In Silico Design, Analysis and Structure Prediction of Tow New Chimeras of VK210/VK247CSP Antigen for Development of a Malaria Pre-erythrocytic Subunit Vaccine Candidate

SH. Shabani[a,b], S. Zakeri[a]*, AH. Salmanian[c], J. Amani[d], AA. Mehrizi[a], Y. Mortazavi[b], ND. Djadid[a]

[a] Pasteur Institute, Tehran, Iran

[b] Zanjan University of Medical Sciences, Zanjan, Iran

[c] National Institute of Genetic Engineering and Biotechnology, Tehran, Iran

[d] Baqiyatallah University of Medical Sciences, Tehran, Iran
*zakeris@yahoo.com

Abstract: *Plasmodium vivax* is the most widespread species of Plasmodium and responsible for approximately 50% of malaria cases outside Africa. An effective vaccine is vital for successful control, elimination and eradication of malaria in endemic areas. Due to molecular and cell biology, epidemiology and immunology, progress towards an effective malaria vaccine has been disappointing and a subunit vaccine against all vivax strains is the only option in the absence of long-term culture and special biological characterization of *P. vivax* parasites. The circumsporozoite protein (CSP) that covers the surface of Plasmodium sporozoites is a well-characterized vaccine candidate. The main objective of the present study is to design and compare of structure and folding impact of two new constructs based on different forms of CSP on effective stimulating of immune system to produced neutralizing antibodies.

In this study, tow constructs were designed based on reference sequences (Sal-1 and PNG). CS127 and CS712 encompassing repeats from the two major alleles, VK210 and VK247 as equal as minimum number of each repeats variants in nature and entire N- and C-terminal to compare structure and antigenicity potent of each construct. *In silico* techniques, have launched to characterize the properties and structure of the proteins. Biochemical properties and multiple alignments were predicted by appropriate web servers. Secondary molecular structures were predicted based on Chou and Fasman, Garnier-Osguthorpe-Robson, and Neural Network methods. Tertiary modeling elucidated conformational properties of the chimeric proteins. Ramchandran Plot analysis were used to confirm predicted tertiary model. Linear and conformational B-cell antigenic epitopes, were predicted using bioinformatic web servers. Ligand-receptor interaction of chimeric proteins and mRNA secondary structure prediction was performed by means of computational bioinformatics tools and servers.

Our results have indicated codon adaptation index of both chimeric genes has increased and the overall GC content, was improved to 63.22% for both constructs in comparison with the reference sequences. The mfold data has shown that CS127 and CS712 mRNA was stable enough for efficient translation in the prokaryotic host. One 3D structure out of five predicted by I-TASSER for each construct was selected based on the highest value for C-score. Ramchandran Plot analysis of both constructs also represented that most residues were fallen in favorable regions and classified both constructs as stable protein. The linear and conformational B-cell epitopes in the chimeric proteins are surfaced and likely to induce the B-cell mediated immune responses. Finally based on ligand-receptor docking the binding ability of both CS127 and CS712 was strong enough to their receptor, so these constructs could be assigned as tow new subunit vaccines against malaria.

In conclusion, *in silico* study showed that both CS127 and CS712 design constructs has a constant structure and are effectively able to elucidate the effective immune responses. Therefore, data reported in this paper represents the first step toward developing of an effective subunit vaccine candidate based on CSP against malaria infection.

Keywords: *Plasmodium vivax*; malaria; subunit vaccine; circumsporozoite protein; sporozoites