

KORESPONDENSI ARTIKEL
JURNAL KEDOKTERAN HEWAN - *INDONESIAN JOURNAL OF VETERINARY*
SCIENCES
VOL. 15 NO. 3 SEPTEMBER 2021
URL : <https://jurnal.usk.ac.id/JKH/article/view/19774>

**The Effect of Black Cumin (*Nigella sativa*)
Supplementation Through Drinking Water on The
Histology of Small Intestine and Large Intestine of
Broiler Chickens**



Rendy Sirat <m.mirandy@fp.unila.ac.id>

[J.KED.HEWAN] Submission Acknowledgement

1 message

Drh. T. Armansyah TR, M.Kes. <jurnal@unsyiah.ac.id>

Sun, Jan 31, 2021 at 10:14 AM

To: "drh., M.Sc. Muhammad Mirandy Pratama Sirat" <m.mirandy@fp.unila.ac.id>

drh., M.Sc. Muhammad Mirandy Pratama Sirat:

Thank you for submitting the manuscript, "EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER" to Jurnal Kedokteran Hewan - Indonesian Journal of Veterinary Sciences. With the online journal management system that we are using, you will be able to track its progress through the editorial process by logging in to the journal web site:

Manuscript URL: <http://jurnal.unsyiah.ac.id/JKH/author/submission/19774>

Username: rendy

If you have any questions, please contact me. Thank you for considering this journal as a venue for your work.

Drh. T. Armansyah TR, M.Kes.

Jurnal Kedokteran Hewan - Indonesian Journal of Veterinary Sciences

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP

Chief Editor E-mail: siregar@unsyiah.ac.idWebsite: <http://jurnal.unsyiah.ac.id/JKH>e-mail : jkh@unsyiah.ac.id

1 **EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH**
2 **DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND**
3 **LARGE INTESTINE OF BROILER**

4
5 **¹Madi Hartono, ¹Purnama Edy Santosa, ¹Ratna Ermawati, ¹Muhammad Mirandy**
6 **Pratama Sirat***

7
8 ¹Department of Animal Husbandry, Faculty of Agriculture, University of Lampung,
9 Indonesia

10 Jl. Soemantri Brodjonegoro No. 1, Gedong Meneng, Rajabasa, Bandar Lampung, Lampung,
11 Indonesia 35145

12 *Corresponding author: m.mirandy@fp.unila.ac.id

13
14 **ABSTRACT**

15
16 This study aimed to determine the effect of Black Cumin (*Nigella sativa*)
17 supplementation through drinking water on the histology of small intestine and large intestine
18 of broiler. The research was conducted from April September 2020 in cage facility of Integrated
19 Field Laboratory, Faculty of Agriculture, University of Lampung. This research was using a
20 completely randomized design (CRD) with four treatment groups and three replications (five
21 heads per replication) with a total of 60 male broilers. The treatment were drinking water
22 without Black Cumin (P0, control); drinking water with Black Cumin 36 mg/kg BW/day (P1);
23 72 mg/kg BW/day (P2); and 144 mg/kg BW/day (P3). Three broilers from each group were
24 randomly necropsed at 31st days old, and samples of the small intestine (duodenum, jejunum,
25 ileum) and large intestine were fixed with 10% formalin solution and sent to the Lampung

26 Disease Investigation Center for histological preparations. The observation of preparations was
27 carried out microscopically using the Leica DM500® Binocular Microscope to accurately
28 calculate various parameter sizes. The results were analyze descriptively. The conclusion of
29 this study was the supplementation of Black Cumin 36 mg/kg BW/day through drinking water
30 could increase the average sizes of villi height, villi apex width, basal villi width, villi area, and
31 gland diameter of small intestine (duodenum, jejunum, ileum) and large intestine of broiler.

32

33 Key words : black cumin, broiler, Leica DM500 microscope, large intestine, small intestine.

34

35

ABSTRAK

36

37 *Penelitian ini bertujuan untuk mengetahui pengaruh pengaruh suplementasi Jintan*
38 *Hitam (Nigella sativa) melalui air minum terhadap histologi usus halus dan usus besar broiler.*
39 *Penelitian dilakukan pada April-September 2020 di unit kandang Laboratorium Lapang*
40 *Terpadu, Fakultas Pertanian, Universitas Lampung. Penelitian bersifat eksperimental*
41 *menggunakan Rancangan Acak Lengkap (RAL) dengan empat kelompok perlakuan dan tiga*
42 *ulangan (lima ekor tiap ulangan) sehingga total 60 ekor broiler jantan. Perlakuan yang*
43 *diberikan yaitu pemberian air minum tanpa Jintan Hitam (P0, kontrol); air minum dengan*
44 *Jintan Hitam 36 mg/kg BB/hari (P1); 72 mg/kg BB/hari (P2); dan 144 mg/kg BB/hari (P3).*
45 *Tiga ekor dari tiap kelompok secara acak dinekropsi pada hari ke-31 dan diambil sampel*
46 *organ usus halus (duodenum, jejunum, ileum) dan usus besar (colon) kemudian difiksasi*
47 *dengan larutan formalin 10% dan dikirim ke Balai Veteriner Lampung untuk pembuatan*
48 *preparat histologi. Pengamatan preparat dilakukan secara mikroskopis menggunakan*
49 *Mikroskop Binokuler Leica DM500® untuk menghitung berbagai ukuran parameter secara*
50 *akurat. Analsis hasil dilakukan secara deskriptif. Kesimpulan penelitian ini yaitu suplementasi*

51 *Jintan Hitam (Nigella sativa) 36 mg/kg BB/hari meningkatkan rata-rata ukuran tinggi vili,*
52 *lebar apeks vili, lebar basal vili, luas vili, dan diameter kelenjar pada organ saluran*
53 *pencernaan yaitu usus halus (duodenum, jejunum, ileum) dan usus besar broiler.*

54

55 *Kata kunci : broiler, jintan hitam, mikroskop Leica DM500, usus besar, usus halus.*

56

57

58 **INTRODUCTION**

59

60 The development of poultry farming in Indonesia is increasing. Broilers are one of the
61 fastest growing poultry. The relatively short period of time and the relatively lower
62 maintenance costs compared to ruminants make breeders prefer to cultivate broilers. In 2018,
63 the broiler population in Indonesia reached 1.89 billion heads and an increase of 2.26% from
64 the broiler population in 2017 of 1.85 billion heads (BPS, 2019).

65 Disease is a serious obstacle in the broiler farming industry. The high incidence of disease
66 can cause a decrease in productivity and even death of livestock which causes significant losses
67 for breeders. Administration of antibiotics in the livestock industry is used for the treatment of
68 livestock so as to reduce the risk of death and restore the condition of the livestock to health,
69 however, giving antibiotics for a long period of time can cause residual buildup which has
70 negative effects if consumed by humans.

71 The use of antibiotics needs to be reduced to prevent negative effects by providing natural
72 ingredients as immunomodulators. Immunomodulators can be defined as biological or
73 synthetic substances that can stimulate the innate immune system, adaptive or both. One of the
74 herbs that can act as an immunomodulator is Black Cumin (*Nigella sativa*). *Nigella sativa* is
75 a plant that has the potential as an immunostimulant that can stimulate and strengthen the

76 system by increasing the number, quality and activity of the body's immune cells (Hendrik,
77 2009). *Nigella sativa* contains *thymoquinone*, saponins, zinc or zinc, *alpha-linolenic acid*
78 (Omega 3) and *linoleic acid* (Omega 6) which functions in cell formation, maintains the
79 immune system, and helps in the process of blood formation (Yusuf, 2014).

80 The solution needed for the above problems is to examine the effect of black cumin
81 (*Nigella sativa*) supplementation through histological studies of the digestive organs of broilers
82 which are expected to have the potential to increase the size of villi height, apex villi width,
83 basal villi width, villi area, crypt depth and gland diameter of small intestine (duodenum,
84 jejunum, ileum) and large intestine.

85

86 MATERIALS AND METHODS

87

88 Materials

89 This study used broiler cages, sprayer for cage disinfection, bamboo to make 12 cage
90 plots, plastic tarpaulin for curtains, newspaper and used husks as litter, 12 bulbs of 15 bulbs
91 watt as a heating source for the area. *brooding*, *a hanging feeder* 12 pieces, *chick feeder tray* 12
92 pieces, 12 pieces of chicken drinking places; 1 bucket, 1 *hand spray*, 1 water tray for *dipping*,
93 1 electric scale, *thermohygrometer* for measuring temperature and humidity, sack and plastic.
94 Organ sampling equipment, namely necropsy equipment, *object glass*, *cover glass*,
95 *refrigerator*, microtome, light microscope, camera technology and *software* Optilab® along
96 with a laptop for taking tissue images and measuring the parameters of each organ.

97 Materials used in the study were 60 Day Old Chicks (DOC) male broiler Cobb CP 707
98 strains kept for 30 days, rations, drinking water, extract of Black Cumin (*Nigella sativa*),
99 vaccines of Newcastle Disease (ND), Avian Influenza (AI) and Infectious Bursal Disease
100 (IBD), 10% formalin solution.

101

102 **Methods**

103 This research carried out for 6 months (April - September 2020) in the Integrated Field
104 Laboratory enclosure unit, Faculty of Agriculture, University of Lampung. This research was
105 experimental in nature using a completely randomized design (CRD) with four treatment
106 groups and three replications (five heads per replication) so a total of 60 male broilers. The
107 treatment dose was according to the broiler body weight, namely 1) drinking water without
108 black cumin (P0, control); drinking water with Jintan Hitam 36 mg / kg BW / day (P1); drinking
109 water with Jintan Hitam 72 mg / kg BW / day (P2); and drinking water with cumin 144 mg /
110 kg BW / day (P3). On the 31st day, three from each group were randomly necropsed and
111 samples of the small intestine (duodenum, jejunum, ileum) and large intestine were then fixed
112 with 10% formalin solution and sent to the Lampung Veterinary Center for making histological
113 preparations. with Hematoxylin Eosin (HE) staining. The observation of preparations was
114 carried out microscopically using the Leica DM500® Binocular Microscope Technology to
115 accurately calculate various parameter sizes.

116 The research parameters were villi height, villi apex width, basal villi width, villi area,
117 crypt depth and gland diameter. Observation of histological preparations using 10x
118 magnification objective lens. The calculation of each parameter was carried out as many as
119 three villi (villiheight, villi apex width, basal villi width, villi area, crypt depth) and nine gland
120 diameters in each digestive tract organ (duodenum, jejunum, ileum, large intestine). There were
121 three replications per organ, so that the total for each organ was obtained an average of the nine
122 villi and twenty-seven glands.

123 The calculation of the surface area of the intestinal villi using the method of Iji *et al.*
124 (2001) modified with the assumption that the villi model is an analogue of the trapezium shape

125 so that the average number of the apical widths of the villi plus the average basal width of the
126 villi is divided by two then multiplied by the height of the villi by the following formula.

127

$$128 \text{ Surface area} = \frac{\mathbf{b + c}}{2} \times \mathbf{a}$$

129

130 Description:

131 a = height of intestine villi

132 b = width of apex of intestine villi

133 c = width of basal of intestine villi

134

135 **Data analysis**

136 Measurement data for various research parameters were calculated the average of all
137 replications of each digestive tract organs (duodenum, jejunum, ileum, and large intestine)
138 then analyzed descriptively .

139

140 **RESULTS AND DISCUSSION**

141

142 **Parameter Measurements**

143 The average measurements of villi height, villi apex width, basal villi width, villi area,
144 crypt depth and gland diameter of the digestive tract organs (duodenum, jejunum, ileum, large
145 intestine) are presented in Table 1. Calculation of each parameter Three villi were performed
146 (height of villi, width of villi apex, basal width of villi, area of villi, depth of crypt) and nine
147 gland diameters on histopathological preparations in each digestive tract organ (duodenum,
148 jejunum, ileum, large intestine) in each treatment. There were three replications for each organ,
149 so that the total for each organ from each treatment was obtained an average calculation of the
150 nine villi and twenty-seven glands.

151 Based on Table 1, it is known that the treatment of giving black cumin (*Nigella sativa*)
 152 to the digestive organs of broilers through drinking water resulted in an increase in the size of
 153 the villi height, the width of the villi apex, the basal width of the villi, the area of the villi, and
 154 the diameter of the glands compared to the control (P0). Treatment P1 with a dose of 36 mg /
 155 kg of broiler body weight gave the highest effect on the increase in the average size of the villi
 156 height, villi apex width, basal villi width, villi area, and gland diameter in the broiler digestive
 157 tract organs, namely the small intestine (duodenum, jejunum, ileum) and large intestine.

158

159 Table 1. Average measurement of each parameter in each treatment, giving Black Cumin
 160 (*Nigella sativa*) to the digestive organs of broilers through drinking water (in
 161 millimeter)

	Villi Height	Basal Villi Width	Apex Villi Width	Vili Size	Crypt Depth	Gland Diameter
Duodenum						
P0	0.5438	0.1716	0.0696	0.0616	0.2872	0.0497
P1	0.8781	0.2489	0.1449	0.1746	0.2728	0.0576
P2	0.7096	0.2251	0.0979	0.1161	0.3312	0.0523
P3	0.6446	0.1498	0.0838	0.0757	0.2473	0.0513
Jejunum						
P0	0.3789	0.1571	0.1439	0.0540	0.2366	0.0561
P1	0.8741	0.1836	0.1325	0.1264	0.2651	0.0611
P2	0.5516	0.1055	0.0448	0.0422	0.1910	0.0560
P3	0.3423	0.1480	0.0864	0.0423	0.2312	0.0529
Ileum						

P0	0.2890	0.1916	0.1060	0.0454	0.1781	0.0511
P1	0.4572	0.2448	0.1623	0.0910	0.1743	0.0529
P2	0.3256	0.1371	0.0923	0.0370	0.1314	0.0461
P3	0.4234	0.1554	0.0656	0.0454	0.1403	0.0500
<hr/>						
Large						
Intestinum						
P0	0.1914	0.1736	0.0666	0.0231	0.2051	0.0493
P1	0.3126	0.1936	0.1174	0.0493	0.1571	0.0511
P2	0.2266	0.1185	0.0510	0.0190	0.1381	0.0500
P3	0.2592	0.1328	0.0649	0.0251	0.1288	0.0602

162 P0 (drinking water without Black Cumin (*Nigella sativa*));

163 P1 (drinking water with Black Cumin (*Nigella sativa*) 36 mg/kg BW/day);

164 P2 (drinking water with Black Cumin (*Nigella sativa*) 72 mg/kg BW/day);

165 P3 (drinking water with Black Cumin (*Nigella sativa*) 144 mg/kg BW/day)

166 Highlight text (highest measurement for each treatment).

167

168 The ability of digestion and absorption of food substances could be affected by the
 169 surface area of the intestinal epithelium, the number of folds, and the number of villi and
 170 microvilli that expand the absorption field (Austic and Nesheim, 1990; Ibrahim 2008) and also
 171 influenced by the height and surface area of the villi organs. digestive tract (Sugito et al., 2007;
 172 Ibrahim 2008). These villi function to expand the surface of the intestine which affects the
 173 process of absorption of food (Alfiansyah, 2011). The development of the intestinal villi in
 174 broiler chickens is related to the function of the intestine and growth of the chicken (Sun, 2004).
 175 Villi are places for absorption of nutrients, the wider the villi, the more food substances that

176 will be absorbed, in the end it can have an impact on the growth of organs and increased carcass
177 (Asmawati, 2014).

178 The increase in villi height in the broiler intestine is closely related to an increase in
179 digestive function and absorption function due to the expansion of the absorption area and is
180 an expression of the smooth transportation system of nutrients throughout the body (Awad et
181 al., 2008). One of the parameters that can be used to measure the quality of growth is the
182 morphological structure of the intestine. The height of villi in all parts of the small intestine
183 (duodenum, jejunum, ileum) and large intestine in general increases (Ningtias, 2013).
184 Increasing the villi width and the height of the villi can expand the absorption area of the villi.
185 According to Asmawati (2014), the wider the villi the more food substances that will be
186 absorbed in the end can have an impact on the growth of the body's organs and according to
187 Rahmawati (2016) the higher the size of the villi, the wider the area of nutrient absorption by
188 the small intestine wall so that it will trigger Increased growth, according to Guyton (1997),
189 the more villous surface area indicates the more efficient absorption of nutrients that occurs.
190 Efficiency of nutrient absorption cannot be separated from the work of hormonal, nervous and
191 digestive glands in the digestive tract and its accessory glands.

192 Food, environment, and metabolic activity affects the number of intestinal glands.
193 Chickens generally eat food consisting of granules and are hard, so that a more active secretion
194 of intestinal glands is needed, to support the development of epithelial cells that make up the
195 villi (Mardhiah, 1991). Crypts contained in the intestinal villi, which are composed of inline
196 cylindrical epithelial cells. These glands produce mucus and several enzymes for the
197 metabolism of peptides, fats, carbohydrates, and intestinal juices (mucin) which function to
198 protect the intestinal mucosa (Aughey and Frye, 2001 The increase in the average size of the
199 gland diameter in the treatment with black cumin in drinking water showed an increase in the
200 size of the gland diameter in each organ teruta. However, in P1 treatment with the highest

201 increase in size compared to other treatments, it can support the development of epithelial cells
202 that make up the villi, which will increase the absorption of nutrients in the digestive tract.

203 The highest mean measurement results for the depth of crypto varied in each treatment
204 in each organ (Table 1). According to Sun et al. (2005) and Smirnov et al. (2005) that into
205 crypto has no effect after broilers are more than 28 days old. Broilers in this study collected
206 samples of digestive tract organs at the age of 31 days. It is assumed that the development of
207 intestinal morphology is closely related to the role of micronutrients in line with the increasing
208 age of broilers (Harimurti and Rahayu, 2009).

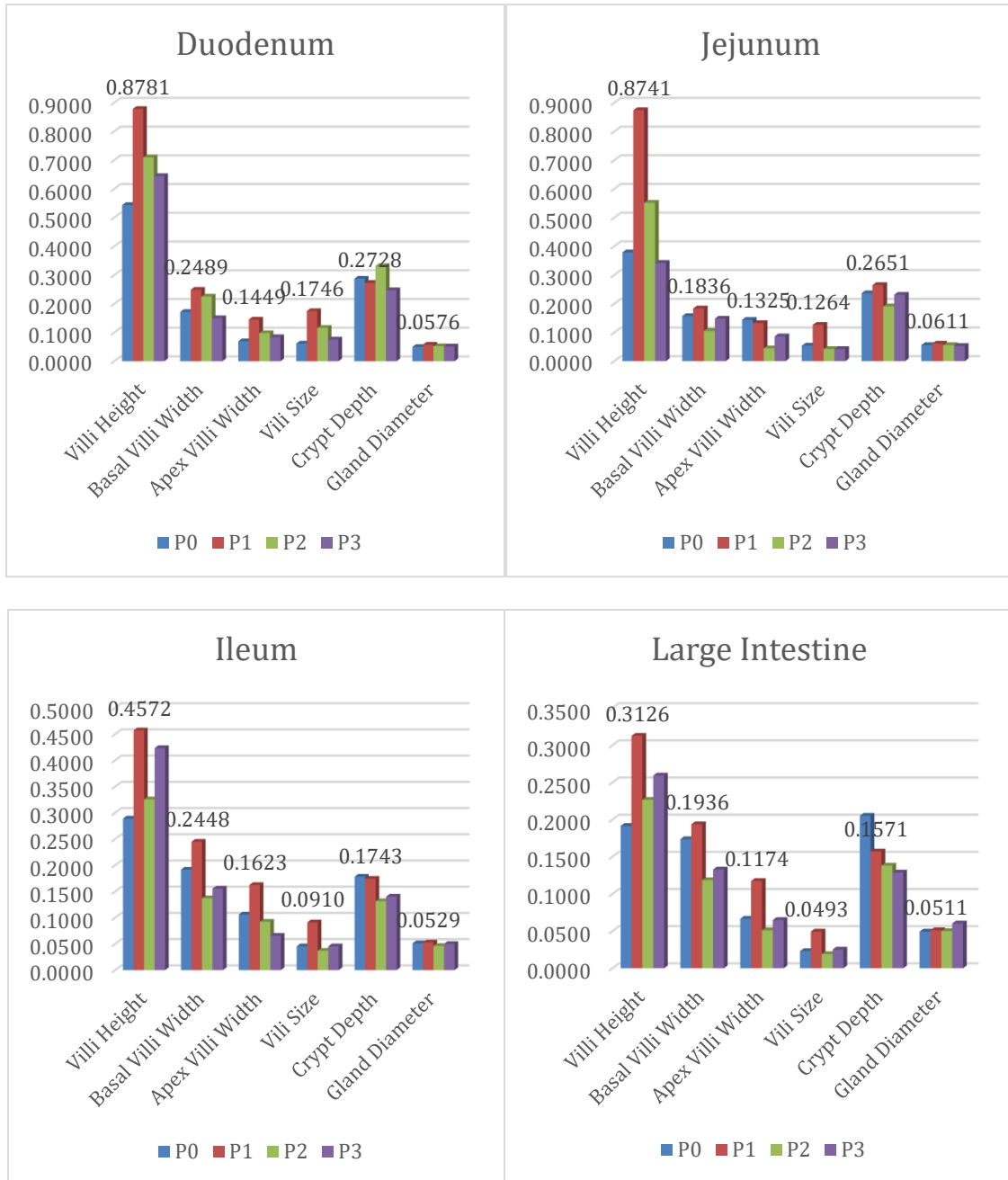
209 Based on the data presented in Figure 1, supplementation of Black Cumin (*Nigella*
210 *sativa*) could increase the size of the research parameters, namely the size of the villi height,
211 the width of the villi apex, the basal width of the villi, the area of the villi, and the gland
212 diameter of all digestive tract organs compared to controls. P1 treatment with a dose of 36
213 mg/kg of broiler body weight had the highest effect on increasing the size of each of these
214 parameters.

215 One of the parameters that can be used to measure the quality of growth is the
216 morphological structure of the intestine (Wang *et al.*, 2008; Ningtias 2013). According to
217 Suprijatna *et al.*, (2008) the small intestine is the main organ for digestion and absorption of
218 digestive products. Various enzymes that enter this channel function to accelerate and
219 streamline the breakdown of carbohydrates, proteins and fats to facilitate the absorption
220 process. In adult chickens, the length of the small intestine is about 62 inches or 1.5 meters.

221 The digestive tract organs are supported by villi which are a special shape in the mucosa.
222 The villi are finger-shaped protrusions of the mucosa and are characteristic of the small
223 intestine. Increasing the number of villi will increase food absorption. Villi function to expand
224 the surface of the intestine which affects the process of absorption of food (Alfiansyah, 2011).
225 The development of the intestinal villi in broiler chickens is related to the function of the

226 intestine and growth of the chicken (Sun, 2004). The increase in villi causes more villi surface
 227 area to absorb nutrients into the bloodstream (Mile *et al.* 2006; Rostinawati 2008).

228



229

230

231 Figure 1. Drinking water with Black Cumin (*Nigella sativa*) 36 mg/kg BW/day (P1) was the
 232 highest measurement of each research parameter in the small intestine
 233 (duodenum, jejunum, ileum) and large intestine

234

235 High villi indicate that the intestines are better off than short villi. Awad *et al.* (2008)
236 stated that the increase in the height of the villi in the intestine with digestive and absorption
237 functions occurs because of the intact villi form which is a smooth expression of the nutrient
238 transport system throughout the body. Rofiq (2003) states that the absorption of nutrients in
239 the intestine is influenced by the inner surface area of the intestine (folds, villi and microvilli)
240 and the length of transit of the digesta in the intestine.

241 The surface area of the intestine such as the height of the villi describes the area for
242 absorption of nutrients. Villi are small finger or leaf-like protrusions found on the mucous
243 membrane, 0.5 to 1.5 mm long and found only in the small intestine. The villi in the ileum are
244 finger-like in shape and shorter than the villi found on the duodenum and jejunum. One of the
245 parameters used to measure the quality of growth is the intestinal morphological structure
246 (Wang and Peng, 2008).

247 The intestinal gland (Lieberkuhn's gland) has a small hole that becomes the mouth of the
248 simplek tubulose gland. The intestinal glands are scattered between the villi, attached to the
249 mucous membrane. The intestinal glands and intestinal villi are covered by an epithelium,
250 consisting of goblet cells and enterocytes, among others. Goblet cells secrete mucus to
251 lubricate and protect the surface of the intestine, while the enterocytes in crypt secrete large
252 amounts of water and electrolytes (Pfeiffer and Macpherson, 1990).

253

254

CONCLUSION

255

256 Supplementation of Black Cumin 36 mg/kg BW/day through drinking water could
257 increasing the average sizes of villi height, villi apex width, basal villi width, villi area, and
258 gland diameter of small intestine (duodenum, jejunum, ileum) and large intestine of broiler.

259

ACKNOWLEDGMENTS

260
261
262
263
264
265
266
267
268
269
270
271
272
273
274
275
276
277
278
279
280
281
282
283

The authors would like to thank University of Lampung for providing research funding sourced from the BLU University of Lampung 2020.

REFERENCES

- Abun. 2007. **Pengukuran nilai kecernaan pakan yang mengandung limbah udang windu produk fermentasi pada ayam broiler**. Research report. Padjajaran University, Jatinangor.
- Al-Beitawi N., S.S.E. Ghousein. 2008. Effect of feeding different levels of Nigella sativa seed (black cumin) on performance, blood constituents and carcass characteristic of broiler chick. **Int. J. Poult. Sci.** 7:775-778.
- Alfiansyah, M. 2011. Anatomi dan Pencernaan Usus Halus. [http://www. sentra-edukasi.com/](http://www.sentra-edukasi.com/). Diakses tanggal 20 Agustus 2020
- Alzeer. 2008. **Diseases of Poultry**. 12th Ed. Blackwell, New Jersey.
- Asmawati. 2014. The Effect of In Ovo Feeding on Hatching Weight and Small Intestinal Tissue Development of Native Chicken. **Dissertation**. Hasanuddin University, Makassar.
- Awad, W.A., K. Ghareeb, S. Nitclu, S. Pasteiner, S.A. Raheem, and J. Bohm. 2008. Effect of dietary inclusion of probiotic, prebiotic and symbiotic on intestinal glucose absorption of broiler chickens. **J. Poult. Sci.** 7: 688-691.
- Badan Pusat Statistik. 2019. Populasi Ayam Ras Pedaging menurut Provinsi (Ekor) Tahun 2009-2018. <https://www.bps.go.id/dynamic/table/2015/12/18/1034/populasi-ayam-ras-pedaging-menurut-provinsi-2009-2018.html> Diakses pada tanggal 13 Agustus 2020.

