



# EVALUATION OF EFFICACY OF TRANSDERMAL DELIVERY OF CHLOROQUINE ON *PLASMODIUM BERGHEI*-INFECTED MALE SPRAGUE-DAWLEY RATS AND EFFECTS ON BLOOD GLUCOSE AND RENAL ELECTROLYTE HANDLING

<sup>1</sup>Cephas T Musabayane, <sup>2</sup>Samson Mukaratirwa, <sup>1</sup>Happiness Sibiya and <sup>1</sup>Pretty Murambiwa

Schools of <sup>1</sup>Laboratory Medicine and Medical Sciences and <sup>2</sup>Life Sciences, University of KwaZulu-Natal, Private Bag X54001, Durban 4000, South Africa.

## INTRODUCTION

Clinical evidence indicates that oral administration of chloroquine (CHQ) evokes adverse effects on glucose homeostasis and kidney function in African children (Jarzyna et al., 2001). These complications are partly ascribed to transiently high plasma CHQ concentration and/or malaria parasites (Gustafsson et al., 1983). We have, however, reported that topical application of pectin CHQ matrix patch formulation sustains controlled release of CHQ into the bloodstream (Musabayane et al., 2003).

## OBJECTIVES

The purpose of the study was to determine whether CHQ delivered via the transdermal route can reduce malaria parasites and ameliorate the side effects associated with oral CHQ.

## MATERIALS AND METHODS

### Patch preparation

The pectin-CHQ matrix patch was prepared using a protocol similar to that previously described with slight modifications (Musabayane et al., 2003). Briefly, pectin was dissolved in de-ionised water followed by agitation at 700 rpm. Chloroquine disphosphate was then added to the mixture and stirred at 37°C for 30 minutes. Following this, dimethyl sulphoxide, vitamin E and eucalyptus oil were added to the mixture. Following this, an aliquot of the mixture (1.1 ml) was transferred to separate petri dishes and frozen at -50°C for 18 hours following which a 2% CaCl<sub>2</sub> solution was added to the mixture to allow for cross linking. The patches were then stored in the refrigerator at 2°C until use.

### Animals

Male Sprague-Dawley rats (90-190 g body weight) bred and maintained at Biomedical Research Unit, University of KwaZulu-Natal were used in this study.

### Induction of malaria

Malaria was induced in male Sprague-Dawley rats with a single intraperitoneal injection of *P. berghei* (105 parasitised RBC).

### Oral glucose tolerance (OGT) responses

Oral glucose tolerance responses (OGT) to CHQ delivered orally (60mg/kg) or transdermally (53mg/kg) were monitored in groups of non-infected and *Plasmodium berghei*-infected male Sprague-Dawley rats given a glucose load after an 18-h fast. Rats treated with deionized water (3 ml/kg p.o.) or drug free pectin acted as controls. Blood glucose was monitored at 15 min intervals for the first hour, and hourly thereafter for 3 h.

### Sub-chronic effects of CHQ

Blood glucose and renal function were monitored over a 21-day period divided into pre-treatment (days 0-7), treatment (days 8-12) and post-treatment (days 13-21) in separate groups of animals. Oral CHQ was administered twice daily (60 mg/kg, p.o.) by means of a bulbed steel needle whilst the CHQ patch (53 mg/kg) was applied once at the beginning of the treatment period.

### Statistical analysis

All data presented as means ±SEM. Graph Pad Instat software (version 5) using one way analysis of variance (ANOVA) followed by Tukey-Kramer multiple comparison test was used. p values < 0.05 were considered significant.

## RESULTS AND DISCUSSION

### CHQ effects on blood glucose

Both acute and long-term oral administration of CHQ significantly (p<0.05) decreased blood glucose concentrations of non-infected and infected animals (Figures 1). A decrease in blood glucose following acute topical application of the CHQ patch was observed.

However, the blood glucose concentration remained stable throughout the experiment following long-term application of the CHQ patch by comparison to both non infected and infected baseline values (Figure 2).

### CHQ effects on renal electrolytes handling

An elevation of mean urinary Na<sup>+</sup> output in non-infected and infected animals following oral administration of CHQ was observed (Figure 3). The current study has also shown hyperkalaemia and elevated urinary K<sup>+</sup> outputs in malaria infected control animals. Elevated urinary K<sup>+</sup> outputs were also shown in *P. berghei* infected animals treated with oral CHQ. However, transdermal application of CHQ had no significant effect on renal electrolyte handling (Figure 4).

