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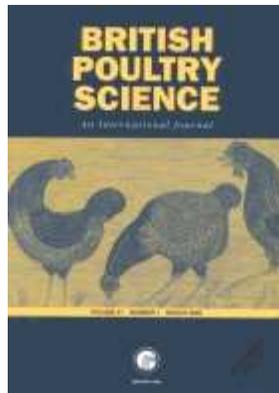
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Chemical composition and biological value of spray dried porcine blood by-products and bone protein hydrolysate for young chickens

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9 3 **Chemical composition and biological value of spray dried porcine blood by-**
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11 4 **products and bone protein hydrolysate for young chickens**
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2
3 31 **Abstract:** 1. The chemical composition of spray dried porcine blood by-products is
4
5 32 characterised by wide variation in crude protein contents. In spray dried porcine blood plasma
6
7 33 (SDBP) it varied between 670-780 g/kg, in spray dried blood cells (SDBC) between 830-930
8
9 34 g/kg, and in bone protein hydrolysate (BPH) in a range of 740-780 g/kg. Compared with fish
10
11 35 meal, these feeds are poor in Met and Lys. Moreover, in BPH deep deficits of Met, Cys, Thr and
12
13 36 other amino acids were found.

14
15
16
17 37 2. The experiment comprised 7 dietary treatments: SDBP, SDBC, and BPH, each at an inclusion
18
19 38 rate of 20 or 40 g/kg diet, plus a control. The addition of 20 or 40 g/kg of the analysed meals
20
21 39 into feeds for very young chickens (1-28 d post hatch) significantly decreased the body weight
22
23 40 (BW) of birds. Only the treatments with 40 g/kg of SDBP and SDBC showed no significant
24
25 41 difference in BW as compared with the control. There were no significant differences between
26
27 42 treatments and type of meal for feed intake, haematocrit and haemoglobin concentrations in
28
29 43 blood.

30
31
32
33 44 3. Addition of bone protein and blood cell meals to feed decreased the IgG concentration in
34
35 45 blood and caused shortening of the femur and tibia bones. However, changes in the mineral
36
37 46 composition of bones were not significantly affected by the type of meal used.

38
39
40 47 4. The blood by-products, which are rich in microelements, improved retention of Ca and Cu
41
42 48 only. In comparison to control chickens, significantly better accretion of these minerals was
43
44 49 found in treatments containing 20 g/kg of SDBP or 40 g/kg of SDBC.

45
46
47 50 5. Great variability in apparent ileal amino acid digestibility in chickens was determined. In this
48
49 51 respect, some significant differences related to the type of meal fed were confirmed for Asp,
50
51 52 Pro, Val, Tyr and His. In general, the apparent ileal digestibility of amino acids was about 2-3
52
53 53 percentage units better in chickens fed on diets containing the animal by products than in control
54
55 54 birds.
56
57
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59
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INTRODUCTION

55 Studies on the determination of the biological value of blood meal and blood by-products as
56 alternative protein sources instead of fish meal, to a great extent have been carried out with pigs,
57 especially with early-weaned animals. The main purpose of these investigations was to
58 recognise the effects of using these feed components on intestinal functions and modulation of
59 immune response (Cain *et al.*, 1992; DeRouchey *et al.*, 2002; Campbell *et al.*, 2009; Moretó
60 and Pérez-Bosque, 2009). Supplementation of diets for piglets and pigs with spray dried porcine
61 plasma has positively influenced collagen synthesis and small intestine growth, improved the
62 activity of insulin-like growth factor, and improved the performance of animals (De Rodas *et*
63 *al.*, 1995; Jiang *et al.*, 2000; Rodriguez *et al.*, 2007; King *et al.*, 2008). The number of
64 publications on the physiological effects of these feed supplements in poultry is small, and the
65 results are unclear (Orda *et al.*, 1988; Mazurkiewicz *et al.*, 1990; Bregendahl *et al.*, 2005; Vasan
66 *et al.*, 2007).

67
68 The amino acid composition of blood or bone proteins (mainly collagen) is
69 characterised by the huge deficit of Met in blood cell meal – that, plus a deficit of Ile and
70 relatively high amounts of Val, Glu and Phen have been stated. The bone protein is rich in Gly,
71 Pro and Arg. So, the use of these by-products leads to an amino acids imbalance in prepared
72 feeds (Moughan *et al.*, 1999; Pieper *et al.*, 2002; Kerr *et al.*, 2004; Marquez *et al.*, 2005). Blood
73 and particularly bone proteins are relatively resistant to enzymatic degradation (Vasan *et al.*,
74 2007), and both the content and availability of some amino acids for animals can be heavily
75 modified as an effect of higher temperatures during the drying process (Moughan *et al.*, 1999;
76 Marichal *et al.*, 2000).

77 The porcine blood and bone by-products obtained by the modified spray drying method
78 can be treated as a potentially beneficial source of proteins, amino acids, microelements and
79 some biologically active substances for monogastric animals.

1
2
3 80 The purpose of the investigation was to collect more data about the variability in
4
5 81 chemical composition of both porcine blood and bone by-products and to determine their
6
7 82 usefulness in the diet of young chickens.
8

9 83 MATERIAL AND METHODS

10 84 **Characteristics of spray dried porcine blood by-products and bone protein hydrolysate**

11
12 85 Raw materials for the study were collected from the Polish feedstuffs market or obtained
13
14
15
16
17 86 directly from different Polish producers. In total, 48 different production batches (samples) were
18
19 87 examined. Within these, 21 samples of spray dried porcine blood plasma (SDBP), 16 blood cell
20
21 88 meals (SDBC) and 11 samples of bone protein hydrolysates (BPH) were chemically analysed
22
23
24 89 (Tables 1 and 2). According to the information obtained from manufacturers, all the animal
25
26 90 meals were produced using similar technologies. The functional properties of these feeds were
27
28
29 91 examined using young chickens as sensitive model animals.
30

31 92 The technology of meal production comprised the following procedures: porcine blood
32
33 93 was cooled to 3°C, mixed with trisodium citrate solution as an anticoagulant, and centrifuged to
34
35
36 94 separate the plasma and blood cells. The process of substrate spray drying was conducted using
37
38 95 a carrier, in order to increase the drying surface. The applied temperature was as follows: for
39
40
41 96 plasma, from 80-98°C at the start down to 38-45°C at final stage of drying (mean about 50°C);
42
43 97 and for blood cells about 120–130°C and 40°C, respectively. Tables 1 & 2 near here
44

45 98 The bone pulp was treated with lactic acid (as a catalyst) and alkaline solutions at a
46
47
48 99 temperature of 50–150°C. The next step was hydrolysis at 130-140°C and a pressure of 0.3-0.4
49
50
51 100 MPa. Decanting was carried out at 90°C for fat separation. The final product was dried. The
52
53 101 substantial part of this information was obtained from the meal producers; however, details of
54
55
56 102 specific modifications to production methods were not available.
57
58 103
59
60

104 **Investigation with chickens**

105 One d old Hubbard Flex male broiler chicks with an average initial body weight of 41.6 g were
106 randomly assigned to 7 treatments and kept in battery cages. Each treatment comprised 4
107 replicates (cages) with 10 birds per cage. The room temperature was gradually reduced from an
108 initial 32°C to 20°C during the first 4 weeks post hatch. Relative humidity inside the room
109 varied from 64 to 70 %. The lighting programme during the experiment was 24 h light per d.
110 The chickens had free access to drinking water given via nipple system. During the 4 weeks
111 post-hatch the chicks were fed *ad libitum* on one of 7 isoenergetic (12.5 MJ/kg) and isoprotein
112 (215.7-218.4 g/kg) starter mash diets. The 7 diets were supplemented with SDBP, SDBC, or
113 BPH, at an inclusion rate of either 20 or 40 g/kg diet; plus a control diet (Tables 3 and 4).

Tables 3 &
4 near here

114 Control chickens were fed on a diet free of animal meals, in which soya bean meal was
115 the main protein source. The composition of feed mixtures was calculated on the basis of the
116 authors' own analytical data for feed components using simple linear optimisation. The
117 vitamin/mineral premix was free of feed supplements. Only the coccidiostat Diclazuril was
118 used.

119 **Balance trial**

120 The balance trials, in which retention of mineral elements was determined (4 cages with 9-10
121 birds per treatment), were carried out at 22-26 d of age. The feed intake was precisely controlled
122 and the excreta were quantitatively collected for each cage-replication twice a day, for 5 d. The
123 mineral composition of both feed and excreta was recorded.

124 **Assays of amino acid digestibility**

125 On d 27 post hatch, precisely two hours after feeding, 12 chickens from each treatment (three
126 birds per cage) were randomly selected, weighed and killed by cervical dislocation, and then the
127 digestive tracts were immediately removed from the body. The contents of that part of the ileum
128 from Meckel's diverticulum to the junction with the caecum were collected for determination of
129 apparent ileal amino acids digestibility (7 treatments x 4 replications, each sample pooled from

1
2
3 130 three birds per cage). As an indicator, Cr₂O₅ (5 g/kg in diets) was used. Such procedures have
4
5 131 been effectively used in many investigations in which intestine contents have been analysed
6
7 132 (Ten Doeschate *et al.*,1993; Ravindran *et al.*,1999, 2004; Jamroz *et al.*, 2001, 2002; Lemme *et*
8
9 133 *al.*, 2004).

12 134 **Blood collection and bone characteristics**

14 135 On d 28, the next 12 chickens from each treatment (three birds per cage) were randomly selected
15
16 136 for blood sampling, in which haematocrit, haemoglobin and immunoglobulin G concentrations
17
18 137 were determined. Moreover after killing, both legs were removed, the muscles separated from
19
20 138 the bones and the tibia and femur were cleaned. The metric parameters were estimated in the
21
22 139 fresh bones and these were saved from drying and stored in the refrigerator. In defatted (ether
23
24 140 extract at 45°C for 24 h according to Soxhlet method) and dried bones the crude ash, Ca, P and
25
26 141 Zn contents were determined (samples pooled from both femurs and both tibias from each
27
28 142 chicken). All analyses were done in duplicate.

33 143 **Histological examinations**

34 144 Prior to the leg and bone preparation, the digestive tracts of the chickens were removed and
35
36 145 from each of 6 birds per treatment, the central portions of the ileum were collected for
37
38 146 histological examination of the intestine walls. The samples of ileum were stabilised and fixed
39
40 147 in aqueous formalin solution 9:1 (v/v) for 24 h, the samples were then rinsed in tap water,
41
42 148 dehydrated in alcoholic series and embedded in paraffin. Material was cut into 7 µm thick strips
43
44 149 that were stained with haematoxylin according to Delafield's method, with eosin (cell
45
46 150 cytoplasm), and by the AZAN-NOVUM method according to Geidies (Gable,1968). The
47
48 151 histological preparations were examined using a light microscope (Nicon Eclipse 80I) and for
49
50 152 each sample, 20 microphotographs taken with a Canon PS66 camera were reviewed.

