

Full Length Research Paper

Selected hematological and immunological parameters in pigs transferred from the rearing unit to the finishing house

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The aim of the study was to determine how selected hematological and immunological parameters are affected when growing pigs are transferred from the rearing unit to the finishing house. Blood was collected from 64 healthy growing pigs one week before and one week after they were transferred to the finishing house. The following tests were performed: complete blood count with machine differential, immunoglobulin levels, C-reactive protein (CRP) level and cortisol level. Pigs were divided into two groups. Group A contained those pigs with a normal white blood cell (WBC) count, and Group B those with an elevated WBC count. Throughout the experiment, body weight and indoor microclimate conditions were also monitored. After transfer, the neutrophil count increased, while the lymphocyte count decreased. Hematocrit (HCT), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) also decreased. CRP level and cortisol level increased. Red blood cell (RBC) count was higher and mean corpuscular volume (MCV) was lower in Group B. CPR level and immunoglobulin G (IgG) level were also higher. On the other hand, average daily gain (ADG) was higher in Group A. Although the presence of subclinical infections cannot be ruled out, the changes observed were probably caused by other stressogenic factors such as transport, adaptation to a new maintenance system, and worse sanitary conditions. Those with elevated WBC counts before transport were most susceptible to adaptive stress.

Key words: Pig, transfer, immunity, C-reactive protein (CRP), cortisol, immunoglobulin, stress.

INTRODUCTION

Recent changes in animal turnover on pig farms (a new system of feeding and breeding conditions, management and especially genetic progress), have produced conditions that promote the growth of pathogenic microorganisms where pigs are kept, as well as select for more virulent mutant microbial strains (Kołacz et al., 2009). Together with the stress to which the animals are subjected to during rearing, these conditions can impair or

suppress their immune systems (Heinonen et al., 2010), and these systems all influence health and well-being in response to environmental and management conditions.

Transferred pigs following the rearing phase are a common management practice in the swine industry, and it imposes an additional stress on the young pig. The advent of wean-to-finish facilities has potentially alleviated commingling after the nursery phase; however,

the wasted space of placing weanling pigs in pens with space allowances for market hogs has led to the practice of double-stocking pens at weaning and later moving half the pigs to another pen, which introduces a commingling stress at an older age (Wolter et al., 2002). In addition to the peri-weaning period, transfer to the finishing house is of key importance. At this time, the pigs are subjected to intense stress induced by adaptation to the new feeding strategy and management conditions, as well as by the establishment and nature of the dominance hierarchy in the new herd. Sub-optimal management practices add to the stress the animals experience when adapting to a new location. This stress can also reduce the integrity of the immune response, which increases the incidence of microbial diseases in the herd (Ekkel et al., 1996).

Poor air quality in pig houses can also increase the number of infections, the prevalence of disease, and the development of clinical manifestations (Borell et al., 2007). Monitoring the health status of the pigs can be an important help in an early warning system from which you can take action and start up control measures. Among the parameters that have proven useful in such surveillance programs are levels of immunoglobulins and the kinetics of acute phase proteins (APP). Immunoglobulin levels can be affected by pollutants in the air, feed contaminated with mycotoxins, as well as by stress associated with relocation, isolation and physical confinement (Leek et al., 2004).

The kinetics of acute phase proteins can be used to diagnose and monitor inflammatory processes in an entire herd or in individual pigs at slaughter time (Parra et al., 2006; Berg et al., 2008). They are objective, non-specific markers that correlate well with the effectiveness of therapy. They can also be used as a prognostic tool. In disease-free herds, the kinetics of acute phase proteins can provide useful information about environmental stressogenic factors (Salamano et al., 2010). C-reactive protein (CRP), amyloid A (SAA) and haptoglobin (Hp) are important indicators of inflammation in pigs (Parra et al., 2006).

Excessive stimulation of the immune system is inversely correlated with growth rate (Chmielowiec-Korzeniowska, 2011; Kołacz and Bodak, 2000). The time that it takes for pigs to reach market weight can be prolonged by exposure to environmental stress. More specifically, high ambient temperature, social mixing, and restricted floor space reduce feed intake and body weight (BW) gain (Sutherland et al., 2006). Additionally, stimulation of the immune system excitation induces a hypercatabolic state,

while at the same time reducing synthesis of tissue proteins and increasing synthesis of acute phase proteins. This results in a negative energy and nitrogen balance (Kołacz and Bodak, 2000).

The impact of housing conditions and management procedures on physiological traits depends on the duration and intensity of the factor that acts on the animal (Borell, 2001). There are many publications that reported both short stressful events that is, direct handling, isolation, transportation (Berg et al., 2008; Borell, 2001; Heinonen et al., 2010; Salamano et al., 2010) and chronic stressful events, that is, long-acting heat stress or restraint (Leek et al., 2004; McGlone, 1994; Sutherland et al., 2006). There is limited scientific information on the combined effects of the movement of animals and management conditions on performance and immune function of swine.