Topical application of pectin CHQ matrix patch did not alter the urinary Na<sup>+</sup> and K<sup>+</sup> outputs of both non infected and infected animals, thus confirming the ability of the CHQ patch to provide a sustained controlled release of CHQ to the systemic circulation, evading the adverse effects of oral CHQ administration.

### Effects on % parasitaemia

The peak percentage parasitaemia of control animals was reached at day 14 51.5 ± 16.5%. The once off topical application of CHQ and twice oral administration of CHQ equally reduced *P. berghei* parasites (Figure 5). The once of topical application of CHQ also sustained plasma CHQ concentration.

## CONCLUSION

Transdermally delivered CHQ equally reduced *P. berghei* parasites by comparison with twice daily oral CHQ. Also, CHQ patch has the potential circumvent the adverse effects of oral CHQ.

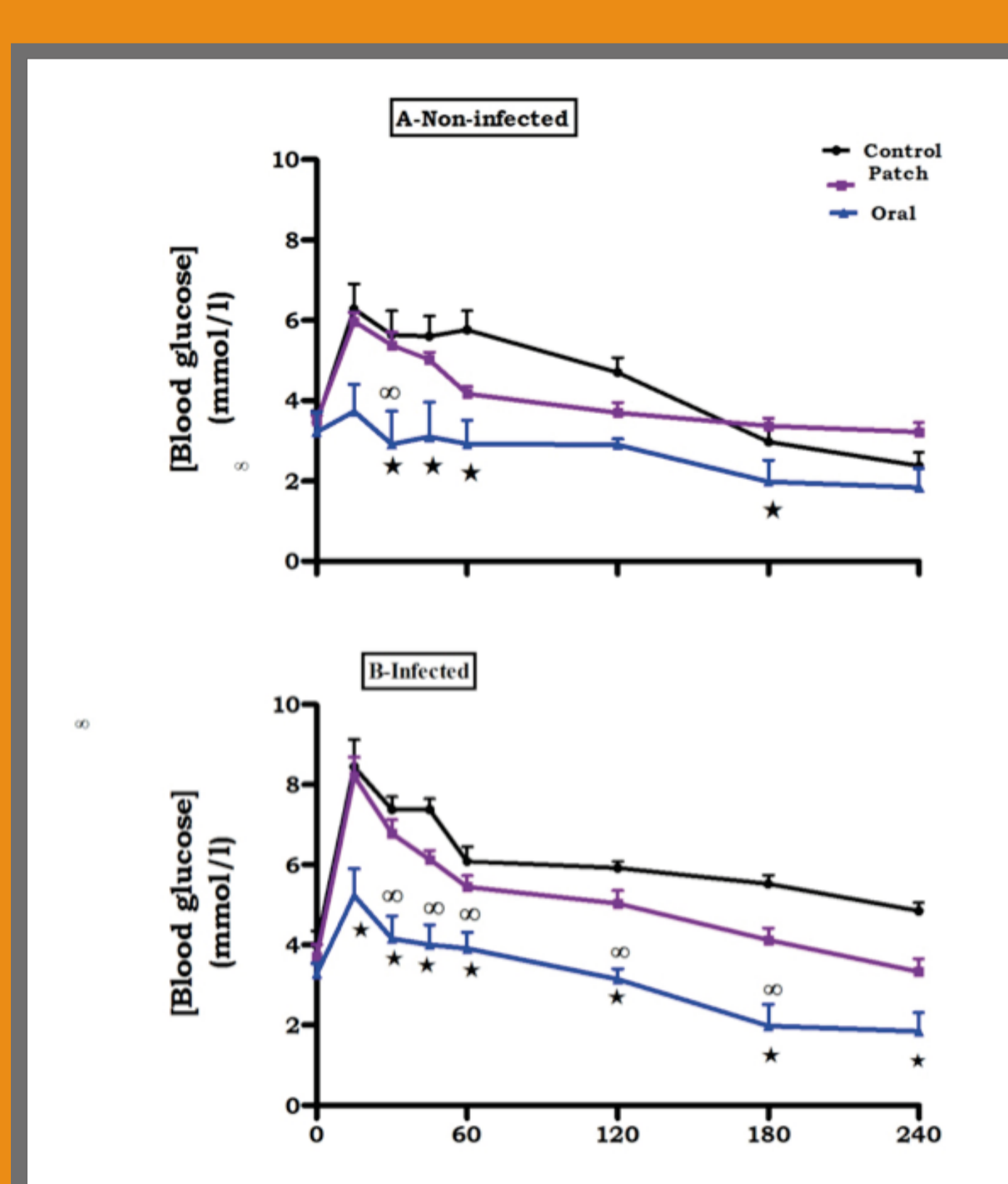


Figure 1: OGT responses to orally and transdermally delivered CHQ in non-infected (A) and infected (B) rats. \* = p<0.05 by comparison with control animals. \*\* = p<0.05 by comparison with oral CHQ treated animals.

