1 **JAST (Journal of Animal Science and Technology) TITLE PAGE**

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Authors' contributions Please specify the authors' role using this form.	Conceptualization: Kim JE, Choi YH, Kim JS Data curation: Park HJ, Min YJ, Jeong YD, Jeon DY, Kim JE, Choi YH Formal analysis: Kim JE, Choi YH, Mun JY, Ha SH Methodology: Kim JE, Choi YH, Kim JS Software: Park HJ, Min YJ, Choi YH, Hosseindoust A Validation: Choi YH, Jin HJ, Hosseindoust A Investigation: Kim JE, Kim JS, Mun JY, Ha SH Writing - original draft: Kim JE, Choi YH, Kim JS
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Abstract

The study evaluated the effects of dietary fiber and energy levels administered during two growing periods (0-28 d and 29-56 d) for pigs exposed to a high temperature. A total of 96 growing pigs were used in six treatments as: Two treatments in thermoneutral temperature (21-24 ºC) with dietary energy of 3,300 and the inclusion of high or low fiber, two treatments in heat stress (30-34 ºC) with dietary energy of 3,300 and the inclusion of high or low fiber, and two treatments in heat stress with dietary energy of 3,450 and the inclusion of high or low fiber. Among standard energy level treatments, heat-stressed pigs showed lower average daily gain (ADG), feed intake, digestibility of dry matter, gross energy, crude protein, and crude fiber in phases 1 and 2. Moreover, higher concentrations of acetate, propionate, butyrate, and total short-chain fatty acid (SCFA) in feces were shown in pigs fed high fiber diets. There was a negative interaction between dietary fiber and energy for the fecal concentration of isobutyrate in phase 1 and valerate in phase 2. Pigs in heat stress treatments showed a higher rectal temperature, respiratory rate, hair cortisol, plasma zonulin, and fecal lipocalin-2. Among heat stress treatments, the overall ADG was increased in pigs fed high fiber. Pigs fed high dietary fiber showed a greater concentration of acetate, propionate, butyrate, and total SCFA. High fiber treatments decreased plasma zonulin. In conclusion, the inclusion of beet pulp, soluble fiber, at the level of 4% looks necessary in pigs diet during heat stress.

Keywords: Intestine, fermentation, zonulin, cortisol, stress, soluble fiber

Introduction

Global warming is one of the most significant obstacles faced by the swine industry during hot and semi-hot seasons [1,2]. Heat stress compromises the growth performance and meat quality of growing pigs [3]. The first responses of animals to diminish metabolic heat increment are decreased average daily feed intake (ADFI) and metabolic interactions [1,2]. Low feed intake caused by heat stress in swine leads to imbalanced energy, body composition, and growth performance [4,5]. In heat-stressed pigs, not only feed intake but also intestinal integrity can be disrupted [3]. Additionally, intestinal fermentation order and short-chain fatty acid production can be affected [6]. In normal conditions, performance, welfare, and intestinal integrity were all increased in pigs fed 3% dietary fiber [7]. However, during heat stress, it has long been a practice to decrease dietary fiber and protein to lessen heat increment and mitigate the negative effects of high temperatures [4,8].

There is a lack of insight into how supplementing sources or levels of fiber during thermal stress affects the growth performance and health of growing pigs. Based on physical and chemical properties, dietary fiber is classified as soluble fiber (SF) or insoluble fiber (ISF) [1]. Pectin, gum, β-glucan, and hemicellulose are present in the SF, while cellulose, lignin, and insoluble arabinoxylan are found in the ISF [9]. The interaction between SF with water in digesta increases the volume of stools, resulting in more typical bowel motions, which may contribute to the mitigation of constipation incidence [7]. Furthermore, SF reduces the occurrence of diarrhea and improves water-holding capacity, gut health, and short-chain fatty acid (SCFA) production, leading to a stable environment in the intestine [1]. Understanding how dietary SF can provide interactive advantages during heat stress is crucial due to the benefits of fiber in promoting intestinal growth in growing pigs. To analyze the effects on growth performance, nutrient digestibility, stress status, and metabolic changes in the gut, the diets in the current study were designed based on SF supplementation and different dietary energy levels in heat-stressed pigs.

Material and methods

This experiment was carried out at the Republic of Korea's National Institute of Animal Science Farm.

Animals and experimental design

The Institutional Animal care and Use Committee at the National Institute of Animal Science acknowledged the experiment's adequate observance of ethical norms and institutional protocols (NIAS2020-459). All experiments were conducted in conformity with the necessary legislation and guidelines. Growing pigs (Landrace × Yorkshire × Duroc) were placed in partially slatted and concrete floor cages (2.8 × 2.0 m). A nipple drinker and a self-feeder were provided in each pen to allow ad libitum access to feed and water. A total of 96 growing pigs (initial BW, 56.6 ± 2.23 kg) were selected to be distributed randomly to one of six treatments. Each treatment had eight replicate pens with two pigs per pen. Two treatments in thermoneutral (TN) temperature (21-24 ºC) with dietary energy of 3,300 and the inclusion of high or low fiber, two treatments in heat stress (30-34 ºC) with dietary energy of 3,300 and the inclusion of high or low fiber, and two treatments in heat stress with dietary energy of 3,450 and the inclusion of high or low fiber in two growing periods (0-28 d and 29-56 d). Beet pulp was used as the source of fiber including 72.5% total fiber, 23.4% SF, and 49.1% ISF. According to the National Research Council [10], all of the diets met or surpassed the nutritional requirements for grower pigs. The pens were equipped with temperature and humidity monitoring devices (Campbell Scientific Ltd., Shepshed, U.K.) that were positioned 1 m above the floor and utilized to continually record temperatures and relative humidity every 10 minutes. The recorded humidity and temperature were used for calculating the temperature and humidity index (THI) as described by Ataallahi et al. [11]. The THI was calculated using the following formula: THI = [(1.8 × temperature (ºC) + 32) – (0.55 - 0.0055 × humidity) × (1.8 × temperature (ºC) – 26)]. The recorded temperature and THI are shown in Fig. 1**.**

Rectal temperature and respiratory rate

Throughout the trial, the rectal temperature was recorded every morning and evening at 8:00 to 9:00 and 19:00 to 20:00, respectively, using a digital thermometer that was inserted 2 cm into the rectum [2].

