

## NIH AIDS Reagent Program

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## DATA SHEET

Reagent: 293T-CEP4SIVtat Cells

Catalog Number: 8123

Lot Number: Aug 20 2002

C Release Category:

Provided: 1 vial frozen cells in DMEM, 20% FCS and 10% DMSO.

Complete Dulbeccos minimal essential medium (DMEM) with 10% fetal calf serum (FCS) **Propagation** Medium:

and 1X penicillin-streptomycin and hygromycin B (250 µg/ml)

Freeze Medium: Propagation medium, 90%; DMSO, 10%.

Sterility: Negative for bacteria, mycoplasma, and fungi.

**Description:** 293T cells constitutively express SIVmne Tat expression driven by CMV IE promoter in

vector pCEP4SIVTat.

**Special** SIVmne mRNA coding for Tat was amplified from M. fascicularis PBMC using RT-PCR with specific primers beginning at the AUG initiation codon and ending at the UAG stop codon. Characteristics:

The SIV tat gene is 396 bases in length, comprised of exon 1 (1-296) and exon 2 (297-396). The complete SIV Tat gene PCR product was cloned into pCRII (Invitrogen) and then subcloned into pCEP4 (Invitrogen) between the *XhoI* and *Bam*HI sites. pCEP4SIVTat was transfected into 293T cells and cells were positively selected in

hygromycin B (250  $\mu$ g/ml) media for 26 passages.

Can be made semi-permanent through maintenance of episomal plasmid by selection in hygromycin B. Expression of Tat can be indirectly measured by co-transfection with SIV or HIV LTR- $\beta$ -gal plasmid and measurement of  $\beta$ -gal levels. Expression of Tat can be enhanced by cotransfection with plasmid expressing CMV IE protein (Peter Barry, UC,

Davis). Reagent pCEP4SIVTat in E. coli JM109 is also available (Cat# 8124).

Care and feeding of 293T and 293T-CEP4SIVtat cells

ALL RECIPIENTS OF THIS MATERIAL MUST COMPLY WITH ALL APPLICABLE BIOLOGICAL, CHEMICAL, AND/OR RADIOCHEMICAL SAFETY STANDARDS INCLUDING SPECIAL PRACTICES, EQUIPMENT, FACILITIES, AND REGULATIONS. NOT FOR USE IN HUMANS.

REV: 07/03/2018 Page 1 of 2 Recommended Storage:

Liquid nitrogen.

**Contributor:** Dr. Richard Grant.

**References:** Grant RF, Stevens Y, Wright N, Agy M, Thouless M, Morton W. Stable SIV Tat Cell Lines

Increase Expression Of Transfected Genes. J Med Primatol 28:(abstract), 1999.

**NOTE:** Acknowledgment for publications should read "The following reagent was obtained

through the NIH AIDS Reagent Program, Division of AIDS, NIAID, NIH: 293T-CEP4SIVtat Cells from Dr. Richard Grant." Also include the reference cited above in any publications.

Requests from commercial organizations should be directed to Ariadna A. Santander, Program Operations Coordinator, Office of Technology Licensing, University of Washington, 1107 N.E 45th Street, Suite 200, Seattle, WA 98195.

Last Updated July 03, 2018

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