TECHNOLOGICAL PROPERTIES, NUTRITIONAL QUALITY OF MEAT AND BLOOD CELLS COUNT OF BROILER CHICKENS FED WITH CONVENTIONAL AND UNCONVENTIONAL DIETS.

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Abstract

The study aims to determine the impact of unconventional feedstuffs use (Azolla, Moringa, rice bran, fish and chicken viscera) on haematological and quality traits of broiler chicken meat. Therefore, 180 day old Marshal Broiler chicks were randomly divided into six dietary treatments of 30 chicks per treatment and reared in cage. Each treatment was replicated 3 times with 10 chicks per replicate and they were randomly all allocated to one conventional control diet (D0) and 5 experimental unconventional diets (D1, D2, D3, D4 and D5). Data on haematological and quality traits of broiler chickens were then collected and analysed. It appears that the red blood cell (RBC) counts didn’t vary significantly according to the dietary treatment and were ranged between 2.35-2.60 x 10⁶/L. Chicken fed unconventional diet recorded the highest RBC count while those of the control group fed with conventional diet had recorded the lowest value. Haemoglobin rates were similar and ranged between 8.20 and 9 g/dl. PCV values were not significantly affected by the dietary treatment (P > 0.05) and ranged between 26 and 28%. The MCHC, MCH and MCV values obtained were respectively 30.8 – 31.4 g/dl, 44.38 – 56.15Pg and 106.4 to 116.6 fl.

Technologically, the pH, an indicator of meat quality, differed between treatments in the current study (P < 0.05). The cooking loss of breast meat fed conventional diet was higher than those of unconventional diet. The drip loss and the water holding capacity were not affected by the diet (P > 0.05), and fluctuate respectively from 5.84 to 10.76 and from 40.44 to 48.79.

Nutritionally, the contents in dry matter, organic matter, protein, ash and fat were affected by the dietary treatment (P < 0.01). The highest
Introduction:-

Poultry meat and breast muscles in particular are characterized by high nutritive values. Consumers expect meat and meat products characterized not only by appropriate flavor values, high nutritive value, and low fat content but also by high concentrations of vitamins and minerals. The chemical composition of muscles is modified by genetic and environmental factors (Tougan et al., 2013a). Consumers express an increasing interest in food products which they perceive as having been produced with natural methods and being environment friendly, that assure a high nutritive value, are characterized by good taste and are manufactured with respect for bird’s welfare (Fanatico et al., 2007). The use of unconventional feed resources in poultry nutrition is one of the ways to overcome this feed crisis in the poultry industry. Alternative feed sources have proved valuable in supporting the performance of livestock and poultry at low cost of production (Babatunde and Oluyemi, 2000). Organic meat production undoubtedly reduces the risk of potential public health problems occurring by prohibiting the use of antibiotics, hormones and pesticides, which are suspected to have endocrine disrupting, carcinogenic, teratogenic, immunosuppressive and nervous effects (Lee et al., 2001). Haematological parameters are good indicators of the physiological status of animals (Adenkola et al., 2004; Adenkola et al., 2008) and changes are of value in assessing the response of animals to various physiological situations (Adenkola et al., 2008) and disease conditions. Haematological parameters have been evaluated in many different species of animals, including poultry, by many researchers. These parameters are known to change with many factors, one of which is nutrition (Amakiri et al., 2009). Dietary components affect the blood profile of healthy birds (Ihekwumere and Herbert 2012). It is often very difficult to assess the current health status of animals without detailed examination of blood (Amakiri et al., 2009). Examination of blood provides the opportunity to clinically investigate the presence of several metabolites and other constituents in the body and it plays a vital role in the physiological, nutritional and pathological status of the animal. It also helps to distinguish normal state from state of stress which can be nutritional (Aderemi, 2004). According to Togun and Oseni (2005) haematological studies have been found useful for disease prognosis and for the therapeutic and feed stress monitoring (Adamu et al., 2006) observed that nutrition had significant effect on haematological values like PCV, Hb and RBC (Togun et al., 2007) reported that when the haematological values fall within the normal range reported for the animal, it is an indication that diets not show any adverse effect on haematological parameters during the experimental period but when the values fall below the normal range, it is an indication of anaemia. Low values for haematological parameters as reported by Bawala et al. (2007) could be due to the harmful effects of high dietary contents. The objective of this research was to determine the impact of using unconventional diets (Azolla, Moringa, rice bran, fish and chicken viscera) on haematological and quality traits of poultry meat. Specifically, it is to:

1. Compare the blood cells counts of broiler chickens according to the dietary treatment;
2. Determine the technological nutritional quality of meat of broiler chickens fed with conventional and unconventional diets.