By counting the number of breaths per minute while watching the thoracic movement of the sow in a lying down position, the respiratory rate was simultaneously measured.

Nutrient digestibility

The digestibility of nutrients was evaluated as previously mentioned [12]. The nutrient digestibility evaluation was conducted to determine the apparent digestibility of dry matter (DM), gross energy (GE), and crude protein (CP) at the end of phases 1 and 2. On day 28 of phase 1 and day 56 of phase 2, the two chosen pigs were put in separate cages (one pig per cage) in order to collect fecal samples. Chromium oxide, an indigestible marker, was given to the feed of the chosen pigs at a rate of 2.5 g kg^{-1}. For phase 1 and phase 2, feces samples were taken on days 26 through 28 and 54 through 56 respectively. Following sample collection, the drying (60℃, 72 h) step was carried out in a forced air-drying oven, and the material was thereafter ground with a mill (Thomas Model 4 Wiley Mill, Thomas Scientific, Swedesboro, NJ, USA) using a 1 mm screen. DM and CP diet and feces analyses were carried out according to the AOAC [13] technique. The concentration of Cr_2O_3 was measured using a calorimeter bomb (Model 1261, Parr Instrument, Moline, IL, USA) and a spectrophotometer (Jasco V-650, Jasco, Tokyo, Japan). Every day at 7:00 a.m. and 15:30 a.m., feces were collected, combined, sealed in plastic bags to be frozen at -20 ℃. The fecal samples from each pig were collected, frozen, and pooled, dried for 72 hours in a forced-draft oven (65 ℃), crushed through a 1-mm screen, and well mixed, and then a subsample was obtained for chemical analysis.

Gut barrier biomarkers

Blood samples (10 mL) were collected by puncture of the ear veins and transferred in vacuum tubes (5 mL) and heparinized tubes (5 mL) for zonulin, lipopolysaccharide (LPS), and lipopolysaccharide-binding protein (LBP) analysis. In order to obtain serum, the samples were centrifuged at 3,000 rpm for 15 minutes at 4 ℃ in vacuum tubes. The serum samples were then kept at -80 ℃ for further analysis. Using an enzyme-linked immunosorbent assay technique, pigs serum and feces samples were examined for LPS, zonulin (Cat # MBS2607498, MyBioSource, San Diego, CA, US), and binding protein LBP

(Nanjing Jiancheng Bioengineering Institute, Nanjing, China). Fecal samples were used for lipocalin-2 determination.

Hair cortisol concentration

The concentrations of cortisol in the hair was determined as previously mentioned [14]. At days 90 and 112 of gestation, hair samples from the sows' foreheads were taken. Aluminum foil was used to collect and store the hair samples before they were dried in polypropylene tubes (HM Hyundai Micro Co., Korea). Samples were cleaned of contaminants by being washed three times in 5 mL of isopropyl alcohol and then allowed to dry for seven days at room temperature (24 ± ℃). According to the manufacturer's instructions, cortisol was extracted using a methanol dilution technique after drying (Cayman Chemical, Ann Arbor, MI).

Fecal SCFAs concentration

As stated by Jeong et al. [15], gas chromatography (GC) was used to measure the SCFA levels in fecal samples. The samples were divided using a TRACE 1310 GC equipped with a flame ionization detector, and they were examined on an HP-88 column (100-m length, 0.25-mm diameter, and 0.2-m film thickness from the producer). The temperature schedule was as follows: 70℃ for 1 minute, 180℃ maintained at 25℃ for 1 minute, 200℃ maintained at 10℃ for 1 minute, 220℃ maintained at 2℃ for 10 minutes, and finally 240℃ maintained at 20℃ for 6 minutes. The sample was run at a split ratio of 20:1 with a column flow rate of 1.3 mL/min. The use of hydrogen as a carrier gas is made. The use of hydrogen as a carrier gas was considered and the injector and detector were both set at 270℃ and 290℃, respectively.

Statistical analyzes

The MIXED procedure was applied to perform the statistical analysis (SAS Inst. Inc. Cary, NC), based on two 2 × 2 factorial arrangement of treatments as room temperature×dietary fiber level and dietary energy level vs dietary fiber level in randomised blocks. Initial BW was included as a covariate factor.

Using Turkey's Honestly Significant Difference technique, statistical differences between the treatments were defined as those with significant differences ($P < 0.05$). For all parameters, a single pig was regarded as the experimental unit.

Results

Pigs performance

Among standard energy level treatments (Table 2), heat-stressed pigs showed lower final BW, average daily gain (ADG), and ADFI in phase 1, phase 2, and overall. Moreover, the growth-to-feed ratio (G:F) was decreased in heat-stressed pigs in phase 1 and phase 2, however, the overall G:F was unaffected. Dietary fiber level showed no effects on the final BW, ADG, ADFI, and G:F during the experiment. There was a positive interaction for ADG between room temperature and dietary fiber.

