USE OF IVERMECTIN FOR TRANSMISSION BLOCKING OF *Plasmodium vivax* IN *Anopheles aquasalis* AND *Anopheles darlingi*

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Abstract

Background. The use of insecticide-treated nets and indoor residual insecticides targeting adult mosquito vectors is a key element in malaria control programs. However, mosquito resistance to the insecticides threatens malaria control efforts. Alternative methods like ivermectin (IVM) administration to humans has been suggested as a possible vector control tool to reduce *Plasmodium* transmission. *Anopheles aquasalis* and *Anopheles darlingi* are competent vectors for *Plasmodium vivax* in the coast of Brazil and the Amazon region, respectively.

Material and Methods. (A) One single IVM dose (200 μg/mL) was ingested by volunteers and blood samples were drawn at distinct times (0, 4 hours, 1, 5, 10, 14 days). *Anopheles aquasalis* and *An. darlingi* were infected by membrane feeding *P. vivax* from malaria patients. Patient plasma was removed and replaced with plasma from IVM-treated volunteers. Seven days after the infection, the mosquitoes were dissected to check the oocyst presence and for *An. darlingi* also the sporozoite presence at day 14. (B) The *ex vivo* effect of the addition of ivermectin on cultivated *P. vivax* was observed. (C) The effect of IVM on malaria vivax patients infectivity to *Anopheles* was analyzed: different regimes treatment Chloroquine (CQ)+IVM, CQ+Primaquine (PQ) and CQ+IVM+PQ.

Results. IVM significantly reduced the prevalence of *An. aquasalis* that developed oocysts (40, 20 and 10ng/mL compound; 4 hours, 1 and 5-day plasma). In *An. darlingi* oocyst prevalence and intensitywas only reduced with plasma from 4 hours and 1 day. Mosquito mortality was increased in *Anopheles aquasalis* that ingested 40 ng/mL ivermectin compound and plasma from 4 hours, 1, 5, 10 and 14 days post-intake, and in *An. darlingi* that ingested plasma from 4 hours and 1 day post-intake, recording 100% mortality 14 days post-feeding with 4 hour,1 and 5-day plasma. The double fled of IVM by the mosquitos has a significant impact on the proportion of infected mosquitos. The oocyst prevalence and

intensity was significantly reduced in comparison to control on mosquitos fed with unprocessed blood from patients that undertook CQ+IVM, CQ+PQ and CQ+IVM+PQ. In the *ex vivo* cultures, ivermectin (plasma 4 hours) significantly inhibited *P. vivax* asexual development, reducing the number of schizonts.
Conclusion. Ivermectin reduces the oocyst prevalence and intensity of *P. vivax* in *An. aquasalis* and *An. darlingi*, and increased the mortality of mosquitos. These findings support that ivermectin is useful to reduce *P. vivax* transmission.

Key words: Ivermectin, *Plasmodium vivax*, *Anopheles aquasalis*, *Anopheles darlingi* Malaria, Vector Control.