Advances in Bioresearch
Volume 3 [4] December 2012: 17- 21
© 2012 Society of Education, India
[ISO 9001: 2008 Certified Organization]
Website: http://www.soeagra.com/abr/abr.htm

A B R
ISSN 0976-4585

Original Article

Effect of Canola oil on Mucosal Amylase and Lipase Activity Enzymes in Small Intestine of Turkey Chicks

Jamshid Ghiasi Ghaleh-kandi[1*], Manish Kumar[2], Yahya Ebrahimnezhad [1], Rahim Beheshti [3]
1- Department of Animal Science, Faculty of Agriculture, Shabestar Branch, Islamic Azad University, Shabestar, East Azerbaijan, 5381637181, Iran
2 Academy for Environment and Life Sciences, Agra, (U.P.) INDIA
3- Department of Veterinary Science, Islamic Azad University, Shabestar Branch, 53815-159 Shabestar, Iran
*** Corresponding author**: ghiasi_jam@yahoo.com

ABSTRACT
Canola is one of the rapeseed varieties and is in temperate and cold climate areas. Contains 94 % unsaturated fatty acid and 6 % saturated fatty acid, thus, has best fatty acid composition among other oils. Canola oil causes alteration in pancreatic enzymes such as amylase and lipase activity. The aim of this study was assessment of mucosal amylase and lipase activity enzymes subsequently using of canola oil on turkey chicks diet. In this study 135 Iranian native turkey chicks were allocated into 3 treatments and 3 replications of 15 chicks and were fed in separate cage with 0, 2.5 and 5 % of canola oil. The end of 20th week after 5 hour fasting 2 chicks from each replicate were selected and slaughtered and sampling were done from parts of 1, 10, 30, 50, 70 and 90 % of small intestine. In lab, the activity of amylase were measured and recorded. According to this survey results revealed that using of canola oil in turkey chicks diet causes increasing of lipase activity (in 5 % treatment than control and 2.5 % treatments) and decreasing of amylase activity (in 2.5 % treatment than control and 5% treatments). Therefore, using of different amounts of canola oil in turkey chicks diet causes increasing of lipase and reducing of amylase activity.
***Key words**: Canola oil, Amylase, Lipase and Calorimetric.*

INTRODUCTION

Canola is one of the rapeseed varieties and is in temperate and cold climate areas. Canola is rich of sulfurous amino acids and vitamins. Canola meal has about 40% protein. Its erucic acid is less than 2% of total fatty acids [1]. Contains less than 30 micromole glucosinolate per oil free dry matter. Contains 94% unsaturated fatty acid and 6% saturated fatty acid, thus, has best fatty acid composition among other oils. Because of having 61% of oleic acid, considered as full resources of unsaturated fatty acids and from this aspect, occupied in second class after olive oil. Its tocopherol is higher than olive and soya oils, that from this aspect can be attribute high antioxidant effect to it. Canola oil combination is inserted in table 1 [1,2].

Lipase is enzyme which is able to hydrolyzing of esters and plays an important role in converting of triglycerides to glycerol and fatty acids that called lipolysis. Lipases are of esterase subclass. Lipase is type of glycoprotein which found in most fauna bodies (such as most viruses). These water-soluble enzymes have roles in triglycerides metabolism and causes of their digestion in body. Pancreatic lipase is enzyme which able to converting of fats to small and digestible molecules. Thus, triglycerides convert to monoglycerides and fatty acids. Other type of lipase include: Phospholipase and sphingomyelinase. Lipases, like other hydrolyzing enzymes are susceptible to temperature, environment pH, moisture and polarity of solvent. Amylase considered as hydrolyzing enzymes that emprises starch breaking. Amylase exists on microbial, Herbal and animal structures [3]. Amylase term is attributing to enzymes that are able to hydrolyzing of α-1,4 bonds, these bonds are found in amylose, amylopectin, glycogen and these products. Amylase hydrolyses bonds which are adjacent with glucose units. Amylase is produce in exocrine pancreas and parotid glands and its action is hydrolyzing of starch and converting to maltose. There are two types of amylase. β-amylase or bacterial amylase which is hydrolyzed polysaccharides chains (starch and glycogen) from reducer end and in each time produces one maltose molecule. α-

amylase hydrolyzing action is not regular and act on different parts of chain. This research was done to survey of canola oil effects on lipase and amylase activity [4].

