IDEIA™ *Borrelia burgdorferi* IgG

REF **K602911-2**

Σ **96** IVD CE

An enzyme immunoassay for determination of human serum IgG antibodies to *Borrelia burgdorferi* sensu lato.

Un dosage immunoenzymatique pour déterminer la Presence d'anticorps IgG dirigés contre *Borrelia burgdorferi* sensu lato dans le sérum humain.

Enzymimmunoassay zur Bestimmung von IgG-Antikörpern gegen *Borrelia burgdorferi* sensu lato in Humanserum.

p 1-12 in English (incl.)
p 13-25 in French
p 26-38 in German
p 39 references (incl.)

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IDEIA™ *Borrelia burgdorferi* IgG
November 2006 9807057EFG

1 INTENDED USE

The IDEIA™ *Borrelia burgdorferi* IgG test is an enzyme immunoassay for the detection of human serum IgG antibodies to *Borrelia burgdorferi* sensu lato. The kit is intended as an aid in the diagnosis of Lyme borreliosis.

2 SUMMARY

Lyme borreliosis is a multisystem infection caused by the tickborne spirochete *B. burgdorferi* sensu lato[1,2]. Lyme borreliosis is the most common vector-transmitted human disease in Europe and North America. The disease has also been recognized in the former USSR, China and Japan.

Infection with *B. burgdorferi* is characterized by a variety of clinical symptoms and is divided into three stages[3].

In stage I, a characteristic skin rash, erythema migrans (EM), develops within days to some weeks after the tick bite. The infection may be accompanied by non-specific symptoms such as headache, malaise, fever, myalgia and arthralgia; In stage II - appearing weeks to months later - the disease usually presents with lymphocytic meningoradiculitis with or without cranial nerve palsies and paresis of the extremities (Bannwarth's syndrome), myocardis or arthritis; Stage III, involving skin, central nervous system or joints, may present as acrodermatitis chronica atrophicans (ACA), chronic progressive encephalomyelitis or arthritis occurring up to several years after infection.

Owing to the extreme paucity of *B. burgdorferi* in pathological lesions and body fluids, culture and the most recent PCR techniques for direct detection of spirochetal DNA have not been successful in the routine diagnosis of Lyme borreliosis. At present, therefore, the most suitable method is still the measurement of *B. burgdorferi*-specific antibodies.

IDEIA™ *Borrelia burgdorferi* IgG uses purified, native *B. afzelii* strain DK1 flagellum as test antigen. The flagellum is highly immunogenic and elicits an early, strong and persistent immune response[4,5] Compared with the conventional and currently most widely used test antigen, which is based on whole cell extract of the spirochete, the purified, native flagellar antigen improves the diagnostic sensitivity and specificity of serological assays for Lyme borreliosis[6,7]. Furthermore, the use of flagellum as test antigen is appropriate in all geographical areas as the flagellum shows no significant variation between *B. burgdorferi* strains.

3 PRINCIPLE OF THE TEST

The IDEIA™ *Borrelia burgdorferi* IgG utilises microwells coated with purified, native *Borrelia* flagellum. Antibodies present in human serum specimens will bind to the flagellum-coated microwells during the first incubation. After washing of the microwells to remove unbound serum proteins, peroxidase-Conjugated antibody to human IgG is added to the microwells. The Conjugate binds specifically to human IgG antibodies attached to the flagellum antigen. Excess Conjugate is removed by washing, and after addition of chromogen the bound peroxidase catalyses the development of a blue colour. The reaction is stopped by addition of acid, changing the blue colour to yellow. The intensity of the colour corresponds to the concentration of *B. burgdorferi*-specific antibodies in the specimen. Colour intensity is determined spectrophotometrically at 450nm and the absorbance value of the specimen is compared with the absorbance values of the controls.

4 DEFINITIONS

The following symbols have been used throughout the product information.

REF	Product code and catalogue number
	Consult the instructions for use
	Contains sufficient for <N> tests
	Manufactured by
IVD	*In vitro* diagnostic medical device
	Use by
LOT	Batch Code
2°C 8°C	Storage temperature limitations

5 REAGENTS PROVIDED

96 - Each kit contains sufficient materials for 96 determinations. - The shelf life of the kit is as indicated on the outer box label.

5.1 IDEIA™ *BORRELIA BURGDORFERI*, IgG TEST CONTENTS

	One Instructions For Use booklet.
MICROTITRATION PLATE	96 well Microtitration Plate (12 strips of 8 microwells) coated with native *Borrelia* flagellum. A resealable plastic pouch is provided for storage of unused microwells.

