



RESEARCH PAPER

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Effect of different levels of probio-enzyme on head, brain, lung, testes, kidney, crop, proventriculus and pancreas characteristics of broilers

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Abstract

An experiment was conducted in order to investigate the impact of different levels of Probio-enzyme on some organ characteristics of broilers based on a completely randomized design as the factorial of 2×4 arrangement. Experiment included 8 treatment and also a control treatment including 270 broiler chickens. Experiment included 4 level of probio-enzyme amount (250, 500, 750, and 1000 g/ton) and also 2 usage duration (14, and 42 day). From obtained results, it is showed that usage duration of probio-enzyme had not significant effect on weight of pancreas ($P>0.05$), although usage duration of probio-enzyme as long as 42 days had the highest pancreas weight numerically (5.117 g). Probio-enzyme amount also had not significant on pancreas weight ($P>0.05$), however 1000 g/ton probio-enzyme resulted to the highest pancreas weight numerically (5.487 g). Meanwhile statistical differences between nine studied treatments for weight of pancreas were significant ($P\leq0.05$). Amount of weight of pancreas in nine studied treatments were between 4.233-7.013 g. Among studied treatments, the highest level of pancreas weight belonged to treatment 3 (usage duration as long as 14 days, and probio-enzyme level as amount as 750 g/ton), and treatment 9 (usage duration as long as 42 days, and probio-enzyme level as amount as 1000 g/ton) remained at lower level than other treatments. Other treatments were between these treatments.

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Introduction

Broiler performance is affected by dietary supplements. Probiotics and symbiotic have positive effects on broiler performance (Aziz Mousavi *et al.*, 2012; Dibaji *et al.*, 2012;). Enzymes also have positive effects on broiler performance (Sittiya *et al.*, 2014). There are not report about simultaneous effects of dietary probiotics and enzymes on organ characteristics of broiler.

There are little information The objective of this study was to determine the effect of duration and amount of probio-enzyme on some gastrointestinal organ characteristics of commercial broilers.

Materials and methods

A total of 270 day-old male Ross 308 broiler chicks were allocated to 27 land cages (100 cm× 100 cm× 60 cm; 9 groups x 3 replicates; 10 birds in each group) and fed 9 diets for 42 days during 2014. The mean birth body weights was similar between each cages (41.37 g). They were given free access to water and a feed throughout experiment period.

At placement time, ambient temperature was controlled at 33°C and decreased periodically to 23°C at 3 weeks of age and was maintained at 23°C until the termination of the investigation.

Humidity was added to the barn atmosphere via a water spray to maintain relative humidity between 55-65%.

Air circulation within the poultry barn was facilitated by 3 wall-mounted 60 cm diameter fans on one end of the barn and 160 cm diameter wall-mounted fans on the other end of the barn to establish tunnel ventilation.

Lighting was provided by 23 watt fluorescent tubes in ceiling fixtures. Constant light was provided on day 1, but on day 2, lighting was established at 21 hours per day until the end of the study.

A vaccination program against Infectious Bronchitis disease (1st, and 19th day of age; Infectious Bronchitis

Virus -H120; Razi Co, Karaj, Iran), IBD (14th, and 25th day of age; Gumboro IBD071IR; Razi Co, Karaj, Iran) and Newcastle disease virus (1st, 11st and 19th day of age; Newcastle lentogenic vaccine; strains Hitchner B1 and La Sota; Razi Co, Karaj, Iran) was practiced, in order to simulate a classical vaccination program.

All procedures have been approved by the Authors' Institution's Ethic Committee, and care was taken to minimize the number of animals used.

A three phase feeding program was used in this investigation and consisted of provision of starter feed from 1st to 10th day, grower feed from 11th to 24th day and finisher feed from 25th to 42nd day. The diets met or exceeded Ross 308 catalogue recommendations. All experimental diets were provided in mash form.

At the 42nd day of age, a bird of each replicate was scarified and head, brain, lung, testes, kidney, crop, proventriculus and pancreas weighted.

The experimental design, a 4×2 factorial arrangement, included 8 treatments with 3 replicates for each treatment. Four levels of probio-enzyme amount were as 250, 500, 750, and 1000 g/ton, and also 2 levels of usage duration were as 14, and 42 day. A control treatment (without dietary probio-enzyme) was also investigated.