57 153 **Analytical methods**

58 154 The chemical composition of the spray dried animal meals, feed compounds, complete diets and
59
60 155 tibia bones was determined according to the standard methods AOAC (2005): the nitrogen

1
2
3 156 content by Kjeldahl-method (984.13) using Kjeltec 2300 Foss Tecator apparatus (Sweden);
4
5 157 crude protein by multiplying the N-content by 6.25; crude fat by ether extraction (920.39); and
6
7 158 crude fibre by the Henneberg-Stohmann method (978.10) using Fibertec Tecator (Sweden)
8
9
10 159 apparatus. The mineralisation of samples was performed with MarsX apparatus (CEM
11
12 160 Corporation) with nitric acid. Phosphorus was analysed, after previous mineralisation with nitric
13
14 161 acid (HNO₃) and perchloric acid (HClO₄), by the ammonium vanadomolybdate method using a
15
16 162 spectrophotometer (Specol 11, Carl Zeiss, Jena) at a wave length of 470 nm (995.11); calcium and
17
18 163 other minerals were determined by atomic absorption spectrophotometry using AA 240 FS
19
20 164 apparatus with SIPS 20 (Varian) (968.08).
21
22
23

24 165 For the determination of the amino acids (994.12), samples of components were
25
26 166 hydrolysed with 6M hydrochloric acid (HCl) for 24 h at 110°C, and the amino acids separated
27
28 167 according to the Moore (1963) and Moore and Stein method (1963). For sulphur amino acids
29
30 168 determination, the feed samples were oxidised (0°C, 16 h) with formic acid and hydrogen
31
32 169 peroxide (H₂O₂) (9:1/v:v) prior to HCl hydrolysis, and then were separated using an Analysator
33
34 170 AAA 400 Ingos (Prague, Czech Republic) (985.25). For tryptophan content, after alkaline
35
36 171 hydrolysis with lithium hydroxide (LiOH) (110°C, 16 h) and 4-dimethylaminobenzaldehyde
37
38 172 (DMAB), the samples were examined colorimetrically at a wave length of 590 nm according to
39
40 173 Landry and Delhaye (1992) (988.15). On the basis of obtained results the amino acid contents in
41
42 174 feed mixtures were optimised, then subsequently analysed in the finished feeds, because the
43
44 175 correct amino acid content in feed is important for regular synthesis of organic matrix – bone
45
46 176 proteins.
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53 177 The gross energy density in the spray dried animal meals was determined using an
54
55 178 automatic calorimeter KL-10f Precyzja Bydgoszcz (Poland). The energy value of the diets was
56
57 179 calculated on the basis of the determined nutrients, and according to the formula published in
58
59 180 European Tables of Energy Values of Feeds for Poultry, WPSA (1989).
60

181

182 **Statistical analysis**

183 All data were evaluated statistically by one or two-factorial ANOVA using StatSoft Statistica®
184 software (2005). The differences for all parameters were tested according to the following
185 statistical model:

$$186 \quad y_{ij} = \mu + a_i + e_{ij}$$

187 or

$$188 \quad y_{ijk} = \mu + a_i + b_j + (ab)_{ij} + e_{ijk}$$

189 where y_{ij} or y_{ijk} is the variance associated with parameter a , μ is the overall mean, a_i is the
190 treatment effect, b_j is the kind or inclusion rate of meal (plasma or blood cells) in the feed
191 mixture, $(ab)_{ij}$ is the interaction effect, and e_{ij} or e_{ijk} is the error term. The individual
192 measurements or averages for cages (replicates) (for digestibility data) were treated as the
193 experimental units, and differences between treatment means were analysed for significance
194 ($P < 0.01$ or $P < 0.05$) using Tukey's test. The data are presented as average values and
195 accompanied by SEM (standard error).

196 All procedures that were carried out with animals have been approved by the Local Ethic
197 Commission for animal experiments.

198 **RESULTS**

199 **Animal meal composition**

200 The amino acid contents in blood by-products and bone protein hydrolysate were characterised
201 by great variability (Table 1). The greatest divergence between production batches was observed
202 for Cys, Met, Try and Ile.

203 The best composition, considered from the point of view of limited amino acids in the
204 diets for monogastric animals (such as Met + Cys, Lys, Try and Thr) was found in dried plasma;
205 however, blood cell meal contains more Lys, two times more Leu, and more Val, His and Phe.
206 Bone protein hydrolysate was rich in Gly, Pro and Arg, but poorer overall due to a lack of Met +
207 Cys, Lys, Thr, Try, Val and His (Table 1).

1
2
3 208 The crude ash content in analysed porcine blood by-products was lower than in fish meal, and
4
5 209 the Ca, P, and Mg concentrations were many times lower. In blood cell meal, a high Fe content
6
7 210 was found (Table 2).

11 211 **Results of investigation on chickens**

12 212 Inclusion of 20 or 40 g/kg of spray dried porcine blood by-products and bone protein
13
14 213 hydrolysate in feed mixtures significantly ($P<0.01$) modified the body weight of chickens. The
15
16
17 214 best results were obtained in the control treatment, and a drastic reduction of chickens' body
18
19 215 weight was noted in treatments with BPH, especially at the dose of 40 g/kg (Table 5). For
20
21 216 average feed intake no significant differences among treatments or experimental factors were
22
23
24 217 observed

Table 5 near here

25
26 218 During the first 5 d of life any weak birds were removed from each treatment. The
27
28
29 219 dietary treatments had no effect on health status of birds and the culling of chickens was not
30
31 220 connected with the type of diet.

32
33 221 For the selected blood parameters – haematocrit (Ht), haemoglobin (Hb) and
34
35
36 222 immunoglobulin G – the significantly ($P<0.05$) lower Hb and IgG concentrations were
37
38 223 observed in chickens fed on diets with BPH; however, these were observed in individual cases
39
40 224 only. The Ht level was similar in all treatments. The femur and tibia bones of chickens were
41
42
43 225 significantly shorter ($P<0.01$) for the hydrolysate treatments than in other chickens. However, in
44
45 226 their chemical composition – in crude ash as well as in Ca, P and Zn content – no significant
46
47
48 227 differences between treatments and for experimental factors were observed (Table 5). The
49
50 228 interaction between the type of animal meal used and the inclusion rate in the treatments was
51
52 229 significant ($P<0.01$) for body weight and tibia length, only.

53
54
55 230 The relative accretion of minerals in 22-26 d old chickens was not high, and for Ca it
56
57 231 varied between 42-56; for P 38-51; for Na 69-78; for K 75-80; for Cu 63-76; for Zn 16-36; for
58
59 232 Fe 10-23; and for from Mn 9-28%, however the differences between treatments were significant
60
233 ($P<0.05$) for Ca and Cu only. In two-factorial ANOVA, non-significant differences for type and

1
2
3 234 inclusion rate of animal meals were confirmed (Table 6), except of Cu, for which the interaction
4
5 235 between kind and share of animal meals was significant. The highest Cu retention in chickens
6
7 236 was found in treatments fed on diets with plasma and blood cell meal ($P<0.05$).

9
10 237 Marked and, for many amino acids, significant differences in apparent ileal digestibility
11
12 238 were found as an effect of the type of animal meal used in one-factorial ANOVA, specially for
13
14 239 Asp, Ser, Pro, Gly, Ala, Met, Tyr, His and Arg. In two-factorial analysis of variance,
15
16 240 significantly higher apparent ileal digestibility of numerous amino acids was noted in chickens
17
18 241 fed on diets containing SDBP. Incorporation of BPH into diets resulted in higher amino acid
19
20 242 digestibility than in other treatments for Pro, Gly, Ala, Met and Arg only (Table 7). The diets
21
22 243 with 40 g/kg of animal meal enhanced the apparent ileal digestibility of many amino acids, but
23
24 244 significant differences were observed for Ala, Cys and Met only. For numerous amino acids the
25
26 245 interaction between type and proportion of animal meals was significant ($P<0.05$). Table 7 near here

31 246 **Histological assays**

32
33 247 The histological photomicrographs of jejunum of control chickens displayed structures that were
34
35 248 characteristic for a normally functioning organ (Figures 1 and 2). The mucosa was correctly
36
37 249 formed into villi, which in turn formed the finger-shaped prominences covered with monolayer,
38
39 250 cylindrical epithelium, containing enterocytes, goblets cells, distinctly different unicellular,
40
41 251 intraepithelial endocrine glands (cells of APUT system), and single cells with light cytoplasm.
42
43 252 In the mucosa were observed fibroblasts, fibrocytes, mastocytes and white blood cells of the
44
45 253 immune system (lymphocytes, neutrophils). The zone of subepithelial connective tissue was
46
47 254 intensively vascularised and innervated. The central part of the intestinal villus was occupied by
48
49 255 the lymphatic space. The submucosa was formed from the flaccid connective tissue with
50
51 256 abundant blood vessels and nerves. Sporadically the single lymphatic nodules were observed.
52
53 257 The muscular membrane was formed from two layers of muscles: the longitudinal and the
54
55 258 circular layer. In the space between these layers the aggregations of the neural cells creating the
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1
2
3 259 intramuscular complex were observed. The external layer of the intestine wall was covered by
4
5 260 serosa with squamous epithelium.

Figures 1 & 2 near here

7 261 The photomicrographs of jejunum wall taken from chickens fed on diets containing
8
9 262 SDBP show higher finger formed villi than in control animals (Figures 3 and 4). The zone of
10
11 263 endocrine glands was similar to the intestine wall of control chickens. In single places in the
12
13 264 deep zone of villi, the proliferation of lymphoreticular tissue, which leads to the formation of
14
15 265 single lymphatic nodules, was observed. The proliferation of reticular tissue was found in some
16
17 266 of the villi. On the surface zone of intestinal villi, numerous enterocytes and mucogenic cells
18
19 267 were observed. The histological pictures of jejunum mucosa show a mobilisation of the zone of
20
21 268 the mucosa membrane (epithelium and subepithelial connective tissue). This is the expression of
22
23 269 the activation of a local immunological area (GALT). Other layers of the intestine wall were
24
25
26
27
28 270 unchanged compared with the pictures in control chickens.

Figures 3 & 4 near here

31 271 The histological characteristics of the intestine wall of chickens that consumed SDBC in
32
33 272 the diet were, in principle, similar to the above described pictures (Figures 5 and 6); however
34
35 273 some differences were observed. For instance, between very high villi some shorter ones, or villi
36
37 274 in the leafy form, were observed. On the surface of the epithelium covering the intestinal villi
38
39 275 the greatest number of mucinogenous cells were present. Most of the epithelium layer shows
40
41 276 the normal picture of connective tissue without numerous centres of proliferating
42
43 277 lymphoreticular tissue. The intensity of this proliferation was lower than in chickens of the
44
45 278 control and plasma treatments. The glandular zone was similar to the picture observed in control
46
47
48
49 279 birds.

