NS5 FROM YELLOW FEVER VIRUS INTERACTS WITH NUCLEAR CELLULAR PROTEINS .

Madrid, M.C.F.S.¹; Bronzoni, R.V.M.¹; Carmo, A.C.V.^{1,2}; Zanelli, C.F.³; Valentini, S.R.³; Carmo, A.C.V.^{1,2}; <u>Nogueira, M.L.¹</u>

¹FAMERP, São José do Rio Preto-SP, Brasil; ² UNESP/IBILCE, São José do Rio Preto-SP, Brasil; ³ UNESP-FCF, Araraquara-SP, Brasil.

Introduction and Objectives: Yellow Fever is a member of the flaviviridae family. NS5 is a viral dual function protein that has a methyltransferase and a RNA polymerase domain, and is detected in the nucleus of infected cells. However, its nuclear function is not known. The aim of this study was to identify and characterize the interactions of NS5 protein with cellular proteins. Results: YFV NS5 RNA polymerase fragment of NS5 gene was cloned in a DNA-BD plasmid. For the two-hybrid system screening, RNApol bait and the prey, (AD vector/human cDNA library), were transformed into AH109 strain of *Saccharomyces cerevisiae*. The transformants, initially selected in SD-HIS-LEU-TRP drop-out media, were tested for the activation of three AH109 yeast reporters' genes. Ten out of 100 positive colonies for HIS3, ADE2 and lacZ reporters were selected for further studies. These clones included protein involved in RNA splicing and translation, transcription activation and regulation, apoptosis control, and cytokines. Deletions in NS5 showed that these interactions use different parts of the protein. Conclusion: These data suggests that NS5 is a multifunctional protein and are indeed related to several activities including actions in the nucleus of the cell.

Financial Support: FAPESP (04/11098-2 and 05/00976-1)

Key words: Yellow Fever Virus, RNA polymerase, yeast 2-hybrid