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Effect of Dietary Supplementation of Plant Seeds as Fatty Acids Source on Nutrients Digestibility, Performance, Carcass and Cutting Percentage and Physical Properties Evaluation of Meat of Broiler

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Abstract. This feeding trial was conducted to investigate the effects of three kinds of plant seeds as fatty acids source on in vitro nutrient digestibility, growth performance, and physical evaluation of meat of broiler. Two hundred broiler chicks were assigned to 4 dietary treatments for 5 weeks. The birds were randomly allocated into five treatments with four replicates, and each replicate containing 10 broilers. The treatments contained 1% palm oil in diet, 1% pumpkin seeds in diet, 1% candlenut in diet, 1% nutmeg in diet, and 1% mix pumpkin seed, candlenut and nutmeg in diet. The based diet consisted of commercial diet 80%, yellow corn 11%, rice bran 8%. The variables were nutrients digestibility, growth performance, carcass characteristics, giblet, and physical properties of meat. Data were analyzed by one-way analysis of variance. The treatment means were compared using Duncan's multiple range test. The results showed that birds fed these plant seeds in diet had similar feed intake, BWG, FCR, SGR, GE, abdominal fat percentage, carcass characteristics though that of control, however had significantly different on heart, spleen, and bilein giblet, WHC and cooking loss of meat. It can be concluded that these plants seeds can be used to broiler diet for improving the performance.

Keywords: Broiler · Performance · Plant Seed

1 Introduction

Plant phytobiotics contain secondary compounds consist of essential oils, bitter, dyes, and phenolic compounds [1]. According to Oloruntola et al. [2] and Oloruntola et al. [3] the application of herbs in poultry diets has been reported to stimulate endogenous antioxidants, facilitate nutrient metabolism and improve meat quality by lowering cholesterol levels.

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Cucurbita moschata Duchesne, called pumpkin, contain seeds. Pumpkin seeds, which are rich in zinc. Trigonelin and nicotinic acid, isolated from pumpkin paste caused significant reductions in blood glucose, cholesterol and triglycerides, showing improvement in diabetic conditions [4]. Pumpkin also has antioxidant activity and is considered a natural source of beta-carotene, vitamin E and vitamin C [5].

Pumpkin is a natural source of carotenoids which are closely related to beta carotene and vitamin A and is important sources of carbohydrates [6, 7]. Pumpkin seeds are a good source of protein and fat, also contains vitamins E and A, and essential nutrients such as Zn, Mg, P, Cu, K, niacin, folic acid, riboflavin, thiamine, and high quality protein [8].

The bioactivity offers stimulates nutrition and reduces abdominal fat and serum levels of harmful lipids, while increasing serum levels of beneficial lipids [9]. In female rabbits, the antioxidant effect of pumpkin seed oil was influential [10]. Martinez et al. [11] reported that 6% *Cucurbita moschata* seed flour in the diet increased body weight, breast meat, and reduced broiler abdominal fat. In laying hens the use of pumpkin seed flour does not affect egg laying rate or egg quality [12, 13].

Aleurites moluccana L. (Wild), called candlenut, is native to Indonesia and Malaysia and has well adapted to the climatic conditions of southern and southeastern of Brazil [14]. Candlenut contains many lipid components linolenic acid, eicosanoic acid methyl ester and -tocopherol [15]. Medium-chain unsaturated fatty acids such as linoleic acid and eicosanoid acid have antibacterial and anti-inflammatory activities [16]. The use of 5% candlenut meal in diet had a significant effect on feed intake, body weight gains and intestinal weight of native chickens. However, it had no significant effect on feed conversion, carcass weight and liver weight [17]. The addition of 2.5% of candlenut seeds in the diet did not improve the performance, carcass yield, chemical compound of broiler meat, and fatty acid of breast and thigh muscles of broilers [18]. The use of 1% candlenut meal produced better blood profile and lowered cholesterol levels in broiler meat [19].

Myristica fragrans Houtt. (Myristicaceae), called nutmeg, is a medium sized, aromatic tree with slender branches. The seeds are oval with a hard shell. Found in the forests of Malaya, Penang, Sumatra, Singapore and China [20]. The nutmeg tree is native to the island of Banda, Maluku in Eastern of Indonesia [21]. Nutmeg, is an aromatic perennial tropical plant with a distinctive aroma that has several medicinal and antioxidant properties [22]. The ethno-botanical nutmeg was studied in Maluku, Central Java and East Java [23].

Nutmeg seeds and mace are used as spices, containing 4% myristicin [24]. The seeds are widely used as herbs and spices in many traditional and conventional food industries around the world [25]. The seeds contain phenolic, flavonoid, and other important bioactive compounds [26, 27]. Potential phytochemicals in the plant nutmeg have been reported to have antioxidant, antimicrobial, ant pain, ant obesity, and hepatoprotective activities in biological systems [25, 27].

