# BLOOD PICTURES OF PULLETS TREATED WITH METHANOLIC AND AQUEOUS EXTRACT OF CUCUMIS METULIFERUS FRUITS AGAINST NEWCASTLE DISEASE KUDU 113 STRAIN

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## **ABSTRACT**

The study was conducted to determine the effect of aqueous and methanolic extracts of Cucumis metuliferus fruit on blood pictures of pullets to determine the level of prevention and/or cure of Newcastle disease (NCD) in pullets. Ninety (90) pullets were used for the experiment. The pullets were divided into 9 groups of 10 birds per group, 4 prophylactic, 4 curative and one control group. For prophylactic groups aqueous extract of the fruit at concentration of 2.5g and 5.0g per litre of water and methanolic extract of the fruit at 75 and 50mg/l were given to the pullets ad-libitum for two weeks and then challenged with Newcastle disease virus kudu 113 strain via intra ocular route. For curative groups the pullets were challenged with the virus and then treated with the aqueous extracts of the fruits at the same concentrations. The results showed that methanolic extract of C. metuliferus at concentration of 75mg/l showed superior prophylactic ability recording higher Packed cell volume (PCV), Red blood cell (RBC) count as well as higher White blood cell (WBC) activity with a statistically significant (p<0.05) difference when compared with control and other groups and also exhibited very good promise as a cure as it recorded better indices than all groups. The results showed proof that C. metuliferus does improve haematologic parameters in pullets and it was thus recommended that methanolic extract of C. metuliferus at 75mg/l can be used for the control of NCD to farmers and that further studies be conducted to isolate the active ingredients of C. metuliferus responsible for these findings.

**Key words** – Newcastle disease, *Cucumis metuliferus*, pullets, Methanolic and Aqueous extracts.

# INTRODUCTION

Poultry production is the fastest growing segments of the agricultural subsector in Nigeria due to the increased demand of animal protein in Nigeria with the ever growing population (Salami *et al.*, 1989). However, one of the constraints to the development of the industry is outbreak of diseases (Salami *et al.*, 1989). Plants have been used for various purposes since prehistoric times (Lawrence and Bennett, 1995; Evans, 2009) and medicinal herbs are being increasingly studied by researchers (Sinclair, 1998). More than 80% of the world's populations rely on traditional medicine for their primary healthcare with majority being plants or their active principles (Gupta *et al.*, 2005).

Cucumis metuliferus, locally called 'gautar kaji' in hausa language, belongs to the family Cucurbitaceae and is a monoecious, climbing, annual herb that can be grown practically anywhere, provided the season is warm (Benzioni et al., 1993). It is commonly known as African horned cucumber, melano, Jelly melon, and kiwano. It is reported that the fruits and seeds of Cucumis metuliferus are eaten raw as supplements by local populations of Africa (Bruecher, 1977; Keith and Renew, 1975). Reports have also shown that the seeds can be ground into fine flour, made into an

emulsion with water, and then eaten to expel parasites from the body (Chiej, 1984). Newcastle disease still remains a serious economic challenge to all segments of the poultry industry because of its high morbidity and mortality rates (Alexander, 2003). The only control method so far still remains vaccination, which does not confer 100% immunity in all vaccinated birds. It has also been reported that vaccination itself may result in the disease and also reduces growth in vaccinated birds (Alexander, 2003; Alexander, 2003; Burridge *et al.*, 1975). Investigations show that local farmers in the study area use *Cucumis metuliferus* fruits to treat their birds for various diseases including Newcastle disease by immersing the whole fruits in drinking water of birds (personal communication, 2017). This information further fueled by the need to identify a cheaper and more accessible means of prevention and treatment of Newcastle disease gave rise to this research work. This study is aimed at testing the effect of methanolic and aqueous extract of *Cucumis metuliferus* fruit as a cure and/or prevention aginst Newcastle disease.

## MATERIALS AND METHODS

# Study area

The study was conducted at the poultry unit of the Department of Animal Science farm, Federal University Dutsinma, Katsina State, Nigeria. Dutsinma lies within the coordinates of latitude 12.45°N and longitude 7.49°E and also on the altitude of 605m above the sea level (Isah, 2009).