The aim of the study was to determine how selected hematological and immunological parameters are affected when growing pigs are transferred from the rearing unit to the finishing house.

MATERIALS AND METHODS

Facility and animals

The study was conducted on a private farm specialized in pig production with an average of 105 LU. Production was carried out in a closed system with a full-empty facility cycle. Pigs were housed in three separate units: a building with two boar stalls; a farrowing and rearing unit; and a finishing house.

The rearing unit contained eight pens holding an average of twenty growing pigs per pen, with 0.3 m² of floor surface per pig. The floor was concrete-slatted. In the finishing house, the pigs were managed using the straw-bedded deep litter system. Litter was removed once at the end of each production cycle. Throughout the experimental period, there were an average of 100 pigs per chamber, with 0.8 m² of floor surface per pig. The ventilation system in rearing unit and the finishing house was based on natural-mechanical ventilation. In both units, feeding was fully automated and mechanized. Pigs were fed *ad libitum* using a balanced full-ration feed tailored to age and body weight. In each pen, the pigs had unhampered access to automatic waterers and feeders.

The pigs received constant veterinary care and underwent routine prevention procedures. Ferric iron was administered when the pigs were 3 and 13 days old. Piglets were castrated and had their needle teeth clipped when they were 5 days old. They were dewormed and vaccinated against *Mycoplasma hyopneumoniae* when they were two weeks old. None of the pigs used in the study were genetically predisposed to stress. *RYR1CC* genotype was confirmed by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP).

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Abbreviations: CRP, C-Reactive protein; SAA, amyloid A; Hp, haptoglobin; WBC, white blood cells; PCR-RFLP, polymerase chain reaction-restriction length fragment polymorphism; EDTA, ethylenediaminetetra acetate; HGB, hemoglobin; HCT, hematocrit; RBC, red blood cell count; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; PLT, blood platelet count; MPV, mean platelet volume, Lym, lymphocyte count; Neu, neutrophil count; ELISA, enzyme linked immuno sorbent assay; ADG, average daily gain; TSA, trypticase soy agar; MEA, malt extract agar; CFU, colony-forming units; APP, acute phase proteins.

Piglets were weaned when they were 28 days old. At this time, they had an average body weight of 7.0 kg. The pigs were kept in the rearing unit until they were 10 weeks old, when they were transferred to the finishing house.

Experimental procedure

The study was conducted on 64 healthy growing pigs that did not exhibit any clinical symptoms. There were equal numbers of males and females. The pigs were F1 hybrids of Polish Large White and Polish Landrace (PLW × PL). The effect of the transfer from the rearing unit to the finishing house on health status was assessed using hematological and immunological indices (National Ethics Commission No. 39/2006). Body weight was monitored throughout the study. Before the pigs were transferred, they underwent hematological testing. Based on the results, the pigs were divided into two groups. Group A contained those pigs with white blood cells (WBC) counts below $20.0 \times 10^9/l$, that is, within the reference values reported by Winnicka (2009). Group B contained those pigs with higher WBC counts.

Sample collection

Blood for hematological and immunological testing was collected when the pigs were 9 and 11 weeks old, that is, one week before and one week after the transfer to the finishing house. Blood was collected from the jugular vein into 4.9 ml S-Monovette clot tubes (Sarstedt AG and Co., Numbrecht, Germany) and into tubes containing K₂-ethylenediaminetetra acetate (EDTA) (Profilab Sc., Warsaw, Poland). Blood was collected in a isolated room within 22 s of the animal entering the room and the end of the procedure. A total of 120 blood samples were taken from 64 pigs.

Hematological determinations

Samples collected in EDTA were analyzed using a hematology analyzer (Melet Schloesing Laboratories, Osny, France). The following parameters were recorded: hemoglobin (HGB), hematocrit (HCT), red blood cell count (RBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), blood platelet count (PLT), mean platelet volume (MPV), white blood cell count (WBC), lymphocyte count (Lym) and neutrophil count (Neu).

Immunological determinations

Clot tubes were centrifuged at $3000 \times g$ for 15 min. Serum immunoglobulin levels were measured using a single radial immunodiffusion assay. Data recorded included the total immunoglobulin complex level, as well as the levels of IgA, IgM, IgG. Serum cortisol and C-reactive protein (CRP) were measured using commercial enzyme linked immuno sorbent assay (ELISA) kits (IBL International GmbH, Hamburg, Germany).

Body weight

Body weight was monitored throughout the study. Animals were first weighed when they were 3 weeks old. They were weighed again upon transfer to the finishing unit when they were 10 weeks old, and third time at the end of the first stage of fattening when they

were 17 weeks old. The weights recorded were used to calculate average daily gain (ADG) in the rearing unit (Weeks 3 to 10) and in the finishing house (Weeks 10 to 17).