Among heat stress treatments, dietary energy level did not affect final BW, ADG, ADFI, and G:F during the whole experiment. Pigs fed high dietary fiber showed no change in final BW, ADG, ADFI, and G:F in phase 1 and phase 2, however, the overall ADG was increased in pigs fed a high fiber. There were no interactive effects between room temperature and dietary fiber.

Nutrient digestibility

Among standard energy level treatments (Table 3), heat-stressed pigs showed lower digestibility of DM, GE, CP, crude fiber (CF), neutral detergent fiber (NDF), and acid detergent fiber (ADF) in phase 1 and phase 2. Dietary fiber level showed no effects on the digestibility of DM, GE, CP, CF, NDF, and ADG in phase 1 and phase 2. There was no interaction for nutrient digestibility parameters between room temperature and dietary fiber.

Among heat stress treatments, dietary energy level did not affect the digestibility of DM, GE, CP, CF, NDF, and ADF during the whole experiment. Pigs fed high dietary fiber showed no change in

digestibility of DM, GE, CP, CF, NDF, and ADF in phase 1 and phase 2. There were no interactive effects on nutrient digestibility between room temperature and dietary fiber.

Short-chain fatty acid production

Among standard energy level treatments (Table 4), heat-stressed pigs showed lower concentrations of acetate, butyrate, propionate, valerate, and total SCFA in feces in phase 1. Moreover, the concentration of acetate, propionate, butyrate, valerate, isovalerate, and total SCFA in feces was reduced in heat stressed-pigs in phase 2. Heat-stressed pigs showed a lower concentration of isobutyrate in feces in phase 2. The concentration of acetate, propionate, butyrate, and total SCFA in feces was increased in pigs fed high dietary fiber in phases 1 and 2. There was a negative interaction between dietary fiber and energy for the fecal concentration of isobutyrate in phase 1 and valerate in phase 2.

Among heat stress treatments, dietary energy level did not affect the concentration of acetate, propionate, butyrate, valerate, isovalerate, and total SCFA in feces during phases 1 and 2. Pigs fed high dietary fiber showed a greater concentration of acetate, propionate, butyrate, and total SCFA in phase 1. There were no interactive effects on nutrient digestibility between room temperature and dietary fiber. Moreover, the concentration of acetate, propionate, butyrate, valerate, isovalerate, and total SCFA were increased in feces of pigs fed high dietary fiber in phase 2. However, a lower concentration of isobutyrate was shown in pigs fed high dietary fiber in phase 2.

Stress indicators

Among standard energy level treatments, pigs in heat stress treatments showed a higher ($P < 0.05$) rectal temperature on wk 1 to 8 compared with pigs in TN groups (Figure 2). Among heat stress treatments, high fiber treatments showed a lower respiratory rate at wk 7 and 8. Among standard energy level treatments, pigs in heat stress treatments showed a higher ($P < 0.05$) respiratory rate on wk 1 to 8 compared with pigs in the TN groups (Figure 3). Among heat stress treatments, there was no change in

respiratory rate between the pigs fed diets with different energy levels. Moreover, high-fiber treatments showed a lower rectal temperature rate at wk 7 and 8. There was no change in rectal temperature between the pigs fed diets with different energy levels. Among standard energy level treatments, pigs in heat stress treatments showed a higher ($P < 0.05$) hair cortisol in phases 1 and 2 compared with pigs in the TN groups (Table 5), however, dietary fiber did not show any changes in hair cortisol. Among heat stress treatments, dietary fiber levels and energy levels showed no change in hair cortisol.

Gut permeability index, lipopolysaccharide, and lipopolysaccharide-binding protein

Among standard energy level treatments, pigs in heat stress treatments showed a higher ($P < 0.05$) plasma zonulin in phase 1, and a higher plasma zonulin and fecal lipocalin-2 in phase 2 compared with pigs in TN groups (Table 6). Moreover, dietary high fiber reduced plasma zonulin in phase 1. Among heat stress treatments, dietary energy level did not affect plasma zonulin and fecal lipocalin-2, however, high fiber treatments decreased plasma zonulin in phases 1 and 2. Among standard energy level treatments, pigs in heat stress treatments showed a higher ($P < 0.05$) plasma LPS and LBP in phases 1 and 2 compared with pigs in TN groups (Table 7). Moreover, dietary high fiber reduced plasma LBP in phases 1 and 2. Among heat stress treatments, dietary energy level did not affect plasma LPS and LBP, however, high fiber treatments decreased plasma LPS and LBP in phases 1.

Discussion

Chronic heat stress has been linked to decreased growth performance in growing pigs [3] via compromising gut health and feed consumption. According to the current study, prolonged heat stress markedly reduced ADFI, ADG, and G:F. Although the fiber level groups consumed the same amount of feed, the ADG was higher in pigs fed high-fiber diet than those of the low-fiber group, suggesting that the high temperature and reduced feed intake together are the reasons for the decline in growth performance. Decreased feed consumption has negative effects on pigs growth performance and meat

198 quality. Addressing low feed intake and poor nutrient absorption in heat-stressed pigs, the aim of the
199 current feeding strategy was to offer a high-energy concentrated meal dense in nutrients while limiting
200 the quantity of undigestible fibre. Along with financial losses imposed by performance declines, the
201 sow's health is also harmed. Increased medicine expenditures to treat the rise in inflammation and an
202 increase in mortality further reduce the performance of farms. The pigs benefit greatly from the
203 alternative strategy of providing significant amounts of fiber since the conventional method only
204 focuses on nutrient absorption impairment, but the fibre strategy optimizes intestinal integrity as well.
205 In contrast, it has been reported that the decrease of SF that leads to excessive fermentation is taken into
206 account when formulating recommendations for fiber supplementation under heat-stress circumstances
207 [7]. High levels of fiber-bound protein are also present in wheat bran and sugar beet pulp, which favors
208 unfavorable protein fermentation in the large intestine[6,16,17]. However, the result of our study
209 showed the positive role of fermentation on intestinal health, digestibility of nutrients, and growth
210 performance.