Materials and Methods:-

Area of study

The study was conducted simultaneously at the Laboratory of Research on Wet Zones of the University of Abomey-Calavi and the Laboratory of Avian and Zoo-Economical Research, Department of Animal Production at the Faculty of Agronomical Sciences/University of Abomey-Calavi located in Atlantic Department of Benin. Situated at latitude of 6° 27’ north and at a longitude of 2° 21’ east, the Commune of Abomey-Calavi covers an area of 650 km2 with a population of 307745 inhabitants (INSAE 2010). This area exhibits climatic conditions of sub-equatorial type, characterized by two rainy seasons with an uneven spatial and temporal distribution of rainfall: major (from April to July) and minor (from September to November). These two seasons are separated by a dry season. Average rainfall is close to 1200 mm per year. The monthly average temperatures vary between 27° and 31°C and the relative air humidity fluctuates between 65%, from January to March, and 97%, from June to July.
Birds and Diets:
For this study, 180 day old Marshal Broiler chicks were randomly divided into six dietary treatments of 30 chicks per treatment and reared in cage. Each treatment was replicated 3 times with 10 chicks per replicate and they were randomly allocated to one conventional control diet (D0) and 5 experimental unconventional diets (D1, D2, D3, D4 and D5). The experimental diets 1, 2, 3, and 4 contain Azolla, Moringa meals, rice bran with graded level of fish and chicken viscera (Table 1). The management of production system used was described by Tougan et al. (2013a).

Blood collection for analysis:
At the 7th day of the trial, blood was collected aseptically with a sterile syringe and needle from the wing vein of 6 birds of each dietary treatment. Before blood collection and slaughtering process, a feed withdrawal of 12 hours was observed in the group of intensively kept chickens to avoid excessive bleeding, microbial contamination and severe stress. Two millilitres of blood was collected through the wing vein from each bird, and was dispensed into clean bijou bottles containing anticoagulant EDTA (ethylene diamine tetracetic acid). The anticoagulated blood was used to determine: red blood cell (RBC), white blood cell (WBC) count, packet cell volume (PVC), haemoglobin (Hb) concentration and power of hydrogen (pH) out of which values, mean corpuscular haemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were calculated according to the method described by Aholou et al. (2016; 2017).

Slaughtering process and analytical
The slaughtering process used in the current study is described by Tougan et al. (2013b). After slaughtering, the cuts of breast were used to evaluate the technological properties (pH, drip loss, cooking loss and water holding capacity (WHC) in accordance with the method used by Tougan et al. (2013c) and chemical composition (moisture, protein, fat, and ash) of meat. The dry matter content, protein content, fat content, and ash content were determined as described by Tougan et al. (2013d). Those different parameters evaluated herein were expressed as the mean of the different values recorded from breast muscles.

Statistical Analysis
The statistical analyses were done using the software Statistical Analysis System (SAS 2006). The model used was a factorial ANOVA; means were compared using the Student-Newman-Keuls test. For all evaluations, differences between means were considered significant at the threshold of 5%.

Results:
Blood cells count and meat technological properties of broiler chickens by diet
The blood cells count and meat technological properties of broiler chickens are given by diet respectively in Table 2 and Table 3. Indeed, it appears that the red blood cell (RBC) counts didn’t vary significantly according to the dietary treatment and were ranged between 2.35-2.60 x 106/L. Chicken fed unconventional diet recorded the highest RBC count while those of the control group fed with conventional diet had recorded the lowest value. Haemoglobin rates were similar and ranged between 8.20 and 9 g/dl. PCV values were not significantly affected by the dietary treatment (P > 0.05) and ranged between 26 and 28%. The MCHC, MCH and MCV values obtained were respectively 30.8 – 31.4 g/dl, 44.38 – 56.15Pg and 106.4 to 116.6 fl.

