INSTRUCTIONS FOR USE

EN

ANA Screen

REF 2Z29001G
SM2Z29001G

IVD

CE

Rx Only

Σ 96

INTENDED USE

The ANA Screen is a qualitative screening assay designed to detect anti-nuclear antibodies (ANA) in human sera. When performed according to these instructions, this test system is capable of detecting all ANAs commonly tested for, such as those against double stranded DNA (dsDNA), Jo-1, Sm, Sm/RNP, SSA, SSB, and Scl-70. The test is also capable of detecting ANA demonstrating centromere, nucleolar, peripheral, and spindle indirect immunofluorescence antibody (IFA) patterns. This test is for *In Vitro* diagnostic use.

SIGNIFICANCE AND BACKGROUND

In recent years, it has become clear that autoantibodies to a number of nuclear constituents have proven to be useful in the diagnosis of various connective tissue diseases. Antibodies to dsDNA are highly specific for active systemic lupus erythematosus (SLE), and correlate closely with the onset of lupus nephritis. The Jo-1 autoantibody is one of a family of characteristic autoantibodies seen in myositis patients (19). Scientists also find them specifically in patients with myositis, and associate them with a high incidence of accompanying interstitial lung disease (10). Doctors consider antibodies directed against the Sm marker a diagnostic criterion for SLE due to high specificity for patients with SLE (1, 2). The presence of high level RNP antibodies alone are considered diagnostic of mixed connective tissue disease (MCTD) and are usually associated with a more benign disease course (3), while patients with low levels of RNP antibodies, together with other autoantibodies, may be observed in the serum of patients with progressive systemic sclerosis, Sjögren's Syndrome, and rheumatoid arthritis. The presence of RNP antibodies in the serum of SLE patients is usually associated with a lower incidence of renal involvement and a more benign disease course. To the contrary, patients with Sm antibodies experience a higher frequency of renal and central nervous system complications (4). Studies have observed autoantibodies directed against SSA and SSB in patients with SLE (5, 6), and Sjögren's disease (7 - 9). SSA antibodies are frequently present in the serum of ANA negative SLE patients, such as subacute cutaneous lupus erythematosus (12), a lupus-like syndrome associated with a homozygous C2 deficiency (13), and in a subset of patients who lack anti-dsDNA antibodies (11). Scl-70 antibodies are highly specific for scleroderma (11). Research also observes these antibodies in a minority of SLE patients. Scl-70 positive scleroderma patients tend to have a more severe disease course, more internal organ involvement and diffuse rather than limited skin involvement (14). Scientists rarely find Scl-70 antibodies in other autoimmune diseases, and thus, their detection in a patient with the recent onset of Raynaud's phenomenon is highly significant (15). The following table summarizes the various autoantibodies noted above with respect to disease association (16):

Antibody	Disease State	Relative Frequency of Antibody Detection %
Anti-Jo-1	Myositis	25 - 44 (19)
Anti-Sm	SLE	30*
Anti-RNP	MCTD, SLE	100** and >40, respectively
Anti-SSA (Ro)	SLE, Sjögren's	15 and 30-40, respectively
Anti-SSB (La)	SLE, Sjögren's	15 and 60 - 70, respectively
Anti-Scl-70	Systemic sclerosis	20 - 28*
Anti-dsDNA	SLE	40 - 60*
*Highly specific		
**Highly specific when present alone at high titer.		

Until recently, testing of autoantibodies occurred using indirect immunofluorescence, ouchterlony gel diffusion, hemagglutination, radioimmunoassay, or enzyme-linked immunosorbent assay (ELISA). Unlike several other systems, the ELISA methodology offers sensitive, objective, and rapid evaluation of specimens, and therefore is suitable for screening a large number of samples for total ANA.

The exact etiology of autoimmune diseases is unknown, and the specific role played by autoantibodies in the onset of various autoimmune connective tissue diseases is obscure. The ANA Screen offers an efficient test procedure for the laboratory workup of patients with suspected various connective tissue diseases using the association and frequency of detection of these antibodies.

