

# Potential biomarkers for chronic seasonal heat stress in Kagoshima Berkshire pigs reared in the subtropical region

Moe Ijiri<sup>1</sup>, Kenji Odo<sup>2</sup>, Motohiko Sato<sup>3</sup>, Maiko Kawaguchi<sup>1,4</sup>,  
Yoshikazu Fujimoto<sup>1</sup>, Naoki Miura<sup>1</sup>, Tomohide Matsuo<sup>1</sup>, De-Xing Hou<sup>5</sup>,  
Osamu Yamato<sup>1</sup>, Takashi Tanabe<sup>2</sup>, Hiroaki Kawaguchi<sup>4,6</sup>✉

<sup>1</sup>Joint Faculty of Veterinary Medicine, Kagoshima University, 890-0065 Kagoshima, Japan

<sup>2</sup>Kagoshima City Meat Inspection Center, 891-0144 Kagoshima, Japan

<sup>3</sup>Department of Surgery, Kyoto University Graduate School of Medicine, 606-8501 Kyoto, Japan

<sup>4</sup>Kagoshima University Graduate School of Medical and Dental Sciences, Kagoshima University, 890-8544 Kagoshima, Japan

<sup>5</sup>Department of Food Science and Biotechnology, Faculty of Agriculture, Kagoshima University, 890-0065 Kagoshima, Japan

<sup>6</sup>Laboratory of Veterinary Pathology, School of Veterinary Medicine, Kitasato University, 034-0021 Aomori, Japan

hiroka@vmas.kitasato-u.ac.jp

*Received: December 08, 2021 Accepted: May 10, 2022*

## Abstract

**Introduction:** Potential biomarkers for chronic seasonal heat stress in Kagoshima Berkshire pigs reared in the subtropical region were investigated by comparing the biomarker changes in the summer (a period of chronic heat stress) and winter (a thermoneutral period) seasons. **Material and Methods:** Pigs were allocated to summer- and winter-finishing cohorts, 12 each. The evaluations included assessment of carcass traits and internal organs' normality carried out at the time of slaughter, and measurement of biomarkers in whole blood: derivatives of reactive oxygen metabolites (d-ROMs) and biological antioxidant potential as markers of oxidative stress, and serum amyloid A and albumin/globulin (A/G) ratio as markers of acute and chronic inflammation, respectively. **Results:** The summer-finished pigs reared under subtropical field conditions showed lower carcass quality than the winter-finished pigs, indicating a potential adverse effect of summer temperatures on the swine industry. Marginal changes were observed in d-ROMs and the A/G ratio between the summer- and winter-finishing cohorts. **Conclusion:** The results demonstrate that d-ROMs and the A/G ratio could be used as sensitive markers for heat stress under field conditions.

**Keywords:** albumin/globulin ratio, derivatives of reactive oxygen metabolites (d-ROMs), biological antioxidant potential (BAP), heat stress, Kagoshima Berkshire pig, oxidative stress.

## Introduction

Heat stress has an adverse impact on livestock production as elevation of temperature can result in compensatory changes in animal behaviour, physiology, and metabolism. This impact is particularly marked in pigs as they are more poorly adapted to heat dissipation than other livestock species because of the paucity of sweat glands and the thick subcutaneous fat. The porcine response to increasing environmental temperatures appears to be based on reducing metabolic heat production to maintain eutheria (16). One example of a behavioural response in pigs is the

reduction of metabolic heat by decreasing feed intake, which is effective because digestion and nutrient absorption generate heat (17). Physiologically, elevations in environmental temperature also directly alter pigs' nutrient utilisation, energy metabolism, and hormonal regulation (5, 9, 24). Consequently, heat stress can cause substantial economic losses for the swine industry through its effect on growth rates, carcass composition, sow performance, mortality and morbidity (2, 22, 27).

Researchers have proposed a number of potential markers of porcine heat stress, often based on studies of it at elevated temperatures under laboratory conditions.

