BioChek
SMART VETERINARY DIAGNOSTICS

NDV

Data Pack

Newcastle Disease Antibody Test Kit
(Detects antibodies to PMV 1)

CONTENTS

SUMMARY

Kit

- 5 plates
- Indirect ELISA
- Run at room temperature
- Incubation times: 30-30-15
- Read at: 405nm
- 1:500 dilution

Key Performance Features

General:

- Detects antibodies to avian paramyxo virus type 1 in Chicken or Turkey sera
- Same kit for Turkeys and chickens
- Picks up antibodies to NDV 7 - 14 days after challenge or vaccination. Good affinity for both Hitchner B1, LaSota and Ulster strain vaccines.

Correlation mean Flocktiters with HI		Correlation individual titers with HI	
Commercial layers	93%	Commercial layers	83%
Broilers	91%	Broilers	80%
Turkeys	92%	Turkeys	85%

Specificity:

Specificity (broilers, SPF Layers)	99-100%

Sensitivity:

Sensitivity live vaccinations (2 x)	100 %

Applications

Vaccination check:
Test flock after vaccination in order to establish efficiency of vaccination. Answers to key questions like “did the vaccine actually stimulate the immune system” and how well the vaccine spread in the flock can be found by testing 2 - 5 weeks after vaccination. See our interpretation manual for details on expected response to vaccination.

Determine best moment to vaccinate:
When vaccinating through drinking water, it's very important that maternal antibodies are at a low level. When vaccinating at a high maternal antibody level, the vaccine will be partially neutralized and no or little serological response will take place. However you don't want the chicks to be unprotected, therefor you would want to vaccinate while there still are protective antibodies. Users should determine their optimum moment to vaccinate. We expect this will be at a BC NDV titre between 1000 and 2000.

Field infection:
About 10 - 20 days after infection seroconversion will show.

BioChek Poultry Immunoassays

Newcastle Disease Antibody Test Kit

Catalogue Code CK 116

Description of Test

The NDV ELISA kit will measure the amount of antibody to NDV in the serum of chickens. Microtitre plates have been pre-coated with inactivated NDV antigen. Chicken serum samples are diluted and added to the microtitre wells where any anti- NDV antibodies present will bind and form an antigen-antibody complex. Non specific antibodies and other serum proteins are then washed away. Anti-chicken IgG labelled with the enzyme alkaline phosphatase is then added to the wells and binds to any chicken anti- NDV antibodies originally bound to the antigen. After another wash to remove unreacted conjugate, substrate is added in the form of pNPP chromogen. A yellow colour is developed if anti- NDV antibody is present and the intensity is directly related to the amount of anti- NDV present in the sample.

Reagents provided

1. **NDV Coated plates**. Inactivated viral antigen on microtitre wells.
2. **Conjugate reagent**. Sheep anti-Chicken: Alkaline Phosphatase in Tris buffer with protein stabilisers, inert red dye and sodium azide preservative (0.1% w/v)
3. **Substrate tablets**. PNPP (p-Nitrophenyl Phosphate) tablets to dissolve with Substrate buffer.
4. **Substrate buffer**. Diethanolamine buffer with enzyme co-factors
5. **Stop Solution**. Sodium Hydroxide in Diethanolamine buffer
6. **Sample Diluent**. Phosphate buffer with protein stabilisers and sodium azide preservative (0.1% w/v)
7. **Wash Buffer**. Powdered Phosphate Buffered Saline with Tween
8. **Negative control**. Specific Pathogen Free serum in Phosphate Buffer with protein stabilisers and sodium azide preservative (0.1% w/v)
9. **Positive Control**. Antibodies specific to NDV in Phosphate Buffer with protein stabilisers and sodium azide preservative (0.1% w/v)

Materials and Equipment Required (not provided with kit)

Precision Pipettors and disposable tips
8 or 12 channel pipette / repeater pipette
Plastic tubes for sample dilution
Distilled or deionised water
Microtitre Plate Reader with 405 nm filter
Microtitre Plate Washer

Warnings and Precautions

1. Handle all reagents with care. STOP SOLUTION contains STRONG ALKALI which can be CAUSTIC. If in contact with skin or eyes, wash with copious amounts of water.
2. Treat all biological materials as potentially biohazardous, including all field samples. Decontaminate used plates and waste including washings with bleach or other strong oxidising agent before disposal.
3. NEVER pipette anything by mouth. There should be no eating, drinking or smoking in areas designated for using kit reagents and handling field samples.
4. This kit is for IN VITRO use only.
5. Strict adherence to the test protocol will lead to achieving best results.