PRINCIPLE OF THE ASSAY

The ANA Screen is designed to detect IgG class antibodies to a variety of common nuclear antigens in human sera. Creation of the sensitized wells of the plastic microwell strips occurred using passive adsorption with antigen. The test procedure involves three incubation steps:

1. Test sera (properly diluted) are incubated in antigen coated microwells. Any antigen specific antibody in the sample will bind to the immobilized antigen. The plate is washed to remove unbound antibody and other serum components.
2. Peroxidase Conjugated goat anti-human IgG is added to the wells and the plate is incubated. The Conjugate will react with the antibody immobilized on the solid phase in step 1. The wells are washed to remove unreacted Conjugate.
3. The microwells containing immobilized peroxidase Conjugate are incubated with peroxidase Substrate Solution. Hydrolysis of the Substrate by peroxidase produces a color change. After a period of time the reaction is stopped and the color intensity of the solution is measured photometrically. The color intensity of the solution depends upon the antibody concentration in the original test sample.

TEST SYSTEM COMPONENTS

Materials Provided:

Each Test System contains the following components in sufficient quantities to perform the number of tests indicated on the packaging label.

Kit Component	Quantity (Σ 96)	Description
PLATE	1	Plate: 96 wells configured in twelve, 1x8-well, strips coated with inactivated antigen. The strips are packaged in a strip holder and sealed in an envelope with desiccant.
CONJ	1	Conjugate: Conjugated (horseradish peroxidase) goat anti-human IgG (Fc chain specific). 15mL, white-capped bottle. Ready to use.
CTRL +	1	Positive Control (Human Serum): 0.35mL, red-capped vial. 21X concentrate.
CAL	1	Calibrator (Human Serum): 0.5mL, blue-capped vial. 21X concentrate.
CTRL -	1	Negative Control (Human Serum): 0.35mL, green-capped vial. 21X concentrate.
DIL SPE	1	Sample Diluent: 30mL, green-capped bottle containing Tween-20, bovine serum albumin and phosphate-buffered-saline. Green solution. Ready to use.
SOLN TMB	1	TMB: 15mL, amber-capped, amber bottle containing 3, 3', 5, 5' - tetramethylbenzidine (TMB). Ready to use.
SOLN STOP	1	Stop Solution: 15mL, red-capped bottle containing 1M H_2SO_4, 0.7M HCl. Ready to use.
WASH 10X	1	Wash Buffer Concentrate (10X): Dilute 1 part concentrate + 9 parts deionized or distilled water. 100mL, clear-capped bottle containing a 10X concentrated phosphate-buffered-saline and Tween-20 solution (blue solution). **NOTE: 1X solution will have a pH of 7.2 ± 0.2.**

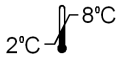
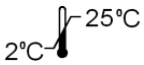
NOTE: The following components are not Test System Lot Number dependent and may be used interchangeably with the ZEUS ELISA Test Systems: TMB, Stop Solution, and Wash Buffer.

MATERIALS REQUIRED BUT NOT PROVIDED

1. ELISA microwell reader capable of reading at a wavelength of 450nm. **NOTE: Use of a single (450nm), or dual (450/620 - 650nm), wavelength reader is acceptable. Dual wavelength is preferred, as the additional reference filter has been determined to reduce potential interference from anomalies that may absorb light.**
2. Pipettes capable of accurately delivering 10 - 200µL.
3. Multichannel pipette capable of accurately delivering 50 - 200µL.

4. Reagent reservoirs for multichannel pipettes.
5. Wash bottle or microwell washing system.
6. Distilled or deionized water.
7. One-liter graduated cylinder.
8. Serological pipettes.
9. Disposable pipette tips.
10. Paper towels.
11. Laboratory timer to monitor incubation steps.
12. Disposal basin and disinfectant (i.e., 10% household bleach - 0.5% sodium hypochlorite).