One bottle of each of the following:

SAMPLE DILUENT	100mL Sample Diluent: Buffered solution with detergent, antimicrobial agent and coloured red dye.
WASH BUFFER (X25)	50mL Wash Buffer concentrate x25: Tris Buffered solution with detergent and antimicrobial agent.
IgG CUT-OFF CONTROL	2.5mL IgG Cut-off Control: Human serum in buffer with antimicrobial agent and coloured light green.
IgG POSITIVE CONTROL	1.5mL IgG Positive Control: Human serum in buffer with antimicrobial agent and coloured dark green.
ANTI-IgG CONJUGATE	13mL anti-IgG Conjugate: Rabbit anti-human IgG conjugated with horseradish peroxidase and coloured light blue.
SUBSTRATE TMB	12mL Substrate: stabilized peroxide and 3,3'-5,5'-tetramethylbenzidine in a dilute buffer solution. TMB has been reported to be non-carcinogenic. However, personal protective equipment is recommended to avoid direct exposure.
STOP SOLUTION	25mL Stop Solution: 0.46mol/L sulphuric acid.

5.2 PREPARATION, STORAGE AND RE-USE OF KIT COMPONENTS

The IDEIA™ *Borrelia burgdorferi* IgG kit format allows for up to 10 individual runs during any 3 month period within the expiry date. In order to ensure optimal kit performance, it is important that unused kit components are stored according to the following instructions:

5.2.1 Microwells - MICROTITRATION PLATE

Open the plate pouch by cutting along the seal. Break-off the required number of microwells and relocate them into the frame. One strip allows double determination of one patient specimen; the remaining 6 microwells of the strip are used for Sample Diluent, IgG Cut-off Control and IgG Positive Control. Each additional strip allows for testing of specimens from 4 patients. When all strips are used at one time, specimens from 45 patients can be tested.

Remove unwanted microwell strips from the plate frame and immediately place in the resealable plastic pouch with the dessicant. Carefully reseal the pouch and store at 2-8°C. Microwells may be used for up to 12 weeks after initial opening, provided they are stored in this manner.

5.2.2 Sample Diluent - SAMPLE DILUENT

Ready to use. Store unused Sample Diluent at 2-8°C.

5.2.3 Wash Buffer Concentrate - WASH BUFFER (X25)

Provided x25 concentrate. Prepare working strength Wash Buffer by adding 1 part of Wash Buffer concentrate to 24 parts of fresh deionised or distilled water (or add the contents of Wash Buffer concentrate to 1200mL fresh deionised or distilled water). **Prepare working strength Wash Buffer as required on the day of use**. Store unused concentrate at 2-8°C.

Do not store unused working strength Wash Buffer for subsequent use (see Section 8.2.11).

5.2.4 IgG Cut-off Control - IgG CUT-OFF CONTROL

Ready to use. Store unused IgG Cut-off Control at 2-8°C.

5.2.5 IgG Positive Control - IgG POSITIVE CONTROL

Ready to use. Store unused IgG Positive Control at 2-8°C.

5.2.6 Anti-IgG Conjugate - ANTI-IgG CONJUGATE

Ready to use. Store unused anti-IgG Conjugate at 2-8°C.

5.2.7 Substrate - SUBSTRATE TMB

Ready to use. Store unused Substrate at 2-8°C.

5.2.8 Stop Solution - STOP SOLUTION

Ready to use. Store unused Stop Solution at 2-8°C.

6 ADDITIONAL REAGENTS

6.1 REAGENTS

Fresh deionised or distilled water for preparation of working strength Washing Buffer.

7 EQUIPMENT

The following equipment is required:

Measuring cylinder (2L)

Test tubes of approximately 5mL capacity

Precision micropipettes and disposable tips to deliver 10-1000µL volumes

8-channel pipette to deliver 100µL (optional)

Reagent reservoirs for 8-channel pipette (optional)

Clean absorbent paper (onto which microwells can be tapped dry)

Plastic lid for microwell plate

Microtitration plate shaker capable of a minimum speed of 500rpm with an orbital diameter of 3-4mm. For information on suitability of plate shakers contact your local Oxoid subsidiary or distributor

Timer

Automated plate washer (optional) or suitable equipment for washing 8 microwell strips (Section 10.2.3)
Note: If washing less than 8 test microwells in a strip using an automated washer with an 8 microwell head, it is important to completely fill the strip with blank microwells

Spectrophotometer or EIA plate reader able to read a 96 microwell plate of 8 microwell strips at an absorbance of 450nm with a reference at 620-650nm. (Optional, Section 10.3, Reading the Test Results)

Application notes for use on open automated systems are available for this assay. Contact your local Oxoid subsidiary or distributor

8 PRECAUTIONS

IVD - For *in vitro* diagnostic use. Anyone performing an assay with this product must be trained in its use and must be experienced in laboratory procedures.