Figures 5 & 6 near here

52 280 Feeding the chickens with diets containing BPH caused some changes in histological
53
54 281 pictures, from finger villi to the leafy form, with an increased number of mucinogenous cells in
55
56 282 the epithelium covering the intestinal villi (Figures 7 and 8). Large numbers of these cells were
57
58 283 present in the distal and subdistal zone of villi. In the deep zone and on the fundus of the
59
60 284 intestinal gland, the number of mucinogenous cells was smaller. Here the enterocytes

1
2
3 285 predominated, but in the specific layer of villi mucosa a clear mobilisation of immunocells and a
4
5 286 proliferation of lymphoreticular tissue were observed. Arising from these, the lymphatic nodules
6
7 287 were present in the corresponding layers of villi mucosa. The glandular zone of intestine wall
8
9 shows evidence of increased secretion. Figures 7 and 8 near here

11 289 DISCUSSION

12
13
14 290 The chemical composition of the porcine blood by-products was greatly variable, *i.e.* the crude
15
16 291 protein in dried plasma varied between 670-780 g/kg, and in dried blood cells it ranged from
17
18 292 830 to 930 g/kg. This changeability resulted in different amino acid contents. As compared with
19
20 293 fish meal, in the analysed raw materials a lower amount of Met, and clearly lower concentration
21
22 294 of Lys were observed. However, in comparison with the soya bean meal, dried plasma is rich in
23
24 295 Cys, Lys, Thr, Leu and Val. Large deficits of Met, Cys, Lys, Thr, Try, Leu, His and Tyr can be
25
26 296 seen in bone protein compared with fish meal. Both SDBP and SDBC are widely used as a good
27
28 297 protein source for weaned pigs. The blood protein contains fibrinogen, immunoreactive
29
30 298 globulins and albumins (Del Rio de Reys *et al.*, 1980; De Rodas *et al.*, 1995; Gómez-Juárez,
31
32 299 1999; Pierce *et al.*, 2005), as well as immune stimulating nucleotides (Martinez-Puig *et al.*,
33
34 300 2007). The content of nucleotides in atomised blood cells amounted to 55 ppm, in atomised
35
36 301 porcine plasma to 7 ppm, and in serum protein concentrate to 282 ppm. Antibacterial activity of
37
38 302 SDBP was stated in investigations carried out by Campbell *et al.* (2009). A progressive increase
39
40 303 in small intestine mass, and improvements in feed efficiency and nutrient retention in pigs and
41
42 304 mice were obtained in the studies of Hansen *et al.* (1993), De Rouche *et al.* (2002) and Pierce
43
44 305 *et al.* (2005), in which spray dried blood by-products were used as protein sources in feed
45
46 306 mixtures. No similar results were found in the investigations of other authors (Gatnau *et al.*,
47
48 307 1989; Kats *et al.*, 1994; Coffey and Cromwell, 1995; De Rodas *et al.*, 1995). In the opinion of
49
50 308 Goodfredson-Kisic and Johnson (1997) improvement of performance in weanling pigs depends
51
52 309 on the immunoglobulins, but not on the fibrinogen or albumin fractions.
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2
3 310 Immunoglobulins that are present in animal blood by-products may contain some
4
5 311 antibodies active against some bacterial pathogens (Rump *et al.*, 1992; Owusu-Asiedu *et al.*,
6
7 312 2002), although the antibacterial action of spray dried plasma has not been confirmed.

9
10 313 In the numerous investigations carried out with weaned pigs and growing animals, a
11
12 314 beneficial response to porcine plasma as a replacement for blood meal and meat meal were
13
14 315 obtained; however, such positive effects were not observed in the authors' own studies
15
16 316 conducted on very young chickens. The use of SDBC or BPH as partial replacement for soya
17
18 317 bean meal (SBM) significantly decreased the body weight of chickens by d 28 post hatch. In
19
20 318 contrast, the use of SDBP at a rate of 40 g/kg in feed mixtures, non-significantly reduced body
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22 319 weight and feed intake in chickens compared with control birds. These results are contrary to
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24 320 the effects obtained in young pigs (Hansen *et al.*, 1993; De Rodas *et al.*, 1995). The findings of
25
26 321 this study may be confirmed by the research by Donkoh *et al.* (1999) in which similar results
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28 322 were obtained: there was no impact on feed intake, but better weight gain in broilers fed on
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30 323 blood by-products was observed. Kerr *et al.* (2004) reported better results by increasing the
31
32 324 amount of SDBC in feed for pigs from 4 to 6%; in other experiments the use of 2 or 5% SDBC
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34 325 resulted in improved performance. These authors explain such findings as due to insufficient
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36 326 isoleucine content, caused by low concentrations of this animal meal in feed mixtures. Results
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38 327 from this study cannot be explained in the same way, because the experimental diets were
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40 328 variable in isoleucine content to a small extent only. In this experiment, the use of animal meals
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42 329 had no effect on haematocrit, but had a small effect on haemoglobin content in chicken blood.
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44 330 Similar results were presented by Donkoh *et al.* (1999) who added dried blood meal into broiler
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46 331 diets. In the research of these authors, and in this study as well, the mortality of chickens was
47
48 332 also unaffected by the treatment. The IgG concentration in the blood of young chickens fed on
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50 333 diets containing SDBC and BPH was found to be significantly lower in comparison with birds
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52 334 fed on control or dried plasma diets. Different results, i.e. reduced immune response in pigs fed
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60 335 on diets with dried plasma, were noted by Campbell *et al.* (2009).

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3 336 The relatively large amounts of glycine, prolamine and alanine in bone protein may
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5 337 suggest the possibility of a beneficial effect from these feed materials on the formation of the
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7 338 organic matrix of bones. In the present investigation, in chickens fed on diets containing BPH
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10 339 the decrease in body weight was accompanied by a simultaneous significant reduction in bone
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12 340 length. Moreover, non-significant differences among treatments were found in the crude ash
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15 341 content of bone, and the Ca, P and Zn concentrations in ash.

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17 342 In total, improved Ca and Cu retention was observed in chickens fed on diets with
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19 343 animal meals (significant differences were found between control and plasma treatments only).
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21 344 Published information which analysed the relation between mineral composition of spray dried
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24 345 blood by-products or bone protein hydrolysate, and its probable impact on bone quality, was not
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26 346 found in the available literature.

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28
29 347 Inclusion of blood by-products and bone protein into experimental feed mixtures resulted
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31 348 in moderate variability in amino acid composition. Nevertheless, great variability and improved
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33 349 coefficients of apparent ileal digestibility of amino acids was found compared with controls in
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36 350 chickens receiving the animal meals. Some differences between treatments were found to be
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38 351 significant for Asp, Pro, Val, Tyr and His. The influence of inclusion rate of blood or bone by-
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40 352 products on amino acid digestibility was significant only in individual cases. Overall, the
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43 353 digestibility of amino acids from diets enriched with animal meals was better by 2-3 %-units
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45 354 when compared with the control feed.

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48 355 In poultry diets, methionine and cystine are the first, and lysine the second, limiting
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50 356 amino acids. The blood by-products are poor in sulphur amino acids, but in comparison with
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52 357 soya bean meal are relatively rich in lysine. Dried blood cells are poor in isoleucine (Kerr *et al.*,
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54
55 358 2004). The concentration of this amino acid (AA) in bone protein is low. In total, the best AA
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57 359 composition was found in the SDBP. High values for ileal digestibility coefficients of AA from
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59 360 evaluated feed mixtures were compatible with the data estimated for meat-bone and fish meals
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361 by Farrell *et al.* (1999) and Huang *et al.* (2005). Very efficient digestibilities of lysine in

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3 362 experimental diets, estimated at 86-88%, demonstrate that the technology of production of spray
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5 363 dried animal meals has not decreased (in comparison with control) the digestibility of this amino
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7 364 acid. The use of older technologies of drying (Batterham *et al.*, 1986) indicated lower nutritional
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10 365 values for both bone and meat meals in pigs, and also in poultry; however, chickens were more
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12 366 efficient in utilisation of lysine than pigs. The availability of lysine in blood meal to poultry
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14 367 varied between 68-88 %, and for pigs between 48-88 %, of total lysine (Batterham *et al.*, 1986).
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17 368 The great variability in chemical composition of analysed animal meals (Tables 1, 2 and 8, 9,
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19 369 10) can result from non-standardised raw materials. Some differences in technological processes
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21 370 – duration of heating, denaturation of protein, or Maillard reaction, can explain the effects of
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24 371 some biologically active substances (Marichal *et al.*, 2000). Tables 8, 9, 10 near here

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26 372 The histological assays indicate that the structure of jejunum walls in chickens fed on
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29 373 diets containing soya bean meal, and spray dried porcine blood and bone by-products was to a
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31 374 great extent similar to the histological picture that was characteristic for a normally functioning
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33 375 intestine wall. After inclusion of spray dried porcine plasma into the feed mix, higher villi than
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35 376 in control animals were observed. In these the mobilisation of the mucosa membrane and
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37 377 expression of the activation of immunological areas was seen. The use of dried blood cells as a
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39 378 component of feed mixes, caused some changes in histology of the jejunum such as variability
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41 379 in villus height, the appearance of greater numbers of mucinogenous cells, and lowered
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43 380 proliferation of lympho-reticular tissues. In principle, the histological picture indicated the
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45 381 normality of intestine wall functions.

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47 382 Inclusion of BPH in feed mixes led to formation of leafy shaped villi, increased numbers
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50 383 of mucinogenous cells in the epithelium of intestinal villi, and the mobilisation of immunocells.

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52 384 The histological findings of other authors, obtained in pigs, indicate that dried plasma as
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55 385 a protein source has no or little effect on the morphology of the intestine wall (Nofrarias *et al.*,
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57 386 2006), or on villus height, crypt depth, and villus and crypt goblet cell density (King *et al.*,
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3 387 2008). Also, in an experiment carried out by Jiang *et al.* (2000) no significant differences in
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5 388 mucosa villus height, crypt depth or cell proliferation index, were observed.

7 389 In conclusion, it could be stated that the presence of dried plasma in feed mixes given to
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9 390 monogastric animals, did not reduce the functions of the epithelial cells of the small intestine
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11 391 wall surface. The substantial differences observed between pigs (especially very young animals)
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13 392 and poultry, in efficiency of utilisation of the biologically active proteins in dried porcine
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15 393 plasma and blood cells, probably can be explained by the ability of porcine peptides or amino
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17 394 acids to pass across the jejunum wall and their activity in immune modulation and nitrogen
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19 395 metabolism. Probably the size of porcine blood proteins or peptide molecules limits or even
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21 396 renders impossible their utilisation in the intestinal tract and wall, and hence their participation
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23 397 in biological functions inside the bird itself.

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26 398 The mode of action of these specific feed compounds need to be more precisely
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28 399 explained by further investigation.