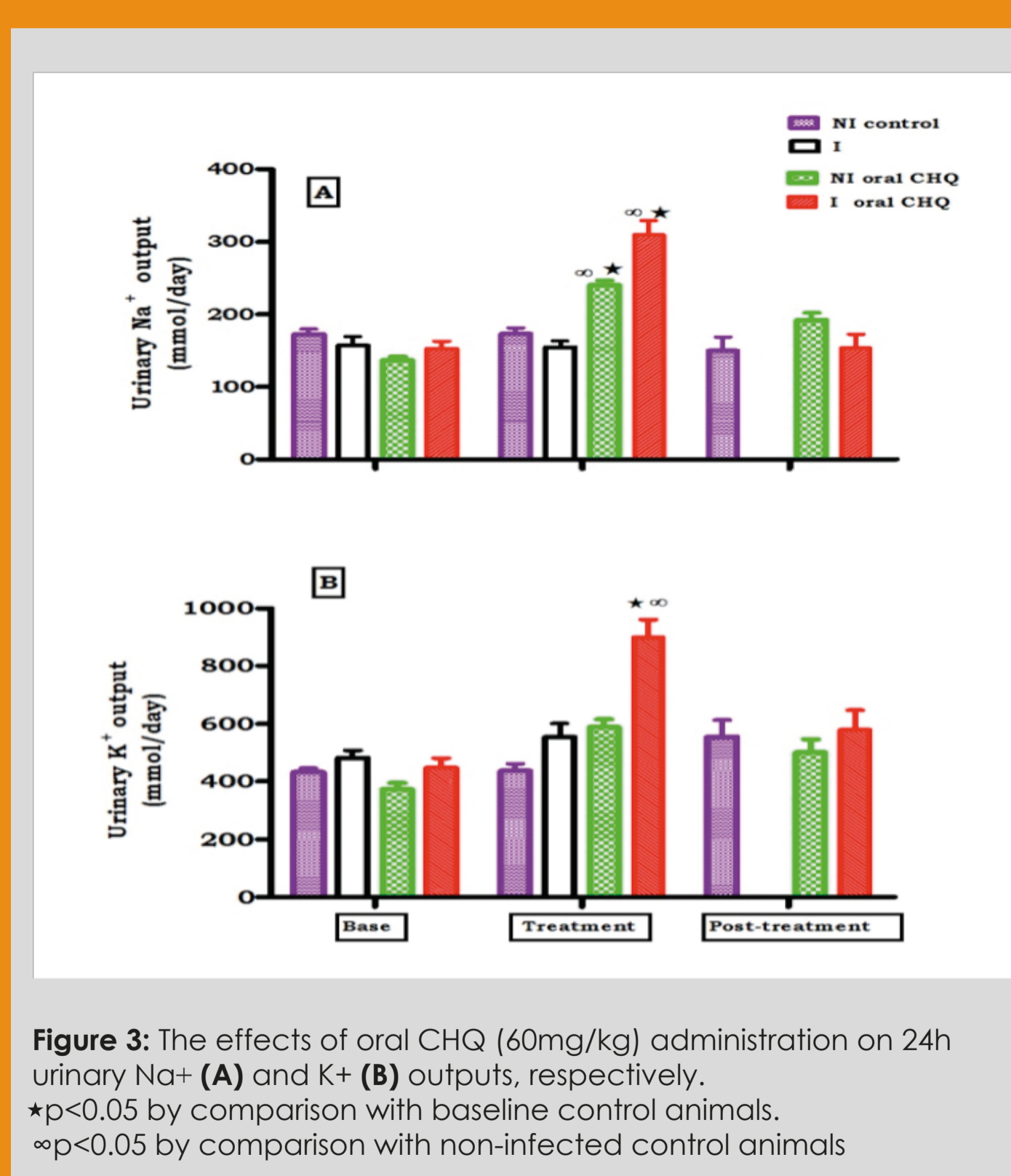


Figure 3: The effects of oral CHQ (60mg/kg) administration on 24h urinary Na<sup>+</sup> (A) and K<sup>+</sup> (B) outputs, respectively. \*p<0.05 by comparison with baseline control animals. \*\*p<0.05 by comparison with non-infected control animals.