Table 1: fatty acids contents of canola oil

Fatty acid	Percent
C12:0	0
C14:0	0
C16:0	4,73
C16:1	0,13
C18:0	2,31
C18:1cis9	61,1
C18:1cis11	0
C18:1trans9	0
C18:2cis	19,73
C18:2trans	1,78
C18:3cis	7,35
C18:3trans	0,71
C20:0	0,53
C20:1	1,18
C20:4	0
C22:0	0,25
C22:1	0,21

MATERIALS AND METHODS

Animals and Diet

This research was performed one 135 Iranian native turkey chicks (from 4th to 20th week of age). In this study, the turkey chicks by chance divided into 3 treatments and each treatment divided into 3 replicates and each replicate was contained 15 turkey chicks and were fed in separate cage with 0, 2.5 and 5 percent of canola oil. The experimental diets formulated isonitrogenouse and isoenergetic and balanced according to 1994 national research council [5]. The birds were given access to water and diets ad-libitum. The composition and calculated nutrient composition of the mixture of treatment is shown in Table 2.

Table 2: Percentage composition of experimental diets in four periods

	4 -8 week			8-12 week			12-16 week			16-20 week		
	T_1	T_2	T_3	T_1	T_2	T_3	T_1	T_2	T_3	T_1	T_2	T_3
Corn	42.50	38.00	36.00	45.60	43.00	35.00	56.64	48.50	40.00	64.41	58.00	48.00
SBM	34.40	36.00	31.15	28.25	27.30	28.24	26.00	27.00	27.50	21.00	21.00	21.00
Oil	0.00	1.25	2.50	0.00	2.50	5.00	0.00	2.50	5.00	0.00	2.50	5.00
Fish	4.80	3.70	6.60	8.00	8.00	8.00	2.64	1.82	1.50	0.65	0.70	0.67
Starch	3.10	3.22	1.56	7.46	3.32	3.37	6.57	6.51	6.50	7.10	5.56	6.71
Alfalfa	3.47	5.00	6.00	3.00	5.00	6.00	1.50	4.00	6.00	1.00	3.80	6.00
DCP	1.38	1.52	1.11	0.63	0.61	0.62	1.03	1.15	1.18	1.17	1.15	1.15
Met	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Lys	1.50	1.50	1.50	1.50	1.50	1.50	1.40	1.50	1.50	1.50	1.50	1.50
Oyster	1.02	1.02	0.86	0.73	0.67	0.62	0.92	0.87	0.82	0.90	0.81	0.73
wheat bran	2.00	3.00	6.00	2.50	5.00	6.00	1.00	3.00	6.00	0.00	1.70	5.00
Vit supp1	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Min supp2	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Salt	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Sand	3.58	3.54	4.47	0.08	0.85	3.40	0.05	0.90	1.75	0.02	1.03	1.99
	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
Calculated nutrient content												
ME kcal/kg	2755	2755	2755	2850	2850	2850	2945	2945	2945	3040	3040	3040
Crude protein (%)	24.7	24.7	24.7	20.9	20.9	20.9	18.1	18.2	18.1	15.7	15.7	15.7
Calcium (%)	0.95	0.95	0.95	0.81	0.81	0.81	0.71	0.71	0.71	0.62	0.62	0.62
Available P (%)	0.48	0.48	0.48	0.40	0.40	0.40	0.36	0.36	0.36	0.31	0.31	0.31
ME/CP	112	112	112	136	136	136	163	162	163	194	194	194
Ca/P	2	2	2	2	2	2	2	2	2	2	2	2

1Vitamin content of diets provided per kilogram of diet: vitamin A,D, E and K.

