The goals of this research were to estimate the phenotypic relationship among various breast meat quality traits, blood parameters and tonic immobility from a broiler and to describe the relation among these variables. Broiler chicks were divided into different groups: (1) colour clothing groups represented four different colours as red, blue, green, and gray and (2) the chicks that were subjected to visual human contact for 60 and 300 sec. A total of 192 chicks, one day-old, were randomly assigned to eight treatment groups according to the colour clothing and visual human contact. Hatching weights for red, blue, green and gray clothing colour groups were found as 47.10, 48.35, 46.67, and 47.05 g, and for 60 and 300 sec visual human contact groups were determined as 47.34 and 47.24 g, respectively. Cooking loss had a significant negative relationship with pH at 15 minutes after slaughter (pH 15) in the red clothes group. Water holding capacity had a negative correlations (r=-0.355 and -0.489) with pH at 24 hours after slaughter (pH 24) in the 60 and 300 sec groups, respectively. Total protein level had a high degree and positive correlation (r=0.675) with cholesterol level in the red clothes group. Tonic immobility (TI) duration had significant relationship with TI induction in the green clothes group. The a* colour value of breast meat showed a moderate negative and significant correlation with the variables L* value of breast meat (-0.574 (P<0.01) and -0.373 (P<0.05), respectively) in gray clothing and 300 sec visual contact group. These finding have a lot of implications on the use of gray colour clothing and 300 s visual contact to improve meat quality for welfare in broilers.

**KEY WORDS** blood parameters, meat quality, relationship, tonic immobility, visual contact.

**INTRODUCTION**

Broiler breeding in intensive management systems may expose human eye contact (Zulkifli et al. 2002). Human factors affect fear of poultry which may lead to a reduction in the productivity of poultry performance. Normally, since the routine rearing procedure, poultry fears humans because of (Vasdal et al. 2018) different kinds of stressors (e.g. stockpersons, noise, and lighting) (Waiblinger et al. 2006). However, poultry can easily adapt to human presence and human visual and physical contact. Studies have demonstrated that human contact has a positive effect in reducing fear in poultry (Hemsworth et al. 2002; Zulkifli and Siti Nor Azah, 2004; Bassler et al. 2013; Edwards et al. 2013), and this causes to improve the production and immune function, which are two indicators for determining reduced stress response (Hemsworth et al. 2002; Zulkifli et al. 2002; Zulkifli and Siti Nor Azah, 2004; Lentfer et al. 2015). There is also a study reporting the evidence that poultry are somewhat sensitive to visual contact with humans (Hemsworth, 2009). Vision is important to bird, and this reflected in the fact that the avian eye is particularly large in relation to both the head and the brain. Studying the visual perception of birds have shown that they respond to
visual stimuli very much like human beings. The color preference is important in the domestic chickens (Zhang et al. 2012). Studies on this subject are usually on stockpersons’ clothing, coloured feeders and food, water, enrichment devices, illuminated targets, and imprinting objects. Kovach and Kabai (1993) reported that birds are adjudged via long wavelengths, giving colours that they see as red, yellow or orange and this condition is responce spectra of visual markers. Welfare has been assessed from blood glucose, triglyceride, lactate, cholesterol, and lactate dehydrogenase (LDH), total protein, and meat quality traits (Zhang et al. 2009; Deep et al. 2010). In poultry, color variations in meat have received considerable attention from researchers because of their direct influence on consumer acceptance and high relationship with the functional characteristics of meat (Alkan et al. 2010; Narinç et al. 2013; Narinç et al. 2015). Fresh raw breast meat has a pale pink color. Also, sustaining birds in good health, with high welfare and health standards, results in good quality meat products (Sundrum, 2001). Producers should be concerned with factors that may negatively affect these important meat quality traits characteristics (Qiao et al. 2002). Fletcher et al. (2000) established a marked relationship between pH and extreme colour variations, while Salakova et al. (2009) reported that negative relationship between breast meat yellowness (b*), lightness (L*), and pH values, but there were positive relationship between breast meat L*, b* and redness (a*). The aim of this study was to determine relationships between breast meat quality traits, some blood parameters, and TI in broilers under the regular visual contact by the stockperson wearing clothes in different colours.