Nutmeg seed extract and its quercetin compound have anti-inflammatory potential shown through inhibition of the secretion of TNF-, IL-6, IL-1β and NO [28]. The methanol extract of nutmeg seeds and mace can manage moderate oxidative stress and relieve pain. Extending treatment can increase antihyperglycemic activity [29].

Nutmeg oil contains high levels of unsaturated oils, linoleic acid and linoleic acid, which have many beneficial effects on human health [30]. The mineral content of nutmeg estimation indicated the major presence of Ca (30.95%) [31]. Nutritionally, nutmeg is rich in energy, carbohydrates, protein, dietary fiber, Vitamins A, C, and E, and minerals [32]. Administration of *Myristica fragrans* extract in the diet has been reported to inhibit lipid digestion, absorption and accumulation of adipose tissue in rats [27]. Supplementation of 200 mg/kg BW ethanol extract of nutmeg did not cause hematological parameters in rats [33]. Nutmeg plant is one of the herbal plants that has a lot of potential that can be used as a feed additive in poultry production [22].

This feeding trial was conducted to investigate the effects of the seeds of pumpkin, candlenut and nutmeg as fatty acids source on in vitro nutrient digestibility, growth performance, physical evaluation of meat of broiler.

2 Materials and Methods

2.1 Materials

The plant seed of pumpkin, candlenut, and nutmeg that were used in this research were collected from the local market in Manado.

2.2 Methods

The research was carried out by laboratory analysis in phase I, consisting of proximate and fatty acids of pumpkin seeds, candlenut and nutmeg, and nutrient digestibility of the diets.

Proximate Analysis. Pumpkin seed, candlenut and nutmeg then in the proximate analysis for the content of water, ash, crude protein, crude fat and crude fiber were carried out according to the standard method of AOAC (2000).

The Fatty Acid Analysis. The analysis of fatty acid was carried out by A.O.A.C. Official Methods 2012.13:991.33 (fatty acid in oils and fats), and fat content analysis according to A.O.A.C 2012:991.36. The pumpkin, candlenut and nutmeg was collected from the local market. The samples were dried and powdered. Fat and fatty acid were extracted from pumpkin, candlenut and nutmeg by hydrolytic method. Fat was extracted into ether, then methylated to fatty acid methyl esters (FAMEs).

In vitro Digestibility. In vitro digestibility test of the diets using the Tilley and Terry method [34] stage 2 (pepsin digestibility). The Tilley and Terry method in stage 2 is imitating digestion in the stomach, namely digestion with pepsin and hydrochloric acid with an incubation time of 48 h, the tube is centrifuged at 3,000 rpm for 15 min, the filtrate is discarded and the residue is given a solution of pepsin HCl 0, 5% as much as 50 ml and incubated for 48, the solution was then filtered using Whitman paper no 41, then dried for 48 h at 60 °C for analysis of nutrient levels.

Table 1. Proximate analysis of pumpkin, candlenut and nutmeg seeds

Component	Pumpkin Seed	Candlenut	Nutmeg
Dry matter (%)	95.79	89.83	88.99
Ash (%)	3.47	3.63	2.90
Crude Protein (%)	32.72	28.63	25.34
Crude Fibre (%)	18.23	7.27	26.08
Crude Fat (%)	20.47	10.56	28.45
Ca (%)	1.10	1.38	0.67
P (%)	0.60	0.67	0.18
Gross Energy (%)	6712.97	7834.70	6115.70

Table 2. Proximate analysis of the diets

	Water Contents	Ash	Fat	Protein	Crude Fibre
	%				
T0	11.32	6.90	5.43	17.38	11.88
T1	11.29	6.83	5.20	17.60	12.75
T2	11.29	6.93	6.49	16.67	15.53
T3	11.27	7.14	5.85	14.72	15.30
T4	11.25	6.89	5.55	16.27	16.05

In phase II of the research, two hundred broiler chicks were assigned to 5 dietary treatments for 5 weeks. The birds were allocated into five treatments with four replicates, and each replicate containing 10 broilers. The treatments contained 1% palm oil in diet (T0), 1% pumpkin seeds in diet (T1), 1% candlenut in diet (T2), 1% nutmeg in diet (T3), and 1% mix pumpkin seed, candlenut and nutmeg in diet (T4). The based diet consisted of commercial diet 80%, yellow corn 11%, rice bran 8%. Proxymate analysis of pumpkin, candlenut and nutmeg seeds, and the diets shown in Table 1 and 2. The variables were nutrients digestibility, growth performance, carcass characteristics, giblet and physical properties of meat. Data were analyzed by one-way analysis of variance. The treatment means were compared using Duncan's multiple range test.