Reagent preparation

1. **Substrate Reagent**. To make Substrate Reagent, add 1 tablet to 5.5 ml of Substrate Buffer and allow to mix for 3 minutes or until fully dissolved. The prepared reagent should be made on day of use *but will be stable for one week if kept in dark at +4 ℃.*

*D*rop tablets into clean container and add appropriate volume of Substrate Buffer
DO NOT HANDLE TABLETS WITH BARE FINGERS

2. **Wash Buffer**. Empty the contents of one wash buffer sachet into one litre of distilled or deionised water and allow to dissolve fully by mixing. Wash buffer will remain stable for use for 1 month if stored at *+4 ℃.*

3. All other kit components are ready to use but allow to come to room temperature (22-27°C) before use.

Sample preparation
Dilute each test sample 1 : 500 by adding 1 ul to .5 ml of sample diluent

1. Mix well by vortexing or shaking the tube
2. A fresh pipette tip must be used for each separate sample.
3. Identify dilution tube clearly with sample number

POSITIVE AND NEGATIVE KIT CONTROLS DO NOT REQUIRE DILUTING !!

Test procedure:
1. Remove NDV coated plate from sealed bag and record location of samples on template.
2. Add 100 µl of negative control into wells A1 and B1
3. Add 100 µl of positive control into wells C1 and D1
4. Add 100 µl of diluted samples into the appropriate wells. Cover plate with lid and incubate at room temperature (22-27°C) for **30 minutes**.
5. Aspirate contents of wells and wash 4 times with wash buffer (300µl per well). Invert plate and tap firmly on absorbent paper.
6. Add 100 µl of Conjugate Reagent into the appropriate wells. Cover plate with lid and incubate at room temperature (22-27°C) for **30 minutes**.
7. Repeat wash procedure as in 5.
8. Add 100 µl of Substrate Reagent into the appropriate wells. Cover plate with lid and incubate at room temperature (22-27°C) for **15 minutes**.
9. Add 100 µl of Stop Solution to appropriate wells to stop reaction.
10. Blank the microtitre plate reader on air and record the absorbance of controls and the samples by reading at 405 nm.

Results:

For the test result to be valid the mean negative control absorbance should read below 0.3 and the difference between the mean negative control and the mean positive control should be greater than 0.2.

Variance in lab temperatures will lead to lower or higher absorbance values. Test sample values will be relative to the control values and the test will still be valid.

The NDV positive control has been carefully standardised to represent significant amounts of antibody to NDV in Chicken serum.
The relative amounts of antibodies in chicken samples can then be calculated by reference to the positive control. This relationship is expressed as S/P ratio (Sample to Positive Ratio)

Interpretation of results

Samples with an S/P of .350 or greater contain anti- NDV antibodies and are considered POSITIVE.

1. **Calculation of S/P ratio**

$$\frac{\text{Mean of Test Sample - Mean of negative control}}{\text{Mean of Positive control - Mean of negative control}} = \text{S/P}$$

2. **Calculation of Antibody Titre**

The following equation relates the S/P of a samples at a 1 : 500 dilution to an end point titre

Log10 Titre = 1.0 * Log(SP) + 3.52

Antilog = Titre

S/P value	Titre Range	Antibody status
.249 or less	827 or less	Negative
.250 - 0.349	828 - 1158	Suspect
.350 or greater	1159 or greater	Positive

Each Laboratory should establish its own criteria for non protected and protected

BioChek has available a software programme which can be used with the NDV kit to calculate S/P values, titres and provide general flock profiling.

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DATA SHEETS

SPECIFICITY

Purpose

To determine the distribution and characteristics of chicken serum originating from SPF (Specific Pathogen Free) chickens, when tested on the BioChek NDV ELISA.