- 284 Diamita, A.A. 2009. Pengaruh pemberian minyak wijen (*Sesamum indicum Linn.*) dengan cold
285 press bertingkat terhadap kerusakan histologis lambung mencit (*Mus musculus*) yang
286 diinduksi aspirin. **Thesis**. Faculty of Medical Universitas Sebelas Maret, Surakarta.
- 287 El-Dakhakhny M., N.J. Madi, N. Lambert, H.P. Ammon. 2002. Nigella sativa oil, nigellone
288 and derived thymoquinone inhibit synthesis of 5-lipoxygenase products in
289 polymorphonuclear leukocytes from rats. **J. Ethnopharmacol.** 81:161-164.
- 290 Fadillah, R. 2004. **Panduan Mengelola Peternakan Ayam Broiler Komersial**. PT
291 AgroMedia Pustaka, Tangerang.
- 292 Frappier, B.L. 2006. **Digestive System**. In: J.A. Eurell dan B.L. Frappier, Editor. Dellmann's
293 **Texbook of Veterinary Histology**. 6th Ed. Blackwell, Oxford.
- 294 Hamzah. 2013. Respon usus dan karakteristik karkas pada ayam ras pedaging dengan berat
295 badan awal berbeda yang dipuaskan setelah menetas. **Thesis**. Faculty of Animal
296 Science. Hasanuddin University, Makassar.
- 297 Harimurti,, S. dan E.S. Rahayu. 2009. Morfologi Usus Ayam Broiler yang disuplementasi
298 dengan Probiotik Strain Tunggal dan Campuran. **Agritech**. 29(3): 179-183.
- 299 Hendrik. 2009. **Habbatus Sauda**. Pustaka Iltazam, Solo.
- 300 Ibrahim, S. 2008. Hubungan ukuran-ukuran usus halus dengan berat badan broiler. **Agripet**.
301 8(2): 42-46.
- 302 Iji, P. A., R. J. Hughes, M. Choct and D. R. Tivey. 2001. Intestinal structure and function of
303 broiler chickens on wheat-based diets supplemented with microbial enzyme. Asian-Aust.
304 **J. Anim. Sci.** 14(1):54-60.
- 305 Kabir, S.M.L. 2010. Avian colibacillosis and salmonellosis: a closer look at epidemiology,
306 pathogenesis, diagnosis, control and public health concerns. **Int. J. Environ. Res. Public**
307 **Health**. 7:89-114.

- 308 Mile, R.D., G.D. Butcher, P.R. Henry, R.C. Littlel. 2006. Effect of antibiotic growth promoters
309 on broiler performance, intestinal growth parameters, and quantitative morphology. **J.**
310 **Poultry Sci.** 85:476-485.
- 311 Ningtias, A.S. 2013. Comparison of Growth Performance of Broilers, Kampong, and
312 Backcross3 (*Gallus gallus domesticus* Linnaeus, 1758) Based on Morphometri and
313 Histological Structure of Ileum and Breast Muscle. **Thesis.** Fakultas Biologi Universitas
314 Gajah Mada. Yogyakarta
- 315 Rostinawati T. 2008. Aktivitas antibakteri ekstrak etanol dan ekstrak air kelopak bunga rosela
316 (*Hibiscus sabdariffa* L.) terhadap *Mycobacterium tuberculosis* galur Labkes-026 (Multi
317 Drug Resisten) dan *Mycobacterium tuberculosis* galur H37Rv secara in vitro. **Thesis.**
318 Faculty of Pharmacy, Padjadjaran University, Bandung.
- 319 Siagian, Y.A. 2016. Gambaran Histologis dan Tinggi Vili Usus Halus Bagian Ileum Ayam Ras
320 Pedaging yang diberi Tepung Daun Kelor (*Moringa oleifera*) dalam ransum. **Thesis.**
321 Fakultas of Animal Science Hasanuddin University, Makasar.
- 322 Smirnov, A., R. Perez, E. Amit-Romach, D. Sklan, and Z. Uni. 2005. Mucin dynamics and
323 microbial populations in chicken small intestine changed by dietary probiotic and
324 antibiotic growth promotor supplementation. **J. of Nutrition.** 135: 187-192.
- 325 Sturkie, P.D., G.C. Whittow. 2000. **Sturkie's Avian Physiology.** Academic Press, Waltham.
- 326 Sugito, M.W., D.A. Astuti, E. Handharyani, Chairul. 2007. Histopatologi hati dan ginjal pada
327 ayam broiler yang dipapar cekaman panas dan diberi ekstrak kulitbatang Jaloh (*Salix*
328 *tetrasperma* Roxb). **JITV.** 12:6873.
- 329 Sun, X. 2004. Broiler performance and intestinal alterations when fed drug-free diets. **Thesis.**
330 Animal and Poultry Science. Blacksburg, Virginia
- 331 Sun, X., A. McElroy, Jr. Webb, A.E.K.E. Sefton and C. Novak. 2005. Broiler performance and
332 intestinal alterations when fed drug-free diets. **J. of Poult. Sci.,** 84: 1294-1302.

- 333 Suprijatna, E., U. Atmomarsono, R. Kartasudjana. 2008. **Ilmu Dasar Ternak Unggas**. Penebar
334 Swadaya, Jakarta.
- 335 Thippeswammy N.B., K.A. Naidu. 2005. Antioxidant potency of cumini varieties cumini, black
336 cumini and bitter cumini on antioxidant systems. **Euro Food Res Tech.** 220: 472-476
- 337 Topozoda H.H., H.A. Mazloun, M. El-Dakhakhny. 1965. The antibacterial properties of
338 *Nigella sativa* seeds. **J. Egypt Med.** 48:187-202.
- 339 Yusuf, M.S. 2014. Efektivitas Penggunaan Jintan Hitam (*Nigella sativa*) dalam Proses
340 Percepatan Penyembuhan Luka Setelah Pencabutan Gigi. **Thesis**. Faculty of Dentistry
341 Hasanuddin University, Makassar.
- 342 Wang, J.X. and K.M. Peng. 2008. Molecular, Cellular, and Developmental Biology
343 Developmental Morphology of the Small Intestine of African Ostrich Chicks. **J. Poultry**
344 **Sci.** 87: 2629-2635.

Editor/Author Correspondence

Editor
2021-03-10 04:01 PM

Subject: [J.KED.HEWAN] Editor Decision

[DELETE](#)

drh., M.Sc. Muhammad Mirandy Pratama Sirat:

We have reached a decision regarding your submission to Jurnal Kedokteran Hewan - Indonesian Journal of Veterinary Sciences, "EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER".

Our decision is: Revisions Required

- Pendahuluan perlu direvisi agar lebih detail. masih terdapat dalam satu alinea terdiri atas satu kalimat.
- Komposisi pustaka hari minimal 70% dari jurnal ilmiah.
- Mohon dikirimkan 2 calom reviewer dari luar institusi penulis

Drh. T Armansyah TR, M.Kes.
Departement of Veterinary Medicine Faculty of Veterinary Medicine Syiah Kuala University
Phone 085260153935
jkh@unsyiah.ac.id

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

Author
2021-03-17 03:26 PM

Subject: EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH [DELETE](#) DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER

Terimakasih atas kesempatan yang diberikan untuk kami dapat merevisi naskah.





Rendy Sirat <m.mirandy@fp.unila.ac.id>

[J.KED.HEWAN] Editor Decision

2 messages

Drh. T Armansyah TR, M.Kes. <jurnal@unsyiah.ac.id>

Wed, Mar 10, 2021 at 4:01 PM

To: "drh., M.Sc. Muhammad Mirandy Pratama Sirat" <m.mirandy@fp.unila.ac.id>

Cc: Madi Hartono <madi.hartono@fp.unila.ac.id>, Purnama Edy Santosa <purnama.edy@fp.unila.ac.id>, Ratna Ermawati <ratna.ermawati@fp.unila.ac.id>

drh., M.Sc. Muhammad Mirandy Pratama Sirat:

We have reached a decision regarding your submission to Jurnal Kedokteran Hewan - Indonesian Journal of Veterinary Sciences, "EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER".

Our decision is: Revisions Required

- Pendahuluan perlu direvisi agar lebih detail. masih terdapat dalam satu alinea terdiri atas satu kalimat.
- Komposisi pustaka hari minimal 70% dari jurnal ilmiah.
- Mohon dikirimkan 2 calon reviewer dari luar institusi penulis

Drh. T Armansyah TR, M.Kes.

Departement of Veterinary Medicine Faculty of Veterinary Medicine Syiah

Kuala University

Phone 085260153935

jkh@unsyiah.ac.id-----
Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP

Chief Editor E-mail: siregar@unsyiah.ac.idWebsite: <http://jurnal.unsyiah.ac.id/JKH>e-mail : jkh@unsyiah.ac.id**Rendy Sirat** <m.mirandy@fp.unila.ac.id>

Thu, Mar 11, 2021 at 1:16 PM

To: "Drh. T Armansyah TR, M.Kes." <jurnal@unsyiah.ac.id>

Kepada Yth. Editor JKH

Dengan hormat,

Terimakasih kami ucapkan atas kesempatan yang diberikan. Kami akan segera melengkapi berdasarkan saran-saran yang telah diberikan.

[Quoted text hidden]

--

drh. Muhammad Mirandy Pratama Sirat, M.Sc.***Department of Animal Husbandry******Faculty of Agriculture University of Lampung*****082226238837**

Author
2021-03-17 03:26 PM

Subject: EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH [DELETE](#)
DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE
OF BROILER

Terimakasih atas kesempatan yang diberikan untuk kami dapat merevisi naskah.
Naskah akan kami perbaiki segera sesuai saran-saran editor.

Berikut kami lampirkan Form A terkait usulan 2 calon reviewer dari luar institusi.

Atas kerjasama yang diberikan, kami ucapkan terimakasih.

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

Author
2021-03-17 03:31 PM

Subject: EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH [DELETE](#)
DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE
OF BROILER

Terimakasih atas kesempatan yang diberikan untuk kami dapat merevisi naskah.
Naskah akan kami perbaiki segera sesuai saran-saran editor.

Berikut kami lampirkan Form A terkait usulan 2 calon reviewer dari luar institusi.

Atas kerjasama yang diberikan, kami ucapkan terimakasih.

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

Editor
2021-04-16 02:31 PM

Subject: [J.KED.HEWAN] Editor Decision [DELETE](#)

drh., M.Sc. Muhammad Mirandy Pratama Sirat:



Suggest Reviewers

No.	Name	Institute	Address	Telephone	E-mail
1.	drh. Fajar Shodiq Permata, M.Biotech.	Faculty of Veterinary Medicine Brawijaya University	Puncak Dieng Eksklusif, Kalisongo, Kec. Danau, Kab. Malang 65151	085878214102	drh.fajar@ub.ac.id
2.	drh. Filphin Adolfin Amalo, M.Sc.	Faculty of Veterinary Medicine University of Nusa Cendana	Jl. Adi Sucipto Penfui, Lasiana, Klp. Lima, Kota Kupang, Nusa Tenggara Tim.	085237940993	drh.filphin.amalo@gmail.com

Editor
2021-04-16 02:31 PM

Subject: [J.KED.HEWAN] Editor Decision

[DELETE](#)

drh., M.Sc. Muhammad Mirandy Pratama Sirat:

We have reached a decision regarding your submission to Jurnal Kedokteran Hewan - Indonesian Journal of Veterinary Sciences, "EFFECT OF BLACK CUMIN (Nigella sativa) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER".

Our decision is: Revisions Required

Reviewer A: it is better if using OneWAY ANOVA statistic continue Tukey test to comparing among small intestine

Reviewer B: See in Online Journal JKH

Drh. T Armansyah TR, M.Kes.
Departement of Veterinary Medicine Faculty of Veterinary Medicine Syiah Kuala University
Phone 085260153935
jkh@unsyiah.ac.id

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

Author
2021-04-18 08:47 AM

Subject: EFFECT OF BLACK CUMIN (Nigella sativa) SUPPLEMENTATION THROUGH [DELETE](#) DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER

Dear Mr. Tongku

Regarding reviewers results of our article, could you help me how can I see the review results from reviewers as you said in email before.

Reviewer A: it is better if using OneWAY ANOVA statistic continue Tukey test to comparina among small intestine

Author
2021-04-18 08:47 AM

Subject: EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH ~~DELETE~~ DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER

Dear Mr. Tongku

Regarding reviewers results of our article, could you help me how can I see the review results from reviewers as you said in email before.

Reviewer A: it is better if using OneWAY ANOVA statistic continue Tukey test to comparing among small intestine

Reviewer B: See in Online Journal JKH

Because I can't see any suggestions from reviewers in article file 19774-60754-1-RV or in Online Journal JKH.

Thank you very much for your help

Best regards

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

Author
2021-04-25 04:09 PM

Subject: EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH ~~DELETE~~ DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER

Dear Mr. Tongku

I have been submitted revision of our article and have adjusted it according to the reviewers' suggestions.

Thank you for your attention and cooperation





Rendy Sirat <m.mirandy@fp.unila.ac.id>

[J.KED.HEWAN] Editor Decision

1 message

Drh. T Armansyah TR, M.Kes. <jurnal@unsyiah.ac.id>

Fri, Apr 16, 2021 at 2:31 PM

To: "drh., M.Sc. Muhammad Mirandy Pratama Sirat" <m.mirandy@fp.unila.ac.id>

Cc: Madi Hartono <madi.hartono@fp.unila.ac.id>, Purnama Edy Santosa <purnama.edy@fp.unila.ac.id>, Ratna Ermawati <ratna.ermawati@fp.unila.ac.id>

drh., M.Sc. Muhammad Mirandy Pratama Sirat:

We have reached a decision regarding your submission to Jurnal Kedokteran Hewan - Indonesian Journal of Veterinary Sciences, "EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER".

Our decision is: Revisions Required

Reviewer A: it is better if using OneWAY ANOVA statistic continue Tukey test to comparing among small intestine

Reviewer B: See in Online Journal JKH

Drh. T Armansyah TR, M.Kes.
Departement of Veterinary Medicine Faculty of Veterinary Medicine Syiah Kuala University
Phone 085260153935
jkh@unsyiah.ac.id

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

1 **EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH**
 2 **DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND**
 3 **LARGE INTESTINE OF BROILER**

4
 5 **Authors**

6
 7
 8 **ABSTRACT**

9
 10 This study aimed to determine the effect of Black Cumin (*Nigella sativa*)
 11 supplementation through drinking water on the histology of small intestine and large intestine
 12 of broiler. The research was conducted from April September 2020 in cage facility of Integrated
 13 Field Laboratory, Faculty of Agriculture, University of Lampung. This research was using a
 14 completely randomized design (CRD) with four treatment groups and three replications (five
 15 heads per replication) with a total of 60 male broilers. The treatment were drinking water
 16 without Black Cumin (P0, control); drinking water with Black Cumin 36 mg/kg BW/day (P1);
 17 72 mg/kg BW/day (P2); and 144 mg/kg BW/day (P3). Three broilers from each group were
 18 randomly necropsied at 31st days old, and samples of the small intestine (duodenum, jejunum,
 19 ileum) and large intestine were fixed with 10% formalin solution and sent to the Lampung
 20 Disease Investigation Center for histological preparations. The observation of preparations was
 21 carried out microscopically using the Leica DM500® Binocular Microscope to accurately
 22 calculate various parameter sizes. The results were analyze descriptively. The conclusion of
 23 this study was the supplementation of Black Cumin 36 mg/kg BW/day through drinking water
 24 could increase the average sizes of villi height, villi apex width, basal villi width, villi area, and
 25 gland diameter of small intestine (duodenum, jejunum, ileum) and large intestine of broiler.

Commented [a11]: This is dose but application is in water which concentration

Commented [a12]: Check necropsied

Commented [a13]: Analyzed

descriptively but the results are quantitative variables including villi height, width

Please write the method for histomorphometry measurement of villi

Commented [a14]: Using statistic analysis or not?

26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51

Key words : black cumin, broiler, Leica DM500 microscope, large intestine, small intestine.

ABSTRAK

Penelitian ini bertujuan untuk mengetahui pengaruh suplementasi Jintan Hitam (Nigella sativa) melalui air minum terhadap histologi usus halus dan usus besar broiler. Penelitian dilakukan pada April-September 2020 di unit kandang Laboratorium Lapang Terpadu, Fakultas Pertanian, Universitas Lampung. Penelitian bersifat eksperimental menggunakan Rancangan Acak Lengkap (RAL) dengan empat kelompok perlakuan dan tiga ulangan (lima ekor tiap ulangan) sehingga total 60 ekor broiler jantan. Perlakuan yang diberikan yaitu pemberian air minum tanpa Jintan Hitam (P0, kontrol); air minum dengan Jintan Hitam 36 mg/kg BB/hari (P1); 72 mg/kg BB/hari (P2); dan 144 mg/kg BB/hari (P3). Tiga ekor dari tiap kelompok secara acak dinekropsi pada hari ke-31 dan diambil sampel organ usus halus (duodenum, jejunum, ileum) dan usus besar (colon) kemudian difiksasi dengan larutan formalin 10% dan dikirim ke Balai Veteriner Lampung untuk pembuatan preparat histologi. Pengamatan preparat dilakukan secara mikroskopis menggunakan Mikroskop Binokuler Leica DM500® untuk menghitung berbagai ukuran parameter secara akurat. Analisis hasil dilakukan secara deskriptif. Kesimpulan penelitian ini yaitu suplementasi Jintan Hitam (Nigella sativa) 36 mg/kg BB/hari meningkatkan rata-rata ukuran tinggi vili, lebar apeks vili, lebar basal vili, luas vili, dan diameter kelenjar pada organ saluran pencernaan yaitu usus halus (duodenum, jejunum, ileum) dan usus besar broiler.

Kata kunci : broiler, jintan hitam, mikroskop Leica DM500, usus besar, usus halus.

INTRODUCTION

The development of poultry farming in Indonesia is increasing. Broilers are one of the fastest growing poultry. The relatively short period of time and the relatively lower maintenance costs compared to ruminants make breeders prefer to cultivate broilers. In 2018, the broiler population in Indonesia reached 1.89 billion heads and an increase of 2.26% from the broiler population in 2017 of 1.85 billion heads (BPS, 2019).

Disease is a serious obstacle in the broiler farming industry. The high incidence of disease can cause a decrease in productivity and even death of livestock which causes significant losses for breeders. Administration of antibiotics in the livestock industry is used for the treatment of livestock so as to reduce the risk of death and restore the condition of the livestock to health, however, giving antibiotics for a long period of time can cause residual buildup which has negative effects if consumed by humans.

The use of antibiotics needs to be reduced to prevent negative effects by providing natural ingredients as immunomodulators. Immunomodulators can be defined as biological or synthetic substances that can stimulate the innate immune system, adaptive or both. One of the herbs that can act as an immunomodulator is Black Cumin (*Nigella sativa*). *Nigella sativa* is a plant that has the potential as an immunostimulant that can stimulate and strengthen the system by increasing the number, quality and activity of the body's immune cells (Hendrik, 2009). *Nigella sativa* contains *thymoquinone*, saponins, zinc or zinc, *alpha-linolenic acid* (Omega 3) and *linoleic acid* (Omega 6) which functions in cell formation, maintains the immune system, and helps in the process of blood formation (Yusuf, 2014).

The solution needed for the above problems is to examine the effect of black cumin (*Nigella sativa*) supplementation through histological studies of the digestive organs of broilers which are expected to have the potential to increase the size of villi height, apex villi width,

Commented [a15]: To farm

Commented [a16]: So as well as to

Commented [a17]: Please add the references

77 basal villi width, villi area, crypt depth and gland diameter of small intestine (duodenum,
78 jejunum, ileum) and large intestine.

Commented [a18]: The introduction is unclear related with background of problem and offering solution

79

80 MATERIALS AND METHODS

81

82 **Materials**

83 This study used broiler cages, sprayer for cage disinfection, bamboo to make 12 cage
84 plots, plastic tarpaulin for curtains, newspaper and used husks as litter, 12 bulbs of 15 bulbs
85 watt as a heating source for the area. *brooding*, a *hanging feeder* 12 pieces, *chick feeder tray* 12
86 pieces, 12 pieces of chicken drinking places; 1 bucket, 1 *hand spray*, 1 water tray for *dipping*,
87 1 electric scale, *thermohygrometer* for measuring temperature and humidity, sack and plastic.
88 Organ sampling equipment, namely necropsy equipment, *object glass*, *cover glass*,
89 *refrigerator*, microtome, light microscope, camera technology and *software* Optilab® along
90 with a laptop for taking tissue images and measuring the parameters of each organ.

91 Materials used in the study were 60 Day Old Chicks (DOC) male broiler Cobb CP 707
92 strains kept for 30 days, rations, drinking water, extract of Black Cumin (*Nigella sativa*),
93 vaccines of Newcastle Disease (ND), Avian Influenza (AI) and Infectious Bursal Disease
94 (IBD), 10% formalin solution.

95

96 **Methods**

97 This research carried out for 6 months (April - September 2020) in the Integrated Field
98 Laboratory enclosure unit, Faculty of Agriculture, University of Lampung. This research was
99 experimental in nature using a completely randomized design (CRD) with four treatment
100 groups and three replications (five heads per replication) so a total of 60 male broilers. The
101 treatment dose was according to the broiler body weight, namely 1) drinking water without

102 black cumin (P0, control); drinking water with Jintan Hitam 36 mg / kg BW / day (P1); drinking
103 water with Jintan Hitam 72 mg / kg BW / day (P2); and drinking water with cumin 144 mg /
104 kg BW / day (P3). On the 31st day, three from each group were randomly necropsed and
105 samples of the small intestine (duodenum, jejunum, ileum) and large intestine were then fixed
106 with 10% formalin solution and sent to the Lampung Veterinary Center for making histological
107 preparations. with Hematoxylin Eosin (HE) staining. The observation of preparations was
108 carried out microscopically using the Leica DM500® Binocular Microscope Technology to
109 accurately calculate various parameter sizes.

110 The research parameters were villi height, villi apex width, basal villi width, villi area,
111 crypt depth and gland diameter. Observation of histological preparations using 10x
112 magnification objective lens. The calculation of each parameter was carried out as many as
113 three villi (villi height, villi apex width, basal villi width, villi area, crypt depth) and nine gland
114 diameters in each digestive tract organ (duodenum, jejunum, ileum, large intestine). There were
115 three replications per organ, so that the total for each organ was obtained an average of the nine
116 villi and twenty-seven glands.

117 The calculation of the surface area of the intestinal villi using the method of Iji *et al.*
118 (2001) modified with the assumption that the villi model is an analogue of the trapezium shape
119 so that the average number of the apical widths of the villi plus the average basal width of the
120 villi is divided by two then multiplied by the height of the villi by the following formula.

121

$$122 \text{ Surface area} = \frac{b + c}{2} \times a$$

123 Description:

125 a = height of intestine villi

126 b = width of apex of intestine villi

127 c = width of basal of intestine villi

128

129 **Data analysis**

130 Measurement data for various research parameters were calculated the average of all
131 replications of each digestive tract organs (duodenum, jejunum, ileum, and large intestine)
132 then analyzed descriptively .

Commented [a19]: It is better using One WAY ANOVA statistic

133

134 **RESULTS AND DISCUSSION**

135

136 **Parameter Measurements**

137 The average measurements of villi height, villi apex width, basal villi width, villi area,
138 crypt depth and gland diameter of the digestive tract organs (duodenum, jejunum, ileum, large
139 intestine) are presented in Table 1. Calculation of each parameter Three villi were performed
140 (height of villi, width of villi apex, basal width of villi, area of villi, depth of crypt) and nine
141 gland diameters on histopathological preparations in each digestive tract organ (duodenum,
142 jejunum, ileum, large intestine) in each treatment. There were three replications for each organ,
143 so that the total for each organ from each treatment was obtained an average calculation of the
144 nine villi and twenty-seven glands.

145 Based on Table 1, it is known that the treatment of giving black cumin (*Nigella sativa*)
146 to the digestive organs of broilers through drinking water resulted in an increase in the size of
147 the villi height, the width of the villi apex, the basal width of the villi, the area of the villi, and
148 the diameter of the glands compared to the control (P0). Treatment P1 with a dose of 36 mg /
149 kg of broiler body weight gave the highest effect on the increase in the average size of the villi
150 height, villi apex width, basal villi width, villi area, and gland diameter in the broiler digestive
151 tract organs, namely the small intestine (duodenum, jejunum, ileum) and large intestine.

152

153 Table 1. Average measurement of each parameter in each treatment, giving Black Cumin
 154 (*Nigella sativa*) to the digestive organs of broilers through drinking water (in
 155 millimeter)

	Villi Height	Basal Villi Width	Apex Villi Width	Vili Size	Crypt Depth	Gland Diameter
Duodenum						
P0	0.5438	0.1716	0.0696	0.0616	0.2872	0.0497
P1	0.8781	0.2489	0.1449	0.1746	0.2728	0.0576
P2	0.7096	0.2251	0.0979	0.1161	0.3312	0.0523
P3	0.6446	0.1498	0.0838	0.0757	0.2473	0.0513
Jejunum						
P0	0.3789	0.1571	0.1439	0.0540	0.2366	0.0561
P1	0.8741	0.1836	0.1325	0.1264	0.2651	0.0611
P2	0.5516	0.1055	0.0448	0.0422	0.1910	0.0560
P3	0.3423	0.1480	0.0864	0.0423	0.2312	0.0529
Ileum						
P0	0.2890	0.1916	0.1060	0.0454	0.1781	0.0511
P1	0.4572	0.2448	0.1623	0.0910	0.1743	0.0529
P2	0.3256	0.1371	0.0923	0.0370	0.1314	0.0461
P3	0.4234	0.1554	0.0656	0.0454	0.1403	0.0500
Large Intestinum						
P0	0.1914	0.1736	0.0666	0.0231	0.2051	0.0493
P1	0.3126	0.1936	0.1174	0.0493	0.1571	0.0511

Commented [a110]: Please add \pm Standar deviation (ad is it better used micrometer)

P2	0.2266	0.1185	0.0510	0.0190	0.1381	0.0500
P3	0.2592	0.1328	0.0649	0.0251	0.1288	0.0602

156 P0 (drinking water without Black Cumin (*Nigella sativa*));
 157 P1 (drinking water with Black Cumin (*Nigella sativa*) 36 mg/kg BW/day);
 158 P2 (drinking water with Black Cumin (*Nigella sativa*) 72 mg/kg BW/day);
 159 P3 (drinking water with Black Cumin (*Nigella sativa*) 144 mg/kg BW/day)
 160 Highlight text (highest measurement for each treatment).

161

162 The ability of digestion and absorption of food substances could be affected by the
 163 surface area of the intestinal epithelium, the number of folds, and the number of villi and
 164 microvilli that expand the absorption field (Austic and Nesheim, 1990; Ibrahim 2008) and also
 165 influenced by the height and surface area of the villi organs. digestive tract (Sugito et al., 2007;
 166 Ibrahim 2008). These villi function to expand the surface of the intestine which affects the
 167 process of absorption of food (Alfiansyah, 2011). The development of the intestinal villi in
 168 broiler chickens is related to the function of the intestine and growth of the chicken (Sun, 2004).
 169 Villi are places for absorption of nutrients, the wider the villi, the more food substances that
 170 will be absorbed, in the end it can have an impact on the growth of organs and increased carcass
 171 (Asmawati, 2014).

172 The increase in villi height in the broiler intestine is closely related to an increase in
 173 digestive function and absorption function due to the expansion of the absorption area and is
 174 an expression of the smooth transportation system of nutrients throughout the body (Awad et
 175 al., 2008). One of the parameters that can be used to measure the quality of growth is the
 176 morphological structure of the intestine. The height of villi in all parts of the small intestine
 177 (duodenum, jejunum, ileum) and large intestine in general increases (Ningtias, 2013).
 178 Increasing the villi width and the height of the villi can expand the absorption area of the villi.

179 According to Asmawati (2014), the wider the villi the more food substances that will be
180 absorbed in the end can have an impact on the growth of the body's organs and according to
181 Rahmawati (2016) the higher the size of the villi, the wider the area of nutrient absorption by
182 the small intestine wall so that it will trigger Increased growth, according to Guyton (1997),
183 the more villous surface area indicates the more efficient absorption of nutrients that occurs.
184 Efficiency of nutrient absorption cannot be separated from the work of hormonal, nervous and
185 digestive glands in the digestive tract and its accessory glands.

