Excellence in Separations

DENSITY GRADIENT MEDIA

Why should I change to OptiPrep for the purification of virus?

Compared to CsCl and sucrose there are procedural advantages to the use of OptiPrep

- OptiPrep is a sterile solution of 60% (w/v) iodixanol; simply dilute it with saline to prepare sterile gradient solutions. It is the only gradient medium manufactured under strict FDA and EU cGMP compliance.
- CsCl and sucrose are toxic to cells and must be removed prior to infection of cells.
- Iodixanol is non-toxic to cells; it has very low endotoxin levels (<1 EU/ml); measured levels on each batch are usually <0.13 EU/ml. No dialysis is required before re-infection of cells[1].
- CsCl must be removed prior to HPLC or gel electrophoresis; iodixanol rarely needs removing prior to further processing.

CsCl gradients lead to major reductions in infectivity of all viruses

- The recovery of rAAV from iodixanol gradients is at least 10x greater than from CsCl gradients[2].
- Infectivity titre is up to 10x higher in iodixanol isolates compared to CsCl isolates of rAAV[2] and adenovirus[3].

Sucrose gradients cause changes to the virus surface and poor resolution from cellular components

- Iodixanol gradients give much better resolution of HIV-1[4] and Herpes virus (HHV-6A)[5] from membrane vesicles; in iodixanol gradients contamination of HIV-1 by extracellular Vif is also substantially reduced, while infectivity is unimpaired[4].
- Sucrose gradients lead to loss of surface glycoproteins from retroviruses[6]. Electron microscopy of human endogenous retrovirus particles recovered from iodixanol gradients shows a much better retention of surface architecture[7]. Sucrose gradients strip the majority of glycoprotein gB from Kaposi's sarcoma associated herpes virus; this did not occur in Nycodenz gradients[8] (iodixanol is a close derivative of Nycodenz).

The Axis-Shield virus database

- Over 35 different viruses have been purified using OptiPrep. Presently 25 Application Sheets giving detailed protocols for virus purification (with full bibliographical back-up) are available from the Axis-Shield website: go to www.axis-shield-density-gradient-media.com and click on "Viruses", then select the virus of interest from the Index.

Web:
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