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Effect of Different Methods of Processing and Antioxidant Supplementation on the Storage Quality of Hatchery Waste Meal

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Abstract: Four different levels of antioxidant (Oxygun®) at 0, 100, 200 and 300 mg/kg were added to the ground, processed viz, cooked, autoclaved, extruded and raw hatchery waste. Samples were stored for 50 days and analyzed for peroxide value (PV), thiobarbituric acid value (TBA) and free fatty acids (FFA) with 10-days of storage interval. Linear increase was observed in PV, FFA and TBA values of all treatments throughout the storage period. Comparison within raw hatchery waste (RHW), cooked HW, autoclaved HW and extruded HW revealed that extrusion was found to be the most effective processing technique to check fat oxidation, followed by autoclaving and simple cooking whereas unprocessed HW showed maximum fat deterioration. PV, FFA and TBA decreased with all levels of antioxidant addition when compared with control group. Significantly highest ($p<0.05$) levels of PV, FFA and TBA were observed in control group while least degradation of fat was observed in extruded samples with 300 mg/kg of antioxidant addition. All the processing techniques and antioxidant addition checked the fat rancidity throughout the storage period, compared to control group, however synergistic effect of extrusion and 300 mg/kg of antioxidant addition was found to be most appropriate to keep the samples highly acceptable for 50 days storage period.

Key words: Antioxidant, oxidation, processing technique, rancidity

INTRODUCTION

The scarcity of conventional feed resources coupled with high prices and extreme weather conditions are the limiting factors in the cost effective preparation and supply of poultry feed. Soybean meal (SBM) is the most conventionally used protein feed ingredients in poultry diets. However, the high cost of SBM, (McGill and Firman, 2009) demands non-conventional low cost feed resources. Hatchery waste (HW) is one of such non-conventional feed resource which can be used as cheaper source of protein in poultry feed. Hatchery waste includes shells from hatched chicks, infertile eggs, dead embryos and low grade unsalable and dead chicks. while its composition may vary due to hatch percentage and factors related to it. However, one limiting factor of HW inclusion rates in the poultry feed is its fat contents (18-21%), which depends upon the number of infertile eggs in the final product (Leeson and Summer, 2001). Unsaturated fatty acids present in the improperly stored fats (Nelson and Cox, 2008) are more prone to rancidity which may deteriorate the quality of poultry feed leading to poor growth rate. It affects the organoleptic quality of feed by the formation of many breakdown products like ketones, aldehydes etc; so, there is need to control its deleterious effects by supplementation of antioxidant.

Different methods have been used for the estimation of various intermediate and end products of oxidation, i.e., thiobarbituric acid value (TBA), free fatty acids (FFA) and peroxide value (PV). As these products are “moving targets” so it is very difficult to predict which method is the best indicator of fat oxidation (Hamilton and Kirstein, 2008).

In the present study, HW was processed by different techniques i.e., simple cooking, autoclaving and extrusion cooking, to form hatchery waste meal (HWM) and then further fortified with antioxidant to determine its efficacy at different levels. Although the supplementation of antioxidants cannot stop the process of auto-oxidation but delay it.

Keeping in view, the present study was designed to evaluate the storage quality of HWM at different storage periods with the supplementation of different levels of anti-oxidant.

MATERIALS AND METHODS

Two experiments were conducted at Department of Food and Nutrition, University of Veterinary and Animal Sciences Lahore, Pakistan, to determine the chemical composition of HWM after applying different processing techniques and its keeping quality after t antioxidant supplementation. The raw hatchery waste (RHW), collected from broiler residue, comprised of infertile eggs, shells, dead in shells and low grade unsalable chicks. The RHW was subjected to the following three processing techniques.

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Cooking: The HW was processed by simple cooking in water with a ratio of HW to water as 2:1 (Khan and Bhatti, 2001). The simple cooking process continued for about 3 h at 100°C in an open container with regular stirring. The product after cooking was dried by placing in an oven at 60°C till constant weight and ground through a laboratory mill for further analyses.

Autoclaving: The dried and ground HW was put in the plastic jars, covered with aluminum foil. These plastic jars were then placed in the autoclave and subjected to a temperature of 125°C along with 1.76 Kg/cm^2 pressure for 15 min. Lilburn *et al.* (1997). The autoclaved HWM was stored till further analyses.

