Malaria is a major cause of mortality and morbidity worldwide. According to the latest World Malaria Report, there were 229 million cases of Malaria in 2019 (1). The etiological agent of this disease is transmitted through the bite of infected Anopheles mosquitoes, which introduce the parasites of the genus Plasmodium into the host skin. The circumsporozoite protein (CSP) is the most abundant protein on the surface of Plasmodium sporozoites, the motile and infective stage of malaria parasite transmitted by the mosquitoes. CSP contains central repeating motifs and it mediates sporozoite adhesion to target cells. Antibodies against the central CSP repeats of different plasmodial species are known to block sporozoites infectivity (2). We have recently demonstrated that an anti-repeat monoclonal antibody, capable of sterile protecting mice against infectious sporozoites, kills parasites by stripping off the protective CSP surface coat, rendering the parasite membrane susceptible to the sporozoites pore-forming-like protein secreted to wound and traverse the host cell membrane (3). To further increase the protective anti-CSP antibodies, we screened a Fab library that was generated in our group following intradermal immunization of mice with radiation-attenuated sporozoites combined with Fab phase-display technology. From the CSP-specific Fab library, we selected 43 Fab presenting unique full-length sequences and screened for their capacity to bind CSP repeated region, in particular, minor, major and junctional regions. Periplasmic extracts of 43 Fab were produced and their presence confirmed by Western blot against the myc-tag. The Fab binding capacity to the several regions of the CSP is being assessed by enzyme-linked immunosorbent assay (ELISA). The most effective binders will be purified by affinity chromatography and their functionality at blocking sporozoites infectivity will be further evaluated by in vitro and in vivo assays.

Results

1. Recombinant Fab were detected in 31 out of 43 periplasmic extracts

2. The peptides selection in the P. berghei CSP

3. The anti-CSP Fab clones recognize the major repeats

Conclusions

- The several Fab showed reactivity against the major repeat "PPPNPND".
- None of the Fab clones recognize the minor or the junctional regions of CSP.
- Further work is needed to evaluate the functionality of the most effective binders at blocking sporozoites infectivity.

References


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