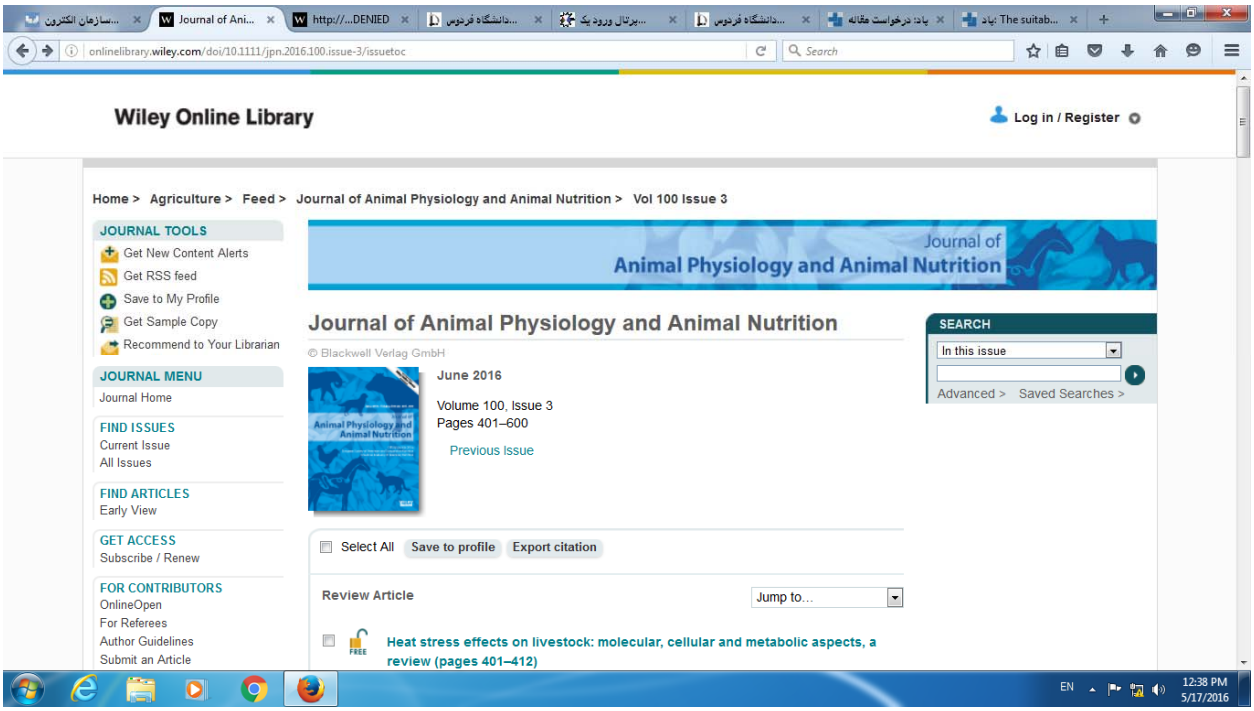

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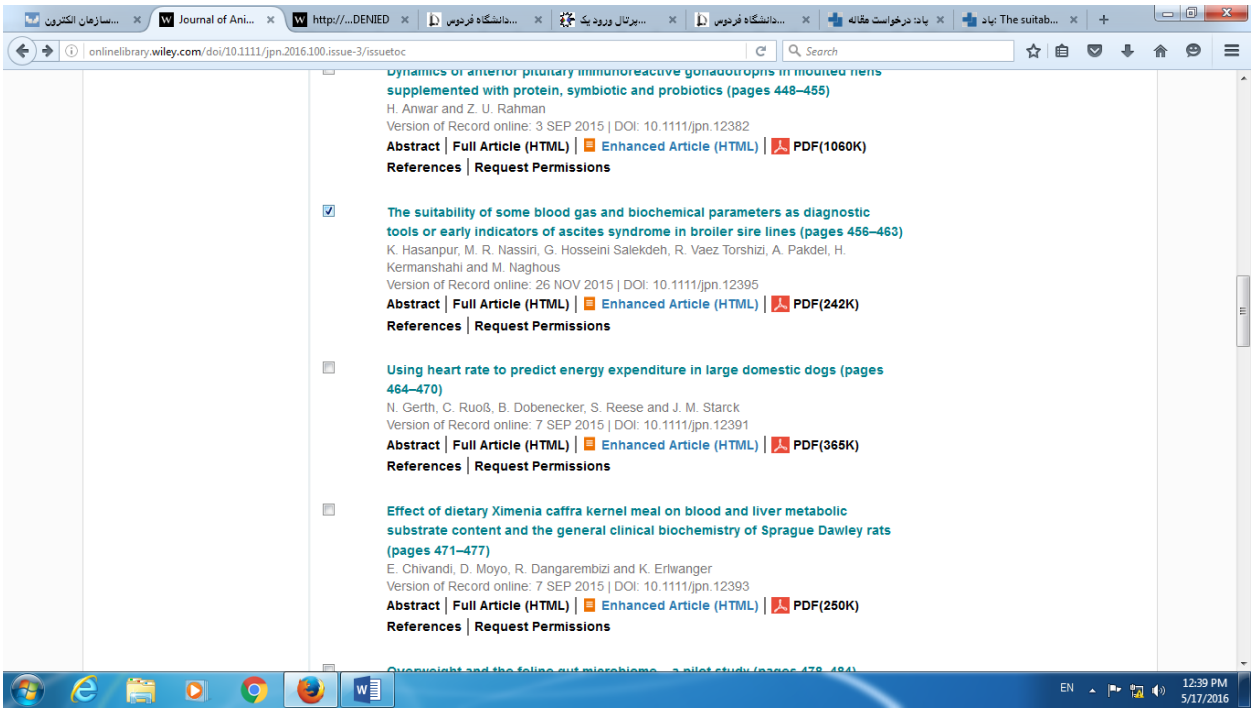

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ORIGINAL ARTICLE

The suitability of some blood gas and biochemical parameters as diagnostic tools or early indicators of ascites syndrome in broiler sire lines

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Summary

In recent few years, there have been some attempts to find a reliable indicator trait as a selection criterion against susceptibility to ascites syndrome (AS). Blood parameters were of great interest as they could be measured in live animals without implementing an ascites-inducing challenge (AIC). In this work, the suitability of some blood parameters was evaluated for diagnosing AS-susceptible chicks in later steps of the disease in trial 1 as well as their early predictive ability in trial 2. In the first trial, one hundred 1-day-old chicks from two pure broiler lines namely S_1 and S_2 and, in the second trial, 226 1-day-old chicks from line S_2 were subjected to AIC. Saline drinking water (1200 mg/l) and lower-than-standard ambient temperatures were the implemented AICs in trials 1 and 2 respectively. The blood parameters including pH, partial pressure of O_2 (pO_2), partial pressure of CO_2 (pCO_2), bicarbonate ion concentration (BIC), percentage of haematocrit (HCT) and saturated haemoglobin (SaO_2) were measured twice per each bird at days 28 and 35 in trial 1 and once in trial 2 at day 21. The results of the first trial revealed that in line S_2 some of the blood parameters differed significantly between the ascitic and non-ascitic groups following exposure to AIC. In this line, the incidence of AS was accompanied by a lower pO_2, SaO_2 and BIC, while with higher pCO_2 and HCT values. In the second trial, however, although almost all of the parameters showed meaningful differences between the ascitic and non-ascitic broilers, only mean difference of BIC parameter was statistically significant. The general conclusion of this study is that the blood parameters can somewhat have diagnostic ability in the condition in which the AIC is already present, whereas the results did not approve their usefulness as early predictors of AS.