Extrusion cooking: The dried and ground HW was passed through dry extruder (frequency 50 Hz, orifice size 8-12 mm, screw speed 536 rpm, feed rate 750 kg/h) using a single screw (Model: 1000-4 Miltenz Auckland, NZ). The internal temperature of the extruder barrel was 115 to 126°C at the point of extrusion. The final extruded product was collected on polythene sheet spread on cardboard piece and placed on the floor (Tadtiyanant *et al.*, 1993).

Chemical composition of HWM: In the first experiment, the representative samples of the processed HWM were subjected to chemical analyses for crude protein (Method no. 984.13 A-D), crude fat (Method no. 920.39 A), crude fiber (Method no. 978.10), total ash (Method no. 942.05), nitrogen free extract (by difference method), calcium (Method no. 968.08), phosphorus (Method no. 965.17), lysine (Method no. 982.30 E chp 45.3.05) and methionine (Method no. 982.30 E chp 45.3.05) according to their respective AOAC (2006) procedures.

Stabilization of fat: In the second experiment, HWM prepared from different processing techniques were stored with four different levels of antioxidant (Oxygun®, Impextraco, Belgium) viz., 0, 100, 200 and 300 mg/kg for 50 days. The antioxidant brand was in powder form based on the synergism between BHA (E-320), BHT (E-321), ethoxyquin (E-324), citric acid (E-330) and ortho-phosphoric acid (E-338) and especially designed for compound feeds and premixes. The samples were then stored at room temperature in air tight sterilized glass jars. There were 15-jars for each treatment and out of those 15 jars three jars were opened at each storage interval after every 10 days to conduct triplicate analysis PV (AOAC Method no. 965.33), TBA (AOCS Method no. Cd 19-90) and FFA (AOCS Method no. Ca 5a-40) (AOCS, 1998; AOAC, 2006). The room temperature variation was noted daily.

The average PV, FFA and TBA values for unprocessed HWM at 0 day was found to be 2.30±0.05 mEq/kg, 5.61±0.09 and 12.1±0.12 mg/kg, respectively.

Statistical analysis: All the parameters at all-time intervals were analyzed in triplicates. The data, thus obtained, were statistically analyzed through two way analysis of variance technique under Randomized Complete Block Design (RCBD) (Steel *et al.*, 1997) Processing techniques were considered as blocks, whereas levels of antioxidant and storage intervals were taken as factors. The level of significance was defined as p = 0.05. The significance comparisons of means were obtained through DMR test. All the data were analyzed through CoStat version 6.303 developed by CoHort Sofwares, Monterey, CA, USA.

RESULTS

In the first experiment chemical analyses of cooked, autoclaved and extruded HWM was performed (Table 1). In the second experiment, PV, FFA, TBA values of HW and HWM were evaluated to determine the effectiveness of different processing techniques i.e., simple cooking, autoclaving and extrusion cooking and various levels of antioxidant (Oxygun®) for different storage periods (Fig. 1, 2 and 3). During the 50 days storage period, variable values of oxidation were observed. The PV, FFA and TBA values of HW and HWM treated with different processing techniques were compared at different storage periods, which depicted significant differences (p = 0.05) with respect to processing techniques, storage intervals and antioxidant levels.

Peroxide value (PV): Oxidative process speeds up when a product contains unsaturated fatty acid which ultimately results in the formation of undesirable products resulting in repulsive smell with toxic properties thereby, leading to nutritional and economic losses (Cheeke, 1991). Dietary fat quality in terms of PV of feed or feed ingredient has significant impact on animal growth performance, metabolism and digestibility (Lin *et al.*, 1989; Enberg *et al.*, 1996; Lin *et al.*, 1989). PV of hatchery waste material was significantly affected ($p<0.05$) with antioxidant level, various processing techniques and storage period (Fig. 1 and 4).

In non-processed as well as the processed HW, PV increased linearly throughout the study intervals (Fig. 2). Overall increase in PV was from 8.17e to 19.23a mEq/kg up to 50 days of hatchery waste storage; however increase was lower in processed and antioxidant fortified samples (Fig. 1 and 4). Inclusion of antioxidant at all levels caused significant ($p<0.05$) reduction in peroxide formation at all storage intervals as compared to samples without antioxidant addition, thereby indicating an improved storage stability of hatchery waste meals (Fig. 4).