Animal management conditions

The microclimate in each facility was assessed using standard zootechnical methods. Temperature, humidity and air motion were measured twice on the day of blood collection, 4 h apart at three sites (P1 to P3) 30 cm off the ground. This minimized sudden increases in the parameters measured, especially temperature, providing a more representative picture of indoor conditions. Temperature and humidity were measured with a RT811 thermohygrometer (Technik, Warsaw, Poland). Air motion was measured using an A-12001 anemometer (OBRAiUP, Łódź, Poland).

At the same time, ammonia, carbon dioxide, volatile organic compounds, total dust, total bacterial count, total actinomycetes count and total fungal count were measured in the rearing unit and the finishing house. Ammonia, carbon dioxide, volatile organic compounds were measured using a QRAE Plus model PGM-2000 multi-gas monitor (RAE Systems, San Jose, USA). Dust level was measured using a gravimetric method. Air samples were collected with a 224-PCEX8 aspirator (SKC, Dorset, England). Microbial counts were determined using an aspiration method in compliance with Polish Norms PN-EN 13098:2007, followed by the dilution plate technique. Bacterial counts were carried out using trypticase soy agar (TSA) with 5% sheep blood. Inoculated plates were incubated for seven days (one day at 37°C, three days at 22°C, and three days at 4°C). Fungal counts were carried out using malt extract agar (MEA). Plates were incubated at 30°C for four days and 25°C for three days. Actinomycete counts were carried out using "agar for actinomycetes". Plates were incubated for seven days at 25°C. After incubation, the colonies on each medium were counted and expressed in terms of colony-forming units (CFU).

Statistical analysis

The effect of animal housing conditions, sex and group determined on the basis of WBC count on the level of the determined indices and the interactions between them was determined on the basis of ANOVA.

RESULTS

Throughout the study, the temperature in the rearing unit and finishing house fell within the specified limits. Average air temperature in the rearing unit was 5°C higher than in the finishing house ($P < 0.01$; Table 1). The dust level was higher in the finishing house ($P < 0.05$). Of the chemical pollutants, only ammonia concentration was higher in the finishing house than in the rearing unit ($P < 0.05$). There were also significant differences in microbial counts, with total bacterial count as much as two orders of magnitude higher in the finishing house than in the rearing unit ($P < 0.05$).

Hematological parameters are summarized in Table 2. HCT, MCH, MCHC and HGB decreased after transfer. Lymphocyte count decreased, and neutrophil count increased ($p < 0.05$). Total WBC also increased slightly, although the difference was not statistically significant.

Table 1. Environmental conditions in rearing unit (RU) and finishing house (FH)[†].

Parameter	Place				P-value
	RU		FH		
	M	SD	M	SD	
Microclimate					
Temperature (°C)	23.7	1.6	18.7	0.4	<0.001
Relative humidity (%)	76.2	6.2	83.0	3.6	0.078
Air flow (m/s)	0.1	0.1	0.1	0.0	0.455
Microbiological pollution					
Total count of bacteria (CFU × 10 ⁵ /m ³)	4.52	1.38	342.7	472.9	0.039
Total count of fungal (CFU × 10 ⁵ /m ³)	0.05	0.01	1518.1	5024.2	0.340
Total count of actinomycete (CFU × 10 ⁵ /m ³)	0.05	0.03	19.9	29.7	0.051
Physico-chemical air pollution					
Total dust (mg/m ³)	1.7	2.4	6.1	1.1	0.025
CO ₂ (%)	0.2	0.0	0.1	0.1	0.083
Ammonia (ppm)	4.5	2.0	18.8	8.1	0.036
VOCs (ppm)	0.2	0.1	0.5	0.2	0.135

RU, Contained 8 pens holding an average of 20 pigs per pen, with 0.3 m² floor/pig; FH, the pigs were managed using the straw-bedded deep litter system; there were an average of 100 pigs per chamber, with 0.8 m² floor/pig. The floor was concrete-slatted.

Table 2. The level of hematological parameters of pigs studied[†].

Parameter	Place				P-value [§]		
	RU (N=64)		FH (N=64)		Site	Sex	Site × Sex
	M	SD	M	SD			
WBC (10 ⁹ /l)	17.87	4.58	21.14	6.36	0.622	0.499	0.494
Lim (10 ⁹ /l)	10.41	0.43	10.09	0.68	<0.001	0.573	0.889
Neu (10 ⁹ /l)	6.02	0.44	8.84	0.70	<0.001	0.252	0.553
RBC (10 ¹² /l)	7.15	0.52	7.19	0.51	0.439	0.922	0.652
MCV (fl)			53.24	2.48	0.148	0.268	0.646
MCH (fmol)	1.03	0.09	0.96	0.05	<0.001	0.460	0.913
MCHC (mmol/l)	18.93	1.44	18.07	0.87	<0.001	0.828	0.699
HCT (l/l)	0.39	0.03	0.38	0.03	0.203	0.566	0.556
HGB (mmol/l)	7.33	0.78	6.90	0.46	<0.001	0.807	0.869
PLT (10 ⁹ /l)	380.6	127.3	390.57	109.72	0.820	0.359	0.479

The pigs were kept in the RU (rearing unit) until 10 week old, when they were transferred to finishing house (FH). Blood collected one week before and one week after the transfer. [†], Pigs F1 hybrids of polish large white and polish landrace (PLW × PL); [§] impact of site, sex and interaction place × sex on the level of indicators calculated on the basis of Anova test.