Candidates have been suggested based on the inflammatory and acute-phase response known to be induced by heat in humans and pigs (5, 26). Among the proteins involved in the acute response, serum amyloid A (SAA) and serum albumin have been identified as positive and negative acute-phase reactants, respectively (6). Acute phase proteins (APPs) such as SAA, haptoglobin, and C-reactive protein are recognised markers of inflammation and have also been proposed as indicators of stress in cattle (14) and pigs (20).

Heat stress under laboratory conditions may not always accurately reflect the field conditions under which pigs are raised. Consequently, the swine industry could benefit from further research on sensitive biomarkers of the type of heat stress experienced by pigs under field conditions. Recent advances in detecting reactive oxygen species (ROS) using derivatives of reactive oxygen metabolites (d-ROMs) and biological antioxidant potential (BAP) have opened potential avenues in biomarker research which could be exploited for strategies to combat porcine heat stress. Heat stress is generally considered to trigger oxidative stress in livestock animals (21), and acute heat stress in pigs has been shown to cause oxidative damage and up-regulation of antioxidant enzymes in their oxidative muscle (21). Chronic exposure to high environmental temperatures accelerates ROS generation, potentially inducing cytotoxicity ranging from lipid peroxidation to oxidative damage to protein damage (1).

Oxidative stress may be involved in various disorders and pathogenesises in humans (7, 10, 11). As demonstrably established markers of oxidative stress in areas of bioresearch such as human lifestyle diseases (7), d-ROMs and BAP represent a promising line of investigation as markers of such stress in animals, but have only been evaluated in a limited number of studies in domestic animals so far (18, 23, 25, 29).

Field conditions vary with season and climate zone, meaning that research during different seasons in diverse areas of the globe can yield data which complement each other. Kagoshima Prefecture in southwest Japan is a major pig-producing area located in the subtropical zone. The Kagoshima Berkshire pigs raised in this prefecture are used by one of the most famous pork brands in Japan, called 'Kagoshima Kurobuta'. Pigs raised in the subtropical summer are likely to be subject to a milder form of heat stress than those investigated previously in many laboratory-based studies. The subtropical summer represents a chronic form of heat stress, and Kagoshima Prefecture is thus a suitable location to elucidate sensitive markers of chronic heat stress in pigs under field conditions.

Accordingly, in this study, we set out to investigate potential markers of chronic heat stress by comparing Kagoshima Berkshire pigs finished in the summer (a period of chronic heat stress) and in the winter (a thermoneutral period) in Kagoshima Prefecture. The evaluations included assessment of

carcass traits (made at the time of slaughter), biomarkers previously established for heat stress, and d-ROMs and BAP as markers of oxidative stress.

## Material and Methods

**Animals and experiment design.** Twenty-four finishing pigs (Kagoshima Berkshire) were reared on commercial pig farms in Kagoshima Prefecture. Twelve pigs were allocated to a summer finishing cohort (August to September with average local temperatures of 29.6°C and 26.4°C, respectively) and 12 pigs were allocated to a winter-finishing cohort (November to December with average local temperatures of 16.0°C and 12.2°C, respectively).

The temperature and relative humidity (RH, %) in the pig housing area were recorded on an hourly basis (Thermo-Hygro Data Logger RX-350TH; AS ONE Corporation, Osaka, Japan) and represented as mean  $\pm$  SD (Table 1).

The pigs were slaughtered at the end of the two-month finishing period and whole blood was collected during the pigs' exsanguination. The research was carried out according to the Institutional Guidelines for Animal Experiments, in compliance with the Japanese Act on Welfare and Management of Animals (Act No. 105 and Notification No. 6).

**Inspection of carcasses and internal organ disposal.** Each slaughtered pig's back fat thickness and carcass weight were measured, and the quality of its carcass was assessed based on these parameters according to the criteria of the Japan Meat Grading Association (under which grade 3 represents high quality, grade 2 represents middle quality, grade 1 low quality, and grade 0 substandard). Organs and tissue were checked as part of the routine meat inspection, and instances of condemned organs were recorded to determine the overall incidence. All these tests were performed blind.

