

## AGED GARLIC EXTRACT AND ALICIN IMPROVE PERFORMANCE AND GASTROINTESTINAL TRACT DEVELOPMENT OF PIGLETS REARED IN ARTIFICIAL SOW

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**Abstract:** This study was performed to investigate whether postnatal administration with aged garlic extract (AGE) and allicin influences performance and systemic development of piglets exposed to early weaning. Twenty-four male piglets were weaned from sows at the age of two days of life, divided into 4 weight-matched groups and kept under conditions of artificial sow for 6 days. The first group consisted of control animals, while piglets that were given AGE daily per os at the dosages of 1 ml and 2 ml/kg body weight, respectively belonged to the second and third group. The fourth group consisted of piglets administered orally with allicin at the dosage of 1.0 mg/kg body weight/day. At the age of 8 days of life all animals were sacrificed. Next to body weight gain and morphological properties of the gastrointestinal tract, the haematological examination was performed, and activity of lysozyme and ceruloplasmin as well as level of gamma-globulins were determined. The obtained results showed that AGE and allicin improved final body weight, morphological properties of intestine villi and non-specific defence mechanisms of pigs. All these results indicate that AGE and allicin induced beneficial effects on health status, performance and systemic development of piglets.

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### INTRODUCTION

Proper function of the gastrointestinal tract in piglets plays a very important role for their adaptation to new environmental conditions present just after the birth and after weaning. After weaning, piglets are exposed to many stressful factors the most important among which are separation from mother, new environmental conditions and changes of diet. All these factors lead to systemic growth

inhibition induced by strongly reduced food consumption and negative changes in the intestine structure [16]. Next to the histological changes expressed as atrophy of the intestinal villi, the decrease of biochemical processes taking place on the intestinal level induces impaired digestion and absorption and may lead to diarrhea. Furthermore, rapid changes of the intestine microflora consequential to the weaning were reported as causative factors responsible for all these negative changes [7, 8, 13, 16, 17, 18].

The antibacterial activity of garlic (*Allium sativum*) has been widely investigated and is well documented. Louis Pasteur was the first to describe the antibacterial effects of onion and garlic juices. Historically, garlic has been used worldwide to fight bacterial infections and showed a broad antibiotic spectrum against both gram-positive and gram-negative bacteria. Raw juice of garlic was found to be effective against many common pathogenic intestinal bacteria, which are responsible for diarrhea in humans and animals. Garlic was reported to be effective even against those strains that have become resistant to antibiotics and the combination of garlic with antibiotics leads to partial or total synergism. Furthermore, lack of bacterial resistance to garlic has been observed [19]. Our previous studies in pigs have shown that maternal treatment with garlic preparations during the last 24 days of pregnancy and 4 weeks of lactation induced immunostimulating effects in both sows and piglets, when humoral non-specific and specific defence mechanisms were investigated [5, 22]. These effects were confirmed by the response of white blood cells of piglets born by sows treated with aged garlic extract (AGE) and allicin [4]. Moreover, these studies showed that maternal administration with AGE and allicin regulates the systemic development of piglets during intrauterine life and early stages of postnatal life [20, 21]. All these effects may be beneficial for improvement of the developmental process and health status of pigs during weaning. Even though the effects of maternal administration of AGE and allicin on the immunological status and systemic development of piglets have been well documented, no data are available on the effects of these substances administered to piglets during postnatal life. Thus, the aim of the study was to investigate effects of postnatal administration with AGE and allicin on the performance and development of the gastrointestinal tract of piglets exposed to the early weaning from sows, starting 48 hours after the birth, and kept under artificial sow conditions.

## MATERIAL AND METHODS

The experimental procedures used throughout this study were approved by the Local Ethics Committee on Animal Experimentation of the Agricultural University of Lublin, Poland.