Technologically, the pH, an indicator of meat quality, differed between treatments in the current study (P < 0.05). The cooking loss of breast meat fed conventional diet was higher than those of unconventional diet (P < 0.05). The drip loss and the water holding capacity were not affected by the diet (P > 0.05), and fluctuate respectively from 5.84 to 10.76 and from 40.44 to 48.79.

Variation of meat nutritional quality of broiler chickens by diet
The nutritional quality of the chicken meat varied according to the diet (Table 4). Overall, the contents in dry matter, organic matter, protein, ash and fat were affected by the dietary treatment (P <0.01). The highest dry matter content and fat content were recorded in the chicken fed with the diet D0 (P <0.01), while the most important content in ash and protein were found respectively in the diet D3 (P <0.01) and D4 (P <0.001).
Discussion:-
Variability of blood cells count and meat technological properties

The red blood cell (RBC) counts ranged between 2.35-2.60 x 106/L. This value is close to the normal value (2.0 x 106/L) reported by Aiello and Mays (1998) in chickens. Chicken fed unconventional diet recorded the highest RBC count while those of the control group fed with conventional diet had recorded the lowest value. No significant (P > 0.05) difference was found between different dietary treatments. RBC counts for the groups on unconventional diets did not show any statistical difference but were generally higher than value obtained for control diet group. These results are not in accordance with the result of Bamgbose et al. (2007) who reported an increased RBC count of turkey with diet made of wheat and maize.

Diets D2andD1 recorded respectively the highest (9g/dl) and lowest average Hb concentration (8.2 g/dl), although the difference was not significant (P > 0.05) (Table 2). Emenalom et al. (2004) observed that poor nutrient utilisation might result in variation in Hb values. It will appear also that the quality of unconventional and local protein was high enough to elicit elevated Hb. However, Hb values were generally within the range of 11.0 and 11.4g/dl reported for turkeys (Aiello and Mays, 1998; Bamgbose et al., 2007).

In the current study, the PCV values obtained were between 26 and 28%. PCV values, was not significantly affected by the dietary treatment. However, unconventional diets had lower PCV values (27.45%) than control diet (28.4%) although the difference was notsignificant. The MCHC, MCH and MCV values obtained were 30.8 – 31.4 g/dl, 44.38 – 56.15Pg and 106.4 to 116.6 fl, respectively. No significant difference was foundbetween MCHC, MCV and MCH values according to the treatment. Fluctuations in values for these parameters were observed in broilers (Talebi et al., 2005; Islam et al., 2004).

Technologically, the pH, an indicator of meat quality, differed between treatments in the current study. Breast pH values recorded herein were similar to those reported for Thai native chicken (5.77) at the same age (Jaturasithaet al., 2008), and to the findings of El Rammouzet al. (2004) on Label chicken (5.73) kept under conventional breeding conditions at the Poultry Research Center (INRA-Nouzilly) until the usual marketing age (12 weeks) and the results of Jehlet al. (2003) and Wattanachantet al. (2004) on slow growing broiler (5.66), while lower than values reported in the literature on thigh and breast meat of Label Rouge chickens bred under improved breeding system until 24 weeks old (Youssaoeet al., 2012) and the ultimate pH value of 6.1 found at 24 hours post-mortem reported by Raach-Moujahedet al.(2008) on tunisian local poultry raised in outdoor access. In normal ante mortem muscle, the pH is approximately 7.2. At the time of slaughter, oxygen and nutrients that are supplied by the way of the circulatory system are stopped. Glycogen, which supplies energy to the muscle, is metabolized postmortem in an anaerobic environment to lactic acid, which reduces muscle pH (Nissen and Young, 2006; Tougan et al., 2013a). The rate and the extent of pH decline will have a large influence on meat quality characteristics. Variation in muscle pH is likely to influence color and the ability of meat to hold water. In the conducted experiment, a higher pH24 value was assayed in the muscles of birds using unconventional diet. In the case of breast muscles, the pH value of 5.8 indicates the proper acidity and fits within the range of values corresponding to quality standards for non-defective meat. The rate of pH decrease depends on glycogen reserves in muscles. Only sufficiently high reserves assure the proper course of meat maturation and pH decrease (Ebreed et al., 2015). Many studies confirm the impact of growth rate of poultry on the final level of pH (Fernandez et al., 2001; Berri et al., 2005). Authors determined a higher level of glycogen reserves in muscles of slowly-growing birds, hence a lower post-mortem pH value was observed in these birds.