186 Food, environment, and metabolic activity affects the number of intestinal glands.
187 Chickens generally eat food consisting of granules and are hard, so that a more active secretion
188 of intestinal glands is needed, to support the development of epithelial cells that make up the
189 villi (Mardhiah, 1991). Crypts contained in the intestinal villi, which are composed of inline
190 cylindrical epithelial cells. These glands produce mucus and several enzymes for the
191 metabolism of peptides, fats, carbohydrates, and intestinal juices (mucin) which function to
192 protect the intestinal mucosa (Aughey and Frye, 2001 The increase in the average size of the
193 gland diameter in the treatment with black cumin in drinking water showed an increase in the
194 size of the gland diameter in each organ teruta. However, in P1 treatment with the highest
195 increase in size compared to other treatments, it can support the development of epithelial cells
196 that make up the villi, which will increase the absorption of nutrients in the digestive tract.

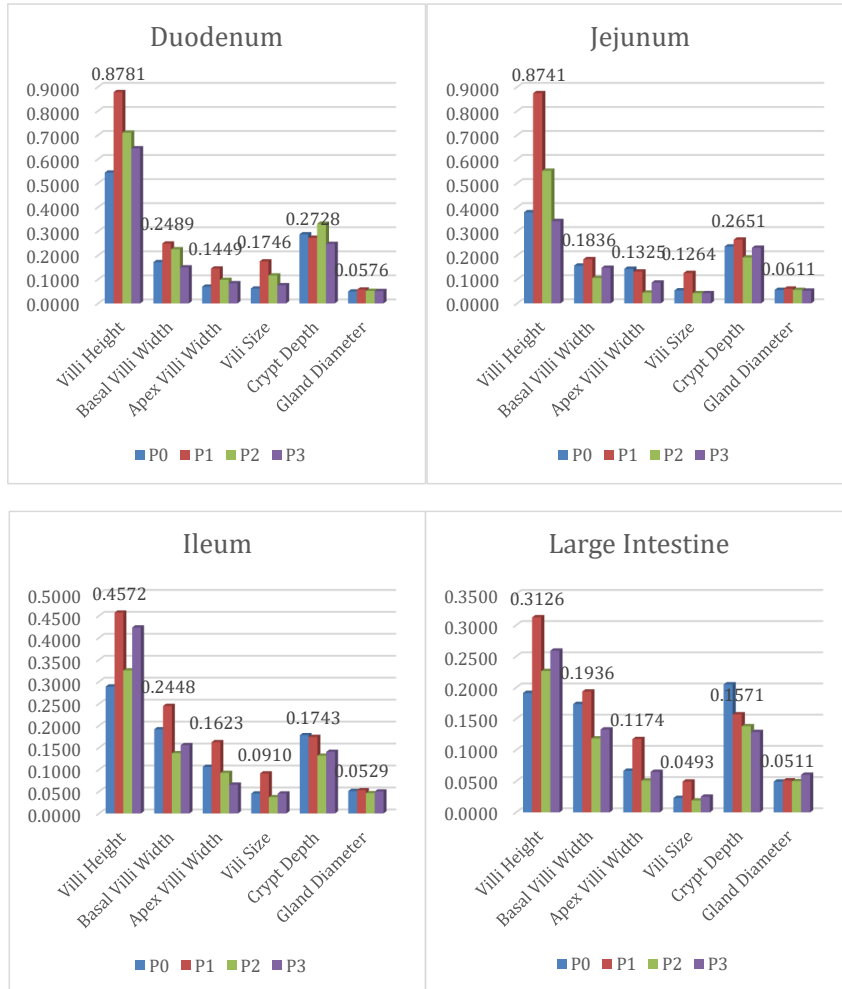
197 The highest mean measurement results for the depth of crypto varied in each treatment
198 in each organ (Table 1). According to Sun et al. (2005) and Smirnov et al. (2005) that into
199 crypto has no effect after broilers are more than 28 days old. Broilers in this study collected
200 samples of digestive tract organs at the age of 31 days. It is assumed that the development of
201 intestinal morphology is closely related to the role of micronutrients in line with the increasing
202 age of broilers (Harimurti and Rahayu, 2009).

203 Based on the data presented in Figure 1, supplementation of Black Cumin (*Nigella*
204 *sativa*) could increase the size of the research parameters, namely the size of the villi height,
205 the width of the villi apex, the basal width of the villi, the area of the villi, and the gland
206 diameter of all digestive tract organs compared to controls. P1 treatment with a dose of 36
207 mg/kg of broiler body weight had the highest effect on increasing the size of each of these
208 parameters.

209 One of the parameters that can be used to measure the quality of growth is the
210 morphological structure of the intestine (Wang *et al.*, 2008; Ningtias 2013). According to
211 Suprijatna *et al.*, (2008) the small intestine is the main organ for digestion and absorption of
212 digestive products. Various enzymes that enter this channel function to accelerate and
213 streamline the breakdown of carbohydrates, proteins and fats to facilitate the absorption
214 process. In adult chickens, the length of the small intestine is about 62 inches or 1.5 meters.

215 The digestive tract organs are supported by villi which are a special shape in the mucosa.
216 The villi are finger-shaped protrusions of the mucosa and are characteristic of the small
217 intestine. Increasing the number of villi will increase food absorption. Villi function to expand
218 the surface of the intestine which affects the process of absorption of food (Alfiansyah, 2011).
219 The development of the intestinal villi in broiler chickens is related to the function of the
220 intestine and growth of the chicken (Sun, 2004). The increase in villi causes more villi surface
221 area to absorb nutrients into the bloodstream (Mile *et al.* 2006; Rostinawati 2008).

222



223

224

225 Figure 1. Drinking water with Black Cumin (*Nigella sativa*) 36 mg/kg BW/day (P1) was the

226 highest measurement of each research parameter in the small intestine

227 (duodenum, jejunum, ileum) and large intestine

228

229 High villi indicate that the intestines are better off than short villi. Awad *et al.* (2008)

230 stated that the increase in the height of the villi in the intestine with digestive and absorption

231 functions occurs because of the intact villi form which is a smooth expression of the nutrient

Commented [a111]: Please add \pm SD and using histogram 2D is better and using * if significant different

232 transport system throughout the body. Rofiq (2003) states that the absorption of nutrients in
233 the intestine is influenced by the inner surface area of the intestine (folds, villi and microvilli)
234 and the length of transit of the digesta in the intestine.

235 The surface area of the intestine such as the height of the villi describes the area for
236 absorption of nutrients. Villi are small finger or leaf-like protrusions found on the mucous
237 membrane, 0.5 to 1.5 mm long and found only in the small intestine. The villi in the ileum are
238 finger-like in shape and shorter than the villi found on the duodenum and jejunum. One of the
239 parameters used to measure the quality of growth is the intestinal morphological structure
240 (Wang and Peng, 2008).

241 The intestinal gland (Lieberkuhn's gland) has a small hole that becomes the mouth of the
242 simplek tubulose gland. The intestinal glands are scattered between the villi, attached to the
243 mucous membrane. The intestinal glands and intestinal villi are covered by an epithelium,
244 consisting of goblet cells and enterocytes, among others. Goblet cells secrete mucus to
245 lubricate and protect the surface of the intestine, while the enterocytes in crypt secrete large
246 amounts of water and electrolytes (Pfeiffer and Macpherson, 1990).

247

248

CONCLUSION

249

250 Supplementation of Black Cumin 36 mg/kg BW/day through drinking water could
251 increasing the average sizes of villi height, villi apex width, basal villi width, villi area, and
252 gland diameter of small intestine (duodenum, jejunum, ileum) and large intestine of broiler.

253

254

ACKNOWLEDGMENTS

255

256 The authors would like to thank University of Lampung for providing research funding
257 sourced from the BLU University of Lampung 2020.

258

259

REFERENCES

260

261 Abun. 2007. **Pengukuran nilai pencernaan pakan yang mengandung limbah udang windu**
262 **produk fermentasi pada ayam broiler**. Research report. Padjajaran University,
263 Jatinangor.

264 Al-Beitawi N., S.S.E. Ghousein. 2008. Effect of feeding different levels of *Nigella sativa* seed
265 (black cumin) on performance, blood constituents and carcass characteristic of broiler
266 chick. **Int. J. Poult. Sci.** 7:775-778.

267 Alfiansyah, M. 2011. Anatomi dan Pencernaan Usus Halus. [http://www. sentra-edukasi.com/](http://www.sentra-edukasi.com/).
268 Diakses tanggal 20 Agustus 2020

269 Alzeer. 2008. **Diseases of Poultry**. 12th Ed. Blackwell, New Jersey.

270 Asmawati. 2014. The Effect of In Ovo Feeding on Hatching Weight and Small Intestinal Tissue
271 Development of Native Chicken. **Dissertation**. Hasanuddin University, Makassar.

272 Awad, W.A., K. Ghareeb, S. Nitclu, S. Pasteiner, S.A. Raheem, and J. Bohm. 2008. Effect of
273 dietary inclusion of probiotic, prebiotic and symbiotic on intestinal glucose absorption
274 of broiler chickens. **J. Poult. Sci.** 7: 688-691.

275 Badan Pusat Statistik. 2019. Populasi Ayam Ras Pedaging menurut Provinsi (Ekor) Tahun
276 2009-2018. [https://www.bps.go.id/dynamic/2015/12/18/1034/populasi-ayam-ras-
277 pedaging-menurut-provinsi-2009-2018.html](https://www.bps.go.id/dynamic/2015/12/18/1034/populasi-ayam-ras-pedaging-menurut-provinsi-2009-2018.html) Diakses pada tanggal 13 Agustus 2020.

278 Diamita, A.A. 2009. Pengaruh pemberian minyak wijen (*Sesamum indicum* Linn.) dengan cold
279 press bertingkat terhadap kerusakan histologis lambung mencit (*Mus musculus*) yang
280 diinduksi aspirin. **Thesis**. Faculty of Medical Universitas Sebelas Maret, Surakarta.

- 281 El-Dakhakhny M., N.J. Madi, N. Lambert, H.P. Ammon. 2002. Nigella sativa oil, nigellone
282 and derived thymoquinone inhibit synthesis of 5-lipoxygenase products in
283 polymorphonuclear leukocytes from rats. **J. Ethnopharmacol.** 81:161-164.
- 284 Fadillah, R. 2004. **Panduan Mengelola Peternakan Ayam Broiler Komersial.** PT
285 AgroMedia Pustaka, Tangerang.
- 286 Frappier, B.L. 2006. **Digestive System.** In: J.A. Eurell dan B.L. Frappier, Editor. Dellmann's
287 Textbook of Veterinary Histology. 6th Ed. Blackwell, Oxford.
- 288 Hamzah. 2013. Respon usus dan karakteristik karkas pada ayam ras pedaging dengan berat
289 badan awal berbeda yang dipuaskan setelah menetas. **Thesis.** Faculty of Animal
290 Science. Hasanuddin University, Makassar.
- 291 Harimurti,, S. dan E.S. Rahayu. 2009. Morfologi Usus Ayam Broiler yang disuplementasi
292 dengan Probiotik Strain Tunggal dan Campuran. **Agritech.** 29(3): 179-183.
- 293 Hendrik. 2009. **Habbatus Sauda.** Pustaka Iltazam, Solo.
- 294 Ibrahim, S. 2008. Hubungan ukuran-ukuran usus halus dengan berat badan broiler. **Agripet.**
295 8(2): 42-46.
- 296 Iji, P. A., R. J. Hughes, M. Choct and D. R. Tivey. 2001. Intestinal structure and function of
297 broiler chickens on wheat-based diets supplemented with microbial enzyme. Asian-Aust.
298 **J. Anim. Sci.** 14(1):54-60.
- 299 Kabir, S.M.L. 2010. Avian colibacillosis and salmonellosis: a closer look at epidemiology,
300 pathogenesis, diagnosis, control and public health concerns. **Int. J. Environ. Res. Public**
301 **Health.** 7:89-114.
- 302 Mile, R.D., G.D. Butcher, P.R. Henry, R.C. Littlel. 2006. Effect of antibiotic growth promoters
303 on broiler performance, intestinal growth parameters, and quantitative morphology. **J.**
304 **Poultry Sci.** 85:476-485.

- 305 Ningtias, A.S. 2013. Comparison of Growth Performance of Broilers, Kampong, and
306 Backcross3 (*Gallus gallus domesticus* Linnaeus, 1758) Based on Morphometri and
307 Histological Structure of Ileum and Breast Muscle. **Thesis**. Fakultas Biologi Universitas
308 Gajah Mada. Yogyakarta
- 309 Rostinawati T. 2008. Aktivitas antibakteri ekstrak etanol dan ekstrak air kelopak bunga rosela
310 (*Hibiscus sabdariffa* L.) terhadap *Mycobacterium tuberculosis* galur Labkes-026 (Multi
311 Drug Resisten) dan *Mycobacterium tuberculosis* galur H37Rv secara in vitro. **Thesis**.
312 Faculty of Pharmacy, Padjadjaran University, Bandung.
- 313 Siagian, Y.A. 2016. Gambaran Histologis dan Tinggi Vili Usus Halus Bagian Ileum Ayam Ras
314 Pedaging yang diberi Tepung Daun Kelor (*Moringa oleifera*) dalam ransum. **Thesis**.
315 Fakultas of Animal Science Hasanuddin University, Makasar.
- 316 Smirnov, A., R. Perez, E. Amit-Romach, D. Sklan, and Z. Uni. 2005. Mucin dynamics and
317 microbial populations in chicken small intestine changed by dietary probiotic and
318 antibiotic growth promotor supplementation. **J. of Nutrition**. 135: 187-192.
- 319 Sturkie, P.D., G.C. Whittow. 2000. **Sturkie's Avian Physiology**. Academic Press, Waltham.
- 320 Sugito, M.W., D.A. Astuti, E. Handharyani, Chairul. 2007. Histopatologi hati dan ginjal pada
321 ayam broiler yang dipapar cekaman panas dan diberi ekstrak kulitbatang Jaloh (*Salix*
322 tetrasperma Roxb). **JITV**. 12:6873.
- 323 Sun, X. 2004. Broiler performance and intestinal alterations when fed drug-free diets. **Thesis**.
324 Animal and Poultry Science. Blacksburg, Virginia
- 325 Sun, X., A. McElroy, Jr. Webb, A.E.K.E. Sefton and C. Novak. 2005. Broiler performance and
326 intestinal alterations when fed drug-free diets. **J. of Poult. Sci.**, 84: 1294-1302.
- 327 Suprijatna, E., U. Atmomarsono, R. Kartasudjana. 2008. **Ilmu Dasar Ternak Unggas**. Penebar
328 Swadaya, Jakarta.

- 329 Thippeswamy N.B., K.A. Naidu. 2005. Antioxidant potency of cumin varieties cumin, black
330 cumin and bitter cumin on antioxidant systems. **Euro Food Res Tech.** 220: 472-476
- 331 Topozoda H.H., H.A. Mazloun, M. El-Dakhkhny. 1965. The antibacterial properties of
332 *Nigella sativa* seeds. **J. Egypt Med.** 48:187-202.
- 333 Yusuf, M.S. 2014. Efektivitas Penggunaan Jintan Hitam (*Nigella sativa*) dalam Proses
334 Percepatan Penyembuhan Luka Setelah Pencabutan Gigi. **Thesis.** Faculty of Dentistry
335 Hasanuddin University, Makassar.
- 336 Wang, J.X. and K.M. Peng. 2008. Molecular, Cellular, and Developmental Biology
337 Developmental Morphology of the Small Intestine of African Ostrich Chicks. **J. Poultry**
338 **Sci.** 87: 2629-2635.

1 **EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH**
2 **DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND**
3 **LARGE INTESTINE OF BROILER**

4
5 **Authors**

6
7
8 **ABSTRACT**

9
10 This study aimed to determine the effect of Black Cumin (*Nigella sativa*)
11 supplementation through drinking water on the histology of small intestine and large intestine
12 of broiler. The research was conducted from April September 2020 in cage facility of
13 Integrated Field Laboratory, Faculty of Agriculture, University of Lampung. This research
14 was using a completely randomized design (CRD) with four treatment groups and three
15 replications (five heads per replication) with a total of 60 male broilers. The **treatment** were
16 drinking water without Black Cumin (P0, control); drinking water with Black Cumin 36
17 mg/kg BW/day (P1); 72 mg/kg BW/day (P2); and 144 mg/kg BW/day (P3). Three broilers
18 from each group were randomly **necropsed** at 31st days old, and samples of the small
19 intestine (duodenum, jejunum, ileum) and large intestine were fixed with 10% formalin
20 solution and sent to the Lampung Disease Investigation Center for histological preparations.
21 The observation of preparations was carried out microscopically using the Leica DM500®
22 Binocular Microscope to accurately calculate various parameter sizes. The results were
23 **analyze** descriptively. **The conclusion** of this study was the supplementation of Black Cumin
24 36 mg/kg BW/day through drinking water could increase the average sizes of villi height,

Commented [A1]: Treatments (plural subject)

Commented [A2]: necropsied

Commented [A3]: analyzed

Commented [A4]: Ini hasil yangdidapat. Perlu ditambahkan kesimpulan yang menerangkan hasil risetnya.

25 villi apex width, basal villi width, villi area, and gland diameter of small intestine
26 (duodenum, jejunum, ileum) and large intestine of broiler.

27

28 Keywords :black cumin, broiler, Leica DM500 microscope, large intestine, small intestine.

Commented [A5]: Tambahkan kata kunci lain yang ada di judul atau yg menjadi inti penelitian

29

30

ABSTRAK

31

32 *Penelitian ini bertujuan untuk mengetahui pengaruh suplementasi Jintan Hitam*
33 *(Nigella sativa) melalui air minum terhadap histologi usus halus dan usus besar broiler.*
34 *Penelitian dilakukan pada April-September 2020 di unit*
35 *kandang Laboratorium Lapang Terpadu, Fakultas Pertanian, Universitas Lampung.*
36 *Penelitian bersifat eksperimental menggunakan Rancangan Acak Lengkap (RAL)*
37 *dengan empat kelompok perlakuan dan tiga ulangan (lima ekor tiap ulangan) sehingga total 60*
38 *ekor broiler jantan. Perlakuan yang diberikannya yaitu pemberian air minum tanpa Jintan Hitam*
39 *(P0, kontrol); air minum dengan Jintan Hitam 36 mg/kg BB/hari (P1); 72 mg/kg BB/hari (P2);*
40 *dan 144 mg/kg BB/hari (P3). Tiga ekor dari tiap kelompok secara acak di nekrops pada hari ke-31*
41 *dan diambil sampel organ usus halus (duodenum, jejunum, ileum) dan usus besar (colon)*
42 *kemudian difiksasi dengan larutan formalin 10% dan dikirim ke Balai Veteriner Lampung*
43 *untuk pembuatan preparat histologi.*
44 *Pengamatan preparat dilakukan secara mikroskopis menggunakan Mikroskop Binokuler Leica*
45 *DM500® untuk menghitung berbagai ukuran parameter secara akurat.*
46 *Analisis hasil dilakukan secara deskriptif. Kesimpulan*
47 *penelitian ini yaitu suplementasi Jintan Hitam (Nigella sativa) 36 mg/kg BB/hari meningkatkan*
48 *rata-rata ukuran tingg vili, lebar apeks vili, lebar basal vili, luas vili, dan diameter kelenjar*

49 pada organ saluranpencernaanyaitu usus halus (duodenum, jejunum, ileum) dan usus besar
50 broiler.

51

52 Kata kunci :broiler, jintanhitam, mikroskopLeica DM500, usus besar, usus halus.

53

54

55

INTRODUCTION

56

57 The development of poultry farming in Indonesia is increasing. Broilers are one of the
58 fastest growing poultry. The relatively short period of time and the relatively lower
59 maintenance costs compared to ruminants make breeders prefer to cultivate broilers. In 2018,
60 the broiler population in Indonesia reached 1.89 billion heads and an increase of 2.26% from
61 the broiler population in 2017 of 1.85 billion heads (BPS, 2019).

62 Disease is a serious obstacle in the broiler farming industry. The high incidence of
63 disease can cause a decrease in productivity and even death of livestock which causes
64 significant losses for breeders. Administration of antibiotics in the livestock industry is used
65 for the treatment of livestock so as to reduce the risk of death and restore the condition of the
66 livestock to health, however, giving antibiotics for a long period of time can cause residual
67 buildup which has negative effects if consumed by humans.

Commented [A6]: The disease

68 The use of antibiotics needs to be reduced to prevent negative effects by providing
69 natural ingredients as immunomodulators. Immunomodulators can be defined as biological or
70 synthetic substances that can stimulate the innate immune system, adaptive or both. One of
71 the herbs that can act as an immunomodulator is Black Cumin (*Nigella sativa*). *Nigella*
72 *sativa* is a plant that has the potential as an immunostimulant that can stimulate and
73 strengthen the system by increasing the number, quality and activity of the body's immune

Commented [A7]: The administration

Commented [A8]: Tambahkan koma sebelum or

Commented [A9]: Tambahkan koma sebelum and

74 cells (Hendrik, 2009). *Nigella sativa* contains *thymoquinone*, saponins, zinc or zinc, *alpha-*
 75 *linolenic acid* (Omega 3) and *linoleic acid* (Omega 6) which functions in cell formation,
 76 maintains the immune system, and helps in the process of blood formation (Yusuf, 2014).

77 The solution needed for the above problems is to examine the effect of black cumin
 78 (*Nigella sativa*) supplementation through histological studies of the digestive organs of
 79 broilers which are expected to have the potential to increase the size of villi height, apex villi
 80 width, basal villi width, villi area, crypt depth and gland diameter of small intestine
 81 (duodenum, jejunum, ileum) and large intestine (colon).

Commented [A10]: Tambahkan koma sebelum and

Commented [A11]: Tambahkan sedikit penjelasan bagaimana kaitan *Nigella sativa* dengan sistem pencernaan (tinggi villi usus dll) sehingga perlu dilihat parameter tsb.

83 MATERIALS AND METHODS

85 Materials

86 This study used broiler cages, sprayer for cage disinfection, bamboo to make 12 cage
 87 plots, plastic tarpaulin for curtains, newspaper and used husks as litter, 12 bulbs of 15bulbs
 88 watt as a heating source for the area. *brooding*, a *hanging feeder* 12 pieces, *chick feeder tray*
 89 12 pieces, 12 pieces of chicken drinking places; 1 bucket, 1 *hand spray*, 1 water tray for
 90 *dipping*, 1 electric scale, *thermohyrometer* for measuring temperature and humidity, sack
 91 and plastic. Organ sampling equipment, namely necropsy equipment, *object glass*, *cover*
 92 *glass*, *refrigerator*, *microtome*, *light microscope*, *camera technology* and
 93 *software*Optilab®along with a laptop for taking tissue images and measuring the parameters
 94 of each organ.

95 Materials used in the study were 60 Day Old Chicks (DOC) male broiler Cobb CP 707
 96 strains kept for 30 days, rations, drinking water, extract of Black Cumin (*Nigella sativa*),
 97 vaccines of Newcastle Disease (ND), Avian Influenza (AI) and Infectious Bursal
 98 Disease (IBD), 10% formalin solution.

99

100 **Methods**

101 This research carried out for 6 months (April - September 2020) in the Integrated Field
102 Laboratory enclosure unit, Faculty of Agriculture, University of Lampung. This research was
103 experimental in nature using a completely randomized design (CRD) with four treatment
104 groups and three replications (five heads per replication) so a total of 60 male broilers. The
105 treatment dose was according to the broiler body weight, namely 1) drinking water without
106 black cumin (P0, control); drinking water with JintanHitam 36 mg / kg BW / day (P1);
107 drinking water with JintanHitam 72 mg / kg BW / day (P2); and drinking water with cumin
108 144 mg / kg BW / day (P3). On the 31st day, three from each group were randomly necropsied
109 and samples of the small intestine (duodenum, jejunum, ileum) and large intestine were then
110 fixed with 10% formalin solution and sent to the Lampung Veterinary Center for making
111 histological preparations. with Hematoxylin Eosin (HE) staining. The observation of
112 preparations was carried out microscopically using the Leica DM500® Binocular Microscope
113 Technology to accurately calculate various parameter sizes.

Commented [A12]: Missing a verb : was carried

114 The research parameters were villi height, villi apex width, basal villi width, villi area,
115 crypt depth and gland diameter. Observation of histological preparations using 10x
116 magnification objective lens. The calculation of each parameter was carried out as many as
117 three villi (villiheight, villi apex width, basal villi width, villi area, crypt depth) and nine
118 gland diameters in each digestive tract organ (duodenum, jejunum, ileum, large intestine).
119 There were three replications per organ, so that the total for each organ was obtained an
120 average of the nine villi and twenty-seven glands.

Commented [A13]: necropsied

121 The calculation of the surface area of the intestinal villi using the method of Ijiet *al.*
122 (2001) modified with the assumption that the villi model is an analogue of the trapezium
123 shape so that the average number of the apical widths of the villi plus the average basal width

124 of the villi is divided by two then multiplied by the height of the villi by the following
125 formula.

126

127 **Surface area=** $\frac{b + c}{2} \times a$
128

129 Description:

130 a = height of intestine villi

131 b = width of apex of intestine villi

132 c = width of basal of intestine villi

133

134 **Data analysis**

135 Measurement data for various research parameters were calculated the average of all
136 replications of each digestive tract organs (duodenum, jejunum, ileum, and large intestine)
137 then analyzed descriptively .

138

139 **RESULTS AND DISCUSSION**

140

141 **Parameter Measurements**

142 The average measurements of villi height, villi apex width, basal villi width, villi area,
143 crypt depth and gland diameter of the digestive tract organs (duodenum, jejunum, ileum,
144 large intestine) are presented in Table 1. Calculation of each parameter Three villi were
145 performed (height of villi, width of villi apex, basal width of villi, area of villi, depth of
146 crypt) and nine gland diameters on histopathological preparations in each digestive tract
147 organ (duodenum, jejunum, ileum, large intestine) in each treatment. There were three
148 replications for each organ, so that the total for each organ from each treatment was obtained
149 an average calculation of the nine villi and twenty-seven glands.

150 Based on Table 1, it is known that the treatment of giving black cumin (*Nigella sativa*)
 151 to the digestive organs of broilers through drinking water resulted in an increase in the size
 152 of the villi height, the width of the villi apex, the basal width of the villi, the area of the villi,
 153 and the diameter of the glands compared to the control (P0). Treatment P1 with a dose of 36
 154 mg / kg of broiler body weight gave the highest effect on the increase in the average size of
 155 the villi height, villi apex width, basal villi width, villi area, and gland diameter in the broiler
 156 digestive tract organs, namely the small intestine (duodenum, jejunum, ileum) and large
 157 intestine.

