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DATA SHEET

Reagent:	Panel of Multi-Protease Inhibitor Resistant Infectious Molecular Clones
Catalog Number:	12740
Lot Number:	150175
Release Category:	A
Provided:	5 µg of each plasmid DNA. See Table 1 for included clones.
Cloning Vector:	See pNL4.3 GenBank accession AF324493.1 for the vector sequence. The vector contains an AmpR marker.
Cloning Site:	Restriction maps match those of pNL4.3 with the exception of a novel Bal I site at nucleotide position 4552 and PflM 1 site at nucleotide position 5302 that were removed and a PflM 1 site added at nucleotide position 3491. The unique Apa I site upstream of protease at nucleotide position 2009 is used as a 5' restriction site; double-digestion with Apa I/Bal I generates a protease-deleted linearized vector. Inserts are amplified from clinically-derived viral cDNA using Pfu; Purified PCR products are digested and ligated into the vector using cycling T4 ligation, and the ligation product is grown in Stbl3 E. coli in LB+carbenicillin, then plated and sequenced. The cloned region contains the entire protease open reading frame; the flanking regions cloned along with protease include 3' end of gag with the gag cleavage site, and the 5' end of RT.
GenBank:	See Table 1 for individual accession numbers.
Host Strain:	Stbl3
Description:	Clones in this panel contain each of the canonical protease inhibitor-resistance pathways in clinically derived HIV-1.

Special Characteristics:	In contrast to site-directed mutants, the mutations are present in their naturally occurring genetic contexts, which may include known accessory drug-resistance mutations, as well as changes at positions that are not currently known to be associated with drug resistance. As the clones are also infectious and replication-competent, they can be used for in vitro susceptibility testing of new protease inhibitors (PIs). PIs that are active against these clones are likely to retain activity against the most clinically relevant, or possibly all, protease inhibitor-resistant variants. Researchers can also create their own recombinant viruses using the pNLPFB vector. A protocol is available upon request (please contact Shafer Lab) TABLE 1. Panel of Multi-Protease Inhibitor Resistant Infectious Molecular Clones
Recommended Storage:	Keep at -20°C or lower. Avoid freeze-thaw cycles as reagent degradation may result.
Contributor:	Robert Shafer
References:	Prototypical Recombinant Multi-Protease-Inhibitor-Resistant Infectious Molecular Clones of Human Immunodeficiency Virus Type 1 Vici Varghese, Yumi Mitsuya, W. Jeffrey Fessel, Tommy F. Liu, George L. Melikian,a David A. Katzenstein, Celia A. Schiffer, Susan P. Holmes, and Robert W. Shafer. Antimicrob Agents Chemother. 2013 September; 57(9): 4290–4299. doi: 10.1128/AAC.00614-13. PMID: 23796938.
NOTE:	Acknowledgment for publications should read "The following reagent was obtained through the NIH AIDS Reagent Program, Division of AIDS, NIAID, NIH from Dr Robert Shafer: Cat# 12464, pV20742-4" Also include the reference cited above in any publications.
Last Updated:	December 21, 2018