Keywords pulmonary hypertension syndrome, broiler sire lines, blood gas parameters

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Introduction

Ascites syndrome (AS) is called to a metabolic disorder in broiler chickens that a great deal of fluids, originated from blood plasma, accumulates in the abdominal cavity or pericardium spaces (Wiedman et al., 2013). It is frequently seen in meat-type, rapid-grower chickens which are allowed to achieve their maximum growth potential (Druyan et al., 2008; Huchzermeyer, 2012). This is because of the inherent inefficacy of cardiopulmonary system of AS-susceptible chickens in provision of the ever-increasing demand for O_2 (Lister, 1997; Wideman et al., 1998; Van As et al., 2010; Huchzermeyer, 2012). Therefore, it is, to some degree, under the control of genetic factors. This means that even within a pure line, there are some families which are considerably resistant, while some others are extremely prone to develop AS (Wideman and French, 2000; Druyan and Cahaner, 2007; Druyan et al., 2007a; Pavlidis et al., 2007; Druyan, 2012; Huchzermeyer, 2012). This is also because of some exogenous factors that trigger the onset of AS (Hassanzadeh, 2010; Druyan, 2012; Huchzermeyer, 2012).

Although there is no consensus about the origin of AS in broilers which are reared at sea level, the most probable mechanism of the syndrome was illustrated

more recently by Huchzermeyer (2012) and Wiedman et al., (2013). Shortly, the effects of endogenous and exogenous factors, alone or in combination, cause hypoxaemia and hypercapnia in the bird's body and these changes, in turn, enforce the body metabolic pathways to begin some compensatory reactions.

These adaptive modifications are somewhat successful, unless the needed O_2 exceeds the cardiovascular capacity. When this threshold reached by, the cardiopulmonary system could no longer supply the body metabolism with fresh O_2. In the case of severity, the compensatory events develop vicious cycle and, therefore, the cascade of events that eventually leads to AS begins to occur (Wideman et al., 1998).

To date, a number of researchers attempted to test for the usefulness of some haematological indices to identify the main trigger of AS. Although they were not completely successful, each of them introduced one of the blood parameters as a predictor of AS or as an indicator trait for indirect selection against AS susceptibility (Maxwell, 1991; Shlosberg et al., 1996, 1998; Van As et al., 2010; Navarro et al., 2006; Druyan et al., 2007b, 2009). Among many blood parameters, the pCO_2 (Scheele et al., 2003, 2005; Hassanzadeh et al., 2010) and SaO_2 parameters (Wideman et al., 1998; Navarro et al., 2006; Wiedman, 2013) have been reported as more reliable criteria in predicting the susceptibility of the chicks to AS. However, because of the varied successes and also because some of the studies failed to find the potential differences between the ascitic and non-ascitic chickens in the rearing condition without AIC, that is in normal commercial condition (NCC), the suitability of the blood parameters has become questionable. For example, in the study of Druyan et al. (2007b, 2009) SaO_2 values were almost similar in the ascitic and non-ascitic groups when the chickens were reared in NCC, but were significantly different after exposing the chicks to AIC. Even more disappointing results were reported by Closter et al. (2009) who rejected the usefulness of blood gas parameters as a good indicator trait for reduced AS susceptibility.

In this research, we aimed (i) to test for the suitability of blood parameters as diagnostic tools in trial 1 in which the blood parameters were measured 3–4 weeks after the commencement of AIC and (ii) to test for the suitability of the blood parameters as early predictors of AS in the condition in which they were measured just before the beginning of the challenge, that is in NCC (trial 2). It is apparent that if and only if the results of later trial would reveal significant differences in the blood parameters between the ascitic and non-ascitic chickens, then they could be introduced as indicator traits for indirect selection against susceptibility to AS.

Materials and methods

Rearing protocol and AS-inducing conditions

Two separate trials were designed for this work using two pure broiler lines, namely S_1 and S_2. These are sire lines of Arian strain that imported to Iran approximately two decades ago in order to provide the broiler hybrid requirement of the country. Lines S_1 and S_2 have been under intense selection for improved FCR and heavy BW, respectively, in the past.

In trial 1, one hundred 1-day-old chicks from line S_1 (51 chicks; 26 males and 25 females) and line S_2 (49 chicks; 25 males and 24 females) were used. These birds were chosen from different families and not allowed to be half- or full-sibs. All of the chicks were wing-banded in the hatchery for individual identification. The house temperature was set around 33–35 °C at days 1 and 2 and was gradually decreased by 1 °C every other day until 21 °C was achieved at day 21 and kept nearly constant until the end. A three-phase feeding regime was applied. Energy (cal/kg of ME) and crude protein (g/kg) contents of the starter, grower and finisher diets were 2800 and 230, 2910 and 205, and 2965 and 187 respectively. The three diets were provided ad libitum from days 1 to 10, 11 to 24, and 25 to 54 respectively. A constant lighting programme was applied in the first 2 days of age and followed by a 23 h/day illumination and 1 h/day darkness regime afterwards. In the first week of age, a standard rearing condition was provided for the birds. AIC was started at day 8 at which the concentration of salt in drinking water was increased from 12 mg/l to 1200 mg/l and kept constant until the end of the rearing period, that is day 54.

