P-ISSN: 2305-6622; E-ISSN: 2306-3599
International Journal of Agriculture and Biosciences
www.ijagbio.com; editor@ijagbio.com

Research Article

Effects of Fermented Mango (*Mangifera indica*) Kernel Composite Meal on the Haematological Parameters of Laying Quails (*Coturnix coturnix japonica*)

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Article History: Received: January 12, 2019 Revised: June 22, 2019 Accepted: July 16, 2019

ABSTRACT

A study was conducted to investigate the effect of Fermented Mango Kernel Composite (FMKCM) on the Haematological parameters of laying Japanese quails. One hundred and ninety five Japanese quails were randomly allotted to five dietary treatments (I –V) of 39 hens each. Each treatment was replicated thrice with 13 hens per replicate. In each of the five diets, FMKCM was used to replace maize at 0%, 10%, 15%, 20% and 25% for i, ii, ii, iv and v, respectively. Haemoglobin, Red blood cell, White blood cell, Packed cell volume, Mean corpuscular volume, Mean corpuscular haemoglobin, Mean corpuscular haemoglobin concentration, Platelet, Neutrophils, Monocytes, Lymphocytes, Eosinophils and Basophils were determined at the 84th day (that is 12 weeks) and the result showed that all the parameters measured were within the normal reference ranges of healthy quails. It is therefore concluded that FMKCM could replace maize up to 25% without any deleterious effect on their haematology.

Key words: Fermented mango kernel, Maize, Japanese quail, Haematological indices

INTRODUCTION

In recent times, a new genus of poultry, Japanese quail (*Coturnix coturnix japonica*) was introduced into Nigeria by National Veterinary Research Institute (NVRI) Vom to expand the poultry sub sector and help supplement the domestic chicken production through meat and egg (Edache *et al*., 2007; Ani *et al*., 2009). The quails have unique characteristics and advantages over other species of poultry which include early attainment of sexual maturity, short generation interval making it possible to have many generations in a year (Anon, 1991), high rate of egg production between 200-300 eggs in 360 days and are very resistant to common epidemics of poultry (NRC, 1991). Quails are birds that which strive very well in cages and are relatively inexpensive to maintain. They are birds that every household can keep without stress. The common Japanese quail matures in about six weeks and are usually in full egg production by 50 days of egg. If properly mated, quail birds have high fertility and hatchability. The quails are hardy birds that can adapt easily to different environments (Haruna *et al*., 1997). Their meat and eggs are renowned for their high quality protein, high biological value and low caloric content, making it a choice product hypertensive patient (Haruna *et al*., 1997; Olubamiwa *at al*., 1999). Despite all this benefits, there are no improved feeding regimes, the most relevant option to arrest the present feed crisis of the livestock industry is by-product utilization (Atteh, 1986). These point clearly to alternative feed stuff for livestock feed production in order to cut down feed prices and make them more affordable by livestock farmers. As a result of shortage of convectional feed stuffs. Livestock nutritionists have continued with their search for alternative feedstuffs. These alternative feed must be cheap, readily available and less competed for by man and industries or not competed at all. (Akinmutimi, 2004). The search for substitute has led to the discovery of non-conventional energy feed such as cocoyam, cassava, mango kernel etc.

Mango kernel is a good source of soluble carbohydrates (Saadany *et al*., 1980; Jansman *et al*., 1995; Teguia 1995; Diarra 2008). The protein of the kernel (7.80-8.00%) is comparable to that of maize but it has higher fats (7.80-9.00%) than maize (Saadany *et al*. 1980). Mango kernel flour is reported to be equal to rice in food if tannin is free (Morton, 1987). Tannins are known to interfere with protein digestibility and render it unavailable. There are other anti-nutrients contain in mango kernel such as;

Cite This Article as: Kolo YS, AF Bette and H Charles, 2019. Effects of fermented mango (*Mangifera indica*) kernel composite meal on the haematological parameters of laying quails (*Coturnix coturnix japonica*). Inter J Agri Biosci, 8(5): 226-230. www.ijagbio.com

phytate, hydrogen cyanide, trypsin inhibitor, oxalate, saponin etc. processing methods such as; boiling, fermentation, drying have been reported to be effective in reducing these anti-nutrients (Abang *et al*., 2013; Diarra *et al*., 2008). Even though, the proportion of dress weight to live weight is used as a measure of productivity in farm animals (Ijaiya and Fasinya, 2004), it is equally very important to consider the health status of animals used in various feeding trials. One of the ways of assessing it, is by the use of haematological studies (Ojebiyi *et al.*, 2007). The aim of this study therefore is to evaluate the effect of replacing maize with fermented mango kernel composite meal on the haematological parameters of laying quails.