All the processing techniques i.e., simple cooking, autoclaving and extrusion cooking were found to be efficient in stabilizing the fat content leading to lower PV

Table 1: Chemical composition (Mean±SD) of raw hatchery waste and meal processed by different techniques

		Hatchery waste meal		
*Nutrient (%)	Raw hatchery waste	Cooked	Autoclaved	Extruded
Crude protein	44.63±2.23	43.67±1.98	44.1±1.61	41.64±0.96
Crude fat	26.46±0.63	27.14±0.85	25.35±0.75	26.85±0.35
Crude fiber	1.05±0.05	1.62±0.09	1.31±0.03	1.47±0.09
Total ash	25.88±0.54	25.81±0.67	26.94±0.92	27.9±1.60
Nitrogen free extract	1.98±0.15	1.76±0.04	2.3±0.12	2.14±0.13
Calcium	17.56±0.88	19.02±1.02	18.62±0.76	18.95±0.57
Phosphorus	1.63±0.05	1.99±0.14	1.44±0.09	3.21±0.16
Lysine	2.93±0.12	2.83±0.18	2.8±0.21	2.79±0.20
Methionine	1.21±0.10	1.19±0.13	1.19±0.09	1.23±0.02

*All values are on dry matter basis

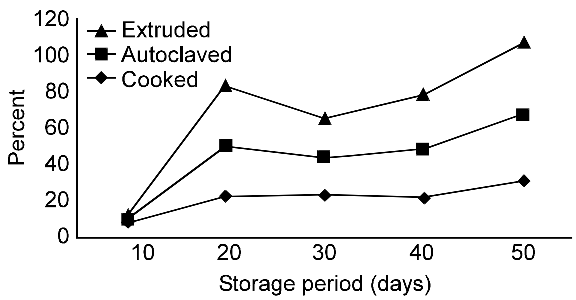

Fig. 1: Percent less increase in PV of hatchery waste meal processed by various techniques as compared to raw hatchery waste

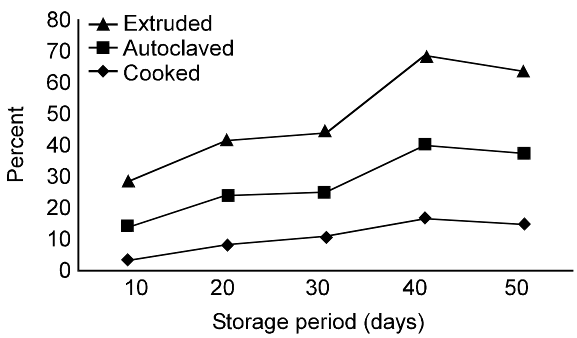

Fig. 2: Percent less increase in FFA of hatchery waste meal processed by various techniques as compared to raw hatchery waste

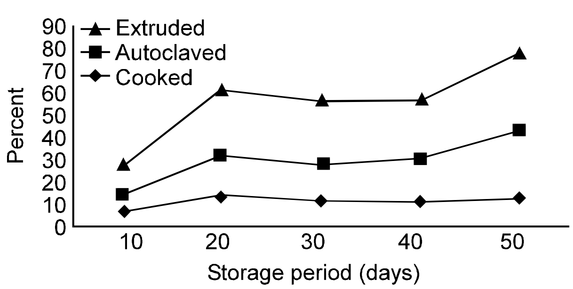

Fig. 3: Percent less increase in TBA of hatchery waste meal processed by various techniques as compared to raw hatchery waste

value in processed hatchery waste meals (Table 1 and Fig. 1). Extrusion cooking was found to be most valuable processing technique, which resulted in 41.80% decreased PV followed by autoclaving (38.60% and simple cooking (33.05%) as compared to raw hatchery waste meal samples after 50-days of storage period (Fig. 1).

It was revealed that with the increasing level of antioxidant, the PV was significantly ($p<0.05$) reduced. Similarly extrusion technique also proved highly efficient ($p<0.05$) to reduce the fat rancidity in hatchery waste samples (Fig. 1). Although, with storage period significant ($p<0.05$) increase in PV was observed yet the synergistic effect of antioxidant and extrusion technique produced the best results to reduce the increase in PV with storage period.