As it will be seen in detail later, the blood parameters were significantly different between the ascitic and non-ascitic chickens of line S_2 in trial 1. In addition, our previous findings showed that the frequency of AS was considerably higher in line S_2 than in line S_1 and, unlike for line S_1, the growth-related traits of line S_2 had no positive genetic relationship with AS (results not shown). Therefore, line S_2 was selected for further investigation in trial 2. This trial was started in late winter; therefore, it was possible to induce AS by exposing the chicks to cool ambient temperature as AIC. A total of 226 1-day-old chicks (107 male and 119 female chicks) were used. The birds were reared under normal temperature protocol in the first 21 days of age as mentioned above for the trial 1 and then exposed to a lower-than-standard

temperature afterwards with ambient temperature fluctuating 15–18 °C during the daytime and 10–15 °C during the night-time (Druyan et al., 2007a,b). To further induce AS in the majority of the AS-susceptible chicks, the energy levels of the three starter, grower and finisher diets were increased by 3000, 3050 and 3100 cal/kg of ME. The remaining experimental protocols were as same as in trial 1.

Measurement and data collection

In trial 1, the AIC was commenced at day 8; however, the blood parameters were measured twice from the bird at days 28 and 35 of age with the aim to study the effectiveness of the blood parameters in diagnosing the ascitic from non-ascitic chicks.

The objective of trial 2 was a bit different in which we aimed to study the suitability of blood parameters as early predictors of AS in the condition in which no earlier AIC was implemented for the chicks. In this trial, the measurement of blood parameters was carried out at day 21 on all 226 chicks and the AIC started soon after the sample collection process finished.

In both of the trials, venous blood was taken from wing vein and collected in heparinized tubes which were already placed on ice. Except for haematocrit (HCT), which were measured by centrifugation of the blood as a volume percentage (12 000 rpm for 5 min), the remaining haematological characteristics including pH, bicarbonate ion concentration (BIC), partial pressure of O_2 (pO_2), partial pressure of CO_2 (pCO_2) and percentage of saturated haemoglobin (SaO_2) were measured using a blood gas analyser (ABL 605; Radiometer systems, Copenhagen, Denmark). It should be noted that some of the birds in trial 1 died before the time of blood sampling. Moreover, some blood samples failed during the measurement. Thus, the number of samples was reduced to 95 in day 28 and 86 in day 35. Chicks were weighed individually once a week in all over of the experiments.

For diagnosis of AS, all of the chicks that died after the commencement of AIC were necropsied and examined to determine the cause of death. Chicks with ascitic fluid or hydropericardium were considered as having died from AS and therefore recorded as AS-susceptible. Moreover, all of the birds that survived until the end of rearing period were examined for AS symptoms after killing by cervical dislocation. Also in here, those with above-mentioned signs of AS were recorded as being AS susceptible, while the remaining were considered as AS-resistant. The experimental protocols were reviewed and approved by the Animal Care Committee of the Ferdowsi University of Mashhad, Iran.

Statistical analyses

Blood parameters of chicks from lines S_1 and S_2 in trial 1 and from line S_2 in trial 2 were analysed separately using the GLM procedure of SAS software (SAS institute Inc., Cary, NC, USA). The used model was as below.

$$y = \mu + \text{Sex} + \text{HS} + e$$

where HS (health status: ascitic or non-ascitic) and sex considered as fixed effects. Tukey's multiple-range method was used for the comparison of the least square mean differences of ascitic and non-ascitic chickens.

In addition to significance test, pairwise Pearson's correlation coefficients between blood parameters and some growth-related traits including body weight (BW) and growth rate (GR) traits were also estimated using the CORR procedure of SAS software.

