



### Certificate of Analysis

#### Rhesus/Cynomolgus Interferon Alpha 2

**Catalog No:** 14110-1

**Lot No:** 7320

**Size:** ≥ 1 x 10<sup>5</sup> units/vial

**Description:** Recombinant Rhesus/Cynomolgus Interferon Alpha 2 (Rhesus/Cynomolgus IFN-α2)

**Volume:** 0.1 ml

**Activity:** 3.60 x 10<sup>6</sup> units/ml (MDBK/VSV)

**Specific Activity:** 1.89 x 10<sup>8</sup> units/mg (MDBK/VSV)

**Buffer:** Phosphate buffered saline (PBS) containing 0.1% bovine serum albumin (BSA)

**Endotoxin:** < 1 EU/μg

**Molecular Weight:** 19.5 kDa

**Purity:** > 98%

**Source:** Rhesus cDNA expressed in *E. coli*

**Accession #:** XM\_001107516

**Assay Used to Measure Bioactivity:** Interferon was titrated with the use of the cytopathic effect inhibition assay as listed. The protein has similar activity to Human IFN Alpha 2 on human, rhesus and bovine cells.

*Bovine (MDBK/VSV)* – performed as described [Rubinstein *et al.* (1981) *J. Virol.* 37(2):755] The EC<sub>50</sub> for interferon in this assay is ~5 U/ml.

*Human (A549/EMCV)* – performed as described [Budd *et al.* (1985) *Canc. Chem. Pharm.* 12:39]. The EC<sub>50</sub> for interferon in this assay is ~1 U/ml. **Activity:** 7.44 x 10<sup>6</sup> units/ml **Specific Activity:** 3.91 x 10<sup>8</sup> units/mg.

*Rhesus (LLC-MK2/VSV)* – Comparative studies using LLC-MK2 cells versus A549 cells have shown that the activity of the protein on A549 cells is indicative of its activity on LLC-MK2 cells.

As there is no international Monkey Interferon Alpha standard, the units are determined by use of a Human IFN Alpha A (Hu-IFN-αA [2a]) laboratory standard calibrated to the international reference standard for Human Interferon Alpha A (Hu-IFN-αA [2a]) provided by the National Institutes of Health [Meager *et al.* (2001). *J Immunol. Methods* 257(1-2):17-33].

**Product Information:** The cDNA for Rhesus/Cynomolgus IFN-α2 was cloned from the Rhesus cell line LLC-MK2. This was found to be identical to the putative Rhesus IFN-α2 (XM\_001107516) identified as part of the Rhesus genome project (<http://www.hgsc.bcm.tmc.edu/projects/rmacaque>). There are 14 AA differences in the mature protein (2 in the signal peptide) between the Rhesus and Human Alpha A (2a) sequence. There are 13 AA differences in the mature protein (2 in the signal peptide) between the Rhesus and Human Alpha 2 (2b) sequence. This is in accord with a previous report [Villinger *et al.* (1995) *J. Immunol.* 155:3946] stating that there are 15-16 amino acids different between the Rhesus IFN alpha 2 and human alpha 2 (2b) and human alpha A(2a) respectively.

This Rhesus (*Macaca mulatta*) sequence is also identical to a Cynomolgus (*Macaca fascicularis*) IFN-α2 cloned from cDNA isolated from primary hepatocytes (PBL unpublished observation).

Comparison of the Mature Sequences of Human IFN Alpha 2b and Rhesus IFN Alpha 2

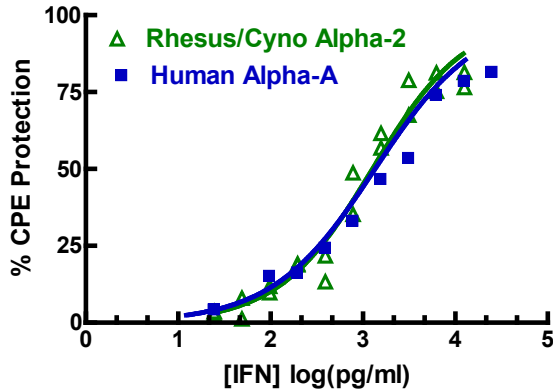
Rhesus 1	CDLPQTHSLG	NRRTLMLLAQ	MRRISLFFCL	KDRHDFEFPQ	EEFGNQFQKA	QTIPVLHEMI
Human 1	.....S.....	.....S.....	.....G....	.....E.....		



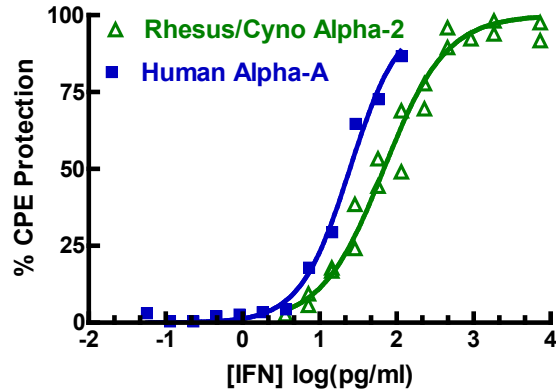
# pbl assay science

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Rhesus 61	QQTFNLFSTK	DSSAAWDETL	LNKFYTELYQ	QLNDLEACVM	QEMGVTETPL	MNKNSILAVR
Human 61	..I.....	.....	.D.....	.....I	.GV.....	.KED.....
Rhesus 121	KYFQRITLYL	KEKKYSLCAW	EVVRAEIMRS	FSLSTNLQES	LSRKE	
Human 121	.....	.....P.....	.....	.....	.....	



**Figure 1:** Activity of Rhesus/Cyno and Human IFN Alpha A on rhesus cells (LLC-MK2) challenged with VSV.



**Figure 2:** Activity of Rhesus/Cyno and Human IFN Alpha A on human cells (A549) challenged with EMCV.

Results are representative and may vary depending upon experimental conditions.

**Shipping Conditions:** Dry Ice

**Physical State of Product During Shipping:** Frozen

**Storage Conditions/Comments:** After receipt, the product may be stored at -20°C for short-term use (≤ 6 months). For long-term storage, we recommend storing the product at -70°C or below for retention of full activity. When thawing, the contents of the tube should be apportioned in separate tubes so that freezing and thawing is kept to a minimum. Refreezing should be done on dry ice or in a dry ice/alcohol bath. Further dilution of the product should be in buffers containing protein such as 0.1% bovine serum albumin (BSA). For more information on protein handling, visit our Resource Library at [www.pbl assaysci.com](http://www.pbl assaysci.com).

### Authorization

Released by: \_\_\_\_\_

Date: January 20, 2021

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