211 As a result, poor nutrient absorption can lower available nutrients to maintain body metabolism,
212 severely impacting growth performance. Energy levels and fiber inclusion did not affect the digestibility
213 of DM, CP, GE, CF, NDF, and ADF, however, the HS decreased the digestibility of DM, CP, GE, CF,
214 NDF, and ADF compared with non-stressed treatments. Hemicellulose and pectin form the soluble
215 portion of fibers, which can be fermented by gut bacteria [1,7]. As a result, the SF enhances gut health,
216 which facilitates nutrition absorption. The ISF component of fiber, on the other hand, is not affected by
217 fermentation and is excreted undigested, as evidenced by the fact that the digestibility of ADF was not
218 enhanced in heat-stressed pigs in the current experiment.

219 The fiber is used as substrates for SCFA production at the distal section of the colon other than dietary
220 starch or protein [2,18]. The major byproducts of fermentation in the large intestine include acetate,
221 propionate, and butyrate [7,15]. The quantity of SCFA synthesis depends on a number of variables,
222 including the environment, nutrition, and intestinal microbiota [3]. Dietary fiber in soluble form is
223 reportedly a more significant substrate for SCFA synthesis [1]. Although SCFA can be synthesized in

all parts of the gastrointestinal tract, it is mostly produced in the colon and cecum [18]. Dietary fiber may enclose carbohydrates, facilitate their transit to the large intestine, and finally enhance fermentation [1]. The immune system, lipid metabolism, digestive organs health, and carbohydrate balance are predicted to be SCFA's primary targets [7]. Short-chain fatty acids prevent histone deacetylation and have a beneficial function in body metabolism by binding to particular receptors in the gut, such as G-protein-coupled receptors 41 and 43 [2]. In our study productions of acetate, propionate, and butyrate were increased in heat-stressed pigs fed SF, however, the acetate production was relatively more regulated. In agreement, SF produces more acetate than butyrate [1]. Effect of fermentation and acetate synthesis in the colon plays a significant role in promoting intestinal development and health by lowering the vulnerability to LPS [1]. Additionally, SCFA production enhances barrier function and decreases intestinal permeability in epithelial cells, which is a crucial factor in modifying intestinal health, inflammation, and nutritional digestibility [3,9]. Soluble fiber fermentation can help to improve gut health by reducing constipation due to excellent water-holding capacity [19]. In addition, starch-based carbohydrates can escape enzymatic digestion by being encapsulated in fiber structures, which indeed supply additional substrates for fermentation in distal regions of the intestine [12]. Our results confirm the necessity of dietary SF as fermentation substrates for SCFA production.

During a stressful condition, the release of cortisol is a common physiological response [20]. According to the findings of our study, an increase in hair cortisol is related to an elevation of room temperature. Fast-growing animals have a substantial reaction to cortisol release under heat stress, due to considerable tissue formation, weight gain, and immunological change [21,22]. Because of the long-term accumulation of cortisol in the hair, earlier studies demonstrated that hair cortisol is a better biomarker of chronic heat stress than blood cortisol [23]. The relationship between chronic stress and hair cortisol has been demonstrated in cattle [24] and sheep [25,26]. The mechanism of action behind the role of SF level on cortisol secretion in growing pigs has not been validated, however, satiety may be associated with decreased stress.

Heat stress impacts the physiological status of animals on several levels: in vitro data for cell cultures showed a heat stress-related lowering of the trans-epithelial electric resistance, which evaluates the integrity of the intestinal barrier in growing pigs. High-fiber treatments significantly reduced zonulin secretion. An increased zonulin concentration indicates a leaky gut, and any decrease in zonulin release is related to intestinal health at the mucosa level [1]. The intestinal tight junction injury was observed following the exposure of pigs to heat stress, which was demonstrated by the rise in zonulin and LPS in the serum. This finding is noteworthy since various authors have shown that pathogenic bacterial translocation associated with the onset of systemic inflammation may be mediated by a loss of intestinal epithelial barrier integrity [9,27,28]. The observed villus integrity may be explained in part by changes in the fermentation order, butyric acid synthesis, LPS penetration, plasma LBP, and zonulin concentration. The poorer the intestinal barrier, the quicker microorganisms can enter the body, causing infections and inflammatory processes as an immunological response. The more energy a pig must spend on immunological reactions, the less energy is available for growth.

Conclusion

In conclusion, based on the growth performance results, our study showed that the supplementation of SF in the pig diet is necessary during heat stress. The improvement in the production of SCFA and the reduction of zonulin, LPS, and LBP release into the blood emphasize antimicrobial activity and intestinal health. These beneficial alterations in gut health appeared in lower stress and cortisol deposition in hair. This finding may offer further insight into the importance of reducing the adverse effects of heat stress during growing stages and the function of SF as a regulator of intestinal integrity.

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361 Table 1. Experimental diets.