STORAGE CONDITIONS

	Coated Microwell Strips: Immediately reseal extra strips with desiccant and return to proper storage. After opening, strips are stable for 60 days, as long as the indicator strips on the desiccant pouch remain blue.
	Conjugate – DO NOT FREEZE.
	Unopened Kit, Calibrator, Positive Control, Negative Control, TMB, Sample Diluent.
	Stop Solution: 2 – 25 °C Wash Buffer (1X) : 20 - 25°C for up to 7 days, 2 - 8°C for 30 days Wash Buffer (10X): 2 - 25°C

PRECAUTIONS

1. For *In Vitro* diagnostic use.
2. Follow normal precautions exercised in handling laboratory reagents. In case of contact with eyes, rinse immediately with plenty of water and seek medical advice. Wear suitable protective clothing, gloves, and eye/face protection. Do not breathe vapor. Dispose of waste observing all local, state, and federal laws.
3. The wells of the ELISA plate do not contain viable organisms. However, consider the strips **potentially biohazardous materials** and handle accordingly.
4. The Controls are **potentially biohazardous materials**. Source materials from which these products were derived were found negative for HIV-1 antigen, HBsAg and for antibodies against HCV and HIV by approved test methods. However, since no test method can offer complete assurance that infectious agents are absent, handle these products at the Biosafety Level 2 as recommended for any potentially infectious human serum or blood specimen in the Centers for Disease Control/National Institutes of Health manual "Biosafety in Microbiological and Biomedical Laboratories": Current Edition; and OSHA's Standard for Bloodborne Pathogens (20).
5. Adherence to the specified time and temperature of incubations is essential for accurate results. **All reagents must be allowed to reach room temperature (20 - 25°C) before starting the assay**. Return unused reagents to refrigerated temperature immediately after use.
6. Improper washing could cause false positive or false negative results. Be sure to minimize the amount of any residual wash solution; (e.g., by blotting or aspiration) before adding Conjugate or Substrate. Do not allow the wells to dry out between incubations.
7. The Sample Diluent, Controls and Calibrator contain Sodium Azide at a concentration of <0.1% (w/v). Sodium Azide has been reported to form lead or copper azides in laboratory plumbing which may cause explosions upon hammering. To prevent, rinse sink thoroughly with water after disposing of solution containing Sodium Azide.
8. The Stop Solution is TOXIC if inhaled, has contact with skin or if swallowed. It can cause burns. In case of accident or ill feelings, seek medical advice immediately.
9. The TMB Solution is HARMFUL. It is irritating to eyes, respiratory system and skin.
10. The Wash Buffer concentrate is an IRRITANT. It is irritating to eyes, respiratory system and skin.
11. Wipe the bottom of the plate free of residual liquid and/or fingerprints that can alter optical density (OD) readings.
12. Dilution or adulteration of these reagents may generate erroneous results.
13. Do not use reagents from other sources or manufacturers.
14. TMB Solution should be colorless, very pale yellow, very pale green, or very pale blue when used. Contamination of the TMB with Conjugate or other oxidants will cause the solution to change color prematurely. Do not use the TMB if it is noticeably blue in color.
15. Never pipette by mouth. Avoid contact of reagents and patient specimens with skin and mucous membranes.
16. Avoid microbial contamination of reagents. Incorrect results may occur.
17. Cross contamination of reagents and/or samples could cause erroneous results.

