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Comparison of the Haemodynamic Effects of Pyrethroid Insecticide and Amodiaquine in Rats

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Abstract: Malaria infection is a common cause of morbidity and mortality especially in the tropics and subtropics. This has led to the increased prophylactic use of pyrethroid insecticides and/or Amodiaquine (Aq) to combat the parasitic protozoan infection. The aim of this study was to investigate the comparative haemodynamic effects of pyrethroid insecticide and amodiaquine in rats. Experimental rats were randomly allocated into seven groups of five rats in each. Groups 1, 2 and 3 were exposed to pyrethroid by inhalation for 1, 2 and 3 min, respectively, while groups 4, 5 and 6 were administered Aq per oral at 5, 10 and 15 mg kg⁻¹ b.wt., respectively. Control rats were neither exposed to pyrethroid nor administered Aq. Pyrethroid insecticide led to reduced systolic, diastolic and mean arterial pressures, but increased pulse pressure. Aq treatment did not cause any significant variation in haemodynamic variables. Heart rate was comparable in all groups. Results from the study provide extended safety/toxicity profile for pyrethroid use and Aq treatment. Aq showed no cardiotoxic potential, while pyrethroids have hypotensive effect. It is thus recommended that exposure to pyrethroids should be minimized.

Key words: Pyrethroids, amodiaquine, arterial pressure, pulse pressure, heart rate

INTRODUCTION

About one million people annually die of malarial infection (Snow *et al.*, 2005), consequently making it a major public health problem (Uneke, 2006; Akhigbe *et al.*, 2011; Saka *et al.*, 2012). Malarial infection occurs in the tropics and subtropics and sometimes in the temperate regions (Bienzle *et al.*, 1981; Werner and Mathys, 1987; Akhigbe *et al.*, 2011). Besides Acquired Immunodeficiency Syndrome (AIDS), malaria is the only disease that shows a significant increasing tendency (Amoo *et al.*, 2008). This has posed a serious public health challenge.

In an attempt to reduce malaria burden, prophylactic measures have been employed to prevent the spread of the infection. These measures include the use of Insecticide-treated Nets (ITN), insecticide sprays and pharmaco-prophylaxis. Commonly used in the tropics and subtropics are pyrethroid insecticides and Amodiaquine (Aq) prophylaxis (Olliaro and Mussano, 2003; WHO, 2006a; Saka *et al.*, 2012).

Insecticides use as in ITN or indoor spraying is the most widely used measure of malaria vector control (WHO, 2006b). Aq has also been used as a common chemoprophylaxis against malaria infection (Olliaro and Mussano, 2003). Pyrethroid insecticides are widely used to control plasmodial vector, mosquito. They are efficient,

broad-spectrum and quick neurotoxic pesticides developed on the base of the researches on the chemical structure of the natural pyrethrins (Cox, 2002; Jayakumar *et al.*, 2008). Aq is a cost-effective and potent schizonticide 4-aminoquinoline common used to treat uncomplicated malaria and to prevent malaria infection (Olliaro *et al.*, 1996; WHO, 1998; Massaga *et al.*, 2008).

None of these commonly implemented preventive measures are without side effects. Though, pyrethroids formulated insecticide has been reported to be safe following normal haematological parameters seen in experimental studies with minimal exposure (Saka *et al.*, 2011), some studies have reported its toxic effects (Inayat *et al.*, 2007; Sangha *et al.*, 2011) such as neurotoxicity, haematotoxicity and hepatotoxicity (Sayim *et al.*, 2005; Altug *et al.*, 2006; Saxena and Saxena, 2010). Similarly, Aq toxicity has been documented to include agranulocytosis and hepatitis (Clarke *et al.*, 1991; Olliaro and Mussano, 2003) although our previous study showed that Aq does not alter haematological variables (Saka *et al.*, 2012).

It is worth noting that the toxic hepatic and potential lethal agranulocytosis in Aq treatment were observed in non-immune travelers on chemoprophylaxis (Hatton *et al.*, 1986), however, it has been documented to be efficacious without serious haematological adverse effects or potential biological hepatotoxicity and nephrotoxicity

when given in required doses for chemotherapeutic (Mengesha and Makonnen, 1999).

