

FATTY ACID CONTENT IN BROILER'S ROSS 308 MEAT MUSCLES AFTER USING BEE POLLEN AND PROBIOTIC AS SUPPLEMENTARY DIET INTO THEIR FEED MIXTURE

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ABSTRACT

The present experiment was aimed to study the effect of the bee pollen and probiotic on broiler's meat fatty acid. A total of 120 one day old chicks, which were divided into 4 groups (n=30). Central, E1 (400 mg.kg⁻¹ bee pollen), E2 (3.3 g probiotic) and E3 (400 mg.kg⁻¹ bee pollen + 3.3 g probiotic) of complete feed mixture, the probiotic has been added through drinking water. After 42 days broiler has been slaughtered to determine meat fatty acid concentrations. We found that the probiotic was increased about (78.9%) of the essential fatty acid and bee pollen was increased about (68.4%) of essential fatty acid in the experimental groups and there were found significant differences (P≤0.05) in monounsaturated acid (MUFA), heptadecanoic acid and omega-6 between control and E1 groups. The mixed between bee pollen and probiotic were reduce the content of the essential fatty acid. However, bee pollen and probiotic were decreased nonessential fatty acid in broiler meat muscles and they were found significant difference (P≤0.05) in saturated fatty acid (SFA) between control and E1, also between control and myristic acid. It was concluded that the bee pollen and probiotic have increased the fatty acid and decrease the non essential fatty acid in broiler meat muscles.

Keywords: Broiler Ross 308, fatty acid, bee pollen, probiotic, monounsaturated

INTRODUCTION

For good humans health and other animals must ingest essential fatty acid, because their body can't synthesize fatty acid (Robert *et al.*, 1980). To support human health, meat should be processed from the meat content high amount of MUFs, by feeding animal with a feed mixture rich of fatty acid (Aronal *et al.*, 2012). Fatty acid are important sources of the body fuel because, when metabolized they yield large quantities of ATP and many cell types can use either glucose or fatty acid for this purpose in particular heart and skeletal muscle prefer fatty acid, despite longstanding assertions to the contrary, the brain can use fatty acid as a source of the fuel in addition to glucose and ketone bodies (Robert *et al.*, 1980; Ebert *et al.*, 2003; Isaac *et al.*, 2012). It has been observed that the consumption of chickens of diets with different fatty acid composition not only changes the degree of saturation, but also modifies the amount of fat deposited in chicken tissues. In particular the intake of the polyunsaturated fatty acid (PUFA) compared to the intake of saturated fatty acid (SFA) causes a lower fat deposition in the animal (Villaverde *et al.*, 2005).

Bee pollen is rich of the a major fatty acid, presented as mean values were C18:3 (25.1%), C16:0 (19.6%), C18:1 (17.3%), C18:2 (8.78%), C22:0 (4.07%) and C18:0 (2.96%) acids. The proportions of C18:3 were generally higher than those of C18:2 and the ratio of total unsaturated fatty acid (TUS) to total saturated fatty acid (TS) was >1.0, except for *Nelumbo nucifera* Gaertn, pollen for the characteristic absence of C18:3 acids (Yang *et al.*, 2013). Probiotics are microorganisms that have claimed health benefits when consumed (Ringø *et al.*, 1998). Probiotic increases the (PUA) fatty acid (Bomba *et al.*, 2002). Despite a substantial amount of the basic and clinical research on the beneficial effects of probiotics all of the evaluated claim applications thus far have received a negative opinion. With the restrictions on the use of the clinical endpoints, validated biomarkers for gut health and immune health in relation to reduction in disease risk are needed. Clear-cut criteria for design as well as evaluation of the future studies are needed. An open dialogue between basic and clinical scientists, regulatory authorities, food and nutrition industry, and consumers could bridge the gap between science and marketing of probiotics (Rijkers *et al.*, 2011).

The study was objected to study the effect of addition the bee pollen into feed mixture and probiotic through drinking water on the broiler's meat essential fatty acid.

