.21 and the highest was 3625.00±1233.75/µl in female local duck and male local duck, respectively. The amount of leukocyte In Nigerian duck was 16,93 x 10^9/l (Olayemi et al., 2006) and 6.58x10^9/l in wet season and 22.55x10^{9x}/l in dry season (Olayemi and Arowolo, 2009). The different leukocyte is caused by genetic and fowl age. The main function of leukocyte is as body immune system, this result indicated that the different leukocyte was due to different heterophile. Heterophile functions as phagocytosis to defend against pathogen microorganism through the lysis of cell wall (Moyes and Schute, 2008), this shows that immune system in female local duck applies phagocytosis mechanism.

Season is strongly related to the stress condition of both Muscovy duck and local duck. In wet season fowls are in a more welfare with lower heterophile/lymphocyte than that in dry season. The research result showed that heterophile/lymphocyte ratio was relatively equal in Muscovy duck and local duck in both sexes. Campo et al., (2000) stated that one of stress indicators in fowl is heterophile/lymphocyte ratio. Fujita et al., (1998) stated that fowls in welfare or in normal physiological condition and proper environment have a relatively lower heterophile/lymphocyte ratio than distressed and under pressure fowls. The result showed that biologically local duck has a higher stress level than Muscovy duck. Stress condition is observable from the restless local duck, and being recluse when approached, Muscovy duck on the other hand is calmer. Bounous and Stedman (2000) reported that heterophile and lymphocyte are the most component from

Tabel 2. Average of leukogram, differential leukocyte, and heterophile/lymphocyte ratio of Muscovy duck and local duck in dry season and wet season

Season	Water fowl	Leukocyte (mil/ul)	Heterophile (%)	Eosinophile (%)	Lymphocyte (%)	Monosit (%)	H/L
Dry season	MMD	6368.75±3279.04^{ab}	49.13±16.23^{ab}	6.25±3.33^{ab}	35.38±13.86^{ab}	7.50±7.75^{ab}	1.81±1.27^{ab}
	FMD	5187.50±3528.33^{a}	52.13±10.96^{ab}	4.63±2.56^{ab}	29.75±9.42^{ab}	11.50±4.63^{b}	2.10±1.45^{ab}
	MLD	3625.00±1233.75^{a}	54.75±18.29^{bc}	14.13±7.90^{b}	25.13±13.58^{a}	6.00±3.16^{ab}	3.14±2.48^{b}
	FLD	9110.00±2845.21^{b}	69.75±8.24^{c}	3.13±3.31^{a}	23.00±7.56^{a}	4.13±3.14^{a}	3.45±1.52^{b}
Wet season	MMD	10795±3546.24^{b}	41.50±8.76^{ab}	14.20±6.43^{b}	36.60±11.46^{ab}	7.70±3.50^{ab}	1.31±0.67^{a}
	FMD	9770.00±2148.15^{b}	33.60±9.81^{a}	11.60±5.91^{ab}	44.80±11.96^{b}	10.00±3.37^{b}	0.90±0.69^{a}
	MLD	9525.00±2285.01^{b}	39.90±13.82^{ab}	11.40±7.32^{ab}	42.30±14.86^{ab}	6.40±2.46^{ab}	1.21±0.92^{a}
	FLD	5520.00±792.74^{a}	39.30±18.26^{ab}	24.50±11.53^{c}	27.50±17.71^{a}	8.70±5.31^{ab}	2.80±2.85^{b}

MMD : male muscovy duck, FMD : female muscovy duck, MLD : male local duck, FLD : female local duck
Values bearing different superscript at the same column differ significantly in Tukey test

leukocyte which is the first defense against infection and breaking down the foreign matter through phagocytosis process. Campo et al., (2000) stated that the optimum ratio of heterophile and lymphocyte will bring welfare to fowl to relieve stress. Shini (2003) and Wang et al., (2003) also reported that H/L ratio in layer chicken and broiler reared in a proper cage had a relatively lower H/L ratio.

Conclusions

Local duck and Muscovy duck had a different haematological condition, and there was interaction of season, duck class and sex. The most significant immune system in both classes was heterophile. Muscovy duck and local duck had more comfort in wet season.

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