Immunological parameters are presented in Table 3. After transfer, CRP levels increased ($p < 0.05$). Cortisol levels also increased slightly, although the difference was not statistically significant. Average daily weight gain in the rearing unit was 470 g for gilts and 459 g for barrows. In the finishing unit, average daily weight gain was 938 g for gilts and 967 g for barrows (Table 4). RBC count was higher in Group B than in Group A. On the other hand, MCV was lower (Table 5). There were also significant differences in CRP and IgG between the groups (Table 6).

Statistical analysis revealed that the WBC differential was affected not only by the transfer, but also by an interaction between site and group (Table 5). Pigs that had elevated WBC counts in the rearing unit had higher neutrophil counts. For Group A, neutrophil counts after transfer increased by $0.77 \times 10^9/l$ in gilts and $0.56 \times 10^9/l$ in barrows. For Group B, they increased by $3.52 \times 10^9/l$ in gilts and $3.48 \times 10^9/l$ in barrows. Neutrophil counts were also affected by an interaction between sex and group. Counts were significantly higher in gilts than in barrows,

Table 3. The concentration of CRP, cortisol, and immunoglobulins in blood serum[†].

Parameter	Place				P-value [§]		
	RU (N=64)		FH (N=64)		Site	Sex	Site × Sex
	M	SD	M	SD			
CRP (µg/ml)	68.67	39.67	104.80	74.53	0.028	0.582	0.785
Crtisol (ng/ml)	51.65	16.99	62.39	23.34	0.378	0.632	0.993
IgA (g/l)	0.40	0.00	0.51	0.32	0.165	0.219	0.219
IgG (g/l)	3.43	4.06	4.87	2.84	0.420	0.580	0.590
IgM (g/l)	2.25	0.82	0.78	0.41	<0.001	0.548	0.285

[†]pigs and houses described in Table 1 and 2.

Table 4. Average daily gain (ADG) in rearing unit (between 3 and 10 weeks of age) and finishing house (between 10 and 17 weeks of age)[†].

Parameter	Total (N=64)		Gilts (N=32)		Barrows (N=32)		P-value
	M	SD	M	SD	M	SD	
Body weight in the 3 week of age (kg)	5.7	2.0	5.6	1.8	5.8	2.2	0.412
Body weight in the 10 week of age (kg)	33.7	7.4	33.9	6.9	33.6	8.0	0.893
ADG in rearing unit, g/day	464.6	77.8	470.3	80.1	459.8	77.1	0.361
Body weight in the 17 week of age (kg)	70.6	8.3	70.5	7.9	70.6	8.7	0.959
ADG in finishing house (g/day)	954.1	148.7	938.5	123.3	967.2	168.5	0.583

[†]pigs and houses described in Table 1 and 2.

regardless of group ($P < 0.05$).

In the rearing unit, CRP was higher in Group B than in Group A. The difference was 77.8 µg/ml for gilts, and 10.6 µg/ml for barrows. After transfer, CRP increased in all pigs, more so in animals in Group B than in Group A (interaction between site and group). CRP levels were higher in Group B than in Group A by 29.52 µg/ml in gilts and 48.4 µg/ml in barrows. Immunoglobulin M (IgM) decreased after transfer, but less so in Group B than in group A (interaction between site and group). Regardless of site, IgG was several times higher in Group B ($p < 0.01$; Table 6). After transfer, weight gain was higher in Group A than in Group B by 5.9 g in gilts and 20.4 g in barrows (Table 7). These differences, however, were not statistically significant.

DISCUSSION

Transfer from the rearing unit to the finishing house could be a serious stressogenic factor for the pigs in this study. This was due both to stress associated with regrouping, as well as to stress associated with new, and worse, maintenance conditions. Ammonia, volatile organic compounds and dust were much higher in the finishing house than in the rearing facility, which had an adverse effect on the health of the pigs. Total dust in the finishing house was twice as high as the level mandated by zoohygienic regulations.

In the finishing house, chemical air pollutants and biological agents could have a negative synergistic effect on animal health. Bacterial counts reached 4.52×10^5 CFU/m³, which was 97% higher than in the rearing unit. Fungal counts were higher by four orders of magnitude. These values were higher than those set by Donham (1991), who stated that total microbial count in the pig house air should not exceed 10^5 CFU/m³, and many times higher than the levels mandated by zoohygienic regulations for finishing houses (8.0×10^4 CFU/m³).

These results agree with a previous study in which the level of microbial pollutants in farm buildings depended not only on the season and housing system, but also on the nature of building utilization (Létourneau et al., 2010). In that study, total bacteria counts were also higher in the finishing house than in the farrowing and rearing units. Microbial counts are higher in buildings contaminated with dust from hay and straw. Litter bedding was a major source of airborne microbes in the finishing house in the present study. The transfer had a significant effect on red blood cell parameters. HGB markedly decreased, as did MCH and MCHC. MCH and MCHC were below the reference range (Meyer and Harvey, 1998; Winnicka, 2009). In Group B, the RBC count was also higher, although the MCV was lower.