158

159 Table 1. Average measurement of each parameter in each treatment, giving Black Cumin
 160 (*Nigella sativa*) to the digestive organs of broilers through drinking water (in
 161 millimeter)

Commented [A14]: The average

Commented [A15]: millimeter

	Villi Height	Basal Villi Width	Apex Villi Width	Vili Size	Crypt Depth	Gland Diameter
Duodenum						
P0	0.5438	0.1716	0.0696	0.0616	0.2872	0.0497
P1	0.8781	0.2489	0.1449	0.1746	0.2728	0.0576
P2	0.7096	0.2251	0.0979	0.1161	0.3312	0.0523
P3	0.6446	0.1498	0.0838	0.0757	0.2473	0.0513
Jejunum						
P0	0.3789	0.1571	0.1439	0.0540	0.2366	0.0561
P1	0.8741	0.1836	0.1325	0.1264	0.2651	0.0611
P2	0.5516	0.1055	0.0448	0.0422	0.1910	0.0560
P3	0.3423	0.1480	0.0864	0.0423	0.2312	0.0529

Ileum

P0	0.2890	0.1916	0.1060	0.0454	0.1781	0.0511
P1	0.4572	0.2448	0.1623	0.0910	0.1743	0.0529
P2	0.3256	0.1371	0.0923	0.0370	0.1314	0.0461
P3	0.4234	0.1554	0.0656	0.0454	0.1403	0.0500

Large

Intestinum

P0	0.1914	0.1736	0.0666	0.0231	0.2051	0.0493
P1	0.3126	0.1936	0.1174	0.0493	0.1571	0.0511
P2	0.2266	0.1185	0.0510	0.0190	0.1381	0.0500
P3	0.2592	0.1328	0.0649	0.0251	0.1288	0.0602

- 162 P0 (drinking water without Black Cumin (*Nigella sativa*));
- 163 P1 (drinking water with Black Cumin (*Nigella sativa*) 36 mg/kg BW/day);
- 164 P2 (drinking water with Black Cumin (*Nigellasativa*) 72 mg/kg BW/day);
- 165 P3 (drinking water with Black Cumin (*Nigellasativa*)144 mg/kg BW/day)
- 166 Highlight text (highest measurement for each treatment).

167

168 The ability of digestion and absorption of food substances could be affected by the

169 surface area of the intestinal epithelium, the number of folds, and the number of villi and

170 microvilli that expand the absorption field (Austic and Nesheim, 1990; Ibrahim 2008) and

171 also influenced by the height and surface area of the villi organs digestive tract (Sugito et al.,

172 2007; Ibrahim 2008). These villi function to expand the surface of the intestine which affects

173 the process of absorption of food (Alfiansyah, 2011). The development of the intestinal villi

174 in broiler chickens is related to the function of the intestine and growth of the chicken (Sun,

175 2004). Villi are places for absorption of nutrients, the wider the villi, the more food

176 substances that will be absorbed, in the end it can have an impact on the growth of organs and
177 increased carcass (Asmawati, 2014).

178 The increase in villi height in the broiler intestine is closely related to an increase in
179 digestive function and absorption function due to the expansion of the absorption area and is
180 an expression of the smooth transportation system of nutrients throughout the body (Awad et
181 al., 2008). One of the parameters that can be used to measure the quality of growth is the
182 morphological structure of the intestine. The height of villi in all parts of the small intestine
183 (duodenum, jejunum, ileum) and large intestine in general increases (Ningtias, 2013).
184 Increasing the villi width and the height of the villi can expand the absorption area of the
185 villi. According to Asmawati (2014), the wider the villi the more food substances that will be
186 absorbed in the end can have an impact on the growth of the body's organs and according to
187 Rahmawati (2016) the higher the size of the villi, the wider the area of nutrient absorption by
188 the small intestine wall so that it will trigger Increased growth, according to Guyton (1997),
189 the more villous surface area indicates the more efficient absorption of nutrients that occurs.
190 **Efficiency** of nutrient absorption cannot be separated from the work of hormonal, nervous
191 and digestive glands in the digestive tract and its accessory glands.

Commented [A16]: The efficiency

192 Food, environment, and metabolic activity affects the number of intestinal glands.
193 Chickens generally eat food consisting of granules and are hard, so that a more active
194 secretion of intestinal glands is needed, to support the development of epithelial cells that
195 make up the villi (Mardhiah, 1991). Crypts contained in the intestinal villi, which are
196 composed of inline cylindrical epithelial cells. These glands produce mucus and several
197 enzymes for the metabolism of peptides, fats, carbohydrates, and intestinal juices (mucin)
198 which function to protect the intestinal mucosa (Aughey and Frye, 2001 The increase in the
199 average size of the gland diameter in the treatment with black cumin in drinking water
200 showed an increase in the size of the gland diameter in each organ **teruta**. However, in P1

Commented [A17]: ???

201 treatment with the highest increase in size compared to other treatments, it can support the
202 development of epithelial cells that make up the villi, which will increase the absorption of
203 nutrients in the digestive tract.

204 The highest mean measurement results for the depth of crypto varied in each treatment
205 in each organ (Table 1). According to Sun et al. (2005) and Smirnov et al. (2005) that into
206 crypto has no effect after broilers are more than 28 days old. Broilers in this study collected
207 samples of digestive tract organs at the age of 31 days. It is assumed that the development of
208 intestinal morphology is closely related to the role of micronutrients in line with the
209 increasing age of broilers (Harimurti and Rahayu, 2009).

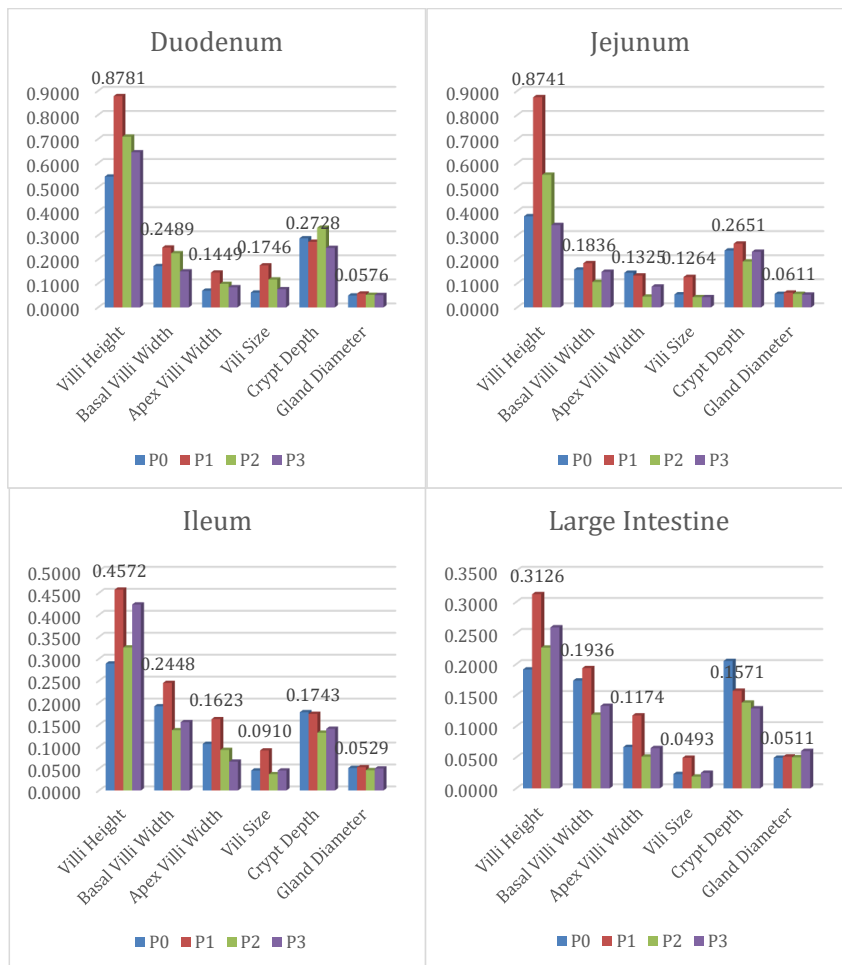
210 Based on the data presented in Figure 1, supplementation of Black Cumin (*Nigella*
211 *sativa*) could increase the size of the research parameters, namely the size of the villi height,
212 the width of the villi apex, the basal width of the villi, the area of the villi, and the gland
213 diameter of all digestive tract organs compared to controls. P1 treatment with a dose of 36
214 mg/kg of broiler body weight had the highest effect on increasing the size of each of these
215 parameters.

216 One of the parameters that can be used to measure the quality of growth is the
217 morphological structure of the intestine (Wang et al., 2008; Ningtias 2013). According to
218 Suprijatna et al., (2008) the small intestine is the main organ for digestion and absorption of
219 digestive products. Various enzymes that enter this channel function to accelerate and
220 streamline the breakdown of carbohydrates, proteins and fats to facilitate the absorption
221 process. In adult chickens, the length of the small intestine is about 62 inches or 1.5 meters.

222 The digestive tract organs are supported by villi which are a special shape in the
223 mucosa. The villi are finger-shaped protrusions of the mucosa and are characteristic of the
224 small intestine. Increasing the number of villi will increase food absorption. Villi function to
225 expand the surface of the intestine which affects the process of absorption of food

226 (Alfiansyah, 2011). The development of the intestinal villi in broiler chickens is related to the
 227 function of the intestine and growth of the chicken (Sun, 2004). The increase in villi causes
 228 more villi surface area to absorb nutrients into the bloodstream (Mile *et al.* 2006; Rostinawati
 229 2008).

230



231

232

233 Figure 1. Drinking water with Black Cumin (*Nigella sativa*) 36 mg/kg BW/day (P1) was the

234 highest measurement of each research parameter in the small intestine

235 (duodenum, jejunum, ileum) and large intestine

236

237 High villi indicate that the intestines are better off than short villi. Awadet *al.* (2008)
238 stated that the increase in the height of the villi in the intestine with digestive and absorption
239 functions occurs because of the intact villi form which is a smooth expression of the nutrient
240 transport system throughout the body. Rofiq (2003) states that the absorption of nutrients in
241 the intestine is influenced by the inner surface area of the intestine (folds, villi and microvilli)
242 and the length of transit of the digesta in the intestine.

243 The surface area of the intestine such as the height of the villi describes the area for
244 absorption of nutrients. Villi are small finger or leaf-like protrusions found on the mucous
245 membrane, 0.5 to 1.5 mm long and found only in the small intestine. The villi in the ileum are
246 finger-like in shape and shorter than the villi found on the duodenum and jejunum. One of
247 the parameters used to measure the quality of growth is the intestinal morphological structure
248 (Wang and Peng, 2008).

249 The intestinal gland (Lieberkuhn's gland) has a small hole that becomes the mouth of
250 the simplektubulose gland. The intestinal glands are scattered between the villi, attached to
251 the mucous membrane. The intestinal glands and intestinal villi are covered by an
252 epithelium, consisting of goblet cells and enterocytes, among others. Goblet cells secrete
253 mucus to lubricate and protect the surface of the intestine, while the enterocytes in crypt
254 secrete large amounts of water and electrolytes (Pfeiffer and Macpherson, 1990).

255

256

CONCLUSION

257

Commented [A18]: jejunum

258 Supplementation of Black Cumin 36 mg/kg BW/day through drinking water could
 259 increasing the average sizes of villi height, villi apex width, basal villi width, villi area, and
 260 gland diameter of small intestine (duodenum, jejunum, ileum) and large intestine of broiler.

Commented [A19]: increase/be increasing

261

262

ACKNOWLEDGMENTS

263

264 The authors would like to thank University of Lampung for providing research funding
 265 sourced from the BLU University of Lampung 2020.

266

267

REFERENCES

268

269 Abun. 2007. Pengukurannilaikecernaanpakan yang
 270 mengandunglimbahandangwinduprodukfermentasi pada ayam broiler. Research
 271 report.Padjajaran University,Jatinangor.

Commented [A20]: Pustaka yang belum ada :

- 1.Austic & Neishen, 1990
- 2.Rahmawati, 2016
- 3.Guyton, 1997
- 4.Mardhiah, 1991
- 5.Aughey & Friye, 2001
- 6.Wang et al, 2008
- 7.Ningtias, 2013
- 8.Rofiq, 2003
- 9.Pfeiffer&Macpherson, 1990

Agar dilengkapi

272 Al-Beitawi N., S.S.E. Ghousein. 2008. Effect of feeding different levels of Nigella sativa
 273 seed (black cumin) on performance, blood constituents and carcass characteristic of
 274 broiler chick. **Int. J. Poult. Sci.** 7:775-778.

Commented [A21]: Tidak ada dlm artikel

Commented [A22]: Tidak ada dlm artikel

275 Alfiansyah, M. 2011. Anatomi dan Pencernaan Usus Halus. [http://www. sentra-edukasi.com/](http://www.sentra-edukasi.com/).
 276 Diaksestanggal 20 Agustus 2020

277 Alzeer. 2008. **Diseases of Poultry**. 12th Ed. Blackwell, New Jersey.

Commented [A23]: Tidak ada dlm artikel

278 Asmawati. 2014. The Effect of In Ovo Feeding on Hatching Weight and Small Intestinal
 279 Tissue Development of Native Chicken. **Dissertation**. Hasanuddin University,
 280 Makassar.

- 281 Awad, W.A., K. Ghareeb, S. Nitclu, S. Pasteiner, S.A. Raheem, and J. Bohm. 2008. Effect of
282 dietary inclusion of probiotic, prebiotic and symbiotic on intestinal glucose absorption
283 of broiler chickens. **J. Poult. Sci.**7: 688-691.
- 284 Badan Pusat Statistik. 2019. Populasi Ayam Ras Pedaging menurut Provinsi (Ekor) Tahun
285 2009-2018. [https://www.bps.go.id/dynamic/ptb/2015/12/18/1034/populasi-ayam-ras-](https://www.bps.go.id/dynamic/ptb/2015/12/18/1034/populasi-ayam-ras-pedaging-menurut-provinsi-2009-2018.html)
286 [pedaging-menurut-provinsi-2009-2018.html](https://www.bps.go.id/dynamic/ptb/2015/12/18/1034/populasi-ayam-ras-pedaging-menurut-provinsi-2009-2018.html) Diakses pada tanggal 13 Agustus 2020.
- 287 **Diamita, A.A.** 2009. Pengaruh pemberian minyak wijen (*Sesamum indicum* Linn.) dengan cold
288 press bertingka terhadap kerusakan histologi lambung mencit (*Mus musculus*) yang
289 diinduksi aspirin. **Thesis**. Faculty of Medical Universitas Sebelas Maret, Surakarta.
- 290 **El-Dakhkhny M., N.J. Madi, N. Lambert, H.P. Ammon.** 2002. Nigella sativa oil, nigellone
291 and derived thymoquinone inhibit synthesis of 5-lipoxygenase products in
292 polymorphonuclear leukocytes from rats. **J. Ethnopharmacol.** 81:161-164.
- 293 **Fadillah, R.** 2004. **Panduan Mengelola Peternakan Ayam Broiler Komersial.** PT
294 AgroMedia Pustaka, Tangerang.
- 295 **Frappier, B.L.** 2006. **Digestive System.** In: J.A. Eurell dan B.L. Frappier, Editor.
296 Dellmann's Textbook of Veterinary Histology. 6th Ed. Blackwell, Oxford.
- 297 **Hamzah.** 2013. Respon usus dan karakteristik karkas pada ayam ras pedaging dengan berat
298 badan awal berbeda yang dipuasakan setelah menetas. **Thesis**. Faculty of Animal Science.
299 Hasanuddin University, Makassar.
- 300 **Harimurti, S. dan E.S. Rahayu.** 2009. Morfologi Usus Ayam Broiler yang
301 disuplementasi dengan Probiotik Strain Tunggal dan Campuran. **Agri-tech.** 29(3): 179-
302 183.
- 303 **Hendrik.** 2009. **Habbatus Sauda.** Pustaka Iltazam, Solo.
- 304 **Ibrahim, S.** 2008. Hubungan ukuran-ukuran usus halus dengan berat badan broiler. **Agripet.**
305 8(2): 42-46.

Commented [A24]: Tidak ada dlm artikel

Commented [A25]: Tidak ada dlm artikel

Commented [A26]: Tidak ada dlm artikel

Commented [A27]: Tidak ada dlm artikel

Commented [A28]: Tidak ada dlm artikel

- 306 Iji, P. A., R. J. Hughes, M. Choct and D. R. Tivey. 2001. Intestinal structure and function of
307 broiler chickens on wheat-based diets supplemented with microbial enzyme. Asian-
308 Aust. **J. Anim. Sci.** 14(1):54-60.
- 309 Kabir, S.M.L. 2010. Avian colibacillosis and salmonellosis: a closer look at epidemiology,
310 pathogenesis, diagnosis, control and public health concerns. **Int. J. Environ. Res.**
311 **Public Health.** 7:89-114.
- 312 Mile, R.D., G.D. Butcher, P.R. Henry, R.C. Littlel. 2006. Effect of antibiotic growth
313 promotors on broiler performance, intestinal growth parameters, and quantitative
314 morphology. **J. Poultry Sci.**85:476-485.
- 315 Ningtias, A.S. 2013. Comparison of Growth Performance of Broilers, Kampong, and
316 Backcross3 (*Gallus gallusdomesticus* Linnaeus, 1758) Based on Morphometri and
317 Histological Structure of Ileum and Breast Muscle. **Thesis.** FakultasBiologi Universitas
318 Gajah Mada. Yogyakarta
- 319 Rostinawati T. 2008. Aktivitasantibakteriekstraketanol dan ekstrak air kelopakbungarosela
320 (*Hibiscus sabdariffa* L.) terhadap Mycobacterium tuberculosis galur Labkes-026 (Multi
321 Drug Resisten) dan Mycobacterium tuberculosis galur H37Rv secara in vitro.**Thesis.**
322 Faculty of Pharmacy, Padjadjaran University, Bandung.
- 323 Siagian, Y.A. 2016. Gambaran Histologis dan Tinggi Vili Usus Halus Bagian Ileum Ayam
324 Ras Pedaging yang diberiTepungDaunKelor (*Moringa oleifera*) dalamransum. **Thesis.**
325 Fakultasof Animal Science HasanuddinUniversity,Makasar.
- 326 Smirnov, A., R. Perez, E. Amit-Romach, D. Sklan, and Z. Uni. 2005. Mucin dynamics and
327 microbial populations in chicken small intestine changed by dietary probiotic and
328 antibiotic growth promotor supplementation. **J. of Nutrition.** 135: 187-192.
- 329 Sturkie, P.D., G.C. Whittow. 2000. **Sturkie's Avian Physiology.** Academic Press, Waltham.

Commented [A29]: Tidak ada dlm artikel

Commented [A30]: Tidak ada dlm artikel

Commented [A31]: Tidak ada dlm artikel

- 330 Sugito, M.W., D.A. Astuti, E. Handharyani, Chairul. 2007. Histopatologi hati dan ginjal pada
331 ayam broiler yang dipaparkan panas dan diberikan ekstrak kulit batang Jaloh (*Salix*
332 *tetrasperma* Roxb). **JITV**, 12:6873.
- 333 Sun, X. 2004. Broiler performance and intestinal alterations when fed drug-free diets. **Thesis**.
334 Animal and Poultry Science. Blacksburg, Virginia
- 335 Sun, X., A. McElroy, Jr. Webb, A.E.K.E. Sefton and C. Novak. 2005. Broiler performance
336 and intestinal alterations when fed drug-free diets. **J. of Poult. Sci.**, 84: 1294-1302.
- 337 Suprijatna, E., U. Atmomarsono, R. Kartasudjana. 2008. **Ilmu Dasar Ternak Unggas**.
338 Penebar Swadaya, Jakarta.
- 339 Thippeswamy N.B., K.A. Naidu. 2005. Antioxidant potency of cumin varieties cumin, black
340 cumin and bitter cumin on antioxidant systems. **Euro Food Res Tech.** 220: 472-476
- 341 Topozoda H.H., H.A. Mazloun, M. El-Dakhakhny. 1965. The antibacterial properties of
342 *Nigella sativa* seeds. **J. Egypt Med.** 48:187-202.
- 343 Yusuf, M.S. 2014. Efektivitas Penggunaan Jintan Hitam (*Nigella sativa*) dalam Proses
344 Percepatan Penyembuhan Luka Setelah Pencabutan Gigi. **Thesis**. Faculty of Dentistry
345 Hasanuddin University, Makassar.
- 346 Wang, J.X. and K.M. Peng. 2008. Molecular, Cellular, and Developmental Biology
347 Developmental Morphology of the Small Intestine of African Ostrich Chicks. **J.**
348 **Poultry Sci.** 87: 2629-2635.

Commented [A32]: Tidak ada dlm artikel

Commented [A33]: Tidak ada dlm artikel

Commented [A34]: Tidak ada dlm artikel

Because I can't see any suggestions from reviewers in article file 19774-60754-1-RV or in Online Journal JKH.

Thank you very much for your help

Best regards

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

Author
2021-04-25 04:09 PM

Subject: EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH ~~DELETE~~ DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER

Dear Mr. Tongku

I have been submitted revision of our article and have adjusted it according to the reviewers' suggestions.

Thank you for your attention and cooperation

Best regards

Chief Editor,

Prof. Dr. Drh. Tongku N. Siregar, MP
Chief Editor E-mail: siregar@unsyiah.ac.id
Website: <http://jurnal.unsyiah.ac.id/JKH>
e-mail : jkh@unsyiah.ac.id

Close



1 **EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH**
2 **DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND**
3 **LARGE INTESTINE OF BROILER**

4
5 **Authors**

6
7 **ABSTRACT**

8
9 This study aimed to determine the effect of Black Cumin (*Nigella sativa*)
10 supplementation through drinking water on the histology of small intestine and large intestine
11 of broiler. The research was conducted from April September 2020 in cage facility of Integrated
12 Field Laboratory, Faculty of Agriculture, University of Lampung. This research was using a
13 completely randomized design with four treatment groups and three replications (five heads
14 per replication) with a total of 60 male broilers. The **treatments** were drinking water without
15 Black Cumin (P0, control); drinking water with Black Cumin 36 mg/kg BW/day (P1); 72
16 mg/kg BW/day (P2); and 144 mg/kg BW/day (P3). Three broilers from each group were
17 randomly **necropsied** at 31st days old, and samples of the small intestine (duodenum, jejunum,
18 ileum) and large intestine were fixed with 10% formalin solution and sent to the Lampung
19 Disease Investigation Center for histological preparations. The observation of preparations was
20 carried out microscopically using the Leica DM500[®] Binocular Microscope to accurately
21 calculate various parameter sizes. The results were **analyzed statistically with one way Analysis**
22 **of Variance at significant level 5% and if significant then continue with Tukey test. The**
23 **conclusions of this study were the supplementation of Black Cumin (*Nigella sativa*) 72 mg/kg**
24 **BW/day through drinking water could increase significantly (P<0.05) to 1) the average sizes**
25 **of villi height and villi area of small intestine (duodenum, jejunum, ileum) and large intestine**

26 (colon) of broiler; 2) the average sizes of basal villi width, villi apex width, and gland diameter
27 of broiler duodenum; and 3) the average size of apex villi width of broiler colon.

28

29 Keywords : Broiler, Histology, Large intestine, *Nigella sativa*, Small intestine.

30

31

ABSTRAK

32

33 *Penelitian ini bertujuan untuk mengetahui pengaruh pengaruh suplementasi Jintan*
34 *Hitam (*Nigella sativa*) melalui air minum terhadap histologi usus halus dan usus besar broiler.*
35 *Penelitian dilakukan pada April-September 2020 di unit kandang Laboratorium Lapang*
36 *Terpadu, Fakultas Pertanian, Universitas Lampung. Penelitian bersifat eksperimental*
37 *menggunakan Rancangan Acak Lengkap dengan empat kelompok perlakuan dan tiga ulangan*
38 *(lima ekor tiap ulangan) sehingga total 60 ekor broiler jantan. Perlakuan yang diberikan yaitu*
39 *pemberian air minum tanpa Jintan Hitam (P0, kontrol); air minum dengan Jintan Hitam 36*
40 *mg/kg BB/hari (P1); 72 mg/kg BB/hari (P2); dan 144 mg/kg BB/hari (P3). Tiga ekor dari tiap*
41 *kelompok secara acak dinekropsi pada hari ke-31 dan diambil sampel organ usus halus*
42 *(duodenum, jejunum, ileum) dan usus besar (colon) kemudian difiksasi dengan larutan*
43 *formalin 10% dan dikirim ke Balai Veteriner Lampung untuk pembuatan preparat histologi.*
44 *Pengamatan preparat dilakukan secara mikroskopis menggunakan Mikroskop Binokuler Leica*
45 *DM500® untuk menghitung berbagai ukuran parameter secara akurat. Analisis hasil dilakukan*
46 *secara statistik menggunakan analisis sidik ragam satu arah dengan taraf signifikansi 5% dan*
47 *jika hasilnya signifikan maka dilanjutkan dengan uji lanjut Tukey. Kesimpulan penelitian ini*
48 *yaitu suplementasi Jintan Hitam (*Nigella sativa*) 72 mg/kg BB/hari dapat meningkatkan secara*
49 *signifikan ($P < 0.05$) terhadap 1) ukuran rata-rata tinggi vili dan luas vili usus halus*
50 *(duodenum, jejunum, ileum) dan usus besar broiler; 2) ukuran rata-rata tinggi vili, lebar vili*

51 *basal, lebar puncak vili, luas vili, dan diameter kelenjar duodenum broiler; dan 3) ukuran*
52 *rata-rata lebar puncak vili usus besar broiler.*

53

54 *Kata kunci : Broiler, Histologi, Nigella sativa, Usus besar, Usus halus.*

55

56

INTRODUCTION

57

58 The development of poultry farming in Indonesia is increasing. Broilers are one of the
59 fastest growing poultry. The relatively short period of time and the relatively lower
60 maintenance costs compared to ruminants make breeders prefer to farm broilers. In 2018, the
61 broiler population in Indonesia reached 1.89 billion heads and an increase of 2.26% from the
62 broiler population in 2017 of 1.85 billion heads (BPS, 2019).

63 Intensive maintenance of broiler can make it easy to experience stress, resulting in a
64 decrease in the ability of the immune system, it will be easy to contract various kinds of
65 diseases. The disease is a serious obstacle in the broiler farming industry. The high incidence
66 of disease can cause a decrease in productivity and even death of livestock which causes
67 significant losses for breeders. The diseases can lead to decrease in the function of body organs,
68 one of which is the digestive organ and the administration of antibiotics for a long period of
69 time can cause residual buildup, antibiotic resistance in bacteria and could be has negative
70 effects if consumed by humans (Marshall and Levy, 2011). One of effort to maintain the
71 quality of the digestive tract to function properly and to reduce the residual of antibiotics in
72 broiler by providing natural herbs such as Black cumin (*Nigella sativa*) that is believed can
73 improve organ function digestion such as research that has been carried out in mice (Rostika,
74 2012).

75 This study was conducted to determine the effect of black cumin supplementation
76 through histological studies of the digestive organs of broilers which are expected to have the
77 potential to increase the size of villi height, apex villi width, basal villi width, villi area, crypt
78 depth and gland diameter of small intestine (duodenum, jejunum, ileum) and large intestine
79 (colon).

80

81 MATERIALS AND METHODS

82

83 Materials

84 This study used broiler cages, sprayer for cage disinfection, bamboo to make 12 cage
85 plots, plastic tarpaulin for curtains, newspaper and used husks as litter, 12 bulbs of 15 bulbs
86 watt as a heating source for the area brooding, 12 pieces hanging feeder, 12 pieces chick feeder
87 tray, 12 pieces of chicken drinking places; 1 bucket, 1 hand spray, 1 water tray for dipping, 1
88 electric scale, thermohygrometer for measuring temperature and humidity, sack and plastic.
89 Organ sampling equipments, namely necropsy equipment, object glass, cover glass,
90 refrigerator, microtome, **Leica DM500® Binocular Microscope Technology connected with a**
91 **computer** for taking tissue images and measuring the parameters of each organ.

92 Materials used in the study were 60 Day Old Chicks (DOC) male broiler Cobb CP 707
93 strains kept for 30 days, rations, drinking water, extract of Black Cumin (*Nigella sativa*),
94 vaccines of Newcastle Disease (ND), Avian Influenza (AI) and Infectious Bursal Disease
95 (IBD), 10% formalin solution.

