



**ANTI-PLASMODIAL PROPERTIES OF METHANOLIC EXTRACT OF *MUSCA
DOMESTICA* MAGGOT ON *P. BERGHEI* – INFECTED MICE**

SHITTU OK^{1*}, OLAYEMI IK², OMALU ICJ² AND ADENIYI AK²

1: Trypanosomosis Research Unit, Department of Biochemistry, Federal University of
Technology, PMB65, Minna, Nigeria

2: Department of Biology, Federal University of Technology, P. M. B. 65, Minna, Nigeria

***Corresponding Author: E Mail: toscue@yahoo.com; toscueyusuf@gmail.com; Tel: +234-
803-388-3658**

ABSTRACT

The antiplasmodial activity of crude methanolic extract of *Musca domestica* (house fly) maggot was investigated in *Plasmodium berghei* infected mice. Fifteen albino mice was intraperitoneally infected with chloroquine sensitive *Plasmodium berghei* strain and divided into three groups. Group I was set up as negative control of 0.2ml normal saline/kg body weight, group II as 5mg chloroquine/kg body weight and group III as 600mg of the extract/kg body weight. The result shows that methanolic extract of *Musca domestica* suppress the level of parasitaemia and there was no significant difference ($p < 0.05$) in the packed cell volume (PCV) of all the groups at Day 0, while at Day 4 there was significant increase ($p < 0.05$) in the packed cell volume (PCV) of the negative control compared to other groups. Whereas at Day 7 of the experiment, there was significant increase ($p < 0.05$) in the packed cell volume (PCV) of the negative control and 5mg received chloroquine compared to group III (600mg of the extract/kg body weight). The crude methanolic extract of *Musca domestica* also show longer mouse survival period relative to the negative control, with mean survival time of 34.00 ± 1.25 for the tested dose of 600mg/kg body weight and 44.00 ± 1.38 for Chloroquine compared to of negative control. This work revealed that methanolic extract of *Musca domestica* maggot has antiplasmodial activity. Therefore, this

shows the significance of insects and their products for the development of new drug for treatment against malarial and other infectious disease.

Keywords: Malaria, *Plasmodium berghei*, House Fly Maggot, Antiplasmodial

INTRODUCTION

Malaria is an important disease in sub-sahara Africa and is number one public health problem in Nigeria, where the disease is hyper-endemic and occurs in all zones [1]. In Nigeria, malaria kills over 300,000 people annually [2], account for 30% childhood mortality [3] 40% public health expenditure and 50% Outpatient Department (OPD) hospital attendance [4]. The disease impacts adversely on the productivity of all sectors of the country, affecting its rate of economic growth and level of development. According to [5], poor families in Nigeria spend up to 25% of their annual income on malaria treatment and prevention. It has also been reported that the cost of malaria treatment in Nigeria is estimated to be 1% of the country Gross National Product [6]. Despite much research on malaria drugs breakthrough remain elusive [7]. Efforts on drug discovery have largely been dominated by plants and two main groups of modern antimalarial drugs (artemisinin and quinine derivatives) have been developed. These antimalarial drugs were derived from plants

and are still effective in treating malaria [8], but they are relatively expensive for the rural dwellers that are predominantly at risk of the disease. Studies have shown that insects and their products also play important roles in folk medicine among native tribes in many part of the world. *Musca domestica* (House fly) is a major domestic medical and veterinary pest. The larva is also called maggot has been used clinically to cure malnutritional stagnation, decubial necrosis, osteomyelitis, ecthyma and lip boil [9-11]. The medical use of live maggots (fly larvae) for cleaning non-healing wounds refers to as Maggot Debridement Therapy (MDT) has been dated to 19th century. Medicinal maggots have been reported to perform antibacterial, immunoactive, antiviral and antitumor action [12-14]. However, the main components are been speculated to be proteins and peptides [15].

Therefore, the objective of this work was to investigate the anti-plasmodial activity of the crude methanolic extract of *Musca domestica* (house fly) maggot.

MATERIALS AND METHODS

Collection of Maggot

Musca domestica (House fly) maggots were collected from poultry dung in Minna, Niger state, Nigeria.

Animals

Swiss albino mice weighing between 18-24g were obtained from Animal House, Department of Pharmacology and Clinical Pharmacy, Ahmadu Bello University, Zaria, Kaduna state. The mice were housed in plastic cages and maintained under standard laboratory conditions with free access to rat pellets and tap water *ad-libitum*.

