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Full Length Research Paper

Assessment of the potency of some selected anti-malaria drugs on the supplements of vitamin B2 and orange fruit juice (combination therapy)

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Current practice in treating cases of malaria is based around the concept of combination therapy, since this offers several advantages - reduced risk of treatment failure, reduced risk of developing resistance, enhanced convenience and reduced side-effects. The effect of the supplements vitamin B2 (Riboflavin) and orange fruit juice on the potency and efficacy of some selected anti-malaria drugs (Armact, Coartem, Waipa and Fansider) as a combination therapy were investigated. 80 patients (adults) infected with malaria parasites were used. The study showed that the simultaneous administration of the drugs with vitamin B2 did not alter the potency of the drugs, while orange fruit juice altered the efficacy of the drugs. Therefore, the concomitant administration of these anti-malaria drugs (combination therapies) with orange fruit juice should be avoided during the period of malaria treatment for the effectiveness of such drugs. The result of this study also showed that concomitant administration of Riboflavin with antimalarial drugs may possess potent antimalarial effects and may therefore offer a potential drug lead for development of a safe, effective and affordable antimalarial.

Key words: Combination therapy, orange fruit juice, riboflavin, antimalarial.

INTRODUCTION

Malaria is a mosquito-borne infectious disease of humans and other animals caused by *Plasmodia*. The disease results from the multiplication of Plasmodium parasites within red blood cells, causing symptoms that typically include fever and headache, in severe cases progressing to coma or death. Five species of *Plasmodium* can infect and be transmitted by humans. Severe disease is largely caused by *Plasmodium falciparum* while the disease caused by *Plasmodium vivax*, *Plasmodium ovale* (Sutherland et al., 2010) and *Plasmodium malariae* is generally a milder disease that is rarely fatal. *Plasmodium knowlesi* is a zoonosis that causes malaria in macaques but can also infect humans (Fong et al., 1971; Singh, et al., 2004).

Malaria transmission can be reduced by preventing mosquito bites by distribution of mosquito nets and insect repellents, or by mosquito-control measures such as spraying insecticides and draining standing water (where mosquitoes breed). Despite a clear need, no vaccine offering a high level of protection currently exists. Efforts to develop one are ongoing (Kilama and Ntoumi, 2009). A number of medications (anti-malaria drugs) are also available to prevent malaria in travelers to malaria-endemic countries (prophylaxis).

A variety of anti-malaria medications are available. Severe malaria is treated with intravenous or intramuscular quinine or, since the mid-2000s, the artemisinin derivative artesunate (Dondorp and Day, 2007), which is superior to quinine in both children and adults (Dondorp et al., 2010). Resistance has developed to several anti-malaria drugs, most notably chloroquine (Kirby, 1989). There were an estimated 225 million cases of malaria worldwide in 2009 (WHO, 2011). An estimated 655,000 people died from malaria in 2010 (WHO, 2011),

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Table 1. Effects of Armact only on patients infected with malaria parasite.

Patients	Weight(kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight(kg) after treatment	Remark
Female	60	++	Armact only	_	58	Cleared
Female	65	+++	Armact only	_	64	Cleared
Male	70	++	Armact only	_	65	Cleared
Male	70	++	Armact only	_	67	Cleared

+ = Positive (Malaria parasite present), ++ = Moderately severe parasite present, +++ = Severe malaria parasite, - = Negative (malaria parasite absent).

a decrease from the 781,000 who died in 2009 according to the World Health Organization's 2011 World Malaria Report, accounting for 2.23% of deaths worldwide. However, a 2012 meta-study from the University of Washington and University of Queensland estimates that malaria deaths are significantly higher. Published in *The Lancet*, the study estimates that 1,238,000 people died from malaria in 2010 (Christopher et al., 2012). Ninety percent of malaria-related deaths occur in sub-Saharan Africa, with ~60% of deaths being young children under the age of five (Christopher et al., 2012). *P. falciparum*, the most severe form of specie, is responsible for the vast majority of deaths associated with the disease (Snow, 2005). This work looked at the effects of some supplements (grape fruit juice, orange fruit juice, multivitamin and vitamin C) on the efficacy and potency of some selected anti-malaria drugs-combination therapy (Armact, Coartem, Waipa and Fansider).

