

The Synbiotics ProFLOK® **Newcastle Disease Virus (NDV) ELISA kit** is a rapid and specific USDA-licensed test for the detection of NDV antibodies in turkey serum.

NDV infections vary from mild subclinical infection to fulminating disease with high mortality<sup>1</sup>. The Synbiotics T-NDV ELISA kit is designed for monitoring NDV vaccination programs and detecting NDV infected turkey flocks.

## Kit Performance

The Synbiotics T-NDV ELISA kit has been extensively tested for the following parameters:

### 1. Specificity

The results shown in Table 1 below indicate that the Synbiotics T-NDV ELISA kit demonstrated excellent specificity (i.e. no false positive reactions) to reference turkey sera for numerous infections agents of turkeys. The Synbiotics T-NDV ELISA kit, like all Synbiotics ELISA kits, is highly specific to provide valid, reproducible test results.

**Table 1. Specificity.**  
Average sample-to-positive (SP) ratio and titer values for a reference serum panel assayed with the SBIO T-NDV ELISA test.

Reference Turkey Serum: Ref. No.	Average SP <sup>A,B</sup> Ratio Values	SBIO T-NDV ELISA SP Range <sup>C</sup>	SBIO T-NDV ELISA Titer Values <sup>D</sup>
Normal turkey control (NTS):193	0.000	-	0
Specific pathogen free (SPF):118	0.000	-	0
<u>Bordetella avium</u> (BA):109	0.007	-	0
Hemorrhagic enteritis virus (HEV):112	0.018	-	0
Avian influenza virus (AIV):116	0.035	-	0
Avian influenza virus (AIV):117	0.000	-	0
<u>Mycoplasma meleagridis</u> (Mm):209; 20 HI Titer	0.007	-	0
<u>Mycoplasma synoviae</u> (Ms):111; NA HI Titer	0.016	-	0
<u>Mycoplasma gallisepticum</u> (Mg):201; 80 HI Titer	0.010	-	0
Newcastle disease virus (NDV):195; 20 HI Titer	<b>1.000</b>	<b>+</b>	<b>8054</b>
Newcastle disease virus (NDV):113; NA HI Titer	<b>1.330</b>	<b>+</b>	<b>9881</b>
Newcastle disease virus (NDV):198; 160 HI Titer	<b>2.720</b>	<b>+</b>	<b>16503</b>

<sup>A</sup>Values are the arithmetic mean of 5 replicate samples

<sup>B</sup>Sample SP Ratio =  $\frac{\text{Optical Density (OD) sample} - \text{Avg. OD Normal Control}}{\text{Avg. OD Positive Control} - \text{Avg. OD Normal Control}}$

<sup>C</sup>SBIO T-NDV ELISA SP threshold ranges: Negative (-) =  $\leq 0.150$   
Positive (+) =  $> 0.150$

<sup>D</sup>Log<sub>10</sub> titer =  $(0.717 \times \text{Log}_{10} \text{SP}) + 3.906$

NA = not available

### 2. Sensitivity

The data shown in Table 2 below indicate that the Synbiotics T-NDV ELISA demonstrated excellent sensitivity when compared to the conventional NDV HI test results from two laboratories. Additionally, the Synbiotics T-NDV ELISA kit provides standardized test reagents and consistent results for improved inter-laboratory results:

**Table 2. Sensitivity.**  
Comparison of SBIO T-NDV ELISA sample-to-positive (SP) ratio and titer values, and NDV hemagglutination-inhibition (HI) titers for an NDV sensitivity serum panel assayed with SBIO T-NDV ELISA kit and the NDV HI test in two commercial laboratories.

