

Data Sheet

DESCRIPTION:	Recombinant Nef protein (B-clade consensus sequence)
REPOSITORY REFERENCE:	ARP692.1 ARP692.2 (Low Endotoxin)
CHARACTERISTICS:	Recombinant Nef-B protein is expressed as His tag-fusion protein in <i>E. coli</i> using expression vector pET24 and purified using Ni-agarose columns (Qiagen). Endotoxins were removed by phase separation using Triton X-114 (described by Liu et al. <i>Clinical Biochem.</i> 30, 1997, p. 455)
SOURCE:	FIT Biotech Oyj Plc Eesti Filiaal, Tartu, Estonia
PRESENTATION:	100ug. In PBS +10% glycerol (may contain traces of Triton X-114). ARP692.1 (1.7mg/ml) ARP692.2 (1.7mg/ml)
NOTE:	Prepared for AVIP collaborative study. Non AVIP members are restricted to 1 vial per request.
PURITY:	See Fig.1 for SDS-PAGE and Fig.2 for Western blot analysis.
STORAGE:	Store at -70°C or less. Avoid multiple freeze-thaw cycles as product degradation may occur.

ACKNOWLEDGEMENTS:

Publications should acknowledge the donor of the reagent and the Programme EVA Centre for AIDS Reagents. Suggested wording can be found on our website at <http://www.nibsc.ac.uk/spotlight/aidsreagent/index.html> in the “Acknowledgements” section.

Please also ensure that you send us a copy of any papers resulting from work using reagents acquired through CFAR (this can be electronically or as a paper copy)

PROTEIN SEQUENCE:

MGGKWSKRSVVGWPTVRERMRAEPAADGVGAVSRDLEKHGAISSNTAANNADCAWLEAQE
EEEVGFVVRPQVPLRPMTYKAAVDLSHFLKEKGGLEGLIYSQKRQDILDLWVYHTQGYFPDWQN
YTPGPGIRYPLTFGWCFKLVPEPEKVEEANEGENNSLLHPMSLHGMDDPEREVLVWKFDSRLAF
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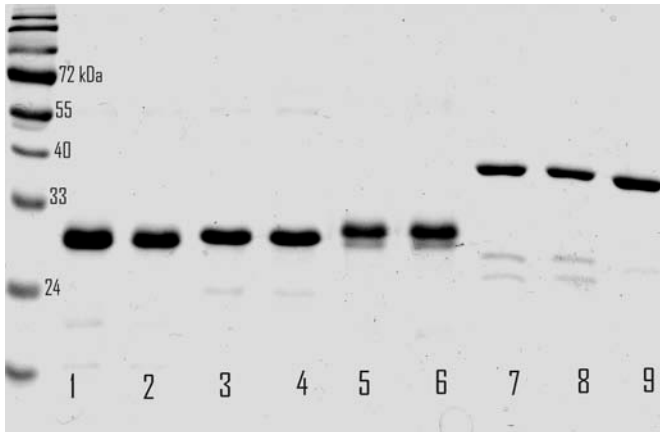


Figure 1. SDS-PAGE analysis of purified proteins (Coomassie staining)

Lane 1- Nef-A,	1 μ g
Lane 2- Nef-A low endo,	1 μ g
Lane 3- Nef-B	1 μg
Lane 4- Nef-B low endo	1 μg
Lane 5- Nef-C	1 μ g
Lane 6- Nef-C low endo	1 μ g
Lane 7- p17/24-B	1 μ g
Lane 8- p17/24-B low endo	0.8 μ g
Lane 9- p17/24-C	1 μ g

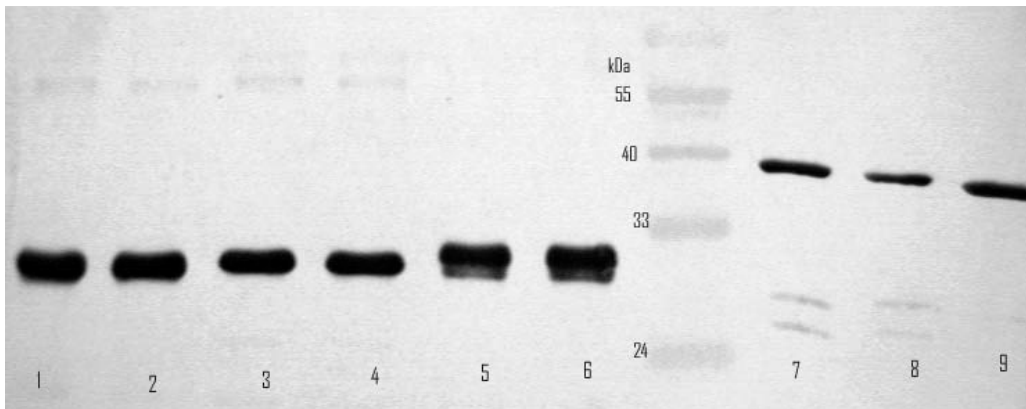


Figure 2. Western blot analysis of purified proteins, detected by monoclonal antibody against His-tag.

Lane 1- Nef-A	0.5 μ g
Lane 2- Nef-A low endo	0.5 μ g
Lane 3- Nef-B	0.5 μg
Lane 4- Nef-B low endo	0.5 μg
Lane 5- Nef-C	0.5 μ g
Lane 6- Nef-C low endo	0.5 μ g
Lane 7- p17/24-B	0.5 μ g
Lane 8- p17/24-B low endo	0.4 μ g
Lane 9- p17/24-C 1 μ g	0.5 μ g