Free fatty acids: The mean value of free fatty acids (FFA) content of raw, cooked, autoclaved and extruded hatchery waste with 100 mg/kg, 200 mg/kg and 300 mg/kg of antioxidant with 50 days of storage period are presented in (Fig. 5). The FFA content of HWM was significantly ($p<0.05$) affected with respect to processing technique, antioxidant level and storage period.

In present study storing raw hatchery waste without antioxidant addition for 50 days resulted almost in 3 fold increase from baseline values (9.70±0.02 to 34.57±0.03%) (Fig. 5). Raw hatchery waste contained the highest amount of FFA (15.08a%). Although all the processing techniques resulted in decrease in FFA content of hatchery waste, however, extrusion cooking was found to be the most appropriate processing technique to reduce the fat degradation by lowering the FFA content, thus resulting in improved quality of the product (Fig. 2). Addition of antioxidant resulted significant decrease ($p<0.05$) in FFA content at all augmentation levels (Fig. 5). It was observed that inclusion of antioxidant significantly checked the FFA content of hatchery waste with storage.

Addition of 100 mg/kg of antioxidant resulted in decrease of 18% in FFA content as compared, whereas addition of further 100 mg/kg and 200 mg/kg resulted in additional decrease of 5 and 8%, respectively. Therefore 100 mg/kg was found to be most cost effective tool to

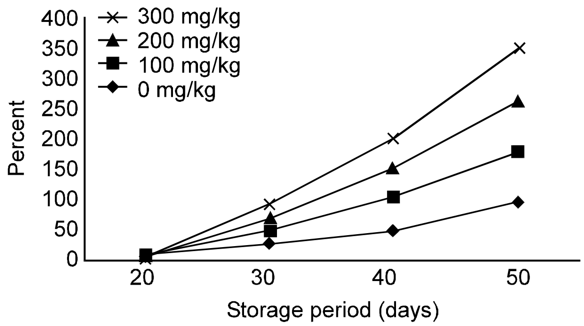

Fig. 4: Percent less increase in PV of hatchery waste meal fortified with various antioxidant levels as compared to control

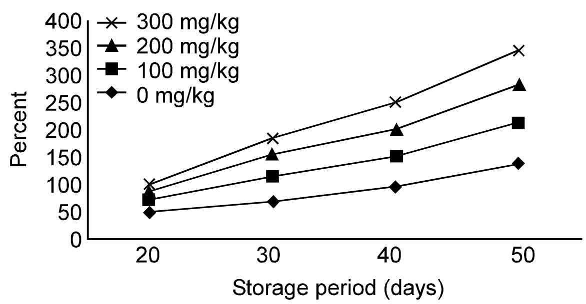

Fig. 5: Percent less increase in FFA of hatchery waste meal fortified with various antioxidant levels as compared to control

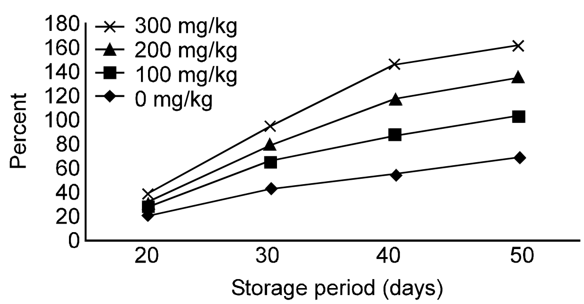

Fig. 6: Percent less increase in TBA of hatchery waste meal fortified with various antioxidant levels as compared to control

decrease the deterioration of fat content in hatchery waste material. FFA concentration in hatchery waste linearly connected with storage time; increased from 9.10 to 16.97% from 10 days to 50 days of storage period. Although, the similar general pattern was noted in all the samples with storage yet the increase in FFA content was lower in processed and antioxidant treated samples as compared to raw samples and with no antioxidant added samples.

TBA value: The statistical analysis of various treatments showed significant effect ($p<0.05$) of antioxidants, processing technique and storage period on TBA values (Fig. 3).

The TBA values increased linearly and significantly throughout the storage period, however, the increase was variable with respect to antioxidant treatment and processing technique employed initially to stabilize the hatchery wastes (Fig. 3 and 6). Extrusion cooking was found to be the most appropriate stabilization technique followed by autoclaving and simple cooking (Fig. 3). TBA value decreased up to 30% with extrusion cooking, 21.4% with autoclaving, while the decrease was 12% with simple cooking as compared to control with 50 days of hatchery waste storage. Addition of antioxidant resulted in improved quality of hatchery waste as it decreased the fat degradation at all levels (Fig. 6). The significantly highest TBA value was observed in hatchery waste without the augmentation of antioxidant (16.21a mg/kg) followed by 100 mg/kg of antioxidant level (13.50b) whereas; the minimum TBA value was recorded in the waste materials with 300 mg/kg of antioxidant addition.