Data were analyzed by analysis of variance using a 4×2 factorial, using a two-way ANOVA procedure (SPSS, 1997) and based on $Y_{ijk} = \mu + A_i + B_j + AB_{ij} + e_{ijk}$ formula. The Duncan post hoc test was used if the initial test result was significant at $P < 0.05$. Statements of significance were based on $P < 0.05$.

Results and discussion

Obtained results are summarized in Tables 1-3. From obtained results, it is showed that usage duration of probio-enzyme had not significant effect on weight of head ($P > 0.05$), although usage duration of probio-enzyme as long as 14 days had the highest head weight numerically (52.877 g). Probio-enzyme

amount also had not significant on head weight ($P>0.05$), however 750 g/ton probio-enzyme resulted to the highest head weight numerically (53.512 g). Meanwhile statistical differences between nine studied treatments for weight of head were not significant ($P>0.05$). Amount of weight of head in nine studied treatments were between 48.990-56.233 g. Among studied treatments, the highest level of

head weight belonged to treatment 2 (usage duration as long as 14 days, and probio-enzyme level as amount as 500 g/ton), and treatment 6 (usage duration as long as 42 days, and probio-enzyme level as amount as 500 g/ton) remained at lower level than other treatments. Other treatments were between these treatments.

Table 1. Mean (\pm SEM) of invaluable body parts at 42nd days of age in Ross 308 broilers affected the four different amounts of probio-enzyme and two different durations of its usage*.

Trait Treatment	Head weight (gr)	Relative weight of head (%)	Brain weight (gr)	Relative weight of brain (%)	Lungs (gr)	Relative weight of lungs (%)
Usage duration 14 (day)	52.877 ^a 51.187 ^a	2.814 ^a 2.773 ^a	2.698 ^a 2.738 ^a	0.144 ^a 0.148 ^a	10.559 ^a 10.520 ^a	0.560 ^a 0.570 ^a
P	0.333	0.589	0.684	0.423	0.923	0.674
SEM (Standard Error of Mean)	1.190	0.052	0.068	0.004	0.283	0.018
Probio-enzyme amount (g/ton)	250 500 750 1000	52.003 ^a 52.612 ^a 53.512 ^a 50.000 ^a	2.617 ^c 2.962 ^a 2.902 ^{ab} 2.693 ^{bc}	2.817 ^a 2.708 ^{ab} 2.895 ^a 2.453 ^b	0.142 ^{ab} 0.153 ^a 0.157 ^a 0.133 ^b	10.840 ^a 10.848 ^a 10.380 ^a 10.090 ^a
P	0.523	0.015	0.030	0.029	0.485	0.197
SEM (Standard Error of Mean)	1.684	0.074	0.096	0.005	0.400	0.025
Duration (14)- amount (250)	54.463 ^a	2.600 ^b	2.713 ^{abc}	0.129 ^b	12.350 ^a	0.590 ^{ab}
Duration (14)- amount (500)	56.233 ^a	3.152 ^a	2.667 ^{abc}	0.151 ^{ab}	10.313 ^{bcd}	0.580 ^{abc}
Duration (14)- amount (750)	51.567 ^a	2.724 ^b	2.990 ^a	0.158 ^a	10.963 ^{abc}	0.580 ^{abc}
Duration (14)- amount (1000)	49.243 ^a	2.781 ^{bc}	2.423 ^c	0.138 ^{ab}	8.610 ^d	0.488 ^{bc}
Duration (42)- amount (250)	49.543 ^a	2.635 ^b	2.920 ^{ab}	0.155 ^a	9.330 ^{cd}	0.498 ^{bc}
Duration (42)- amount (500)	48.990 ^a	2.772 ^{bc}	2.750 ^{abc}	0.156 ^a	11.383 ^{ab}	0.647 ^a
Duration (42)- amount (750)	55.457 ^a	3.081 ^{ab}	2.800 ^{abc}	0.155 ^a	9.797 ^{bed}	0.545 ^{abc}
Duration (42)- amount (1000)	50.757 ^a	2.605 ^b	2.483 ^{bc}	0.128 ^b	11.570 ^{ab}	0.592 ^{ab}
Control: Duration (0)- amount (0)	54.093 ^a	2.449 ^b	2.820 ^{abc}	0.128 ^b	10.533 ^{bc}	0.476 ^c
P	0.244	0.002	0.113	0.032	0.004	0.032
SEM (Standard Error of Mean)	2.337	0.100	0.131	0.008	0.540	0.033

* Means (\pm standard error) within each column of dietary treatments with no common superscript differ significantly at $P<0.05$.