MATERIAL AND METHODS

The experiment was implemented at the test poultry station of Slovak University of Agriculture in Nitra. The tested chickens were broiler Ross 308. The experiment included 120 chicks in one day-old, which were divided into 4 groups (n=30): control, E1 (400 mg.kg⁻¹ natural bee pollen), E2 (3.3 g probiotic) E3 (400 mg.kg⁻¹ natural bee pollen + 3.3 g probiotic) the probiotic has been added through drinking water. In all the investigated groups the chickens were fed by same starter complete feed mixture (CFM) HYD-01 (loose structure) from 1st day to 21st days of their age and grower from the 22nd to 42nd days of their age (Table 1).

The chickens were bred in cage conditions. Each cage was equipped with feed dispenser and water intake was ensured ad libitum through a self feed-pump. The temperature was controlled during the fattening period and it was 33 °C at the first day and every week was reduced about 2 °C and final temperature was 19 °C. The lighting during the experimental period was continuous. The experiment duration was 42 days.

At the end of the fattening period from each experimental group were selected 20 pieces of chickens for slaughter analysis (10 ♀ pieces and 10 pieces ♂), to evaluate the count of fatty acid of the broiler's meat muscles. The experimental analyses in Animal Production Research Centre Nitra (APRC Nitra) by using Agilent 7890A Gas Chromatograph (USA).

Statistical analysis

The results of meat performance (arithmetic mean, standard deviation) were statistically analysed by the statistic program Statgraphics Plus version 5.1 (AV Trading Umex, Dresden, Germany). For the determination of significant differences among the tested groups was used analysis of variance.

Table 1 Composition of the broiler feed mixture

Ingredients (%)	Starter	Grower
	(1 to 21 days of age)	(22 to 42 days of age)
Wheat	35.00	35.00
Maize	35.00	40.00
Soybean meal (48 % N)	21.30	18.70
Fish meal (71 % N)	3.80	2.00
Dried blood	1.25	1.25
Ground limestone	1.00	1.05
Monocalcium phosphate	1.00	0.70
Fodder salt	0.10	0.15
Sodium bicarbonate	0.15	0.20
Sodium bicarbonate	0.15	0.20
Lysine	0.05	0.07
Methionine	0.15	0.22
Palm kernel oil Bergafat	0.70	0.16
¹ Premix Euromix BR 0,5 %	0.50	0.50
Analysed composition (g.kg⁻¹)		
Crude protein	210.76	190.42
Fibre	30.19	29.93
Ash	24.24	19.94
Ca	8.16	7.28
P	6.76	5.71
Mg	1.41	1.36
ME (MJ.kg ⁻¹)	12.02	12.03

¹active substances per kilogram of premix: vitamin A 2 500 000 IU; vitamin E 50 000 mg; vitamin D3 800 000 IU; niacin 12 000 mg; d-pantothenic acid 3 000 mg; riboflavin 1 800 mg; pyridoxine 1200 mg; thiamine 600 mg; menadione 800 mg; ascorbic acid 50000 mg; folic acid 400 mg; biotin 40 mg; vitamin B12 10.0 mg; choline 100000 mg; betaine 50000 mg; Mn 20 000 mg; Zn 16 000 mg; Fe 14 000 mg; Cu 2 400 mg; Co 80 mg; I 200 mg; Se 50 mg.

RESULTS

In humans the fatty acid is a principle energy substrate and structural components of cell membranes (phospholipids) and second messengers, fatty acid are also ligands of nuclear receptors affecting gene expression (Kremmyda et al., 2011). However, table (2) shows the fatty acid content of the broiler's meat muscles after were used bee pollen as a supplemental diet into broiler's feed mixture and probiotic were added through drinking water. It was found that the palmitoleic acid, oleic acid, monounsaturated (MUFAs) and omega-3 were higher in the experimental groups compared to control group and they found significant differences (P<0.05) in monounsaturated (MUFAs) between control and E1 groups. Also they were found that the myristoleic acid, arachidonic acid, heptadecanoic acid, nervonic acid, cis-11,14-eicosadienoic acid, cis-8,11,14-eicosatrienoic acid and cis-5,8,11,14,17 eicosapentaenoic in the experimental groups were higher than control group except E3 it was lower than the control group and they were found significant differences (P<0.05) in myristoleic acid between E1 and E3 groups, also in heptadecanoic acid there were found significant differences (P<0.05) between E1 and control, E2 and E3 groups. On the other hand, in elaidic acid they were found that the experimental E1 was higher than the control, but E2, E3 groups it were a lower than the control group and there were found significant differences (P<0.05) between E1 and E2 groups. However, the linoleic acid and alpha-linolenic acid were higher in control than E1, E2 and E3 groups. Further, the polyunsaturated (PUFAs) were higher in E2 group compared to control, except E1, E3 it were a lower than control. The linoelaidic acid and pentadecanoic acid were higher in the E1, E3 groups compared to the control group except the E1 group. Moreover, the cis-11-eicosenoic acid was higher in the E1 group than the control group, but E2 and E3 were a lower compared to control group. Otherwise, the omega-6 was higher in E2 that control and experimental E1, E3 were a lower than the control group. Table (3) shows the effect of the bee pollen and probiotic on broiler's meat saturated fatty acid, where were found that the myristic acid, palmitic acid, stearic acid and saturated fatty acid (SFA) in the control group were higher compared to experimental groups and it were found significant differences (P<0.05) in myristic acid between E3 and control and E1, also in saturated fatty acid (SFA) were found significant differences (P<0.05) between control and E1 group.