3 Results and Discussion

Table 3 showed the effect of supplementation of plant seed on the diet on the digestibility of nutrients (*in vitro*) and Table 4 showed the fatty acids composition of pumpkin, candlenut and nutmeg seeds. In general, that plants seeds supplementation improved the digestibility of nutrients of the diets (*in vitro*).

Table 3. Effect of supplementation of plant seed on diet on the digestibility of nutrients (in vitro)

	T0	T1	T2	T3	T4
	%				
Dry Matter Digestibility	83.10	88.33	85.66	87.32	86.79
Organic Matter Digestibility	82.68	88.03	84.85	86.51	86.70
Protein Digestibility	79.49	85.68	80.10	83.72	84.65
Crude Fibre Digestibility	65.20	69.14	67.74	66.74	66.89
Fat Digestibility	73.10	77.93	73.56	75.04	76.12

Table 4 showed that the three kinds of the plant seed have differ in the amount and type of fatty acids. Gamma linoleic acid (GLA) (rich in candlenut) is an important omega-6 PUFA of medicinal, and essential for the formation of metabolites from precursor essential fatty acid. GLA was reported to induce lipid peroxidation in tumor cells and lead to altered mitochondrial metabolism and ultrastructure, cytochrome-c release, caspase activation, and apoptosis [35].

Meristic acid methyl ester is less water soluble but more amenable for the formulation of myristate-containing diets and dietary supplements. It is named for nutmeg (*Myristica fragrans*), from which it was first isolated in 1841 by Playfair [36]. Myristic acid have positive effects on HDL cholesterol and improve the HDL to total cholesterol ratio [37]. Myristoleic acid methyl ester is a more hydrophobic form of the free acid.

Palmitoleic acid (rich in pumpkin seed) is an omega-7 MUFA that is found in plants [38]. The MUFA palmitoleic acid or palmitoleate has received a lot of attention in recent years, even though its metabolism was described in the 1960s [39]. The cis isoform (cis-palmitoleate) has been associated with increased insulin sensitivity and decreased lipid accumulation in the liver [40].

Linolelaidic acid (rich in pumpkin seed) is an omega-6 trans fatty acid (TFA), isomer of linoleic acid. It is found in partially hydrogenated vegetable oils [41]. Arachidic acid, also known as icosanoic acid, is a saturated fatty acid with a 20 C-chain. The salts and esters of arachidic acid are known as arachidates (found in pumpkin and candlenut seed) [42].

Heneicosanoic acid (foundin candlenut), showed significant inhibitory effects towards human p53 DNA binding domain [43]. Methyl henicosanoate is a fatty acid methyl ester which has a role as a plant metabolite. It is functionally related to a henicosanoic acid.

Table 5 showed the effect of adding the various of plant seed to the diet on feed intake, average weight gain (g) FCR, GE, and SGR for broiler. In general, no statistically significant differences on the growth performance.

Table 6 showed the effect of adding the various of plant seed to the diet on carcass cutting, abdominal fat, and giblet for broiler. That there were no significant differences on carcass cutting, abdominal fat, liver and gizzard. However, there were significant difference ($P < 0.05$) for heart, spleen, and bile.

Table 4. The fatty acids composition of three kinds of plant seeds

No	Parameter	Pumpkin Seed	Candlenut	Nutmeg
1	Total Fat (%)	14.98	45.43	14.39
	Octanoate	-	1.50	-
	Undecanoate	2.48	4.85	10.40
	Laurite	-	0.14	1.24
	Hexanoate	-	0.14	-
	Tridecanoate	-	0.43	-
	Myristate	0.12	0.24	0.56
	Myristoleic Acid Methyl Ester	-	-	64,91
	Palmitate	-	7.98	7.58
	Palmitoleate	22.80	-	-
	Cis-9-Oleate	5.53	0.12	2.30
	Heptadecanoate	-	-	0.18
	Stearate	-	0.22	5.57
	Trans-9-elaidate	-	0.78	0.54
	Linolelaidate	34.76	8.53	1.49
	Gamma-Linolenic Acid Methyl Ester	-	26.36	-
	Arachidate	32.43	13.82	-
	Linolenate	0.67	8.30	0.30
	Linoleate	-	-	1.56
	Heneicosanoate	0.12	23.91	-
	Cis-13-docosenoate	0.15	-	0.15
	Cis-5-8-11-14-eicosatetraenoate	0.11	-	-
	Cis-13-16-docosadienoate	0.25	-	-
	Cis-5-8-11-14-17-eicosapentanoate	0.26	0.26	2.27
	Lignocerate	0.29	-	0.24
	Nervonate	0.13	-	-

Dietary inclusion of pumpkin, candlenut and nutmeg meal at level of 1% each in the diets did not affect to the body weight, slaughter weight, carcass weight and yields of carcass cutting (Table 5 and 6). Based on the carcass data, it is indicated that there were no negative effects of including these plants seeds meal to broiler chicken diets. The inclusion of plants seeds meal in diet caused decreasing of abdominal fat. The decreased of abdominal fat weight is in agreement with the previous studies conducted by Ferrini et al. [44], who have reported that abdominal fat significantly decreased in chickens fed diets rich in n-3 PUFA.

