

# Influenza A Virus (IAV) Nucleoprotein Antigen Calibration Kit

## Product Insert

FOR RESEARCH USE ONLY

Catalog No.: NCP-1050

### Introduction:

The ImmTech Influenza A Nucleoprotein Antigen Capture ELISA kits (IAV-1192, IAV-1480) are highly sensitive and specific semi-quantitative assays for the detection of Influenza A nucleoprotein in a wide variety of samples. To enhance the analytical utility of the ELISA kits, ImmTech has developed the Influenza A Nucleoprotein Antigen Calibration Kit, which permits the generation of a nucleoprotein calibration curve and estimation of nucleoprotein in a sample.

The Influenza A Nucleoprotein Antigen Concentrate supplied in this product was produced using A/Beijing/262/95 (H1N1). The virus was grown in embryonated eggs, detergent-treated to remove the viral envelope and concentrated by centrifugation. Following SDS-PAGE, nucleoprotein concentration was calculated as a percentage of the total protein in the preparation.

### Intended Use:

The ImmTech Influenza A Nucleoprotein Antigen Calibration Kit is intended for the production of a Nucleoprotein Antigen calibration curve and is designed for use with the ImmTech Influenza A Nucleoprotein Antigen Capture ELISA Kits (IAV-1192, IAV-1480).

### Kit Components:

1. Influenza A Virus Nucleoprotein Antigen Concentrate (250 ng/ml)-0.2 ml  
Store at -20 deg. C
2. Influenza A Virus Nucleoprotein Antigen Diluent (1x)-50 ml  
Store at 2-10 deg. C

### Procedure:

The ImmTech Influenza A Nucleoprotein Antigen Concentrate is supplied at 250 ng/ml and requires dilution prior to use. For the generation of a calibration curve, prepare an initial dilution of 1:50 in the diluent supplied. This will result in a nucleoprotein concentration of 5 ng/ml. Serial 2-fold dilutions should then be prepared from the initial dilution (see chart below). Depending upon the nature of the sample being tested, it may be necessary to prepare dilutions in the diluent/buffer used for the experiment (tissue culture medium, for example).

NP (ng/ml)	NP Concentrate	Diluent
5	20 ul @ 250 ng/ml	980 ul
2.5	500 ul @ 5 ng/ml	500 ul
1.25	500 ul @ 2.5 ng/ml	500 ul
0.625	500 ul @ 1.25 ng/ml	500 ul
0.313	500 ul @ 0.625 ng/ml	500 ul
0.156	500 ul @ 0.313 ng/ml	500 ul
0.078	500 ul @ 0.156 ng/ml	500 ul
0	-----	500 ul

Test each dilution in duplicate following the product insert for the appropriate ImmTech Influenza A Nucleoprotein Antigen Capture ELISA Kit.

To determine the amount of nucleoprotein in a sample, a standard curve is constructed as follows:

1. Calculate the average value for each antigen concentration and then subtract the 0 ng/ml value from each.
2. Plot the resulting data and read the unknown values by extrapolation. Alternatively, enter the data into a calculator or a software program and calculate the sample concentrations by linear regression.

### Representative Calibration Curve:

