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RESPONSE OF BROILER CHICKENS FROM THREE GENETIC GROUPS TO DIFFERENT STOCKING DENSITIES*

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Abstract

The aim of the present study was to determine the effect of stocking density as a stress factor on production results, blood corticosterone levels and duration of tonic immobility (TI) in broiler chickens from three genetic groups. Chickens were assigned to 6 groups: in groups 1, 2 and 3, Ross 308, Hubbard Flex and Hybro broiler chickens were kept at a stocking density of 13 birds/m^2, and in groups 4, 5 and 6 Ross 308, Hubbard Flex and Hybro broiler chickens were kept at a density of 17 birds/m^2, respectively. During the experiment, individual body weight of the chickens, feed consumption, and mortality were recorded at weekly intervals. Blood corticosterone levels were determined and tonic immobility (TI) measured in 7 birds from each group. The results obtained showed that genetic background had no effect on the final body weight of the chickens or feed conversion (kg feed/kg gain), but origin had an effect on blood corticosterone levels. Stocking density had a significant effect on broiler productivity. Increasing the stocking density from 13 to 17 birds/m^2 had a negative effect on most production parameters of broilers from all three genetic groups. However, Ross 308 broilers showed the smallest decrease in productivity after stocking density per m^2 was increased, possibly suggesting that these birds are less sensitive to this stress factor compared to Hubbard Flex and Hybro chickens. Stocking density had no clear effect on the duration of tonic immobility. There was a tendency towards increased blood corticosterone levels in broilers subjected to higher stocking density per m^2.

Key words: broiler chickens, productivity, tonic immobility (TI), corticosterone, stocking density

Modern broiler chickens, which show rapid weight gains and good feed conversion, have become more demanding in terms of management, feeding and handling conditions. The optimization and stabilization of rearing conditions made birds more delicate and the absence of necessary environmental stimuli (weather conditions, foraging activity) suppressed the body's adaptive mechanisms. For this reason, modern broilers have lower resistance to environmental factors while being more susceptible to stress states, which has an adverse impact on their productivity and health (Al-Murrani et al., 2006; Sosnówka-Czajka et al., 2006).

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One of the stress factors of birds is high stocking density, which is often found in commercial large-scale systems. Campo et al. (2005) report that high stocking density per m^2 makes birds more fearful and susceptible to stress. Increased stocking density is accompanied by reduced body weight, poorer feed conversion and increased mortality (Skomorucha et al., 2004; Sosnówka-Czajka et al., 2005; Estevez, 2007; Onbaşılar et al., 2008).

Stress factors that affect birds also increase blood corticosterone levels (Puvadolpirod and Thaxton, 2000; Olanrewaju et al., 2006) and increase the duration of tonic immobility, which reflects the birds' welfare and stress levels (Andrews et al., 1997; Campo et al., 2005; Onbaşılar et al., 2008)

Sensitivity to environmental factors and susceptibility to stress may be genetically determined (Campo and Carnicer, 1994; Campo et al., 2008; Sosnówka-Czajka et al., 2005).

The aim of the present study was to determine the effect of stocking density as a stress factor on production results, blood corticosterone levels and duration of tonic immobility (TI) in broiler chickens from three genetic groups.

Material and methods

The experiment was carried out at a poultry farm in Aleksandrowice using 810 unsexed broiler chickens of three commercial lines (Ross 308, Hubbard Flex, Hybro) originating from Poultry Hatcheries in Gardawice and Łężkowice.

After weighing and tagging, chicks were placed in cage batteries, in which each cage with an area of 1 m^2 was a replicate:

	Genetic group					
	Ross 308	Hubbard Flex	Hybro	Ross 308	Hubbard Flex	Hybro
Group	I	II	III	IV	V	VI
No. of replicates	9	9	9	9	9	9
Stocking density (bird/m^2)	13	13	13	17	17	17

Chickens were fed *ad libitum* diets based on concentrates: starter diet to 21 days of age (3000 kcal metabolizable energy, crude protein 21.7%, lysine 1.15%, methionine + cystine 0.82% per kg feed), grower diet from 22 to 35 days (ME 3100 kcal, CP 19.8%, lysine 0.96%, methionine + cystine 0.78% per kg feed), and finisher diet to 42 days (ME 3100 kcal, CP 18.5%, lysine 0.82%, methionine + cystine 0.71% per kg feed). Throughout the experiment, birds had free access to water drinkers.

During the study, individual body weight of the chickens, feed consumption in groups (replicates) and mortality were recorded every week. At 1, 21 and 42 days of rearing, blood was collected from 7 birds of each group to determine corticosterone levels, which were tested immunoenzymatically using a Diagnostic System Laboratories kit (USA) and a SIRIO S immunoenzymatic analyser.

At 21, 28, 35 and 42 days of rearing, tonic immobility (TI) was measured according to the method of Akşit et al. (2006) in 7 birds from each group.

The results were analysed statistically by two-way analysis of variance and significant differences were estimated using Duncan's test.

Results

Tables 1–3 give mean results for the experimental groups.

Broilers' rearing results are shown in Table 1. On day 1 of rearing, the lowest body weight was characteristic of Hybro broilers compared to the other groups ($P \leq 0.01$). At 21 days of rearing, the highest body weight at a stocking density of 13 birds/m^2 was found in Hubbard Flex broilers compared to the other groups ($P \leq 0.01$), and at a density of 17 birds/m^2 Ross 308 and Hubbard Flex broilers weighed 911 g compared to 860 g in Hybro chickens, with a highly significant difference. Comparison of body weight in chickens from the same genetic group that were kept at different stocking densities per m^2 showed a highly significant difference between Hubbard Flex chickens stocked at 13 birds/m^2 (group II) and 17 birds/m^2 (group V) and between Hybro chickens stocked at 13 birds/m^2 (group III) and 17 birds/m^2 (group VI). On the final day of the experiment, broiler chickens reared at a stocking density of 13 birds/m^2 obtained final body weights of 2527, 2540 and 2532 g, respectively, and when stocking density per m^2 was higher, Hubbard Flex chickens were the lightest and Ross 308 chickens the heaviest ($P \leq 0.01$). Comparison of the birds from the same genetic group that were kept at different stocking densities showed differences in body weight between Hubbard Flex chickens ($P \leq 0.01$) and between Hybro chickens ($P \leq 0.05$).

The poorest feed conversion per kg weight gain (Table 1) in groups at a stocking density of 13 birds/m^2 was found in Hybro broilers and the highest in Ross 308 broilers ($P \leq 0.01$). At 17 birds/m^2 no statistically significant differences were found between the groups. A highly significant difference was found when comparing Hybro broilers at densities of 13 and 17 birds/m^2. In the grower period, a difference emerged between Hubbard Flex and Hybro chickens raised at a density of 13 birds/m^2 ($P \leq 0.01$). At a density of 17 birds/m^2, no significant differences were found between the groups. Comparison of broilers in terms of stocking density per m^2 showed a highly significant difference in Hubbard Flex chickens between densities of 13 birds/m^2 (group II) and 17 birds/m^2 (group V), and a significant difference in Ross 308 chickens between groups I (stocking density of 13 birds/m^2) and IV (stocking density of 17 birds/m^2). For the whole rearing period, the best feed conversion per kg weight gain at a density of 13 birds/m^2 was found in Hubbard Flex broilers and the poorest in Hybro broilers ($P \leq 0.01$). No statistically significant differences emerged between the groups for a density of 17 birds/m^2. Highly significant differences in feed conversion were found between Hubbard Flex and Hybro chickens stocked at a density of 13 birds/m^2 and Hubbard Flex and Hybro chickens stocked at a density of 17 birds/m^2. Ross 308 broilers raised at a density of 13 birds/m^2 were characterized by better feed conversion per kg weight gain compared to chickens of the same genetic group that were reared at a higher stocking density per m^2 ($P \leq 0.05$).

Table 1. Productive results of broiler chickens

Item	Days of rearing	Group						Genetic group (A)	Stocking density (B)	A×B
		13 birds/m^2			17 birds/m^2					
		I Ross 308	II Hubbard Flex	III Hybro	IV Ross 308	V Hubbard Flex	VI Hybro			
Body weight (g)	1	45.36±0.43 B	44.73±0.41 BX	39.13±0.34 A	45.47±0.36 B	46.21±0.39 B Y	39.58±0.32 A	<0.01		
	21	899.61±10.72 A	975.36±15.04 B X	926.97±9.24 AX	911.19±7.58 B	910.71±13.44 B Y	859.68±8.85 AY	<0.01	<0.01	<0.01
	42	2527.21±35.21	2540.32±30.05 X	2532.46±34.77 x	2468.93±31.67 B	2324.81±29.39 Aa Y	2436.45±34.94 by	NS	<0.01	<0.05
Feed conversion (g feed/kg gain)	1–21	1494.00±42.38 Bb	1412.00±20.35 a	1376.00±12.88 A X	1454.00±9.27	1442.00±9.69	1498.00±21.31 Y	NS	<0.05	<0.01
	22–42	1776.00±36.55 x	1710.00±15.81 A X	1910.00±59.16 B	1956.00±36.28 y	2042.00±27.46 Y	2000.00±80.12	NS	<0.01	NS
	1–42	1674.00±16.00 Bx	1600.00±10.95 Aa X	1702.00±34.55 B X	1768.00±18.28 y	1808.00±17.15 Y	1812.00±40.42 Y	NS	<0.01	NS
Mortality (%)	1–21	0	0	1.5	0	0	2.3			
	22–42	3	1.5	4.6	1.2	2.4	6.9			
	1–42	3	1.5	6.1	1.2	2.4	9.2			

A,b,A,B – significant differences between genetic groups of broilers within stocking density.
x,y,X,Y – significant differences between different stocking densities.
a,b,x,y – values in rows with different letters differ significantly (P≤0.05).
A,B,X,Y – values in rows with different letters differ highly significantly (P≤0.01).

Table 2. Tonic immobility of broiler chickens (s)

Days of rearing	Group						Genetic group (A)	Stocking density (B)	A × B
	13 birds/m²			17 birds/m²					
	I Ross 308	II Hubbard Flex	III Hybro	IV Ross 308	V Hubbard Flex	VI Hybro			
21	163.57±57.26	113.14±47.29 X	168.14±58.67	283.00±67.90	430.00±74.32 BY	167.71±45.68 A	NS	≤0.01	≤0.05
28	216.29±67.75	313.14±100.78	148.43±83.33	386.71±88.05	174.43±76.52	234.29±82.01	NS	NS	NS
35	318.71±95.31	198.29±67.62	287.29±87.07	303.43±85.48	189.71±74.12	341.86±94.07	NS	NS	NS
42	307.71±66.41	307.57±92.15 x	301.43±79.85	198.00±69.13	94.29±28.49 y	185.57±54.05	NS	≤0.05	NS

A,b,A,B – significant differences between genetic groups of broilers within stocking density.
x,y,X,Y – significant differences between different stocking densities.
a,b,x,y – values in rows with different letters differ significantly ($P \leq 0.05$).
A,B,X,Y – values in rows with different letters differ highly significantly ($P \leq 0.01$.)

Table 3. Blood corticosterone levels in broiler chickens (ng/ml)

Days of rearing	Group						Genetic group (A)	Stocking density (B)	A × B
	13 birds/m²			17 birds/ m²					
	I Ross 308	II Hubbard flex	III Hybro	IV Ross 308	V Hubbard flex	VI Hybro			
1	315.48±23.04 B	94.96±36.19 A	59.70±57.94 A	321.42±49.56 B	93.86±62.81 A	46.12±13.63 A	≤0.01	-	-
21	264.27±71.31 x	181.47±65.26	126.06±63.33	498.34±96.60 By	321.93±69.44	152.27±60.71 A	≤0.01	≤0.05	NS
42	232.17±60.93 b	96.16±31.44 AaX	365.11±20.85 Bc	309.00±42.50	297.94±29.20 Y	345.67±23.47	≤0.01	≤0.01	≤0.05

For significant differences, see tables above.

The highest mortality was characteristic of Hybro chickens stocked at both densities (Table 1). The lowest mortality was found in group II (Hubbard Flex) for a density of 13 birds/m^2 and in group IV (Ross 308) for a density of 17 birds/m^2. Among Hubbard Flex and Hybro broilers, mortality was lower when birds were stocked at 13 birds/m^2 compared to 17 birds/m^2.

The duration of tonic immobility is presented in Table 2. At 21 days of the study, no significant differences were found between the groups for a stocking density of 13 birds/m^2. At a density of 17 birds/m^2, a highly significant difference emerged between Hubbard Flex and Hybro chickens. Comparison of broiler chickens from the same genetic group that were kept at different densities per m^2 showed a difference between Hubbard Flex broilers ($P \leq 0.01$). At 28 and 35 days of the experiment, no statistically significant differences were found between the groups in the duration of tonic immobility. At 42 days of the experiment, a difference emerged between Hubbard Flex chickens reared at different densities per m^2 ($P \leq 0.05$).