**Experimental design and sampling procedure.** The study was performed on 24 male piglets born by 6 sows of the Large Polish White breed that were weaned 48 hours after the birth and divided into 4 weight-matched groups. To avoid litter-dependent effects connected with various genotype and intrauterine conditions, 4 male piglets with the most similar body weight were obtained from each sow and ascribed to 4 different groups [3, 9]. After weaning, the piglets were kept under conditions of artificial sow; they were housed in a special pen equipped with 2 heaters providing optimal temperature and bottle-fed commercially

**Table 1.** Composition of diet administered to piglets during the study\*.

Ingredient	Value
Metabolic energy (MJ)	17.70
Crude protein (%)	20.00
Crude fat (%)	18.00
Crude fiber (%)	0.67
Lactose (%)	38.00
L-Lysine-HCl (%)	1.41
DL-Methionine (%)	0.87
L-Threonine (%)	0.92
L-Tryptophan (%)	0.29
Calcium (%)	1.00
Phosphorus (%)	0.89
Sodium (%)	0.40

\*Milk-replacement feed was administered to piglets as the water solution prepared in the weight ratio of 1:7 and temperature of 40°C.

available standard milk-replacement feed (Lakti R, Trouw Nutrition, Poland) every 4 h throughout 6 days (Tab. 1). The first group ( $n = 6$ ) consisted of control animals (CON group), while piglets that were given aged garlic extract (AGE) daily per os at the dosages of 1 ml and 2 ml/kg body weight, respectively, belonged to the second (AGE 1 group;  $n = 6$ ) and the third (AGE 2 group;  $n = 6$ ) groups. The fourth group (ALL group;  $n = 6$ ) consisted of piglets administered orally with allicin (Alliomax, Herbapol Lublin, Poland) at the dosage of 1.0 mg/kg body weight. The vehiculum (soybean oil) in the controls, as well as AGE and allicin in the experimental groups, were administered every 24 hours, starting on the day of weaning. All the piglets were weighed every day to determine body weight gain. Blood samples were collected at the age of 8 days of life and analyzed immediately with the use of automatic haematological analyzer MS9 (Melet Schloesing Laboratories, France). The lysozyme activity in the serum was assessed with the turbidimetric method using lysis of *Micrococcus lysodeicticus*. The serum ceruloplasmin activity was determined with the use of spectrophotometric method. The total protein level and gamma-globulin concentration was determined as described previously [5, 22]. The animals were sacrificed at the age of 8 days of life and their gastrointestinal tracts were isolated for analysis. In each piglet, the weight of empty stomach, liver and pancreas was taken, the mesentery from the intestine was removed gently and the length of the duodenum and total small intestine was measured. In the next step, transverse sections of the duodenum, jejunum (25%, 50%, and 75% of total small intestine length) and ileum, 1 cm in length, were collected similarly for all samples for histological analysis.

**Table 2.** Body weight (g) of piglets kept under conditions of artificial sow from 2<sup>nd</sup>–8<sup>th</sup> day of life from control group (CON) and groups treated with aged garlic extract (AGE 1, AGE 2) and allicin (ALL).

Group	2 days old (Weaning)	3 days old	4 days old	5 days old	6 days old	7 days old	8 days old
CON	1700 ± 121	1644 ± 137	1527 ± 155	1756 ± 134	1761 ± 134	1871 ± 141	1952 ± 144
AGE 1	1699 <sup>a</sup> ± 73	1638 <sup>a</sup> ± 60	1699 <sup>a</sup> ± 66	1832 <sup>ab</sup> ± 75	1897 <sup>ab</sup> ± 54	1937 <sup>ab</sup> ± 56	2019 <sup>b</sup> ± 56
AGE 2	1703 <sup>ab</sup> ± 125	1653 <sup>a</sup> ± 113	1774 <sup>ac</sup> ± 115	1834 <sup>ac</sup> ± 109	1916 <sup>ac</sup> ± 113	1995 <sup>bc</sup> ± 102	2056 <sup>c</sup> ± 107
ALL	1690 <sup>ab</sup> ± 88	1651 <sup>a</sup> ± 84	1741 <sup>ac</sup> ± 108	1833 <sup>ac</sup> ± 107	1900 <sup>ac</sup> ± 117	1978 <sup>bc</sup> ± 113	2031 <sup>c</sup> ± 101

<sup>a, b, c</sup>Values within rows that do not share common superscripts differ significantly for  $p \leq 0.05$ .