Parasites

A chloroquine-sensitive strain of *Plasmodium berghei* (NK-65) was obtained from the National Institute for Pharmaceutical Research and Development (NIPRD), Idu, Abuja, Nigeria and maintained by re-infestation via intraperitoneal with infected blood suspension (0.2 ml) containing about 1×10^7 suspension of *P. berghei* parasitized red blood cells.

Extract Preparation

Matured House flies were allowed to lay on the poultry dung and developed to fourth instar stage and later collected. They were washed and killed in salt water. These were dried in the shade at room temperature until constant weight was attained and pulverized to powder

using an electric blender. 200 g of the powder was percolated in 1600ml of absolute methanol and kept in the shade for 48 hours after which it was filtered. The filtrate was collected in a beaker, exposed to air and allowed to evaporate at room temperature to yield the extract concentrate [16].

Antiplasmodial Studies

Curative Test

Fifteen mice were inoculated as described above and left untreated. Three days later, the mice were randomized into three groups of five mice each. Group I mice were given normal saline/kg body weight intraperitoneally. Groups II were given 600 mg extract/kg body weight intraperitoneally, while group III mice received 5mg chloroquine /kg body weight intraperitoneally daily for 5 days. On each day, about drops of blood were collected from the tail of each mouse smeared onto a microscope slide to make a thin film, stained with 10% Giemsa stain and examined microscopically to monitor the parasitaemia level.

Mean Survival Period Test

The mean survival time (MST) for each group was determined by finding the mean survival time (days) of the mice (post-inoculation) in each group over a period of 29 days (D₀–D₂₈) [17]. The thin blood smear of the mice that survived beyond 28 days was also monitored.

Estimation of Packed Cell Volume (PCV)

Packed cell volume (PCV) was measured before infection (0 day), 72 hrs post infection (day 4) and four days after confirmation of infection (7 day) and was determined by a micro-method, using a Hawksley microhaematocrit centrifuge.

Statistical Analysis

Values are expressed as mean \pm SEM. The data were statistically analyzed using one-way analysis of variance (ANOVA) and Duncan Multiple Range Test [18]. Data from the test groups were compared with their respective controls and differences at $P < 0.05$ were considered significant.

RESULTS AND DISCUSSION

A number of studies have reported that *Musca domestica* maggot has therapeutic properties. *Musca domestica* has been reported to have anti-bacterial and immunoactive substance such as prophenol oxidase, antibacterial protein/peptide, lysozyme and some other secretion [19-21].

The result from **Figure 1** shows that the average daily parasitaemia for both chloroquine disphosphate treated and methanolic extract of *Musca domestica* maggot was significantly reduced when compared with negative control over the period of the experiment. This implies that the extract at 600mg/kg bw have positive activity

against *P. berghei* infection and the insignificant progressive reduction in parasitaemia of both chloroquine disphosphate treated and methanolic extract of *Musca domestica* maggot is an attribute that shows the potential of the extract as an antiplasmodial drug. **Figure 2** shows the changes in percentage of packed cell volume (PCV) for day 0, 4 and 7 of the experiment. At day zero and four, there was no significant difference ($p < 0.05$) in percentage PCV for all the groups. Whereas at day four (7) there was significant decrease ($p < 0.05$) in percentage PCV of the negative control and the methanolic extract of *Musca domestica* maggot when compared with chloroquine disphosphate treated. Anaemia is a fairly common problem encountered in malaria. The haemolysis may be due to the growing parasite consumes and degrades the intracellular proteins which are mainly hemoglobin [22]. These decreases however were considerably reversed in the infected extract-treated and infected chloroquine-treated groups on day 7 post-infection. The mean survival result shows extension of life span beyond 28 days for infected extract-treated and infected chloroquine-treated groups. While negative control mean survival period was 8.3 days (**Table 1**).