MATERIALS AND METHODS

Experiment site

This experiment was conducted at the Poultry Unit of the Teaching and Research Farm of the Federal University of Agriculture, Makurdi, Benue state. Makurdi is located at the longitude 6º 10' East and latitude 6º8' North. The area is warm with a minimum temperature range of 29.8-35.6ºC. Rainfall is between 508-1016mm and relative humidity is 47%-87% (Anon, 1995). One important geographical features of this area is the river Benue which divides Makurdi into the Northern and Southern parts. Makurdi local Government has an area of 16km radius. It lies within the Guinea savannah region of the Nigeria vegetative belt located in the Benue valley. Makurdi experiences a typical tropical climate with two distinct seasons (dry and wet). The dry season begins in November and ends in March while the wet season starts in April and ends in October. Harmathan with cool weather is experienced from December to early February (Anon, 1995).

Preparation of Experimental Materials

Different cultivars of both indigenous and improved mango were collected during the month of May (peak of the mango season) in Gboko and Makurdi area of Benue state, Nigeria. Mango kernel was removed by cracking manually with the aid of hammer. The fresh kernels were soaked in water at room temperature to allow it ferment for a period of 2 days (48hrs) in order to reduce the anti-nutrients to a more tolerable level and rinsed thoroughly with clean cool water. The fermented kernel was sundried in order to reduce the moisture content to less than 10% to prevent microbial build up and for prolonged storage. The ingredients were crushed separately into fine grit and were later mixed at varying inclusion levels with other ingredients to formulate the various diets.

Chemical analysis

Chemical analysis of fermented mango kernel and experimental diets were analyzed using (AOAC, 1995).

Formulation of diets

Feeds were formulated to meet the nutritional requirements for quails during the growing phase. Fermented mango kernel composite meal replaced maize at 0% (control diets was compounded with 100% maize and 0% FMKCM) 10% (diet was compounded with 90% and 10% FMKCM) 15% (diets was compounded with 85% maize and 15% FMKCM) 20% (diet was compounded with 80% maize and 20% FMKCM) and 25% (diet was compounded with 75% maize and 25% FMKCM) in treatments I, II, III, IV, V respectively.

Animal grouping

A total of one hundred and ninety five two weeks old un-sexed Japanese quails of about 33.60g of weight purchased with the national veterinary research institute Vom –Jos, Nigeria. At the start of the feeding trial, three groups were allotted to five dietary treatments of 39 quails each. Each treatment was replicated thrice with 13 quails per replicate.

Housing

The birds were managed intensively in cages of three tiers. Each tier was separated with wood. Wire mesh was used for the walls and doors to allow adequate ventilation/lighting. The dimension of each tier was ($1.0m^2$ x $0.78m^2$). Litter materials (wood shaving) were used on the wooden floors. Each tier was equipped with adequate drinkers and feeding troughs. A floor space of about 0.007 m^2 to 0.009 m^2 per quail was provided. Artificial lighting was provided with the use of one battery lantern for each tier to ensure adequate feed intake.

Routine operations

Feeds were weighed with a micro scale balance of 5kg before serving to ensure a uniform amount across treatments. Quails were served with 250grams of feeds for the first week at about 8 am on daily basis, the quantity was increased by 50grams on weekly basis. Fresh clean water was supplied ad-libitum. Drinkers and feeders were washed and disinfected using izal when appropriate. Litter materials were changed when due and replaced accordingly.

Design and analysis

At the end of the experiment (12 weeks of age) two (2) laying quails per replicate (amounting to 6 quails per treatment) were randomly selected. Quails were slaughtered by cutting the jugular vein with sharp knife. The blood samples were collected at slaughter into bottles coated with ethylene diamine tetracetic acid (EDTA) as an anti-coagulant for haematological essay. The samples were then analyzed for packed cell volume (PCV) using the micro haematocrit method (Coles, 1986). Haemoglobin concentration (HB) was determined by the cyanomet haemoglobin method (Kachmar, 1970). Total white blood cell (WBC) count was determined using the standard method described by Schalm *et al*. (1975). Differential leucocytes count was obtained by Lieshman staining techniques and counting with differential cell counter (Schalm *et al*., 1975). The results were interpreted using the reference range of Haematological test for female Japanese quails (Campbell, 1988). The data obtained on all the parameters studied were subjected to one-way analysis of variance (ANOVA) using Minitab statistical software version 14 (Minitab, 2014) and least significant method was used to separate means that differed significantly (Steel and Torries, 1980).