**Morphological examination.** The intestine samples were immediately fixed in fresh 4% buffered formaldehyde, dehydrated in growing concentration of ethyl alcohol, cleared in xylene, and embedded in paraplast (Sigma-Aldrich). Serial sections were cut at 4.5 µm, stained with haematoxylin and eosin and closed with the use of DPX (Sigma-Aldrich). The samples were examined using light and confocal (AXIOVERT 200 M equipped with an LSM 5 Pascal laser scanning head, Zeiss) microscopes. Simultaneous collection of fluorescent and Nomarsky contrast images was performed using an argon laser, with wave length of 514 nm. Morphometric measurements of villus height (VH), villus width at villus base (VW), villus section area (VSA), crypt (gland) depth (CD), and mucosa thickness (MT) were performed automatically under confocal microscope using LSM 5 Image Examiner software. Moreover, villus number per cm (VN) of cross section of each investigated intestine sample was estimated.

**Statistical analysis.** Statistical analysis was performed using Statistica software (version 6.0). All the data were presented as means ± SEM. All the examined parameters were found to be normally distributed in accordance with Kolmogorov-Smirnov test. Body weights of piglets measured at each day of the experiment were analyzed by ANOVA with repeated measures and *post-hoc* Tukey test. Differences

between all other mean values were tested using one-way ANOVA and *post-hoc* Duncan test. Differences showing  $p \leq 0.05$  were considered statistically significant.

## RESULTS

Body weight values of the control piglets did not change significantly during the whole period of the experiment, which was in contrast to the results obtained in all the experimental groups. The final body weight was significantly increased in piglets treated with both dosages of AGE and allicin when compared to the values measured at the weaning ( $p \leq 0.05$ ; Tab. 2).

Weight of the internal organs did not differ significantly between investigated groups, except for weight of stomach and liver in AGE 2 and AGE 1 groups, respectively. AGE and allicin administration did not change the length of the duodenum and the small intestine. The ratio of stomach to body weight in the ALL group was significantly decreased when compared to the AGE 2 group. Furthermore, decreased liver to body weight ratio was noted in AGE 2 group, when compared to all the other groups of the experiment (Tab. 3).

The results of morphological examination in several segments of small intestine of the piglets are shown in Table 4. Aged garlic extract (AGE) administration to piglets reared

**Table 3.** Weight of internal organs and length of duodenum and small intestine of piglets at age of 8 days of life from control group (CON) and groups treated with aged garlic extract (AGE 1, AGE 2) and allicin (ALL).

Investigated parameter	CON	AGE 1	AGE 2	ALL
Weight of stomach (g)	12.18 <sup>a</sup> ± 0.77	12.18 <sup>a</sup> ± 0.48	14.5 <sup>b</sup> ± 0.97	11.6 <sup>a</sup> ± 0.64
Weight of liver (g)	62.9 <sup>a</sup> ± 0.95	74.4 <sup>b</sup> ± 4.13	61.4 <sup>a</sup> ± 2.41	71.0 <sup>ab</sup> ± 3.24
Weight of pancreas (g)	2.51 ± 0.36	2.23 ± 0.16	2.65 ± 0.26	2.80 ± 0.23
Length of duodenum (cm)	15.8 ± 1.36	16.8 ± 0.83	18.0 ± 0.85	17.8 ± 0.87
Length of small intestine (cm)	418 ± 21.70	459 ± 9.25	447 ± 10.13	446 ± 10.19
Ratio of stomach to body weight	0.00629 <sup>abc</sup> ± 0.000125	0.00625 <sup>abc</sup> ± 0.000239	0.00712 <sup>b</sup> ± 0.000436	0.00580 <sup>c</sup> ± 0.000578
Ratio of liver to body weight	0.0327 <sup>a</sup> ± 0.0017	0.0370 <sup>a</sup> ± 0.0015	0.0298 <sup>b</sup> ± 0.0014	0.0355 <sup>a</sup> ± 0.0025
Ratio of pancreas to body weight	0.00181 ± 0.000125	0.00202 ± 0.000109	0.00179 ± 0.000147	0.00190 ± 0.000200

<sup>a, b, c</sup>Values within rows that do not share common superscripts differ significantly for  $p \leq 0.05$ .