96

97 Methods

98 This research was carried out for 6 months (April - September 2020) in the Integrated
99 Field Laboratory enclosure unit, Faculty of Agriculture, University of Lampung. This research

100 was experimental in nature using a completely randomized design (CRD) with four treatment
 101 groups and three replications (five heads per replication) so a total of 60 male broilers. The
 102 treatment dose was according to the broiler body weight, namely 1) drinking water without
 103 black cumin (P0, control); drinking water with Jintan Hitam 36 mg / kg BW / day (P1); drinking
 104 water with Jintan Hitam 72 mg / kg BW / day (P2); and drinking water with cumin 144 mg /
 105 kg BW / day (P3). On the 31st day, three from each group were randomly necropsied and
 106 samples of the small intestine (duodenum, jejunum, ileum) and large intestine (colon) were
 107 then fixed with 10% formalin solution and sent to the Lampung Veterinary Disease
 108 Investigation Center for making histological preparations. with Hematoxylin Eosin (HE)
 109 staining. The observation of preparations was carried out microscopically using the Leica
 110 DM500@ Binocular Microscope Technology to accurately calculate various parameter sizes.

111 The research parameters were villi height, villi apex width, basal villi width, villi area,
 112 crypt depth and gland diameter. Observation of histological preparations using 10x
 113 magnification objective lens. The calculation of each parameter was carried out as many as
 114 three villi (villi height, villi apex width, basal villi width, villi area, crypt depth) and three gland
 115 diameters in each digestive tract organ (duodenum, jejunum, ileum, colon). There were three
 116 replications per organ, so that the total for each organ was obtained an average of the nine villi
 117 and nine gland diameters.

118 The calculation of the surface area of the intestinal villi using the method of Iji *et al.*
 119 (2001) modified with the assumption that the villi model is an analogue of the trapezium shape
 120 so that the average number of the apical widths of the villi plus the average basal width of the
 121 villi is divided by two then multiplied by the height of the villi by the following formula.

$$122 \quad \text{Surface area} = \frac{b + c}{2} \times a$$

123 Description:

124 a = height of intestine villi

126 b = width of apex of intestine villi

127 c = width of basal of intestine villi

128

129 **Data analysis**

130 Measurement data for various research parameters were calculated the average of all
131 replications of each digestive tract organs (duodenum, jejunum, ileum, and colon) then
132 analyzed with *one way Analysis of Variance* then continue with Tukey test. Different
133 supercripts with letters in the same column indicate significant differences ($P<0.05$).

134

135 **RESULTS AND DISCUSSION**

136

137 The average measurements of villi height, villi apex width, basal villi width, villi area,
138 crypt depth and gland diameter of the digestive tract organs (duodenum, jejunum, ileum, colon)
139 presented in each table. Calculation of each parameter nine villi were performed (villi height,
140 basal villi width, apex villi width, villi area, crypt depth) and nine gland diameters on
141 histopathological preparations in each digestive tract organ (duodenum, jejunum, ileum, large
142 intestine) in each treatment.

143 The average measurement of each parameter in broiler duodenum that supplemented by
144 black cumin (*Nigella sativa*) through drinking water presented in Table 1. Supplementation of
145 black cumin (*Nigella sativa*) with a dose of 72 mg/kg of broiler body weight (P2) gave the
146 significant effect (Figure 1) on the increase in the average size of villi height, basal villi width,
147 villi apex width, villi area, and gland diameter of duodenum.

148

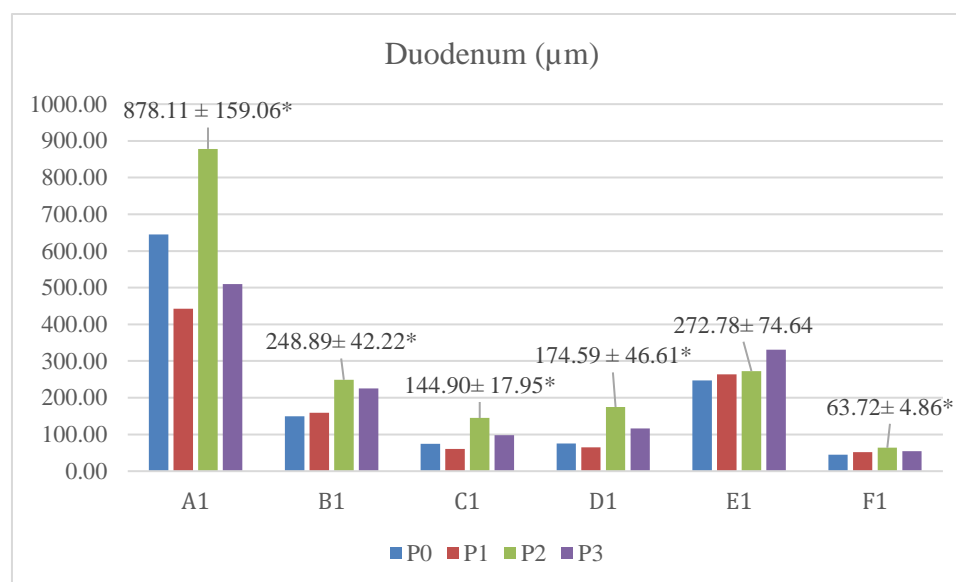
149

150

151 Table 1. The average measurement of each parameter in broiler duodenum supplemented by
 152 black cumin (*Nigella sativa*) through drinking water.

Treatment	Duodenum					
	A1	B1	C1	D1	E1	F1
	-----Mean \pm SD (μm)-----					
P0	644.55 \pm 71.73 ^{ab}	149.78 \pm 7.72 ^a	74.96 \pm 8.11 ^a	75.70 \pm 1.35 ^{ab}	247.33 \pm 37.83 ^a	44.44 \pm 4.67 ^a
P1	442.78 \pm 2.67 ^a	158.89 \pm 32.35 ^a	60.81 \pm 20.43 ^a	65.24 \pm 33.66 ^a	263.78 \pm 51.42 ^a	52.09 \pm 3.37 ^{ab}
P2	878.11 \pm 159.06 ^b	248.89 \pm 42.22 ^b	144.90 \pm 17.95 ^b	174.59 \pm 46.61 ^b	272.78 \pm 74.64 ^a	63.72 \pm 4.86 ^b
P3	509.55 \pm 135.72 ^a	225.07 \pm 26.34 ^{ab}	97.93 \pm 19.65 ^a	116.13 \pm 50.90 ^{ab}	331.22 \pm 112.74 ^a	54.1 \pm 7.02 ^{ab}

153 Information : Drinking water without *Nigella sativa* (P0), Drinking water with *Nigella sativa*
 154 36 mg/kg BW/day (P1), 72 mg/kg BW/day (P2), 144 mg/kg BW/day (P3); A1
 155 (villi height of duodenum), B1 (basal villi width of duodenum), C1 (apex villi
 156 width of duodenum), D1 (vili area of duodenum), E1 (crypt depth of duodenum),
 157 F1 (gland diameter of duodenum). Different supercripts with letters in the same
 158 column indicate significant differences (P<0.05).
 159



160

161 Figure 1. The supplementation of black cumin (*Nigella sativa*) 72 mg/kg BW (P2) in drinking
 162 water increasing average sizes of villi height (A1), basal villi width (B1), villi apex
 163 width (C1), villi area (D1), and gland diameter (F1) of broiler duodenum.
 164 Asterisk (*) indicates the most significant effect (P<0.05).
 165

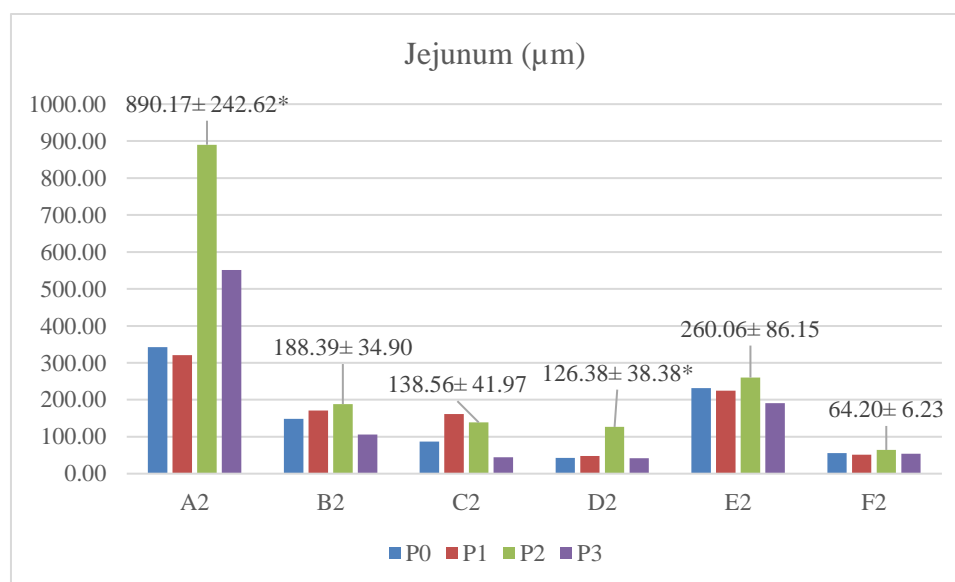
166 The average measurement of each parameter in broiler jejunum that supplemented by
 167 black cumin (*Nigella sativa*) through drinking water presented in Table 2. Supplementation of
 168 black cumin (*Nigella sativa*) with a dose of 72 mg/kg of broiler body weight (P2) gave the
 169 significant effect on the increase in the average size of villi height and villi area of jejunum.

170

171 Table 2. The average measurement of each parameter in broiler jejunum supplemented by black
 172 cumin (*Nigella sativa*) through drinking water.

Treatment	Jejunum					
	A1	B1	C1	D1	E1	F1
	-----Mean \pm SD (μm)-----					
P0	342.34 \pm 111.40 ^a	148.01 \pm 19.47 ^a	86.43 \pm 39.58 ^a	42.28 \pm 22.93 ^a	231.22 \pm 134.32 ^a	55.83 \pm 2.87 ^a
P1	320.67 \pm 176.51 ^a	170.63 \pm 58.27 ^a	161.23 \pm 93.12 ^a	47.71 \pm 32.24 ^a	224.44 \pm 95.06 ^a	51.52 \pm 3.82 ^a
P2	890.17 \pm 242.62 ^b	188.39 \pm 34.90 ^a	138.56 \pm 41.97 ^a	126.38 \pm 38.38 ^b	260.06 \pm 86.15 ^a	64.20 \pm 6.23 ^a
P3	551.53 \pm 128.51 ^{ab}	105.5 \pm 20.63 ^a	44.80 \pm 8.44 ^a	42.15 \pm 17.44 ^a	191.00 \pm 38.40 ^a	54.16 \pm 7.41 ^a

173 Information : Drinking water without *Nigella sativa* (P0), Drinking water with *Nigella sativa*
 174 36 mg/kg BW/day (P1), 72 mg/kg BW/day (P2), 144 mg/kg BW/day (P3); A1
 175 (villi height of jejunum), B1 (basal villi width of jejunum), C1 (apex villi width
 176 of jejunum), D1 (villi area of jejunum), E1 (crypt depth of jejunum), F1 (gland
 177 diameter of jejunum). Different supercripts with letters in the same column
 178 indicate significant differences (P<0.05).
 179



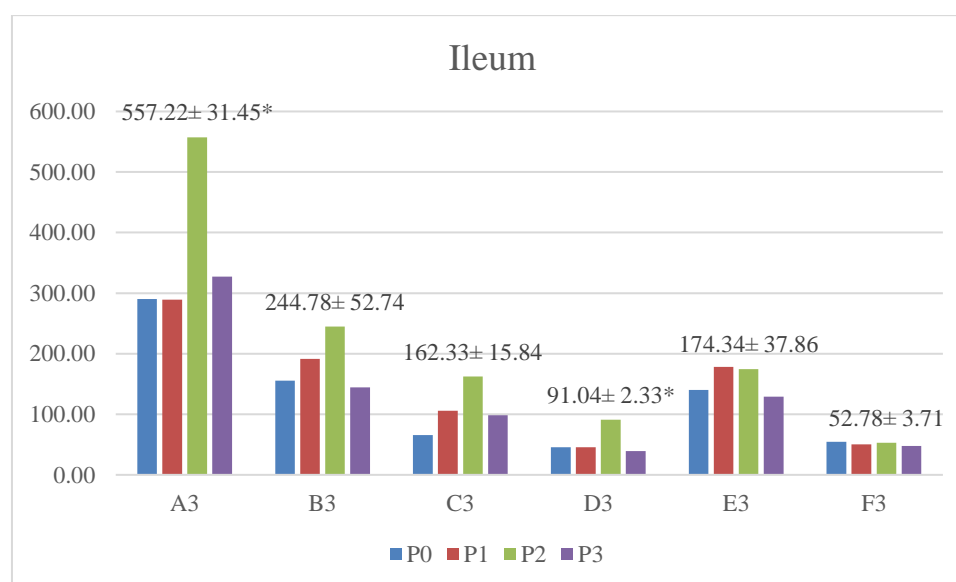
180
 181 Figure 2. The supplementation of black cumin (*Nigella sativa*) 72 mg/kg BW (P2) in drinking
 182 water increase average sizes of villi height (A2) and villi area (D1) of broiler jejunum.
 183 Asterisk (*) indicates the most significant effect (P<0.05).
 184

185 The average measurement of each parameter in broiler ileum that supplemented by black
 186 cumin (*Nigella sativa*) through drinking water presented in Table 3. Supplementation of black
 187 cumin (*Nigella sativa*) with a dose of 72 mg/kg of broiler body weight (P2) gave the significant
 188 effect on the increase in the average size of villi height and villi area of ileum.
 189

190 Table 3. The average measurement of each parameter in broiler ileum supplemented by black
 191 cumin (*Nigella sativa*) through drinking water.

Treatment	Ileum					
	A1	B1	C1	D1	E1	F1
	-----Mean \pm SD (μm)-----					
P0	290.11 \pm 25.06 ^a	155.40 \pm 9.59 ^a	65.64 \pm 16.48 ^a	45.43 \pm 18.92 ^a	140.26 \pm 18.97 ^a	54.43 \pm 9.23 ^a
P1	289.00 \pm 88.88 ^a	191.56 \pm 37.02 ^a	106.00 \pm 59.13 ^a	45.41 \pm 26.65 ^a	178.11 \pm 52.14 ^a	50.56 \pm 3.02 ^a
P2	557.22 \pm 31.45 ^b	244.78 \pm 52.74 ^a	162.33 \pm 15.84 ^a	91.04 \pm 2.33 ^b	174.34 \pm 37.86 ^a	52.78 \pm 3.71 ^a
P3	327.05 \pm 87.56 ^a	144.68 \pm 52.87 ^a	98.26 \pm 43.50 ^a	39.33 \pm 18.74 ^a	128.93 \pm 22.06 ^a	47.60 \pm 12.7 ^a

192 Information : Drinking water without *Nigella sativa* (P0), Drinking water with *Nigella sativa*
 193 36 mg/kg BW/day (P1), 72 mg/kg BW/day (P2), 144 mg/kg BW/day (P3); A1 (villi height of jejunum), B1 (basal villi width of jejunum), C1 (apex villi width
 194 of jejunum), D1 (villi area of jejunum), E1 (crypt depth of jejunum), F1 (gland
 195 diameter of jejunum). Different supercripts with letters in the same column
 196 indicate significant differences (P<0.05).
 197
 198



199
 200 Figure 3. The supplementation of black cumin (*Nigella sativa*) 72 mg/kg BW (P2) in drinking
 201 water increase average sizes of villi height (A2) and villi area (D1) of broiler ileum.
 202 Asterisk (*) indicates the most significant effect (P<0.05).
 203

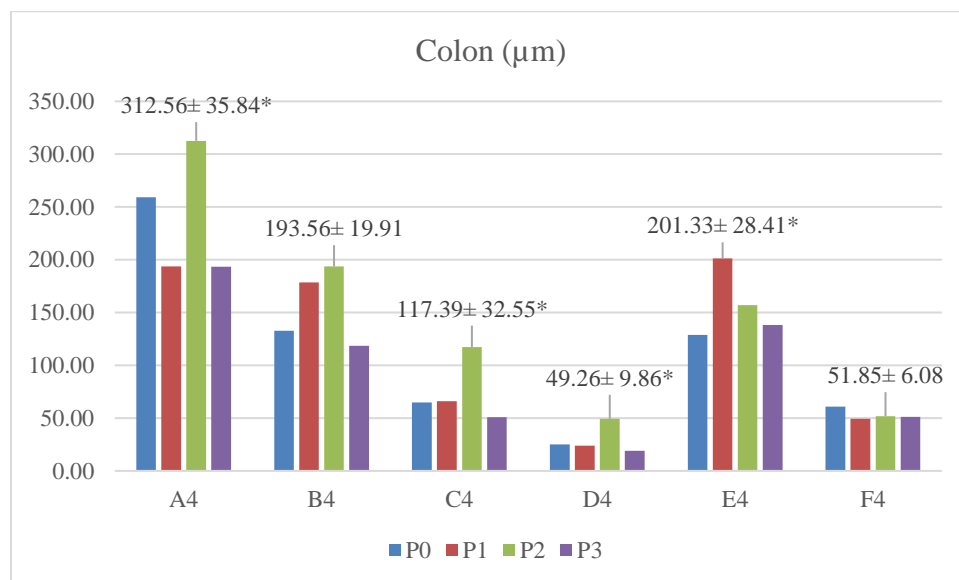
204 The average measurement of each parameter in broiler colon that supplemented by black
 205 cumin (*Nigella sativa*) through drinking water presented in Table 4. Supplementation of black
 206 cumin (*Nigella sativa*) with a dose of 72 mg/kg of broiler body weight (P2) gave the significant
 207 effect on the increase in the average size of villi height, apex villi width, villi area of colon and
 208 in parameter of crypt depth showed that dose of P1 (36 mg/kg BW) had a significant effect
 209 but was not different from the dose of P2.

210

211 Table 4. Average measurement of each parameter in broiler colon supplemented by black
 212 cumin (*Nigella sativa*) through drinking water.

Treatment	Colon					
	A1	B1	C1	D1	E1	F1
-----Mean \pm SD (μm)-----						
P0	259.22 \pm 33.07 ^{ab}	132.82 \pm 44.40 ^a	64.95 \pm 4.74 ^{ab}	25.15 \pm 3.85 ^a	128.81 \pm 28.46 ^a	60.80 \pm 11.19 ^a
P1	193.56 \pm 25.20 ^a	178.39 \pm 38.25 ^a	66.10 \pm 27.36 ^{ab}	23.76 \pm 5.23 ^a	201.33 \pm 28.41 ^b	49.29 \pm 7.77 ^a
P2	312.56 \pm 35.84 ^b	193.56 \pm 19.91 ^a	117.39 \pm 32.55 ^b	49.26 \pm 9.86 ^b	157.11 \pm 12.36 ^{ab}	51.85 \pm 6.08 ^a
P3	193.22 \pm 45.30 ^a	118.53 \pm 13.63 ^a	50.97 \pm 8.34 ^a	19.03 \pm 7.77 ^a	138.07 \pm 29.86 ^{ab}	51.14 \pm 7.58 ^a

213 Information : Drinking water without *Nigella sativa* (P0), Drinking water with *Nigella sativa*
 214 36 mg/kg BW/day (P1), 72 mg/kg BW/day (P2), 144 mg/kg BW/day (P3); A1 (villi height of colon), B1 (basal villi width of colon), C1 (apex villi width of
 215 colon), D1 (villi area of colon), E1 (crypt depth of colon), F1 (gland diameter of
 216 colon). Different superscripts with letters in the same column indicate significant
 217 differences (P<0.05).
 218
 219



220

221 Figure 4. The supplementation of black cumin (*Nigella sativa*) 72 mg/kg BW (P2) in drinking
 222 water increasing average sizes of villi height (A1), villi apex width (C1), villi area (D1), and
 223 crypt depth (E1) of broiler colon.

224 Asterisk (*) indicates the most significant effect (P<0.05).
 225

226 The ability of digestion and absorption of food substances could be affected by the
 227 surface area of the intestinal epithelium, the number of folds, and the number of villi and
 228 microvilli that expand the absorption field (Austic and Nesheim, 1990; Ibrahim 2008) and also
 229 influenced by the height and surface area of the villi organs. digestive tract (Sugito et al., 2007;

230 Ibrahim 2008). These villi function to expand the surface of the intestine which affects the
231 process of absorption of food (Alfiansyah, 2011). The development of the intestinal villi in
232 broiler chickens is related to the function of the intestine and growth of the chicken (Sun, 2004).
233 Villi are places for absorption of nutrients, the wider the villi, the more food substances that
234 will be absorbed, in the end it can have an impact on the growth of organs and increased carcass
235 (Asmawati, 2014).

236 Treatment with a dose of 72 mg/kg BW/day (P2) gave the most significant increase
237 ($P < 0.05$) in villi height and villi area of all broiler digestive organs (duodenum, jejunum,
238 ileum, colon) compared to other treatments. The increase in villi height in the broiler intestine
239 is closely related to an increase in digestive function and absorption function due to the
240 expansion of the absorption area and is an expression of the smooth transportation system of
241 nutrients throughout the body (Awad et al., 2008). One of the parameters that can be used to
242 measure the quality of growth is the morphological structure of the intestine. The height of villi
243 in all parts of the small intestine (duodenum, jejunum, ileum) and large intestine in general
244 increases (Ningtias, 2013). Increasing the villi width and the villi height can expand the
245 absorption area of the villi. According to Asmawati (2014), the wider the villi the more food
246 substances that will be absorbed in the end can have an impact on the growth of the body's
247 organs and according to Rahmawati (2016) the higher the size of the villi, the wider the area
248 of nutrient absorption by the small intestine wall so that it will trigger increased growth,
249 according to Guyton (1997), the more villous surface area indicates the more efficient
250 absorption of nutrients that occurs. The efficiency of nutrient absorption cannot be separated
251 from the work of hormonal, nervous and digestive glands in the digestive tract and its accessory
252 glands.

253 Food, environment, and metabolic activity affects the number of intestinal glands.
254 Chickens generally eat food consisting of granules and are hard, so that a more active secretion

255 of intestinal glands is needed, to support the development of epithelial cells that make up the
256 villi (Mardhiah, 1991). Crypts contained in the intestinal villi, which are composed of inline
257 cylindrical epithelial cells. These glands produce mucus and several enzymes for the
258 metabolism of peptides, fats, carbohydrates, and intestinal juices (mucin) which function to
259 protect the intestinal mucosa (Aughey and Frye, 2001). The increase in the average size of the
260 gland diameter in the treatment with supplementation of black cumin in drinking water showed
261 an increase in the size of the gland diameter in duodenum. P2 treatment with the significant
262 increase ($P<0.05$) in size compared to other treatments, it can support the development of
263 epithelial cells that make up the villi, which will increase the absorption of nutrients in the
264 digestive tract.

265 Supplementation of black cumin (*Nigella sativa*) 36 mg/kg BW/day (P1) gave the most
266 significant increase ($P<0.05$) in parameter of crypt depth of colon but was not different from
267 the dose of P2 (Table 4). Supplementation of black cumin (*Nigella sativa*) had no significant
268 effect for other organs (duodenum, jejunum, and ileum) that presented in other tables.
269 According to Sun et al. (2005) and Smirnov et al. (2005) that into crypt has no effect after
270 broilers are more than 28 days old. Broilers in this study collected samples of digestive tract
271 organs at the age of 31 days. It is assumed that the development of intestinal morphology is
272 closely related to the role of micronutrients in line with the increasing age of broilers
273 (Harimurti and Rahayu, 2009).

274 Based on the data presented in Figure 1, Figure 2, Figure 3, Figure 4, supplementation
275 of Black Cumin (*Nigella sativa*) 72 mg/kg BW/day had most significant effect to the size of
276 the villi height and villi area of all digestive tract organs compared to controls. One of the
277 parameters that can be used to measure the quality of growth is the morphological structure of
278 the intestine (Wang *et al.*, 2008; Ningtias 2013). According to Suprijatna *et al.*, (2008) the
279 small intestine is the main organ for digestion and absorption of digestive products. Various

280 enzymes that enter this channel function to accelerate and streamline the breakdown of
281 carbohydrates, proteins and fats to facilitate the absorption process. In adult chickens, the
282 length of the small intestine is about 62 inches or 1.5 meters.

283 The digestive tract organs are supported by villi which are a special shape in the mucosa.
284 The villi are finger-shaped protrusions of the mucosa and are characteristic of the small
285 intestine. Increasing the number of villi will increase food absorption. Villi function to expand
286 the surface of the intestine which affects the process of absorption of food (Alfiansyah, 2011).
287 The development of the intestinal villi in broiler chickens is related to the function of the
288 intestine and growth of the chicken (Sun, 2004). The increase in villi causes more villi surface
289 area to absorb nutrients into the bloodstream (Mile *et al.* 2006; Rostinawati 2008).

290 High villi indicate that the intestines are better off than short villi. Awad *et al.* (2008)
291 stated that the increase in the height of the villi in the intestine with digestive and absorption
292 functions occurs because of the intact villi form which is a smooth expression of the nutrient
293 transport system throughout the body. Rofiq (2003) states that the absorption of nutrients in
294 the intestine is influenced by the inner surface area of the intestine (folds, villi and microvilli)
295 and the length of transit of the digesta in the intestine.

296 The surface area of the intestine such as the height of the villi describes the area for
297 absorption of nutrients. Villi are small finger or leaf-like protrusions found on the mucous
298 membrane, 0.5 to 1.5 mm long and found only in the small intestine. The villi in the ileum are
299 finger-like in shape and shorter than the villi found on the duodenum and jejunum. One of the
300 parameters used to measure the quality of growth is the intestinal morphological structure
301 (Wang and Peng, 2008).

302 The intestinal gland (Lieberkuhn's gland) has a small hole that becomes the mouth of the
303 glandular simplex tubule. The intestinal glands are scattered between the villi, attached to the
304 mucous membrane. The intestinal glands and intestinal villi are covered by an epithelium,

305 consisting of goblet cells and enterocytes, among others. Goblet cells secrete mucus to
306 lubricate and protect the surface of the intestine, while the enterocytes in crypt secrete large
307 amounts of water and electrolytes. Goblet cells secrete a kind of mucus, namely mucin which
308 functions to coat the intestinal tract and protect pathogens that can damage intestinal epithelial
309 cells so that the number of goblet cells is important for the health of broilers (Forder et al.,
310 2007). The presence of goblet cells in the broiler duodenum was available in sufficient
311 numbers in the body of broilers since before hatching, but the number of goblet cells in the
312 jejunum and ileum was only reached after the broilers have hatched (Reynold et al., 2020).

313

314

CONCLUSION

315

316 The conclusions of this study were the supplementation of Black Cumin (*Nigella sativa*)
317 72 mg/kg BW/day through drinking water could increase significantly ($P < 0.05$) to 1) the
318 average sizes of villi height and villi area of small intestine (duodenum, jejunum, ileum) and
319 large intestine (colon) of broiler; 2) the average sizes of basal villi width, villi apex width, and
320 gland diameter of broiler duodenum; and 3) the average size of apex villi width of broiler
321 colon.

322

323

ACKNOWLEDGMENTS

324

325 The author would like to thank University of Lampung for providing a source of research
326 funding from the University of Lampung Grants in 2020.