MATERIALS AND METHODS

Experimental animals

A total of 80 persons (40 males and 40 females) infected with malaria parasite residing in Umuguma in Owerri west local government area of Imo state, Nigeria were used during the experiment after a general malarial test on all the individuals and their body weights were taking before and after drug administration (treatment).

Collection of blood sample

The methanol was used as a disinfectant to swab the big thumb and a lancet used to puncture it for blood collection. Two drops of blood was placed on free grease slide, a thick film was made and allowed to air-dry. The dried thick blood film slide was laid on a staining rack, and Giemsa stained and allowed for 30-40 min, washed off with clean water, drained and allowed to dry at room temperature. Then viewed under the microscope using 10x objectives for focusing and 40x objective for identifying the plasmodium involved. Blood samples of subjects were all confirmed to be malaria parasite infected via malaria parasite test as described by Sibley (2001). The research had the approval of the concerned institutional medicinal ethics boards.

Drugs and supplements administered

The drugs used were Armact (Artesunate and Amodiquine), Coartem (Artemether and Lumefantrine), both purchased from Novartis pharmaceutica, Waipa (Dihydroartemisinin and Piperaquine) and Fansider (Sulfadoxin and Pyrimethamine) both purchased from Swiss Pharma Nigeria limited. The supplements used were vitamin B2 and orange fruit juice.

RESULTS

The results of the test on the blood samples before and after administration of the anti-malaria drugs to the patients are shown in Tables 1 to 12.

DISCUSSION

From the assessment of the potency of some selected anti-malaria drugs on the supplements of orange fruit juice and vitamin B2 (combination therapy), the result of the effects of the administration of antimalarial drugs such as Armact only, Coartem only, Waipa only and Fansider on patients infected with malaria parasite showed that the malaria parasite in all the groups were absent. The absence of malarial symptoms or death in the oral administration of those antimalarial drugs observed in the patients suggests that the drug is practically non-toxic acutely (Salawu et al., 2009; Russell, 2008). This could also explain the safe use of the drugs by the local people, for the treatment of malaria, in the eastern part of Nigeria. This also suggests the findings of Ajaiyeoba et al. (2006) that the use of these drugs for the treatment of malaria was due to the presence of alkaloids. Also, this is similar to the effect of the extract reported by previous studies on Alstonia boonei (Iyiola et al., 2011). From Tables 1, 5, 8 and 11, it showed that the concomitant administration of the antimalarial drugs (Armact, Coartem, Waipa and Fansider) and orange fruit juice indicated the presence of the Plasmodium in all the treated groups. This could be as a result of the anti-oxidant properties present in the fruit which may increase plasma concentrations of the drug and delays reaching

Table 2. Effects of Armact and orange juice (OJ) on patients infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/Supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	85	++	Armact /OJ	+	85	Cleared
Female	80	++	Armact /OJ	+	80	Cleared
Male	55	+	Armact /OJ	+	55	Cleared
Male	65	+++	Armact /OJ	+	65	Cleared

Table 3. Effects of Armact and vitamin B2 on patients infected with malaria parasite.

Patients	Weight(kg) before treatment	Malaria parasite before treatment	Drug/Supplements	Malaria parasite/after treatment	Weight(kg) after treatment	Remark
Female	60	++	Armact/Vit B2	_	60	Not cleared
Female	60	++	Armact / Vit B2	_	60	Not cleared
Male	85	++	Armact / Vit B2	_	85	Not cleared
Male	85	++	Armact / Vit B2	_	85	Not cleared

Table 4. Effects of Coartem (Coart) only on patients infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	65	++	Coart only	_	60	Cleared
Female	65	+	Coart only	_	65	Cleared
Male	60	+	Coart only	_	60	Cleared
Male	60	+	Coart only	_	60	Cleared

Table 5. Effects of Coartem and orange juice (OJ) on patients infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	60	++	Coart /OJ	+	60	Cleared
Female	65	+	Coart /OJ	+	65	Cleared
Male	70	++	Coart /OJ	+	70	Cleared
Male	70	+++	Coart /OJ	+	70	Cleared