Despite data reporting the safety and toxicity of pyrethroids and Aq use, it is still pertinent to assess their haemodynamic effects. This study sought to evaluate the comparative haemodynamic effects of pyrethroids formulated insecticide and Aq as commonly used in malarious regions.

MATERIALS AND METHODS

Experimental animal: Sprague Dawley rats of both sexes and comparable weights were used for the study. Animals were procured and bred in the Animal Holding Unit of the Department of Physiology, Ladoko Akintola University of Technology, LAUTECH, Ogbomoso, Nigeria. They were maintained in standard laboratory conditions with 12 h day/light cycle in standard rat cages and fed rat chow and water without restriction. They were acclimatized for two weeks prior to the onset of the experimental study (Eghoghosa *et al.*, 2011).

Treatment: Animals were randomly allocated into seven groups, with five rats per group. Groups 1, 2 and 3 were exposed to 30 mg m⁻³ of pyrethroids formulated insecticide containing 0.02%/w/w imiprothrin, 0.03%/w/w d-phenothrin and 0.10%/w/w d-trans allethrin, (®Mortein-Reckitt Benckiser, Nigeria) by inhalation for 1, 2 and 3 minutes daily respectively for three weeks (Saka *et al.*, 2012). Groups 4, 5 and 6 were administered Aq (Camoquine®, Parke-Davis Laboratories, United Kingdom) at varying doses of 5 mg kg⁻¹ b.wt. (low dose), 10 mg kg⁻¹ b.wt. (normal dose) and 15 mg kg⁻¹ b.wt. (high dose) *per oral* (p.o.) daily for 14 days, respectively (Saka *et al.*, 2012). Control rats were neither exposed to pyrethroid insecticide nor administered Aq.

All animals received humane care in compliance with the institution's guideline and criteria for humane care as outlined in the National Institute of Health Guidelines for the Care and Use of Laboratory Animals.

Anaesthesia, Dissection and cannulation and balancing and calibration of the polygraph: Animals were anaesthetized with 25% urethane and 1% alpha chloralose. The anaesthesia was administered at a dose of 3 mL kg⁻¹ intraperitoneally and when necessary, additional 0.5 mL was given to maintain surgical anaesthesia. The anaesthesia was warm in water bath before use. After administration of the anaesthetic agent, animals were kept in rat cages for some min until the anaesthetic effect sets in. The degree of anaesthesia was assessed to be suitable when there was lack of response

to painful stimuli, diminished or absence of muscular tone, absence of corneal reflex and presence of normal respiration in terms of frequency and amplitude.

The anaesthetized animal was placed on the dissecting board with ventral surface facing upward and its limbs pinned down to the board. A fold of skin was excised with a pair of scissors from the chin to the upper part of the sternum to expose the underlying connective tissue. The trachea was exposed by blunt dissection. The trachea was semi-transected between the two rings half way in the neck. A polyethylene cannula with internal diameter similar to the trachea was inserted and firmly tied with a ligature to ensure clear airway.

The skin over the right femoral artery and vein was cleared and the artery was exposed by blunt dissection and cannulated using a polyethylene catheter filled with heparinized salt solution. The femoral artery was used to record the blood pressure and heart rate on a tracing paper. The polygraph was calibrated at 40, 80, 120, 160 and 200 mmHg for recording blood pressure tracing.

The Grass 7D polygraph was set on calibration and the baseline control knob was set to adjust the baseline at convenient position on the recording paper. The pen writer was adjusted to a standard deflection of 2 cm representing 100 mV. The control knob on the DC amplifier was set on 'use'. The sensitivity and balance voltage on low level pre-amplifiers was maintained at zero and the calibration was re-adjusted to 2 cm/100 mV deflection. The sensitivity of the low level amplifier was set to maximum and the balance voltage was increased for the experiment (2 mL cm⁻¹). The calibration of blood pressure and balancing of the polygraph machine was done using a manometer after which it was disconnected.