2 Composition of mineral premix provided as follows per kilogram of premix: Mn, 120,000mg; Zn, 80,000 mg; Fe, 90,000 mg; Cu, 15,000 mg; I, 1,600
mg; Se, 500 mg; Co, 600 mg

Sample collection

In the Rearing period, all conditions such as temperature, humidity, light, ventilation and management were appropriate and similar for all broilers and 20th week of age of end the rearing period, after 5 hours of starvation, 2 broilers from every group (totally 18 chickens of sampling) which weighed nearly equal to the average weight of each replicate have been chosen and slaughtered. The abdominal cavity was opened, and the entire gastrointestinal tract was removed. The small intestine was isolated, and the length of intestine was determined by a graduate ruler. The positions at 1, 10, 30, 50, 70 and 90 % of the length of small intestine for analyzing the Enzymes activity were separated with specific scissors (an 8-cm sample was taken). The samples for enzymes determination were cut open lengthwise, rinsed carefully with phosphate buffer saline (pH=7), blotted dry, then samples envelop in vacuum packed and stored at −80°C until enzymes analysis [6].

Enzyme assay

After thawing, all of vacuum packed were opened and then using a sensitive scale, 0.05 gram of the mucosal small intestine was weighed and along with 10 ml liter phosphate buffer saline (pH=7) was formed into a homogenized solution using sonic Vibracell Sonics (VCX 130 TE USA) device [6]. Enzymes activity of Lipase and Amylase were measured according to the procedure (calorimetric method). For detection of enzymes activity it was needed to measure total protein which Pirogallol (calorimetric) method was used [7]. The level of activity of enzymes of each sample is divided into the amount of its total protein so the activity level of the enzyme is calculated according to the IU in liter/gram protein [6].

Statistical analyses

The results of the research have been statistically analyzed using the linear model of SAS software [8].

Analysis of variance according to the model,

$$x_{ij} = \mu + T_j + e_{ij}$$

Where,

x_{ij} = All dependent variable

μ = Overall mean

Ti = The fixed effect of RRO levels ($_i$ = 1, 2, 3)

e_{ij} = The effect of experimental error

Values of different parameters were expressed as the mean ± standard deviation (X±SD). When significant difference among means was found, means were separated using Duncan's multiple range tests.

RESULT

According to table 3, adding different levels of canola oil to turkey chick's diet have different effects on amylase activity on several regions of small intestine. ,amylase activity in part of 1% of small intestine in 2.5% treatment has significant increasing than control treatment, and in 2.5% treatment has significant increasing than 5% treatment, whereas, amylase activity in part of 50% of small intestine has significant increasing in 2.5% treatment than 5%, and 5% treatment has significant increasing than control treatment. There are significant increasing in parts of 30% and 70% of small intestine in control and 2.5% treatments than 5% treatment and in part of 10% of small intestine there is a significant increasing in 2.5% treatment than 5% ($p<0.05$).

According to table 4 adding different levels of canola oil to turkey chick's diet have different effects on lipase activity on several regions of small intestine. So that, lipase activity has significant increasing in parts of 10, 30, 50, 70 and 90 percent of small intestine in 5% treatment than control and 2.5% treatments and in part of 1% of small intestine has significant increasing in 5% treatment than control and 2.5% treatments

DISCUSSION

according to this survey results revealed that using of canola oil in turkey chicks diet causes increasing of lipase activity (in 5% treatment than control and 2.5% treatments) and decreasing of amylase activity (in 2.5% treatment than control and 5% treatments). In other research were done