As the main objective of this study was to identify the reliable parameters to be introduced as indirect selection criteria against AS susceptibility, the first type error level was considered as 0.10 rather than 0.05. Therefore, the significance level of 90% will be called 'suggestive', while the significance level of 95% will be called 'significant' in the remainder of this manuscript.

Results and discussion

Data description

The average venous blood pH, BIC, pO_2, SaO_2 and HCT values of chickens in trial 1 were higher than in trial 2 (7.6, 38.7 mmol/l, 46.3 mmHg, 88.2% and 33.8% vs. 7.4, 27.7 mmol/l, 43.2 mmHg, 74.8% and 33.4% respectively), while average pCO_2 value was lower (42.1 mmHg vs. 43.7 mmHg). These differences could be resulted from the effects of age, line and the rearing condition factors. In trial 1, the blood parameters were measured on two lines, at 28 and 35 days of age and almost 3–4 weeks after exposing the chicks to AIC, while in trial 2, the blood parameters were measured only on line S_2 under NCC and at 21 days of age. Also, chickens of trial 1 were on average heavier in almost all over of the rearing period as compared to the chickens of trial 2 (results not shown). Because of these confounding effects, the comparison of blood parameters between the two trials was not carried out. The average blood parameter values of this study

were almost as in a similar range as the reports of Closter et al. (2009).

Average BW and GR of lines S_1 and S_2 at 21, 28 and 35 days of age are reported in Table 1. For line S_1, the average BW and GR traits were 1335 and 46.0 g at day 28 and 1918 and 53.5 g at day 35 respectively. For line S_2 in trial 1, these two traits were 1449 and 50.3 g at day 28 and 2066 and 57.9 g at day 35 and, in trial 2, the average BW and GR at day 21 were 611 and 27.6 g respectively.

The difference of blood gas parameters between ascitic and non-ascitic chickens

The results revealed that the blood parameters' mean differences of ascitic and non-ascitic chickens were more obvious in trial 1 than in trial 2. In line S_1, none of the blood parameters differed significantly between the ascitic and non-ascitic chickens, neither at day 28 nor at day 35 (Table 2). Whereas there were significant differences between the two groups in line S_2. BIC and HCT mean values of ascitic chickens on day 28 were significantly lower and higher, respectively, than those of non-ascitic chickens. Also, SaO_2 mean value of ascitic chickens on day 35 was significantly lower than that of non-ascitic chickens ($p < 0.05$). Furthermore, the differences of pO_2 ($p = 0.06$) and pCO_2 ($p = 0.06$) parameters were suggestive between the two groups (Table 3).

In trial 2, although there were a little meaningful differences between the mean values of ascitic and non-ascitic groups, except for BIC ($p = 0.03$), the remaining parameters were not statistically significant ($p > 0.10$; Table 4).

As mentioned above, the blood parameters in trial 1 were measured under the condition that the AIC was already present; thereby, the differences between the blood parameters' mean values in the two non-ascitic and ascitic groups were, undoubtedly, due to the combined effect of both genetic susceptibility/resistance to AS and bad rearing environment. Indeed, AS is known to be the result of the genetic–environment interaction effect that the bad rearing condition stimulates the incidence of AS in birds that are genetically prone to develop AS (Pakdel et al., 2005a,b; Hassanzadeh, 2010). The results of these kinds of experimental conditions can solely be used to clinically identify the birds with advance, developed stages of AS.

In trial 2, however, because the blood parameters were measured under the absence of AIC, the existing differences were merely due to the effect of genetic factors. As it can be seen in Tables 2–4, the differences of blood parameters' mean values between the ascitic and non-ascitic groups were large enough only in trial 1 (absolute difference of 0.4–20.6% in blood parameters between the two groups) to be statistically significant ($p < 0.10$). In the second trial, however, although there were seemingly meaningful differences, almost all of them were small and were not statistically significant. These results indicate that tracing the blood parameters could not be as effective as early predictors of AS. It might be noteworthy that making use of advanced instruments to measure blood parameters can enable us to measure them more precisely and with small standard errors. Therefore, using the blood parameters might be more useful in near future even as early predictors of AS when measured at NCCs.