Body weight	50-75				75-100			
ME	Standard		High		Standard		High	
Fiber	Low	High	Low	High	Low	High	Low	High
Corn	70.93	66.68	68.2	63.95	74.84	70.59	72.11	67.86
Wheat	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Oil	1.71	2.03	4.13	4.45	1.60	1.92	4.02	4.34
SBM (44%)	19.38	19.42	19.70	19.74	15.86	15.90	16.18	16.22
Sugar beet pulp	-	4.00	-	4.00	-	4.00	-	4.00
DL-Met (98%)	0.03	0.04	0.03	0.04	0.02	0.03	0.02	0.03
L-Lys (78.8%)	0.22	0.20	0.21	0.19	0.21	0.19	0.20	0.18
Thr (99%)	0.03	0.02	0.03	0.02	0.01	-	0.01	-
Limestone	0.72	0.62	0.72	0.62	0.67	0.57	0.67	0.57
DCP	1.22	1.23	1.22	1.23	1.03	1.04	1.03	1.04
Salt	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Choline chloride	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Vitamine	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Mineral	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
$NaCO_3$	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16
Phytase	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Sum	100	100	100	100	100	100	100	100
ME	3300	3300	3450	3450	3300	3300	3450	3450
CP	15.2	15.2	15.2	15.2	14.1	14.1	14.1	14.1
Ca	0.62	0.62	0.62	0.62	0.55	0.55	0.55	0.55
P	0.30	0.30	0.30	0.30	0.26	0.26	0.26	0.26
Lys	0.90	0.90	0.90	0.90	0.80	0.80	0.80	0.80
Met	0.26	0.27	0.27	0.28	0.24	0.24	0.24	0.24
Met+Cys	0.51	0.51	0.51	0.51	0.48	0.48	0.48	0.48
Thr	0.56	0.56	0.56	0.56	0.56	0.56	0.56	0.56
Trp	0.17	0.17	0.17	0.17	0.30	0.30	0.16	0.16

362 DCP, di-calcium phosphate; ME, metabolizable energy; CP, crude protein.

363 [1]Supplied per kilogram of diet: 16,000 IU vitamin A, 3,000 IU vitamin D3, 5.0 mg vitamin B1, 20 mg vitamin B2, 4 mg
364 vitamin B6, 0.08 mg vitamin B12, 40 IU vitamin E, 5.0 mg vitamin K3, 75 mg niacin, 40 mg pantothenic acid, 0.15 mg
365 biotin, 0.65 mg folic acid.

366 [2] Supplied per kilogram of diet: 0.25 mg Co, 50 mg Cu, 15 mg Mn, 25 mg Zn, 45 mg Fe, 0.35 mg I, 0.13 mg Se.

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Table 2. Effect of energy level and dietary beet pulp on growth performance of growing-finishing pigs under heat stress conditions.

Temperature (T)	TN		HS				SEM	p-value					
ME, kcal/kg (E)	3,300		3,300		3,450			3,300			HS		
Fiber level (F)	Low	High	Low	High	Low	High		T	F	T×F	E	F	E×F
BW, kg													
Initial	56.43	57.27	56.80	56.48	56.44	56.59	0.38	0.428	0.526	0.263	0.730	0.778	0.532
Final	116.3	116.6	99.4	101.9	100.5	102.7	1.31	<0.001	0.333	0.243	0.438	0.069	0.905
Phase 1 (d 0~28)													
ADG, g	1,169	1,122	767	808	775	833	28.97	<0.001	0.895	0.048	0.568	0.410	0.751
ADFI, g	2,223	2,189	1,755	1,767	1,739	1,712	61.68	<0.001	0.728	0.489	0.635	0.916	0.796
G:F	0.526	0.513	0.437	0.457	0.448	0.501	0.02	<0.001	0.777	0.147	0.272	0.154	0.493
Phase 2 (d 29~56)													
ADG, g	968	975	754	819	800	816	37.9	<0.001	0.349	0.452	0.477	0.186	0.413
ADFI, g	3,295	3,213	2,331	2,317	2,344	2,221	126.3	<0.001	0.657	0.756	0.770	0.628	0.701
G:F	0.294	0.303	0.323	0.353	0.351	0.388	0.03	0.015	0.335	0.685	0.405	0.306	0.824
Overall (d 0~56)													
ADG, g	1,069	1,049	761	814	788	825	22.6	<0.001	0.437	0.081	0.364	0.049	0.734
ADFI, g	2,759	2,701	2,043	2,042	2,041	1,967	64.28	<0.001	0.576	0.592	0.598	0.601	0.614
G:F	0.387	0.388	0.372	0.398	0.389	0.423	0.01	0.912	0.208	0.244	0.197	0.080	0.797

TN: thermoneutral, 21~24℃; HS: heat stress, 30~34℃; SEM: standard error of mean; ME: metabolizable Energy; BW: body weight; ADG, average daily gain; ADFI: average daily feed intake; G:F, gain to feed intake ratio

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Table 3. Effect of energy level and dietary beet pulp on nutrient digestibility of growing-finishing pigs under heat stress conditions.

Temperature (T)	TN		HS				SEM	p-value					
ME, kcal/kg (E)	3,300		3,300		3,450			3,300			HS		
Fiber level (F)	Low	High	Low	High	Low	High		T	F	T×F	E	F	E×F
Phase 1 (d 0~28), %													
DM	86.62	86.19	82.88	82.47	82.64	82.98	0.61	<0.001	0.290	0.982	0.604	0.824	0.214
GE	90.07	91.26	86.69	87.10	86.07	87.49	0.73	<0.001	0.154	0.594	0.838	0.111	0.359
CP	88.56	88.53	83.72	84.49	83.62	84.40	0.97	<0.001	0.964	0.231	0.869	0.205	0.996
CF	56.39	54.52	49.89	51.90	49.31	50.01	1.89	0.005	0.957	0.170	0.383	0.340	0.638
NDF	54.73	54.69	48.76	50.20	47.19	49.24	1.82	0.001	0.591	0.572	0.352	0.207	0.821
ADF	48.48	47.25	40.14	42.28	39.49	40.55	1.52	<0.001	0.119	0.654	0.337	0.202	0.658
Phase 2 (d 29~56), , %													
DM	86.00	85.52	82.91	83.47	82.64	82.98	0.64	<0.001	0.917	0.181	0.324	0.255	0.764
GE	90.45	90.79	85.32	86.18	84.97	85.53	0.80	<0.001	0.253	0.420	0.451	0.288	0.818
CP	88.4	88.45	83.02	83.63	83.25	84.11	1.12	<0.001	0.768	0.634	0.661	0.365	0.880
CF	55.76	55.61	52.50	51.29	50.47	52.47	1.98	<0.001	0.596	0.679	0.760	0.775	0.260
NDF	52.15	53.06	48.28	49.44	48.34	49.59	1.17	<0.001	0.272	0.896	0.900	0.169	0.958
ADF	49.07	50.20	44.46	45.50	45.94	46.13	1.55	<0.001	0.397	0.968	0.341	0.571	0.694