**Table 4.** Morphometric measurements of villus height (VH), villus width (VW), villus section area (VSA), crypt depth (CD), mucosa thickness (MT) and villus number per cm (VN) of the small intestine in the piglets at age of 8 days of life from control group (CON) and groups treated with aged garlic extract (AGE 1, AGE 2) and allicin (ALL).

Investigated parameter	CON group	AGE 1 group	AGE 2 group	ALL group
Duodenum				
VH (μm)	422 <sup>a</sup> ± 24	930 <sup>b</sup> ± 73	590 <sup>c</sup> ± 19	490 <sup>ac</sup> ± 21
VW (μm)	90 <sup>a</sup> ± 4	93 <sup>a</sup> ± 8	127 <sup>b</sup> ± 9	90 <sup>a</sup> ± 8
VSA (μm <sup>2</sup> )	34782 <sup>a</sup> ± 3661	64336 <sup>b</sup> ± 2436	39651 <sup>a</sup> ± 3432	43277 <sup>a</sup> ± 4374
CD (μm)	163 <sup>ab</sup> ± 7	148 <sup>a</sup> ± 10	178 <sup>b</sup> ± 12	116 <sup>c</sup> ± 7
MT	656 <sup>a</sup> ± 39	986 <sup>b</sup> ± 62	825 <sup>c</sup> ± 23	595 <sup>ad</sup> ± 17
VN	108 <sup>ab</sup> ± 12	97 <sup>a</sup> ± 8	89 <sup>a</sup> ± 9	131 <sup>b</sup> ± 8
25% of small intestine				
VH (μm)	358 <sup>a</sup> ± 15	588 <sup>bc</sup> ± 57	511 <sup>c</sup> ± 10	652 <sup>b</sup> ± 26
VW (μm)	69 <sup>a</sup> ± 4	89 <sup>b</sup> ± 7	110 <sup>c</sup> ± 7	108 <sup>c</sup> ± 4
VSA (μm <sup>2</sup> )	26564 <sup>a</sup> ± 1709	47828 <sup>b</sup> ± 4324	37196 <sup>c</sup> ± 2440	52906 <sup>a</sup> ± 4548
CD (μm)	108 ± 4	117 ± 3	115 ± 8	123 ± 12
MT	512 <sup>a</sup> ± 17	720 <sup>b</sup> ± 63	702 <sup>b</sup> ± 36	774 <sup>b</sup> ± 21
VN	100 ± 7	104 ± 3	89 ± 5	104 ± 6
50% of small intestine				
VH (μm)	385 <sup>a</sup> ± 28	762 <sup>b</sup> ± 27	566 <sup>c</sup> ± 19	951 <sup>d</sup> ± 49
VW (μm)	78 <sup>a</sup> ± 7	101 <sup>b</sup> ± 4	124 <sup>c</sup> ± 4	67 <sup>ad</sup> ± 5
VSA (μm <sup>2</sup> )	35424 <sup>a</sup> ± 4681	63021 <sup>b</sup> ± 2469	52983 <sup>b</sup> ± 3667	54828 <sup>b</sup> ± 5409
CD (μm)	98 <sup>a</sup> ± 5	129 <sup>b</sup> ± 10	147 <sup>b</sup> ± 8	90 <sup>a</sup> ± 7
MT	541 <sup>a</sup> ± 40	846 <sup>b</sup> ± 24	731 <sup>c</sup> ± 29	1092 <sup>d</sup> ± 42
VN	118 ± 10	95 ± 10	97 ± 6	119 ± 14
75% of small intestine				
VH (μm)	344 <sup>a</sup> ± 14	675 <sup>b</sup> ± 20	614 <sup>b</sup> ± 42	637 <sup>b</sup> ± 87
VW (μm)	78 <sup>a</sup> ± 3	90 <sup>a</sup> ± 7	111 <sup>b</sup> ± 4	63 <sup>c</sup> ± 6
VSA (μm <sup>2</sup> )	23967 <sup>a</sup> ± 764	54897 <sup>b</sup> ± 3654	54841 <sup>b</sup> ± 5340	42911 <sup>c</sup> ± 2563
CD (μm)	111 <sup>a</sup> ± 3	84 <sup>b</sup> ± 5	129 <sup>c</sup> ± 4	81 <sup>bd</sup> ± 6
MT	475 <sup>a</sup> ± 15	844 <sup>b</sup> ± 23	759 <sup>b</sup> ± 30	754 <sup>b</sup> ± 84
VN	113 <sup>a</sup> ± 11	103 <sup>a</sup> ± 5	108 <sup>a</sup> ± 6	153 <sup>b</sup> ± 21
Ileum				
VH (μm)	343 <sup>a</sup> ± 10	462 <sup>b</sup> ± 18	459 <sup>b</sup> ± 44	407 <sup>ab</sup> ± 32
VW (μm)	133 <sup>a</sup> ± 9	121 <sup>a</sup> ± 8	128 <sup>a</sup> ± 7	74 <sup>b</sup> ± 4
VSA (μm <sup>2</sup> )	33408 ± 2108	40551 ± 2815	43942 ± 6055	39425 ± 4332
CD (μm)	128 <sup>a</sup> ± 12	124 <sup>a</sup> ± 5	163 <sup>b</sup> ± 11	127 <sup>a</sup> ± 7
MT	554 ± 14	626 ± 27	659 ± 52	638 ± 42
VN	89 ± 6	92 ± 5	78 ± 5	93 ± 4