Concurrent efforts have recently been focused on reliably identifying the ascitic broilers from those non-ascitic ones based on the haematological characteristics and to find a reliable indicator trait for indirect selection against AS susceptibility (Scheele et al., 2003; Wideman et al., 2003; Navarro et al., 2006; Van As et al., 2010). Of course, some of these researchers were successful and introduced some favourable results, but some others failed to find a reliable indicator trait (Closter et al., 2009). The study of Olkowski et al. (1999) indicated that hypoxaemia was not the critical symptom leading to AS. They showed that hypercapnia (high pCO_2) was a feature of ascitic broilers. Scheele et al. (2003, 2005) found that birds with higher mean values of pCO_2 on day 11 coincide with higher AS susceptibility. Recent findings showed that high pCO_2 values together with low pH values (males) or high pH values (females) in the venous blood of young broilers (day 11) could predict AS (Van As et al., 2010). Similar suggestions can be found in literature for haematocrit (Shlosberg et al., 1996) and SaO_2 (Navarro et al., 2006; Druyan et al., 2007b, 2009). In total, the findings of the present study are not in agreement with the reports of Scheele et al.

Table 1 Average body weight (BW) and growth rate (GR) (standard error) of lines S_1 and S_2 at 21, 28 and 35 days of age

Line*	S_1	S_{21}	S_{22}
BW_{21}	839 (13)	910 (12)	611 (7)
BW_{28}	1335 (22)	1449 (22)	1077 (12)
BW_{35}	1918 (32)	2066 (40)	1674 (16)
GR_{21}	37.8 (0.61)	41.2 (0.58)	27.6 (0.37)
GR_{28}	46.0 (0.79)	50.3 (0.81)	37.7 (0.45)
GR_{35}	53.5 (0.93)	57.9 (1.2)	47.5 (0.48)

*S_{21} and S_{22} represent the averages of line S_2 from trials 1 and 2 respectively.

Table 2 Difference of blood parameters between ascitic and non-ascitic chickens of line S_1 in trial 1 measured at 28 and 35 days of age

	Day 28				Day 35			
Blood parameters*	Non-ascitic (44)	Ascitic (4)	Diff %†	Prob‡	Non-ascitic (42)	Ascitic (2)	Diff %	Prob
pH	7.59	7.61	−0.3	NS	7.57	7.54	0.4	NS
pCO_2	41.4	40.3	2.8	NS	50.1	50.6	−1.0	NS
pO_2	53.7	47.7	12.6	NS	43.7	39.0	12.0	NS
BIC	39.8	40.4	−1.4	NS	35.1	35.1	0.1	NS
SaO_2	90.1	90.1	−0.1	NS	86.9	80.3	8.2	NS
HCT	33.4	31.8	4.8	NS	34.2	33.6	1.7	NS

*pCO_2, pO_2, BIC, SaO_2 and HCT are partial pressure of CO_2 (mmHg), partial pressure of O_2 (mmHg), bicarbonate ion concentration (mmol/l), percentage of saturated O_2 and percentage of haematocrit, respectively, all measured from venous blood.
†The difference between means of non-ascitic and ascitic chickens [((non-ascitic-ascitic)/ascitic) *100].
‡Probability of F statistics (NS; non-significant at 0.10 significance level).

Table 3 Difference of blood parameters between ascitic and non-ascitic chickens of line S_2 in trial 1 measured at 28 and 35 days of age

	Day 28				Day 35			
Blood parameters*	Non-ascitic (38)	Ascitic (9)	Diff %†	Prob‡	Non-ascitic (34)	Ascitic (8)	Diff %	Prob
pH	7.61	7.58	0.4	NS	7.60	7.57	0.4	NS
pCO_2	39.4	38.7	1.8	NS	37.0	42.0	−12.0	0.06
pO_2	43.8	41.6	5.3	NS	45.8	39.8	15.0	0.06
BIC	38.8	34.5	12.4	0.01	43.2	35.8	20.6	NS
SaO_2	89.7	84.8	5.7	NS	88.8	81.5	8.9	0.04
HCT	32.8	35.8	−8.3	0.01	34.6	33.7	2.6	NS

*pCO_2, pO_2, BIC, SaO_2 and HCT are partial pressure of CO_2 (mmHg), partial pressure of O_2 (mmHg), bicarbonate ion concentration (mmol/l), percentage of saturated O_2 and percentage of haematocrit, respectively, all measured from venous blood.
†The difference between means of non-ascitic and ascitic chickens [((non-ascitic-ascitic)/ascitic) *100].
‡Probability of F statistics (NS; non-significant at 0.10 significance level).