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600 FIGURE LEGENDS

601
602 **Figure 1.** *Glandular zone of jejunum wall in chicken of control treatment (H+E x400).*

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605 **Figure 2.** *Distal zone of villi in chicken of control treatment (H + E, x 400).*

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609 **Figure 3.** *Glandular zone of jejunum of chicken fed on diets with plasma meal, with single*
610 *centers of proliferation of lymphoreticular tissue (H + E, x 400).*

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614 **Figure 4.** *Distal part of jejunum villi with greater number of monogenous cells and*
615 *proliferating enterocytes in chickens fed on diets with plasma meal (H + E, x 400)*

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618 **Figure 5.** *Glandular zone of jejunum wall from chicken fed on blood cell diets. Picture similar*
619 *to that in control animals (H + E, x 400).*

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622 **Figure 6.** *Distal part of jejunum wall of chickens fed on blood cell meal diets. Picture similar to*
623 *control (H + E, x 400).*

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625
626 **Figure 7.** *Glandular zone of chickens fed on diets with bone protein hydrolysate, with visible*
627 *proliferation of enterocytes and mobilisation of lymphoreticular cells, single lymphotic goblet*
628 *cells (H + E, x 400).*

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631 **Figure 8.** *Distal part of jejunum wall with greater number of mucogenous cells and enterocytes*
632 *proliferation (H + E, x 400).*

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Table 1. *Amino acid content of spray-dried porcine blood by-products and bone protein hydrolysate (g/kg) collected from different producers (own results)*

Amino acids	Plasma		Blood cells		Bone protein hydrolysate	
	mean	± SD	mean	± SD	mean	± SD
Asp	69.38	4.74	98.07	10.54	55.50	7.05
Thr	38.78	4.77	30.64	1.81	17.39	1.84
Ser	40.02	4.22	40.28	2.71	29.34	2.32
Glu	83.20	8.75	68.68	2.85	82.27	6.69
Pro	38.98	4.23	32.75	2.94	116.05	8.98
Cys	17.95	3.45	9.36	2.09	0.43	0.36
Gly	26.54	1.80	39.23	4.79	192.34	12.03
Ala	40.56	3.15	64.87	8.20	79.13	5.22
Val	44.68	3.72	65.94	8.43	21.45	2.92
Met	5.50	1.21	6.46	1.07	5.83	3.57
Ile	20.33	3.50	7.32	2.20	10.20	1.44
Leu	68.63	5.11	105.70	12.25	28.46	2.34
Tyr	29.25	3.94	19.56	2.02	7.15	1.83
Phe	39.12	2.68	55.40	5.85	19.47	2.01
His	26.84	3.31	57.94	8.62	9.78	1.14
Lys	59.43	5.19	75.97	6.75	33.04	3.16
Arg	44.68	4.35	42.29	3.82	77.20	8.78
Try	10.06	1.28	10.17	4.67	0.63	0.01
Met+Cys	23.44	4.42	15.82	2.84	6.00	3.78
Total	703.94	52.41	830.62	70.63	784.96	59.73
Number of samples - production batches	21		16		11	

647 **Table 2.** Contents of minerals and gross energy value of spray-dried porcine blood by-products and bone protein hydrolysate

Mineral elements	Plasma		Blood cells		Bone protein hydrolysate		Fish meal**, 2005	Blood meal*	
	mean	± SD	mean	± SD	mean	± SD		mean	± SD
Crude ash, g/kg	162.2	40.4	73.6	28.2	43.1	28.9	162-210	93.0	2.7
Ca, g/kg	1.03	0.44	0.46	0.20	1.55	0.56	33-38	3.0	0.13
P	1.80	0.29	2.50	0.20	0.82	0.34	27-28	1.7	0.11
Mg	1.48	0.77	0.73	0.47	0.21	0.10	2.0	2.0	0.21
Cl	21.90	3.17	14.08	2.74	15.93	23.20	-	2.5	0.37
Na	70.61	29.86	34.18	17.63	4.62	3.12	-	3.1	0.84
K	13.35	3.73	8.78	2.20	3.30	2.01	-	0.9	0.27
Fe, mg/kg	998.47	310.70	1770.0	400.0	169.01	71.60	-	2948	380.0
Mn	11.87	7.35	6.45	1.88	7.19	2.71	36	5.0	0.73
Cu	50.25	17.49	19.33	21.61	12.62	20.50	-	8.2	3.10
Zn	79.88	39.69	67.92	20.02	19.77	8.83	148	8.9	2.51
Estimated gross energy value, MJ/ kg	30.83	2.85	32.20	3.10	30.78	2.22	-	-	-
Number of samples – product batches	21		16		11		-	7	

648 *means for own laboratory (Wroclaw University of Environmental and Life Sciences).

649 ** Polish Recommendations for Poultry Nutrition, 2005.

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Table 3. *Composition of experimental feed mixes, g/kg*

Components	Feeding groups - treatments						
	Control	Plasma		Blood cells		Bone protein hydrolysate	
			20	40	20	40	20
Maize	200	200	200	200	200	200	200
Wheat	250	273	295	280	309	286	321
Barley	100	100	100	100	100	100	100
Rapeseed oil	70	65	60	64	59	64	59
Soya bean meal	337	301	265	294	251	287	237
Dried porcine plasma	-	20	40	-	-	-	-
Dried porcine blood cells	-	-	-	20	40	-	-
Bone protein hydrolysate	-	-	-	-	-	20	40
Dicalcium phosphate	17.6	17.9	18.5	18.8	19.2	18.2	18.4
Chalk	3.9	3.9	4.0	4.0	4.1	3.8	3.7
NaCl	3.5	1.8	-	1.8	0.2	3.4	3.3
Cr ₂ O ₃	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Premix DKA S*	10.0	10.0	10.0	10.0	10.0	10.0	10.0
DL-methionine 98%	2.4	2.4	2.5	2.4	2.5	2.5	2.6
L-lysine 98%	0.6	-	-	-	-	0.1	-

* In 1 kg of premix: retinylpalmitate 693.750 µg, cholecalciferol 7.500 µg, α-tocopherol acetate, 5.0 g, menadione 0.3 g, thiamin 0.2 g, riboflavin 0.6 g, pyridoxine 0.35 g, cyanocobalamin 0.002 g, Ca-pantothenate 1.5 g, biotin 0.06 g, nicotinic acid 5g, folic acid 0.15 g, choline chloride 6.0 g, Fe 6 g, Cu 0.8 g, Mn 9.0 g, Zn 8 g, Co 0.04 g, I 0.08 g, Se 0.025 g, Diclazuril 100 mg.

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675**Table 4.** *Chemical composition of experimental mixtures*

Item	Treatments						
	Control	plasma		blood cells		bone hydrolyzate	
		g/kg					
		20	40	20	40	20	40
Metabolisable energy, MJ/kg (calculated)	12.5	12.5	12.5	12.5	12.5	12.5	12.5
Crude protein, g/kg	218.4	217.0	215.7	218.0	217.4	217.6	216.8
Crude fibre	37.7	36.1	34.5	35.9	34.1	35.6	33.5
Crude fat	85.7	80.7	75.8	79.8	75.0	79.9	75.1
Crude ash	30.0	31.6	33.3	29.1	28.2	28.2	26.4
Ca, g/kg	9.33	9.46	9.36	9.25	9.53	9.30	9.25
P-total	7.17	7.30	7.18	7.12	7.33	7.18	7.12
Mg	1.62	1.60	1.58	1.57	1.51	1.55	1.48
Na	1.60	2.11	2.66	1.39	1.22	1.64	1.63
K	7.83	7.59	7.35	7.35	6.87	7.17	6.51
Mn, mg/kg	78.8	78.5	77.9	78.5	78.0	78.2	77.5
Cu	9.00	9.53	10.06	8.46	7.92	8.41	7.82
Fe	335.9	372.2	390.7	382.4	418.0	342.0	344.2
Zn	59.0	59.6	60.2	59.3	59.5	58.0	56.8
Asp, g/kg	19.07	18.65	18.24	18.91	18.75	17.87	16.66
Thr	8.00	8.09	8.19	7.87	7.74	7.58	7.16
Ser	10.39	10.31	10.23	10.23	10.06	9.96	9.52
Glu	46.90	45.77	44.67	45.20	43.45	45.23	43.52
Pro	12.81	13.01	13.23	12.88	12.94	14.55	16.28
Cys	3.78	3.71	3.70	3.68	3.63	3.48	3.23
Gly	8.33	8.20	8.08	8.34	8.36	11.40	14.47
Ala	10.55	10.49	10.44	10.82	11.09	11.10	11.64
Val	9.82	9.88	9.95	10.16	10.50	9.34	8.87
Met	5.55	5.41	5.36	5.46	5.47	5.59	5.63
Ile	9.07	8.76	8.46	8.41	7.74	8.38	7.69
Leu	16.83	16.94	17.06	17.44	18.06	15.94	15.04
Tyr	6.86	6.87	6.88	6.63	6.40	6.35	5.84
Phe	11.39	11.29	11.20	11.47	11.55	10.74	10.09
His	5.59	5.69	5.80	6.20	6.80	5.31	5.02
Lys	11.81	11.53	11.35	11.38	11.63	10.73	9.94
Arg	14.35	14.03	13.72	13.83	13.30	14.53	14.71
Try	2.69	2.66	2.63	2.70	2.72	2.44	2.19
Total	213.7	211.3	209.2	211.6	210.2	210.5	207.5

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All dietary components were determined chemically.

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Table 5. *Body weight, blood parameters and bone characteristics of chickens¹*