It is showed that usage duration of probio-enzyme had not significant effect on weight of brain ($P>0.05$), although usage duration of probio-enzyme as long as 14 days had the highest brain weight numerically (2.814 g). Probio-enzyme amount had significant on brain weight ($P\leq0.05$), so 500 g/ton probio-enzyme resulted to the highest brain weight numerically (2.962 g). Meanwhile statistical differences between nine studied treatments for weight of brain were significant ($P\leq0.05$). Amount of weight of brain in nine studied treatments were between 2.449-3.152 g. Among studied treatments, the highest level of brain

weight belonged to treatment 2 (usage duration as long as 14 days, and probio-enzyme level as amount as 500 g/ton), and treatment 9 (control) remained at lower level than other treatments. Other treatments were between these treatments.

Usage duration of probio-enzyme had not significant effect on weight of lungs ($P>0.05$), although usage duration of probio-enzyme as long as 14 days had the highest lungs weight numerically (10.559 g). Probio-enzyme amount had not significant on lungs weight ($P>0.05$), however 250 and 500 g/ton probio-enzyme

resulted to the highest lungs weight numerically (10.840 g). Meanwhile statistical differences between nine studied treatments for weight of lungs were significant ($P \leq 0.05$). Amount of weight of lungs in nine studied treatments were between 8.610-12.350 g. Among studied treatments, the highest level of lungs weight belonged to treatment 1 (usage duration

as long as 14 days, and probio-enzyme level as amount as 250 g/ton), and treatment 4 (usage duration as long as 14 days, and probio-enzyme level as amount as 1000 g/ton) remained at lower level than other treatments. Other treatments were between these treatments.

Table 2. Mean (\pm SEM) of other invaluable body parts at 42nd days of age in Ross 308 broilers affected the four different amounts of probio-enzyme and two different durations of its usage*.

Trait	Testes weight (gr)	Relative weight of testes (%)	Kidneys weight (gr)	Relative weight of kidneys (%)	Back thoracic vertebrae (notarium) weight (gr)	Relative weight of thoracic vertebrae (notarium) (%)
Treatment						
Usage duration 14 (day)	0.507 ^a	0.027 ^a	14.471 ^a	0.769 ^a	34.779 ^a	1.851 ^a
42	0.562 ^a	0.030 ^a	13.861 ^a	0.751 ^a	32.260 ^a	1.746 ^a
P	0.552	0.502	0.420	0.692	0.302	0.403
SEM (Standard Error of Mean)	0.064	0.003	0.519	0.033	1.662	0.086
Probio-enzyme amount (g/ton)	250	0.612 ^a	0.031 ^a	15.762 ^a	0.795 ^a	35.248 ^a
500	0.480 ^a	0.027 ^a	13.408 ^b	0.756 ^a	33.577 ^a	1.897 ^a
750	0.600 ^a	0.032 ^a	13.253 ^b	0.719 ^a	31.665 ^a	1.708 ^a
1000	0.448 ^a	0.024 ^a	14.240 ^{ab}	0.769 ^a	33.588 ^a	1.822 ^a
P	0.493	0.634	0.107	0.706	0.763	0.734
SEM (Standard Error of Mean)	0.090	0.005	0.734	0.046	2.351	0.122
Duration (14)- amount (250)	0.650 ^a	0.031 ^a	16.613 ^a	0.793 ^a	36.363 ^a	1.717 ^a
Duration (14)- amount (500)	0.417 ^a	0.026 ^a	14.247 ^{ab}	0.799 ^a	31.213 ^a	1.824 ^a
Duration (14)- amount (750)	0.550 ^a	0.029 ^a	13.930 ^{ab}	0.738 ^a	34.767 ^a	1.839 ^a
Duration (14)- amount (1000)	0.413 ^a	0.024 ^a	13.093 ^{ab}	0.747 ^a	35.130 ^a	1.995 ^a
Duration (42)- amount (250)	0.573 ^a	0.030 ^a	14.910 ^{ab}	0.798 ^a	34.133 ^a	1.820 ^a
Duration (42)- amount (500)	0.543 ^a	0.031 ^a	12.570 ^b	0.714 ^a	34.297 ^a	1.940 ^a
Duration (42)- amount (750)	0.650 ^a	0.036 ^a	12.577 ^b	0.700 ^a	28.563 ^a	1.577 ^a
Duration (42)- amount (1000)	0.483 ^a	0.025 ^a	15.387 ^{ab}	0.790 ^a	32.047 ^a	1.648 ^a
Control: Duration (0)- amount (0)	0.623 ^a	0.029 ^a	14.900 ^{ab}	0.671 ^a	39.047 ^a	1.774 ^a
P	0.904	0.983	0.223	0.796	0.598	0.761
SEM (Standard Error of Mean)	0.144	0.008	1.100	0.064	3.299	0.170