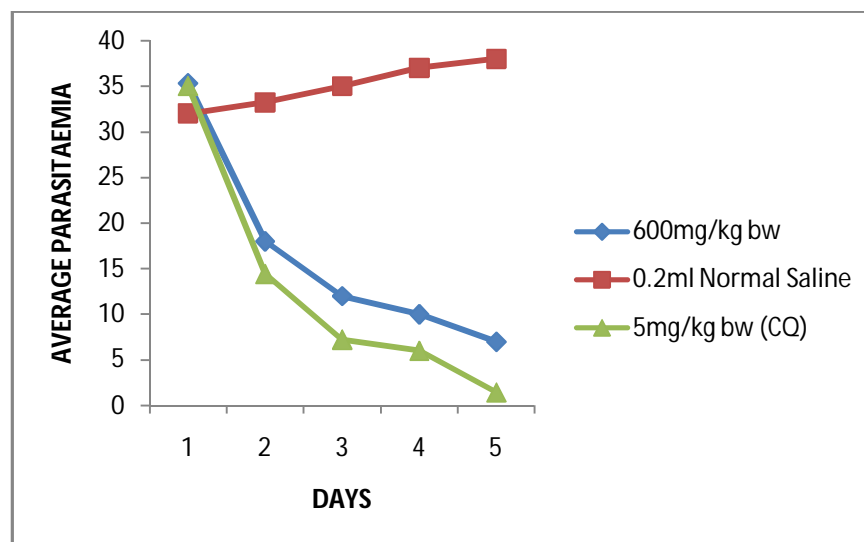


Figure 1: In Vivo Antiplasmodial Activity of Methanolic Crude Extract of House Fly Maggot Against *P. berghei* infected mice and Each Point is an Average Count From Five Mice

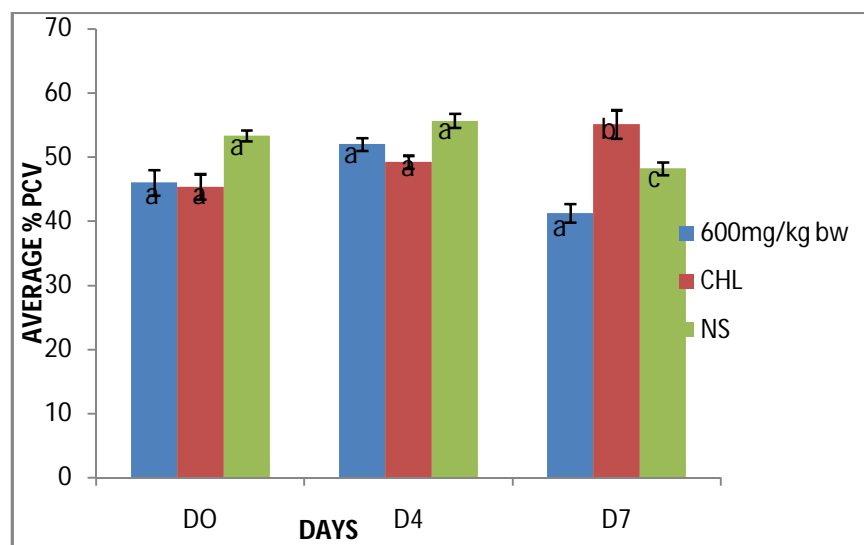


Figure 2: Changes in Pack Cell Volume (PCV) of Methanolic Crude Extract of House Fly Maggot Against *P. berghei* infected Mice and Each Value is an Average of Five Determinations + SEM. Values with Different Superscript are Significantly Different

NOTE: Day0: Before Infection; Day4: 72 Hours After Infection; Day7: 4 Days After Confirmation of Infection

Table 1: Mean Survival Period of Mice Treated with Crude Extract of House Fly Maggot

| S. No. | GROUPS | SURVIVAL DAYS |
|--------|--------------------------|---------------|
| 1. | Normal saline (NL) 0.2ml | 8.3 ± 0.50 |
| 2. | Chloroquine (5mg/kg) | 44 ± 1.38 |
| 3. | Extract (6005mg/kg) | 34 ± 1.25 |

NOTE: Each Result is an Average Count from Five Mice

CONCLUSION

We conclude that crude methanolic extract of *Musca domestica* maggot (600mg/kg body weight) has a potential in the development of active antiplasmodial substance that do not acquire resistance. We further suggest from the results in this work that crude methanolic extract of *Musca domestica* maggot has ameliorates the effects on symptoms of *P. berghei* infection in mice.