Item	Body weight, kg	Average feed intake, g/head/d in period 2	Blood parameters			Bone parameters					
			Haemato-crit %	Haemoglo-bin g%	Immuno-globulins G mg/ml	Length (average for both bones), cm		crude ash g/kg	in 1 kg of crude ash		
						femur	tibia		Ca, g	P, g	Zn, mg
Feeding groups treatments²											
Control	1.05 ^A	40.0	26.1	15.8	2.77 ^a	5.61 ^A	7.45 ^A	462	295.1	171.0	510.5
Plasma 20 g/kg	0.91 ^A	40.7	26.1	17.1	2.71	5.55 ^A	7.36 ^A	467	289.1	172.7	564.0
Plasma 40 g/kg	0.95 ^A	35.9	26.0	18.2 ^a	2.80 ^a	5.49	7.45 ^A	474	290.8	167.3	561.6
Blood cells 20 g/kg	0.74 ^B	40.4	25.0	17.6	1.69 ^b	5.38	7.08	456	306.1	168.7	546.3
Blood cells 40 g/kg	0.92 ^A	33.5	27.0	16.0	2.57	5.50	7.23 ^A	470	282.1	171.4	620.2
Bone hydrolysate 20 g/kg	0.67 ^B	32.0	25.6	12.9 ^b	2.24	5.26	7.00	468	304.8	167.1	578.6
Bone hydrolysate 40 g/kg	0.61 ^B	38.7	27.5	16.0	2.05 ^b	5.08 ^B	6.65 ^B	458	301.1	169.5	569.6
SEM	0.02	1.10	0.37	0.51	0.11	0.04	0.05	2.50	2.99	0.77	10.44
<i>P</i> -value for feeding group	0.000	0.169	0.654	0.134	0.069	0.001	0.000	0.398	0.274	0.377	0.192
Type of animal meal											
Control	1.05 ^A	40.0	26.1	15.8	2.77 ^a	5.61 ^A	7.45 ^A	462	295.1	171.0	510.5
Plasma	0.93 ^{AB}	38.3	26.1	17.6	2.76 ^a	5.52 ^A	7.41 ^A	471	290.0	170.0	562.8
Blood cells	0.83 ^B	37.0	26.0	16.8	2.13 ^b	5.44 ^b	7.15 ^A	463	294.1	170.1	583.3
Bone hydrolysate	0.64 ^C	35.3	26.6	14.5	2.15 ^b	5.17 ^{Ba}	6.83 ^B	463	303.0	168.3	574.1
<i>P</i> -value for type of animal meal	0.000	0.561	0.827	0.052	0.059	0.001	0.000	0.393	0.244	0.616	0.748
Incl. rate of animal meal											
Control	1.05	40.0	26.1	15.8	2.77	5.61	7.45	462	295.1	171.0	510.5
20 g/kg	0.77	37.7	25.6	15.9	2.22	5.40	7.15	464	300.0	169.5	563.0
40 g/kg	0.83	36.0	26.8	16.8	2.48	5.35	7.11	468	291.3	169.4	583.8
<i>P</i> -value for incl. rate of animal meals	0.055	0.465	0.131	0.397	0.279	0.574	0.605	0.456	0.180	0.943	0.347
Interaction type x incl. rate of animal meal	0.004	0.043	0.494	0.208	0.175	0.228	0.013	0.176	0.228	0.088	0.240

681 Differences in column marked with a, b: significant by $P < 0.05$; marked with A, B: significant by $P < 0.01$.682 ¹Means for individuals.683 ²Means for 4 replications (cages) with 9-10 birds.

Table 6. Retention of mineral elements in chickens¹

Item	Ca	P	Na	K	Cu	Zn	Fe	Mn
Feeding groups - treatments								
Control	0.42 ^a	0.40	0.71	0.75	0.64 ^A	0.16	0.12	0.11
Plasma 20 g/kg	0.56 ^b	0.51	0.73	0.79	0.77 ^B	0.24	0.16	0.17
Plasma 40 g/kg	0.52	0.49	0.75	0.79	0.72	0.34	0.17	0.20
Blood cells 20 g/kg	0.53 ^b	0.46	0.77	0.80	0.63 ^A	0.26	0.18	0.28
Blood cells 40 g/kg	0.49	0.38	0.69	0.77	0.76 ^B	0.36	0.23	0.10
Bone hydrolysate 20 g/kg	0.46 ^a	0.42	0.77	0.77	0.68	0.22	0.11	0.22
Bone hydrolysate 40 g/kg	0.51	0.45	0.78	0.80	0.66	0.22	0.15	0.09
SEM	0.01	0.01	0.01	0.007	0.01	0.02	0.02	0.02
P-value for feeding groups	0.063	0.122	0.089	0.464	0.010	0.081	0.896	0.252
Type of animal meal								
Control	0.42	0.40	0.71	0.75	0.64 ^a	0.16	0.12	0.11
Plasma	0.54	0.50	0.74	0.79	0.74 ^b	0.29	0.16	0.18
Blood cells	0.51	0.42	0.73	0.79	0.69	0.31	0.20	0.19
Bone hydrolysate	0.48	0.43	0.77	0.78	0.67	0.22	0.13	0.15
P-value for type of animal meal	0.182	0.063	0.199	0.893	0.046	0.172	0.534	0.813
Incl. rate of animal meal								
Control	0.42	0.40	0.71	0.75	0.64	0.16	0.12	0.11
20 g/kg	0.52	0.46	0.76	0.79	0.69	0.24	0.15	0.22
40 g/kg	0.51	0.44	0.74	0.79	0.71	0.31	0.18	0.13
P-value for incl. rate of animal meal	0.706	0.358	0.488	0.972	0.309	0.099	0.519	0.077
Interaction type x incl. rate animal meals	0.253	0.306	0.066	0.332	0.015	0.409	0.974	0.194

Differences in column marked with a,b significant by $P < 0.05$

Differences in column marked with A,B significant by $P < 0.01$

¹n for each treatment, 4 replications (cages), excreta samples pooled from 9-10 birds.

Table 7. Apparent ileal digestibility of amino acids in diets (determined by indicator method)¹

Item	Feeding group									Type of animal meal					Incl. rate of animal meal				Interaction kind x share
	Control	Plasma 20 g/kg	Plasma 40 g/kg	Blood cells 20 g/kg	Blood cells 40 g/kg	Bone hydrolysate 20 g/kg	Bone hydrolysate 40 g/kg	SEM	P – value Feeding groups	Control	Plasma	Blood cells	Bone hydrolysate	P – value Type of animal meal	Control	20 g/kg	40 g/kg	P – value Incl. rate of animal meal	
Asp	0.77 ^{Aa}	0.85 ^B	0.82 ^b	0.78 ^A	0.83 ^{Bc}	0.81	0.81	0.000	0.000	0.77 ^A	0.83 ^{Ba}	0.81 ^b	0.81 ^b	0.015	0.77	0.81	0.82	0.477	0.002
Thr	0.78	0.81 ^a	0.80	0.77 ^b	0.80	0.79	0.77	0.005	0.037	0.78	0.80	0.78	0.78	0.101	0.78	0.79	0.79	0.943	0.019
Ser	0.82 ^{Aa}	0.88 ^B	0.86 ^b	0.82 ^A	0.86	0.85	0.85	0.004	0.000	0.82 ^A	0.87 ^B	0.84	0.85	0.002	0.82	0.85	0.85	0.432	0.009
Glu	0.89 ^a	0.91 ^b	0.90 ^b	0.89 ^a	0.91 ^b	0.90 ^b	0.90 ^b	0.004	0.011	0.89	0.91	0.90	0.90	0.213	0.89	0.90	0.90	0.298	0.050
Pro	0.82 ^A	0.86	0.85	0.81 ^A	0.85	0.86 ^B	0.88 ^B	0.002	0.000	0.82 ^A	0.85	0.83 ^A	0.87 ^B	0.000	0.82	0.85	0.86	0.144	0.033
Gly	0.73 ^A	0.80	0.79	0.74	0.78	0.84 ^B	0.88 ^C	0.004	0.000	0.73	0.80	0.76	0.86	0.000	0.73	0.79	0.82	0.076	0.136
Ala	0.80 ^A	0.84	0.84	0.81	0.86 ^B	0.84 ^B	0.86 ^C	0.008	0.000	0.80	0.84	0.84	0.85	0.130	0.80 ^A	0.83	0.85 ^B	0.007	0.016
Cys	0.62	0.62	0.67	0.55 ^A	0.70 ^B	0.67	0.61	0.004	0.006	0.62	0.64	0.63	0.64	0.869	0.62	0.61 ^a	0.66 ^b	0.030	0.001
Val	0.79 ^A	0.82	0.82	0.82	0.86 ^B	0.82	0.80	0.011	0.001	0.79 ^a	0.82	0.84 ^b	0.81	0.037	0.79	0.82	0.83	0.302	0.029
Met	0.82 ^A	0.80	0.83 ^A	0.74 ^B	0.87	0.84 ^A	0.83 ^A	0.004	0.000	0.82	0.81	0.80 ^b	0.84 ^a	0.020	0.82	0.79 ^A	0.84 ^B	0.000	0.000
Ile	0.81	0.83	0.82	0.81	0.84	0.82	0.81	0.006	0.365	0.81	0.82	0.82	0.82	0.845	0.81	0.82	0.82	0.875	0.109
Leu	0.84 ^A	0.86	0.86	0.85	0.89 ^B	0.85	0.85	0.004	0.004	0.84	0.86	0.87	0.85	0.077	0.84	0.86	0.86	0.229	0.024
Tyr	0.72 ^A	0.80 ^B	0.77	0.71 ^A	0.74	0.76	0.75	0.004	0.000	0.72 ^A	0.79 ^B	0.72 ^A	0.75	0.000	0.72	0.76	0.75	0.635	0.148
Phe	0.84 ^a	0.86	0.86	0.85	0.88 ^b	0.85	0.84	0.006	0.021	0.84	0.86	0.87	0.85	0.145	0.84	0.86	0.86	0.395	0.024
His	0.82 ^A	0.85	0.86	0.85	0.89 ^B	0.84 ^A	0.83 ^A	0.003	0.000	0.82 ^A	0.85	0.87 ^B	0.84	0.000	0.82	0.85	0.86	0.151	0.011
Lys	0.86 ^a	0.88	0.87	0.87	0.89 ^b	0.87	0.87	0.004	0.040	0.86	0.87	0.88	0.87	0.346	0.86	0.87	0.88	0.338	0.048
Arg	0.87 ^a	0.88 ^a	0.88	0.87 ^a	0.88 ^a	0.89 ^b	0.89 ^b	0.003	0.034	0.87	0.88	0.87	0.89	0.144	0.87	0.88	0.88	0.386	0.097
Try	0.67	0.67	0.70	0.65	0.71	0.71	0.62	0.002	0.160	0.67	0.69	0.68	0.66	0.655	0.67	0.68	0.68	0.986	0.017
Total	0.82 ^A	0.86	0.85	0.82 ^A	0.86 ^B	0.85	0.85	0.010	0.001	0.82	0.85	0.84	0.85	0.281	0.82	0.84	0.85	0.146	0.011

Differences in rows marked with a, b, c: significant by $P < 0.05$; marked with A, B, C: significant by $P < 0.01$.

¹n for each treatment, 4 replications (cages), ileal content samples pooled from 3 birds per cage).

Table 8. Amino acid contents in spray-dried porcine blood plasma, g/kg (data from literature)

Amino acids	Hansen <i>et al.</i> , 1993	NRC, 1998	Degussa, 2006	Duda, 2007	Jamroz <i>et al.</i> , 2008*	Fish meal*
Crude protein	670	780	770	700	704 ± 52.4	706
Asp	NDA	NDA	76.0	71.0	69.4 ± 4.7	71.7
Thr	40.4	47.2	47.0	43.0	38.8 ± 4.8	36.5
Ser	NDA	NDA	45.0	42.0	40.0 ± 4.2	33.6
Glu	NDA	NDA	113.0	106.0	83.2 ± 8.7	98.8
Pro	NDA	NDA	126.0	115.0	39.0 ± 4.2	30.9
Cys	22.9	26.3	27.0	25.0	17.9 ± 3.4	5.8
Gly	NDA	NDA	29.0	27.0	26.5 ± 18.0	70.0
Ala	NDA	NDA	40.0	38.0	40.5 ± 3.1	49.7
Val	46.6	49.4	51.0	48.0	44.6 ± 3.7	25.3
Met	4.8	7.50	7.0	6.0	5.5 ± 1.2	24.2
Ile	26.0	27.1	28.0	26.0	20.3 ± 3.5	18.1
Leu	67.0	76.1	75.0	70.0	68.6 ± 5.1	58.9
Tyr	36.4	35.3	35.0	33.0	29.2 ± 3.9	19.5
Phen	40.0	44.2	44.0	41.0	39.1 ± 2.7	25.9
His	23.5	25.5	27.0	25.0	26.8 ± 3.0	26.2
Lys	61.0	68.4	65.0	61.0	59.4 ± 5.2	76.7
Arg	42.3	45.5	45.0	42.0	44.7 ± 4.3	52.4
Try	13.8	13.6	13.0	12.0	10.0 ± 1.3	7.2
Met+Cys	27.7	25.0	34.0	31.0	23.4 ± 4.4	30.0

* means from own laboratory (Wroclaw University of Environmental and Life Sciences).