From obtained results, it is showed that usage duration of probio-enzyme had not significant effect on weight of testes ($P > 0.05$), although usage duration of probio-enzyme as long as 42 days had the highest testes weight numerically (0.562 g). Probio-enzyme amount also had not significant on testes weight ($P > 0.05$), however 250 g/ton probio-enzyme resulted to the highest testes weight numerically (0.612 g). Meanwhile statistical differences between nine studied treatments for weight of testes were not

significant ($P > 0.05$). Amount of weight of testes in nine studied treatments were between 0.413-0.650 g. Among studied treatments, the highest level of testes weight belonged to treatments 1 and 7 (usage duration as long as 14 and 42 days, and probio-enzyme level as amount as 250 and 750 g/ton respectively), and treatment 4 (usage duration as long as 14 days, and probio-enzyme level as amount as 1000 g/ton) remained at lower level than other

treatments. Other treatments were between these treatments.

It is showed that usage duration of probio-enzyme had not significant effect on weight of kidneys ($P>0.05$), although usage duration of probio-enzyme as long as 14 days had the highest kidneys weight numerically (14.471 g). Probio-enzyme amount had significant on kidneys weight ($P\le0.05$), so 250 g/ton probio-enzyme resulted to the highest kidneys weight numerically (15.762 g). Meanwhile statistical

differences between nine studied treatments for weight of kidneys were significant ($P\le0.05$). Amount of weight of kidneys in nine studied treatments were between 12.570-16.613 g. Among studied treatments, the highest level of kidneys weight belonged to treatment 1 (control), and treatment 6 (usage duration as long as 42 days, and probio-enzyme level as amount as 500 g/ton) remained at lower level than other treatments. Other treatments were between these treatments.

Table 3. Mean (\pm SEM) of cranial gut segments at 42nd days of age in Ross 308 broilers affected the four different amounts of probio-enzyme and two different durations of its usage*.

Trait	Crop weight (gr)	Relative weight of crop (%)	Proventriculus weight (gr)	Relative weight of proventriculus (%)	Pancreas weight (gr)	Relative weight of pancreas (%)
Treatment						
Usage duration (day)	14	7.767 ^a	0.409 ^a	9.230 ^a	0.490 ^a	4.899 ^a
	42	7.135 ^a	0.385 ^a	9.617 ^a	0.519 ^a	5.117 ^a
P		0.547	0.662	0.559	0.397	0.461
SEM (Standard Error of Mean)		0.746	0.039	0.457	0.024	0.204
Probio-enzyme amount (g/ton)	250	8.760 ^a	0.433 ^a	9.913 ^a	0.496 ^a	4.982 ^a
	500	6.163 ^a	0.353 ^a	8.977 ^a	0.506 ^a	4.745 ^a
	750	6.410 ^a	0.347 ^a	8.497 ^a	0.461 ^a	4.820 ^a
	1000	8.470 ^a	0.456 ^a	10.307 ^a	0.554 ^a	5.487 ^a
P		0.207	0.380	0.221	0.315	0.301
SEM (Standard Error of Mean)		0.986	0.051	0.646	0.034	0.288
Duration (14)- amount (250)		11.077 ^b	0.523 ^{ab}	10.590 ^{ab}	0.499 ^a	4.677 ^b
Duration (14)- amount (500)		6.641 ^{cd}	0.383 ^b	9.231 ^b	0.535 ^a	5.117 ^b
Duration (14)- amount (750)		6.623 ^{cd}	0.348 ^b	8.230 ^b	0.436 ^a	4.233 ^b
Duration (14)- amount (1000)		6.660 ^{cd}	0.376 ^b	8.837 ^b	0.503 ^a	5.570 ^b
Duration (42)- amount (250)		6.443 ^{cd}	0.342 ^b	9.237 ^b	0.493 ^a	5.287 ^b
Duration (42)- amount (500)		5.620 ^d	0.317 ^b	8.690 ^b	0.491 ^a	4.373 ^b
Duration (42)- amount (750)		6.197 ^{cd}	0.346 ^b	8.763 ^b	0.487 ^a	5.407 ^b
Duration (42)- amount (1000)		10.280 ^{bc}	0.536 ^{ab}	11.777 ^{ab}	0.606 ^a	5.403 ^b
Control: Duration (0)- amount (0)		15.257 ^a	0.694 ^a	12.730 ^a	0.572 ^a	7.013 ^a
P		0.001	0.023	0.085	0.438	0.007
SEM (Standard Error of Mean)		1.311	0.070	10.038	0.049	0.401
						0.024