Blood corticosterone levels were dependent on both the commercial line and stocking density (Table 3). On day 1 of rearing, the highest corticosterone level was characteristic of Ross 308 chickens compared to the other groups ($P \leq 0.01$). At 21 days of the experiment, corticosterone level was still the highest in Ross 308 broilers, with a highly significant difference found for a density of 17 birds/m^2 between Ross 308 and Hybro chickens. Birds from the groups stocked at 17 birds/m^2 tended to have higher blood corticosterone levels, but a significant difference emerged only between Ross 308 chickens. At 42 days of the experiment, the highest corticosterone level at a density of 13 birds/m^2 was characteristic of Hybro broilers and the lowest of Hubbard Flex broilers ($P \leq 0.01$). At a density of 17 birds/m^2 no statistically significant differences were found between the groups in blood corticosterone levels. Comparison of broilers stocked at 13 and 17 birds/m^2 showed a highly significant difference in Hubbard Flex chickens.

Discussion

Good results in poultry production are largely dependent on the choice of proper genetic material and appropriate stocking density per m^2. The literature suggests that broiler chickens of different origin have different production results (Sosnówka-Czajka et al., 2005; Skomorucha et al., 2007). According to Smith et al. (1998), broiler chickens from different genetic groups differ in rate of growth, final body weight, feed consumption, and feed conversion (kg feed/kg gain). In our study, genetic group had a statistically significant effect on the body weight of chickens at 1 and 21 days of the experiment. At 42 days of rearing, chickens stocked at 13 birds/m^2 were characterized by similar body weight, and for those stocked at 17 birds/m^2, the lowest body weight was found for Hubbard Flex broilers and the highest for Ross 308 broilers ($P \leq 0.01$). No differences in weight gains and body weight of chickens from three different genetic groups were reported by Reiter and Kutritz (2001). In our study, genetic group had no effect on feed conversion. However, for lower stocking density per m^2, highly significant differences were found in feed conversion between Hybro and Ross

308 broilers during the starter period and between Hybro and Hubbard Flex broilers during the grower period and for the whole experimental period. Skomorucha et al. (2007) showed the effect of genetic group on feed conversion in broiler chickens. Likewise, Sosnówka-Czajka et al. (2005) found differences in feed conversion between Hybro G and Hybro PN chickens. Reiter and Kutritz (2001) also found differences in feed conversion between Ross and Lohmann Meat and Hubbard broilers, but observed no differences between Lohmann Meat and Hubbard broiler chickens.

Campo et al. (2008) showed differences in the duration of tonic immobility (TI) between chickens of Red Villafranquina and White-Faced Spanish breeds. In our study, genetic group had no effect on the duration of TI, but origin had an effect on blood corticosterone levels.

Increasing the stocking density per m^2 in intensive poultry production systems is aimed to reduce production costs, but often causes stress to the birds, which may affect their productivity. Onbaşılar et al. (2008) showed that stocking rate has an effect on the final body weight of broiler chickens ($P \leq 0.01$), feed consumption ($P \leq 0.05$) and feed conversion (kg feed/kg gain) ($P \leq 0.05$). Likewise, Feddes et al. (2002) reported lower body weights in broilers when stocking density increased from 14 to 18 birds/m^2. Also Dozier et al. (2006) stated that densities exceeding 30 kg body weight per m^2 of floor space had a negative effect on weight gains and dressing percentage of pullets, but had no effect on the physiological indicators of stress. In our study, we found decreases in the body weight of Hubbard Flex (by 6.67%) and Hybro broilers (by 7.23%) at 21 days of the experiment and in the final body weight (by 8.46% at $P \leq 0.01$ and by 3.79% at $P \leq 0.05$) when stocking density increased from 13 to 17 birds/m^2. This was paralleled by increases in feed conversion (kg feed/kg gain) and mortality of broiler chickens from two genetic groups. Skomorucha et al. (2004) found a decrease in feed consumption by broilers as stocking density increased from 12 to 18 birds/m^2. In the present study, the productivity of Ross 308 broilers did not deteriorate when stocking density per m^2 increased. Likewise, Thomas et al. (2004) reported similar body weights, feed conversion and mortality for broiler chickens stocked at 10, 15 and 20 birds/m^2. Also Pettit-Riley and Estevez (2001) found no effect of increasing stocking rate from 10 to 20 birds/m^2 on body weight and feed conversion.