Table 2 The effect of bee pollen (400 mg.kg⁻¹) and probiotic (3.3 g) on broiler Ross 308 unsaturated fatty acids (%)

Indicators	Control	E1 pollen	E2 probiotic	E3 pollen+probiotic
Myristoleic acid	0.211±0.18 ^{ab}	0.225±0.20 ^a	0.230±0.03 ^{ab}	0.209±0.01 ^b
Palmitoleic acid	7.011±0.63	7.317±0.73	7.452±0.81	7.291±0.37
Oleic acid	40.733±0.88	41.239±0.8	40.791±1.50	41.356±0.92
Elaidic acid	0.119±0.05 ^{ab}	0.146±0.06 ^a	0.984±0.05 ^b	0.113±0.01 ^{ab}
Linoleic acid	11.491±0.53	11.148±0.58	11.385±1.01	11.077±0.82
Linoelaidic acid	0.121±0.03	0.106±0.05	0.134±0.01	0.135±0.02
α-Linolenic acid	0.753±0.02	0.743±0.044	0.730±0.07	0.737±0.05
Arachidonic acid	0.372±0.20	0.377±0.18	0.416±0.21	0.308±0.11
Pentadecanoic acid	0.087±0.04	0.052±0.05	0.088±0.03	0.839±0.04
Heptadecanoic acid	0.105±0.04 ^a	0.061±0.05 ^b	0.109±0.01 ^a	0.101±0.02 ^a
Nervonic acid	0.317±0.17	0.367±0.19	0.350±0.18	0.270±0.10
cis-11-eicosenoic acid	0.454±0.04	0.483±0.07	0.446±0.04	0.447±0.03
Cis-11,14-Eicosadienoic acid	0.208±0.07	0.228±0.08	0.222±0.07	0.193±0.05
Cis-8,11,14-eicosatrienoic	0.160±0.06	0.166±0.09	0.181±0.07	0.152±0.05
Cis-5,8,11,14,17 Eicosapentaenoic	0.146±0.04	0.160±0.03	0.162±0.04	0.145±0.02
Polyunsaturated (PUFAs)	13.252±0.75	12.924±0.71	13.320±1.18	12.749±0.97
Monounsaturated (MUFAs)	48.845±1.02 ^a	49.776 ±0.04 ^b	49.357±1.47 ^{ab}	49.689±1.05 ^{ab}
Omega-3	0.072±0.001	0.075±0.002	0.073±0.002	0.074±0.002
Omega-6	13.736±0.31 ^a	13.398±0.36 ^b	13.822±0.48 ^{ab}	13.427±0.43 ^{ab}

E1, E2, E3: experimental groups; ^{a,b} – means with different superscripts differ significantly; (P ≤ 0.05) significant

Table 3 The effect of bee pollen (400 mg.kg⁻¹) and probiotic (3.3 g) on the broiler Ross 308 saturated fatty acid (%)

Indicators	Control	E1 pollen	E2 probiotic	E3 pollen+probiotic
Myristic acid	0.694±0.02 ^a	0.684±0.03 ^a	0.663±0.05 ^{ab}	0.641±0.01 ^b
Palmitic acid	26.887±0.51	26.610±0.47	26.633±0.80	26.902±0.47
Stearic acid	6.394±0.53	6.046±0.67	6.092±0.65	6.052±0.38
Saturated fatty acids (SFA)	34.168±0.68 ^a	33.453±0.78 ^b	33.583±0.58 ^{ab}	33.787±0.74 ^{ab}