by Xu et al., [9], demonstrated that adding 4g/kg fructooligosaccharide to broiler diets causes significant increasing on daily weight gain. Also seen that adding 2-4g/kg fructooligosaccharide to broiler diets causes improvement of amylase activity after 49 days of age in compared with control group. Also in that study described that lipolitic activity of intestine don't affect by diet treatments, these results not compatible with our results. In one study accomplished by Cau et al., [10], revealed that there aren't relationship among essential oils and digestive enzymes on material digestibility in ileum. In that study expressed that on day 21 of nurturing essential oils causes improvement of digestibility of dry matter and energy in ileum about 6.5 and 6.9 percent in compared with control group. On 24 days of age essential oils haven't effects on enzymes and volatile fatty acids of digested materials in ileum. On day 22 of research with measurement of volatile fatty acids observed that adding fats to diet haven't effect on this material concentration in Cecum. These results are not compatible with our results. In other study were done by Edem et al. [11], revealed that adding palm and corn oils to rats diet causes increasing of lipase activity that is compatible with our results. In other study were done by Jang et al., [12] observed that adding essential oils to broiler diets causes alteration in pancreatic enzymes activity. Thus, those chicks were fed with essential oils than those were fed with corn oil had increased α-amylase and maltase activity that is not compatible with our research results. In other research were done by Elisabeth et al., [13] revealed that using of ciglitazone causes alteration of amylase and lipase activity. Ciglitazone is one of the diabetic drugs that causes increasing in LDL and HDL and conversely causes reducing of triglycerides. In that study revealed that adding this drug to rats diet causes decreasing in amylase and increasing in lipase concentrations. In one study carried out by Kalpan et al., [14] Demonstrated that adding dietary spices to chicks diet causes increasing in lipase, maltase and disaccharides and also causes reduce in phosphates and Sucrase activity. In one study fulfilled by Ducasse-cabanot et al., [15] delineated that adding fish oil to rainbow trout diet causes reducing in alkaline phosphatase, amylase, aminopeptidase N and γ-glutamyl peptidase. Also observed with removing of fish oil from diet, lipase activity was decreased. In other research were done by Mir et al., [16], obtained that amylase activity in different regions of intestine (proximal, mid and distal regions) in those sheep was fed with control diet was 5.67±1.57, 6.74±1.38 and 2.40±1.46 Respectively, but In those sheep were fed with sunflower oil was 6.40±1.57, 6.40±1.57 and 1.71±1.31 Respectively. Thus, can be deducing that this enzyme activity in proximal and distal regions were not affected by added oil but in middle region were decreased. Therefore, using of different amounts of canola oil in turkey chicks diet causes increasing of lipase and reducing of amylase activity. The experimental diets formulated isonitrogenouse and isoenergetic. But when we added different levels of canola oil to diet, consequently, energy increased and carbohydrate decreased thus, amylase activity decreased and lipase activity increased.

Table 3: comparison of average Amylase activity between treatments in different periods and segments of small intestine in broiler chicks (IU/g protein)

Intestine length Groups	1 %	10%	30 %	50%	70 %	90%
0 % canola oil	$7632.9^{b}\pm1151.4$	$14754.3^{ab}\pm7642.3$	$15541.7^{a}\pm4672$	$5145^{c}\pm1186.9$	$12347^{a}\pm1660.3$	5703.8±919.9
2.5% canola oil	$9190.2^{a}\pm1687.7$	$18411.3^{a}\pm1194.7$	$14364.8^{a}\pm1065$	$13779.1^{a}\pm1558.6$	$12395.3^{a}\pm3781$	4520.3±854.5
5 % canola oil	$3956.4^{c}\pm267.9$	$9328.6^{b}\pm905.4$	$7613.5^{b}\pm341.8$	$11251^{b}\pm1344.7$	$6794.4^{b}\pm1119.4$	4024.6±992.6

a,b Means in the same column with different superscripts differ significantly X ± SD (P<0.05).

Table 4: comparison of average Lipase activity between treatments in different periods and segments of small intestine in broiler chicks (IU/g protein)

Intestine length Groups	1 %	10%	30 %	50%	70 %	90%
0 % canola oil	$1696.1^{c}\pm120.7$	$1412^{b}\pm200.6$	$1630.5^{b}\pm83.2$	$1561.1^{b}\pm166.4$	$2430.9^{b}\pm345.1$	$2491.9^{b}\pm438.3$
2.5%canola oil	$2612.4^{b}\pm441.5$	$2907^{b}\pm653.4$	$2463.1^{b}\pm497.1$	$2794.6^{b}\pm438.2$	$2337.5^{b}\pm414.2$	$2450.4^{b}\pm563.3$
5 % canola oil	$5396^{a}\pm396.2$	$8348.1^{a}\pm881.5$	$8143.2^{a}\pm1287.6$	$8210^{a}\pm1371.1$	$9151^{a}\pm1600.6$	$7415.8^{a}\pm647.5$

a,b Means in the same column with different superscripts differ significantly X ± SD (P<0.05).