**Blood sample analysis.** The plasma was obtained by centrifugation (4°C, 3,000 rpm, 15 min) and used for the measurement of catecholamines. The serum was obtained by centrifugation (room temperature, 3000 rpm, 15 min) and used for the measurement of other parameters indicated in Tables 3–5. Analyses of the cortisol and catecholamine biochemical parameters were commissioned to the Clinical Pathology Laboratory, Co. Ltd. (Kagoshima, Japan). The concentration of serum globulin (Glb) and albumin/globulin (A/G) ratio were calculated from the measured values for total protein (TP) and serum albumin (Alb).

**Measurement of serum amyloid A (SAA) concentration.** The concentration of SAA was measured using a Pentra C200 automated biochemical analyser (HORIBA ABX SAS, Montpellier, France) with an SAA reagent specialised for animal serum or plasma (VET-SAA 'Eiken' Reagent; Eiken Chemical Co., Ltd., Tokyo, Japan). In addition, the concentration

of SAA was calculated on the basis of a standard curve made by a VET-SAA Calibrator Set (Eiken Chemical Co., Ltd.).

**Measurement of blood oxidative stress.** Serum d-ROMs and BAP represent reactive oxygen metabolites and antioxidant capacity, respectively. Serum levels of these two markers were measured using a FREE Carrio Duo Redox Analyzer system (Wismerll, Tokyo, Japan), which included a photometer and a thermostatically regulated mini-centrifuge. Both the d-ROMs and BAP tests were conducted according to the manufacturer's instructions.

**Statistical analysis.** Numerical data other than the incidences of condemned organs are expressed as mean  $\pm$  standard deviation. All data were analysed using IBM SPSS Statistics 25 software (IBM, Armonk, NY, USA). Data on carcass quality and incidences of condemned organs were analysed using Pearson's chi-square test and Fisher's exact test, respectively. Other data were analysed using the Mann-Whitney U test. Values of  $P < 0.05$  were considered statistically significant.

## Results

**Environmental condition.** Summer-finished pigs were reared under higher average whole-day temperatures and relative humidity than winter-finished

pigs. Nocturnal temperatures were around 2°C lower than daytime temperatures for summer-finished pigs and around 4°C lower for winter-finished pigs. Average relative humidity varied within a narrow range for summer- and winter-finished pigs (Table 1).

**Inspection of carcasses and internal organs.** Data collected at slaughter were compared between summer-finished and winter-finished pigs (Table 2). Summer-finished pigs tended to have lower carcass quality ( $P = 0.065$ ) and significantly greater back fat thickness. Incidences of condemned internal organs did not differ between summer-finished and winter-finished pigs.

**Blood biochemistry and stress markers.** In blood biochemistry (Table 3), summer-finished pigs showed significantly lower mean Na and Cl concentrations than those in winter-finished pigs, and tended to have a higher TP value ( $P = 0.052$ ). Globulin and the A/G ratio also differed significantly between the summer-finished and the winter-finished pigs. On the other hand, there was no difference in blood stress markers between the groups of pigs (Table 4).

We investigated d-ROMs and BAP as indices of ROS production and antioxidant potential, respectively. The summer-finished pigs' levels of d-ROMs were significantly higher than those of the winter-finished pigs, although BAP and BAP/d-ROMs did not differ between the two seasonal cohorts (Table 5).