18. Reusable glassware must be washed and thoroughly rinsed free of all detergents.
19. Avoid splashing or generation of aerosols.
20. Do not expose reagents to strong light during storage or incubation.
21. Allowing the microwell strips and holder to equilibrate to room temperature prior to opening the protective envelope will protect the wells from condensation.
22. Collect the wash solution in a disposal basin. Treat the waste solution with disinfectant (i.e.: 10% household bleach - 0.5% Sodium Hypochlorite). Avoid exposure of reagents to bleach fumes.
23. Caution: Neutralize any liquid waste at an acidic pH before adding to a bleach solution.
24. Do not use ELISA plate if the indicator strip on the desiccant pouch has turned from blue to pink.
25. Do not allow the Conjugate to come in contact with containers or instruments that may have previously contained a solution utilizing Sodium Azide as a preservative. Residual amounts of Sodium Azide may destroy the Conjugate's enzymatic activity.
26. Do not expose any of the reactive reagents to bleach-containing solutions or to any strong odors from bleach-containing solutions. Trace amounts of bleach (sodium hypochlorite) may destroy the biological activity of many of the reactive reagents within this Test System.

SPECIMEN COLLECTION

1. ZEUS Scientific recommends that the user carry out specimen collection in accordance with CLSI document M29: Protection of Laboratory Workers from Infectious Disease (Current Edition).
2. No known test method can offer complete assurance that human blood samples will not transmit infection. Therefore, consider all blood derivatives potentially infectious.
3. Use only freshly drawn and properly refrigerated sera obtained by approved aseptic venipuncture procedures in this assay (17, 18). Do not use if there are any added anticoagulants or preservatives. Avoid using hemolyzed, lipemic, or bacterially contaminated sera.
4. Store sample at room temperature for no longer than 8 hours. If testing is not performed within 8 hours, sera may be stored between 2 - 8°C, for no longer than 48 hours. If delay in testing is anticipated, store test sera at –20°C or lower. Avoid multiple freeze/thaw cycles which may cause loss of antibody activity and give erroneous results. It is the responsibility of the individual laboratory to use all available references and/or its own studies to determine stability criteria for its laboratory (21).

ASSAY PROCEDURE

1. Remove the individual components from storage and allow them to warm to room temperature (20 - 25°C).
2. Determine the number of microwells needed. Allow for six Control/Calibrator determinations (one Reagent Blank, one Negative Control, three Calibrators and one Positive Control) per run. Run a Reagent Blank on each assay. Check software and reader requirements for the correct Controls/Calibrator configurations. Return unused strips to the resealable pouch with desiccant, seal, and return to storage between 2 - 8°C.

EXAMPLE PLATE SET-UP		
	1	2
A	Blank	Patient 3
B	Negative Control	Patient 4
C	Calibrator	Etc.
D	Calibrator	
E	Calibrator	
F	Positive Control	
G	Patient 1	
H	Patient 2	

3. Prepare a 1:21 dilution (e.g.: 10µL of serum + 200µL of Sample Diluent) of the Negative Control, Calibrator, Positive Control, and each patient serum.
4. To individual wells, add 100µL of each diluted Control, Calibrator and patient specimen. Ensure that the samples are properly mixed. Use a different pipette tip for each sample.
5. Add 100µL of Sample Diluent to well A1 as a Reagent Blank. Check software and reader requirements for the correct Reagent Blank well configuration.
6. Incubate the plate at room temperature (20 - 25°C) for 60 - 65 minutes.
7. Wash the microwell strips 5 times.
    a. **Manual Wash Procedure**:
        1. Vigorously shake out the liquid from the wells.

2. Fill each microwell with Wash Buffer. Make sure no air bubbles are trapped in the wells.
3. Repeat steps 1. and 2. for a total of 5 washes.
4. Shake out the wash solution from all the wells. Invert the plate over a paper towel and tap firmly to remove any residual wash solution from the wells. Visually inspect the plate to ensure that no residual wash solution remains. Collect wash solution in a disposable basin and treat with disinfectant at the end of the day's run.

b. **Automated Wash Procedure**:
If using an automated microwell wash system, set the dispensing volume to 300 - 350µL/well. Set the wash cycle for 5 washes with no delay between washes. If necessary, the microwell plate may be removed from the washer, inverted over a paper towel and tapped firmly to remove any residual wash solution from the microwells.