Table 1: Composition of Diet with Fermented Mango (*Mangifera spp*) Kernel Composite Meal (FMKCM) for Laying Japanese Quails (*Coturnix coturnix japonica*)

Ingredients	(0%)	(10%)	(15%)	(20%)	(25%)
Maize	52.00	47.00	45.10	42.80	40.50
FMKCM	0.00	5.00	6.90	9.20	11.50
Soybean meal	23.00	23.00	23.00	23.00	23.00
Groundnut cake	16.00	16.00	16.00	16.00	16.00
Bone meal	7.00	7.00	7.00	7.00	7.00
Lysine	0.50	0.50	0.50	0.50	0.50
Methionine	0.50	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50	0.50
Vit/min premix	0.50	0.05	0.50	0.50	0.50
Total	100.00	100.00	100.00	100.00	100.00
analyzed nutrients:					
Crude protein	20.85	20.96	21.02	21.07	21.13
M.E(Kcal/kg)	3043.42	3050.36	3051.09	3061.22	3069.61

Key: M.E= Metabolizable energy; FMKCM= Fermented mango kernel composite meal.

RESULTS AND DISCUSSION

The results obtained in this study showed no significant (P>0.05) difference in Red blood cell, White blood cell, MCV, MCH, and Monocytes. The Haemoglobin, Packed cell volume, MCHC, Platelets, Neutrophils, Eosinophils and Lymphocytes were significantly (P<0.05) different across treatments. The result for haemoglobin concentration ranged from 13.63-15.03g/dL. Haemoglobin was significantly (P<0.05) different across the treatment groups. The values were within the normal reference range (12.00-15.20g/dL) reported by Terry and Campbell (1988). This agrees with the findings of Kayode *et al*. (2013). Who reported significant difference in haemoglobin when broilers chickens were fed mono-culture fungal fermented mango kernel meal. But disagree with the assertion of Diarra *et al*. (2008) who reported no significant difference when broiler chickens were fed with sundried mango kernel. Haemoglobin is an iron containing compound found in the red blood cell, which transport oxygen around the body. Measuring the concentration of haemoglobin in the blood can help diagnose anaemia, a condition caused by a deficiency of haemoglobin. Red blood cell count in this study ranged from 4.44- 4.65x 10^{-12}/L. The red blood cells had no significant (P>0.05) difference across the treatment groups. The values were within the normal reference (3.50-5.50 x10^{-6}/L) by Terry and Campbell (1988). The result is in agreement with the findings of Diarra *et al*. (2011). Who fed broiler chicken with boiled mango kernel meal. But in contrast with the findings of Nse-Abasi *et al*. (2014) and Kayode *et al*. (2013) who reported significant (P<0.05) difference in red blood cell of broiler chicken fed different processed mango kernel meal and mono-culture fungal fermented mango kernel meal respectively. The red blood cell also known as erythrocytes specializes in transportation of oxygen to the presence of haemoglobin within the erythrocytes. The result for packed cell volume ranged from 41.00- 46.67%. The values were within the normal reference range (40.00- 46.90%) by Terry and Campbell (1988). The significant (P<0.05) difference observed in the packed cell volume agrees with the findings of Daudu *et al*. (2015) and Kayode *et al*. (2013) who fed broiler chicken with sundried mango kernel meal and mono-culture fungal fermented mango kernel meal respectively. But disagree with the findings of Zendesha (2017) and Diarra *et al*. (2010) who recorded no significant difference when broiler chickens were fed fermented mango kernel composite meal and boiled mango kernel meal. According to Campbell (1988) packed cell volumes of less than 37% indicate anaemia and one more 67% indicate dehydration. White blood cell count of Japanese quails ranged from 23.43- 28.27 x10^{9}/L. The white blood cell in this study showed no significant (P>0.05) difference across the treatment groups. The result were within the normal reference ranged (24.43-28.27 x10^{9}/L) reported by Ganti (2009). The trend or pattern observed with white blood cell simply suggests that the immune system of birds gradually increase with increased supplementation of FMKCM. The results correspond with the findings Diarra *et al*. (2010) and Zendesha (2017) who reported no significant (P>0.05) difference when broiler chicken were fed boiled mango kernel meal and FMKCM. But disagree with the findings of Daudu *et al*. (2015) and Kayode *et al*. (2013) who observed significant (P<0.05) different when broiler chickens were fed sundried and mono-culture fungal fermented mango kernel. Animal with low white blood cell count are exposed to high risk of disease infection, while those with high counts are capable of generating antibodies in the presence of phagocytosis and have higher degree of resistance to diseases (Soetan *et al*., 2013). The mean corpuscular volume (MCV) ranged from 128.07-134.00FL. There was not significant (P>0.05) difference in MCV across the treatment groups. The results were within the normal references range (90-140FL) by Rick (2004). The non significant (P>0.05) difference observed in this study agrees with the findings of Diarra *et al*. (2011) and Zendesha (2017) when broiler chicken were fed boiled mango kernel meal and FMKCM. However the result disagree with the findings of Daudu *et al*. (2015) who recorded significant (P<0.05) difference when broiler chickens were fed sundried mango kernel meal. Mean corpuscular volume is the expression of the average volume of individual erythrocytes. A low MCV may indicate iron deficiency, chronic disease and anaemia due to blood cell destruction or bone marrow disorder. A high MCV may indicate anaemia due to nutritional deficiencies, bone marrow abnormalities. The mean corpuscular haemoglobin (MCH) values ranged from 44.50- 46.60Pg. The MCH had no significant (P>0.05) difference across the treatment groups. The results were within the normal reference range (33- 47Pg) by Rick (2004). This agrees with the report of Diarra (2008) who fed broiler chicken with raw or sundried mango kernel meal. MCH is the average mass of haemoglobin per red blood cell in a sample of blood. MCH values diminish in hypochromic anaemia (Ganti, 2009). A low MCH signifies decreased production of haemoglobin. The result for mean corpuscular haemoglobin concentration (MCHC) ranged from 31.43-33.30g/dL. This result was within the normal reference ranged (26-35g/dL) by Rick (2004). MCHC is significantly (P<0.05) different across the treatment groups. This agrees with the finding of Kayode *et al.* (2013) who recorded significant (P<0.05) difference when broiler chickens were fed mono-culture fungal fermented mango kernel meal.