Table 4 Difference of blood parameters between ascitic and non-ascitic chickens of line S_2 in trial 2 measured at 21 days of age

Blood parameters*	Non-ascitic (156)	Ascitic (70)	Diff %†	Prob‡
pH	7.40	7.37	0.4	NS
pCO_2	43.4	44.5	−2.5	NS
pO_2	44.1	41.2	6.9	NS
BIC	27.2	28.7	−5.3	0.03
SaO_2	75.3	73.4	2.5	NS
HCT	33.2	33.6	−1.2	NS

*pCO_2, pO_2, BIC, SaO_2 and HCT are partial pressure of CO_2 (mmHg), partial pressure of O_2 (mmHg), bicarbonate ion concentration (mmol/l), percentage of saturated O_2 and percentage of haematocrit, respectively, all measured from venous blood.
†The difference between means of non-ascitic and ascitic chickens [((non-ascitic-ascitic)/ascitic)*100].
‡Probability of F statistics (NS; non-significant at 0.10 significance level).

(2003, 2005) and Van As et al. (2010), while these findings are partially in accordance with the reports of Druyan et al. (2007b) and Closter et al. (2009) who did not approve the early predictive characteristics of the blood parameters.

Estimation of Pearson's correlation coefficients between blood parameters and growth-related traits

In trial 1, pairwise correlation coefficients between both 28-day BW and 28-day GR traits and HCT were statistically significant in line S_1 ($p < 0.05$). Furthermore, these growth-related traits were suggestively correlated with BIC parameter ($p < 0.10$). The remaining correlations were not statistically significant in this line (Table 5). In line S_2, pH, pO_2 and HCT parameters were significantly correlated with 28-day BW and 28-day GR traits ($p < 0.05$). Also, pO_2, BIC and SaO_2 parameters were suggestively correlated with 35-day BW and 35-day GR traits ($p < 0.10$). The remaining correlation coefficients were not statistically significant ($p > 0.10$; Table 6).

Table 5 Estimation of pairwise Pearson's correlations between blood parameters measured at 28 and 35 days of age and corresponding body weight (BW) and growth rate (GR) traits in line S_1 of trial 1

	BW_{28}		GR_{28}		BW_{35}		GR_{35}	
Blood parameters*	Corr†	Prob‡	Corr	Prob	Corr	Prob	Corr	Prob
pH	0.14	NS	0.15	NS	0.09	NS	0.09	NS
pCO_2	0.01	NS	0.01	NS	0.22	NS	0.22	NS
pO_2	0.15	NS	0.15	NS	−0.06	NS	−0.06	NS
BIC	0.25	0.09	0.25	0.10	0.20	NS	0.20	NS
SaO_2	0.10	NS	0.09	NS	0.03	NS	0.02	NS
HCT	−0.39	0.01	−0.39	0.01	0.01	NS	0.01	NS

*pCO_2, pO_2, BIC, SaO_2 and HCT are partial pressure of CO_2 (mmHg), partial pressure of O_2 (mmHg), bicarbonate ion concentration (mmol/l), percentage of saturated O_2 and percentage of haematocrit, respectively, all measured from venous blood.
†Pearson's correlation.
‡NS; non-significant at 0.10 significance level.