NDA = no data available.

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715**Table 9.** Amino acid content in spray-dried porcine blood cells, g/kg (data from literature)

Amino acid	Kats <i>et al.</i> 1994	Degussa, 1995	Amino acid content according to:				Fish meal UP, 2006	
			USA - Degussa, 1995	NRC, 1998	De Rouchey <i>et al.</i> , 2002	Degussa, 2006		Jamroz <i>et al.</i> , 2008
Crude protein	898.0	905.0	933.0	920.0	921.0	920.0	830.0 ± 70.6	706
Asp	NDA	100.5	105.5	NDA	NDA	114.0	98.1 ± 10.5	71.7
Thr	36.2	38.9	35.9	33.8	39.8	36.0	30.6 ± 1.8	36.5
Ser	NDA	44.4	43.6	NDA	NDA	44.0	40.3 ± 2.7	33.6
Glu	NDA	74.2	75.5	NDA	NDA	87.0	68.7 ± 2.8	98.8
Cys	10.1	6.2	6.2	6.1	6.4	6.0	9.4 ± 2.1	5.8
Gly	NDA	41.3	42.1	NDA	NDA	47.0	39.2 ± 4.7	70.0
Ala	NDA	76.3	75.2	NDA	NDA	76.0	64.9 ± 8.2	49.7
Val	69.1	81.1	79.5	85.0	85.0	92.0	65.9 ± 8.4	25.3
Met	8.8	11.8	10.3	8.1	12.0	8.0	6.5 ± 1.1	24.2
Ile	9.1	4.5	5.5	4.9	3.7	6.0	7.3 ± 2.2	18.1
Leu	99.6	121.3	124.1	127.0	126.0	134.0	105.7 ± 12.2	58.9
Tyr	24.8	21.2	NDA	21.4	23.3	22.0	19.5 ± 2.0	19.5
Phe	54.0	65.5	NDA	66.9	69.3	71.0	55.4 ± 5.8	25.9
His	49.1	63.2	NDA	69.9	59.8	75.0	57.9 ± 8.6	26.2
Lys	75.3	83.3	84.3	85.1	87.4	90.0	75.9 ± 6.7	76.7
Arg	34.5	34.1	36.6	37.7	31.5	40.0	42.3 ± 3.8	52.4
Try	16.7	15.9	NDA	13.7	17.8	12.0	10.2 ± 4.6	7.2
Met + Cys	18.9	18.0	16.5	NDA	18.4	14.0	15.9 ± 2.8	30.0

716 NDA = no data available.

Table 10. Amino acid contents in dried porcine bone protein hydrolysate, g/kg (data from literature)

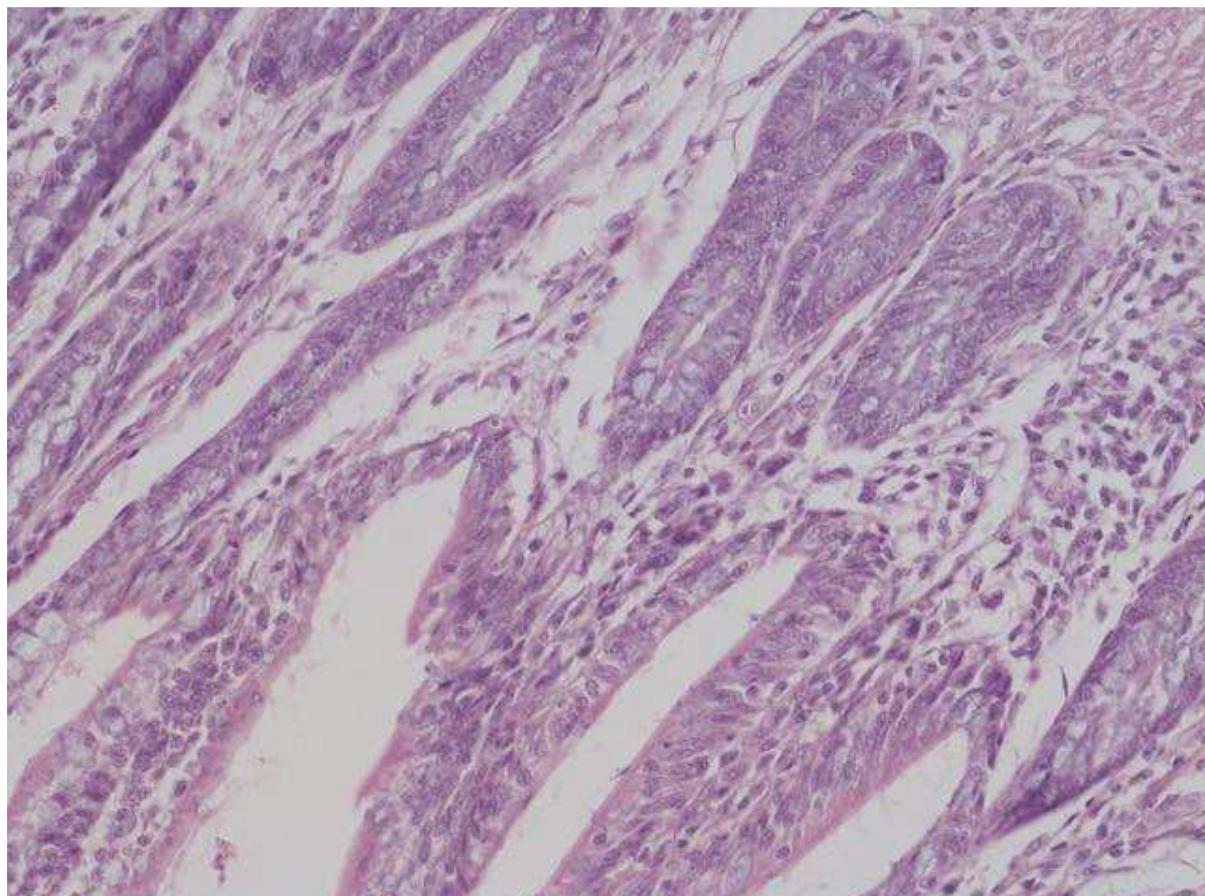
Amino acids	Degussa, 1995	Jamroz <i>et al.</i> , 2008	Fish meal*
Crude protein	742	785 ± 54.2	706
Asp	52.6	55.0 ± 7.0	71.7
Thr	16.6	17.4 ± 1.8	36.5
Ser	25.4	29.3 ± 2.3	33.6
Glu	88.3	82.3 ± 6.7	98.8
Pro	96.0	116.0 ± 8.9	30.9
Cys	1.9	0.4 ± 0.3	5.8
Gly	146.6	192.3 ± 12.0	70.0
Ala	71.9	79.1 ± 5.2	49.7
Val	21.9	21.4 ± 2.9	25.3
Met	7.9	5.8 ± 3.6	24.2
Ile	13.2	10.2 ± 1.4	18.1
Leu	28.9	28.4 ± 2.3	58.9
Tyr	NDA	7.1 ± 1.8	19.5
Phe	19.6	19.5 ± 2.0	25.9
His	14.5	9.8 ± 1.1	26.2
Lys	33.7	33.0 ± 3.2	76.7
Arg	57.9	77.2 ± 8.8	52.4
Try	1.5	0.6 ± 0.1	7.2
Met+Cys	9.8	6.0 ± 3.8	30.0

* means from own laboratory (Wroclaw University of Environmental and Life Sciences).

NDA = no data available.

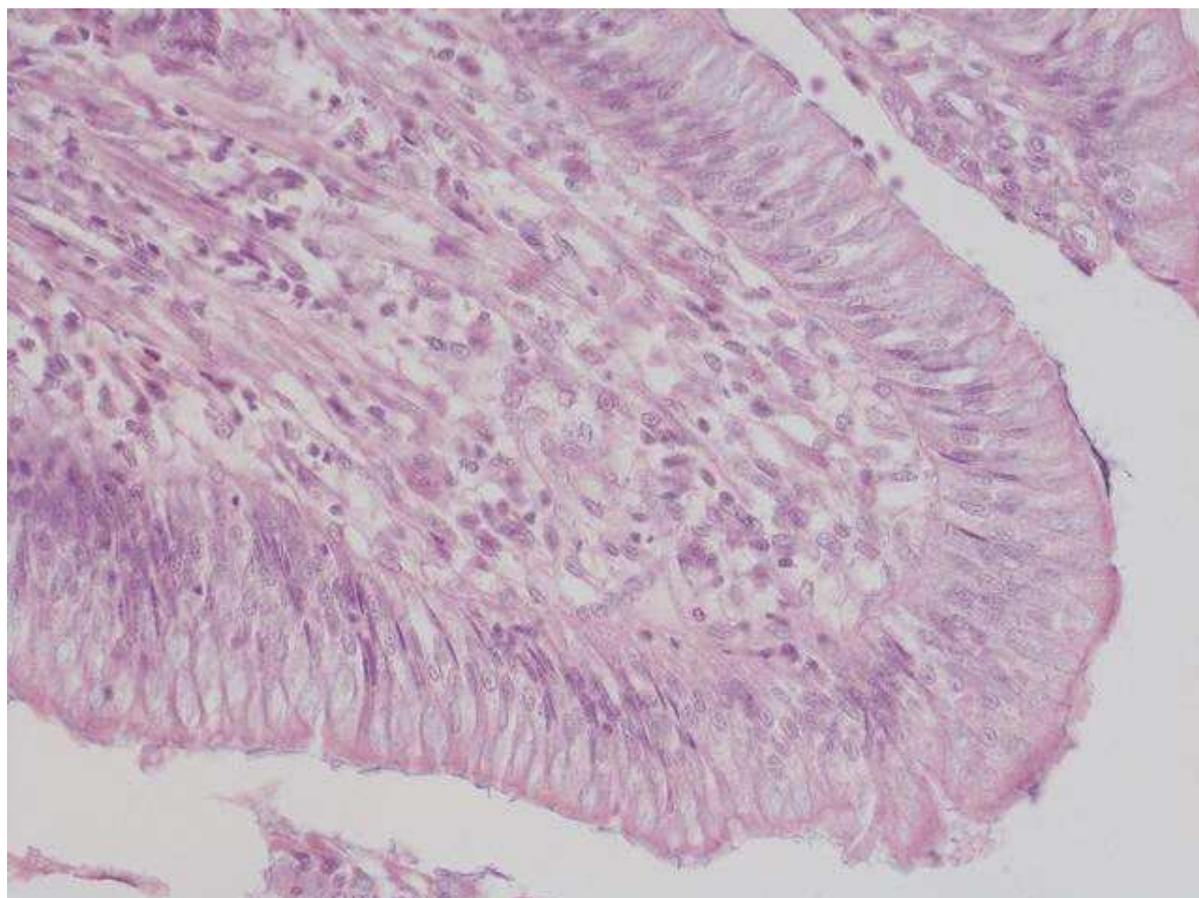
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Figure 1