# Study design

The research was carried out using a Completely Randomized Design with 9 treatments, each treatment/group were made up of 5 pullets. Day old pullets were sourced from reputable a hatchery and brooded under strict biosecurity measures to prevent entry of pathogens. The birds were vaccinated against Infectious bursal disease at day seven of the arrival but not against Newcastle disease. They were fed with chick-mash. Experimental groups consisted of Curative, Prophylactic and Control groups. Aqueous solutions of Cucumis metuliferus was made at 2.5grams per liter of water and 5grams per liter of water. Methanolic extract was gotten using a soxylet extractor. The extract was given in water at a concentration of 75 and 50 grams per liter of drinking water. The curative groups were T1 and T2 (administered 2.5g/l and 5g/l aqueous extract respectively) and T3 and T4 (administered 50g/l and 75g/l methanolic extract respectively). These groups were initially challenged with the Newcastle disease virus Kudu 113 strain at 8weeks and treated as indicated depending on the groups ad libitum in drinking water as soon as clinical signs were observed. The prophylactic groups were T5 and T6 (administered 2.5g/l and 5g/l aqueous extract respectively) and T7 and T8 (administered 50g/l and 75g/l methanolic extract respectively). These groups were treated with indicated treatments ad-libitum in drinking water for 2weeks and then subsequently challenged with Newcastle disease virus Kudu 113 strain to observe if the treatments would prevent or reduce the intensity of infection. The control group was challenged with the Newcastle disease virus Kudu 113 strain but was not treated with any extracts at 8 weeks of age.

# Laboratory analysis

Blood was collected via wing vein of birds at fourth week of the experiment.

# Data analysis

Data obtained from the experiment was presented in table as Staandard Errorss of Mean (SEM). Numerical variables were subjected to One Way ANOVA (analysis of variance) using Statistical Package for Social Sciences analysis software SPSS version 20.0. P values of <0.05 were considered significant.

#### RESULTS AND DISCUSSION

Groups T4 and T8 recorded the highest PCVs (36.67; P<0.033 and 44.00; P<0.013), total white blood cell count (16.00; P<0.041 and 16.33; P<0.035) as well as total red blood cell counts (5.73 and 7.87; P<0.033) for prophylactic and curative treatments respectively (Tables 6 and 7).

Table 6: Blood Picture/Heamatology of Pullets Treated with *Cucumis metuliferus Fruit Extracts* (Curative Treatments)

| Parameters         | T1                 | T2                 | T3                 | T4                 | Control            | SEM     |
|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|---------|
| PCV                | $31.00^{a}$        | 24.33 <sup>b</sup> | 25.33 <sup>b</sup> | 36.67 <sup>a</sup> | 16.00 <sup>c</sup> | 2.62926 |
| HGB (g/dl)         | 10.20 a            | $8.07^{a}$         | $8.43^{a}$         | 5.37 <sup>b</sup>  | 5.53 <sup>b</sup>  | 0.81666 |
| TP                 | $4.73^{a}$         | 3.53 <sup>b</sup>  | $4.87^{\rm a}$     | 5.27 <sup>a</sup>  | $3.87^{\rm b}$     | 0.15333 |
| TWBC x $10^9/l$    | 14.27 <sup>a</sup> | 14.33 <sup>a</sup> | 11.67 <sup>b</sup> | 16.00°a            | 11.13 <sup>b</sup> | 0.85076 |
| TRBC x $10^{12}/1$ | 5.23 <sup>a</sup>  | 4.23 <sup>a</sup>  | 4.33 <sup>a</sup>  | 5.73 <sup>a</sup>  | $3.27^{\rm b}$     | 0.37389 |
| HETERO (%)         | 13 <sup>a</sup>    | 9 <sup>b</sup>     | 8 <sup>b</sup>     | 9 <sup>a</sup>     | 5 <sup>c</sup>     | 1.03494 |
| LYMPHO (%)         | 82 <sup>a</sup>    | 89 <sup>a</sup>    | 90 <sup>a</sup>    | 86 <sup>a</sup>    | 95 <sup>a</sup>    | 1.47745 |
| MONO (%)           | 0                  | 2                  | 1                  | 0                  | 0                  | 0.39841 |
| EOSINO (%)         | 0                  | 0                  | 0                  | 4                  | 0                  | 0.23231 |
| BASO (%)           | 0                  | 0                  | 0                  | 0                  | 0                  | 0.00000 |
| BAND (%)           | 3                  | 0                  | 1                  | 1                  | 0                  | 0.40237 |

Means with the same letter are not significantly different (P>0.05).