TN: thermoneutral, 21~24°C; HS: heat stress, 30~34°C; SEM: standard error of mean; ME: metabolizable energy; DM, dry matter, GE, gross energy, CP, crude protein, CF, crude fiber, NDF, neutral detergent fiber; ADF, acid detergent fiber.

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Table 4. Effect of energy level and dietary beet pulp on short-chain fatty acid of growing-finishing pigs under heat stress conditions (μmol/g).

Temperature (T)	TN		HS				SEM	p-value					
ME, kcal/kg (E)	3,300		3,300		3,450			3,300			HS		
Fiber level (F)	Low	High	Low	High	Low	High		T	F	T×F	E	F	E×F
Phase 1 (d 0~28), μmol/g													
Acetate	68.89	75.68	55.69	60.21	53.9	59.54	0.93	<0.001	0.027	0.622	0.514	0.011	0.763
Propionate	13.91	15.11	11.67	12.47	11.47	13.07	0.28	<0.001	0.041	0.665	0.722	0.041	0.489
Butyrate	7.62	9.14	5.87	6.67	6.00	6.73	0.16	<0.001	<0.001	0.099	0.775	0.026	0.918
Isobutyrate	3.40	2.35	3.30	3.41	3.15	3.09	0.14	0.106	0.109	0.045	0.420	0.922	0.783
Valerate	3.26	3.08	2.92	2.85	2.81	2.84	0.07	0.038	0.331	0.664	0.713	0.883	0.726
Isovalerate	4.77	4.89	4.63	4.14	4.25	4.21	0.12	0.117	0.491	0.259	0.540	0.293	0.379
Total SCFA	104.4	112.36	85.07	92.18	82.65	89.48	1.42	<0.001	0.023	0.607	0.375	0.021	0.960
Phase 2 (d 29~56), , μmol/g													
Acetate	80.38	84.54	60.76	63.36	60.09	63.73	0.64	<0.001	0.020	0.548	0.908	0.022	0.686
Propionate	15.26	16.96	12.19	13.74	12.67	13.67	0.27	<0.001	0.008	0.887	0.704	0.027	0.622
Butyrate	8.75	10.37	6.04	7.08	6.02	7.10	0.22	<0.001	0.004	0.454	0.996	0.027	0.973
Isobutyrate	2.57	2.57	3.62	3.55	3.61	3.51	0.01	<0.001	0.457	0.485	0.411	0.006	0.728
Valerate	3.72	3.48	2.99	3.25	2.95	3.13	0.04	<0.001	0.920	0.013	0.422	0.034	0.687
Isovalerate	6.10	5.49	4.26	4.54	4.23	4.77	0.09	<0.001	0.465	0.061	0.616	0.045	0.532
Total SCFA	118.1	124.6	89.91	95.69	89.88	96.32	0.91	<0.001	0.018	0.465	0.948	0.026	0.572

TN: Thermoneutral, 21~24℃; HS: Heat stress, 30~34℃; SEM: Standard error of mean; ME: Metabolizable Energy; SCFA: Short-chain fatty acid.

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Table 5. Effect of energy level and dietary beet pulp on hair cortisol levels of growing-finishing pigs under heat stress conditions.

Temperature (T)	TN		HS				SEM	p-value					
ME, kcal/kg (E)	3,300		3,300		3,450			3,300			HS		
Fiber level (F)	Low	High	Low	High	Low	High		T	F	T×F	E	F	E×F
Phase 1 (d 0~28)													
Cortisol, pg/mL	14.09	13.37	22.23	22.73	22.39	20.17	0.68	<0.001	0.940	0.674	0.384	0.534	0.327
Phase 2 (d 29~56)													
Cortisol, pg/mL	20.53	21.26	29.04	27.69	29.63	26.59	0.60	<0.001	0.806	0.406	0.833	0.079	0.487

TN: Thermoneutral, 21~24℃; HS: Heat stress, 30~34℃; SEM: Standard error of mean; ME: Metabolizable Energy.

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Table 6. Effect of energy level and dietary beet pulp on gut permeability index of growing-finishing pigs under heat stress conditions.

Temperature (T)	TN		HS				SEM	p-value					
ME, kcal/kg (E)	3,300		3,300		3,450			3,300			HS		
Fiber level (F)	Low	High	Low	High	Low	High		T	F	T×F	E	F	E×F
Phase 1 (d 0~28)													
Plasma zonulin, pg/mL	246.9	219.8	281.0	251.4	285.8	258.6	4.02	<0.001	<0.001	0.868	0.464	0.010	0.884
Fecal lipocalin-2, μg/g	24.38	23.15	25.73	23.63	24.75	22.92	0.55	0.318	0.074	0.633	0.454	0.086	0.904
Phase 2 (d 29~56)													
Plasma zonulin, pg/mL	279.9	267.1	316.5	296.5	313.5	292.1	4.17	<0.001	0.056	0.668	0.659	0.020	0.930
Fecal lipocalin-2, μg/g	25.67	24.78	29.69	27.23	29.58	27.77	0.52	<0.001	0.062	0.369	0.837	0.053	0.761

TN: Thermoneutral, 21~24℃; HS: Heat stress, 30~34℃; SEM: standard error of mean; ME: Metabolizable Energy.