327

328

REFERENCES

329

- 330 Alfiansyah, M. 2011. Anatomi dan Pencernaan Usus Halus. [http://www. sentra-edukasi.com/](http://www.sentra-edukasi.com/).
- 331 Diakses tanggal 20 Agustus 2020
- 332 Aughey, E. and F.L. Frye. 2001. **Comparative Veterinary Histology with Clinical**
- 333 **Correlates**. CRC Press, Florida.
- 334 Asmawati. 2014. The Effect of In Ovo Feeding on Hatching Weight and Small Intestinal Tissue
- 335 Development of Native Chicken. **Dissertation**. Hasanuddin University, Makassar.
- 336 Austic, R.E. and M.C. Nesheim. 1990. **Poultry Production**. Lea & Febiger. Philadelphia
- 337 Awad, W.A., K. Ghareeb, S. Nitclu, S. Pasteiner, S.A. Raheem, and J. Bohm. 2008. Efect of
- 338 dietary inclusion of probiotic, prebiotic and symbiotic on intestinal glucose absorb'tion
- 339 of broiler chickens. **J. Poult. Sci.** 7: 688-691.
- 340 Badan Pusat Statistik. 2019. Populasi Ayam Ras Pedaging menurut Provinsi (Ekor) Tahun
- 341 2009-2018. [https://www.bps.go.id/dynamictable/2015/12/18/1034/populasi-ayam-ras-](https://www.bps.go.id/dynamictable/2015/12/18/1034/populasi-ayam-ras-pedaging-menurut-provinsi-2009-2018.html)
- 342 [pedaging-menurut-provinsi-2009-2018.html](https://www.bps.go.id/dynamictable/2015/12/18/1034/populasi-ayam-ras-pedaging-menurut-provinsi-2009-2018.html) Diakses pada tanggal 13 Agustus 2020.
- 343 Forder, R.E.A., G.S. Howarth, D.R. Tivey, R.J. Hughes. 2007. Bacterial modulation of small
- 344 intestinal goblet cells and mucin composition during early posthatch development of
- 345 poultry. **Poult. Sci.** 86(11): 2396-2403.
- 346 Guyton, A.C. 1997. **Fisiologi Kedokteran Edisi ke-9**. Universitas Indonesia Press, Jakarta.
- 347 Harimurti,, S. dan E.S. Rahayu. 2009. Morfologi Usus Ayam Broiler yang disuplementasi
- 348 dengan Probiotik Strain Tunggal dan Campuran. **Agritech.** 29(3): 179-183.
- 349 Hendrik. 2009. **Habbatus Sauda**. Pustaka Iltazam, Solo.
- 350 Ibrahim, S. 2008. Hubungan ukuran-ukuran usus halus dengan berat badan broiler. **Agripet.**
- 351 8(2): 42-46.
- 352 Iji, P. A., R. J. Hughes, M. Choct and D. R. Tivey. 2001. Intestinal structure and function of
- 353 broiler chickens on wheat-based diets supplemented with microbial enzyme. *Asian-Aust.*
- 354 **J. Anim. Sci.** 14(1):54-60.

- 355 Kabir, S.M.L. 2010. Avian colibacillosis and salmonellosis: a closer look at epidemiology,
356 pathogenesis, diagnosis, control and public health concerns. **Int. J. Environ. Res. Public**
357 **Health.** 7:89-114.
- 358 Marshall, R.M. and S.B. Levy. 2011. Food Animals and Antimicrobials: Impacts on Human
359 **Health. Clin. Microbiol. Rev.** 24(4): 718-733.
- 360 Mile, R.D., G.D. Butcher, P.R. Henry, R.C. Littlel. 2006. Effect of antibiotic growth promoters
361 on broiler performance, intestinal growth parameters, and quantitative morphology. **J.**
362 **Poultry Sci.** 85:476-485.
- 363 Ningtias, A.S. 2013. Comparison of Growth Performance of Broilers, Kampong, and
364 Backcross3 (*Gallus gallus domesticus* Linnaeus, 1758) Based on Morphometri and
365 Histological Structure of Ileum and Breast Muscle. **Thesis.** Fakultas Biologi Universitas
366 Gajah Mada. Yogyakarta
- 367 Rahmawati. 2016. Histologis saluran pencernaan ayam buras hasil in ovo feeding asam amino
368 l-arginine. **Thesis.** Fakultas Peternakan Universitas Hasanuddin. Makassar.
- 369 Reynold, K.L., S.E. Cloft., E.A. Wong. 2020. Changes with age in density of goblet cells in
370 the small intestine of broiler chicks. **Poult. Sci.** 99(5): 2342-2348.
- 371 Rostika N. 2012. Pengaruh pemberian ekstrak minyak jintan hitam (*Nigella sativa*) terhadap
372 gambaran histologi organ lambung dan usus halus mencit (*Mus musculus*). **Thesis.**
373 **Fakultas Kedokteran Hewan Institut Pertanian Bogor, Bogor.**
- 374 Rostinawati T. 2008. Aktivitas antibakteri ekstrak etanol dan ekstrak air kelopak bunga rosela
375 (*Hibiscus sabdariffa* L.) terhadap Mycobacterium tuberculosis galur Labkes-026 (Multi
376 Drug Resisten) dan Mycobacterium tuberculosis galur H37Rv secara in vitro. **Thesis.**
377 **Faculty of Pharmacy, Padjadjaran University, Bandung.**

- 378 Siagian, Y.A. 2016. Gambaran Histologis dan Tinggi Vili Usus Halus Bagian Ileum Ayam Ras
379 Pedaging yang diberi Tepung Daun Kelor (*Moringa oleifera*) dalam ransum. **Thesis**.
380 Fakultas of Animal Science Hasanuddin University, Makasar.
- 381 Smirnov, A., R. Perez, E. Amit-Romach, D. Sklan, and Z. Uni. 2005. Mucin dynamics and
382 microbial populations in chicken small intestine changed by dietary probiotic and
383 antibiotic growth promotor supplementation. **J. of Nutrition**. 135: 187-192.
- 384 Sturkie, P.D., G.C. Whittow. 2000. **Sturkie's Avian Physiology**. Academic Press, Waltham.
- 385 Sugito, M.W., D.A. Astuti, E. Handharyani, Chairul. 2007. Histopatologi hati dan ginjal pada
386 ayam broiler yang dipapar cekaman panas dan diberi ekstrak kulitbatang Jaloh (*Salix*
387 *tetrasperma* Roxb). **JITV**. 12:6873.
- 388 Sun, X. 2004. Broiler performance and intestinal alterations when fed drug-free diets. **Thesis**.
389 Animal and Poultry Science. Blacksburg, Virginia
- 390 Sun, X., A. McElroy, Jr. Webb, A.E.K.E. Sefton and C. Novak. 2005. Broiler performance and
391 intestinal alterations when fed drug-free diets. **J. of Poult. Sci.**, 84: 1294-1302.
- 392 Suprijatna, E., U. Atmomarsono, R. Kartasudjana. 2008. **Ilmu Dasar Ternak Unggas**. Penebar
393 Swadaya, Jakarta.
- 394 Thippeswammy N.B., K.A. Naidu. 2005. Antioxidant potency of cumin varieties cumin, black
395 cumin and bitter cumin on antioxidant systems. **Euro Food Res Tech**. 220: 472-476
- 396 Topozoda H.H., H.A. Mazloun, M. El-Dakhakhny. 1965. The antibacterial properties of
397 *Nigella sativa* seeds. **J. Egypt Med**. 48:187-202.
- 398 Yusuf, M.S. 2014. Efektivitas Penggunaan Jintan Hitam (*Nigella sativa*) dalam Proses
399 Percepatan Penyembuhan Luka Setelah Pencabutan Gigi. **Thesis**. Faculty of Dentistry
400 Hasanuddin Univerity, Makassar.

- 401 Wang, J.X. and K.M. Peng. 2008. Molecular, Cellular, and Developmental Biology
402 Developmental Morphology of the Small Intestine of African Ostrich Chicks. **J. Poultry**
403 **Sci.** 87: 2629-2635.



Akreditasi No. 200/M/KPTS/2020

JURNAL KEDOKTERAN HEWAN

FAKULTAS KEDOKTERAN HEWAN UNIVERSITAS SYIAH KUALA

Jl. Tgk. Hasan Krueng Kalee No. 4, Kampus Unsyiah

Darussalam Banda Aceh, Aceh

Telp./Fax. 0651-7410247 E-mail : jkh@unsyiah.ac.id; jurnal_khusk@yahoo.com

home page: <http://jurnal.unsyiah.ac.id/JKH>

SURAT KETERANGAN

Nomor : 041/UN11.1.22/JKH/IV/2021

Redaksi Jurnal Kedokteran Hewan menerangkan tentang status terbitan artikel untuk :

Nama Penulis : M. Hartono, PE. Santosa, R. Ermawati, MMP. Sirat

Judul : Effect of black cumin (*nigella sativa*) supplementation through drinking water on the histology of small intestine and large intestine of broiler

Dengan ini dinyatakan telah **accepted** untuk diterbitkan pada Jurnal Kedokteran Hewan Vol. 15 No. 2 Juni 2021.

Demikian surat keterangan ini kami perbuat untuk dapat dipergunakan seperlunya.

Banda Aceh, 27 April 2021

Ketua Dewan Penyunting



Prof. Dr. drh. Tongku N Siregar, MP.
NIP. 19690901 199403 1 003



JURNAL KEDOKTERAN HEWAN

FAKULTAS KEDOKTERAN HEWAN UNIVERSITAS SYIAH KUALA

Jl. Tgk. Hasan Krueng Kalee No. 4, Kampus Unsyiah
Darussalam Banda Aceh, Aceh

Telp./Fax. 0651-7410247 E-mail : jkh@unsyiah.ac.id; jurnal_khusk@yahoo.com
home page: <http://jurnal.unsyiah.ac.id/JKH>

Akreditasi No. 200/M/KPTS/2020

PEMBERITAHUAN BIAYA PENERBITAN

Nomor. 054/UN11.1.22/BP/VI/2021

Untuk Bpk/Ibu/Sdr. Madi Hartono

Judul:

EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING
WATER ON THE HISTOLOGY OF SMALL INTESTINE AND
LARGE INTESTINE OF BROILER

No.	Deskripsi	Satuan (Rp)	Jumlah	Total (Rp)
1	Biaya penerbitan naskah	1.000.000	1	1.000.000
2	Biaya cetak	-	-	-
3	Biaya translasi	-	-	-
4	Biaya editing Bahasa Inggris	425.000	1	425.000
5	Biaya yang harus dibayarkan	-	-	1.425.000

Harap dikirimkan biaya tersebut, melalui Bank Syariah Mandiri (BSM)/Bank Syariah Indonesia (BSI)
(Kode Bank 451):

Atas nama : **BUDIANTO PANJAITAN**
Nomor rekening : **7 1 5 5 2 4 4 8 2 2**

Bukti transfer harap dikirimkan ke:

Alamat email : jkh@unsyiah.ac.id
Konfirmasi ke nomor : **0877 4476 0115 (Budianto Panjaitan)**

Terima kasih atas partisipasi dan kerjasamanya

=====



Rendy Sirat <m.mirandy@fp.unila.ac.id>

Revisi Naskah Siap Terbit dan Bukti Transfer Biaya Penerbitan

1 message

Rendy Sirat <m.mirandy@fp.unila.ac.id>
To: "Drh. T Armansyah TR, M.Kes." <jurnal@unsyiah.ac.id>

Wed, Jun 23, 2021 at 4:54 PM

Kepada Yth. Editor Jurnal Kedokteran Hewan
Fakultas Kedokteran Hewan Universitas Syiah Kuala

Assalamu'alaikum wrwb.

Kami mengucapkan terimakasih atas kesempatan yang telah diberikan kepada kami untuk dapat menerbitkan artikel pada JKH FKH Unsyiah yang berjudul :

THE EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER CHICKENS

Berikut kami lampirkan :

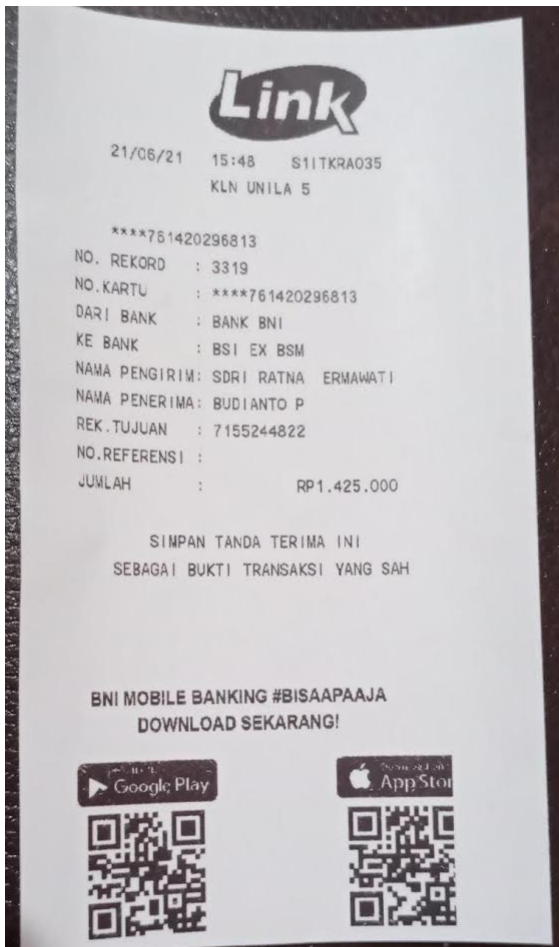
1. Revisi Naskah Siap Terbit (file word 4_JKH_EDITED JKH 1 (revisi))
2. Bukti Transfer Biaya Penerbitan (sesuai surat pemberitahuan biaya penerbitan)

Atas perhatian dan kerjasama Bapak/Ibu, kami ucapkan terimakasih.

Wassalamu'alaikum wrwb.


--

drh. Muhammad Mirandy Pratama Sirat, M.Sc.
Department of Animal Husbandry
Faculty of Agriculture University of Lampung
082226238837



2 attachments

 **4_JKH_EDITED JKH 1 (revisi).docx**
94K

 **054_2021_Madi Hartono et_al (Biaya Penerbitan).pdf**
81K



21/06/21 15:48 S11TKRA035
KLN UNILA 5

****761420296813

NO. REKORD : 3319
NO. KARTU : ****761420296813
DARI BANK : BANK BNI
KE BANK : BSI EX BSM
NAMA PENGIRIM: SDRI RATNA ERMAWATI
NAMA PENERIMA: BUDIANTO P
REK. TUJUAN : 7155244822
NO. REFERENSI :
JUMLAH : RP1.425.000

SIMPAN TANDA TERIMA INI
SEBAGAI BUKTI TRANSAKSI YANG SAH

**BNI MOBILE BANKING #BISAAPAAJA
DOWNLOAD SEKARANG!**



THE EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER CHICKENS

Madi Hartono¹, Purnama Edy Santosa², Ratna Ermawati²,
Muhammad Mirandy Pratama Sirat^{1*}

¹ Study Program of Animal Nutrition and Feed Technology, Department of Animal Husbandry, Faculty of Agriculture, University of Lampung, Bandar Lampung, Indonesia

² Study Program of Animal Husbandry, Department of Animal Husbandry, Faculty of Agriculture, University of Lampung, Bandar Lampung, Indonesia

*Corresponding author: m.mirandy@fp.unila.ac.id

ABSTRACT

This study aimed to determine the effect of Black Cumin (*Nigella sativa*) supplementation through drinking water on the histology of broiler chickens' small intestine and large intestine. The research was conducted from April–September 2020 in a cage facility of the Integrated Field Laboratory, Faculty of Agriculture, University of Lampung. This research used a completely randomized design with four treatment groups and three replications (five broilers per replication) with a total of 60 male broilers. The treatments were drinking water without Black Cumin (P0, control); drinking water with Black Cumin 36 mg/kg bw/day (P1); 72 mg/kg bw/day (P2); and 144 mg/kg bw/day (P3). Three broilers from each group were randomly necropsied at 31 days old, and samples of the small intestine (duodenum, jejunum, ileum) and large intestine were fixed with 10% formalin solution and sent to the Lampung Disease Investigation Center for histological preparations. The observation of preparations was carried out microscopically using the Leica DM500[®] Binocular Microscope to accurately calculate various parameter sizes. The results were analyzed statistically with one way Analysis of Variance at significant level 5% and if proven significant, then a Tukey test was conducted. The results of this study were that the supplementation of Black Cumin (*Nigella sativa*) 72 mg/kg bw/day through drinking water could significantly increase (P<0.05) 1) the average sizes of villi height and villi area of small intestine (duodenum, jejunum, ileum) and large intestine (colon) of broiler chickens; 2) the average sizes of basal villi width, villi apex width, and gland diameter of broiler duodenum; and 3) the average size of apex villi width of broiler colon. The conclusion of this study was the supplementation of black cumin (*Nigella sativa*) at dose of 72 mg/kg bw/day through drinking water could increase the histological sizes of the small intestine and large intestine of broilers.

Key words: broiler, histology, large intestine, *Nigella sativa*, poultry, small intestine

ABSTRAK

Penelitian ini bertujuan mengetahui pengaruh pengaruh suplementasi jintan hitam (*Nigella sativa*) melalui air minum terhadap histologi usus halus dan usus besar broiler. Penelitian dilakukan pada April–September 2020 di unit kandang Laboratorium Lapang Terpadu, Fakultas Pertanian, Universitas Lampung. Penelitian bersifat eksperimental menggunakan Rancangan Acak Lengkap dengan empat kelompok perlakuan dan tiga ulangan (lima ekor tiap ulangan) sehingga total 60 ekor broiler jantan. Perlakuan yang diberikan yaitu pemberian air minum tanpa jintan hitam (P0, kontrol); air minum dengan jintan hitam 36 mg/kg bobot badan/hari (P1); 72 mg/kg bobot badan/hari (P2); dan 144 mg/kg bobot badan/hari (P3). Tiga ekor dari tiap kelompok secara acak dinekrops pada hari ke-31 dan diambil sampel organ usus halus (duodenum, jejunum, ileum) dan usus besar (colon) kemudian difiksasi dengan larutan formalin 10% dan dikirim ke Balai Veteriner Lampung untuk pembuatan preparat histologi. Pengamatan preparat dilakukan secara mikroskopis menggunakan Mikroskop Binokuler Leica DM500[®] untuk menghitung berbagai ukuran parameter secara akurat. Analisis hasil dilakukan secara statistik menggunakan analisis sidik ragam satu arah dengan taraf signifikansi 5% dan dilanjutkan dengan uji lanjut Tukey. Hasil penelitian ini yaitu suplementasi jintan hitam 72 mg/kg bobot badan/hari dapat meningkatkan secara signifikan (P<0,05) terhadap 1) ukuran rata-rata tinggi vili dan luas vili usus halus (duodenum, jejunum, ileum) dan usus besar broiler; 2) ukuran rata-rata tinggi vili, lebar vili basal, lebar puncak vili, luas vili, dan diameter kelenjar duodenum broiler; dan 3) ukuran rata-rata lebar puncak vili usus besar broiler. Kesimpulan penelitian ini bahwa suplementasi jintan hitam (*Nigella sativa*) dosis 72 mg/kg bb/hari melalui air minum dapat meningkatkan ukuran histologi usus halus dan usus besar broiler.

Kata kunci: broiler, histologi, usus besar, *Nigella sativa*, unggas, usus halus

Formatted: Space After: 0 pt, Line spacing: single

Formatted: Centered, Space After: 0 pt, Line spacing: single

Formatted: Font: 12 pt

Formatted: Not Superscript/ Subscript

Formatted: Centered, Indent: Left: 0 cm, Hanging: 0.25 cm

Formatted: Indent: Left: 0.25 cm, Hanging: 0.25 cm, Line spacing: single

Formatted: Font: 12 pt

Deleted: Madi Hartono, Purnama Edy Santosa, Ratna Ermawati, Muhammad Mirandy Pratama Sirat (mohon dilengkapi beserta afiliasinya)¶

Formatted: Font color: Green

Deleted: (mohon dituliskan hasil penelitian disini).

Deleted: conclusions

Formatted: Font: 8 pt, Font color: Green

Formatted: Font: 8 pt, Font color: Green

Deleted: Kesimpulan

INTRODUCTION

The development of poultry farming in Indonesia is increasing. Broilers are one of the fastest growing poultry. Many Indonesian breeders prefer to farm broilers because of their relatively short growth period and low maintenance costs compared to ruminants such as cattle, sheep, or goats. In 2018, the broiler population in Indonesia reached 1.89 billion, increasing 2.26% from the 2017 broiler population of 1.85 billion heads (BPS, 2019).

Intensive maintenance of broiler can often induce stress for the animal, resulting in a decreased responsiveness in the immune system, which increases the likelihood for disease and illness. Disease outbreaks are a serious obstacle in the broiler farming industry. The high incidence of disease can decrease productivity and even kill livestock which results in significant losses for breeders. Diseases can lead to decrease in body organ function, particularly the digestive organ. While antibiotics might prevent these diseases, the administration of antibiotics for long periods might cause residual buildup and antibiotic resistance in bacteria, which could have negative effects if consumed by humans (Marshall and Levy, 2011). One strategy to maintain digestive tract quality and function, and to reduce the residual antibiotics in broilers is to provide them with natural herbs such as black cumin (*Nigella sativa*), as immunomodulator (Sulistiawati and Radji, 2014) and antibiotic growth promoter substitution (Miraghaee et al., 2011) with a high content of antioxidants thymoquinone as anti-oxidant that could eliminate free radicals (Kruk et al., 2000), anti-inflammatory effect of several inflammation (Salem, 2005). *Nigella sativa* also has the ability to protect potentially different tissues and organs including liver, kidney, heart, blood, brain, lungs, reproductive system and gastrointestinal against chemical poison (Tavakkoli et al. 2017) and administration of *Nigella sativa* also can minimize heat stress in broilers by reducing cortisol and levels minimize histopathological changes in the liver (Hasan et al., 2019).

This study was conducted to determine the effect of black cumin supplementation through histological studies of the digestive organs of broiler chickens which are expected to potentially increase the villi height, apex villi width, basal villi width, villi area, crypt depth, and gland diameter of small intestine (duodenum, jejunum, ileum) and large intestine (colon).

MATERIALS AND METHODS

This research was carried out for six months (April–September 2020) in the Integrated Field Laboratory enclosure unit of the Faculty of Agriculture, University of Lampung. This research was designed using a completely randomized design (CRD) with four treatment groups and three replications, with a total of 60 male broilers were used (five broilers per replication). The treatment dose was according to the broiler body weight, namely 1) drinking water without black cumin (P0, control); drinking water with black cumin at dose of 36 mg/kg bw/day (P1); drinking water with black cumin at dose of 72 mg/kg bw/day (P2); and drinking water with black cumin at dose of 144 mg/kg bw/day (P3). On the 31st day, three broilers from each group were randomly sacrificed and samples of the small intestine (duodenum, jejunum, ileum) and large intestine (colon) were then fixed with 10% formalin solution and sent to the Lampung Veterinary Disease Investigation Center for histological preparations with Hematoxylin Eosin (HE) staining. The preparation observation was carried out microscopically using the Leica DM500® Binocular Microscope Technology to accurately calculate various parameter sizes.

The research parameters were villi height, villi apex width, basal villi width, villi area, crypt depth, and gland diameter. The observation of histological preparations was done with a 10x magnification objective lens. The calculation of each parameter was carried out as many as three villi (villi height, villi apex width, basal villi width, villi area, crypt depth) and three gland diameters in each digestive tract organ (duodenum, jejunum, ileum, and colon). There were three replications per organ, so that the total for each organ was obtained through an average of the nine villi and nine gland diameters.

The calculation of the surface area of the intestinal villi using the method of Iji et al. (2001) which was slightly modified with the assumption that the villi model is an analogue of the trapezium shape so that the average number of the apical widths of the villi plus the average basal width of the villi is divided by two then multiplied by the height of the villi by the following formula.

Deleted: heads,

Formatted: Font color: Green

Deleted: bodily

Deleted: can

Deleted: that is believed to improve digestion function based on research that was carried out in mice in 2012 (Rostika, 2012)....

Formatted: Font: Italic

Formatted: Font: Italic

Formatted: Font color: Red

Deleted: ¶

Deleted: Materials¶

This study used broiler cages, a sprayer for cage disinfection, bamboo to make 12 cage plots, plastic tarpaulin for curtains, newspaper and used husks as litter, 12 15-watt bulbs used as a heating source for the brooding area, 12 hanging feeders, 12 chick feeder trays, 12 chicken drinking apparatuses; 1 bucket, 1 hand spray, 1 water tray for dipping, 1 electric scale, thermohydrometer for measuring temperature and humidity, sack, and plastic. Organ sampling equipment was also used, namely necropsy equipment, object glass, cover glass, a refrigerator, a microtome, and a Leica DM500® Binocular Microscope Technology connected with a computer for taking tissue images and measuring the parameters of each organ. ¶ Materials used in the study were 60 Day Old Chicks (DOC) male broiler Cobb CP 707 strains kept for 30 days, food rations, drinking water, Black Cumin extracts (*Nigella sativa*), vaccines for Newcastle Disease (ND), Avian Influenza (AI) and Infectious Bursal Disease (IBD), and a 10% formalin solution. (Delete)¶

¶ Methods (Delete)¶

Deleted: (five heads per replication)

Deleted: so

Deleted:

Deleted: necropsied and

Deleted: to undergo

$$\text{Surface area} = \frac{b+c}{2} \times a$$

a = height of intestine villi
 b = apex width of intestine villi
 c = basal width of basal of intestine villi

Data Analysis

The average measurement data for various parameters of each digestive tract organ (duodenum, jejunum, ileum, and colon) were analyzed using analysis of variance, and followed by Tukey test.

RESULTS AND DISCUSSION

The average measurements of villi height, villi apex width, basal villi width, villi area, crypt depth, and gland diameter of the digestive tract organs (duodenum, jejunum, ileum, colon) are presented in each table. Each shows the calculation of each parameter from the nine villi (villi height, basal villi width, apex villi width, villi area, crypt depth) and nine gland diameters on histopathological preparations in each digestive tract organ (duodenum, jejunum, ileum, large intestine) in each treatment.

The average measurement of each parameter in broiler duodenum, jejunum, ileum, and colon were presented in Table 1, Table 2, Table 3, Table 4 respectively. Supplementation of black cumin with a dose of 72 mg/kg of broiler body weight (P2) had a significant effect on the increase in the average villi height, basal villi width, villi apex width, villi area, and gland diameter of the duodenum; the average villi height and villi area of jejunum; the average villi height and villi area of the ileum; and the average size of villi height, apex villi width, villi area of colon.

The ability of digestion and absorption of food substances could be affected by the surface area of the intestinal epithelium, the number of folds, and the number of villi and microvilli that expand the absorption field (Austic and Nesheim, 1990; Ibrahim, 2008). It could also be influenced by the height and surface area of the villi organs or digestive tract (Sugito *et al.*, 2007; Ibrahim, 2008). The development of the intestinal villi in broiler chickens is related to the function of the intestine and growth of the chicken (Sun *et al.*, 2005). Villi are places for absorption of nutrients, the wider the villi, the more food substances that will be absorbed, in the end it can have an impact on the growth of organs and increased carcass (Asmawati, 2014).

