

NIH AIDS Reagent Program

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DATA SHEET

Reagent:	HIV-1 NL4-3 Integrase Expression Vector (pINSD.His.Sol)
Catalog Number:	2958
Lot Number:	190104
Release Category:	C
Provided:	5 μg of dried purified DNA stabilized in DNAstable Plus
Cloning Vector:	pET15B
	Ampicillin resistant
Description:	An expression vector which produces HIV-1 NL4-3 integrase protein that was modified to include the amino acid substitutions F185K and C280S.
Special Characteristics:	This construct is 6565 bp including the insert. HIV-1 NL4-3 integrase from pINSD (ARP Cat# 2820) with an amino-terminal polyhistidine tag and F185K and C280S amino acid substitutions is expressed upon transformation of pINSD.His.Sol into <i>E. coli</i> BL21(DE3) and IPTG induction. The two amino acid substitutions greatly improve the solubility of HIV-1 integrase without affecting <i>in vitro</i> activities. The expressed integrase may be purified by Ni-affinity chromatography with a high yield (5-10 mg/liter of culture) from the soluble fraction of <i>E. coli</i> lysates. The polyhistidine tag may be subsequently removed by cleavage with thrombin. The integrase is functional in <i>in vitro</i> integration assays. <i>E. coli</i> strain BL21(DE3) must be used for integrase expression, but the plasmid may be unstable in this host. Sequence file lot 190104 Plasmids can be propagated in STBL2 cells and grown at 37°C. Larger plasmids may benefit from growth at 30°C. This construct may also be grown in other competent cells. This reagent is currently being provided as dried purified DNA stabilized in DNAstable <i>PLUS</i> . Please see the notice for additional information and the protocol for reconstitution of dried DNA reagents. <u>Dried DNA Notice</u>

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.

Recommended Storage:	Keep the reagent at room temperature in a dry storage cabinet or in a moisture barrier bag.
Contributor:	Dr. Robert Craigie
References:	Jenkins, T. M., Engelman, A., Ghirlando, R. and Craigie, R. (1996). A soluble active mutant of HIV-1 integrase: involvement of both the core and carboxyl-terminal domains in multimerization. J Biol Chem, 271(13), 7712-8. <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 NL4-3 Integrase Expression Vector (pINSD.His.Sol) from Dr. Robert Craigie (cat# 2958)." Also include the reference cited above in any publications.
	Scientists at for-profit institutions or who intend commercial use of this reagent must contact the Technology Advancement Office (TAO) at the following email address: <u>MTA@niddk.nih.gov</u> , before the reagent can be released.
Last Updated:	March 26, 2020

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