

NIH AIDS Reagent Program

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DATA SHEET

Reagent: HIV-1 HXB2 GST-Tat Expression Vector (GST-Tat 1 (72R) C22G)

Catalog Number: 2362

Lot Number: 3

В Release Category:

Provided: Each clone is provided as 1 vial of ampicillin-resistant, transformed BL21 bacteria.

Cloning Vector: pGEX2T or pGEX2TK (Pharmacia).

Description: See attached Table. These constructs contain full length wild type or mutant HIV-1

(HXB2) or HIV-2 (ROD) tat genes. The clones contain a thrombin proteolytic site between the Schistosoma japonicum glutathione S-transferase (GST) sequence and the tat insert. Tat is expressed as a GST fusion protein. Constructs cloned into the pGEX2TK vector contain the phosphorylation site for the catalytic subunit of camp-dependent heart muscle kinase. GST-Tat fusions in this vector can thus be labeled directly in vitro to high

specific activities with (g32P)ATP and commercially available kinase (Sigma).

Table. HIV Tat Expression Vectors

Special

The HIV-1 constructs encode Tat in full length (86 aa), first exon (72 aa), activation domain (first 48 aa), or mutated forms. HIV-2 Tat is encoded as full length (130 aa), one exon (99 aa), or mutated protein. The GST-Tat fusion proteins can be easily purified Characteristics:

from E. coli lysates using a single-step procedure under non-denaturing conditions (see protocol, page 8). Tat can be cleaved from the fusion proteins and purified by thrombin

proteolytic digestion.

Protocol: Preparation and Purification of HIV-1/2 Tat

Recommended Storage:

-90°C.

Contributor: Dr. Andrew Rice.

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: 07/12/2018 Page 1 of 2 References: Herrmann CH, Rice AP. Lentivirus Tat proteins specifically associate with a cellular protein

kinase, TAK, that hyperphosphates the carboxyl-terminal domain of the large subunit of

RNA polymerase II: candidate for a Tat cofactor. J Virol 69:1612-1620, 1995.

Rhim H, Echetebu CO, Herrmann CH, Rice AP. Wild type and mutant HIV-1 and HIV-2 Tat

proteins expressed in Escherechia coli as fusions with glutathione S-transferase. J

Acquired Immune Defic Syndr 7:1116-1121, 1994.

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

through the NIH AIDS Reagent Program, Division of AIDS, NIAID, NIH: (specify reagent) from Dr. Andrew Rice." Also include the references cited above in any publications.

Last Updated: July 12, 2018

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