**Table 1.** Environmental temperature and relative humidity in the pig house

Group	Temperature (°C)			Relative humidity (%)		
	Whole day	Daytime (time)	Night (time)	Whole day	Daytime(time)	Night (time)
Summer <sup>1</sup>	28.3 $\pm$ 2.4	29.3 $\pm$ 2.6	27.1 $\pm$ 1.6	80.7 $\pm$ 6.4	80.5 $\pm$ 6.8	81.0 $\pm$ 6.0
		(6:00–18:00)	(19:00–5:00)		(6:00–18:00)	(19:00–5:00)
Winter <sup>2</sup>	20.2 $\pm$ 4.9	22.2 $\pm$ 4.5	18.5 $\pm$ 4.6	71.7 $\pm$ 4.6	71.4 $\pm$ 4.7	71.9 $\pm$ 4.5
		(7:00–17:00)	(18:00–6:00)		(7:00–17:00)	(18:00–6:00)

<sup>1</sup> – Summer-finished pigs. The data for this group was recorded from July to September

<sup>2</sup> – Winter-finished pigs. The data for this group was recorded from October to December

**Table 2.** Results of inspection of carcasses and incidences of internal organ condemnation

Group	Carcass result score*			Internal organs condemnation <sup>#</sup> (Incidence, %)		
	Result (Score <sup>§</sup> )	Back fat thickness (cm)	Weight (kg)	Heart	Liver	Stomach and intestines
Summer <sup>1</sup>	1.1 $\pm$ 0.3	4.1 $\pm$ 0.8	78.7 $\pm$ 3.3	0	50.0	41.7
Winter <sup>2</sup>	1.8 $\pm$ 0.9	2.5 $\pm$ 0.8	80.6 $\pm$ 4.3	0	50.0	33.3
P value	0.065	0.000	0.340		1.00	1.00

<sup>1</sup> – Summer-finished pigs slaughtered from August to September

<sup>2</sup> – Winter-finished pigs slaughtered from November to December

<sup>§</sup> – 3: high grade, 2: middle grade, 1: low grade

Data were analysed using Pearson's chi-square test (\*) and Fisher's exact test (<sup>#</sup>)

**Table 3.** Common blood biochemistry analyte concentrations

Analyte	Unit	Summer <sup>1</sup>	Winter <sup>2</sup>	P value
LDH	U/L	1143.6 ± 829.8	737.9 ± 207.4	0.119
CK	U/L	11149.8 ± 23757.1	3778.4 ± 4034.7	0.453
TP	g/dL	8.2 ± 0.5	7.9 ± 0.6	0.052
Alb	g/dL	4.5 ± 0.4	4.8 ± 0.3	0.163
Glb	g/dL	3.7 ± 0.6	3.1 ± 0.5	0.015*
A/G	ratio	1.3 ± 0.3	1.6 ± 0.2	0.019*
Na	mEq/L	148.5 ± 4.0	151.8 ± 4.3	0.032*
Cl	mEq/L	98.6 ± 2.3	101.1 ± 3.4	0.048*

<sup>1</sup> – Summer-finished pigs slaughtered from August to September

<sup>2</sup> – Winter-finished pigs slaughtered from November to December

LDH – Lactate dehydrogenase; CK – Creatinine kinase; TP – Total protein;

Alb – Albumin; Glb – Globulin; A/G – Albumin/globulin ratio

\* – P < 0.05; significantly different between summer and winter

**Table 4.** Blood stress marker concentrations

Marker	Unit	Summer <sup>1</sup>	Winter <sup>2</sup>	P value
Cortisol	µg/dL	4.6 ± 2.3	4.5 ± 2.4	0.931
Adrenalin	ng/mL	42.9 ± 15.7	46.2 ± 28.7	0.729
Noradrenaline	ng/mL	49.3 ± 19.9	41.2 ± 26.5	0.299
Dopamine	ng/mL	not detected	not detected	
Serum amyloid A	mg/L	6.0 ± 4.7	6.0 ± 5.1	0.773