Estevez (2007) reports that stocking density is associated not only with production results but also with physiological stress in birds. Onbaşılar et al. (2008) showed longer duration of tonic immobility in broiler chickens stocked at 17.5 birds/m^2 compared to 11.9 birds/m^2. Likewise, Andrews et al. (1997) found that higher density per m^2 has an effect on longer duration of TI in broiler chickens. In our study, longer duration of TI was only found at 21 days of the experiment for the higher stocking density of Hubbard Flex chickens. At 42 days of rearing, broilers from all three genetic groups tended to show short duration of TI at 17 birds/m^2 compared to 13 birds/m^2, although the difference occurred for Hubbard Flex chickens only. Also Skomorucha and Muchacka (2007) showed no effect of stocking density on the duration of TI. Meanwhile, Thaxton et al. (2006) found that stocking density increased from 20 to 50 kg body weight per m^2 has no effect on the blood concentrations of corticosterone, glucose and cholesterol in broiler chickens, which is evidence that these birds show no stress reaction. In our study, broiler chickens showed a tendency towards higher blood corticosterone concentrations when stocked at the higher density per m^2. On the final day of the experiment, chicken body weight per unit area was about 33 kg/m^2 in the groups stocked at the lower density and ranged from 39.5 to 42 kg/m^2 in the groups stocked at the higher density. Statistically significant differences were found at 21 days of rearing between Ross 308 broilers (P≤0.05) and at 42 days of rearing between Hubbard Flex broilers (P≤0.01).

In summary, the genetic background of the broiler chickens had no effect on the final body weight or feed conversion (kg feed/kg gain), but origin had an effect on blood corticosterone levels.

Stocking density had a significant effect on broiler productivity. Increasing the stocking density from 13 to 17 birds/m^2 had a negative effect on most production parameters of broilers from all three genetic groups. However, Ross 308 broilers showed the smallest decrease in productivity after stocking density per m^2 was increased, possibly suggesting that these birds are less sensitive to this stress factor compared to Hubbard Flex and Hybro chickens.

Stocking density had no clear effect on the duration of tonic immobility, but there was a tendency towards increased blood corticosterone levels in broilers subjected to higher stocking density per m^2.

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Reakcja kurcząt brojlerów z trzech grup genetycznych na zróżnicowaną gęstość obsady podłogi

STRESZCZENIE

Celem przeprowadzonych badań było określenie wpływu gęstości obsady, jako czynnika stresowego, na wyniki produkcyjne, poziom kortykosteronu we krwi oraz czas trwania tonicznego bezruchu (TI) kurcząt brojlerów trzech grup genetycznych.

Kurczęta brojlery przydzielono do 6 grup: w grupie 1, 2 i 3 utrzymywano odpowiednio: kurczęta brojlery Ross 308, Hubbard flex i Hybro o obsadzie 13 szt./m^2, natomiast w grupie 4, 5 i 6 odpowiednio: kurczęta brojlery Ross 308, Hubbard flex i Hybro o obsadzie 17 szt./m^2. Podczas doświadczenia co tydzień kontrolowano indywidualnie masę ciała kurcząt, zużycie paszy oraz liczbę padłych kurcząt. Oznaczono również poziom kortykosteronu we krwi i przeprowadzono pomiar tonicznego bezruchu (TI) u 7 ptaków z każdej grupy.

Na podstawie uzyskanych wyników stwierdzono, że uwarunkowania genetyczne nie miały wpływu na końcową masę ciała kurcząt brojlerów oraz na wykorzystanie paszy na przyrost 1 kg masy ciała, odnotowano natomiast wpływ pochodzenia na poziom kortykosteronu we krwi ptaków. Wielkość obsady miała statystyczny wpływ na produkcyjność kurcząt brojlerów. Zwiększenie obsady z 13 do 17 szt./m^2 pogorszyło większość badanych parametrów produkcyjnych kurcząt brojlerów wszystkich trzech grup genetycznych. Kurczęta brojlery Ross 308 odznaczały się jednak najmniejszym spadkiem produkcyjności

przy zwiększeniu obsady na 1 m^2 powierzchni, co może świadczyć, że są one mniej wrażliwe na ten czynnik stresowy w porównaniu z kurczętami Hubbard flex i Hybro. Nie wykazano jednoznacznie wpływu wielkości obsady na czas trwania tonicznego bezruchu, stwierdzono natomiast tendencje do wzrostu poziomu kortykosteronu we krwi ptaków przy wyższej obsadzie na 1 m^2 powierzchni.