**MATERIALS AND METHODS**

The following procedures related to animal handling and sample collections were approved by the ADU Animal Experiments Local Ethic Council (No:64583101/2014/049). A total of 192 male broiler chicks, one-day old (Ross 308) were purchased from a commercial hatchery. On day one, broiler chicks were housed in floor pens covered with wood shavings.

Broilers reared at 12 broilers/m². Hatching weights for red, blue, green, and gray clothing colour groups were 47.10, 48.35, 46.67, and 47.05 g, respectively. In addition, the hatching weights were 47.34 and 47.24 g in both visual human contact groups (60 and 300 sec). Feed and water were given ad libitum throughout the study. The broiler chick ration given between days 1-21 contained 22% of crude protein and 3100 kcal/metabolic energy/kg, while the broiler chicken ration given between days 22-42 contained 21% of crude protein and 3250 kcal/metabolic energy/kg.

The ambient temperature was gradually decreased from 32 ± 1 °C on d 1 to 23 ± 1 °C until end of the study (day 42) (NRC, 1994).

The study was conducted as 4 × 2 factorial experimental design: (i) clothing groups including four different colours (red, blue, green and gray). (ii) two visual human contact groups at a duration of 60 and 300 s. Each group consisted of 2 replicates of 12 broilers per pen. The size of pen was 110 × 103 cm. The chickens were subjected to regular visual human contact once a day by the same stockperson who wore the different colour clothings from day 15 to end of the study.

In the visual contact procedure, the stockperson wearing clothes with different colours entered the pens slowly with the minimal noise and stood up in the centre of the pens without any physical contact with chickens. On day 41, 13 broilers of each group (N=104 broilers in total) were randomly selected to perform TI test. Chickens was gently carried to a quiet room. Each chicken was carefully restrained for 15 s by covering the head with one hand, while placing the other hand on the sternum. A chronometer was used to record latencies until the chicken righted itself. If the chicken righted itself in < 10 s, the procedure was repeated.

If TI test was not happened after three trials the TI duration was noted as zero. The maximum duration of TI allowed was 10 minutes (Jones and Faure, 1981). On day 42, blood samples were obtained between 08:00 am and 09:00 am by brachial vein of 11 birds from each group. Blood parameters were measured using commercial reagents by a spectrophotometry (Shimadzu UV-1601, Germany). To determine meat quality traits, on day 42, 11 birds from each group (total 88) were slaughtered by exsanguination through a neck cut. Meat quality analysis was done using breast muscle (pectoralis major). The pH value was measured with a portable pH meter 15 min (initial pH, pH15) and 24 hours (ultimate pH, pHu) after the right pectoralis major was collected. Meat colour of skinless breast meat samples were determined using a Minolta CR 400 colorimeter. The cooking loss (CL) was evaluated in the meat samples according to Honikel (1998). The holding capacity (WHC) was calculated 24 h after slaughter, using the methodology as previously described by Barton-Gade et al. (1993). Statistical analyses were done by using SPSS 22.0 (SPSS, 2013).

General lineer model (GLM) was designed to reveal the effects of the colour of clothing and duration of visual contact on blood variables, meat and carcass traits, and TI durations. The relationship between meat and carcass traits, blood parameters and TI durations were determined using Person's correlation coefficients.
RESULTS AND DISCUSSION

In study, the variable a* showed a moderate negative and significant correlation with the variables L* (-0.574 (P<0.01) and -0.373 (P<0.05), respectively) in gray clothing and 300 sec visual contact group (Tables 2 and 3). It was showed that a high a* value, probably due to the high heme pigment that may cause a low relationship between L* and meat quality traits in gray clothing and 300 s visual contact group. This correlation agrees with those reported by Barbut (2009), Bianchi et al. (2007) who reported that dark broiler breast meat significantly lower L* and higher a* value in breast fillets. Zhuang and Savage (2010) found that for breast fillets pre-sorted according to their L* values, redness of paler fillets was decreased. Qiao et al. (2002) reported that increased mean broiler fillet lightness values were associated with decreased a* values among light, normal, and dark categories. Overall, this indicates an inverse relationship between redness and lightness on a given surface. According to these results, it can be said that the broiler breast meat used for the study sensory appearance comparisons had characteristic physical and functional properties for each meat color parameter. A researcher have concluded that selection for rapid growth resulted in a higher L* but a lower a* in broiler breast meat (Narinç et al. 2013).

