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Prediction of potential vaccine candidates from *Plasmodium yeolii* through reverse vaccinology approach

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Against *Plasmodium*, causative agent of malaria causing hundreds of millions of clinical infections and at least a million deaths per annum; a number of drugs have been designed but have encountered resistance, hence vaccination appears to be an efficient way for controlling spread of malaria. Rodent malaria parasite *Plasmodium yeolii* is considered because it reproduces many of the biological characteristics of the human malaria parasite whilst falciparum and vivax are particularly tricky as the complete life cycle cannot be maintained in vitro. Reverse vaccinology is an innovative approach to design efficient vaccines to overcome many difficulties of conventional vaccine development including culture of pathogen in lab that pose a chance to be hazardous. RV counts on the collective use of immunological and genomic information to recognize relevant protein antigens for vaccine development. Here in this work antigen determinants were predicted and MAPPP (MHC-I Antigenic Peptide Processing Prediction) made binding and proteasome cleavage prediction to identify possible antigenic peptides present on the cell surfaces. Molecule having greater LCV value was designed and showed potential to be a successful vaccine. This work demonstrated that Reverse Vaccinology approach is more likely to be discovering various immunogenic antigens from computational analysis of pathogen's genome/proteome instead of culturing the whole organism by conventional methods.

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High frequency and poor prognosis of late childhood BCR-ABL positive and MLL-AF4 positive ALL define the need for advanced molecular diagnostics and improved therapeutic strategies in pediatric B-ALL in Pakistan

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Background: Fusion oncogenes (FO's) resulting from chromosomal abnormalities have important role in leukemogenesis in pediatric B-cell acute lymphoblastic leukemia (ALL). The most common FO's are BCR-ABL, MLL-AF4, ETV6-RUNX1 and TCF3-PBX1 which have important prognostic and drug selection implications. Moreover, frequencies of FO's have ethnic variations. We studied frequencies of FO's, clinical pattern and outcome in pediatric B-ALL.

Methods: FO's were studied in 188 patients at diagnosis using RT-PCR and inter-phase FISH. Data were analyzed using SPSS version 17.

Results: FO's were detected in 87.2% of patients. Mean overall survival was 70.9 weeks, 3-year survival 31.9% and 3-year relapse free survival 18.1%. Four patients died of drug toxicities. ETV6-RUNX1 (19.14%) had better survival (110.9 weeks, p=0.03). TCF3-PBX1 (2.1%) was associated with inferior outcome and higher CNS relapse risk. MLL-AF4 (18.1%) was more common in 8-15 year age group (24/34, p=0.001) and associated with organomegaly, low platelet count and poor survival. BCR-ABL (47.9%) was associated with older age (7-15 years, 52/90), lower remission rates, shorter survival (43.73± 4.24 weeks) and higher white cell count. Overall, MLL-AF4 and BCR-ABL were detected in 66% of B-ALL presented at later childhood and were associated with poor prognosis and inferior survival.

Conclusions: Study reports highest frequency of BCR-ABL FO in pediatric ALL consistent with the previous reports from our region. Poor prognosis BCR-ABL and MLL-AF4 detected in two third pediatric B-ALL are likely to be the reason for already-reported poor survival in our part of the world. Furthermore, MLL-AF4 usually most common in infants presented at late childhood in most of ALL patients which is one of the unique findings in our study. This highlights the need for mandatory inclusion of molecular testing for pediatric ALL patients in clinical decision making and incorporation of TKIs as well as HSCT facilities to improve treatment outcome in developing countries.

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