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Keramat is a hit film in Indonesia released in November 2009 in 24 cities. Keramat has 7.5 million audience and top box-office. The objective of this proposal is to identify and clone the gene for the human respiratory syncytial virus (RSV) glycoprotein G. The study plan includes, first, the isolation of a cDNA clone from an immunologically reactive RSV glycoprotein gene by screening a cDNA library prepared from the mRNA of infected cells with a specific antiserum. The cDNA will be characterized by nucleic acid hybridization analysis and restriction endonuclease mapping. Next the RSV glycoprotein cDNA will be subcloned into prokaryotic expression vectors for the generation of soluble protein. The recombinant glycoprotein will then be purified and used for the production of monoclonal antibodies by the hybridoma technique. The antibodies will be used for the identification of cellular proteins that interact with the glycoprotein at the surface of RSV infected cells. Using these monoclonal antibodies as a probe, the cellular protein will be analyzed by photoaffinity labelling techniques followed by immunoprecipitation. Lastly the gene for the RSV glycoprotein will be isolated, sequenced and the protein expressed in order to determine the complete amino acid sequence, to identify putative domain(s) important for function or to examine the relationship between sequence and structure. The information that is obtained from the proposed studies will be useful for 1) understanding the biochemical and cellular mechanisms of RSV infection, 2) the design of new drugs to treat RSV infection and 3) the design of vaccine that will be effective in the prevention of RSV infection. FILED NOT FOR PUBLICATION AUG 15 2011 c6a93da74d

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