An important factor that may have adverse effects on sensory perception by consumers is cooking loss which indicates the quantity of meat juices lost during thermal treatment. The high value of cooking loss may intensify the sensation of a lack of juiciness or even dryness of meat, which significantly affects the overall sensory acceptability of meat. In the current study, the cooking loss value of breast of conventional diet was higher than thoseof unconventional diet. When looking at the effect of production system on cooking loss, meat from organically raised animals showing lower cooking losses than the conventional meat (Olsson et al., 2003).These cooking values were higher than those recorded by Ebreed et al., (2015). In addition, these authors emphasize that the discussed parameter is significantly affected by genotype. According to Fanaticoet al. (2005), breast muscles of slow growing birds are characterized by higher cooking loss than those of the fast-growing ones, which was also confirmed in a research by Debut et al. (2003). A significant physicochemical parameter is water ability to bind with meat. A high water holding capacity (WHC) value can positively influence meat juiciness.
The lack of significant differences in the discussed parameter shows that the different dietary treatments have a limited influence. This may result in the lack of differences in sensory assessment of meat from different rearing systems. Wang et al. (2009) observed a similar tendency as that reported in our study. They also did not confirm statistical significance of their results (P>0.05). Opposite results were described by Castellini et al. (2002) who noted a significantly lower WHC in muscles of chickens allowed to use free ranges. The sensory evaluation of meat is also directly influenced by tenderness.

**Variation of meat nutritional quality**
Overall, the contents in dry matter, organic matter, protein, ash and fat were affected by the dietary treatment. Our results on moisture, dry matter, fat and proteins contents in breast and thigh-drumstick meat are within the limits given by Demb and Cunningham (1980) and reported by Tougan et al. (2013d) in chicken meat. These authors reported that the water content of raw chicken meat varied from 60.4 to 75.4 %, the protein content from 17 to 23.3 %, the fat content from 1.0 to 17.4 % and the ash content from 0.7 % to 3.6 %, with averages of 71.1, 19.8 and 7.5 %, respectively, which left 1.6 % for ash. The values obtained for dry matter, protein and ash in this study are slightly lower than those recorded by Franco et al. (2012) who reported that water content, protein and ash value were 74.2%, 22.97% and 1.32% respectively in broiler strain Mos and 73.88%, 23.68% and 1.27% in broiler strain Sasso T-44; they are also weaker than those reported by Konrad and Gaal (2009) on the Hungarian Yellow chickens and Bogosavljevic-Boskovic et al. (2010) on broilers.

Many variables such as breed, feed, age, system of production, sex, processing and type of meat that can affect nutritional values may account for a large part of the wide range in data (Holcman et al., 2003).

**Conclusion:**
Overall, unconventional feedstuffs such as Azolla, Moringa, rice bran, fish and chicken viscera improve meat nutritional quality trait in broiler chicken without affecting their blood cells count. Further study is needed for the evaluation of their production cost.

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**Table 1:** composition of dietary treatment

| Foodstuffs (%) | Control diet | Experimental diets |
|----------------|--------------|---------------------|
|                | D0           | D1      | D2    | D3    | D4    |
| Maize          | 61.3         | 0       | 0     | 0     | 0     |
| Wheat bran     | 3            | 0       | 0     | 0     | 0     |
| Soya cake      | 15           | 0       | 0     | 0     | 0     |
| Cotton seed cake | 15       | 0       | 0     | 0     | 0     |
| Shell          | 1            | 0       | 0     | 0     | 0     |
| Idafix         | 0.2          | 0       | 0     | 0     | 0     |
| Premix Junior 2.5 (methionine, lysine) | 2.5 | 0 | 0 | 0 | 0 |
| Moringameal    | 0            | 42.7    | 42.7  | 42.7  | 42.7  |
| Azollameal     | 0            | 45      | 45    | 45    | 45    |
| Rice bran      | 0            | 5       | 5     | 5     | 5     |
| Chicken viscera | 0       | 5       | 3.5   | 1.5   | 0     |
| Fish viscera   | 0            | 0       | 1.5   | 3.5   | 5     |
| Palm oil       | 2            | 2       | 2     | 2     | 2     |
| Salt           | 0            | 0.3     | 0.3   | 0.3   | 0.3   |
| Total          | 100          | 100     | 100   | 100   | 100   |
| Dry matter content (%) | 89 | 86 | 87 | 87 | 88 |
| Crude protein content (%) | 19.5 | 22.5 | 23 | 23.5 | 23.8 |
| Crude fiber content (%) | 4 | 22 | 22 | 22 | 22 |
| Metabolizable energy (Kcal/Kg of DM) | 2848 | 2453 | 2484 | 2525 | 2556 |
| Cost/ Kg diet | 300          | 180     | 185   | 190   | 195   |
Table 2: Variation of blood cells count by diet