Procedure

79 samples from 60 week old SPF Leghorns were obtained (Deventer, Holland) and assayed using the standard protocol for the BioChek NDV ELISA

Results / Conclusion

The results are shown in Table 1.
The results have been plotted on Graph1 showing S/P value against sample number.
The mean S/P value is

The data demonstrates that the BioChek NDV ELISA has 100% specificity on this sample panel.

Table 1 NDV ELISA Specificity, Negative Population

Interpretation

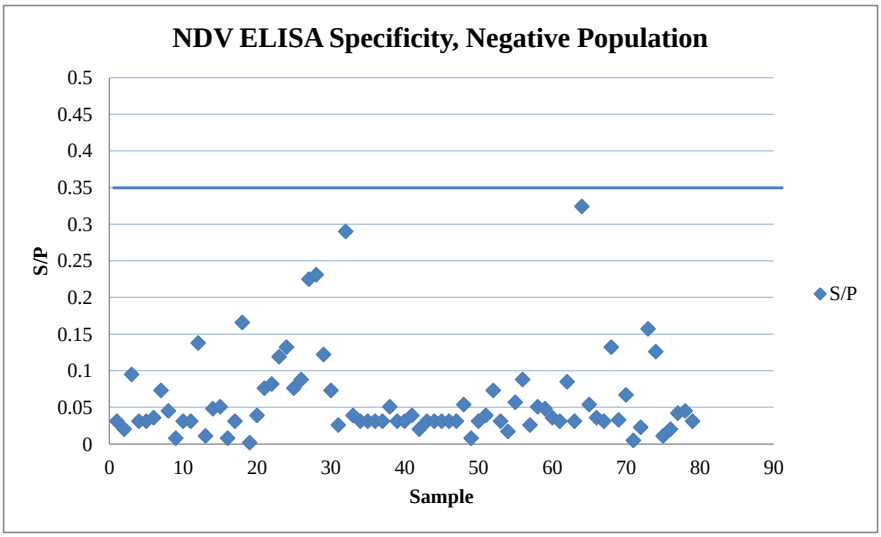
S/P value: Antibody status:

.349 or less Negative

.350 or greater Positive

Sample ID	S/P	Result	Sample ID	S/P	Result	Sample ID	S/P	Result	Sample ID	S/P	Result
01	0.031	NEG -	25	0.076	NEG -	49	0.008	NEG -	73	0.157	NEG -
02	0.02	NEG -	26	0.088	NEG -	50	0.031	NEG -	74	0.126	NEG -
03	0.095	NEG -	27	0.225	NEG -	51	0.039	NEG -	75	0.011	NEG -
04	0.031	NEG -	28	0.231	NEG -	52	0.073	NEG -	76	0.02	NEG -
05	0.031	NEG -	29	0.122	NEG -	53	0.031	NEG -	77	0.042	NEG -
06	0.036	NEG -	30	0.073	NEG -	54	0.017	NEG -	78	0.045	NEG -
07	0.073	NEG -	31	0.026	NEG -	55	0.057	NEG -	79	0.031	NEG -
08	0.045	NEG -	32	0.29	NEG -	56	0.088	NEG -			
09	0.008	NEG -	33	0.039	NEG -	57	0.026	NEG -			
10	0.031	NEG -	34	0.031	NEG -	58	0.051	NEG -			
11	0.031	NEG -	35	0.031	NEG -	59	0.048	NEG -			
12	0.138	NEG -	36	0.031	NEG -	60	0.036	NEG -			
13	0.011	NEG -	37	0.031	NEG -	61	0.031	NEG -			
14	0.048	NEG -	38	0.051	NEG -	62	0.085	NEG -			
15	0.051	NEG -	39	0.031	NEG -	63	0.031	NEG -			
16	0.008	NEG -	40	0.031	NEG -	64	0.324	NEG -			
17	0.031	NEG -	41	0.039	NEG -	65	0.054	NEG -			
18	0.166	NEG -	42	0.02	NEG -	66	0.036	NEG -			
19	0.002	NEG -	43	0.031	NEG -	67	0.031	NEG -			
20	0.039	NEG -	44	0.031	NEG -	68	0.132	NEG -			
21	0.076	NEG -	45	0.031	NEG -	69	0.033	NEG -			
22	0.082	NEG -	46	0.031	NEG -	70	0.067	NEG -			
23	0.119	NEG -	47	0.031	NEG -	71	0.005	NEG -			
24	0.132	NEG -	48	0.054	NEG -	72	0.023	NEG -			

Graph 1 NDV ELISA Specificity, negative population



>0.35 Pos
Cut Off

DATA SHEETS

MONOSPECIFIC SAMPLE PANEL
Monospecific samples containing antibodies to various viruses.