<sup>a, b, c</sup> Values within rows that do not share common superscripts differ significantly for p < 0.05.

under conditions of artificial sow for 6 days increased villus height (VH) in all the investigated segments of the small intestine when compared to the control group. Except for the ileum, a similar effect was observed in the small intestine of piglets that were given allicin. The villus width (VW) was increased in piglets receiving 1 ml/kg of body weight per day of AGE in 25% and 50% of the length of small intestine, while the double dosage of AGE increased this parameter in all the investigated segments except for the ileum. The piglets from the ALL group reached higher

values of VW in 25% of the small intestine length, when compared to the controls. Villus section area (VSA) of the duodenum was significantly increased in the AGE 1 group of piglets, when compared to the control group. However, the analysis of VSA showed its higher values in the segments of 25%, 50% and 75% of the small intestine length in piglets treated with both doses of AGE and allicin. The measurement of crypt depth (CD) in the duodenum and 75% of the small intestine length of piglets from AGE 2 group showed significant increase of this parameter when

**Table 5.** Selected haematological and immunological parameters assessed in serum of piglets at age of 8 days of life from control group (CON) and groups treated with aged garlic extract (AGE 1, AGE 2) and allicin (ALL).

Investigated parameter	CON	AGE 1	AGE 2	ALL
Lysozyme activity (mg/l)	4.64 <sup>a</sup> ± 0.25	5.17 <sup>ab</sup> ± 0.31	6.03 <sup>b</sup> ± 0.68	5.97 <sup>b</sup> ± 0.41
Ceruloplasmin activity (IU)	69.78 <sup>a</sup> ± 3.26	70.31 <sup>ab</sup> ± 2.52	70.46 <sup>ab</sup> ± 1.68	76.95 <sup>b</sup> ± 1.25
Gamma-globulins concentration (g/l)	24.14 ± 1.25	21.08 ± 0.71	20.39 ± 1.77	22.00 ± 1.54
Total protein concentration (g/l)	60.98 ± 1.29	57.27 ± 1.40	59.45 ± 2.49	60.12 ± 1.24
Red blood cells (10 <sup>12</sup> /l)	4.56 <sup>ab</sup> ± 0.16	4.11 <sup>b</sup> ± 0.13	4.86 <sup>a</sup> ± 0.15	4.82 <sup>a</sup> ± 0.20
Haematocrit (%)	29.95 <sup>a</sup> ± 0.95	26.57 <sup>b</sup> ± 0.62	31.83 <sup>a</sup> ± 0.15	29.58 <sup>a</sup> ± 1.03
Haemoglobin concentration (mg/dl)	9.45 <sup>ab</sup> ± 0.22	8.86 <sup>b</sup> ± 0.18	10.10 <sup>c</sup> ± 0.18	8.71 <sup>ab</sup> ± 0.41
White blood cells (10 <sup>9</sup> /l)	11.50 ± 0.37	10.58 ± 0.70	10.46 ± 0.49	10.29 ± 0.54

<sup>a, b, c</sup> Values within rows that do not share common superscripts differ significantly for p ≤ 0.05.

compared to the controls. The analysis of CD in 50% of the small intestine length showed its higher values in AGE 1 and AGE 2 groups, when compared to the control piglets. However, significant differences between the AGE 2 and all other groups were noted in the ileum. Analysis of mucosa thickness (MT) in the duodenum revealed a significant increase of this value in the piglets treated with AGE for 6 days, when compared to the controls. A similar increase of MT was observed in 25%, 50% and 75% of the small intestine length in all the experimental groups. The villus number per centimeter (VN) reached higher values after allicin treatment in 75% of the small intestine length, when compared to the controls.

