

NIH AIDS Reagent Program

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DATA SHEET

Reagent:	HIV-1 A244 gp120 Secreting CHO Cells (6.4D)
Catalog Number:	13577
Lot Number:	200304
Release Category:	D
Provided:	600 µL of cells
	Post thaw cell count = 2.96×10^6 cells/vial
	Post thaw cell viability = 87%
Cell Type:	Chinese hamster ovary cell line derived from CHO-S cells.
Propagation Medium:	CD-CHO Medium; 8mM Glutamax
Freeze Medium:	Gibco Recovery™ Cell Culture Freezing Medium
Morphology:	Adherent epithelial-like Cell Line
Sterility:	Negative for mycoplasma, bacteria, and fungi
Description:	Suspension adapted cell line, useful for the production of A244_N322-rgp120.
Special Characteristics:	This high yielding cell line secretes high levels (>1 g/L) A244-N332-rgp120 into cell culture medium when grown in shake flask cultures. gp120 from this cell line binds multiple broadly neutralizing antibodies including PG9, PGT128, 10-1074, VRC01. This protein is expressed with an Nterminal gD purification tag and is very similar to the one that was used in the VaxGen and RV144 HIV vaccine trials. However, it differs from most gp120s used in clinical trials to date in that exhibits superior binding of broadly neutralizing monoclonal antibodies by virtue of the fact that N-linked glycosylation is restricted

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.

	primarily to mannose-5 and earlier intermediates in the N-linked glycosylation pathway. Because of restricted glycosylation, gp120 from this cell line can be purified by conventional column chromatography without the need for antibody or lectin affinity chromatography. HIV-1 A244 gp120 Secreting CHO Cells (6.4D) are potentially suitable for GMP production.
Recommended Storage:	Keep the reagent in liquid nitrogen.
Contributor:	Dr. Phillip Berman
References:	Byrne, G., O'Rourke, S. M., Alexander, D. L., Yu, B., Doran, R. C., Wright, M., Chen, Q., Azadi, P. and Berman, P. W. (2018). CRISPR/Cas9 gene editing for the creation of an MGAT1-deficient CHO cell line to control HIV-1 vaccine glycosylation. PLoS Biol, (8), e2005817. doi:10.1371/journal.pbio.2005817 <u>PUBMED</u>
	O'Rourke, S. M., Byrne, G., Tatsuno, G., Wright, M., Yu, B., Mesa, K. A., Doran, R. C., Alexander, D. and Berman, P. W. (2018). Robotic selection for the rapid development of stable CHO cell lines for HIV vaccine production. PLoS ONE, (8), e0197656. doi:10.1371/journal.pone.0197656 <u>PUBMED</u>
	O'Rourke, S. M., Yu, B., Morales, J. F., Didinger, C. M., Alexander, D. L., Vollmers, C. and Berman, P. W. (2019). Production of a recombinant monoclonal antibody to Herpes Simplex Virus glycoprotein D for immunoaffinity purification of tagged proteins. J Immunol Methods, 31-38. doi:10.1016/j.jim.2018.11.015 <u>PUBMED</u>
	Doran, R. C., Yu, B., Wright, M., O'Rourke, S. M., Yin, L., Richardson, J. M., Byrne, G., Mesa, K. A. and Berman, P. W. (2018). Development of a Stable MGAT1(-) CHO Cell Line to Produce Clade C gp120 With Improved Binding to Broadly Neutralizing Antibodies. Front Immunol, 2313. doi:10.3389/fimmu.2018.02313 <u>PUBMED</u>
	Li, S. W., Yu, B., Byrne, G., Wright, M., O'Rourke, S., Mesa, K. and Berman, P. W. (2019). Identification and CRISPR/Cas9 Inactivation of the C1s Protease Responsible for Proteolysis of Recombinant Proteins Produced in CHO Cells. Biotechnol Bioeng, (9), 2130-2145. doi:10.1002/bit.27016 <u>PUBMED</u>
NOTE:	Acknowledgment for publications should read "The following reagent was obtained through the NIH AIDS Reagent Program, Division of AIDS, NIAID, NIH: HIV-1 A244 gp120 Secreting CHO Cells (6.4D) from Dr. Phillip Berman (cat# 13577)." Also include the references cited above in any publications.
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