REFERENCES

- [1] Salako L, Malaria-the unending saga, In: Coping with treatment failures in malaria, Mayer and Baker, 1997, 13-25.
- [2] USAID, Health: USAID's Malaria Program, 2005.
- [3] Olukosi YA, Iwalokun BA, Magbagbeola OA, Akinwande AL, Adewale TA, Agomo PU and Awololo TS, Pattern of rural-urban acquisition pfert T76 allele among Nigerian children with acute uncomplicated Plasmodium falciparum malaria, Afri. J. Biotech., 4 (4), 2005, 361-366.
- [4] FMOH (Federal Ministry of Health), Malaria Desk situation analysis, Nigeris, Federal Government Publication, 2000, 27
- [5] Onwujekwe OE, Akpal CO, Shu NE and Okonkwo PO, How do rural households perceive and prioritize malaria and mosquito nets, a study of five communities in Nigeria, Public Health, 114, 2000, 407 – 410.
- [6] Malaney P, Spielman A and Sachs J, The malaria gap, Am. J. Tropical Med. and Hygiene, 2, 2004, 141-146.
- [7] Gillett JD, Mosquitoes, Weidenfeld and Nicholson, London, 1971, 123
- [8] Bodeker G and Willcox ML, Conference report: the first international meeting of the Research Initiative on Traditional Antimalarial Methods (RITAM), J. Alt. Compl. Med., 6, 2000, 195-207.
- [9] Teich S and Myers RAM, Maggot therapy for severe skin infections, Southern Medical J., 79, 1986, 1153.
- [10] Sherman RA and Pechter EA, Maggot Therapy: A review of the therapeutic applications of fly larvae in human medicine, especially for treating osteomyelitis, Medical and Vet. Entomol., 2 (3), 1988, 225-30..
- [11] Sherman, RA, Wyle, F and Vulpe M, Maggot Debridement Therapy for treating pressure ulcers in spinal cord injury patients. Journal of Spinal Cord Medicine, 18 (2), 1995, 71-74
- [12] An C, Li D and Du R, Analysis of antibacterial- relative proteins and

- peptides in housefly larvae, J. Hyg. Res., 33, 2004, 86-88
- [13] Cai H, Choi SI, Lee YM and Heo TR, Antimicrobial effect of herbal medicine extracts Staphylococcus aureus and Escherichia coli O157:H7, Korean J. Biotechnol. Bioeng., 17, 2002, 537-542.
- [14] Wang Y, Li D, Zhao Y, Lei C and Zhu F, Antiviral and antitumor activities of the protein fraction from larvae of the housefly, *Musca domestica*, Af. J. Biotechnol., 11(39), 2012, 9468-9474.
- [15] Chen Y and Li JF, The study of anti-virus activity of *Musca domestica* larva homogenate, J. Trop. Med., (4), 2003, 130-132.
- [16] Adebayo JO, Yakubu MT, Egwim EC, Owoyele BV, Enaibe BU, Effect of ethanolic extract of *Khaya senegalensis* stem bark on some biochemical parameters on rat Kidney, J. Ethnopharmacol., 88, 2003, 69-72.
- [17] Ryley JF and Peters W, The antimalarial activity of some quinoline esters. Annals of Tropical Medicine and Parasitol., 64, 1970, 209-222.
- [18] Mahajan BK, Significance of difference in mean. In: Methods in Biostatistics for medical and research workers, 6th Ed., Jaypee Brothers Medical Publi., New Delhi, 1997, 130-155.
- [19] Lemos FJA and Terra WR, Digestion of bacteria and the role of midgut lysozyme in some insect larvae, Comp. Biochem. Phys. B., 100 (2), 1991, 265- 268.
- [20] Xu XX, Jin FL, Yu XQ, Wang J, Cheng HX, Wang C and Zhang WQ, Expression and purification of a recombinant antibacterial peptide, cecropin from *Escherichia coli*, Protein Express. Purif., 53 (2), 2007, 293-301.
- [21] Cao XH, Zhou MH, Wang CL, Hou LH and Zeng B, Lectin purified from *Musca domestica* pupaup-regulates NO and iNOS production via TLR4/NF-kB signaling pathway in macrophages, Int. immunopharmacol., 11 (4), 2011, 399-405.
- [22] Gavigan CS, Dalton JP and Bell A, The role of aminopeptidases in haemoglobin degradation in *Plasmodium falciparum*-infected erythrocytes, Mol. Biochem. Parasitol., 117 (1), 2001, 37-48.