<sup>1</sup> – Summer-finished pigs slaughtered from August to September

<sup>2</sup> – Winter-finished pigs slaughtered from November to December

**Table 5.** Blood oxidative stress

Parameter	Unit	Summer <sup>1</sup>	Winter <sup>2</sup>	P value
d-ROMs	U. CARR	1106.2 ± 136.6	970.8 ± 83.0	0.017*
BAP	µmol/L	4123.0 ± 771.7	4000.2 ± 306.4	0.356
BAP/d-ROMs		3.8 ± 0.9	4.1 ± 0.4	0.488

<sup>1</sup> – Summer-finished pigs slaughtered from August to September

<sup>2</sup> – Winter-finished pigs slaughtered from November to December

d-ROMs – derivatives of reactive oxygen metabolite-derived compounds

(1 U. CARR = 0.08 mg H<sub>2</sub>O<sub>2</sub>/dL); BAP – biological antioxidant potential

\* – P < 0.05; significantly different between summer and winter

## Discussion

To the best of our knowledge, this study represents the first evaluation of porcine heat stress in a subtropical region by comparing summer-finished and winter-finished pigs slaughtered at the end of a two-month finishing period. Also, we believe this study is a valuable attempt at the relative characterisation of markers of heat stress and particularly of d-ROMs and BAP as indices of ROS derivatives and antioxidant potential, respectively, in farm-reared pigs under seasonal conditions expected to

produce chronic heat stress (summer) and thermoneutral temperatures (winter).

Based on the results of environmental temperature in the pig house and the large summer variance there from the thermoneutrality of the temperature range of 10 to 23.9°C (17) in which the winter-finished pigs were raised, we consider that the summer-finished pigs experienced chronic heat stress. Previous studies have expanded the definition of acute and chronic heat stress in terms of exposure time and temperature for pigs (9, 21, 28). Reportedly, chronic heat stress has been induced under laboratory conditions in finishing pigs

exposed to constant temperatures of 30°C or more for three weeks (5, 6). In this study, the summer-finished pigs experienced a slightly lower daytime temperature (around 29°C), with a further decrease in nocturnal hours, but for a more prolonged period (two months). We thus consider that the conditions of the summer finishing period correspond to chronic heat stress.

The results of carcass quality assessment suggest a chronic heat stress effect. The greater back fat thickness in summer-finished pigs is considered to be a poorer result. More than 90% of the summer-finished pigs were measured at back fat thickness exceeding the upper limit of the criterion of the Japan Meat Grading Association (12). Although the difference in carcass quality was significant, our study results do not allow for definite conclusions on fat accumulation as a heat stress effect under field conditions. However, heat stress has been previously implicated in the upregulation of lipoprotein lipase (13) and fatty acid synthase activities (19), resulting in increased adipose tissue. Further research is needed to elucidate the mechanisms of heat stress-induced metabolic changes under field conditions, and to explain the subsequent alterations in carcass phenotype.

Heat stress alters metabolism, structure and antioxidant mechanisms in skeletal muscle (4). Heat exposure of sows during pregnancy affects foetal development and post-growth carcass quality (3, 27). In growing pigs, it has been reported that acute heat stress disrupts muscle redox balance and inflammatory signalling (8, 21). The differences between summer- and winter-finishing cohorts were considered to reflect adverse effects of seasonal heat stress, with Na and Cl levels suggestive of hypotonic dehydration; however, the blood biochemistry profiles of summer-finished pigs did not correspond to a state of disease. Our data on blood chemistry and blood stress markers provide an interesting contrast to previous studies of chronic heat stress under laboratory conditions. Finishing pigs housed at 30°C for three weeks exhibited increased cortisol, lactate dehydrogenase (LDH), and creatinine kinase (CK) (9). The mean values of LDH and CK in our study were considerably higher in the summer-finished pigs, although the differences were not statistically significant. Our data implies that the adverse effect on muscle tissue was apparently not of sufficient magnitude to be detected in the measurements, including those of cortisol and other blood stress markers.