Log 2	Dilution Ratio	SBIO T-NDV SP Values <sup>A</sup>	SBIO T-NDV SP Range <sup>B</sup>	SBIO T-NDV Titer Values <sup>A,E</sup>	NDV HI Titers (from 2 laboratories)	
					Lab A <sup>C</sup>	Lab B <sup>D</sup>
0	neat	2.66	+	16198	128	160
-1	1:2	1.98	+	13143	128	160
-2	1:4	1.52	+	10874	64	93
-3	1:8	1.07	+	8454	64	40
-4	1:16	0.72	+	6364	32	20
-5	1:32	0.46	+	4615	16	10
-6	1:64	0.27	+	3150	8	10
-7	1:128	0.15	-	0	8	5
					r <sup>2</sup> = .90	r <sup>2</sup> = .88

<sup>A</sup>Values are the arithmetic mean of 30 replicate samples

<sup>B</sup>SBIO T-NDV ELISA SP threshold ranges: Negative (-) =  $\leq 0.150$   
Positive (+) =  $> 0.150$

<sup>C</sup>Values are the arithmetic mean of 2 replicate samples

<sup>D</sup>Values are the arithmetic mean of 3 replicate samples

<sup>E</sup>Log<sub>10</sub> titer =  $(0.717 \times \text{Log}_{10} \text{SP}) + 3.906$

### 3. Reproducibility

The data presented in Table 3 below demonstrate the excellent intra- and inter-plate reproducibility of the Synbiotics T-NDV ELISA kit. All Synbiotics ELISA kits are optimized to provide valid, reproducible results test after test, day after day.

Table 3. Intra-plate and inter-plate reproducibility: Average optical density (OD) values and percent coefficient of variation (CV%) for a single NDV turkey serum assayed 42 times on each of five SBIO T-NDV ELISA plates.

SBIO T-NDV ELISA Plate	Average OD <sup>A</sup>	CV(%)	Avg. Intra-Plate Variation (%)	Avg. Inter-Plate Variation (%)
1	0.694	4.21	4.53	3.77
2	0.548	4.54		
3	0.573	4.04		
4	0.545	4.67		
5	0.537	5.19		

<sup>A</sup>Values are the arithmetic mean of 42 replicate testings of a single NDV turkey serum per each SBIO T-NDV ELISA plate.

### 4. Benefits

The Synbiotics T-NDV ELISA kit offers the following benefits and features:

- Reliable, consistent results
- USDA-licensed
- High-volume, easy-to-use format
- Long shelf life. All Synbiotics kits have an 18-month shelf life.
- Powerful database management software system
- Excellent field-oriented technical support

### Suggested Uses

Suggested uses for the Synbiotics T-NDV ELISA kit:

1. NDV Vaccination Evaluation:
  - Randomly collect and assay 30 or more serum samples per turkey flock immediately prior to NDV vaccine administration (pre-vaccination) and 14 to 18 days post vaccination.
2. Routine NDV Flock Profiling:
  - Collect and assay 30 or more serum samples per turkey flock, particularly breeder hen flocks, on a routine basis (i.e. every four to six weeks).

### Interpreting Results

Evaluate Synbiotics T-NDV ELISA results for the following

1. Uniformity
  - Measured by the coefficient of variation value (CV%).
  - The lower the CV% value for a flock tested, the better the titer value uniformity.
  - Strive for the best 1-3 day old, pre-vaccination, and post-vaccination NDV titer value uniformity (i.e. CV% of less than 50%).
2. Titer Values

SP Threshold: Each Synbiotics ELISA kit has an sample-to-positive (SP) value threshold that clearly separates positive samples from negative samples. A negative sample is one that is not significantly different than the T-NDV kit normal control serum (NCS). The SP threshold for the T-NDV ELISA is as follows:

SP Range	Titer Range
0.150 or less	0
0.151 and greater	2076 and greater

Vaccination Evaluation: Compare pre- and post-vaccination ELISA average mean titers, geometric mean titers and %CV values. A flock geometric mean titer following vaccination of 7000 is considered to be “protective”. Note: Postvaccinal NDV ELISA values depend on a variety of factors such as NDV vaccine strain, route of administration, age of bird, etc. The goal of any vaccination program should be to achieve as uniform postvaccinal titer values for each vaccinated flock (i.e. %CV 45 or less) as possible.

Flock Profiling: Review ELISA flock profiles and correlate NDV ELISA titer levels to vaccination program, flock economic performance data (body weight gain, feed conversion, mortality, etc.) and presence or absence of field infection.

### Reference

1. Beard, C.W. and R.P. Hanson, Newcastle Disease In: Diseases of Poultry, 8th ed. Am. Assoc. of Avian Path. Iowa State Univ. Press, Ames, IA pp. 212-220. 1984.