8. Add 100µL of the Conjugate to each well, including the Reagent Blank well, at the same rate and in the same order as the specimens.
9. Incubate the plate at room temperature (20 - 25°C) for 30 - 35 minutes.
10. Wash the microwells by following the procedure as described in step 7.
11. Add 100µL of TMB to each well, including the Reagent Blank well, at the same rate and in the same order as the specimens.
12. Incubate the plate at room temperature (20 - 25°C) for 30 - 35 minutes.
13. Stop the reaction by adding 50µL of Stop Solution to each well, including the Reagent Blank well, at the same rate and in the same order as the TMB. Positive samples will turn from blue to yellow. After adding the Stop Solution, tap the plate several times to ensure that the samples are thoroughly mixed.
14. Set the microwell reader to read at a wavelength of 450nm and measure the optical density (OD) of each well against the Reagent Blank. Read the plate within 30 minutes of the addition of the Stop Solution.

ABBREVIATED TEST PROCEDURE

1. Dilute Serum 1:21.
2. Add diluted sample to microwell - 100µL/well.
3. ⟶ *Incubate 60 - 65 minutes.*
4. Wash.
5. Add Conjugate - 100µL/well.
6. ⟶ *Incubate 30 - 35 minutes.*
7. Wash.
8. Add TMB - 100µL/well.
9. ⟶ *Incubate 30 - 35 minutes.*
10. Add Stop Solution - 50µL/well - Mix.
11. READ within 30 minutes.

QUALITY CONTROL

1. Each time the assay is performed, the Calibrator must be run in triplicate. A Reagent Blank, Negative Control, and Positive Control must also be included.
2. Calculate the mean of the three Calibrator wells. If any of the three values differ by more than 15% from the mean, discard that value and calculate the mean using the remaining two wells.
3. The mean OD value for the Calibrator, Positive Control, and Negative Control should fall within the following ranges:

	OD Range
Negative Control	≤0.250
Calibrator	≥0.300
Positive Control	≥0.500

   a. The OD of the Negative Control divided by the mean OD of the Calibrator should be ≤0.9.
   b. The OD of the Positive Control divided by the mean OD of the Calibrator should be ≥1.25.
   c. If the above conditions are not met the test should be considered invalid and should be repeated.
4. The Positive Control and Negative Control are intended to monitor for substantial reagent failure, but will not ensure precision at the assay Cutoff.
5. Additional Controls may be tested according to guidelines or requirements of local, state, and/or federal regulations or accrediting organizations.
6. Refer to CLSI document C24: Statistical Quality Control for Quantitative Measurement Procedures for guidance on appropriate QC practices.

INTERPRETATION OF RESULTS

1. **Calculations:**
   a. *Correction Factor*: The manufacturer determined a Cutoff OD Value for positive samples and correlated it to the Calibrator. The Correction Factor (CF) allows for the determination of the Cutoff Value for positive samples. It will also correct for slight day-to-day variations in test results. The Correction Factor is determined for each lot of components and is printed on the Component Label located in the Test System box.
   b. *Cutoff OD Value*: To obtain the Cutoff OD Value, multiply the CF by the mean OD of the Calibrator determined above. *(CF x Mean OD of Calibrator = Cutoff OD Value)*
   c. *Index Values/OD Ratios*: Calculate the Index Value/OD Ratio for each specimen by dividing its OD Value by the Cutoff OD from step 2.

Example:	Mean OD of Calibrator	=	0.793
	Correction Factor (CF)	=	0.25
	Cutoff OD	=	0.793 x 0.25 = 0.198
	Unknown Specimen OD	=	0.432
	Specimen Index Value/OD Ratio	=	0.432/0.198 = 2.18

2. **Interpretations:** Index Values/OD Ratios are interpreted as follows.

