Production of Recombinant Antigens and Antibodies for Diagnosis Purposes Using Synthetic Biology Methods

Zanchin, N.I.T.[1], Krieger M.A.[2,3], Souza C. [2,3], Eugenio D. S. [2,3], Santos K. [2,3], Lima L.F.O. [2,3], Santos N.F.B.[1], Izumi E. [2,3], Teixeira C.S.[2,3], Celedon P.A.F. [2,3], Baschirotto P.T. [2,3], Oliveira U.D. [2,3], Puhl, A.C.[2,3].

[1]Laboratório de Proteômica e Engenharia de Proteínas & [2]Laboratório de Genômica Funcional - Instituto Carlos Chagas, FIOCRUZ, Paraná, Brasil

[3]Instituto de Biologia Molecular do Paraná, Curitiba, Paraná, Brasil

INTRODUCTION: Quality control of blood transfusion and pregnancy risk assessment require millions of diagnosis tests to be performed on a daily bases. This work is motivated by two major points, namely the dependence of Brazil health programs from imported tests, which causes an estimated deficit of 200 million US Dollars/year, and the necessity to improve the biochemical reagents used in immunoassays to include the recently discovered genetic variants of many pathogens.

OBJECTIVES: The objective of this work is to establish production processes and to test antigens and antibodies for diagnosis purposes.

MATERIALS AND METHODS: Antigenic proteins from the pathogens tested in the quality control of blood transfusion and pregnancy risk assessment programs were selected, taking into account their genetic variation. Over one hundred synthetic genes were optimized for their expression in *E. coli*. In some cases, antigenic regions were combined into chimeric proteins in order to reduce the number of bioprocesses. The development of recombinant antibodies involves production of full-length and Fab and scFv fragments.

RESULTS AND DISCUSSION:

Eight pathogens (HIV1, HIV2, HTLV-I, HTLV-II, HCV, HBV, *Trypanosoma cruzi*, *Treponema pallidum*) are tested in quality control of blood transfusion, with *Plasmodium* and CMV being additionally required for malaria endemic areas and immunosuppressed patients, respectively. All of the pathogens above and toxoplasmosis and rubella are generally tested for pregnancy risk assessment. Depending on the region other pathogens must be included, as for example HRSV. In addition to serum antibodies, immunodiagnosis tests of some pathogens (HIV1, HBV, HRSV, malaria) need to include detection of viral proteins, so that antibodies against viral proteins must be developed. Production processes have already been established for recombinant antigens that passed immunological trials for the pathogens HIV1-2, HCV, HBV, *T.cruzi*. *T.pallidum* and *Toxoplasma gondii*. Three recombinant antibodies have been also produced.

CONCLUSION: The approach adopted here allowed successful production of novel recombinant antigen and antibodies for immunodiagnosis.

Funding: FINEP, BNDES, CAPES, CNPq, Fundação Araucária, FIOCRUZ.

Keywords: synthetic genes, biotechnology, immunodiagnosis