Purpose
To determine if the BioChek NDV test kit cross-reacts with antibodies generated by other pathogens common in poultry flocks.

Procedure
A sample panel monospecific for antibodies of pathogens common in poultry was tested on the BioChek NDV ELISA.

Results / Conclusion
The results are shown in Table 2

The data demonstrates that only the monospecific serum sample for NDV tested positive on the BioChek NDV ELISA. This concludes that the test kit does not cross-react with antibodies directed at other avian pathogens.

Table 2 Monospecific Sample Panel

Name : MONOSPECIFICS
Date 11/10/2001
Assay : NDV FS3616

Interpretation
S/P Ratio >=.350 positive

	S/P	result			S/P	result
TRTA	0.01	NEG -		4/91DEV	0.082	NEG -
AE	0.022	NEG -		4/91INT	0.022	NEG -
ILTAGP	0.017	NEG -		793BVLA	0.022	NEG -
PMV3	0.078	NEG -		D274	0.192	NEG -
PMV1	**2.066**	**POS +**		M41INT	0.057	NEG -
IBD	0.075	NEG -		REO1133	0.008	NEG -
Fpox	0.001	NEG -		D3128	0.089	NEG -
adeno	0.024	NEG -		D8880	0.129	NEG -
TRTC	0.006	NEG -		CR88	0.022	NEG -
Mg	0.015	NEG -		CR98	0.008	NEG -
Ms	0.069	NEG -		D1466	0.048	NEG -
ILT	0.015	NEG -		M41	0.08	NEG -
REO2534	0.022	NEG -		D274INT	0.01	NEG -
				D1466INT	0.028	NEG -

DATA SHEETS

SENSITIVITY

Purpose

To establish the time it takes for the BioChek NDV antibody detection assay to detect antibodies after active immunization.

Procedure

A sample set was obtained from AHS, Deventer, Holland. All samples except sample 7 were produced in a vaccination experiment in which SPF White Leghorn birds kept in Horsefall-Bauer isolators were individually vaccinated with a single dose of live NDV vaccine according to the manufacturer's instructions. At 7, 10 or 14 days post vaccination (d.p.v) birds were removed for bleeding. The sera were pooled before freeze drying. Sample #7 is a pooled serum from SPF birds that were inoculated (intra-tracheal) twice with a high dose of NDV LaSota. The birds were bled 4 weeks after the second inoculation.

Sample 1	Clone 30 Vaccination, Pooled sample taken 7 d.p.v
Sample 2	Clone 30 Vaccination, Pooled sample taken 14 d.p.v
Sample 3	NDW Ulster Vaccination, Pooled sample taken 10 d.p.v
Sample 4	SPF serum (1year old layers, Pooled sample)
Sample 5	Avinew VG/GA Vaccination, Pooled sample taken 7 d.p.v
Sample 6	Avinew VG/GA Vaccination, Pooled sample taken 10 d.p.v
Sample 7	Vaccinated and challenged (vvIBDV) chickens
Sample 8	AVIPRO ND HB1 Vaccination, Pooled sample taken 10 d.p.v

Results / Conclusion

The results are shown in Table 3, Table 4 and Table 5

The BioChek NDV antibody detection test has good sensitivity as it detects antibodies 7 – 14 days post vaccination.

