

# High-titer packaging cell lines producing retroviral vectors in suspension and serum-free media

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#### Technology description

Several patients with severe combined immunodeficiency-X1 disease and adenosine deaminase deficiency have been cured by retroviral-mediated gene therapy. Despite the earlier success, the production of retroviral vectors for clinical gene therapy is cumbersome, costly, and lacks safety features because of the adherent nature of packaging cells and the necessity to supplement the culture media with bovine serum. A packaging cell line that could grow in suspension with serum-free media would allow vector production in bioreactors with practically no size limit in a safe manner.

#### Description

Bicistronic vectors containing an internal ribosomal entry site followed by a selection gene were used to express Moloney murine leukemia Gag-pol and amphotropic, RD114 and Gibbon ape leukemia virus envelopes viral proteins in HEK293 cells. We derived 3 clones A2, GLV9 and R30 capable of producing high-titer retroviral vectors. These cells can be cultured in suspension and serum-free media. The production of virus was shown to be stable in suspension and serum-free media for a 3-month period with the A2 packaging cell line (titers were superior to 107 infectious viruses/ml).

#### Market

Gene therapy is still in development with 50 to 100 clinical trials/year, mainly phase I (30% using retroviral vectors). A GMP virus lot for a phase I clinical trial can be estimated between 100,000\$ to 300,000\$ depending on the number of patients.

#### Application area

Retrovirus packaging cell lines are used to produce vectors for gene therapy. These vectors can be used for pre-clinical and clinical applications.

#### Advantages

The manufacturing of retroviral vectors for phase I clinical trials is cumbersome with the existing packaging cell lines, and vector production for late phase clinical trials is almost impossible. The A2, GLV9 and R30 cell lines have the potential for the large-scale biomanufacturing of retroviral vector, and

they should be ideal for the implementation of late phase cancer gene therapy clinical trials. They would also substantially decrease the production cost of vectors for phase I clinical trials.

### Institution

Université Laval

# 联系我们



## 叶先生

电话: 021-65679356 手机: 13414935137

邮箱: yeyingsheng@zf-ym.com