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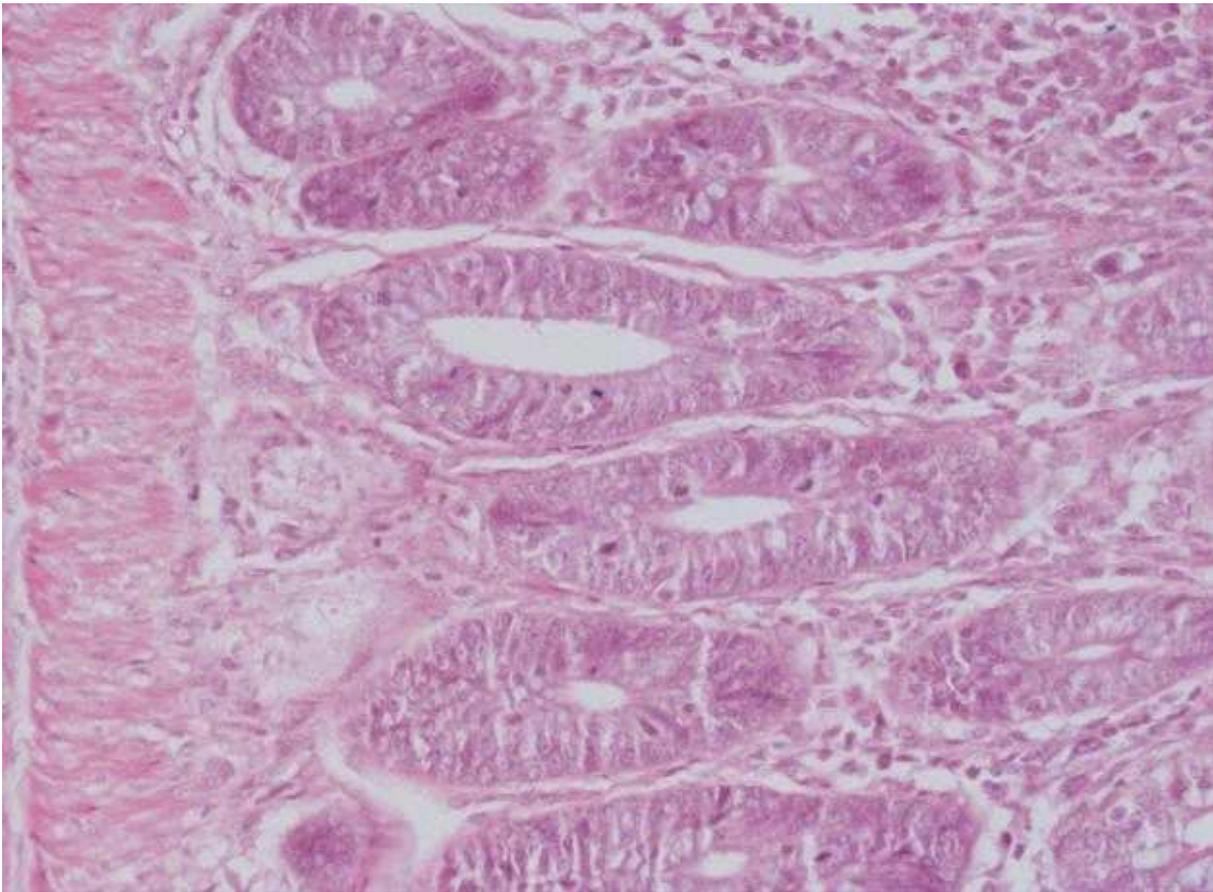
Figure 2



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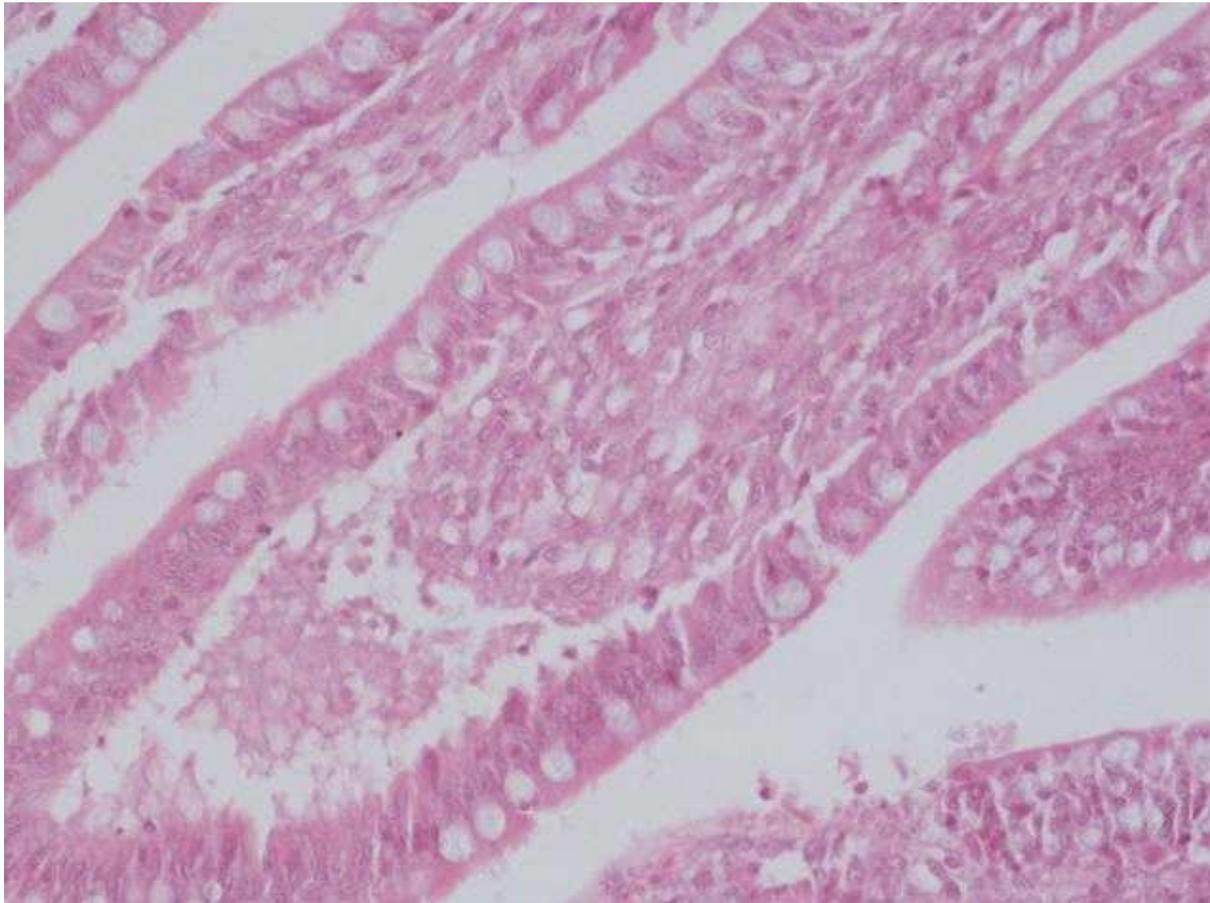
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Figure 3



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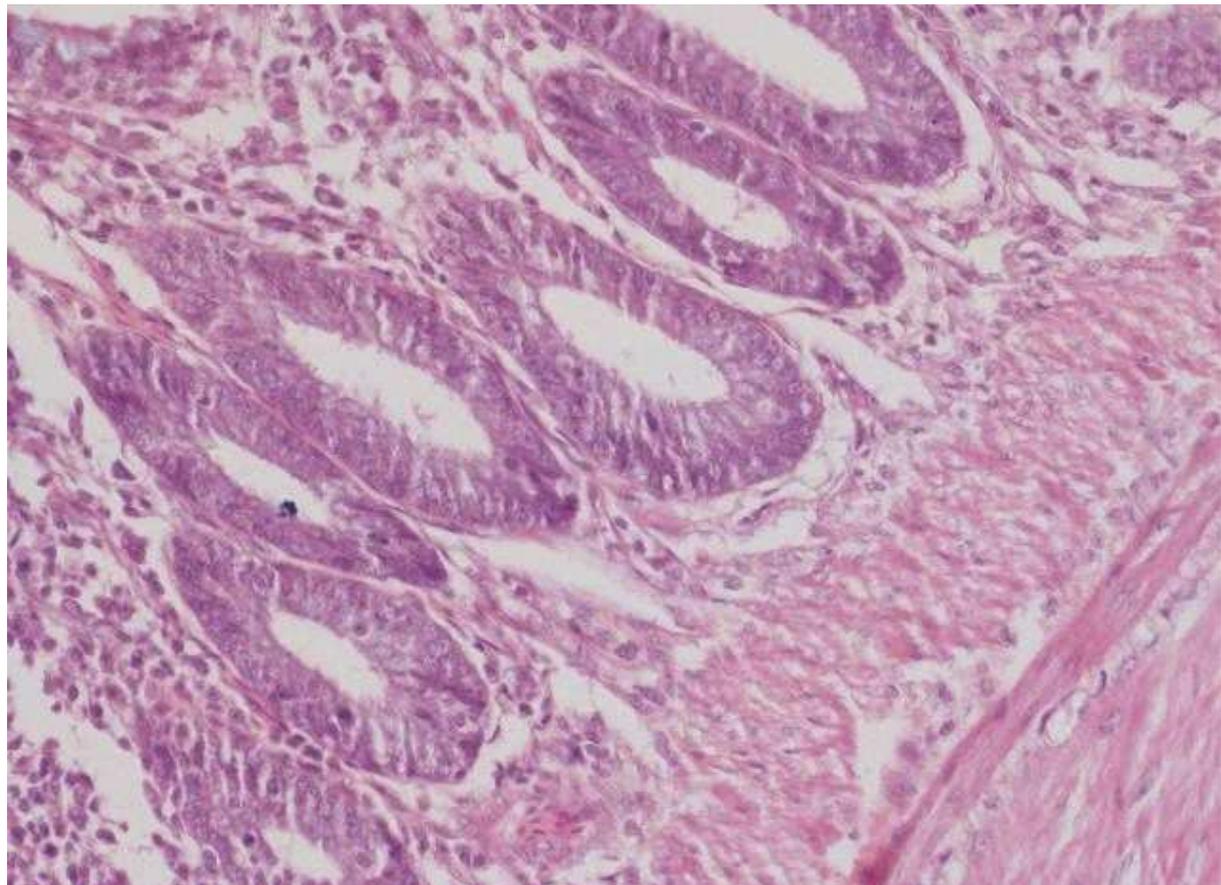
Figure 4