Allicin administration to piglets significantly increased lysozyme and ceruloplasmin activity in serum, when compared to the control group. Moreover, similar changes of lysozyme activity were observed in the piglets ascribed to the AGE 2 group. While the hematocrit value was significantly decreased in piglets treated with single dosage of AGE, an increase of the haemoglobin concentration was observed in the AGE 2 group (Tab. 5).

## DISCUSSION

Investigations of performance and systemic development of piglets kept in artificial sow provide many advantages when compared to the experimental settings involving pigs reared with their mothers. The most important advantages of artificial sow include the identical environmental conditions such as temperature, humidity and nutrition for all the piglets. Furthermore, the composition of the diet may be easily modulated to investigate effects of administration of biologically active substances on processes of organism growth. Due to numerous similarities of the physiology and anatomy of the gastrointestinal tract of man and pigs, the pig model is a very attractive alternative model for human nutritional studies [14]. Investigations performed in humans and pigs showed that the portions of total life required to reach chemical maturity for both these species are nearly identical; 4.4% and 4.6%, respectively. Even though there are species-specific differences of the placenta and

immunological system of pigs and humans, the piglets are optimal experimental model for investigations concerning physiology and pathology of the gastrointestinal tract of human newborns. The chemical composition of fat-free tissue, including water, proteins and ash, at common stages of life is similar for man and pigs [15].

As opposed to our earlier studies on pigs with administration of AGE and allicin to pregnant and lactating sows and its following consequences for the performance and systemic development of the offspring, this study was performed to investigate effects of postnatal administration of biologically active substances present in these garlic preparations. Considering data indicating that the proper functioning of the gastrointestinal tract and immunological system is crucial for performance, health status and optimal growth rate of piglets, this study focused on investigating the effects of AGE and allicin on the macro- and microstructure of the small intestine and selected specific and non-specific defence mechanisms. The microstructural analysis of the different segments of the small intestine in piglets kept in artificial sow has shown positive effects of oral administration with both dosages of AGE. An increased mucosa thickness was observed in all the investigated segments of small intestine except for the ileum and it was mainly manifested by increased villus length. The double dosage of AGE increased crypt depth in the duodenum, 50% and 75% of the small intestine length and ileum, while this parameter reached higher values in the piglets treated with the single dosage of AGE in the middle part of the length of the small intestine. Next to the improved villus height, AGE administration to piglets increased villus width which commonly resulted in enlargement of villus section area and absorption area of the gut. Similarly to the animals treated with AGE, allicin administration to piglets induced higher values of mucosa thickness and villus height and width, resulting in improved villus section area; however, these effects were not observed in the duodenum and ileum. In contrast to the increased crypt depth in the AGE treated piglets, allicin administration did not significantly influence this parameter in the small intestine. The number of villi per cm was not influenced by AGE administration