Treatment with a dose of 72 mg/kg bw/day (P2) showed the most significant increase ($P < 0.05$) in villi height and villi area of all broilers digestive organs (duodenum, jejunum, ileum, colon) compared to other treatments. The increase in villi height in the broiler intestine is closely related to an increase in digestive function and absorption function due to the expansion of the absorption area and is an expression of the smooth transportation system of nutrients throughout the body (Awad *et al.*, 2008). One of the parameters that can be used to measure the quality of growth is the morphological structure of the intestine. The height of villi in all parts of the small intestine (duodenum, jejunum, ileum) and large intestine in general increases (Ningtias, 2013). Increasing the villi width and the villi height can expand the absorption area of the villi. According to Asmawati (2014), the wider the villi, the more food substances will be absorbed, which can have a long-term impact on the growth of the body's organs. Similarly, according to Rahmawati (2016) the higher the size of the villi, the wider the area of nutrient absorption by the small intestine wall, thus will trigger the growth improvement. Guyton (1997) added that a more villous surface area leads to a more efficient nutrient absorption. The efficiency of nutrient absorption cannot be separated from the work of hormonal, nervous, and digestive glands in the digestive tract and its accessory glands.

Crypts are contained in the intestinal villi, which are composed of inline cylindrical epithelial cells. These glands produce mucus and several enzymes for the metabolism of peptides, fats, carbohydrates, and intestinal juices (mucin) which function to protect the intestinal mucosa (Aughey and Frye, 2001). The increase in the average size of the gland diameter in the treatment with supplementation of black cumin in drinking water showed an increase in the size of the duodenum's gland diameter. The P2 treatment showed a significant increase ($P < 0.05$) in size compared to other treatments, it can support the development of epithelial cells that make up the villi, which will increase the absorption of nutrients in the digestive tract.

Supplementation of black cumin 36 mg/kg bw/day (P1) showed the most significant increase ($P < 0.05$) in parameter of crypt depth of colon but was not different from the dose of P2 and P3 (Table 4). According to Sun

Deleted: M

Deleted: research

Deleted: shows the average of all replications of

Deleted: which

Deleted: t

Deleted: hen

Deleted: through the

Deleted: A

Deleted: V

Deleted: then a

Deleted: was conducted

Deleted: Different superscripts with letters in the same column indicate significant differences ($P < 0.05$).

Formatted: Font color: Green

Formatted: Indent: First line: 0 cm

Deleted: supplemented by black cumin through drinking water is

Deleted: (Figure 1)

Deleted: . ¶

The average measurement of each parameter in broiler jejunum supplemented by black cumin through drinking water is presented in Table 2. Supplementation of black cumin with a dose of 72 mg/kg of the broiler's body weight (P2) had a significant effect on the increase of the

Deleted: . ¶

The average measurement of each parameter in broiler ileum supplemented by black cumin through drinking water is presented in Table 3. Supplementation of black cumin with a dose of 72 mg/kg of broiler body weight (P2) had a significant effect on the increase in ...

Deleted: . ¶

The average measurement of each parameter in broiler colon supplemented by black cumin through drinking water is presented in Table 4. Supplementation of black cumin with a dose of 72 mg/kg of broiler body weight (P2) had a significant effect on the increase in ...

Deleted: and in parameter of crypt depth but showed that dose P1 (36 mg/kg bw) had a significant effect but was not different from the dose of P2. ¶

Deleted: These villi function to expand the surface of the intestine which affects the process of food absorption (Alfiansyah, 2011). ...

Deleted: 4

Deleted: '

Deleted: so that it will

Deleted: increased

Deleted: s

Deleted: Food, environment, and metabolic activity affect the number of intestinal glands. Chickens generally eat food consisting of hard granules, so that a more active secretion of intestinal glands is needed to support the development of epithelial cells that make up the villi (Mardiah, 1991). ...

Deleted:). Supplementation of black cumin had no significant effect for other organs (duodenum, jejunum, and ileum) that presented in other tables. ...

et al. (2005) and Smirnov *et al.* (2005) that crypt depth has no effect after broilers are more than 28 days old. In this study, samples of broilers digestive tract organs were collected at the age of 31 days. It is assumed that the development of intestinal morphology is closely related to the role of micronutrients in line with the increasing age of broilers (Harimurti and Rahayu, 2009).

Based on the data presented in Table 1, Table 2, Table 3, and Table 4, the supplementation of black cumin at dose of 72 mg/kg bw/day had most significant effect to the size of the villi height and villi area of all digestive tract organs compared to the control group. One of the parameters that can be used to measure the quality of growth is the morphological structure of the intestine (Wang *et al.*, 2008; Ningtias, 2013). According to Suprijatna *et al.* (2008), the small intestine is the main organ for digestion and absorption of digestive products. Various enzymes that enter this channel function to accelerate and streamline the breakdown of carbohydrates, proteins and fats to facilitate the absorption process. In adult chickens, the length of the small intestine is about 62 inches or 1.5 meters.

The digestive tract organs are supported by villi which are a special shape in the mucosa. The villi are finger-shaped protrusions of the mucosa and are characteristic of the small intestine. Increasing the number of villi will increase food absorption. Villi function to expand the surface of the intestine which affects the process of absorption of food (Alfiansyah, 2011). The development of the intestinal villi in broiler chickens is related to the function of the intestine and growth of the chicken (Sun *et al.*, 2005). The increase in villi causes more villi surface area to absorb nutrients into the bloodstream (Mile *et al.*, 2006).

High villi indicate that the intestines are better off than short villi. Awad *et al.* (2008) stated that the increase in the height of the villi in the intestine with digestive and absorption functions occurs because of the intact villi form which is a smooth expression of the nutrient transport system throughout the body. Rofiq (2003) stated that the absorption of nutrients in the intestine is influenced by the inner surface area of the intestine (folds, villi and microvilli) and the length of transit of the digesta in the intestine. Based on the research of Khedr and Abdel-Fattah (2007) that administration of *Nigella sativa* can increase broiler body weight, it is possible because *Nigella sativa* is rich in essential fatty acids such as oleic, linoleic, and linolenic acids which are essential to help growth and the presence of the active substance thymoquinone which has activities of antimicrobial and antifungal so as to prevent the growth of fungi and inhibit the formation of aflatoxins thereby increasing the efficiency of nutrients in feed.

The surface area of the intestine such as the height of the villi describes the area for absorption of nutrients. Villi are small finger or leaf-like protrusions found on the mucous membrane, 0.5 to 1.5 mm long and found only in the small intestine. The villi in the ileum are finger-like in shape and shorter than the villi found on the duodenum and jejunum. One of the parameters used to measure the quality of growth is the intestinal morphological structure (Wang and Peng, 2008).

The intestinal gland (Lieberkuhn's gland) has a small hole that becomes the mouth of the glandular simplex tubule. The intestinal glands are scattered between the villi attached to the mucous membrane. The intestinal glands and intestinal villi are covered by an epithelium, consisting of goblet cells and enterocytes, among others. Goblet cells secrete mucus to lubricate and protect the surface of the intestine, while the enterocytes in crypt secrete large amounts of water and electrolytes. Goblet cells secrete a kind of mucus, namely mucin which functions to coat the intestinal tract and protect pathogens that can damage intestinal epithelial cells so that the number of goblet cells is important for the health of broilers (Forder *et al.*, 2007). The presence of goblet cells in the broiler duodenum was available in sufficient numbers in the body of broilers since before hatching, but the number of goblet cells in the jejunum and ileum was only reached after the broilers have hatched (Reynold *et al.*, 2020).

CONCLUSION

The supplementation of black cumin (*Nigella sativa*) at dose of 72 mg/kg bw/day through drinking water could increase the histological sizes of the small intestine and large intestine of broilers.

ACKNOWLEDGMENT

Deleted: into crypt

Formatted: Font color: Green

Deleted:

Deleted: '

Deleted: Figure

Deleted: Figure

Deleted: Figure

Deleted: Figure

Formatted: Font color: Green

Deleted: 4

Deleted: ; Rostinawati, 2008

Formatted: Font color: Green

Deleted: s

Formatted: Font color: Green

Formatted: Font: Italic, Font color: Green

Formatted: Font color: Green

Formatted: Font: Italic, Font color: Green

Formatted: Font color: Green

Formatted: Font color: Green

Formatted: Font color: Green

Deleted: The conclusions of this study were the

Formatted: Font color: Green

Deleted: could significantly

Formatted: Font color: Green

Deleted: increase (P<0.05) to 1) the average sizes of villi height and villi area of small intestine (duodenum, jejunum, ileum) and large intestine (colon) of broiler chickens.; 2) (The average sizes of basal villi width, villi apex width, and gland diameter of broiler duodenum; and 3) the average size of apex villi width of broiler colon are also improved...

Authors would like to thank University of Lampung for providing research funding from the University of Lampung Grants in 2020.

Table 1. The average measurement of each parameter in broiler duodenum supplemented by black cumin through drinking water

Treatment	Duodenum					
	A1	B1	C1	D1	E1	F1
	-----Mean ± SD (µm)-----					
P0	644.55±71.73 ^{ab}	149.78±7.72 ^a	74.96±8.11 ^a	75.70±1.35 ^{ab}	247.33±37.83 ^a	44.44±4.67 ^a
P1	442.78±2.67 ^a	158.89±32.35 ^a	60.81±20.43 ^a	65.24±33.66 ^a	263.78±51.42 ^a	52.09±3.37 ^{ab}
P2	878.11±159.06 ^b	248.89±42.22 ^b	144.90±17.95 ^b	174.59±46.61 ^b	272.78±74.64 ^a	63.72±4.86 ^b
P3	509.55±135.72 ^a	225.07±26.34 ^{ab}	97.93±19.65 ^a	116.13±50.90 ^{ab}	331.22±112.74 ^a	54.1±7.02 ^{ab}

P0= Drinking water without *Nigella sativa* , P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P3= Drinking water with *Nigella sativa* 144 mg/kg BW/day, A1= Villi height of duodenum, B1= Basal villi width of duodenum, C1= Apex villi width of duodenum, D1= Villi area of duodenum, E1= Crypt depth of duodenum, F1= Gland diameter of duodenum). ^{a, b, ab}Different superscripts with letters in the same column indicate significant differences (P<0.05)

Table 2. The average measurement of each parameter in broiler jejunum supplemented by black cumin (*Nigella sativa*) through drinking water

Treatment	Jejunum					
	A1	B1	C1	D1	E1	F1
	-----Mean ± SD (µm)-----					
P0	342.34 ± 111.40 ^a	148.01 ± 19.47 ^a	86.43 ± 39.58 ^a	42.28 ± 22.93 ^a	231.22 ± 134.32 ^a	55.83 ± 2.87 ^a
P1	320.67 ± 176.51 ^a	170.63 ± 58.27 ^a	161.23 ± 93.12 ^a	47.71 ± 32.24 ^a	224.44 ± 95.06 ^a	51.52 ± 3.82 ^a
P2	890.17 ± 242.62 ^b	188.39 ± 34.90 ^a	138.56 ± 41.97 ^a	126.38 ± 38.38 ^b	260.06 ± 86.15 ^a	64.20 ± 6.23 ^a
P3	551.53 ± 128.51 ^{ab}	105.5 ± 20.63 ^a	44.80 ± 8.44 ^a	42.15 ± 17.44 ^a	191.00 ± 38.40 ^a	54.16 ± 7.41 ^a

P0= Drinking water without *Nigella sativa* , P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P3= Drinking water with *Nigella sativa* 144 mg/kg BW/day, A1= Villi height of jejunum, B1= Basal villi width of jejunum, C1= Apex villi width of jejunum, D1= Villi area of jejunum, E1= Crypt depth of jejunum, F1= Gland diameter of jejunum). ^{a, b, ab}Different superscripts with letters in the same column indicate significant differences (P<0.05)

Table 3. The average measurement of each parameter in broiler ileum supplemented by black cumin (*Nigella sativa*) through drinking water.

Treatment	Ileum					
	A1	B1	C1	D1	E1	F1
	-----Mean ± SD (µm)-----					
P0	290.11 ± 25.06 ^a	155.40 ± 9.59 ^a	65.64 ± 16.48 ^a	45.43 ± 18.92 ^a	140.26 ± 18.97 ^a	54.43 ± 9.23 ^a
P1	289.00 ± 88.88 ^a	191.56 ± 37.02 ^a	106.00 ± 59.13 ^a	45.41 ± 26.65 ^a	178.11 ± 52.14 ^a	50.56 ± 3.02 ^a
P2	557.22 ± 31.45 ^b	244.78 ± 52.74 ^a	162.33 ± 15.84 ^a	91.04 ± 2.33 ^b	174.34 ± 37.86 ^a	52.78 ± 3.71 ^a
P3	327.05 ± 87.56 ^a	144.68 ± 52.87 ^a	98.26 ± 43.50 ^a	39.33 ± 18.74 ^a	128.93 ± 22.06 ^a	47.60 ± 12.7 ^a

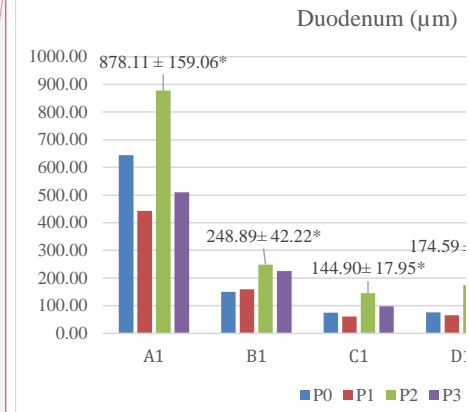
P0= Drinking water without *Nigella sativa* , P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P3= Drinking water with *Nigella sativa* 144 mg/kg BW/day, A1= Villi height of ileum, B1= Basal villi width of ileum, C1= Apex villi width of ileum, D1= Villi area of ileum, E1= Crypt depth of ileum, F1= Gland diameter of ileum). ^{a, b, ab}Different superscripts with letters in the same column indicate significant differences (P<0.05)

Table 4. The average measurement of each parameter in broiler colon supplemented by black cumin (*Nigella sativa*) through drinking water.

Treatment	Colon					
	A1	B1	C1	D1	E1	F1
	-----Mean ± SD (µm)-----					
P0	259.22 ± 33.07 ^{ab}	132.82 ± 44.40 ^a	64.95 ± 4.74 ^{ab}	25.15 ± 3.85 ^a	128.81 ± 28.46 ^a	60.80 ± 11.19 ^a
P1	193.56 ± 25.20 ^a	178.39 ± 38.25 ^a	66.10 ± 27.36 ^{ab}	23.76 ± 5.23 ^a	201.33 ± 28.41 ^b	49.29 ± 7.77 ^a

Deleted: The Deleted: a

Deleted: ¶



¶ **Figure 1.** The supplementation of black cumin (*Nigella sativa*) 72 mg/kg BW (P2) in drinking water increased the average villi height (A1), basal villi width (B1), villi apex width (C1), villi area (D1), and gland diameter (F1) of broiler duodenum. ¶ Asterisks (*) indicate the most significant increase (P<0.05)¶ Figure 1 (delete saja tdk diperlukan lagi sudah ada dalam tabel)¶

Formatted: Indent: Left: 0 cm, Hanging: 1.25 cm

Formatted Table

Formatted: Font color: Green

Formatted: Font: 8 pt, Font color: Green

Formatted: Font color: Green

Formatted: Font: 8 pt, Font color: Green

Formatted: Font color: Green

Formatted: Font: 8 pt, Font color: Green

Formatted: Font color: Green

Formatted: Font: 8 pt, Font color: Green

Formatted: Font color: Green

Formatted: Font: 8 pt, Font color: Green

Formatted: Font color: Green

Deleted: Information: Drinking water without *Nigella sativa* (P0), Drinking water with *Nigella sativa* 36 mg/kg BW/day (P1), 72 mg/kg BW/day (P2), 144 mg/kg BW/day (P3); A1 (villi height of jejunum), B1 (basal villi width of jejunum), C1 (apex villi width of jejunum), D1 (villi area of jejunum), E1 (crypt depth of jejunum), F1 (gland diameter of jejunum). Different superscripts with letters in the same column indicate significant differences (P<0.05).¶

¶

Formatted Table

Formatted: Font color: Green

Formatted: Indent: First line: 0 cm

Deleted: Information: Drinking water without *Nigella sativa* (P0), Drinking water with *Nigella sativa* 36 mg/kg BW/day (P1), 72 mg/kg BW/day (P2), 144 mg/kg BW/day (P3); A1 (villi height of jejunum), B1 (basal villi width of jejunum), C1 (apex villi width of jejunum), D1 (villi area of jejunum), E1 (crypt depth of jejunum), F1 (gland diameter of jejunum). Different superscripts with letters in the same column indicate significant differences (P<0.05).¶

Formatted Table

Treatment	Colon					
	A1	B1	C1	D1	E1	F1
	-----Mean ± SD (µm)-----					
P2	312.56 ± 35.84 ^b	193.56 ± 19.91 ^a	117.39 ± 32.55 ^b	49.26 ± 9.86 ^b	157.11 ± 12.36 ^{ab}	51.85 ± 6.08 ^a
P3	193.22 ± 45.30 ^a	118.53 ± 13.63 ^a	50.97 ± 8.34 ^a	19.03 ± 7.77 ^a	138.07 ± 29.86 ^{ab}	51.14 ± 7.58 ^a

P0= Drinking water without *Nigella sativa* . P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P1= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 144 mg/kg BW/day. A1= Villi height of colon, B1= Basal villi width of colon, C1= Apex villi width of colon, D1= Villi area of colon, E1= Crypt depth of colon, F1= Gland diameter of colon). ^{a, b, ab}Different superscripts with letters in the same column indicate significant differences (P<0.05).

REFERENCES

- Aughey, E. and F.L. Frye. 2001. **Comparative Veterinary Histology with Clinical Correlates**. CRC Press, Florida.
- Asmawati. 2014. The Effect of In Ovo Feeding on Hatching Weight and Small Intestinal Tissue Development of Native Chicken. **Dissertation**. Hasanuddin University, Makassar.
- Austic, R.E. and M.C. Nesheim. 1990. **Poultry Production**. Lea & Febiger, Philadelphia.
- Awad, W.A., K. Ghareeb, S. Nitclu, S. Pasteiner, S.A. Raheem, and J. Bohm. 2008. Effect of dietary inclusion of probiotic, prebiotic and symbiotic on intestinal glucose absorption of broiler chickens. **J. Poult. Sci.** 7:688-691.
- BPS [Badan Pusat Statistik]. 2019. Populasi Ayam Ras Pedaging menurut Provinsi (Ekor) Tahun 2009-2018. <https://www.bps.go.id/dynamic/ta/2015/12/18/1034/populasi-ayam-ras-pedaging-menurut-provinsi-2009-2018.html>. Diakses pada tanggal 13 Agustus 2020.
- Forder, R.E.A., G.S. Howarth, D.R. Tivey, and R.J. Hughes. 2007. Bacterial modulation of small intestinal goblet cells and mucin composition during early posthatch development of poultry. **Poult. Sci.** 86(11):2396-2403.
- Guyton, A.C. 1997. **Fisiologi Kedokteran Edisi ke-9**. Universitas Indonesia Press, Jakarta.
- Harimurti, S. dan E.S. Rahayu. 2009. Morfologi usus ayam broiler yang disuplementasi dengan probiotik strain tunggal dan campuran. **Agritech**. 29(3): 179-183.
- Hasan, D.I., Sugito, M. Sabri, M. Hambal, and U. Balqis. 2019. Effect of Black Cumin Oil Administration on Cortisol Level and Liver Histopathology of Heat Stressed Broiler Chickens. **Jurnal Kedokteran Hewan**. 13(1): 31-36
- Ibrahim, S. 2008. Hubungan ukuran-ukuran usus halus dengan berat badan broiler. **Agripet**. 8(2):42-46.
- Iji, P.A., R.J. Hughes, M. Choct, and D. R. Tivey. 2001. Intestinal structure and function of broiler chickens on wheat-based diets supplemented with microbial enzyme. **Asian-Aust. J. Anim. Sci.** 14(1):54-60.
- Khedr, N., F. Abdel-Fattah. 2007. Response of Broiler Chickens to Diet Containing Black Seed (*Nigella sativa* L.) as Medical Plant. **Benha Vet. Med. J.** 17(2): 323-341
- Kruk, L. T. Michalska, K. Lichszeld, A. Kladna, and H.Y. Aboul-Enein. 2000. The effect of thymol and its derivatives on reactions on generating reactive oxygen species. **Chemosphere**. 41:1059-1064.
- Marshall, R.M. and S.B. Levy. 2011. Food animals and antimicrobials: Impacts on human health. **Clin. Microbiol. Rev.** 24(4):718-733.
- Mile, R.D., G.D. Butcher, P.R. Henry, R.C. Little. 2006. Effect of antibiotic growth promoters on broiler performance, intestinal growth parameters, and quantitative morphology. **J. Poultry Sci.** 85:476-485.
- Miraghaee, S.S., B. Heidary, H. Almasi, A. Shabani, M. Elahi, and M. H. M. Nia. 2011. The effects of *Nigella sativa* powder (black seed) and *Echinacea purpurea* (L.) Moench extract on performance, some blood biochemical and hematological parameters in broiler chickens. **African Journal of Biotechnology**. 10(82): 19249-19254
- Ningtias, A.S. 2013. Comparison of Growth Performance of Broilers, Kampong, and Backcross3 (Gallus gallus domesticus Linnaeus, 1758) Based on Morphometri and Histological Structure of Ileum and Breast Muscle. **Thesis**. Fakultas Biologi Universitas Gajah Mada. Yogyakarta.
- Rahmawati. 2016. Histologis saluran pencernaan ayam buras hasil in ovo feeding asam amino l-arginine. **Thesis**. Fakultas Peternakan Universitas Hasanuddin. Makassar.
- Reynold, K.L., S.E. Cloft., and E.A. Wong. 2020. Changes with age in density of goblet cells in the small intestine of broiler chicks. **Poult. Sci.** 99(5):2342-2348.
- Rofiq, M. N. 2003. Pengaruh Pakan Berbahan Baku Local Terhadap Performans Vili Usus. **J. Sains dan Teknologi**. 5(5): 190-194.
- Salem, M.L. 2005. Immunomodulatory and therapeutic properties of the *Nigella sativa* L. seed. **Int. Immunopharmacol.** 5(13-14): 1749-1770.
- Smirnov, A., R. Perez, E. Amit-Romach, D. Sklan, and Z. Uni. 2005. Mucin dynamics and microbial populations in chicken small intestine changed by dietary probiotic and antibiotic growth promoter supplementation. **J. Nutrition**. 135:187-192.
- Sugito, W. Manalu, D.A. Astuti, E. Handharyani, and Chairul. 2007. Histopatologi hati dan ginjal pada ayam broiler yang dipapar cekaman panas dan diberi ekstrak kulit batang Jaloh (*Salix tetrasperma* Roxb). **J. Ilmu Ternak dan Veteriner**. 12(1):68-72.
- Sulistiawati, F., M. Radji. 2014. Potensi Pemanfaatan *Nigella sativa* L. sebagai Immunomodulator dan Antiinflamasi. **Pharm. Sci. Res.** 1(2): 65-77
- Sun, X., A. McElroy, Jr. Webb, A.E.K.E. Sefton, and C. Novak. 2005. Broiler performance and intestinal alterations when fed drug-free diets. **J. Poult. Sci.** 84:1294-1302.
- Suprijatna, E., U. Atmomasrono, and R. Kartasudjana. 2008. **Ilmu Dasar Ternak Unggas**. Penebar Swadaya, Jakarta.

Formatted Table

Formatted: Indent: Left: 0 cm, First line: 0 cm

Deleted: Information: Drinking water without *Nigella sativa* (P0), Drinking water with *Nigella sativa* 36 mg/kg BW/day (P1), 72 mg/kg BW/day (P2), 144 mg/kg BW/day (P3); A1 (villi height of colon), B1 (basal villi width of colon), C1 (apex villi width of colon), D1 (villi area of colon), E1 (crypt depth of colon), F1 (gland diameter of colon). Different superscripts with letters in the same column indicate significant differences (P<0.05).¶

Deleted: ¶

Deleted: Alfiansyah, M. 2011. Anatomi dan Pencernaan Usus

Deleted: ¶

Formatted: Font color: Green

Formatted: Font color: Green

Formatted: Font color: Green

Formatted: Font color: Green

Formatted: Font color: Green

Formatted: Font: Bold, Font color: Green

Formatted: Font color: Green

Formatted: Font: Italic

Formatted: Font: Bold

Formatted: Font color: Green

Formatted: Font color: Green

Formatted: Font: Bold, Font color: Green

Formatted: Font color: Green

Deleted: Kabir, S.M.L. 2010. Avian colibacillosis and

Formatted: Font color: Green

Formatted: Font: Bold, Font color: Green

Formatted: Font color: Green

Deleted: Rostika N. 2012. Pengaruh pemberian ekstrak minyak

Deleted: Siagian, Y.A. 2016. Gambaran Histologis dan Tingg

Formatted: Font color: Green

Formatted: Font: Bold, Font color: Green

Formatted: Font color: Green

Deleted: Sturkie, P.D., G.C. Whitlow. 2000. Sturkie's Avian

Formatted: Font color: Green

Formatted: Font: Italic, Font color: Green

Formatted: Font color: Green

Formatted: Font: Bold, Font color: Green

Formatted: Font color: Green

Deleted: Sun, X. 2004. Broiler performance and intestinal

Tavakkoli, A., A. Ahmadi, B.M. Razavi, H. Hosseinzadeh. 2017. Black Seed (Nigella Sativa) and its Constituent Thymoquinone as an Antidote or a Protective Agent Against Natural or Chemical Toxicities. *Jranian Journal of Pharmaceutical Research*. 16(Suppl): 2-23.

Wang, J.X. and K.M. Peng. 2008. Molecular, cellular, and developmental biology developmental morphology of the small intestine of African ostrich chicks. *J. Poultry Sci*. 87: 2629-2635.

Wang, H., Y. Guo, J.C.H.H. Shih, 2008. Effects of dietary supplementation of keratinase on growth performance, nitrogen retention and intestinal morphology of broiler chickens fed diets with soybean and cottonseed meals. *Anim. Feed. Sci. Tech*. 140 (3-4): 376-384.

Formatted: Font: Bold

Deleted: Thippeswamy N.B., K.A. Naidu. 2005. Antioxidant potency of cumin varieties cumin, black cumin and bitter cumin on antioxidant systems. *Euro Food Res Tech*. 220: 472-476 (Delete tidak dikutip)

Deleted: Topozoda H.H., H.A. Mazloun, M. El-Dakhkhny. 1965. The antibacterial properties of Nigella sativa seeds. *J. Egypt Med*. 48:187-202. (Delete tidak dikutip)
 Yusuf, M.S. 2014. Efektivitas Penggunaan Jintan Hitam (Nigella sativa) dalam Proses Percepatan Penyembuhan Luka Setelah Pencabutan Gigi. *Thesis*. Faculty of Dentistry Hasanuddin University, Makassar. (Delete tidak dikutip)

Deleted: (Mardhiah, 1991). (Lengkapi)

Wang *et al*

Formatted: Font color: Green

Deleted: ..