Table 6 Estimation of pairwise Pearson's correlations between blood parameters measured at 28 and 35 days of age and corresponding body weight (BW) and growth rate (GR) traits in line S_2 of trial 1

	BW_{28}		GR_{28}		BW_{35}		GR_{35}	
Blood parameters*	Corr†	Prob‡	Corr	Prob	Corr	Prob	Corr	Prob
pH	0.29	0.04	0.22	NS	0.04	NS	0.02	NS
pCO_2	−0.10	NS	−0.01	NS	0.19	NS	0.22	NS
pO_2	0.26	0.07	0.33	0.03	−0.26	0.09	−0.28	0.08
BIC	0.19	NS	0.19	NS	−0.30	0.05	−0.31	0.05
SaO_2	0.19	NS	0.21	NS	−0.28	0.07	−0.30	0.06
HCT	−0.32	0.03	−0.34	0.03	0.21	NS	0.23	NS

*pCO_2, pO_2, BIC, SaO_2 and HCT are partial pressure of CO_2 (mmHg), partial pressure of O_2 (mmHg), bicarbonate ion concentration (mmol/l), percentage of saturated O_2 and percentage of haematocrit, respectively, all measured from venous blood.
†Pearson's correlation.
‡NS; non-significant at 0.10 significance level.

In trial 2, except for the correlation between pCO_2 and 21-day GR traits ($r = 0.14$, $p = 0.04$), the remaining correlations were not statistically significant ($p > 0.10$). Of course all of the correlations were in meaningful directions. Such that, the growth-related traits were negatively correlated with pH, pO_2 and SaO_2 parameters, while the other correlations were positive in sign (Table 7).

A good indicator trait for AS should be easy to measure, strongly correlated with AS and should not have unfavourable genetic correlation with economically more important traits such as GR or FCR (Druyan et al., 2007b; Closter et al., 2009). As the structure of the population used for blood parameters measurement did not allow the estimation of genetic correlations between the blood parameters and growth-related traits, we estimated the phenotypic correlations instead. The results revealed that the pairwise correlations between the growth-related traits and blood parameters were not in similar picture in the two trials, indicating probable changes in the blood parameters or growth-related traits as rearing condition shifts from NCC to AIC. It has been demonstrated that the growth-related traits measured under AIC

Table 7 Estimation of pairwise Pearson's correlations between blood parameters measured at 21 days of age and corresponding body weight (BW) and growth rate (GR) traits in line S_2 of trial 2

	BW_{21}		GR_{21}	
Blood parameters*	Corr†	Prob‡	Corr	Prob
pH	−0.04	NS	−0.04	NS
pCO_2	0.11	NS	0.14	0.04
pO_2	−0.06	NS	−0.07	NS
BIC	0.08	NS	0.08	NS
SaO_2	−0.08	NS	−0.08	NS
HCT	0.06	NS	0.07	NS

*pCO_2, pO_2, BIC, SaO_2 and HCT are partial pressure of CO_2 (mmHg), partial pressure of O_2 (mmHg), bicarbonate ion concentration (mmol/l), percentage of saturated O_2 and percentage of haematocrit, respectively, all measured from venous blood.
†Pearson's correlation.
‡NS; non-significant at 0.10 significance level.

were not as same as the corresponding traits which measured under NCC (Pakdel et al., 2005a,b). The similar pattern could be deemed for the blood parameters.

To our knowledge, the number of studies that were conducted to estimate the genetic and phenotypic relationship between blood parameters and AS/AS-related traits as well as between blood parameters and growth-related traits are rare (Navarro et al., 2006; Druyan et al., 2007b; Closter et al., 2009). Although studied for many years, the exact biochemical and physiological precursor factors related to the genetic propensity to develop AS are still unknown. It is often difficult to determine whether a particular change is primary in nature, and therefore determinative, or is a subsequent secondary manifestation in the development of AS (Druyan, 2012). It is apparent that, based on the results of the current study, we could not find the primary trigger of AS nor a reliable indicator trait to be useful as a selection criterion against AS susceptibility.

Conclusion

In the current study, the suitability of blood parameters was tested to differentiate the ascitic chickens from the non-ascitic ones in two trials which were designed to measure the blood parameters under either AIC or NCC. According to the results, the differences of blood parameters between the ascitic and non-ascitic chickens were, in some cases, significant when measured under AIC, while these differences were not strong enough to be statistically significant when there was no AIC implemented earlier. Therefore, the results did not suggest the blood parameters as reliable indicator traits for indirect selection against AS susceptibility.

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