and significantly increased in 75% of the small intestine length in the piglets that received allicin. Obtained results are in agreement with our earlier studies where maternal administration of AGE and allicin improved morphology of the small intestine on microstructural level in the offspring. Moreover, in the previous studies, the increase of internal organs weight was observed which seems to be in contrast to the current study; however, a significantly longer period of treatment with the garlic preparations was executed previously [20, 21]. Similar positive effects of AGE administration were reported in studies performed by Horie and colleagues, where the reduction of small intestine damage and diminished weight loss of rats exposed to experimental treatment with antitumour drugs such as methotrexate and 5-fluorouracil was observed [10]. Similar values of ratios of the internal organ weight to body weight obtained in all the experimental groups and controls, both in the previous and the current studies, indicate proper systemic development of the piglets subjected to treatment with the garlic preparations [21]. The positive response of non-specific defence mechanisms like lysozyme and ceruloplasmin activity after AGE and allicin treatment in the current study are in accordance with our previous observations in sows and their offspring. However, the maternal administration of the investigated substances to pregnant and lactating sows seems to be more efficient in relation to the immunological system of pigs, especially when considering its effects on gamma-globulin levels [5, 22]. All these positive effects of AGE and allicin administration on gastrointestinal tract development and defence mechanisms resulted in improvement of final body weight of piglets. After 6 days in artificial sow, the increase of body weights in AGE 1, AGE 2 and ALL groups was observed when compared to these values at the beginning of the experiment. These results were opposite to the control group where no significant changes of body weight were obtained. Furthermore, the obtained results are in contrast to the data presented by Horton and colleagues where administration with fresh garlic did not influence body weight in weaned pigs [11]. On the other hand, the administration of garlic during the first 21 days of life in chickens increased their body weight gain [12]. All data from the studies in suckling piglets and hatched chickens support the hypothesis that beneficial effects of garlic compounds and/or their metabolites on systemic development of organisms are much more effective when the administration is performed at early stages of the postnatal life. Furthermore, administration of biologically active substances of garlic at a late period of gestation, when the fetal growth rate is very high seems to be very effective for regulation of organism development, and this phenomenon was confirmed in studies on pigs where AGE and allicin administration was performed between 91<sup>st</sup> and 115<sup>th</sup> days of pregnancy [20, 21].

Even though the comparison of the AGE 1 and AGE 2 groups in the current study showed an increase of the villus height and villus section area in the duodenum and

25% and 50% of the small intestine length of piglets treated with the single dosage of AGE, this study did not provide final response to the question about dose-dependent effectiveness of the aged garlic extract. Furthermore, the question whether water and alcohol soluble ingredients of AGE are more or less effective than oil soluble allicin on pigs' performance, systemic development, and growth rate still remain unanswered. On the other hand, on the basis of the previous and the current studies, it may be concluded that both administered garlic preparations induce similarly positive effects in pigs. These observations seem to be supported by data on body weight gain, macro- and microstructural properties of the gastrointestinal tract and non-specific and specific defence mechanisms.

## CONCLUSIONS

This study showed that aged garlic extract and allicin administered postnatally to pigs improved development of the gastrointestinal tract. These results are complementary to our previous observation, where the maternal administration of the investigated substances was performed. Increased surface of the small intestine, as well as the immunostimulating effect of allicin and AGE on non-specific defense mechanisms of piglets may be postulated as factors responsible for improved systemic development of piglets. Next to the immunostimulating effects on non-specific defence mechanisms, the antibacterial activity of the investigated substances may all be treated as factors responsible for the obtained results. This matter seems to be very interesting, especially considering the antibacterial activity of aged garlic extract and allicin [1, 2, 6]. Moreover, this study showed that aged garlic extract and allicin as a nutritional supplement may have beneficial effects on health status, performance and systemic development of piglets exposed to weaning-related stress. Due to many similarities between the physiology of the gastrointestinal tract of man and pigs, aged garlic extract and allicin may be considered as attractive supplements for humans; however, further studies in this field are needed.

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## REFERENCES

1. Amagase H, Petesch BL, Matsuura H, Kasuga S, Itakura Y: Intake of garlic and its bioactive components. *J Nutr* 2001, **131**, 955S-962S.
2. Ankri S, Mirelman D: Antimicrobial properties of allicin from garlic. *Microbes Infect* 1999, **1**, 125-129.
3. Beck MJ, Padgett EL, Bowman CJ, Wilson DT, Kaufman LE, Varsho BJ, Stump DG, Nemeć MD, Holson JF: Nonclinical juvenile toxicology testing. In: Hood RD (Ed): *Developmental and reproductive toxicology. A practical approach*, 263-328. Tylor & Francis Group, Boca Raton 2006.