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Table 7. Effect of energy level and dietary beet pulp on intestinal LPS and LBP levels of growing-finishing pigs under heat stress conditions.

Temperature (T)	TN		HS				SEM	p-value					
ME, kcal/kg (E)	3,300		3,300		3,450			3,300			HS		
Fiber level (F)	Low	High	Low	High	Low	High		T	F	T×F	E	F	E×F
Phase 1 (d 0~28)													
LPS, ng/mL	22.50	19.69	30.07	27.85	30.85	27.25	0.83	<0.001	0.082	0.836	0.957	0.009	0.684
LBP, ng/mL	23.10	20.99	31.22	27.95	31.58	27.43	0.72	<0.001	0.013	0.570	0.958	0.016	0.764
Phase 2 (d 29~56)													
LPS, ng/mL	25.67	26.25	33.97	31.98	33.81	31.17	0.61	<0.001	0.599	0.343	0.696	0.071	0.794
LBP, ng/mL	25.85	23.35	34.82	32.24	34.24	32.98	0.53	<0.001	0.004	0.967	0.942	0.086	0.549

TN: thermoneutral, 21~24℃; HS: heat stress, 30~34℃; SEM: standard error of mean; ME: metabolizable energy; LPS: lipopolysaccharide; LBP: lipopolysaccharide-binding protein.

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398 **Figure legends**

399 **Figure 1.** Ambient temperature and temperature-humidity index (THI) during the experimental
400 period. TN, thermoneutral; HS, heat stress.

401 **Figure 2**. Effect of fiber inclusion and energy level on respiratory rate of growing-finishing pigs
402 under heat stress conditions. TN, thermoneutral; HS, heat stress.

403 **Figure 3**. Effect of fiber inclusion and energy level on the rectal temperature of growing-finishing
404 pigs under heat stress conditions. TN, thermoneutral; HS, heat stress.

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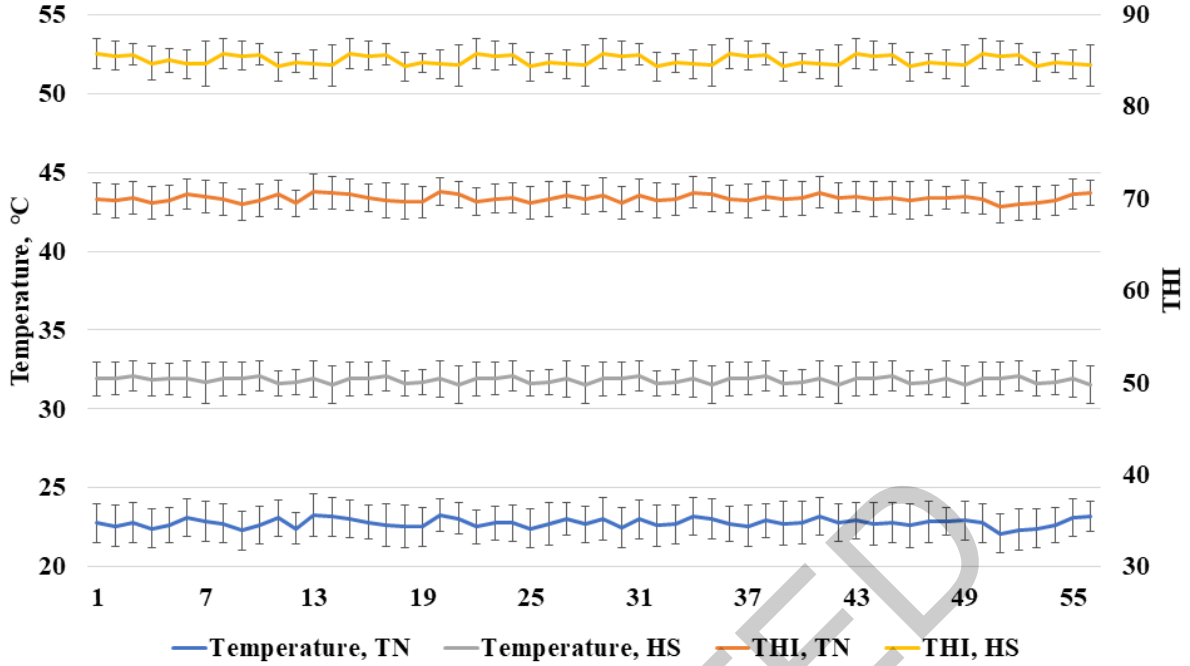

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407 **Figure 1.** Ambient temperature and temperature-humidity index (THI) during the experimental

408 period. TN, thermoneutral (21-24 ºC); HS, heat stress (30-34 ºC).