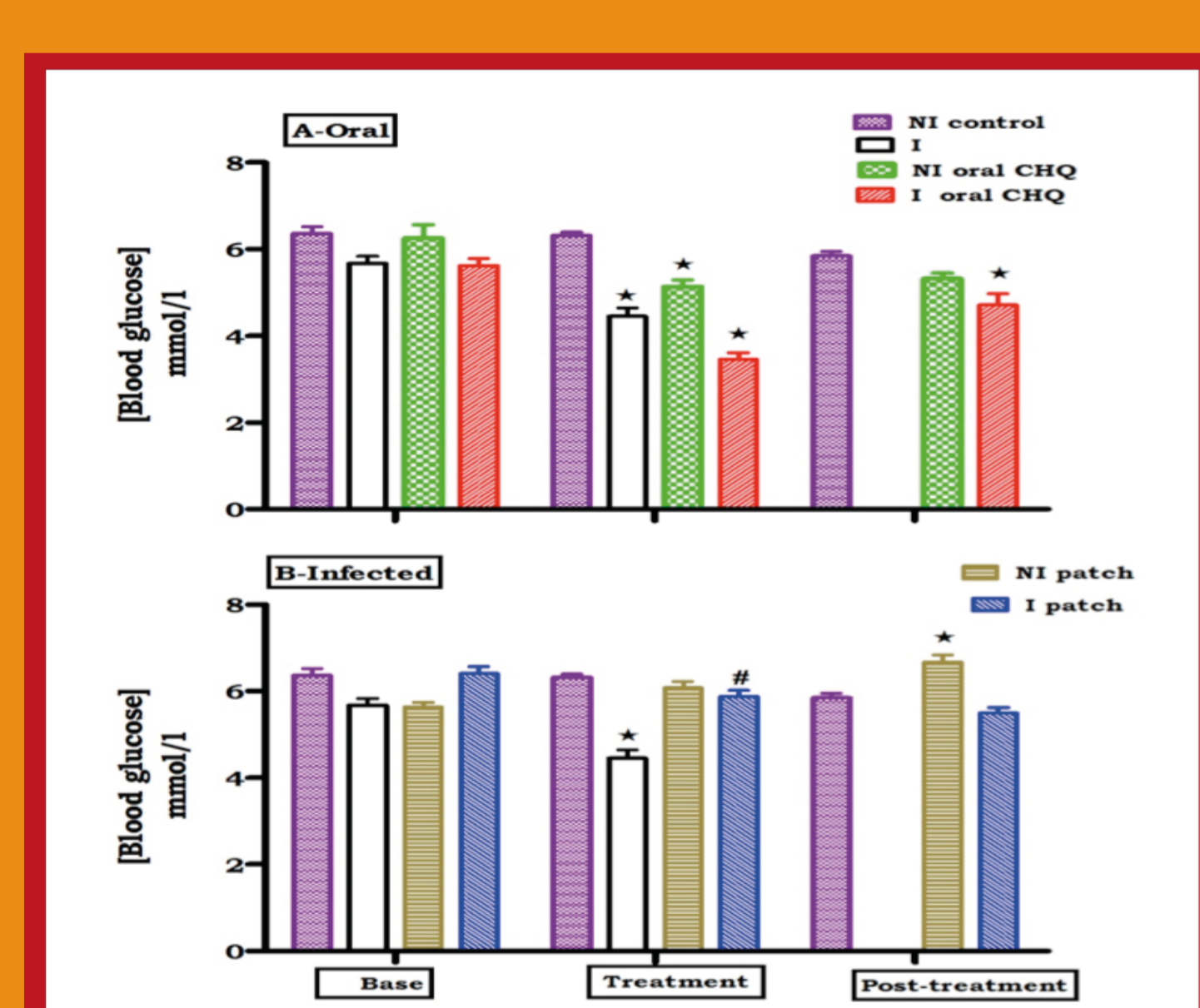


Figure 2: The effects of oral CHQ (60mg/kg) treatment (A) and single topical application of CHQ matrix patch (53mg/kg) (B) on blood glucose concentration. \*p<0.05 by comparison to baseline control animals. # p<0.05 by comparison control animals.