4. Dudek K, Śliwa E, Tatara MR: Changes in blood leukocyte pattern in piglets from sows treated with garlic preparations. *Bull Vet Inst Pulawy* 2006, **50**, 263-267.
5. Dudek K, Tatara MR, Śliwa E, Siwicki A, Łuszczewska-Sierakowska I, Zipser J, Krupski W, Studziński T: Effects of perinatal administration of aged garlic extract (AGE) and allicin on non-specific and specific defence mechanisms in sows. *Pol J Environ Stud* 2005, **14** (Suppl. 2), 69-72.
6. Feldberg RS, Chang SC, Kotik AN, Nadler M, Neuwirth Z, Sundstrom DC, Thompson NH: *In vitro* mechanism of inhibition of bacterial growth by allicin. *Antimicrob Agents Chemother* 1988, **32**, 1763-1768.
7. Hampson DJ: Alterations in piglet small intestine structure at weaning. *Res Vet Sci* 1986, **40**, 32-40.
8. Hampson DJ: Attempts of modified changes in the piglet small intestine after weaning. *Res Vet Sci* 1986, **40**, 313-317.
9. Holson JF, Nemec MD, Stump DG, Kaufman LE, Lindström P, Varsho BJ: Significance, reliability, and interpretation of developmental and reproductive toxicology study findings. In: Hood RD (Ed.): *Developmental and reproductive toxicology. A practical approach*, 329-424. Tylor & Francis Group, Boca Raton 2006.
10. Horie T, Awazu S, Itakura Y, Fuwa T: Alleviation by garlic of antitumor drug-induced damage to the intestine. *J Nutr* 2001, **131**, 1071S-1074S.
11. Horton GMJ, Blethen DB, Prasad BM: The effect of garlic (*Allium sativum*) on feed palatability of horses and feed consumption, selected performance and blood parameters in sheep and swine. *Can J Anim Sci* 1991, **71**, 607-610.
12. Horton GMJ, Fennell MJ, Prasad BM: Effect of dietary garlic (*Allium sativum*) on performance, carcass composition and blood chemistry changes in broiler chickens. *Can J Anim Sci* 1991, **71**, 939-942.
13. Miller BG, James PS, Smith MW, Bourne FJ: Effect of weaning on the capacity of pig intestinal villi to digest and absorb nutrients. *J Agric Sci* 1986, **107**, 579-584.
14. Miller EG, Ullrey DE: The pig as a model for human nutrition. *Ann Rev Nutr* 1987, **7**, 361-382.
15. Moulton CR: Age and chemical development in mammals. *J Biol Chem* 1923, **57**, 79-97.
16. Pluske JR, Hampson DJ, Williams IH: Factors influencing the structure and function of the small intestine in the weaned pigs: a review. *Livestock Prod Sci* 1997, **51**, 215-236.
17. Pluske JR, Williams IH, Aherne FX: Maintenance of villous height and crypt depth in piglets by providing continuous nutrition after weaning. *Anim Sci* 1996, **62**, 131-144.
18. Pluske JR, Williams IH, Aherne FX: Villous height and crypt depth in piglets in response to increase in the intake of cow's milk after weaning. *Anim Sci* 1996, **62**, 145-158.
19. Sivam GP: Protection against *Helicobacter pylori* and other bacterial infections by garlic. *J Nutr* 2001, **131**, 1106S-1108S.
20. Tatara MR, Śliwa E, Dudek K, Kowalik S, Gawron A, Pierasiak T, Dobrowolski P, Studziński T: Effect of aged garlic extract and allicin administration to sows during pregnancy and lactation on body weight gain and gastrointestinal tract development of piglets: Morphological properties of the small intestine. Part II. *Bull Vet Inst Pulawy* 2005, **49**, 455-464.
21. Tatara MR, Śliwa E, Dudek K, Mosiewicz J, Studziński T: Effect of aged garlic extract (AGE) and allicin administration to sows during pregnancy and lactation on body weight gain and gastrointestinal tract development of piglets. Part I. *Bull Vet Inst Pulawy* 2005, **49**, 349-355.
22. Tatara MR, Śliwa E, Dudek K, Siwicki A, Kowalik S, Łuszczewska-Sierakowska I, Krupski W, Zipser J, Studziński T: Influence of perinatal administration of aged garlic extract (AGE) and allicin to sows on some defence mechanisms in their piglets during postnatal life. *Pol J Environ Stud* 2005, **14** (Suppl. 2), 378-381.