	Index Value/OD Ratio
Negative Specimens	≤0.90
Equivocal Specimens	0.91 - 1.09
Positive Specimens	≥1.10

   a. An OD ratio ≤0.90 indicates no significant amount of IgG antibodies to ANA detected.
   b. An OD ratio ≥1.10 indicates that IgG antibodies specific to ANA were detected.
   c. Specimens with OD ratio values in the equivocal range (0.91 - 1.09) should be retested in duplicate. Report any two of the three results which agree. Evaluate repeatedly equivocal specimens using an alternate serological method and/or re-evaluate by drawing another sample one to three weeks later.

LIMITATIONS OF THE ASSAY

1. The ANA Screen is a diagnostic aid. Interpret test results in conjunction with the clinical evaluation and the results of other diagnostic procedures.
2. Positive ANA may be found in apparently healthy people. It is therefore imperative that the results be interpreted in light of the patient's clinical picture by a medical authority.
3. SLE patients undergoing steroid therapy may have negative test results.
4. Many commonly prescribed drugs may induce ANA.
5. The ANA Screen will not identify the specific type of ANA present in a positive specimen. Test positive specimens for individual autoantibodies using more specific reflex tests in combination with the Anti-dsDNA. Alternatively, specific autoantibodies may be detected using a variety of methods including immunodiffusion, western blot or multiplexed fluorescent bead-based assays such as the ZEUS AtheNA Multi-Lyte® ANA II Plus Test System.

EXPECTED RESULTS

The expected value for a normal patient is a negative result. The number of reactives, and the degree of reactivity is dependent upon parameters such as population type being tested, treatment, etc. Each laboratory should establish their own expected values based upon the specimens typically being tested. With respect to disease-state and percent reactivity, the Table in the Significance and Background section of this Package Insert shows the relative frequency of autoantibody activity for various rheumatic disorders.

PERFORMANCE CHARACTERISTICS

1. **Comparative Study**
   A clinical investigation of 270 serum specimens was conducted using the ANA Screen, and a commercial ELISA test system. Specificity was evaluated using 72 asymptomatic normal specimens from southeastern United States, and sensitivity was evaluated using 198 disease-state sera from northeastern United States. Tables 1 through 4 summarize the results of the study.

Table 1: Evaluation of Specificity Performance

		ANA Screen			
		Positive	Negative	Equivocal	Total
Commercial ELISA Kit	Positive	0	1	1	2
	Negative	0	59	8	67
	Equivocal	0	1	2	3
	Total	0	61	11	72

Table 2: Evaluation of Sensitivity Performance

		ANA Screen			
		Positive	Negative	Equivocal	Total
Commercial ELISA Kit	Positive	141	7*	8	156
	Negative	16*	2	2	20
	Equivocal	18	2	2	22
	Total	175	11	12	198
* Represents Discrepant Specimens. See Table 4 for Calculations of Relative Sensitivity.					

Table 3: Summary of Discrepant Specimens

Sample Number	ELISA Results		IFA Hep-2 Results*
	ZEUS ELISA	Other ELISA	
62	0.902/Equivocal	0.87/Negative	Negative
64	0.926/Equivocal	0.65/Negative	Negative
65	0.940/Equivocal	0.74/Negative	Negative
66	0.950/Equivocal	0.53/Negative	Negative
68	1.022/Equivocal	0.92/Negative	Negative
69	1.026/Equivocal	0.74/Negative	Negative
0	1.045/Equivocal	0.43/Negative	Negative
71	1.089/Equivocal	0.46/Negative	Negative
73	0.472/Negative	4.88/Positive	≥ 1:40, Speckled
74	0.482/Negative	4.98/Positive	≥ 1:40, Speckled
76	0.585/Negative	5.47/Positive	≥ 1:40, Speckled
77	0.634/Negative	6.64/Positive	≥ 1:40, Speckled
79	0.714/Negative	3.14/Positive	Negative
81	0.798/Negative	2.67/Positive	≥ 1:40, Centromere
83	0.876/Negative	2.68/Positive	≥ 1:40, Centromere
84	0.979/Equivocal	4.15/Positive	≤ 1:40, Speckled
85	0.984/Equivocal	5.65/Positive	≥ 1:40, Speckled
87	0.992/Equivocal	5.35/Positive	≥ 1:40, Centromere
91	1.023/Equivocal	3.39/Positive	≥ 1:320, Centromere
92	1.053/Equivocal	2.30/Positive	≤ 1:40, Speckled
93	1.065/Equivocal	3.76/Positive	≥ 1:320, Centromere
94	1.073/Equivocal	5.08/Positive	≥ 1:40, Speckled
95	1.091/Equivocal	3.23/Positive	≥ 1:40, Speckled
* ZEUS IFA HEp-2 Test System			