Table 7: Blood Picture/Heamatology of Pullets Treated with *Cucumis metuliferus Fruit Extracts* (Prophylactic Treatments)

| Parameters         | T5                 | T6                 | T7                 | T8                 | Control         | SEM     |
|--------------------|--------------------|--------------------|--------------------|--------------------|-----------------|---------|
| PCV                | 24.33 a            | 25.33 <sup>a</sup> | 37.00°a            | 44.00 <sup>b</sup> | 16.00           | 1.11469 |
| HGB (g/dl)         | 7.97 <sup>a</sup>  | 8.40 <sup>a</sup>  | 13.67 <sup>b</sup> | 12.53 <sup>a</sup> | 5.53            | 0.35017 |
| TP                 | 6.53 <sup>a</sup>  | 4.80 <sup>b</sup>  | 4.33 <sup>b</sup>  | 6.67 <sup>a</sup>  | 3.87            | 0.32403 |
| TWBC x $10^9/l$    | 14.40 <sup>a</sup> | 9.87 <sup>b</sup>  | 11.53 <sup>a</sup> | 16.33 <sup>c</sup> | 10.13           | 1.02003 |
| TRBC x $10^{12}/1$ | 3.97 <sup>a</sup>  | $4.07^{a}$         | 7.37 <sup>b</sup>  | 7.87 <sup>b</sup>  | $3.27^{a}$      | 0.12794 |
| HETERO (%)         | 14 <sup>a</sup>    | 11 <sup>b</sup>    | 9 <sup>b</sup>     | 11 <sup>b</sup>    | 5 <sup>c</sup>  | 1.02291 |
| LYMPHO (%)         | 84 <sup>a</sup>    | 85 <sup>a</sup>    | 89 <sup>a</sup>    | 85 <sup>a</sup>    | 95 <sup>a</sup> | 1.37737 |
| MONO (%)           | 2                  | 3                  | 0                  | 3                  | 0               | 0.47676 |
| EOSINO (%)         | 0                  | 0                  | 2                  | 0                  | 0               | 0.23503 |
| BASO (%)           | 0                  | 0                  | 0                  | 0                  | 0               | 0.00000 |
| BAND (%)           | 0                  | 1                  | 0                  | 1                  | 0               | 0.16330 |

Means with the same letter are not significantly different (P>0.05).

Newcastle disease has been reported to be one of the major diseases bothering poultry with its high cost of management driving up the cost of pullet production (Ibrahim and Tanya, 2001; Bukar-Kolo *et al.*, 2006). The alleviated haematological parameters seen could be as a result of the alkaloids present in *Cucumis metuliferus* which has proven to be beneficial for various ailments (Moore and Pizza, 1992). The result highlighted that the plant *C. metuliferus* increases the level of chicken PCV. The higher white blood cell count seen from blood pictures for group T4 and T8 indicated higher white blood cell activity and thus higher chances of the immune system fighting off the infection when compared with other groups. This could translate to the lower clinical signs and mortality rates observed in these groups. Usman (2014) carried out a study which elucidated that the fruit of C.

metuliferus was shown to increase the values of blood parameters: packed cell volume, haemoglobin, red blood cell and white blood cell counts. This is in agreement with the work of Wannang *et al.* (2007), that the aqueous extract of *C. metuliferus* significantly increased the level of PCV in rats when given at 1000 mg/kg. An increase in the PCV value of rabbits treated with *C. metuliferus* against trypanosomiasis was also reported (Abubakar *et al.*, 2011). Jimam *et al.* (2012) carried out a study using methanolic extract of *C. metuliferus* and observed that birds treated with 200, 400 and 600 mg/kg of the extract exhibited significant (p<0.05) increase in PCV, Hb and RBC count on day 14 and 21 after treatment. Similarly, a significant (p<0.05) increase in RBC count was observed in birds treated with the extract at 400 and 600 mg/kg after 7 days post treatment (Jimam *et al.*, 2012). Usman *et al.* (2018) in their study also showed that *C. metuliferus* may be used as haematinic as well as in the treatment of malnutrition. The presence of iron in the fruit may have stimulated the production of RBC and this therefore confirmed the folkloric use of *C. metuliferus* as a haematinic agent (Usman *et al.*, 2018).

