



Product Information Sheet

Product Name:	WNV NS3 Protease, recombinant
Catalog Number:	AS-72081-100
Size:	100 µg
Concentration:	100 µg/ml
Activity (Unit/µg):	Provided on the label.
Unit definition:	One unit of protease hydrolyzes 1 picomole of Pyr-RTKR-AMC substrate from SensoLyte™ 440 West Nile Virus Protease Assay Kits (AnaSpec Cat# AS-72079) per minute at pH 8.0 at 30° C.
Storage:	Store at -80°C. Avoid multiple freeze-thaw cycles.

Instruction:

West Nile virus (WNV) is a member of the flavivirus genus, which contains many significant human pathogens including Dengue virus (Den), Japanese Encephalitis virus (JE), and Yellow Fever virus (YF). WNV is a small, enveloped virus with a single stranded, positive sense 11kb RNA genome, which encodes a polyprotein precursor. This polyprotein must be cleaved co- and post-translationally to produce ten functional proteins: three structural (C, prM and E) and 7 nonstructural (NS1, NS2A, NS2B, NS3, NS4A, NS4B and NS5).¹ WNV NS3 protease is absolutely essential (along with viral-encoded cofactor NS2B) for post-translational cleavage and viral replication. As a result, this protease is a potential therapeutic target.²⁻⁵

C-terminal his-tagged recombinant WNV NS3 protease (residues 1 to 184) was expressed as a fusion protein with the cofactor NS2b (residues 49 to 96) and a linker sequence (GGGGSGGGG) in E. coli. The apparent Mr on SDS-PAGE is 32-kDa. AnaSpec offers WNV assay kits (Cat#AS-72079, Cat#AS-72080) for screening of WNV inhibitors and for continuous assay of WNV activity. 10-20 ng of enzyme is sufficient for FRET-based assay. WNV NS3 protease is stored in 25 mM Tris-HCl, pH 7.5, 150 mM NaCl. The purity of enzyme is >95% as estimated by SDS-PAGE.

Note: Purified recombinant protein has autocatalytic activity, which can cause protein degradation (faint 22 kD and 11 kD fragments can be visible on SDS PAGE).

Therefore, it is important to store the product below -20° C and to keep it on ice while working with the product.

References:

1. Brinton, MA. *Annu Rev Microbiol.* 56, 371 (2002)
2. Tyndall, JD. et al. *Chem. Rev.* 105, 973 (2005).
3. Loughlin, WA. et al. *Chem. Rev.* 104, 6085 (2004)
4. D'Arcy, A. et al. *Acta Cryst.* F62, 157 (2006).
5. Nall, TA. et al. *J. Biol. Chem.* 279, 48535 (2004).