Table 3

sample no	1	2	3	4	5	6	7	8
	Clone 30	Clone 30	NDW Ulster	SPF	Avinew VG/VA	Avinew VG/VA	LaSota Inoc	Avipro ND HB1
	7 d.p.v.	14 d.p.v.	10 d.p.v.		7 d.p.v.	10 d.p.v.	28 days p.i.	10 d.p.v.
HI	pos	pos	pos	neg	pos	pos	pos	pos
BC ELISA	pos	pos	pos	neg	**pos/neg**	pos	pos	pos
REP 1	pos	pos	pos	neg	**pos/neg**	neg	pos	pos

Table 4

BioChek

sample no: duplicate samples

	4		**7**		**8**		**5**		**6**		**3**		**1**		**2**	
mean	SPF	SPF	14.7	14.6	12.1	12.1	10.2	10.3	12.5	12.6	11.1	11	10.9	11	14	14.1
S/D			0.3	0.3	0.4	0.4	0.6	0.7	0.4	0.4	0.4	0.4	0.5	0.5	0.3	0.3
%CV			2.0	2.1	3.3	3.3	5.9	6.8	3.2	3.2	3.6	3.6	4.6	4.5	2.1	2.1

Table 5

REP 1

sample

	4		**7**		**8**		**5**		**6**		**3**		**1**		**2**	
mean	SPF	SPF	14	13.9	11.7	11.7	8.6	8.6	12.1	12	10.4	10.3	10.1	10	13.6	13.6
S/D			0.8	0.9	0.6	0.7	1.4	1.3	1	1	0.6	0.5	0.7	0.6	1	0.9
%CV			5.7	6.5	5.1	6.0	16.3	15.1	8.3	8.3	5.8	4.9	6.9	6.0	7.4	6.6

DATA SHEETS

REPRODUCIBILITY

Purpose

The final kit assembly is tested by assaying a series of chicken serum samples with varying degrees (standard 1,2,3) of antibody to NDV. These samples must meet set specifications in order to pass and be released for market. All batches of product produced are tested on the same set of samples to help assure lot to lot reproducibility.

Procedure

A standard panel of known NDV samples is assayed on each new kit. Results on new kits should match those of previous kits. Following standard assay protocol, negative control, positive control and the NDV sensitivity panel are tested which results must be within specification.

Results/Conclusion

As can be seen in the corresponding table (table IV stability) the batch variability on the 9 batches is as following:

For the HIGH control sample is:

Mean S/P	4.79
SD	0.59
%CV	12.35

For the MEDIUM control sample the values are:

Mean S/P	1.11
SD	0.10
%CV	8.60

For the LOW control sample the values are:

Mean S/P	0.5
SD	0.04
%CV	8.67

These data demonstrate that there is limited variation when comparing results from various production batches of the BioChek NDV ELISA.

IV Reproducibility of the BioChek NDV ELISA

Date **23-03-2005**

Sample High: prediluted serum containing a high titer of antibodies against NDV.
Sample Medium: prediluted serum containing a intermediate titer of antibodies against NDV.
Sample Low: prediluted serum containing a borderline titer of antibodies against NDV.

				Raw OD values controls + samples								
Name	batch no	manufacturing date	Assay	-	-	+	+	OD	OD	Mean S/P	No. Samples	**Result HIGH**
HIGH	FS4032	March-04	NDV	0.159	0.153	0.625	0.663	2.653	2.639	5.1000	2	**Mean S/P** **4.79**
HIGH	FS4050	April-04	NDV	0.125	0.13	0.463	0.469	2.107	2.176	5.9500	2	**SD** **0.59**
HIGH	FS4067	June-04	NDV	0.119	0.128	0.562	0.556	2.164	2.202	4.7300	2	**%CV** **12.35**
HIGH	FS4082	July-04	NDV	0.121	0.123	0.644	0.628	2.905	2.938	5.4500	2	
HIGH	FS4099	August-04	NDV	0.119	0.115	0.533	0.521	1.865	2.014	4.4500	2	
HIGH	FS4114	September-04	NDV	0.132	0.132	0.574	0.56	1.976	1.952	4.2100	2	
HIGH	FS4132	November-04	NDV	0.16	0.171	0.607	0.624	2.182	2.114	4.4100	2	
HIGH	FS4172	January-05	NDV	0.157	0.153	0.657	0.627	2.321	2.387	4.5200	2	
HIGH	FS4190	February-05	NDV	0.126	0.125	0.652	0.619	2.322	2.309	4.2900	2	