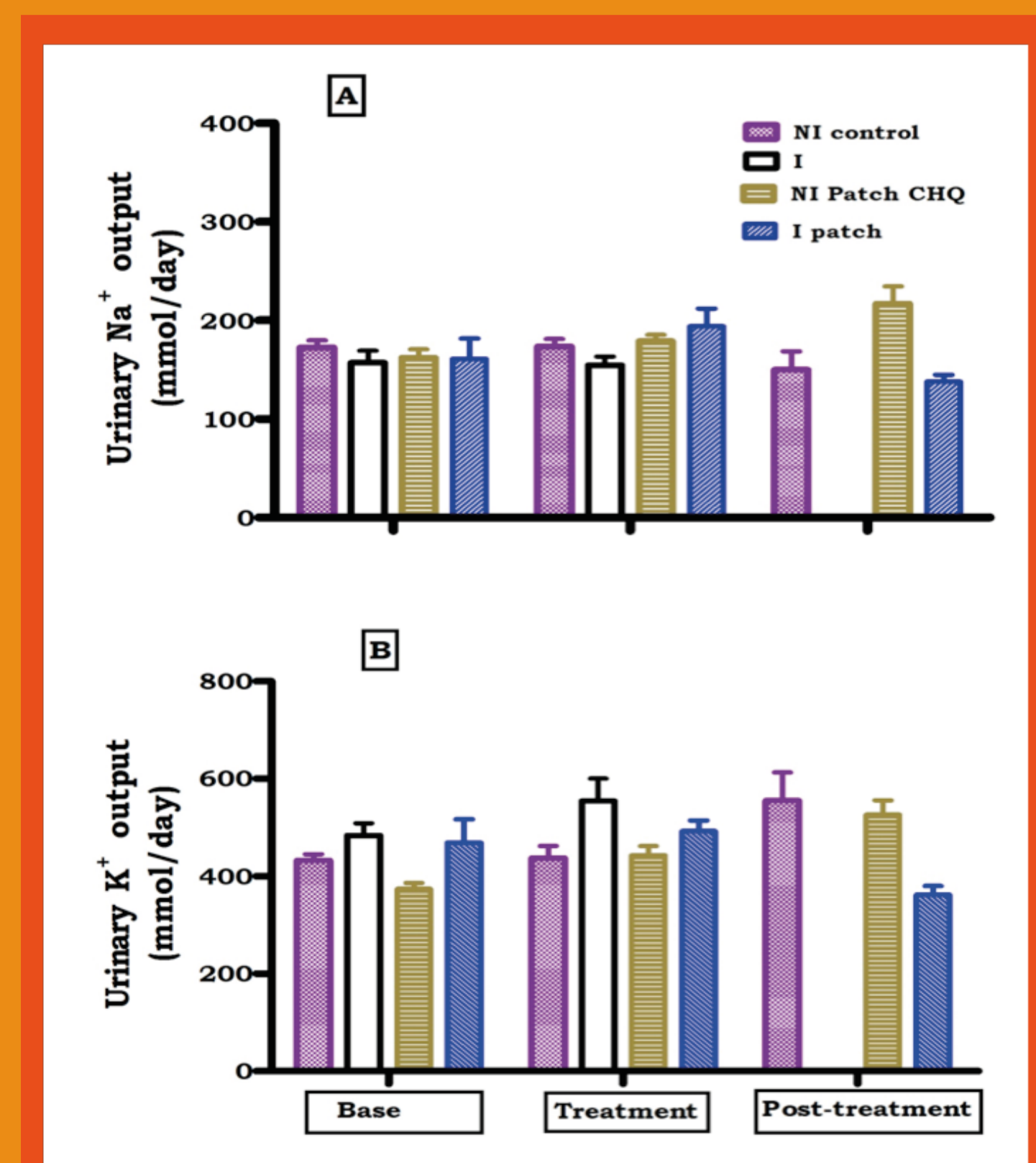


Figure 4: The effects of a single topically applied pectin CHQ matrix patch (53mg/kg) on 24h urinary Na<sup>+</sup> (A) and K<sup>+</sup> (B) outputs, respectively. \*p<0.05 by comparison with baseline control animals.