Table 2: Haematological Parameters of Laying Japanese Quails Fed Fermented Mango Kernel Composite Meal (FMKCM)

Parameters	(0%)	(10%)	(15%)	(20%)	(25%)	SEM	P-Value
Haemoglobin (g/dL)	13.93^{c}	15.03^{a}	14.97^{b}	13.63^{d}	13.97^{c}	11.86	0.03
Red blood cell ($x10^{12}$/L)	4.44	4.57	4.65	4.59	4.48	0.28	0.47
Packed cell volume (%)	44.67^{ab}	46.67^{a}	45.00^{ab}	41.00^{c}	42.00^{bc}	2.85	0.04
White blood cell ($x10^{9}$/L)	23.43	23.90	25.50	27.10	28.27	0.22	0.64
MCV (FL)	134.00	132.67	133.67	128.07	131.73	1.56	0.32
MCH (Pg)	45.30	44.50	46.60	44.90	45.47	0.71	0.58
MCHC (g/dL)	31.43^{b}	33.23^{a}	33.23^{a}	33.27^{a}	33.30^{a}	0.63	0.04
Platelet ($x10^{9}$/L)	305.00^{a}	298.67^{ab}	285.33^{bc}	271.33^{c}	285.67^{bc}	4.82	0.03
Neutrophil (%)	8.00^{a}	7.00^{b}	6.67^{c}	6.69^{c}	6.33^{d}	0.93	0.01
Eosinophils (%)	0.00^{e}	0.33^{d}	1.67^{a}	1.33^{c}	1.35^{b}	0.11	0.01
Monocyte (%)	1.67	1.67	5.00	10.33	8.33	0.28	0.32
Lymphocytes (%)	23.33^{c}	28.00^{b}	29.67^{ab}	30.33^{a}	24.00^{c}	1.21	0.00
Basophils (%)	0.00	0.00	0.00	0.00	0.00	0.00	-

Mean with different superscripts (a, b, c, d, e) within the same row differed significantly (P<0.05); SEM= Standard; error mean; MCV= Mean corpuscular volume; MCH= Mean corpuscular Haemoglobin; MCHC= Mean corpuscular; haemoglobin concentration.