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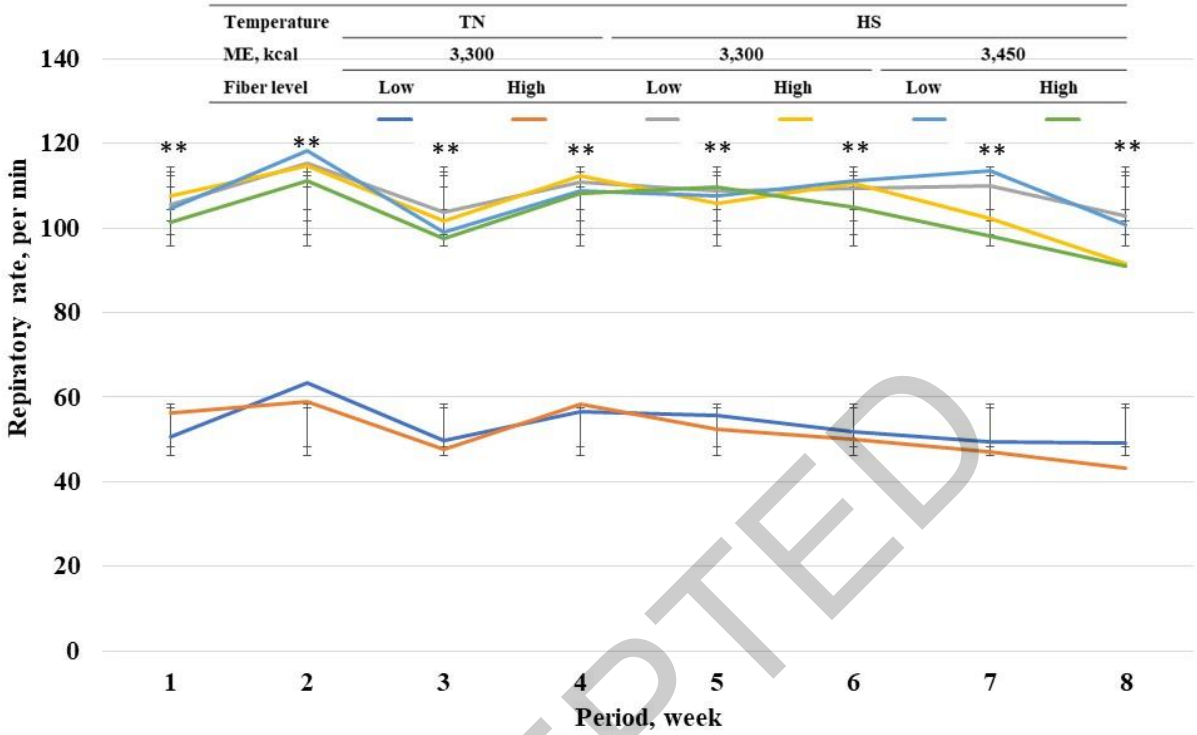

Figure 2. Effect of fiber inclusion and energy level on respiratory rate of growing-finishing pigs under heat stress conditions. TN, thermoneutral (21-24 ºC); HS, heat stress (30-34 ºC).

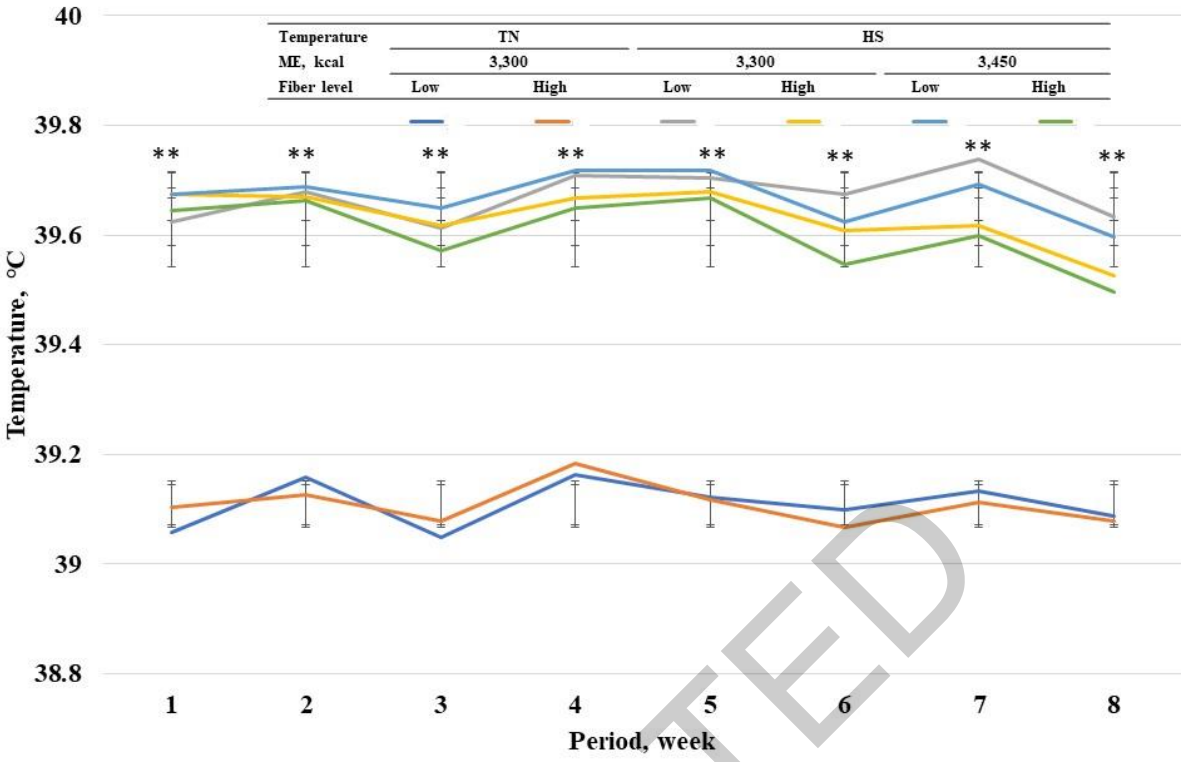

Figure 3. Effect of fiber inclusion and energy level on the rectal temperature of growing-finishing pigs under heat stress conditions. TN, thermoneutral (21-24 ºC); HS, heat stress (30-34 ºC).