CONCLUSIONS AND APPLICATIONS

According to this survey results revealed that using of canola oil in turkey chicks diet causes increasing of lipase activity (in 5% treatment than control and 2.5% treatments) and decreasing of amylase activity (in 2.5% treatment than control and 5% treatments). Therefore, using of different amounts of canola oil in turkey chicks diet causes increasing of lipase and reducing of amylase activity. The experimental diets formulated isonitrogenouse and isoenergetic. But when we added different levels of canola oil to diet, consequently, energy increased and carbohydrate decreased thus, amylase activity decreased and lipase activity increased. The results of this project are used poultry industry and medicine.

REFERENCES

1. Pajohan-Mehr, S., and R. Farahvash. 2008. Comparison of the chemical structure of common cultivars of canola oil (okapi, vanguard or Kobra) in Iran. 18th national congress on food technology (in Persian).
2. Mohammdi, T., M. H. Azizi, and A. Taslimi (2007). Relationship between fatty acid composition with the stability of sunflower oil and canola oil mixture. J. of Food Sci. and Tech. 4: 67-75 (in Persian).
3. Santamrina-Fojo, S., and H. Bryan-Brewer (1994). Lipoprotein lipase: structure, function and mechanism of action. Internat. J. of Clin. & Lab. Res. 24: 143-147.
4. Aiter, V (2005). Amylases and their applications. *Afri. J. of Biotech*. 4: 1525-1529.
5. NRC. 1990. Nutrient requirement of broiler. 8 th rev. ed. Natl. Aca. Press, Washington, DC.
6. Teshfam, M (1984). Comparison of the effects of the high-acid milk replacer with conventional skim milk replacer. Ph.D. Thesis, University of Bristol, UK.
7. Watanba, N., S. Kamal, A. Ohkubo, M. Yanamaka, S. Ohsawas, K. Maikno, K. Tokuda (1986). Method for Assaying Total Protein. Clin. Chem. 32: 1551-1554
8. SAS Institute, (2001). SAS state software: Changes and Enhancement through release 8.2. SAS institute, Inc; Cary, NC.
9. Cau, P. H., F. D. Li, Y. F. Li, Y. J. Ru, A. Peron, H. Schulze, and H. Bento (2010). Effect of Essential Oils and Feed Enzymes on Performance and Nutrient Utilization in Broilers Fed a Corn/Soy-based Diet. Poul. Sci. 9(8): 749-755.
10. Xu, Z. R., C. H. Hu, M. S. Xia, X. A. Zhan, and M. Q. Wang (2003). Effects of dietary Fructooligosaccharide on Digestive Enzyme Activities, Intestinal Microflora and Morphology of Male Broilers. Poul. Sci. 82:1030–1036.
11. Edem, D. O., and M. I. Akpanabiatu (2006). Effects of Palm Oil – Containing Diets on Enzyme Activities of Rats. Paki. J. of Nutr. 5(4): 301-305.
12. Jang, I.S., Y. H. Ko, S. Y. Kang, and C. Y. Lee (2007). Effect of a commercial essential oil on growth performance, digestive enzyme activity and intestinal microflora population in broiler chickens. Ani. Feed Sci. and Tech. 134: 304-315.
13. AOAC, (1995). Association Official Methods of Analysis, 16th ed . Washigton DC.
14. Ducasse-Cabanot, S., J. Zambonino-Infante, N. Richard, F. Medale, G. Corraze, M. Mambrini, J. Robin, C. Cahu, S. Kaushik, and S. Panserat (2007). Reduced lipid intake leads to changes in digestive enzymes in the intestine but has minor effects on key enzymes of hepatic intermediary metabolism in rainbow trout (Oncorhynchus mykiss). The Ani. Cons. 1: 1272–1282.
15. Jensena, M. S., K. E. Bachknudsena, J. Inborr, and K. Jakbsena (1998). Effect of β-glucanase supplementation on pancreatic enzyme activity and nutrient digestibility in piglets fed diets based on hulled and hulless barley varieties. Animal Feed Science and Technology, 72: 329-345.
16. Mir, P.S., M. Ivan, G. J. Mears, B. F. Benkel, C.M. Ross, S. D. Husar, and Z. Mir (2002). Effect of sunflower oil on sheep small intestinal digesta viscosity, composition and amylase activity. Small Rum. Res. 45: 33-38.

[Received: 12.08.12; Accepted: 08.11.2012]