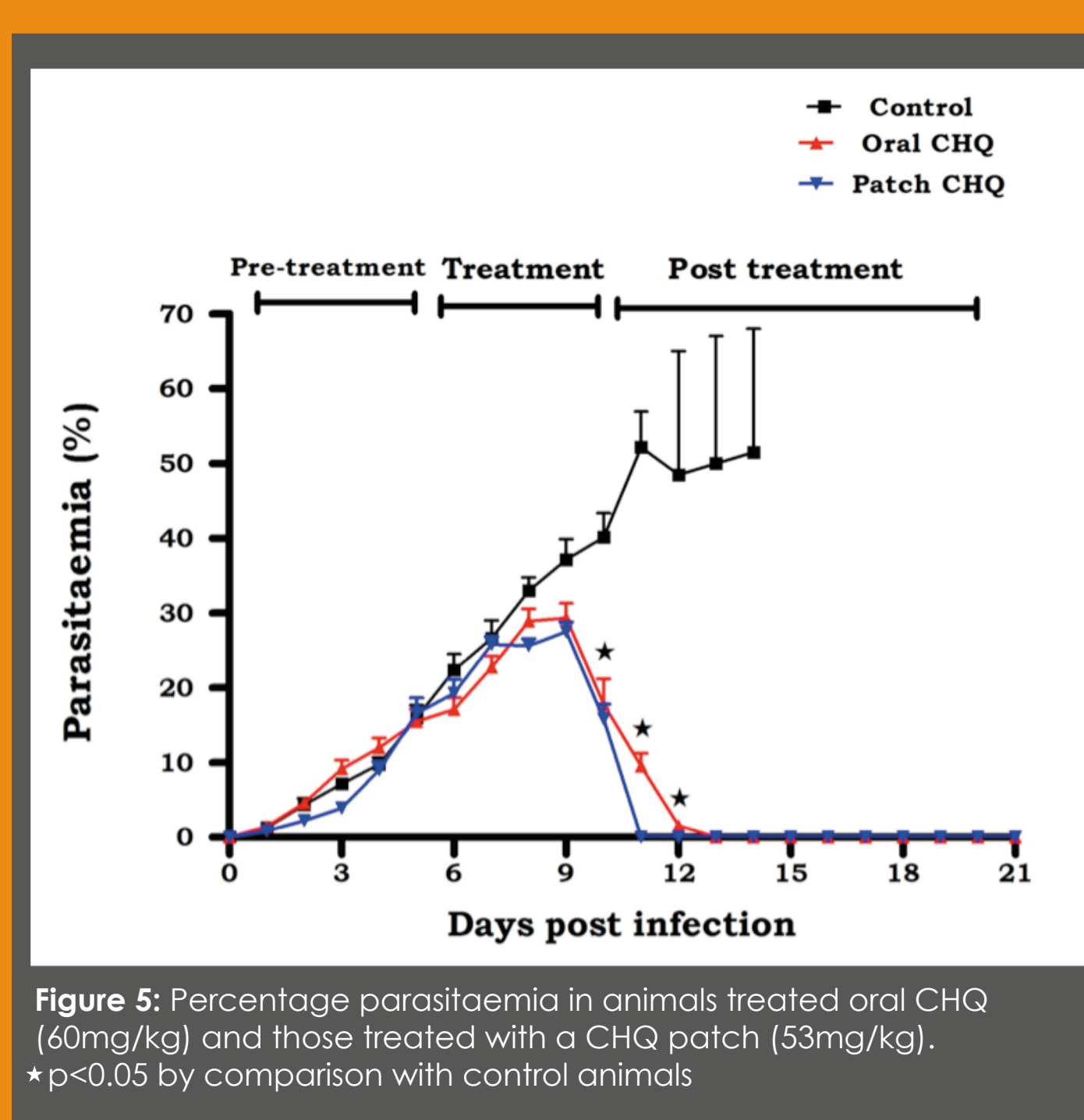


Figure 5: Percentage parasitaemia in animals treated oral CHQ (60mg/kg) and those treated with a CHQ patch (53mg/kg). \*p<0.05 by comparison with control animals.

## REFERENCES

- Musabayane, C. T., Munjeri, O. & Matavire, T. P. (2003). Renal Failure, 25, 525-34.
- Gustafsson, L. L., Walker, O., Alvan, G., Beermann, B., Estevez, F., Gleisner, L., Lindstrom, B. & Sjoqvist, F. (1983). British Journal of Clinical Pharmacology, 15, 471-9.
- Jarzyna, R., Kiersztan, A., Lisowa, O., and Bryla, J. (2001). European Journal of Pharmacology 428, 381-388.