

Poster Presentation

Open Access

Expression of functional recombinant rabies virus glycoprotein in *Drosophila melanogaster* S2 cells

Adriana Y Yokomizo¹, Soraia AC Jorge¹, Renato M Astray¹,
Mariza AG Santos¹, Irene Fernandes², Orlando G Ribeiro³,
Denise SPQ Horton⁴, Aldo Tonso⁵ and Carlos A Pereira*¹

Address: ¹Laboratório de Imunologia Viral, Instituto Butantan, 05503-900 São Paulo, Brasil, ²Laboratório de Imunopatologia, Instituto Butantan, 05503-900 São Paulo, Brasil, ³Laboratório de Imunogenética, Instituto Butantan, 05503-900 São Paulo, Brasil, ⁴Serviço de Controle de Qualidade, Instituto Butantan, 05503-900 São Paulo, Brasil and ⁵Departamento de Engenharia Química, Escola Politécnica, Universidade de São Paulo, Brasil

* Corresponding author

from The 4th Recombinant Protein Production Meeting: a comparative view on host physiology
Barcelona, Spain. 21–23 September 2006

Published: 10 October 2006

Microbial Cell Factories 2006, **5**(Suppl 1):P89 doi:10.1186/1475-2859-5-S1-P89

© 2006 Yokomizo et al; licensee BioMed Central Ltd.

Background

The rabies virus belongs to the genus *Lyssavirus* from the *Rhabdoviridae* family and is widely distributed in nature infecting mammals. Upon infection it can be transmitted to animals or humans and leads to a fatal disease that nowadays has no treatment. Vaccines are commercially available and prevent the disease in animals and humans. Protocols for human or veterinarian vaccine manufacturing evolved from animal tissue homogenates to cell culture technology and today recombinant viral proteins and DNA vaccines are under investigation. The evidence that rabies virus infects and can cause disease in animals and humans, being neutralized by an immune response mounted by very similar vaccines opens a great possibility of testing new vaccines first in experimental animals prior to use in humans [1].

Results

Recombinant rabies virus glycoprotein (rGPV) was expressed in *Drosophila melanogaster* Schneider 2 (S2) cells. The cDNA encoding the GPV gene was cloned in expression plasmids under the control of the inducible metallothionein promoter (Mt) or the constitutive actin promoter (Ac). These were alternatively co-transfected into S2 cells together with a hygromycin selection plasmid. Selected S2 cell populations (S2MtGPV or S2AcGPV) had a decreased ability to grow and consume substrates,

when compared to the non transfected cells (S2). They were shown, by PCR, to express the GPV gene and mRNA as well as, by immunoblotting, to synthesize the rGPV in its expected molecular weight of 65 kDa. ELISA kinetic studies showed the rGPV expression in cell lysates and supernatants attaining concentrations ranging from 150 to 300 µg of rGPV/L. By flow cytometry analysis, about 30% of the cells in these populations were shown to express the rGPV in their membrane. Cell populations selected by limiting dilution expressed higher rGPV yields. Mice immunized with S2MtGPV or S2AcGPV derived rGPV were capable of mounting a protective immune response characterized by the synthesis of antibodies reacting against the rabies virus. Immunization led to protection against rabies virus experimental infection in challenge studies.

Conclusion

The data presented here show that S2 cells are suitable hosts for the rGPV expression allowing its synthesis in a high degree of physical and biological integrity.

Acknowledgements

This work was supported in part by grants and scholarships from the FAPESP, CNPq, CAPES and Fundação Butantan. C. A. Pereira is recipient of CNPq research fellowships. We thank M.J.S. Leme for technical assistance performing "in vivo" challenge assays.

References

1. Warrel MJ, Warrel DA: **Rabies and other lyssavirus diseases.** *Lancet* 2004, **363**:959-969.

Publish with **BioMed Central** and every scientist can read your work free of charge

"BioMed Central will be the most significant development for disseminating the results of biomedical research in our lifetime."

Sir Paul Nurse, Cancer Research UK

Your research papers will be:

- available free of charge to the entire biomedical community
- peer reviewed and published immediately upon acceptance
- cited in PubMed and archived on PubMed Central
- yours — you keep the copyright

Submit your manuscript here:
http://www.biomedcentral.com/info/publishing_adv.asp