Table 6. Effects of Coartem and vitamin B2 on patients infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight(kg) after treatment	Remark
Female	70	+++	Coart /Vit B2	_	70	Not cleared
Female	55	+	Coart / Vit B2	_	55	Not cleared
Male	75	++	Coart / Vit B2	_	75	Not cleared
Male	75	+	Coart / Vit B2	_	75	Cleared

Table 7. Effects of WAIPA (Wp) only on patient infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weigh t(kg) after treatment	Remark
Female	60	+	Wp only	_	60	Cleared
Female	60	++	Wp only	_	58	Cleared
Male	70	++	Wp only	_	68	Cleared
Male	75	+	Wp only	_	72	Cleared

Table 8. Effects of WAIPA (Wp) and orange juice (OJ) on patients infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	75	+	Wp /OJ	+	75	Cleared
Female	65	++	Wp /OJ	+	65	Cleared
Male	70	+	Wp /OJ	+	70	Cleared
Male	75	++	Wp /OJ	+	73	Cleared

Table 9. Effects of WAIPA (Wp) and vitamin B2 on patient with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	60	+	Wp /Vit B2	_	60	Not cleared
Female	68	+	Wp / Vit B2	_	68	Not cleared
Male	75	+	Wp / Vit B2	_	75	Not cleared
Male	70	++	Wp / Vit B2	_	70	Cleared

Table 10. Effects of Fansider (Fans) only on patients infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	75	+	Fans only	_	70	Cleared
Female	70	+	Fans only	_	68	Cleared
Male	65	+	Fans only	_	62	Cleared
Male	60	+	Fans only	_	58	Cleared

Table 11. Effects of Fansider and orange fruit Juice (G.J) on patients infected with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	70	+	Fans /GJ	+	68	Cleared
Female	70	+	Fans /GJ	+	67	Cleared
Male	60	+++	Fans /GJ	+	58	Cleared
Male	55	+	Fans /GJ	+	53	Cleared

Table 12. Effects of Fansider and vitamin B2 on patients with malaria parasite.

Patients	Weight (kg) before treatment	Malaria parasite before treatment	Drug/supplements	Malaria parasite/after treatment	Weight (kg) after treatment	Remark
Female	78	+++	Fans /Vit. B2	_	78	Not cleared
Female	65	+	Fans /Vit. B2	+	65	Not cleared
Male	60	++	Fans /Vit. B2	_	60	Not cleared
Male	65	+	Fans /Vit. B2	_	65	Cleared

peak drug concentration (Owira and Ojewole, 2010). Also it may be due to the presence of orange fruit enzyme that break down medications in the digestive system and cause more of the medications to stay in the body which may increase the risk of serious malarial problems (Stump, 2006).

The concomitant administration of the antimalarial drugs (Armact, Coartem, Waipa and Fansider) and vitamin B2 (Riboflavine) from Tables 2, 6, 9 and 12 showed no malaria parasite presence in all the patients in the various treated groups. This infers that vitamin B2 may play a significant role in antimalarial activity which is similar to the report of Adesokan and Akanji (2010). This could be as a result of its ability to promote appetite, quick recovery and do not hinder the activity of the antimalarial drug (Makarchikov, 2003). This report supports the finding of Basu et al. (2007) that this vitamin may possess health promoting effects, at least under some circumstances.

Conclusion

Successful malaria control depends greatly on treatment with efficacious anti-malarial drugs. The ability of the four drugs (Armact, Coartem, Waipa and Fansider) to reduce the presence of malaria parasite may be due to presence of phytochemically-active components in the drugs which might be responsible for their therapeutic activity as antimalarial drugs. Also, the use of vitamin B2 (Riboflavin) with antimalarial drugs (combination therapy) has potential health promoting effects. Multiple-drug therapies that include a nonantimalarial drug like vitamin E and orange fruit juice to enhance the antimalarial effect of a blood schizontocidal drug are not considered combination therapy.

This finding supports the use of vitamin B2 and antimalarial drugs as a combination therapy which is safe and possess potent antimalarial activity as found in its ability to suppress Plasmodium infection in patients.

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