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Acetylcholinesterase Activity and Histopathological Effect of Caffeine in Spleen of *Plasmodium berghei* ANKA-infected ICR Mice

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Structured Abstract

Background: Malaria has been a significant health concern in Peninsular Malaysia since 2015. Despite a decrease in the number of cases, the disease remains troubling, with an unexpected occurrence of 4,000 to 5,000 cases annually. Antimalarial drug resistance has led to higher rates of morbidity and mortality, weakening efforts to control the disease. Hence plant-based compound like caffeine was explored as a potential drug to treat malaria. The association between caffeine and malaria has been studied, particularly for its potential to improve the efficacy of antimalarial medications. This study aims to determine the AChE activity and the histopathological effects of caffeine on the spleen of malarial infected mice.

Methods: The study methodology involves division of mice into 4 groups: the normal control, *P. berghei* ANKA-infected (negative control), infected CQ-treated (positive control), and infected caffeine-treated. Each group was inoculated with *P. berghei* ANKA followed by administration of 10 mg/kg b.w of chloroquine and 5 mg/kg b.w of caffeine. Next, parasitemia levels were measured to confirm infection, euthanization with carbon dioxide and spleen extraction. Then, histopathological examination and AChE analysis by using Ellman *et al.* (1961) method were conducted.

Results: This study shows that caffeine reduced AChE activity to $1.69\times 10^{-6} \pm 0.05\ \mu M$/min compared to the infected control at $2.37 \times 10^{-6} \pm 0.23\ \mu M$/min. Histological analysis of caffeine demonstrated a decrease in inflammation, hemozoin presence, and PRBCs in the cell structure. Caffeine-treated mice trabeculae also showed less disruption and thickening of the spleen than chloroquine-treated mice, which showed signs of edema and a thicker structure due to fibrosis.

Conclusion: In conclusion, caffeine shows potential as a drug to mitigate the impact of malaria on the spleen. To further assess its efficacy, additional research on other *Plasmodium* strains and different animal models is important to ensure their validity. Genetically modified mouse models could provide insights into the genetic factors influencing the response of caffeine in malarial infection.

Keywords: Malaria, *Plasmodium berghei* ANKA, Caffeine, AChE activity

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