Formatted: Font: Bold

Formatted: Font: Bold

Deleted: (Lengkapi)
 Rofiq (2003) (Lengkapi)

Skripsi tidak boleh dipakai sebagai referensi
 Jumlah jurnal yang dipakai sebagai referensi belum 70%

THE EFFECT OF BLACK CUMIN (*Nigella sativa*) SUPPLEMENTATION THROUGH DRINKING WATER ON THE HISTOLOGY OF SMALL INTESTINE AND LARGE INTESTINE OF BROILER CHICKENS

Madi Hartono¹, Purnama Edy Santosa², Ratna Ermawati², and Muhammad Mirandy Pratama Sirat^{1*}

¹Study Program of Animal Nutrition and Feed Technology, Department of Animal Husbandry, Faculty of Agriculture, University of Lampung, Bandar Lampung, Indonesia

²Study Program of Animal Husbandry, Department of Animal Husbandry, Faculty of Agriculture, University of Lampung, Bandar Lampung, Indonesia

*Corresponding author: m.mirandy@fp.unila.ac.id

ABSTRACT

This study aimed to determine the effect of black cumin (*Nigella sativa*) supplementation through drinking water on the histology of broiler chickens' small intestine and large intestine. The research was conducted from April-September 2020 in a cage facility of the Integrated Field Laboratory, Faculty of Agriculture, University of Lampung. This research used a completely randomized design with four treatment groups and three replications (five broilers per replication) with a total of 60 male broilers. The treatments were drinking water without black cumin (P0, control); drinking water with black cumin 36 mg/kg BW/day (P1); 72 mg/kg BW/day (P2); and 144 mg/kg BW/day (P3). Three broilers from each group were randomly necropsied at 31 days old, and samples of the small intestine (duodenum, jejunum, and ileum) and large intestine were fixed with 10% formalin solution and sent to the Lampung Disease Investigation Center for histological preparations. The observation of preparations was carried out microscopically using the Leica DM500[®] Binocular Microscope to accurately calculate various parameter sizes. The results were analyzed statistically with one way analysis of variance at significant level 5% and if proven significant, then a Tukey test was conducted. The results of this study were that the supplementation of black cumin 72 mg/kg BW/day through drinking water could significantly increase ($P < 0.05$) 1) the average sizes of villi height and villi area of small intestine (duodenum, jejunum, ileum) and large intestine (colon) of broiler chickens; 2) the average sizes of basal villi width, villi apex width, and gland diameter of broiler duodenum; and 3) the average size of apex villi width of broiler colon. The conclusion of this study was the supplementation of black cumin at dose of 72 mg/kg BW/day through drinking water could increase the histological sizes of the small intestine and large intestine of broilers.

Key words: broiler, histology, large intestine, *Nigella sativa*, small intestine

ABSTRAK

Penelitian ini bertujuan mengetahui pengaruh suplementasi jintan hitam (*Nigella sativa*) melalui air minum terhadap histologi usus halus dan usus besar broiler. Penelitian dilakukan pada April-September 2020 di unit kandang Laboratorium Lapang Terpadu, Fakultas Pertanian, Universitas Lampung. Penelitian bersifat eksperimental menggunakan Rancangan Acak Lengkap dengan empat kelompok perlakuan dan tiga ulangan (lima ekor tiap ulangan) sehingga total 60 ekor broiler jantan. Perlakuan yang diberikan yaitu pemberian air minum tanpa jintan hitam (P0, kontrol); air minum dengan jintan hitam 36 mg/kg bobot badan/hari (P1); 72 mg/kg bobot badan/hari (P2); dan 144 mg/kg bobot badan/hari (P3). Tiga ekor dari tiap kelompok secara acak dinekropsi pada hari ke-31 dan diambil sampel organ usus halus (duodenum, jejunum, ileum) dan usus besar (colon) kemudian difiksasi dengan larutan formalin 10% dan dikirim ke Balai Veteriner Lampung untuk pembuatan preparat histologi. Pengamatan preparat dilakukan secara mikroskopis menggunakan Mikroskop Binokuler Leica DM500[®] untuk menghitung berbagai ukuran parameter secara akurat. Analisis hasil dilakukan secara statistik menggunakan analisis sidik ragam satu arah dengan taraf signifikansi 5% dan dilanjutkan dengan uji lanjut Tukey. Suplementasi jintan hitam 72 mg/kg bobot badan/hari dapat meningkatkan secara signifikan ($P < 0,05$) terhadap 1) ukuran rata-rata tinggi vili dan luas vili usus halus (duodenum, jejunum, ileum) dan usus besar broiler; 2) ukuran rata-rata tinggi vili, lebar vili basal, lebar puncak vili, luas vili, dan diameter kelenjar duodenum broiler; dan 3) ukuran rata-rata lebar puncak vili usus besar broiler. Disimpulkan bahwa suplementasi jintan hitam (*Nigella sativa*) dosis 72 mg/kg bobot badan/hari melalui air minum dapat meningkatkan ukuran histologi usus halus dan usus besar broiler.

Kata kunci: broiler, histologi, usus besar, *Nigella sativa*, usus halus

INTRODUCTION

Intensive maintenance of broiler can often induce stress for the animal, resulting in a decreased responsiveness in the immune system, which increases the likelihood for disease and illness. Disease outbreaks are a serious obstacle in the broiler farming industry. The high incidence of disease can decrease productivity and even kill livestock which results in significant losses for breeders. Diseases can lead to decrease in body organ function, particularly the digestive organ. While antibiotics might prevent these diseases, the administration of antibiotics for long periods might cause residual buildup and antibiotic resistance in bacteria, which could have negative effects if consumed by humans (Marshall and Levy, 2011).

One strategy to maintain digestive tract quality and function, and to reduce the residual antibiotics in broilers is to provide them with natural herbs such as black cumin (*Nigella sativa*) as immunomodulation (Sulistiawati and Radji, 2014) and antibiotic growth promoter substitution (Miraghaee *et al.*, 2011) with a high content of antioxidants thymoquinone as antioxidant that could eliminate free radicals (Kruk *et al.*, 2000), anti-inflammatory effects of several inflammation (Salem, 2005). *Nigella sativa* also has the ability to protect potentially different tissues and organs including liver, kidney, heart, blood, brain, lungs, reproductive system and gastrointestinal against chemical poison (Tavakkoli *et al.*, 2017) and administration of *Nigella sativa* also can minimize heat stress in broilers by reducing cortisol and levels

minimize histopathological changes in the liver (Hasan *et al.*, 2019). This study was conducted to determine the effect of black cumin supplementation through histological studies of the digestive organs of broiler chickens which are expected to potentially increase the villi height, apex villi width, basal villi width, villi area, crypt depth, and gland diameter of small intestine (duodenum, jejunum, and ileum) and large intestine (colon).

MATERIALS AND METHODS

This research was carried out for six months (April-September 2020) in the Integrated Field Laboratory enclosure unit of the Faculty of Agriculture, University of Lampung. This research was designed using a completely randomized design (CRD) with four treatment groups and three replications with a total of 60 male broilers were used (five broilers per replication). The treatment dose was according to the broiler body weight, namely 1) drinking water without black cumin (P0, control); drinking water with black cumin at dose of 36 mg/kg BW/day (P1); drinking water with black cumin at dose of 72 mg/kg BW/day (P2); and drinking water with black cumin at dose of 144 mg/kg BW/day (P3). On the 31st day, three broilers from each group were randomly sacrificed and samples of the small intestine (duodenum, jejunum, ileum) and large intestine (colon) were then fixed with 10% formalin solution and sent to the Lampung Veterinary Disease Investigation Center for histological preparations with Hematoxylin Eosin (HE) staining. The preparation observation was carried out microscopically using the Leica DM500@ Binocular Microscope Technology to accurately calculate various parameter sizes.

The research parameters were villi height, villi apex width, basal villi width, villi area, crypt depth, and gland diameter. The observation of histological preparations was done with a 10x magnification objective lens. The calculation of each parameter was carried out as many as three villi (villi height, villi apex width, basal villi width, villi area, crypt depth) and three gland diameters in each digestive tract organ (duodenum, jejunum, ileum, and colon). There were three replications per organ, so that the total for each organ was obtained through an average of the nine villi and nine gland diameters.

The calculation of the surface area of the intestinal villi using the method of Iji *et al.* (2001) which was slightly modified with the assumption that the villi model is an analogue of the trapezium shape so that the average number of the apical widths of the villi plus the average basal width of the villi is divided by two then multiplied by the height of the villi by the following formula.

$$\text{Surface area} = \frac{b + c}{2} \times a$$

a= height of intestine villi

b= apex width of intestine villi

c= basal width of basal of intestine villi

Data Analysis

The average measurement data for various parameters of each digestive tract organ (duodenum, jejunum, ileum, and colon) were analyzed using analysis of variance, and followed by Tukey test.

RESULTS AND DISCUSSION

The average measurements of villi height, villi apex width, basal villi width, villi area, crypt depth, and gland diameter of the digestive tract organs (duodenum, jejunum, ileum, colon) are presented in each table. Each shows the calculation of each parameter from the nine villi (villi height, basal villi width, apex villi width, villi area, crypt depth) and nine gland diameters on histopathological preparations in each digestive tract organ (duodenum, jejunum, ileum, large intestine) in each treatment.

The average measurement of each parameter in broiler duodenum, jejunum, ileum, and colon were presented in Table 1, Table 2, Table 3, Table 4 respectively. Supplementation of black cumin with a dose of 72 mg/kg of broiler body weight (P2) had a significant effect on the increase in the average villi height, basal villi width, villi apex width, villi area, and gland diameter of the duodenum; the average villi height and villi area of jejunum; the average villi height and villi area of the ileum; and the average size of villi height, apex villi width, villi area of colon.

The ability of digestion and absorption of food substances could be affected by the surface area of the intestinal epithelium, the number of folds, and the number of villi and microvilli that expand the absorption field (Austic and Nesheim, 1990; Ibrahim, 2008). It could also be influenced by the height and surface area of the villi organs or digestive tract (Sugito *et al.*, 2007; Ibrahim, 2008). The development of the intestinal villi in broiler chickens is related to the function of the intestine and growth of the chicken (Sun *et al.*, 2005). Villi are places for absorption of nutrients, the wider the villi, the more food substances that will be absorbed, in the end it can have an impact on the growth of organs and increased carcass (Asmawati, 2014).

Treatment with a dose of 72 mg/kg BW/day (P2) showed the most significant increase ($P < 0.05$) in villi height and villi area of all broilers digestive organs (duodenum, jejunum, ileum, colon) compared to other treatments. The increase in villi height in the broiler intestine is closely related to an increase in digestive function and absorption function due to the expansion of the absorption area and is an expression of the smooth transportation system of nutrients throughout the body (Awad *et al.*, 2008). One of the parameters that can be used to measure the quality of growth is the morphological structure of the intestine. The height of villi in all parts of the small intestine (duodenum, jejunum, ileum) and large intestine in general increases (Ningias, 2013). Increasing the villi width and the villi height can expand the absorption area of the villi. According to Asmawati (2014), the wider the villi, the

Table 1. The average measurement of each parameter in broiler duodenum supplemented by black cumin through drinking water

Treatment	Duodenum					
	A1	B1	C1	D1	E1	F1
	Mean±SD (µm)					
P0	644.55±71.73 ^{ab}	149.78±7.72 ^a	74.96±8.11 ^a	75.70±1.35 ^{ab}	247.33±37.83 ^a	44.44±4.67 ^a
P1	442.78±2.67 ^a	158.89±32.35 ^a	60.81±20.43 ^a	65.24±33.66 ^a	263.78±51.42 ^a	52.09±3.37 ^{ab}
P2	878.11±159.06 ^b	248.89±42.22 ^b	144.90±17.95 ^b	174.59±46.61 ^b	272.78±74.64 ^a	63.72±4.86 ^b
P3	509.55±135.72 ^a	225.07±26.34 ^{ab}	97.93±19.65 ^a	116.13±50.90 ^{ab}	331.22±112.74 ^a	54.1±7.02 ^{ab}

^{a, b, ab}Different superscripts in the same column indicate significant differences (P<0.05). P0= Drinking water without *Nigella sativa*, P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P3= Drinking water with *Nigella sativa* 144 mg/kg BW/day, A1= Villi height of duodenum, B1= Basal villi width of duodenum, C1= Apex villi width of duodenum, D1= Villi area of duodenum, E1= Crypt depth of duodenum, F1= Gland diameter of duodenum)

Table 2. The average measurement of each parameter in broiler jejunum supplemented by black cumin (*Nigella sativa*) through drinking water

Treatment	Jejunum					
	A1	B1	C1	D1	E1	F1
	Mean±SD (µm)					
P0	342.34±111.40 ^a	148.01±19.47 ^a	86.43±39.58 ^a	42.28±22.93 ^a	231.22±134.32 ^a	55.83±2.87 ^a
P1	320.67±176.51 ^a	170.63±58.27 ^a	161.23±93.12 ^a	47.71±32.24 ^a	224.44±95.06 ^a	51.52±3.82 ^a
P2	890.17±242.62 ^b	188.39±34.90 ^a	138.56±41.97 ^a	126.38±38.38 ^b	260.06±86.15 ^a	64.20±6.23 ^a
P3	551.53±128.51 ^{ab}	105.5±20.63 ^a	44.80±8.44 ^a	42.15±17.44 ^a	191.00±38.40 ^a	54.16±7.41

^{a, b, ab}Different superscripts in the same column indicate significant differences (P<0.05). P0= Drinking water without *Nigella sativa*, P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P3= Drinking water with *Nigella sativa* 144 mg/kg BW/day, A1= Villi height of jejunum, B1= Basal villi width of jejunum, C1= Apex villi width of jejunum, D1= Villi area of jejunum, E1= Crypt depth of jejunum, F1= Gland diameter of jejunum)

Table 3. The average measurement of each parameter in broiler ileum supplemented by black cumin (*Nigella sativa*) through drinking water

Treatment	Ileum					
	A1	B1	C1	D1	E1	F1
	Mean±SD (µm)					
P0	290.11±25.06 ^a	155.40±9.59 ^a	65.64±16.48 ^a	45.43±18.92 ^a	140.26±18.97 ^a	54.43±9.23 ^a
P1	289.00±88.88 ^a	191.56±37.02 ^a	106.00±59.13 ^a	45.41±26.65 ^a	178.11±52.14 ^a	50.56±3.02 ^a
P2	557.22±31.45 ^b	244.78±52.74 ^a	162.33±15.84 ^a	91.04±2.33 ^b	174.34±37.86 ^a	52.78±3.71 ^a
P3	327.05±87.56 ^a	144.68±52.87 ^a	98.26±43.50 ^a	39.33±18.74 ^a	128.93±22.06 ^a	47.6 ±12.7 ^a

^{a, b, ab}Different superscripts in the same column indicate significant differences (P<0.05). P0= Drinking water without *Nigella sativa*, P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P3= Drinking water with *Nigella sativa* 144 mg/kg BW/day, A1= Villi height of ileum, B1= Basal villi width of ileum, C1= Apex villi width of ileum, D1= Villi area of ileum, E1= Crypt depth of ileum, F1= Gland diameter of ileum)

Table 4. The average measurement of each parameter in broiler colon supplemented by black cumin (*Nigella sativa*) through drinking water

Treatment	Colon					
	A1	B1	C1	D1	E1	F1
	Mean±SD (µm)					
P0	259.22±33.07 ^{ab}	132.82±44.40 ^a	64.95±4.74 ^{ab}	25.15±3.85 ^a	128.81±28.46 ^a	60.80±11.19 ^a
P1	193.56±25.20 ^a	178.39±38.25 ^a	66.10±27.36 ^{ab}	23.76±5.23 ^a	201.33±28.41 ^b	49.29±7.77 ^a
P2	312.56±35.84 ^b	193.56±19.91 ^a	117.39±32.55 ^b	49.26±9.86 ^b	157.11±12.36 ^{ab}	51.85±6.08 ^a
P3	193.22±45.30 ^a	118.53±13.63 ^a	50.97±8.34 ^a	19.03±7.77 ^a	138.07±29.86 ^{ab}	51.14±7.58 ^a

^{a, b, ab}Different superscripts in the same column indicate significant differences (P<0.05). P0= Drinking water without *Nigella sativa*, P1= Drinking water with *Nigella sativa* 36 mg/kg BW/day, P2= Drinking water with *Nigella sativa* 72 mg/kg BW/day, P3= Drinking water with *Nigella sativa* 144 mg/kg BW/day, A1= Villi height of colon, B1= Basal villi width of colon, C1= Apex villi width of colon, D1= Villi area of colon, E1= Crypt depth of colon, F1= Gland diameter of colon)

more food substances will be absorbed, which can have a long-term impact on the growth of the body's organs. Similarly, according to Rahmawati (2016) the higher the size of the villi, the wider the area of nutrient absorption by the small intestine wall, thus will trigger the growth improvement. Guyton (1997) added that a more villous surface area leads to a more efficient nutrient absorption. The efficiency of nutrient absorption cannot be separated from the work of hormonal, nervous, and digestive glands in the digestive tract and its accessory glands.

Crypts are contained in the intestinal villi, which are composed of inline cylindrical epithelial cells. These

glands produce mucus and several enzymes for the metabolism of peptides, fats, carbohydrates, and intestinal juices (mucin) which function to protect the intestinal mucosa (Aughey and Frye, 2001). The increase in the average size of the gland diameter in the treatment with supplementation of black cumin in drinking water showed an increase in the size of the duodenum's gland diameter. The P2 treatment showed a significant increase (P<0.05) in size compared to other treatments, it can support the development of epithelial cells that make up the villi, which will increase the absorption of nutrients in the digestive tract.

Supplementation of black cumin 36 mg/kg BW/day (P1) showed the most significant increase ($P < 0.05$) in parameter of crypt depth of colon but was not different from the dose of P2 and P3 (Table 4). According to Sun *et al.* (2005) and Smirnov *et al.* (2005) that crypt depth has no effect after broilers are more than 28 days old. In this study, samples of broilers digestive tract organs were collected at the age of 31 days. It is assumed that the development of intestinal morphology is closely related to the role of micronutrients in line with the increasing age of broilers (Harimurti and Rahayu, 2009).

Based on the data presented in Table 1, Table 2, Table 3, and Table 4, the supplementation of black cumin at dose of 72 mg/kg BW/day had most significant effect to the size of the villi height and villi area of all digestive tract organs compared to the control group. One of the parameters that can be used to measure the quality of growth is the morphological structure of the intestine (Wang *et al.*, 2008; Ningtias, 2013). According to Suprijatna *et al.* (2008), the small intestine is the main organ for digestion and absorption of digestive products. Various enzymes that enter this channel function to accelerate and streamline the breakdown of carbohydrates, proteins and fats to facilitate the absorption process. In adult chickens, the length of the small intestine is about 62 inches or 1.5 meters.

One of the parameters that can be used to measure the quality of growth is the morphological structure of the intestine (Wang and Peng, 2008). The carrying capacity of the digestive process for the given feed and nutrient absorption can be influenced by the surface area of the intestinal epithelium, the number of folds in it, the height of the villi, the number of villi and microvilli that expand the absorption area (Ruttanavut *et al.*, 2009). The development of the intestinal villi in broiler chickens is related to the function of the intestine and growth of the chicken (Sun *et al.*, 2005). The increase in villi causes more villi surface area to absorb nutrients into the bloodstream (Mile *et al.*, 2006).

High villi indicate that the intestines are better than short villi. Awad *et al.* (2008) stated that the increase in the height of the villi in the intestine with digestive and absorption functions occurs because of the intact villi form which is a smooth expression of the nutrient transport system throughout the body. Rofiq (2003) stated that the absorption of nutrients in the intestine is influenced by the inner surface area of the intestine (folds, villi and microvilli) and the length of transit of the digesta in the intestine. Based on the research of Khedr and Abdel-Fattah (2007) showed that administration of *Nigella sativa* can increase broiler body weight, it is possible because *Nigella sativa* is rich in essential fatty acids such as oleic, linoleic, and linolenic acids which are essential to help growth and the presence of the active substance thymoquinone which has activities of antimicrobial and antifungal so as to prevent the growth of fungi and inhibit the formation of aflatoxins thereby increasing the efficiency of nutrients in feed.

The surface area of the intestine such as the height of the villi describes the area for absorption of nutrients. Villi are small finger or leaf-like protrusions found on the mucous membrane, 0.5 to 1.5 mm long and found only in the small intestine. The villi in the ileum are finger-like in shape and shorter than the villi found on the duodenum and jejunum. One of the parameters used to measure the quality of growth is the intestinal morphological structure (Wang and Peng, 2008).

The intestinal gland (Lieberkuhn's gland) has a small hole that becomes the mouth of the glandular simplex tubule. The intestinal glands are scattered between the villi attached to the mucous membrane. The intestinal glands and intestinal villi are covered by an epithelium, consisting of goblet cells and enterocytes, among others. Goblet cells secrete mucus to lubricate and protect the surface of the intestine, while the enterocytes in crypt secrete large amounts of water and electrolytes. Goblet cells secrete a kind of mucus, namely mucin which functions to coat the intestinal tract and protect pathogens that can damage intestinal epithelial cells so that the number of goblet cells is important for the health of broilers (Forder *et al.*, 2007). The presence of goblet cells in the broiler duodenum was available in sufficient numbers in the body of broilers since before hatching, but the number of goblet cells in the jejunum and ileum was only reached after the broilers have hatched (Reynold *et al.*, 2020).

CONCLUSION

The supplementation of black cumin (*Nigella sativa*) at dose of 72 mg/kg BW/day through drinking water could increase the histological sizes of the small intestine and large intestine of broilers.

ACKNOWLEDGMENT

Authors would like to thank University of Lampung for providing research funding from the University of Lampung Grants in 2020.

REFERENCES

- Aughey, E. and F.L. Frye. 2001. **Comparative Veterinary Histology with Clinical Correlates**. CRC Press, Florida.
- Asmawati. 2014. The Effect of In Ovo Feeding on Hatching Weight and Small Intestinal Tissue Development of Native Chicken. **Dissertation**. Hasanuddin University, Makassar.
- Austic, R.E. and M.C. Nesheim. 1990. **Poultry Production**. Lea & Febiger, Philadelphia.
- Awad, W.A., K. Ghareeb, S. Nitclu, S. Pasteiner, S.A. Raheem, and J. Bohm. 2008. Effect of dietary inclusion of probiotic, prebiotic and symbiotic on intestinal glucose absorption of broiler chickens. **Int. J. Poult. Sci.** 7(7):688-691.
- Forder, R.E.A., G.S. Howarth, D.R. Tivey, and R.J. Hughes. 2007. Bacterial modulation of small intestinal goblet cells and mucin composition during early posthatch development of poultry. **Poult. Sci.** 86(11):2396-2403.
- Guyton, A.C. 1997. **Fisiologi Kedokteran. Edisi ke-9**. Universitas Indonesia Press, Jakarta.
- Harimurti, S. and E.S. Rahayu. 2009. Morfologi usus ayam broiler yang disuplementasi dengan probiotik strain tunggal dan campuran. **Agritech.** 29(3):179-183.

- Hasan, D.I., Sugito, M. Sabri, M. Hambal, and U. Balqis. 2019. Effect of black cumin oil administration on cortisol level and liver histopathology of heat stressed broiler chickens. **J. Kedokt. Hewan.** 13(1):31-36
- Ibrahim, S. 2008. Hubungan ukuran-ukuran usus halus dengan berat badan broiler. **Agripet.** 8(2):42-46.
- Iji, P.A., R.J. Hughes, M. Choct, and D.R. Tivey. 2001. Intestinal structure and function of broiler chickens on wheat-based diets supplemented with microbial enzyme. **Asian-Aust. J. Anim. Sci.** 14(1):54-60.
- Khedr, N. and F. Abdel-Fattah. 2007. Response of broiler chickens to diet containing black seed (*Nigella sativa* L.) as medical plant. **Benha Vet. Med. J.** 17(2): 323-341
- Kruk, I., T. Michalska, K. Lichszeld, A. Kladna, and H.Y. Aboul-Enein. 2000. The effect of thymol and its derivatives on reactions on generating reactive oxygen species. **Chemosphere.** 41:1059-1064.
- Marshall, R.M. and S.B. Levy. 2011. Food animals and antimicrobials: Impacts on human health. **Clin. Microbiol. Rev.** 24(4):718-733.
- Mile, R.D., G.D. Butcher, P.R. Henry, and R.C. Littlel. 2006. Effect of antibiotic growth promoters on broiler performance, intestinal growth parameters, and quantitative morphology. **J. Poultry Sci.** 85:476-485.
- Miraghaee, S.S., B. Heidary, H. Almasi, A. Shabani, M. Elahi, and M.H.M. Nia. 2011. The effects of *Nigella sativa* powder (black seed) and *Echinacea purpurea* (L.) Moench extract on performance, some blood biochemical and hematological parameters in broiler chickens. **Afr. J. Biotechnol.** 10(82):19249-19254.
- Ningtias, A.S. 2013. Comparison of Growth Performance of Broilers, Kampong, and Backcross3 (*Gallus gallus domesticus* Linnaeus, 1758) Based on Morphometri and Histological Structure of Ileum and Breast Muscle. **Thesis.** Fakultas Biologi Universitas Gadjah Mada. Yogyakarta.
- Rahmawati. 2016. Histologis Saluran Pencernaan Ayam Buras Hasil In Ovo Feeding Asam Amino L-Arginine. **Thesis.** Fakultas Peternakan Universitas Hasanuddin. Makassar.
- Reynold, K.L., S.E. Cloft, and E.A. Wong. 2020. Changes with age in density of goblet cells in the small intestine of broiler chicks. **Poult. Sci.** 99(5):2342-2348.
- Rofiq, M.N. 2003. Pengaruh pakan berbahan baku lokal terhadap performans vili usus. **J. Sains dan Teknologi.** 5(5):190-194.
- Ruttanavut, J., K. Yamauchi, H. Goto, and T. Erikawa. 2009. Effects of dietary bamboo charcoal powder including vinegar liquid on growth performance and histological intestinal change in Aigamo ducks. **Int. J. Poult. Sci.** 8(3): 29-236.
- Salem, M.L. 2005. Immunomodulatory and therapeutic properties of the *Nigella sativa* L. seed. **Int. Immunopharmacol.** 5(13-14):1749-1770.
- Smirnov, A., R. Perez, E. Amit-Romach, D. Sklan, and Z. Uni. 2005. Mucin dynamics and microbial populations in chicken small intestine changed by dietary probiotic and antibiotic growth promotor supplementation. **J. Nutr.** 135:187-192.
- Sugito, W. Manalu, D.A. Astuti, E. Handharyani, and Chairul. 2007. Histopatologi hati dan ginjal pada ayam broiler yang dipapar cekaman panas dan diberi ekstrak kulit batang jalloh (*Salix tetrasperma* Roxb). **J. Ilmu Ternak dan Veteriner.** 12(1):68-72.
- Sulistiawati, F. and M. Radji. 2014. Potensi pemanfaatan *Nigella sativa* L. sebagai immunomodulator dan antiinflamasi. **Pharm. Sci. Res.** 1(2):65-77
- Sun, X., A. McElroy, Jr. Webb, A.E.K.E. Sefton, and C. Novak. 2005. Broiler performance and intestinal alterations when fed drug-free diets. **J. Poult. Sci.** 84:1294-1302.
- Suprijatna, E., U. Atmomarsono, and R. Kartasudjana. 2008. **Ilmu Dasar Ternak Unggas.** Penebar Swadaya, Jakarta.
- Tavakkoli, A., A. Ahmadi, B.M. Razavi, and H. Hosseinzadeh. 2017. Black seed (*Nigella sativa*) and its constituent thymoquinone as an antidote or a protective agent against natural or chemical toxicities. **Iran. J. Pharm. Res.** 16:2-23.
- Wang, J.X. and K.M. Peng. 2008. Molecular, cellular, and developmental biology developmental morphology of the small intestine of African ostrich chicks. **J. Poult. Sci.** 87:2629-2635.
- Wang, H., Y. Guo, and J.C.H.H. Shih. 2008. Effects of dietary supplementation of keratinase on growth performance, nitrogen retention and intestinal morphology of broiler chickens fed diets with soybean and cottonseed meals. **Anim. Feed. Sci. Tech.** 140 (3-4):376-384.