These results agree with a study on poultry maintained in a confined space with high atmospheric ammonia, in which both RBC and HGB were elevated (Witkowska et

Table 5. The level of hematological parameters of pigs from Groups A and B[†].

Parameter	Place	Gilt				Barrow				Group	P-value [§]																																																																																																																																																																																																												
		Group A (N=21)		Group B (N=11)		Group A (N=12)		Group B (N=20)			Site× Group	Sex ×Group	Site ×Sex ×Group																																																																																																																																																																																																										
		M	SD	M	SD	M	SD	M	SD																																																																																																																																																																																																														
WBC (10 ⁹ /l)	RU	15.63	2.78	24.80	2.33	16.10	2.87	24.31	2.85	0.000	0.661	0.743	0.335																																																																																																																																																																																																										
	FH	15.88	2.22	24.26	3.41	15.93	2.61	26.26	6.38					Lim (10 ⁹ /l)	RU	8.97	0.30	15.20	0.15	9.68	0.23	13.50	0.28	0.110	0.150	0.217	0.447	FH	8.14	0.25	11.12	0.03	8.19	0.26	11.54	0.61	Neu (10 ⁹ /l)	RU	5.33	0.31	6.94	0.21	5.25	0.22	8.97	0.98	0.109	0.043	0.050	0.624	FH	6.10	0.27	10.46	0.38	5.81	0.19	12.45	0.68	RBC (10 ¹² /l)	RU	7.00	0.49	7.57	0.27	7.12	0.52	7.37	0.55	0.034	0.105	0.677	0.290	FH	7.16	0.54	7.15	0.54	7.15	0.52	7.28	0.51	MCV (fl)	RU	55.41	2.59	54.03	3.83	54.62	3.50	52.87	4.21	0.023	0.810	0.536	0.748	FH	53.88	1.73	53.19	2.16	54.06	2.55	52.21	2.82	MCH (fmol)	RU	1.03	0.10	1.02	0.07	1.03	0.08	1.00	0.09	0.695	0.450	0.662	0.916	FH	0.96	0.04	0.97	0.04	0.95	0.07	0.95	0.05	MCHC (mmol/l)	RU	18.76	1.72	19.05	1.19	19.05	1.23	19.06	1.33	0.166	0.428	0.928	0.634	FH	17.83	0.26	18.29	1.14	17.69	0.62	18.35	0.96	HCT (l/l)	RU	0.39	0.03	0.40	0.02	0.38	0.03	0.39	0.04	0.848	0.219	0.700	0.820	FH	0.38	0.02	0.38	0.03	0.39	0.03	0.38	0.03	HGB (mmol/l)	RU	7.26	0.81	7.60	0.31	7.32	0.72	7.43	0.98	0.253	0.624	0.713	0.617	FH	6.86	0.37	6.93	0.55	6.83	0.58	6.94	0.37	PLT (10 ⁹ /l)	RU	364.1	109.5	402.5	136.1	394.7	147.9	382.2	130.3	0.237	0.089	0.130	0.625	FH	382.3	101.4	360.5
Lim (10 ⁹ /l)	RU	8.97	0.30	15.20	0.15	9.68	0.23	13.50	0.28	0.110	0.150	0.217	0.447																																																																																																																																																																																																										
	FH	8.14	0.25	11.12	0.03	8.19	0.26	11.54	0.61					Neu (10 ⁹ /l)	RU	5.33	0.31	6.94	0.21	5.25	0.22	8.97	0.98	0.109	0.043	0.050	0.624	FH	6.10	0.27	10.46	0.38	5.81	0.19	12.45	0.68	RBC (10 ¹² /l)	RU	7.00	0.49	7.57	0.27	7.12	0.52	7.37	0.55	0.034	0.105	0.677	0.290	FH	7.16	0.54	7.15	0.54	7.15	0.52	7.28	0.51	MCV (fl)	RU	55.41	2.59	54.03	3.83	54.62	3.50	52.87	4.21	0.023	0.810	0.536	0.748	FH	53.88	1.73	53.19	2.16	54.06	2.55	52.21	2.82	MCH (fmol)	RU	1.03	0.10	1.02	0.07	1.03	0.08	1.00	0.09	0.695	0.450	0.662	0.916	FH	0.96	0.04	0.97	0.04	0.95	0.07	0.95	0.05	MCHC (mmol/l)	RU	18.76	1.72	19.05	1.19	19.05	1.23	19.06	1.33	0.166	0.428	0.928	0.634	FH	17.83	0.26	18.29	1.14	17.69	0.62	18.35	0.96	HCT (l/l)	RU	0.39	0.03	0.40	0.02	0.38	0.03	0.39	0.04	0.848	0.219	0.700	0.820	FH	0.38	0.02	0.38	0.03	0.39	0.03	0.38	0.03	HGB (mmol/l)	RU	7.26	0.81	7.60	0.31	7.32	0.72	7.43	0.98	0.253	0.624	0.713	0.617	FH	6.86	0.37	6.93	0.55	6.83	0.58	6.94	0.37	PLT (10 ⁹ /l)	RU	364.1	109.5	402.5	136.1	394.7	147.9	382.2	130.3	0.237	0.089	0.130	0.625	FH	382.3	101.4	360.5	97.9	471.8	107.4	351.1	95.9																		
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[†]pigs and houses described in Tables 1 and 2. Group A, pigs with WBC counts below 20.0 × 10⁹/l; Group B, pigs with high WBC during the transfer.