The presence of a chronic heat stress phenomenon in our study is further substantiated by the A/G ratio and SAA. The A/G ratio decreases with an increase in the globulin level, which implies chronic inflammation (15), and SAA is an acute-phase protein. The significantly lower A/G ratio (Table 3) in summer-finished pigs was not accompanied by any change in SAA (Table 4). Taken together with the other biochemistry and stress marker results, this suggests that the seasonal heat stress was chronic, but not

sufficiently severe to elevate stress marker levels. Furthermore, the A/G ratio might be a potential marker of chronic heat stress under field conditions.

Set against our results of blood biochemistry and stress markers, the novel findings on markers of blood oxidative stress in this study are of considerable interest. Short-term heat exposure reportedly increases oxidative stress marker levels (malondialdehyde, catalase activity and superoxide dismutase) in porcine skeletal muscles (21, 28), which could be consistent with the elevated plasma d-ROM levels in this study. This elevation in d-ROMs was not accompanied by any increase in stress hormone levels, and we consider that this change might be detectable before any changes in hormonal regulation of cortisol and other blood stress markers. Accordingly, we consider that d-ROMs might be more sensitive markers of heat stress than the blood stress markers previously suggested.

In conclusion, the summer-finished pigs reared under subtropical field conditions in this study showed evidence of chronic heat stress, reflected in lower carcass quality than that of winter-finished pigs. This demonstrates the potential adverse effects of such high temperature levels on the swine industry, and underlines the need for sensitive markers to monitor chronic heat stress. We further consider that d-ROMs and the A/G ratio may have utility as sensitive markers for such heat stress under field conditions.

**Conflict of Interests Statement:** The authors declare that there is no conflict of interests regarding the publication of this article.

**Financial Disclosure Statement:** This study was partly supported by a research grant from the Ito Foundation (HK). The funder had no role in designing the research, collecting and analysing data, deciding to publish or preparing the manuscript.

**Animal Rights Statement:** Ethical approval for this study was waived by the Ethics Committees of Animal Care and Experimentation, Kagoshima University, because it did not include any experimental treatment of animals and samples were obtained during commercial pig slaughter. Written informed consent was obtained from the farmers. The research was performed according to the Institutional Guidelines for Animal Experiments and in compliance with the Japanese Act on Welfare and Management of Animals (Act No. 105 and Notification No. 6).

**Acknowledgements:** We thank Mr Naoki Nogami and Mr Yoshiaki Shimoo for raising the Kagoshima Berkshire pigs. We are also grateful to Ms Shiho Suzuki for her technical assistance. We are particularly grateful to Mr Henry Smith for pertinent English revision of the manuscript.