Table 5. Effect of the diet on growth performance

Variable	Treatments					SEM	P Value
	T0	T1	T2	T3	T4		
Feed Intake, g	2426.23	2435.45	2444.53	2431.66	2463.22	36.21	.99
Initial Body Weight, g	115.03	115.00	109.25	112.48	117.33	1.18	.25
Final Body Weight, g	1468.33	1472.18	1441.06	1406.20	1472.20	22.03	.88
Weight Gain, g	1353.83	1357.18	1331.83	1293.73	1354.90	21,29	.89
FCR	1.65	1.66	1.69	1.73	1.68	.014	.44
GE	2.95	2.95	3.05	2.80	2.89	.04	.67
SGR, %	63.75	63.71	63.97	63.16	63.21	.30	.91

Table 6. Carcass cutting, abdominal fat, and giblet of broiler chicken fed various of plant seed on day 35 of age

Variable	Treatments					SEM	P value
	T0	T1	T2	T3	T4		
Body Weight, g	1767.50^{b}	1712.00^{b}	1644.50^{ab}	1588.25^{a}	1680.75^{ab}	23.10	.13
Carcass, %	71.83	71.90	69.50	71.01	70.30	.38	.22
Breast, %	34.32	33.21	31.60	32.67	32.99	.52	.62
Drumstick + Thigh, %	31.47	31.74	31.52	31.27	31.48	.42	.99
Legs, %	5.80	5.80	6.00	6.01	5.36	.19	.86
Back, %	18.12	18.74	20.23	19.46	19.48	.54	.81
Wings, %	10.30	10.51	10.66	10.59	10.70	.11	.85
Abdominal Fat, %	1.76	1.93	1.99	1.71	1.97	.09	.80
Liver, %	1.77	1.91	2.02	1.94	1.96	.06	.75
Gizzard, %	1.40	1.48	1.48	1.45	1.43	.03	.94
Heart, %	0.59^{c}	0.47^{ab}	0.55^{bc}	0.46^{ab}	0.45^{a}	.02	.02
Spleen, %	0.11^{ab}	0.11^{a}	0.12^{ab}	0.16^{b}	0.13^{ab}	.01	.20
Bile, %	0.13^{ab}	0.11^{a}	0.14^{ab}	$0,14^{ab}$	0.17^{b}	.01	.14

Table 7 showed the effect of adding the various of plant seed to the diet on physical properties of broiler meat. There were no significant differences on water content of the meat. However, there were significant difference ($P < 0.05$) for cooking loss, WHC, and pH of the meat.

Table 7. Physical properties of the broiler meat

Variable	Treatments					SEM	P value
	T0	T1	T2	T3	T4		
Cooking Loss, %	36.52^{b}	37.09^{b}	32.97^{ab}	29.86^{ab}	28.15^{a}	1.28	.08
Water, %	75.34	74.56	75.50	75.99	74.96	.25	.46
WHC	46.36^{ab}	43.87^{ab}	42.12^{a}	50.03^{b}	39.39^{a}	1.26	.06
pH	5.71	5.70	5.62	5.72	5.67	.02	.50

Water holding capacity is defined as the ability of meat and meat products to bind water during slicing, mincing, and pressing and also during transport, storage, processing, and cooking [45]. Poor water holding capacity in poultry meat results in diminished visual appeal and inferior palatability for consumers, reduced ingredient retention, protein functionality, and product yields. pH has a direct bearing on the meat quality attributes such as tenderness, water-holding capacity, colour, juiciness and shelf life. The high pH of broiler breast meat has a higher water binding capacity than meat with lower pH. Identification of colour is an easy way to determine the pH of meat. If the meat is very dark, it will have a high pH and if it is very light, it will have a low pH [46].

4 Conclusions

The present study reveal the potential of plant seeds to enhance the growth performance and carcass product when used as a phytogenic feed supplement. That the inclusion 1% of pumpkin, candlenut, and nutmeg in diet had similar feed intake, BWG, FCR, SGR, GE, abdominal fat percentage, carcass characteristics though that of control, however had significantly different on heart, spleen, and bile in giblet, WHC and cooking loss of meat. They didn't have negative effects on carcass quality and cutting yields of broilers, and could improve meat quality of broilers. It can be concluded that these plants seeds can be used to broiler diet for improving the performance.

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