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Genetic Polymorphism and Insilico Study Epitope Block 2 MSP1 Gene of Plasmodium falciparum Isolate Endemic Jayapura

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Abstract : Malaria is an infectious disease caused by Plasmodium sp. This disease has a high prevalence in Indonesia, especially in Jayapura. The vaccine that is currently being developed has not been effective in overcoming malaria. This is due to the high polymorphism in the Plasmodium genome especially in areas that encode Plasmodium surface proteins. Merozoite Surface Protein 1 (MSP1) Plasmodium falciparum is a surface protein that plays a role in the invasion process in human erythrocytes through the interaction of Glycophorin A protein receptors and sialic acid in erythrocytes with Reticulocyte Binding Proteins (RBP) and Duffy Adhesion Protein (DAP) ligands in merozoites. MSP1 can be targeted to be a specific antigen and predicted epitope area which will be used for the development of diagnostic and malaria vaccine therapy. MSP1 consists of 17 blocks, each block is dimorphic, and has been marked as the K1 and MAD20 alleles. Exceptions only in block 2, because it has 3 alleles, among others K1, MAD20 and RO33. These polymorphisms cause allelic variations and implicate the severity of patients infected P. falciparum. In addition, polymorphism of MSP1 in Jayapura isolates has not been reported so it is interesting to be further identified and projected as a specific antigen. Therefore, in this study, we analyzed the allele polymorphism as well as detected the MSP1 epitope antigen candidate on block 2 P. falciparum. Clinical samples of selected malaria patients followed the consecutive sampling method, examining malaria parasites with blood preparations on glass objects observed through a microscope. Plasmodium DNA was isolated from the blood of malarial positive patients. The block 2 MSP1 gene was amplified using PCR method and cloned using the pGEM-T easy vector then transformed to TOP'10 E.coli. Positive colonies selection was performed with blue-white screening. The existence of target DNA was confirmed by PCR colonies and DNA sequencing methods. Furthermore, DNA sequence analysis was done through alignment and formation of a phylogenetic tree using MEGA 6 software and insilico analysis using IEDB software to predict epitope candidate for P. falciparum. A total of 15 patient samples have been isolated from Plasmodium DNA. PCR amplification results show the target gene size about ± 1049 bp. The results of MSP1 nucleotide alignment analysis reveal that block 2 MSP1 genes derived from the sample of malarial patients were distributed in four different allele family groups, K1 (7), MAD20 (1), RO33 (0) and MSP1_Jayapura (10) alleles. The most commonly appears of the detected allele is MSP1_Jayapura single allele. There was no significant association between sex variables, age, the density of parasitemia and alel variation (Mann Whitney, $U > 0.05$), while symptomatic signs have a significant difference as a trigger of detectable allele variation ($U < 0.05$). In this research, insilico study shows that there is a new epitope antigen candidate from the MSP1_Jayapura allele and it is predicted to be recognized by B cells with 17 amino acid lengths in the amino acid sequence 187 to 203.

Keywords : epitope candidate, insilico analysis, MSP1 P. falciparum, polymorphism
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