Overall, the highest TBA value was observed in raw hatchery waste without antioxidant fortification (30.14±0.22 mg/kg) in contrast to the minimum value noted in extruded hatchery waste meal with 300 mg/kg of antioxidant addition (14.06±0.10) (Fig. 3).

DISCUSSION

Oxidative rancidity can be checked by the addition of antioxidants in feed. Antioxidant retards the oxidative spoilage of unsaturated fat, however, spoilage accelerates if exposed to light, air and high temperature (Warraich, 1972). In the presence of light, heat, oxygen or metals, unsaturated fatty acids are converted to radicals of free fatty acids which undergo air oxidation to produce peroxides and decompose to aldehyde and ketone, resulting in bad smell. During storage many kinds of reactions such as oxidation, hydrolysis, polymerization, cyclization and B, scission can occur in the fat and oil leading to production of toxic compound like resins, which are injurious to health (Frankel, 1988; Gotoh and Wada, 2006). Gyula *et al*. (2005) added Vitamin E (natural antioxidant) and found it significantly effective against the harmful effects of lipid oxidation. They inferred that the fortification of food by vitamin E is beneficial in the case of fat supplementations.

The results of present study are in line with those reported earlier by Anjum *et al*. (2004); Waheed *et al*. (2004) reported decrease in fat oxidation with the addition of antioxidants and increase with the fat content of the commodity. Stabilization techniques have also been previously, reported to improve the product quality and storage stability of the fat containing products (Bjorck and Asp, 1983; Fagbenro and Jauncey, 1993; Gajula *et al*., 2009).

The results are also supported by the readings of other parameters i.e., PV and TBA values; also showed the similar pattern with processing, antioxidant addition and storage period. The fat is vulnerable to be broken down and get rancid during storage; more fat content and increased storage period increases the quantity of FFA (Molteberg *et al*., 1995) which can be checked or reduced with antioxidant addition and applying the fat stabilization techniques. Estevez *et al*. (2007) in a study reported severe oxidative deterioration of frankfurters during refrigeration due to gradual degradation of PUFA, with the generation of oxidative products like TBA-RS, hexanal etc. The results obtained during present study are also supported the above mentioned phenomenon. Similar findings have been previously observed by Molteberg *et al*. (1995) who reported reduction in the FFA content by an average of 50% by different processing techniques. This reduction in FFA is probably due to complexing of fatty acids by treating with NaOH according to the acid value. Free fatty acids are reported to be increased in the fats when stored for a longer period of time (Wallace *et al*., 2010). Many studies conducted to demonstrate the effect of FFA on diet performance have shown variable effect of FFA (Austreng and Gjefsen, 1981; Plascencia *et al*., 1999; Schiffelers *et al*., 2001).

The TBA value has been found to be linearly correlated with the fat content and the storage duration of the commodity (Waheed *et al*., 2004; Sharif *et al*., 2005; Ozogul *et al*., 2006). Thus the higher TBA as well the values for other fat oxidation parameters might be due to higher fat content (23.75 to 28.85%) in hatchery waste samples.

The results of present investigation are in line with the earlier findings of Anjum *et al*. (2004) and Hendriks *et al*. (2006) who reported decrease in TBA value with antioxidant addition and increase with fat content and storage period. Extrusion technology showed excellent results compared to the rest of the techniques applied thereby delaying the rancidity of fat compared to other techniques. The lowest values of PV, TBA and FFA attributed to the extruded hatchery waste depicted its advantage upon other techniques. This may be due to the high temperature and sudden drop in pressure after the material left the extruder barrel. It volatilized the fat and moisture quickly, compared to autoclaving and cooking techniques used in this experiment.

Conclusion: All the processing techniques as well as antioxidant addition checked the fat rancidity throughout the storage period as compared to raw samples without antioxidant addition, however synergistic effect of extrusion cooking and 300 mg/kg of antioxidant addition was found to be most appropriate to keep the samples highly acceptable for 50 days storage period.

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