				Raw OD values controls + samples								
Name	batch no	manufacturing date	Assay	-	-	+	+	OD	OD	Mean S/P	No. Samples	**Result Medium**
MEDIUM	FS4032	March-04	NDV	0.159	0.153	0.625	0.663	0.726	0.746	1.1900	2	**Mean S/P** **1.11**
MEDIUM	FS4050	April-04	NDV	0.125	0.13	0.463	0.469	0.551	0.478	1.1400	2	**SD** **0.10**
MEDIUM	FS4067	June-04	NDV	0.119	0.128	0.562	0.556	0.686	0.659	1.2600	2	**%CV** **8.60**
MEDIUM	FS4082	July-04	NDV	0.121	0.123	0.644	0.628	0.694	0.684	1.1000	2	
MEDIUM	FS4099	August-04	NDV	0.119	0.115	0.533	0.521	0.592	0.569	1.1300	2	
MEDIUM	FS4114	September-04	NDV	0.132	0.132	0.574	0.56	0.586	0.535	0.9900	2	
MEDIUM	FS4132	November-04	NDV	0.16	0.171	0.607	0.624	0.703	0.672	1.1600	2	
MEDIUM	FS4172	January-05	NDV	0.157	0.153	0.657	0.607	0.629	0.986	0.9500	2	
MEDIUM	FS4190	February-05	NDV	0.126	0.125	0.652	0.619	0.692	0.669	1.0900	2	

				Raw OD values controls + samples								
Name	batch no	manufacturing date	Assay	-	-	+	+	OD	OD	Mean S/P	No. Samples	**Result Low**
LOW	FS4032	March-04	NDV	0.159	0.153	0.625	0.663	0.414	0.397	0.5100	2	**Mean S/P** **0.50**
LOW	FS4050	April-04	NDV	0.125	0.13	0.463	0.469	0.29	0.297	0.4900	2	**SD** **0.04**
LOW	FS4067	June-04	NDV	0.119	0.128	0.562	0.556	0.356	0.34	0.5200	2	**%CV** **8.67**
LOW	FS4082	July-04	NDV	0.121	0.123	0.644	0.628	0.369	0.35	0.4600	2	
LOW	FS4099	August-04	NDV	0.119	0.115	0.533	0.521	0.334	0.308	0.5000	2	
LOW	FS4114	September-04	NDV	0.132	0.132	0.574	0.56	0.321	0.305	0.4200	2	
LOW	FS4132	November-04	NDV	0.16	0.171	0.607	0.624	0.416	..374	0.5100	2	
LOW	FS4172	January-05	NDV	0.157	0.153	0.657	0.627	0.41	0.423	0.5400	2	
LOW	FS4190	February-05	NDV	0.126	0.125	0.652	0.619	0.416	0.415	0.5700	2	

BioChek R&D
Hounslow UK
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Date : 08-04-2005

Report: Coded Samples - Platelayout

Assay :	NDV	Lot No:	FS4190
Positive Cutoff S/P >=	0.35	Dilution :	500
		Total No. Samples:	92

A prediluted positive sample RF06 was dispensed over 92 wells of the BioChek NDV ELISA test.
Test was performed according to package insert.
The result on the 92 samples is as following:

Mean OD	0.847
st dev	0.05
%CV	5.89

DATA SHEETS

COMPARISON WITH HI

Trial 1

Sensitivity BC-NDV ELISA vs HI AHS Deventer Holland
30 samples were tested BC NDV ELISA and then shipped to AHS in Deventer to be tested on HI. The samples are from Broiler Breeders 14 Weeks of age vaccinated at 3 weeks of age with Hitchner B1 (drinking water) and at 9 weeks of age with Clone 30 (spray).

Result:

100% of the samples tested positive on the BC NDV ELISA

Only 50% of the samples tested positive on HI

		HI
sample #	**BC NDV ELISA**	**IP2001-17847**
	7-Sep	**7-Sep**
1	6950	3
2	13040	4
3	2354	**1**
4	4666	**1**
5	3944	**1**
6	1225	**1**
7	4368	**1**
8	2553	**1**
9	3235	**1**
10	2132	**1**
11	2020	2
12	6692	3
13	8565	3
14	7523	4
15	2156	**1**
16	4381	**1**
17	3076	**1**
18	3113	3
19	3374	2
20	2007	**1**
21	5248	3
22	2219	**1**
23	5523	2
24	4219	2
25	5894	4
26	2887	**1**
27	4791	2
28	3536	2
29	2183	2
30	6590	**1**
Total	**30 x Pos**	**15 x Pos**