The MCHC is a measure of the concentration of haemoglobin in a given volume packed red blood cell. It is reported as part of a standard complete blood count. A low MCHC signifies decreased production of haemoglobin. Platelets result ranged from 271.33- 305.00 $x10^{9}$/L. The results were within the normal reference range for healthy birds (140-450 $x10^{9}$/L) by Campbell (1988). The significant values observed correspond with the findings of Diarra *et al*. (2010) and Kayode *et al*. (2013) who fed broiler chicken with boiled mango kernel and mono-culture fungal fermented mango kernel meal respectively. But disagrees with the reports of Daudu *et al*., 2015 and Zendesha (2017) who reported no significant (P>0.05) difference across the treatments when broiler chicken were fed with sundried mango kernel meal and FMKCM. Platelets also called thrombocytes (blood clots cells) are components of blood whose function is to stop bleeding by clumping and clotting blood vessels. Platelets secrete antibodies that bind to foreign microorganisms in the body tissues and mediate their destruction. The result for Neutrophils ranged from 6.33- 8.00%. The values obtained in this study differed significantly (P<0.05) across the treatments. This result was within the normal reference range (3-10%) by Campbell (1988). The significant (P<0.05) difference observed in this study correspond with the findings of Daudu *et a*l. (2015) and Kayode *et al*. (2013) who observed significant (P<0.05) differences across the treatments when broiler chickens were fed diets containing sundried mango kernel meal and mono-culture fungal fermented mango kernel mea. But disagree with the assertion of (Zendesha, 2017; *Diarra et al*., 2008 and Diarra *et al*., 2011). Who reported a non significant (P>0.05) difference with broiler chicken fed FMKCM, sundried mango kernel meal and boiled mango kernel meal respectively. Neutrophils protect the body against disease and infection. A higher level of Neutrophils may results in a condition known as neutrophilia. The result for Eosinophils ranged from 0.00-1.67%. Eosinophils values in this study differed significantly (P<0.05) across the treatments. The results were within the normal reference ranged (0-7%) by Campbell (1988) and Ganti (2009). The significant difference observed in this study correspond with the findings of Diarra *et al*. (2011) who fed boiled mango kernel meal to broiler chicken. Eosinophils help protect the body against disease and infection, higher than normal level of Eosinophils can lead to a condition known as eosinophilia. Monocytes result ranged from 1.67-10.33%. The results were within the normal reference range of (2-12%) reported by Campbell (1988). The non significant (P>0.05) difference observed in this study corresponds with the findings of Diarra *et al*. (2010). But disagree with the findings of Kayode *et al*. (2013) who reported significant (P<0.05) difference when broiler chicken were fed with mono-culture fungal fermented mango kernel meal. Monocytes are effective in the direct destruction of pathogens and clean up of cellular debris from the sites of infection. An increased number of Monocytes in blood (monocytosis) occur in response to chronic infections, auto immune disorders and in certain cancers. A low number of Monocytes in the blood (monocytopenia) can be caused by anything that decreases the overall white blood cell count, such as blood stream infection and bone marrow disorder. The result for Lymphocytes ranged from 23.33- 30.33%. Lymphocytes was significantly (P<0.05) different across the treatment groups. The results were within the normal reference range (20-50%) by Campbell (1988). The significant difference observed was in line with the findings of Daudu *et al*. (2015) who reported significant (P<0.05) difference when broiler chicken were fed diets containing sundried mango kernel meal. According to Campbell (1988) an increase in lymphocytes is term lymphophilia which could be caused by chronic antigenic stimulation. A decrease is termed lymphopenia which could be caused by radiation, acute infection and toxins. The Basophils values in this study are 0% which is within the normal reference range to (0-2%) reported by Campbell (1988). According to these authors, an increase Basophils is term basophilia which could be caused by severe tissue damage, inflammation, parasitism, severe stress and feed restriction.

Conclusion

It is concluded that FMKCM could replace maize up to 25% without any deleterious effect, as all the haematological parameter of laying Japanese quails measured are within the normal reference range.

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