al., 2007). However, in another study on piglets housed in buildings with highly polluted air, HGB was lower (Chmielowiec-Korzeniowska, 2011). A third study on pigs revealed that chemical air contaminants had no significant effect on red blood cell parameters; HGB, MCV, MCH and MCHC remained unchanged even when the atmospheric ammonia content reached 50 ppm (Borell et al., 2007).

In the present study, WBC counts increased after transfer, although the difference was not statistically significant. The increase can be attributed to stress associated with the transfer as well as to the contaminated air in the finishing house. Although differences in the concentrations of neutrophils and lymphocytes were detected between the two groups (A and B), in group B lymphocytes decreased and increased neutrophils

were more pronounced. This indicates a stronger response to stressogenic factor in debilitated animal.

Before the transfer, CRP and cortisol levels were elevated. CRP averaged 68.7 µg/ml; higher than reported by Burger et al. (1992), but lower than reported by Chen et al. (2003). After transfer, CRP levels increased to 104.8 µg/ml. The persistently elevated CRP suggests chronic

Table 6. The concentration of CRP, cortisol, and immunoglobulins in blood serum of pigs from Groups A and B[†].

Parameter	Place	Gilt				Barrow				P-value																																																																																										
		Group A (N=21)		Group B (N=11)		Group A (N=12)		Group B (N=20)		Group	Site × Group	Sex × Group	Site × Sex × Group																																																																																							
		M	SD	M	SD	M	SD	M	SD																																																																																											
CRP (µg/ml)	RU	31.80	33.54	109.59	6.01	60.39	34.04	71.01	75.81	0.045	0.036	0.546	0.095																																																																																							
	FH	105.43	48.92	134.95	104.10	63.75	38.86	112.17	19.99					Cortisol (ng/ml)	RU	44.40	6.40	69.63	26.25	52.50	21.46	53.78	18.55	0.392	0.420	0.590	0.336	FH	65.50	35.60	62.49	16.04	58.35	20.45	62.16	28.12	IgA (g/l)	RU	0.40	0.00	0.40	0.00	0.40	0.00	0.40	0.00	0.406	0.406	0.319	0.319	FH	0.40	0.00	0.43	0.04	0.80	0.69	0.47	0.13	IgG (g/l)	RU	2.24	3.84	4.41	5.57	0.55	0.10	8.60	1.07	0.002	0.343	0.077	0.460	FH	4.03	2.66	5.71	3.08	2.79	0.16	6.97	3.41	IgM (g/l)	RU	2.47	0.73	2.25	1.65	2.70	0.00	1.27	0.10	0.395	0.008	0.392	0.073	FH	0.63	0.37	0.85
Cortisol (ng/ml)	RU	44.40	6.40	69.63	26.25	52.50	21.46	53.78	18.55	0.392	0.420	0.590	0.336																																																																																							
	FH	65.50	35.60	62.49	16.04	58.35	20.45	62.16	28.12					IgA (g/l)	RU	0.40	0.00	0.40	0.00	0.40	0.00	0.40	0.00	0.406	0.406	0.319	0.319	FH	0.40	0.00	0.43	0.04	0.80	0.69	0.47	0.13	IgG (g/l)	RU	2.24	3.84	4.41	5.57	0.55	0.10	8.60	1.07	0.002	0.343	0.077	0.460	FH	4.03	2.66	5.71	3.08	2.79	0.16	6.97	3.41	IgM (g/l)	RU	2.47	0.73	2.25	1.65	2.70	0.00	1.27	0.10	0.395	0.008	0.392	0.073	FH	0.63	0.37	0.85	0.40	0.51	0.11	1.18	0.46																		
IgA (g/l)	RU	0.40	0.00	0.40	0.00	0.40	0.00	0.40	0.00	0.406	0.406	0.319	0.319																																																																																							
	FH	0.40	0.00	0.43	0.04	0.80	0.69	0.47	0.13					IgG (g/l)	RU	2.24	3.84	4.41	5.57	0.55	0.10	8.60	1.07	0.002	0.343	0.077	0.460	FH	4.03	2.66	5.71	3.08	2.79	0.16	6.97	3.41	IgM (g/l)	RU	2.47	0.73	2.25	1.65	2.70	0.00	1.27	0.10	0.395	0.008	0.392	0.073	FH	0.63	0.37	0.85	0.40	0.51	0.11	1.18	0.46																																									
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	FH	0.63	0.37	0.85	0.40	0.51	0.11	1.18	0.46																																																																																											

[†] pigs and houses described in Table 1, 2 and 5.