E1, E2, E3: experimental groups; ^{a,b} – means with different superscripts differ significantly; (P ≤ 0.05) significant

DISCUSSION

Today the meat fatty acid content has become an important factor to measure the poultry meat quality, because fatty acid affecting on human health (Rahimi et al., 2011). The present study gives us data of the fatty acid content of the broiler's Ross 308 meat after using bee pollen (400 mg.kg⁻¹) into broiler's feed mixture and probiotic (3.3 g) through broiler drinking water.

From table (2) it was found that the bee pollen has increased about 13 essential fatty acids such as myristoleic acid, palmitoleic acid, oleic acid, elaidic acid, linoleic acid, arachidonic acid, heptadecanoic acid, nervonic acid, cis-11-eicosenoic acid, cis-11,14-eicosadienoic acid, cis-5,8,11,14,17 eicosapentaenoic, monounsaturated (MUFAs) and omega-3 and this is about (68.4%) of the total essential fatty acid in broiler's meat muscles. The reason why bee pollen has increased the fatty acid because bee pollen is a rich by fatty acid (Loidland and Crailsheim, 2001; Manning, 2001; Teresa, 2006; Yang et al., 2013).

Moreover, the addition of the probiotic was an increase about 15 essential fatty acids such as myristoleic acid, palmitoleic acid, oleic acid, linoleic acid, arachidonic acid, pentadecanoic acid, heptadecanoic acid, nervonic acid, cis-11,14-eicosadienoic acid, cis-8,11,14-eicosatrienoic acid, cis-5,8,11,14,17 eicosapentaenoic, polyunsaturated (PUFAs), monounsaturated, omega-3 and omega-6, in broiler's meat and this representing approximately about (78.9%) of the total essential fatty acid. This results support by **Kankaanpää et al. (2004)** who was studied the effect of probiotic on broiler's fatty acid. On the other hand, our study are not in agreement with **Zhiping et al. (2003)** who was found that the probiotic reduced the fatty acid in rats. The reason why bee pollen increases the fatty acid, because probiotic improve microorganisms in the small intestine and large intestine, which that led to increased polyunsaturated fatty acid (**Ringø et al., 1998; Kankaanpää et al., 2001; Bomba et al., 2002**).

Otherwise, when we mixed between bee pollen and probiotic they just increase about 6 essential fatty acids such as palmitoleic acid, oleic acid, linoleic acid, pentadecanoic acid, monounsaturated and omega-3 and this representing approximately about 31.6% of the total essential fatty acid. That's because probiotic increase small intestine microorganism (**Eamonn et al., 2006**), but bee pollen put down the microorganisms (**Kačániová et al., 2012**). According to these results we suggest that it's better to use bee pollen and probiotic separately. On the other hand, from the table (3) we found that the bee pollen and probiotic were reduced the saturated fatty acid such as myristic acid, palmitic acid, stearic acid and saturated fatty acid (SFA) in broiler's meat. **Ross et al. (2012)** found that the probiotic decreased (SFA) in pork. Also **Pramote et al. (2011)** support our results who was found that the probiotic had reduced (SFA) on goats. The reason why bee pollen and probiotic decrease the saturated fatty acid (non essential fatty acid) because bee pollen content flavonoids, and the flavonoids are decrease plasma lipid levels, improve glucose tolerance, and attenuate obesity, one possible mechanism underlying these physiological effects is reduction of the hepatic levels of the mRNA for stearoyl-CoA desaturase-1 (SCD1), since repression of this enzyme reduces hyperlipidaemia and adiposity (**Jung et al., 2006; La Nita et al., 2011**).

CONCLUSION

From the recent study it was concluded that the addition of natural bee pollen as a supplement diet into the broiler's feed mixture and probiotic through the drinking water, in the amount (400 mg.kg⁻¹ bee pollen) and (3.3 g probiotic) have a positive effect on broiler meat muscles because bee pollen and probiotic led to an increase the essential fatty acid and decrease the nonessential fatty acid in broiler's meat muscles. However the mixed between bee pollen and probiotic have given a negative effect on broiler's meat muscles which led to decrease the essential fatty acid.

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