The arterial blood pressure was recorded in the anaesthetized animals using the arterial cannula connected to the pressure transducer. The recording was made on the polygraph. The heart rate was obtained from the blood pressure recording.

Statistical analysis: Data were analyzed using the SPSS software (SPSS Inc, Chicago, USA). The data are expressed as Mean±SEM and analyzed for significant differences by one-way analysis of variance and unpaired t-test. Values of p<0.05 was considered statistically significant.

RESULTS

Systolic pressure was significantly reduced in animals exposed to pyrethroid insecticide for 2 and 3 minutes (81.4 and 80 mmHg, respectively) when compared with other groups. This haemodynamic fluctuation was

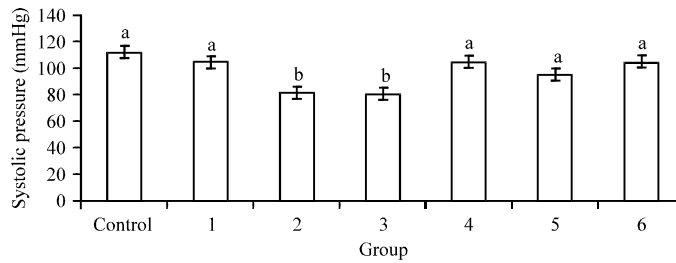


Fig. 1: Effect of pyrethroid insecticide and amodiaquine on systolic pressure, bars carrying same letters are statistically comparable at $p < 0.05$

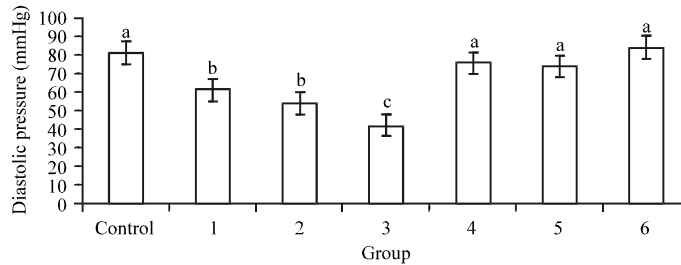


Fig. 2: Effect of pyrethroid insecticide and amodiaquine on diastolic pressure, bars carrying same letters are statistically comparable at $p < 0.05$

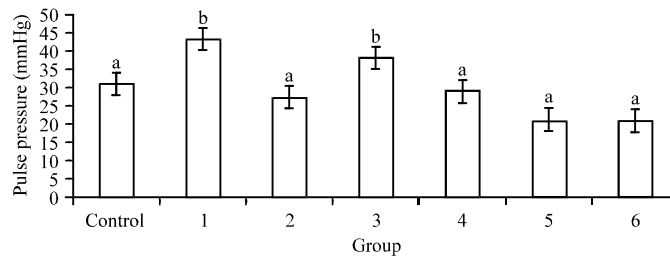


Fig. 3: Effect of pyrethroid insecticide and amodiaquine on pulse pressure, bars carrying same letters are statistically comparable at $p < 0.05$

not in a duration-dependent pattern. Though animals on Aq therapy showed some variations in systolic pressure, these were not statistically significant (Fig. 1). Pyrethroid exposure also led to significant reduction in diastolic pressure in a duration-dependent fashion (61.2, 54 and 41.8 mmHg for animals exposed for 1, 2 and 3 min, respectively). The change in diastolic pressure following Aq-treatment was not significantly different when compared with that of the control (Fig. 2).

Pulse pressures in all animals were similar except in those exposed to pyrethroid insecticide for 1 and 3 min which had a significantly raised pulse pressure (43.2 and 38.2 mmHg, respectively). This is in attendant with the wide range of variation between their systolic and diastolic pressures (Fig. 3).

The mean arterial pressures of rats administered varying doses of Aq were not significantly different when

compared with the control. Though pyrethroid insecticide caused a duration-dependent reduction in the mean arterial pressure of rats, only the rats exposed for 2 and 3 min showed significant fall in the mean arterial pressure (63.13 and 54.53 mmHg, respectively) (Fig. 4).