Usage duration of probio-enzyme had not significant effect on weight of back thoracic vertebrae ($P>0.05$), although usage duration of probio-enzyme as long as 14 days had the highest back thoracic vertebrae weight numerically (34.779 g). Probio-enzyme amount also had not significant on back thoracic vertebrae weight ($P>0.05$), however 250 g/ton probio-enzyme resulted to the highest back thoracic vertebrae weight numerically (35.248 g). Meanwhile statistical differences between nine studied

treatments for weight of back thoracic vertebrae were not significant ($P>0.05$). Amount of weight of back thoracic vertebrae in nine studied treatments were between 28.653-39.047 g. Among studied treatments, the highest level of back thoracic vertebrae weight belonged to treatment 9 (usage duration as long as 42 days, and probio-enzyme level as amount as 1000 g/ton), and treatment 7 (usage duration as long as 42 days, and probio-enzyme level as amount as 750 g/ton) remained at lower level than other treatments.

Other treatments were between these treatments.

From obtained results, it is showed that usage duration of probio-enzyme had not significant effect on weight of crop ($P>0.05$), although usage duration of probio-enzyme as long as 14 days had the highest crop weight numerically (7.767 g). Probio-enzyme amount also had not significant on crop weight ($P>0.05$), however 250 g/ton probio-enzyme resulted to the highest crop weight numerically (8.760 g). Meanwhile statistical differences between nine studied treatments for weight of crop were significant ($P\leq0.05$). Amount of weight of crop in nine studied treatments were between 5.620-11.077 g. Among studied treatments, the highest level of crop weight belonged to treatment 1 (usage duration as long as 14 days, and probio-enzyme level as amount as 250 g/ton), and treatment 6 (usage duration as long as 42 days, and probio-enzyme level as amount as 500 g/ton) remained at lower level than other treatments. Other treatments were between these treatments.

Usage duration of probio-enzyme had not significant effect on weight of proventriculus ($P>0.05$), although usage duration of probio-enzyme as long as 14 days had the highest proventriculus weight numerically (9.230 g). Probio-enzyme amount also had not significant on proventriculus weight ($P>0.05$), however 1000 g/ton probio-enzyme resulted to the highest proventriculus weight numerically (10.307 g). Meanwhile statistical differences between nine studied treatments for weight of proventriculus were significant ($P\leq0.05$). Amount of weight of proventriculus in nine studied treatments were between 8.230-12.730 g. Among studied treatments, the highest level of proventriculus weight belonged to treatment 3 (usage duration as long as 14 days, and probio-enzyme level as amount as 750 g/ton), and treatment 9 (usage duration as long as 42 days, and probio-enzyme level as amount as 1000 g/ton) remained at lower level than other treatments. Other treatments were between these treatments.

It is showed that usage duration of probio-enzyme had not significant effect on weight of pancreas

($P>0.05$), although usage duration of probio-enzyme as long as 42 days had the highest pancreas weight numerically (5.117 g). Probio-enzyme amount also had not significant on pancreas weight ($P>0.05$), however 1000 g/ton probio-enzyme resulted to the highest pancreas weight numerically (5.487 g). Meanwhile statistical differences between nine studied treatments for weight of pancreas were significant ($P\leq0.05$). Amount of weight of pancreas in nine studied treatments were between 4.233-7.013 g. Among studied treatments, the highest level of pancreas weight belonged to treatment 3 (usage duration as long as 14 days, and probio-enzyme level as amount as 750 g/ton), and treatment 9 (usage duration as long as 42 days, and probio-enzyme level as amount as 1000 g/ton) remained at lower level than other treatments. Other treatments were between these treatments.

There are reports on positive effects of probiotics and synbiotics on broiler (Dibaji *et al.*, 2014; Karimi Kivi *et al.*, 2014). Meanwhile there are reports on positive effects of enzymes on broiler (Sateri *et al.*, 2014). However there are not enough reports about simultaneous effects of probiotics and enzymes (probio-enzyme) on broiler. So, findings of this study is novel and demonstrated positive effects of dietary probio-enzyme on organ characteristics of broiler.

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