The b* value had a significant negative correlation with pHu in the red and blue clothes groups (-0.491 and -0.500, respectively) (Tables 1 and 2), and in the 60 and 300 sec visual contact group (-0.371 and -0.444, respectively) (Table 3). The indicated that a high pHu value would decrease b* value (r=-0.491) in the red clothing group. The inverse relationship between yellowness and pHu, there is evidence for a negative phenotypic correlation between yellowness and ultimate pH. In parallel line with the results presented, negative phenotypic relationship between yellowness and ultimate pH were reported (Nadaf et al. 2007).

Some studies (Zulkifli and Siti Nor Azah, 2004; Edwards, 2013) report that regular human contact has an important positive effect on fear and stress. It was thought that there was a decreasing trend in fear and stress level related to human visual contact in broilers. The decline in pH after cessation of the blood flow at slaughter is caused by the accumulation of lactic acid during rigor development. Declining pH induces alterations in the arrangement of myofilaments, thereby expelling fluids. In consequence, reflectance and light scattering could be increased, causing elevated instrumental lightness values due to a decreased light path through meat (Swatland, 2008). We found that relationship between initial and ultimate pH was not significant, probably due to the fact that these two parameters were under the influence of different factors.
The pH15, which is an indicator for the rate of pH decline, had an opposite correlation with L* and a* values compared with pHu. However, similar results were reported by Berri et al. (2005). The pectoralis major muscle between pHu and WHC correlation value (-0.355) were lower in 60 sec birds.
when compared to those of 300 sec birds (-0.489). WHC had a significant moderately negative correlation with pHu in the blue, green, and gray clothing groups, respectively. This relationship agrees with those reported by Silva et al. (2011) who observed that the negative, moderate correlation between breast meat pH at 24 hours postmortem and WHC was significant in poultry.

The determined that WHC values were higher in fillets exhibiting lower pHu in birds similar with result of Silva et al. (2011) who reported a significant negative correlation between pHu and a* values (r=-0.25). The results presented that broiler breast meat with higher ultimate pH is often characterised by increased tenderness, while not tenderness in the blue, green, and gray clothes groups, because it was negative correlation between pHu and WHC value. Le Bihan-Duval et al. (2001) reported that the drip loss of raw meat, a measure of WHC, was highly correlated with pHu (-0.83) and also with L* (0.80). Increasing pHu (or decreasing L*) should contribute to the improvement of the WHC of broiler breast meat.

Increased levels of LDH level were correlated with lower CL (r =-0.480 and -0.429, respectively) in gray clothing and 300 sec visual contact group.

As in this study, increase in lactate concentration is correlated with stress, which indicated a detrimental effect of stress on meat quality (Edwards et al. 2010; Dokmanovic et al. 2015).

These results supported that the statement that LDH level can be used as a marker of meat quality. In study, cholesterol level had a positive correlation with CL in the 300 sec visual contact groups (r=0.366). It was found that the b* value was negatively correlated with the pHu value in 60 and 300 sec visual contact group. A positive correlation can be found between blood total protein level and pH15 in green clothing group (0.529). Dokmanovic et al. (2015) indicated that negative, moderate correlation between breast meat pH at 60 minute postmortem and lactate level (-0.34) in broiler. The correlations between TI duration and induction were found as -0.122, -0.150, 0.398 and -0.049 for red, blue, gren, and gray clothing groups, respectively. The correlations between TI duration and induction were found as -0.108 and 0.167 for 60 and 300 sec visual contact groups, respectively. A significant correlation of 0.398 was observed between TI duration and induction in green clothing group. It has been reported a positive correlation between antecedent fear state and the duration of TI (Hemsworth, 2009).

**CONCLUSION**

There are correlations among the meat colour parameters (L*, b*, and a* colour value) and the other meat quality traits in broilers. Higher LDH level was associated with lower CL suggesting that lactate could be a marker of both meat quality. It can be said that this welfare indicators can be positively affected by the gray clothing and 300 sec visual contact used. This positive effect can be attributed to the improvement of the vision contact duration of the birds by the gray colour clothing and increase in the animal welfare.

References