## References

- Akbarian A., Michiels J., Degroote J., Majdeddin M., Golian A., De Smet S.: Association between heat stress and oxidative stress in poultry; mitochondrial dysfunction and dietary interventions with phytochemicals. *J Anim Sci Biotechnol* 2016, 7, 37, doi: 10.1186/s40104-016-0097-5.
- Baumgard L.H., Rhoads R.P. Jr.: Effects of heat stress on postabsorptive metabolism and energetics. *Annu Rev Anim Biosci* 2013, 1, 311–337, doi: 10.1146/annurev-animal-031412-103644.
- Cruzen S.M., Boddicker R.L., Graves K.L., Johnson T.P., Arkfeld E.K., Baumgard L.H., Ross J.W., Safranski T.J., Lucy M.C., Lonergan S.M.: Carcass composition of market weight pigs subjected to heat stress in utero and during finishing. *J Anim Sci* 2015, 93, 2587–2596, doi: 10.2527/jas.2014-8347.
- Cruzen S.M., Pearce S.C., Baumgard L.H., Gabler N.K., Huff-Lonergan E., Lonergan S.M.: Proteomic changes to the sarcoplasmic fraction of predominantly red or white muscle following acute heat stress. *J Proteomics* 2015, 128, 141–153, doi: 10.1016/j.jprot.2015.07.032.
- Cui Y., Gu X.: Proteomic changes of the porcine small intestine in response to chronic heat stress. *J Mol Endocrinol* 2015, 55, 277–293, doi: 10.1530/JME-15-0161.
- Cui Y., Wang C., Hao Y., Gu X., Wang H.: Chronic heat stress induces acute phase responses and serum metabolome changes in finishing pigs. *Animals* 2019, 9, 395, doi: 10.3390/ani9070395.
- Fukui T., Yamauchi K., Maruyama M., Yasuda T., Kohno M., Abe Y.: Significance of measuring oxidative stress in lifestyle-related diseases from the viewpoint of correlation between d-ROMs and BAP in Japanese subjects. *Hypertens Res* 2011, 34, 1041–1045, doi: 10.1038/hr.2011.76.
- Ganesan S., Reynolds C., Hollinger K., Pearce S.C., Gabler N.K., Baumgard L.H., Rhoads R.P., Selsby J.T.: Twelve hours of heat stress induces inflammatory signaling in porcine skeletal muscle. *Am J Physiol Regul Integr Comp Physiol* 2016, 310, 1288–1296, doi: 10.1152/ajpregu.00494.2015.
- Hao Y., Feng Y., Yang P., Feng J., Lin H., Gu X.: Nutritional and physiological responses of finishing pigs exposed to a permanent heat exposure during three weeks. *Arch Anim Nutr* 2014, 68, 296–308, doi: 10.1080/1745039X.2014.931522.
- Hirata Y., Yamamoto E., Tokitsu T., Fujisue K., Kurokawa H., Sugamura K., Sakamoto K., Tsujita K., Tanaka T., Kaikita K., Hokimoto S., Sugiyama S., Ogawa H.: The pivotal role of a novel biomarker of reactive oxygen species in chronic kidney disease. *Medicine* 2015, 94, e1040, doi: 10.1097/MD.0000000000001040.
- Hirata Y., Yamamoto E., Tokitsu T., Kusaka H., Fujisue K., Kurokawa H., Sugamura K., Maeda H., Tsujita K., Kaikita K., Hokimoto S., Sugiyama S., Ogawa H.: Reactive oxygen metabolites are closely associated with the diagnosis and prognosis of coronary artery disease. *J Am Heart Assoc*, 2015, 4, e001451, doi: 10.1161/JAHA.114.001451.
- Japan Meat Grading Association, Tokyo: Instruction manual of trading standard on carcass of cattle and swine, partial meat of cattle and swine (in Japanese), Japanese Meat Grading Association, Tokyo, 1996, pp. 32–45.
- Kouba M., Hermier D., Le Dividich J.: Influence of a high ambient temperature on lipid metabolism in the growing pig. *J Anim Sci* 2001, 79, 81–87, doi: 10.2527/2001.79181x.
- Lomborg S.R., Nielsen L.R., Heegaard P.M., Jacobsen S.: Acute phase proteins in cattle after exposure to complex stress. *Vet Res Commun* 2008, 32, 575–582, doi: 10.1007/s11259-008-9057-7.