Although exposure to pyrethroid insecticide and Aq treatment caused alterations in heart rate, these were however not statistically significant (Fig. 5).

DISCUSSION

Despite conflicting data documented on the safety of pyrethroids and Aq, no study has reported the haemodynamic changes seen following exposure to these malaria prophylactic measures. As at the time of reporting these findings, no study had documented the comparative haemodynamic effects of pyrethroids formulated

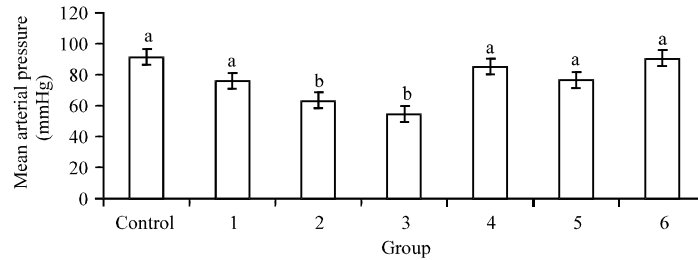


Fig. 4: Effect of pyrethroid insecticide and amodiaquine on mean arterial pressure, bars carrying same letters are statistically comparable at $p < 0.05$

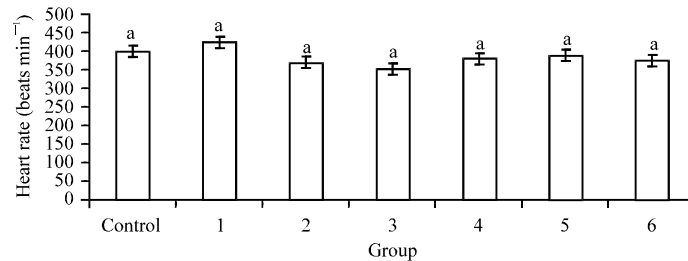


Fig. 5: Effect of pyrethroid insecticide and amodiaquine on heart rate, bars carrying same letters are statistically comparable at $p < 0.05$

insecticides and Aq treatment. Findings in this study revealed that Aq did not alter systolic nor diastolic arterial pressure. However, pyrethroids formulated insecticide caused a significant reduction in systolic and diastolic pressures. These is in consonance with previous study (Massaga *et al.*, 2008) that reported marginal alterations in systolic and diastolic pressures following administration of Aq in parasite negative and parasite positive volunteers.

Similarly, Aq treatment had no significant effect on the mean arterial pressure and pulse pressure in rats. On the other hand, pyrethroid insecticide led to a significant fall in the mean arterial pressure but a rise in pulse pressure. This is inconsistent with previous study Forshaw and Bradbury (1983) that reported that deltamethrin, but not rismethrin (pyrethroids) caused increased mean arterial pressure and differential pressure in pithed rats. This variance could be due to the different pyrethroid constituents of the insecticide used in the studies, as different pyrethroids showed different effects on the mean arterial and pulse pressures.

The significant rise in pulse pressure in association with reductions of systolic and diastolic pressures in pyrethroid-exposed rats suggests that the formulated insecticide had a marked effect on diastole than systole. This showed that pyrethroid perturbed the sympathetic-regulated cardiac activity.

Interestingly, pyrethroid formulation and Aq did not alter heart rate. This disagrees with previous studies that documented the positive inotropic effect of deltamethrin

(Forshaw and Bradbury, 1983) and the bradycardic effect of Aq (Adaramoye and Almeida, 2010). The reduced mean arterial pressure with normal heart rate observed in pyrethroid exposed rats could imply that pyrethroid led to vasodilatation (Flannigan and Tucker, 1985) with resultant fall in total peripheral resistance possibly by opening of opening of vascular K^+ channels or blockade of extracellular Ca^{2+} influx.

CONCLUSION

This study shows a distinction between the cardiovascular activities of pyrethroid formulation and Aq. The findings in this study provide extended safety/toxicity profile for pyrethroid use and Aq treatment. Though this study revealed that pyrethroid formulated insecticide has hypotensive effect, it could still be used with caution by reducing the duration of exposure to the lowest possible. Aq treatment showed no cardiotoxic possibility.

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