Interpretation:	BC NDV ELISA	
S/P value	**Titre Range**	**Antibody status**
.349 or less	1158 or less	Negative
.350 or greater	1159 or greater	Positive

Interpretation:	HI Deventer
HI = 1	**negative**
HI = 2	**suspect**
HI = 3	**positive**

Sensitivity ELISA: 100% positive HI 50% positive

Trial 2

Reproducibility NDV HI as done by the AHS in Deventer Holland
30 samples numbered 1 - 30 were each divided in 4 sealable tubes and stored frozen.
On September 7, 21, 28 and October 4 1 set of 30 samples was taken out of the freezer, thawed, thoroughly mixed and tested first on the BC NDV ELISA and then shipped to Deventer to be tested on HI.

	HI	**HI**	**HI**	
sample ID	3-May	29-Apr	13-May	
	IP2002-12369	**IP2002-11931**	**IP2002-12646**	
1	1	1	2	same
2	ns	1	1	same
3	1	1	2	same
4	1	1	1	same
5	**1**	**1**	**3**	**different**
6	1	1	1	same
7	ns	1	2	same
8	1	1	1	same
9	1	1	1	same
10	5	4	3	same
11	**4**	**2**	**3**	**different**
12	**1**	**1**	**3**	**different**
13	1	1	1	same
14	5	4	4	same
15	4	3	3	same
16	4	3	4	same
17	4	4	4	same
18	1	1	1	same
19	4	3	3	same
20	4	4	4	same
21	1	1	1	same
22	**1**	**4**	**3**	**different**
23	1	1	1	same
24	5	5	3	same
25	1	1	1	same
26	1	1	1	same
27	4	3	3	same
28	1	1	2	same
29	**1**	**1**	**3**	**different**
30	1	1	1	same
				5 x different (17%)

Summary of results:

70% of the samples gave gave the same result at each retest. Definition of same result: Equal interpretation on the bases of negative, suspect and positive status of the sample.

Trial 2

Reproducibility BC-NDV ELISA

30 samples were each divided in 4 sealable tubes and stored frozen. On September 7, 21, 28 and October 4

1 set of 30 samples was taken out of the freezer, thawed, thoroughly mixed and tested first on the

BC NDV ELISA and then shipped to Deventer to be tested on HI.

The samples are from Broiler Breeders 14 Weeks of age vaccinated at 3 weeks of age with Hitchner B1 (drinking water) and at 9 weeks of age with Clone 30 (spray).

sample #	**ELISA**	**ELISA**	**ELISA**	**ELISA**	
	7-Sep	21-Sep	28-Sep	4-Oct	**No of different results on retest**
1	6950	5831	7394	7335	same
2	13040	13199	13689	15815	same
3	2354	1871	2139	2503	same
4	4666	3550	4166	4798	same
5	3944	3169	4179	4831	same
*6**	*1225*	*1189*	***1089***	*1212*	**different**
7	4368	3944	4076	4282	same
8	2553	3027	2785	2616	same
9	3235	4467	3924	3586	same
10	2132	1884	2318	1649	same
11	2020	2487	2139	2745	same
12	6692	7146	6318	7060	same
13	8565	8795	8358	2358	same
14	7523	7762	7003	7593	same
15*	2156	1632	2116	2245	same
16*	4381	3265	4076	4328	same
17*	3076	3629	3089	3490	same
18	3113	3358	2685	3086	same
19	3374	3993	3381	3278	same
20*	2007	1884	1695	1275	same
21	5248	4626	5864	5540	same
22*	2219	1729	2444	1987	same
23	5523	6020	6053	8109	same
24	4219	4404	4282	**162**	**different**
25	5894	6878	6649	8013	same
26*	2887	2788	2583	3070	same
27	4791	3027	4444	5073	same
28	3536	2646	3798	4974	same
29	2183	2407	2089	2424	same
30	6590	7192	6090	7785	same