Table 7. Average daily gain (ADG) in rearing unit (between 3 and 10 weeks of age) and finishing house (between 10 and 17 weeks of age) of pigs from Groups A and B[†].

Parameter	Gilt				Barrow				P-value	
	Group A (N=21)		Group B (N=11)		Group A (N=12)		Group B (N=20)		Group	Sex × Group
	M	SD	M	SD	M	SD	M	SD		
Body weight in the 3 week of age; kg	5.7	1.7	5.3	2.3	5.1	2.0	6.9	2.3	0.281	0.097
Body weight in the 10 week of age; kg	33.7	6.6	34.3	8.4	33.5	7.5	33.8	9.2	0.840	0.946
ADG in rearing unit, g/day	455.7	80.8	506.7	71.8	464.2	63.4	452.0	100.9	0.443	0.214
Body weight in the 17 week of age; kg	69.6	8.0	72.8	8.1	69.4	7.8	72.8	10.4	0.224	0.975
ADG in finishing house; g/day	940.2	146.1	934.3	32.1	974.5	80.5	954.1	154.4	0.788	0.882

[†] pigs and houses described in Tables 1, 2 and 5.

arousal of the immunity system. The lack of clinical manifestations or inflammatory response in the herd in this study indicates that high CRP was associated with other, non-infectious factors such as stress induced by animal regrouping, litter

mingling, adaptation to new feeds, and increased pollutant concentration in the finishing house.

These results are consistent with the findings of previous studies which suggest that the stress associated with transfer and adjustment to new

management practices results in increased APP concentration (Heinonen et al., 2010). Other studies suggest that CRP levels are indicative of increased immune system activity in response to microclimate and sanitary conditions (Parra et al.,

2006; Salamano et al., 2010). In one study, wide fluctuations in temperature increased concentrations of acute phase proteins (Chen et al., 2003). Regrouping with animals from different litters also causes short-term but intense stress when animals were transferred.

Changes in the levels of acute phase proteins, including CRP, and the timing of the active phase response depend on genetics, animal age, and the nature of the initiating stimulus. The results of the present study also indicate that the susceptibility of individual animals plays a role, even in relatively stressing situations such as transfer to a new environment where conditions are worse.

The relationships between the hematological and immunological parameters indicate that neutrophil count and CRP are increased not only by transfer and regrouping, but also by other stress factors that disrupt homeostasis, thereby adversely affecting health and well-being. In the present study, this was manifested by the higher increase in neutrophil count and CRP in the pigs in Group B. Furthermore, the increase in RBC count and IgG were also higher in Group B, whereas IgM was lower. IgG, IgM and IgA mediate the primary immune response of an organism to invasion by pathogens. Persistent elevation of IgG with concomitant suppression of IgM indicates ongoing adaptive responses in an organism. In Group B, the differences in immunoglobulin levels measured in the rearing unit and the finishing house were not large enough to determine that adaptive mechanisms were impaired. However, the interpretation of these findings is not explicit, and several discrepancies are found in the toxicological literature. Pedersen and Rasmussen (1992) reported that air pollutants did not affect immunoglobulin levels. On the other hand, Nowakowicz-Dębek et al. (2005) concluded that increased concentrations of volatile contaminants can elevate serum IgM levels.

In the present study, cortisol levels in the pigs studied were lower than those reported by McGlone et al. (1994). This may have been due to differences between the pig breeds under investigation (Sutherland et al., 2006). After transfer, cortisol levels increased, but the difference was not statistically significant. In other studies, there was a statistically significant increase in cortisol levels in pigs during transport and in pigs housed in facilities with elevated concentrations of atmospheric ammonia (McGlone et al., 1994). When pigs were exposed to an ammonia concentration of 35 ppm, acute phase protein levels increased during the first few days after exposure, whereas cortisol levels increased later, as much as 19 days after exposure (Borell et al., 2007). Production performance is directly correlated with animal welfare standards (Ekkel et al., 1996). In their studies, disruption of animal hierarchy after regrouping affected on pigs with high WBC counts. In those pigs transfer to the finishing house reduced the rate of weight gain. The health status of the pigs was good. Changes in CRP and cortisol levels were not associated with infection or disease. Although the presence of subclinical infections cannot be ruled out,

the changes observed were probably caused by other stressogenic factors such as transport, adaptation to a new maintenance system, and worse sanitary conditions. The intensity of the immune response varied among the pigs studied. Those with elevated WBC counts before transport were most susceptible to adaptive stress.

Improper transport and regrouping increase stress, which negatively affects health and productivity. It is therefore important to perform these operations while minimizing stress. An alternative protocol for doing so was proposed by Ekkel et al. (1996), who concluded that the farrow-to-finish system with pigs housed in the same pen from birth to slaughter improves health, welfare and performance.

REFERENCES

- Berg van den A, Danuser J, Frey J, Regula G (2008). Evaluation of the acute phase protein haptoglobin as an indicator of herd health in slaughter pigs. *Anim. Welfare* 16: 157-159.
- Borell E von (2001). The biology of stress and its application to livestock housing and transportation assessment. *J. Anim. Sci.* 79(E. Suppl.): 260-267.
- Borell E von, Özpınar A, Eslinger KM (2007). Acute and prolonged effects of ammonia on hematological variables, stress responses, performance and behavior of nursery pigs. *J. Swine Health Prod.* 15(3): 137-145.
- Chen HH, Lin JH, Fung HP, Ho L-L, Yang PCh, Lee WCh, Lee YP, Chu RM (2003). Serum acute phase proteins and swine health status. *Can. J. Vet. Res.* 67(4): 283-290.
- Chmielowiec-Korzeniowska A (2011). Examination of air biofiltration performance in a fattening unit and the impact on the health and growth of pigs. *Dissert. Study, UP Lublin* (in Polish).
- Donham KJ (1991). Association of environmental air contaminants with disease and productivity in swine. *Am. J. Vet. Res.* 52(10): 1723-1730.
- Ekkel ED, Savenije B, Schouten WG, Tielen MJ (1996). Health, welfare, and productivity of pigs housed under Specific-Stress-Free conditions. *J. Anim. Sci.* 74: 2081-2087.
- Heinonen M, Orro T, Kokkonen T, Munsterhjelm C, Peltoniemi O, Valros A (2010). Tail biting induces a strong acute phase response and tail-end inflammation in finishing pigs. *Vet. J.* 184: 303-307.
- Kołacz R, Bodak E (2000). Acute phase proteins as a criterium of animal welfare evaluation. *Zesz. Nauk. AR Wrocław* 390: 23-31.
- Kołacz R, Cwynar P, Filistowicz M (2009). Genetic progress and health implications in swine breeding. *Med. Wet.* 65(7): 435-438.
- Leek ABG, Sweeney BT, Duffy P, Beattie VE, O'Doherty JV (2004). The effect of stocking density and social regrouping stressors on growth performance, carcass characteristics, nutrient digestibility and physiological stress responses in pigs. *Animal Sci.* 79: 109-119.
- Létourneau V, Nehmé B, Mériaux A, Massé D, Duchaine C (2010). Impact of production systems on swine confinement buildings bioaerosols. *J. Occup. Environ. Hyg.* 7(2): 94-102.
- McGlone JJ, Salak JL, Lumpkin EA, Nicholson RI, Gibson M, Norman RL (1994). Shipping stress and social status effects on pig performance, plasma cortisol, natural killer cell activity, and leukocyte numbers. *J. Anim. Sci.* 71(4): 888-896.
- Meyer DJ, Harvey JW (1998). *Veterinary laboratory medicine. Interpretation & diagnosis.* W.B. Saunders Comp. Philadelphia. 157-187.
- Nowakowicz-Dębek B, Ondrašovič M, Saba L, Krukowski H, Bis-Wencel H, Vargová M (2005). The effect of volatile air pollutants on the immunoglobulin level in the polar fox. *Folia Vet.* 49(4):198-201.
- Parra MD, Fuentes P, Tecles F, Martínez-Subiela S, Martínez JS, Muñoz A, Cerón JJ (2006). Porcine acute phase protein concentrations in different diseases in field conditions. *J. Vet. Med. B* 53: 488-493.

- Pedersen LM, Rasmussen JM (1992). The haematological and biochemical pattern in occupational organic solvent poisoning and exposure. *Int. Arch. Occup. Environ. Health* 51(2): 113-126.
- Salamano G, Mellia E, Tarantola M, Gennero MS, Doglione L, Schiavone A (2010). Acute phase proteins and heterophil:lymphocyte ratio in laying hens in different housing systems. *Vet. Rec.* 167: 749-751.
- Sutherland MA, Niekamp SR, Rodriguez-Zas SL, Salak-Johnson JL (2006). Impacts of chronic stress and social status on various physiological and performance measures in pigs of different breeds. *J. Anim. Sci.* 84(3): 588-596.
- Winnicka A (2009). The reference values of basic laboratory research in veterinary medicine. Press SGGW. Warszawa (in Polish).
- Witkowska D, Szarek J, Iwańczuk-Czernik K, Sowińska J, Mituniewicz T, Wójcik A, Babińska I (2007). Effect of disinfection of litter on the results of rearing and level of blood indices and the image of the internal organs of broiler chickens. *Med. Wet.* 63(9): 1115-1119.
- Volter BF, Ellis M, DeDecker JM, Curtis SE, Hollis GR, Shanks RD, Parr EN, Webel DN (2002). Effects of double-stocking and weighing frequency on pig performance in wean-to-finish production systems. *J. Anim. Sci.* 80(6):1442-1450.