#### CONCLUSION AND RECOMMENDATION

In conclusion, the results showed proof that *C. metuliferus* does improve heamatologic parameters in pullets and it was thus recommended that methanolic extract of *C. metuliferus* at 75mg/l can be recommended for the control of NCD to farmers and that further studies be conducted to isolate the active ingredientsof *C. metuliferus* responsible for these findings.

## REFERENCES

Abubakar A, Iliyasu B, Ojiegbu FN, Igweh AC, Shamaki BU, Dung EC, et al. Evaluation of the anti trypanosomal activity of *Cucumis metuliferus* pulp extract in rabbits. *Journal of Medicinal Plants Research* 2011; 5(11):2136-2142.

Alexander, D.J. (2003), Newcastle disease and other avian paramyxovirida infections. In: Diseases of Poultry, Saif, Y.M., Barnes, H. J., Glisson, Jr., Fadly AM., McDougal, L.R. and Swayne, D.E., 11th Ed, (Jordan, F. T. W.M., Patttison, M. Eds.). pp. 64-87.

Alh. Bishir Almu (2017). Personal communication, 20-09-2017, 10:30am

Evans W.C. (2009) Trease and Evans Pharmacognosy. Elsevier Health Sciences. pp125

Gupta, M.P, Solís, P.N., Calderón, A.I., Guinneau-Sinclair, F., Correa, M., Galdames, C., Guerra, C., Espinosa, A., Alvenda, G.I., Robles, G. and Ocampo, R. (2005). Medical ethnobotany of the Teribes of Bocas del Toro, Panama. *Journal of Ethnopharmacoogy*. 96:389-401.

Ibrahim, U. I. and Tanya, S. N. (2001): Prevalence of antibodies infectious bursal disease (illD) in village chickens in Sahel Zone of Nigeria. *Bulletin of Animal Health production Africa*, 49: 150-152.

Isah, 2009. Combating water scarcity in Katsina. Retrieved 20 may, 2010 from https,//www.wikipedia.org/wiki/Dutsinma.

Lawrence, D.R. and Bennett, P.N., (1995). Clinical Pharmacology ELBS with Churchill Livingston, Edinburgh, p. 686.

Moore, P.S. and Pizza, C. (1992). Observations on the inhibition of HIV-1 reverse transcriptase by catechins. *Biochemical Journal*. 288:717-719.

Salami, J.O., Egbulett, B.N., Kwaga, J.K.P., Yusufu, H.I. and Abdu, P.A. (1989). Disease diagnosed in poultry in Kaduna State, Nigeria. *Bulletin of Animal Health and Production in Africa*, 18: 123-125.

Sinclair, S. (1998). Chinese herbs: a clinical review of Astragalus, Ligusticum, and Schizandrae. *Alternative Medicine Review*. 3:338–344.

Usman J.G., Sodipo, O.A., Kwaghe, A.V., Wampana B., Umaru N.J.H. and Sandabe U.K. (2018) Effects of crude methanol extract of the fruit of Cucumis metuliferus (Cucurbitaceae) on some haematological parameters in Cockerels. *The Journal of Phytopharmacology*, 7(2): 106-110

Usman, J.G. (2014). Phytochemical Screening and Antibacterial Effects of Cucumis Metuliferus (Jelly Melon) E. Mey. Ex Naudin (Cucurbitaceae) Fruits against Fowl Typhoid. A Dissertation Submitted to the School of Postgraduate Studies, University of Maiduguri, In Partial Fulfilment of the Requirement for the Degree of Master of Science in Pharmacology. 155pp.

Wannang, N., Jimam, N. Simeon, O. Dapar, M. Gyang, S. and Aguiyi, J. (2010). Effects of Cucumis metuliferus (Cucurbitaceae) fruits on enzymes and haematological parameters in albino rats. *African Journal of Biotechnology* 6 (22): 6-10