Sample numbers marked with * are negative on HI

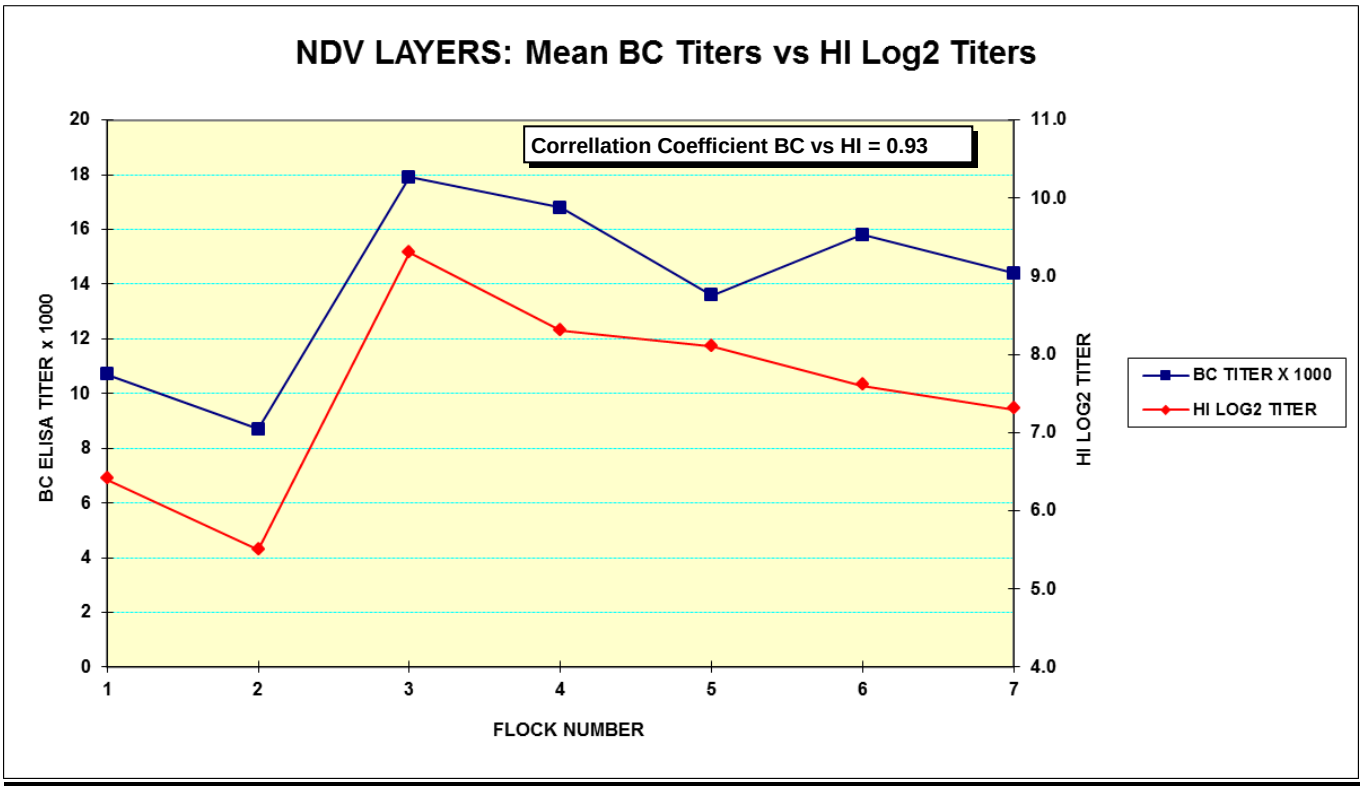
results in **bold** conflict with other results on the same samples

Summary of results:

93% of the samples gave gave the same result at each retest. Definition of same result: Equal interpretation on the bases of negative, suspect and positive status of the sample.

Graph 2 NDV Correlation of BioChek Titers With HI NCD

Mean titers of 7 Layer Flocks, Age 18 – 43W (Total no. of Samples = 89)
Interpretation HI: =>3 positive
BC Titer = >1159 positive (log2 Titer=>10.2)



Graph 3 NDV Correlation of BioChek Titers With HI NCD Deventer

Mean Broiler Flock titers (total no. of samples = 480)
Broilers vaccinated at 01D and 21D with Ulster strain
Interpretation HI: =>3 positive
BC Titer = >1159 positive (log2 Titer = >10.2)