Table 4: Calculations of Relative Specificity and Relative Sensitivity

Relative Specificity:	
1. Calculation including equivocal specimens: 59/67 = 88%	2. Calculation excluding equivocal specimens: 59/59 = 100%
Relative Sensitivity:	
1. Calculation including equivocal specimens; without resolution of discrepant specimens: 141/156 = 90.4%	2. Calculation excluding equivocal specimens; after resolution of discrepant specimens: 141/147 = 95.9%
Percent Agreement: 200/207 = 96.6%	

2. **Reproducibilty**

Reproducibility was performed as outlined in document number EP5-T2: Evaluation of Precision Performance of Clinical Chemistry Devices - Second Edition. as published by National Committee for Clinical Chemistry Laboratory Standards (NCCLS). Briefly, eight specimens were tested; two strong positive samples, two moderately positive specimens, two specimens near the cutoff, and two negative specimens. Each sample was tested in duplicate, two times per day (AM and PM), on each day. Table 5 summarizes the results.

Table 5: Summary of Reproducibility Testing

ID	Mean Ratio	Swr [a]	St [b]	Days Tested	% CV	Total Observations
1	9.86	0.81	1.28	19	12.95	76
2	11.22	1.25	1.63	20	14.60	80
3	4.20	0.43	0.53	18	12.92	72
4	3.77	0.49	0.56	19	14.96	76
B_1	1.24	0.07	0.14	20	11.29	80
B_2	0.94	0.07	0.13	20	14.16	80
5	0.40	0.09	0.14	19	N/A	76
6	0.20	0.05	0.07	18	N/A	72
[a] Point estimate of within run precision standard deviation.						
[b] Point estimate of total precision standard deviation.						

3. **Cross Reactivity**

Specimens negative for ANA by HEp-2 IFA and positive for IgG antibody to various antigens such as EBV-VCA, EBNA, HSV-1, HSV-2, CMV, Rubella, and/or Toxo, were tested for potential cross reactivity using the ANA Screen. All samples were negative on the ELISA, indicating that the potential for cross reactivity with such antibodies is minimal.

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GLOSSARY OF SYMBOLS

The following symbols **may** have been used in the labelling of this product or products associated with this product.

Symbol	Description	Symbol	Description
	Manufacturer		Keep away from sunlight
IVD	*In vitro* diagnostic medical device	PLATE	Plate
REF	Catalogue number	CONJ	Conjugate
Σ n	Sufficient for *n* tests	CTRL +	Positive Control
LOT	Batch code	CTRL -	Negative Control
	Use by	CAL	Calibrator
	Temperature limitation	DIL SPE	Sample Diluent
CONT	Contents	SOLN TMB	TMB
UDI	Unique Device Identifier	SOLN STOP	Stop Solution
	Consult the warnings and precautions	WASH 10X	Wash Buffer Concentrate (10X)
	Consult electronic instructions for use	EN	English
	Store in the upright position	Made in the USA	Made in the USA
RX Only	Applicable for U.S.A: Prescription *in vitro* diagnostic product		Corrosive
	Hazardous Communication	EC REP	European Commission Authorized Representative
CE	Conformity with Directive 98/79		

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EC REP

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