- Maeda S., Takeya Y., Oguro R., Akasaka H., Ryuno H., Kabayama M., Yokoyama S., Nagasawa M., Fujimoto T., Takeda M., Onishi-Takeya M., Itoh N., Takami Y., Yamamoto K., Sugimoto K., Inagaki H., Ogawa M., Nakagawa T., Yasumoto S., Masui Y., Arai Y., Ishizaki T., Ikebe K., Gondo Y., Kamide K., Rakugi H.: Serum albumin/globulin ratio is associated with cognitive function in community-dwelling older people: The Septuagenarians, Octogenarians, Nonagenarians Investigation with Centenarians study. *Geriatr Gerontol Int* 2019, 19, 967–971, doi: 10.1111/ggi.13751.
- Mayorga E.J., Renaudeau D., Ramirez B.C., Ross J.W., Baumgard L.H.: Heat stress adaptations in pigs. *Anim Front* 2018, 9, 54–61, doi: 10.1093/af/vfy035.
- Myer R., Bucklin R.: Influence of hot-humid environment on growth performance and reproduction of swine. Document AN107, Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida, Gainesville, FL, 2001, <https://www.swineweb.com/influence-of-hot-humid-environment-on-growth-performance-and-reproduction-of-swine/>.
- Otomaru K., Oishi S., Fujimura Y., Iwamoto Y., Nagai K., Ijiri M.: Effects of vitamin C supplementation on the blood oxidative stress and antibody titre against *Histophilus somni* vaccination in calves. *J Vet Res* 2021, 65, 73–78, doi: 10.2478/jvetres-2021-0002.
- Pearce S.C., Gabler N.K., Ross J.W., Escobar J., Patience J.F., Rhoads R.P., Baumgard L.H.: The effects of heat stress and plane of nutrition on metabolism in growing pigs. *J Anim Sci* 2013, 91, 2108–2118, doi: 10.2527/jas.2012-5738.
- Piñeiro M., Piñeiro C., Carpintero R., Morales J., Campbell F.M., Eckersall P.D., Toussaint M.J., Lampreave F.: Characterisation of the pig acute phase protein response to road transport. *Vet J* 2007, 173, 669–674, doi: 10.1016/j.tvjl.2006.02.006.
- Rosado Montilla S.I., Johnson T.P., Pearce S.C., Gardan-Salmon D., Gabler N.K., Ross J.W., Rhoads R.P., Baumgard L.H., Lonergan S.M., Selsby J.T.: Heat stress causes oxidative stress but not inflammatory signaling in porcine skeletal muscle. *Temperature (Austin)* 2014, 1, 42–50, doi: 10.4161/temp.28844.
- Ross J.W., Hale B.J., Gabler N.K., Rhoads R.P., Keating A.F., Baumgard L.H.: Physiological consequences of heat stress in pigs. *Anim Prod Sci* 2015, 55, 1381–1390, doi: 10.1071/AN15267.
- Sauerwein H., Schmitz S., Hiss S.: The acute phase protein haptoglobin and its relation to oxidative status in piglets undergoing weaning-induced stress. *Redox Rep* 2005, 10, 295–302, doi: 10.1179/135100005X83725.
- Serviento A.M., Labussière E., Castex M., Renaudeau D.: Effect of heat stress and feeding management on growth performance and physiological responses of finishing pigs. *J Anim Sci* 2020, 98, skaa387, doi: 10.1093/jas/skaa387.
- Shono S., Gin A., Minowa F., Okubo K., Mochizuki M.: The Oxidative Stress Markers of Horses—the Comparison with Other Animals and the Influence of Exercise and Disease. *Animals* 2020, 10, 617, doi: 10.3390/ani10040617.
- Starkie R.L., Hargreaves M., Rolland J., Febbraio M.A.: Heat stress, cytokines, and the immune response to exercise. *Brain Behav Immun* 2005, 19, 404–412, doi: 10.1016/j.bbi.2005.03.005.
- Tuell J.R., Nondorf M.J., Maskal J.M., Johnson J.S., Kim Y.H.B.: Impacts of in Utero Heat Stress on Carcass and Meat Quality Traits of Market Weight Gilts. *Animals* 2021, 11, 717, doi: 10.3390/ani11030717.
- Volodina O., Ganesan S., Pearce S.C., Gabler N.K., Baumgard L.H., Rhoads R.P., Selsby J.T.: Short-term heat stress alters redox balance in porcine skeletal muscle. *Physiol Rep* 2017, 5, e13267, doi: 10.14814/phy2.13267.
- Yun W., Song M., Lee J., Oh H., An J., Kim G., Lee S., Lee S., Kim H.B., Cho J.: Arginine addition in a diet for weaning pigs can improve the growth performance under heat stress. *J Anim Sci Technol* 2020, 62, 460–467, doi: 10.5187/jast.2020.62.4.460.