| Variables                              | D0  | SE  | D1  | SE  | D2  | SE  | D3  | SE  | D4  | SE  | Diet effect |
|----------------------------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-------------|
|                                        | Mean|     | Mean|     | Mean|     | Mean|     | Mean|     |             |
| Red blood cell (10^7/L)                 | 2.45| 0.05| 2.38| 0.09| 2.55| 0.09| 2.35| 0.15| 2.60| 0.06| NS          |
| Haemoglobin (g/dL)                      | 8.86| 0.27| 8.2 | 0.38| 9.0 | 0.43| 8.4 | 0.25| 8.6 | 0.24| NS          |
| Packet cell volume (%)                 | 28.4| 0.68| 26.4| 1.21| 28.4| 1.03| 27.2| 0.97| 27.8| 0.58| NS          |
| Mean corpuscular volume (IL)           | 116.0| 1.41| 110 | 2.62| 111.2| 1.53| 116.6| 4.31| 106.4| 1.94| NS          |
| Mean corpuscular haemoglobin rate (pg/cell) | 36.2| 0.58| 34.4| 0.93| 35.0| 0.63| 36.0| 1.38| 33.0| 0.71| NS          |
| Mean corpuscular haemoglobin concentration (g/dL) | 31.2| 0.20| 30.8| 0.20| 31.4| 0.40| 30.8| 0.20| 30.8| 0.20| NS          |

SE: Standard Error, NS: P>0.05

Table 3: Variability of the technological quality traits of chicken according to the diet

| Variables                              | D0  | SE  | D1  | SE  | D2  | SE  | D3  | SE  | D4  | SE  | Diet effect |
|----------------------------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-------------|
|                                        | Mean|     | Mean|     | Mean|     | Mean|     | Mean|     |             |
| pH 45min                               | 6.39a| 0.01| 6.27b| 0.02| 6.46c| 0.01| 6.51d| 0.01| 6.52d| 0.01| ***         |
| pH 24h                                 | 5.71a| 0.05| 5.70b| 0.07| 5.71c| 0.07| 5.89d| 0.04| 5.80e| 0.06| NS          |
| Cooking loss                           | 38.03a| 1.74| 30.52b| 3.82| 35.76ac| 3.80| 34.60c| 2.84| 37.09a| 1.13| *           |
| Driploss                               | 10.03a| 2.98| 10.13b| 1.67| 7.58c| 2.52| 5.84d| 0.94| 7.48e| 1.69| NS          |
| WHC                                    | 48.79a| 4.36| 40.65b| 4.95| 43.35c| 4.03| 40.44d| 3.36| 44.57e| 2.33| NS          |

WHC: Water holding capacity, SE: Standard Error, ***: P<0.001. The means of the same line followed by different letters differ significantly with the threshold of 5%.

Table 4: Variability of the nutritional quality of chicken according to the diet

| Variables                              | D0  | D1  | D2  | D3  | D4  | RSD | Diet effect |
|----------------------------------------|-----|-----|-----|-----|-----|-----|-------------|
|                                        | Mean|     | Mean|     | Mean|     |             |
| Dry matter (g/100g of FM)              | 23.47b| 22.55c| 23.7a| 23.79a| 23.5ab| 0.78| **          |
| Ash(g/100g of FM)                      | 1.04d| 1.68c| 1.75c| 2.36a| 2.00b| 0.41| **          |
| Crude protein (g/100g of FM)           | 20.5a| 21.45b| 21.8b| 22.0c| 22.5d| 0.8 | ***         |
| Fat (g/100g of FM)                     | 1.21a| 0.42b| 0.55b| 0.85c| 0.95c| 0.44| **          |

FM: Fresh matter; **: P<0.01; ***: P<0.001. The means of the same line followed by different letters differ significantly with the threshold of 5%. RSD: Residual Standard Deviation.

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