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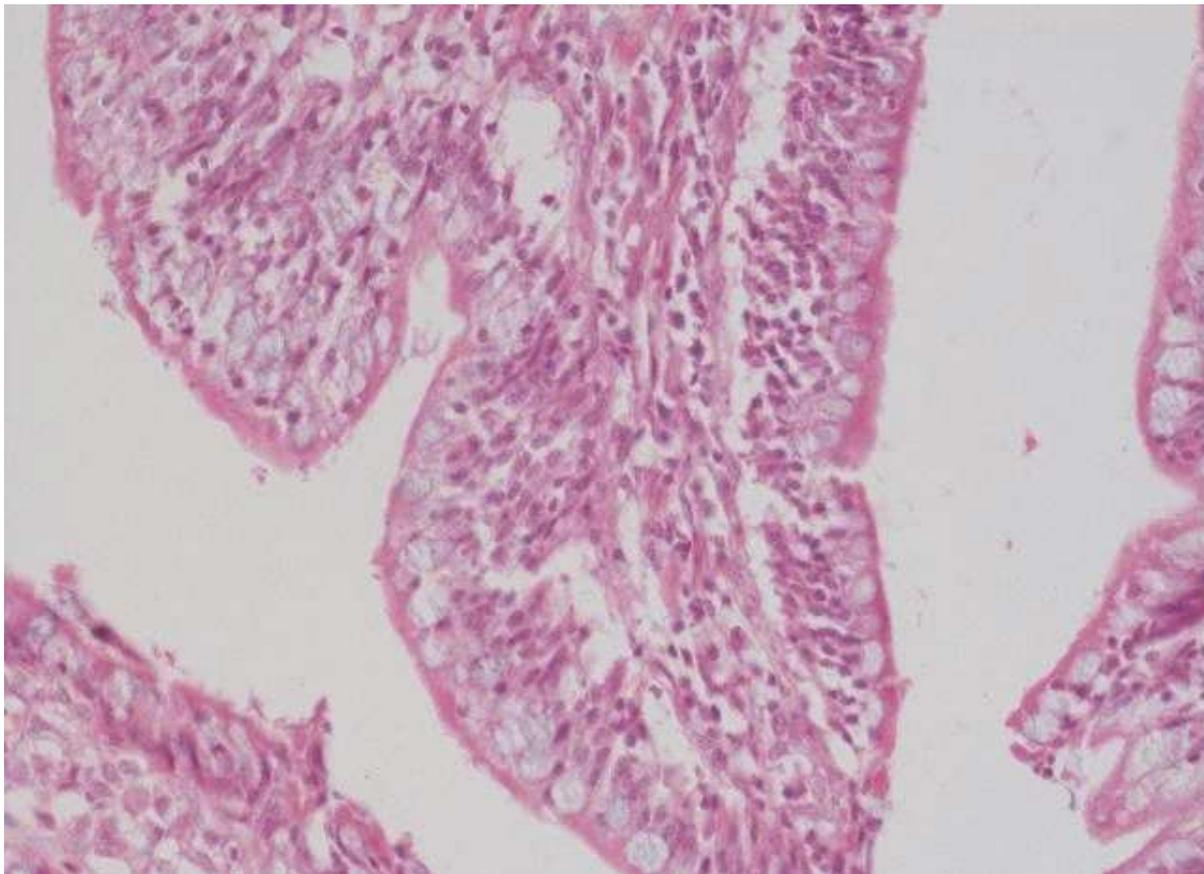
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Figure 5



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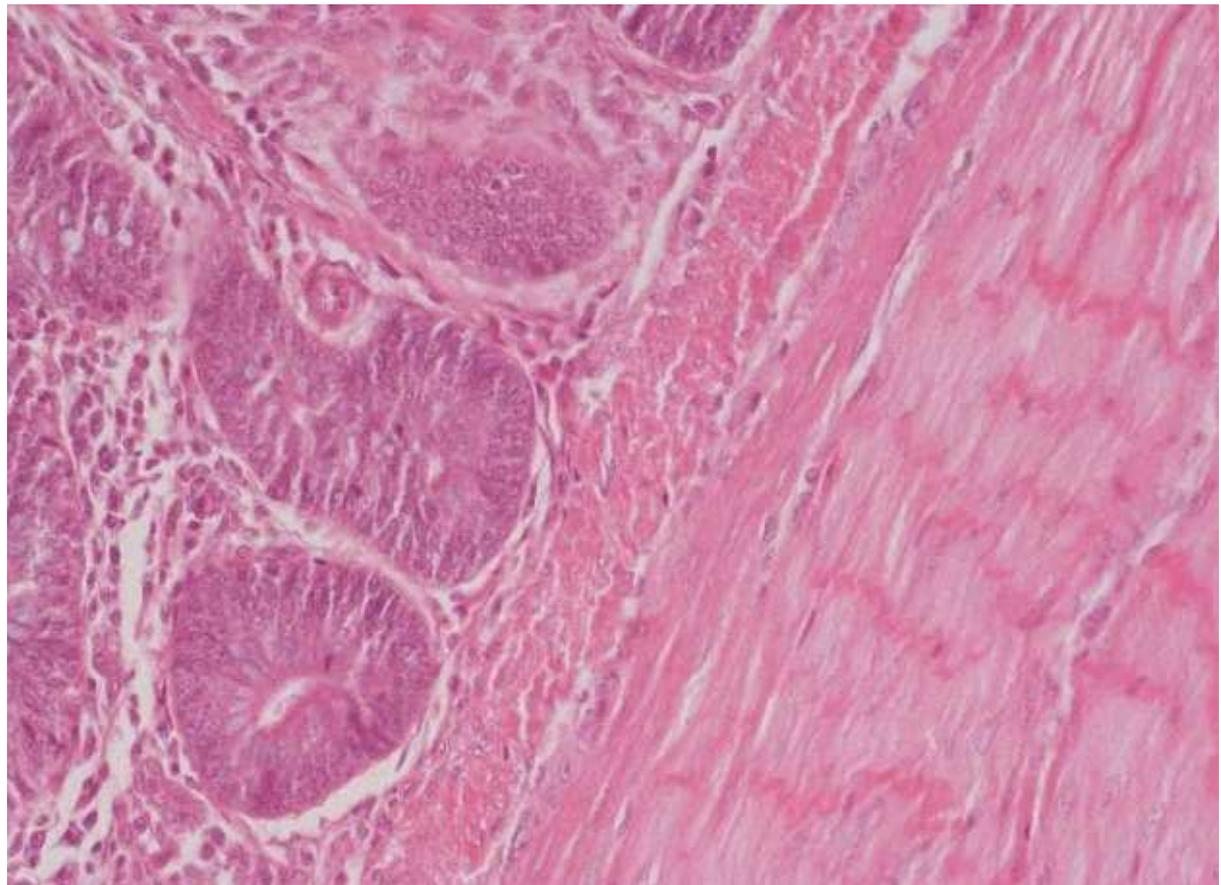
Figure 6



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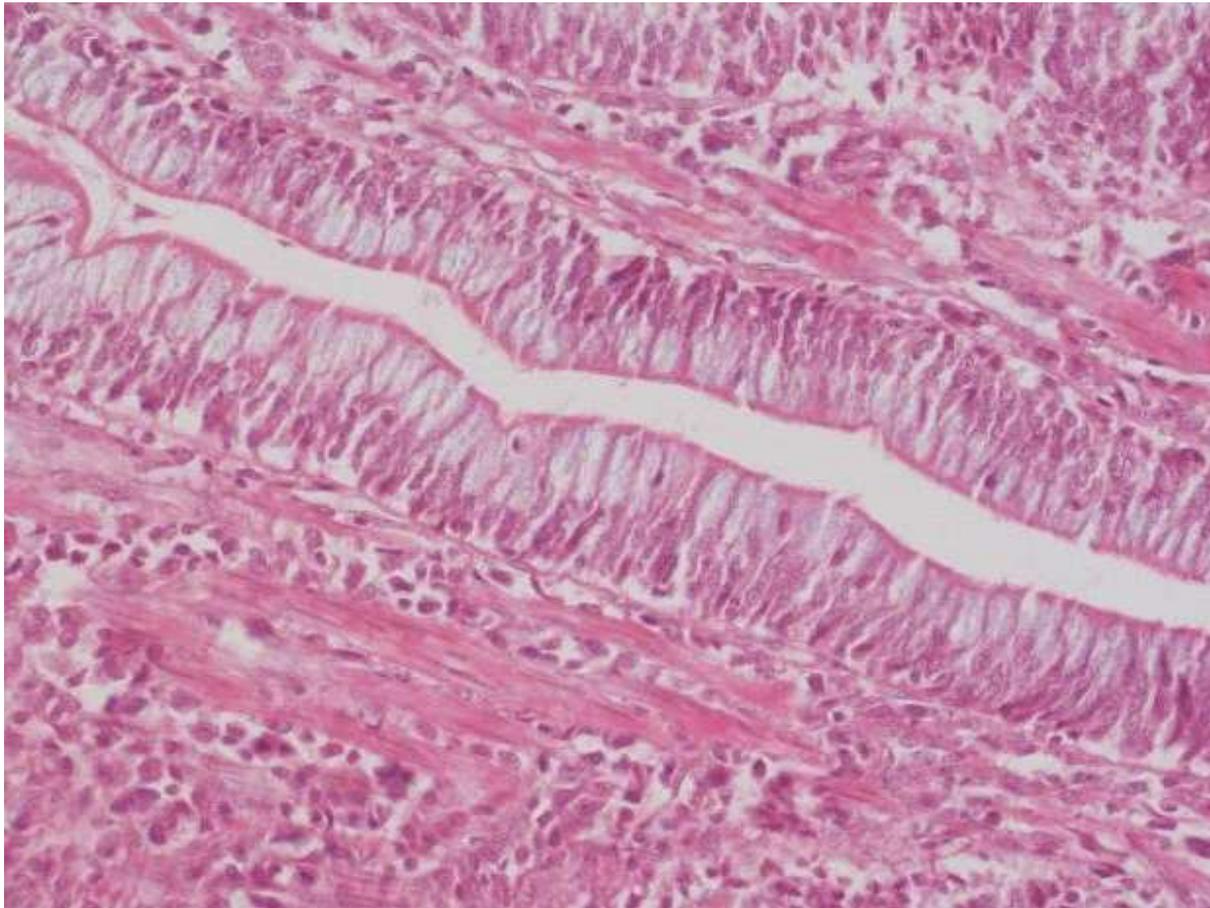
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Figure 7



Review Only

Figure 8



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