8.1 SAFETY PRECAUTIONS

8.1.1 Each donor serum used for the preparation of the IgG controls has been tested individually and found negative for hepatitis B virus surface antigen and antibodies to HIV and hepatitis C.

8.1.2 Stop Solution contains sulphuric acid (0.46mol/L). Avoid eye and skin contact by wearing protective clothing and eye protection.

8.1.3 Do not eat, drink, smoke, store or prepare foods, or apply cosmetics within the designated work area where reagents and samples are handled.

8.1.4 Do not pipette materials by mouth.

8.1.5 Wear disposable gloves whilst handling clinical specimens and always wash hands after working with infectious materials.

8.1.6 Dispose of all clinical specimens in accordance with local legislation.

8.1.7 Safety data sheet available for professional user on request.

8.2 TECHNICAL PRECAUTIONS

8.2.1 Components must not be used after the expiry date printed on the labels. Do not mix or interchange different batches/lots of reagents, with the exception of standard reagents (Sample Diluent, Wash Buffer Concentrate, Substrate and Stop Solution) which are also used in IDEIA™ *Borrelia burgdorferi*, IgM (Code No. K603011-2) and IDEIA™ Lyme Neuroborreliosis (Code No. K602811-2).

8.2.2 The reagents are provided at fixed working concentrations. Test performance will be affected if reagents are modified or stored under conditions other than those detailed in Section 5.2.

8.2.3 Avoid contamination of reagents.

8.2.4 Use separate disposable pipettes or pipette tips for each specimen, control or reagent in order to avoid cross contamination of either specimens, controls or reagents which could cause erroneous results.

8.2.5 Store deionised or distilled water for dilution of concentrated reagent in clean containers to prevent microbial contamination.

8.2.6 Avoid contamination with metal ions and oxidising agents.

8.2.7 Do not use Substrate showing a blue colour prior to its addition to the microwells.

8.2.8 Protect Substrate from light.

8.2.9 Microwells cannot be re-used.

8.2.10 Manual or automated washing equipment, must be free of microbial contamination, be correctly calibrated and maintained according to manufacturer's instructions.

8.2.11 Do not store unused working strength Wash Buffer for subsequent use. When not in use Wash Buffer reservoirs should be rinsed in deionised or distilled water and left to dry.

9 COLLECTION AND PREPARATION OF SPECIMENS

9.1 SPECIMEN COLLECTION AND STORAGE

IDEIA™ *Borrelia burgdorferi* IgG test is for testing of human serum specimens only. Testing of turbid and viscous sera may lead to unreliable results due to pipetting errors. Serum specimens may be stored for 14 days at 2-8°C prior to testing, and for up to 6 months at -20°C or below.

9.2 PREPARATION OF SPECIMENS

Dilute serum specimens 1+200 by adding 10µL of serum to 2mL of Sample Diluent.

10 TEST PROCEDURE

PLEASE REFER TO SECTION 8.2, TECHNICAL PRECAUTIONS, BEFORE PERFORMING THE TEST PROCEDURE.

10.1 PROCEDURAL NOTES

10.1.1 Standard reagents (Sample Diluent, Wash Buffer Concentrate, Substrate and Stop Solution) and a similar assay procedure are also used in IDEIA™ *Borrelia burgdorferi* IgM (Code No. K603011-2) and IDEIA™ Lyme Neuroborreliosis (Code No. K602811-2). This offers the opportunity to run each of the assays in parallel using a single dilution of patient serum.

10.1.2 The validation of the assay performance is based on duplicate testing of all specimens. If the patient specimens are to be run singly, it is recommended that users do so only after becoming familiar with the performance characteristics of the assay. The intra-assay variation (CV%) is approximately 50% higher when based on single determination.

10.1.3 If a shaker capable of 500rpm is not available, the speed of shaking microwells strips during the 2 incubation steps may be reduced to 300rpm without adverse effect on the assay performance, or a static protocol is available. Please contact your local Oxoid subsidiary or distributor for further information on a suitable protocol.

10.2 ASSAY PROCEDURE

NOTE: The assay procedure requires the use of a microtitration plate shaker. For information on suitability of shakers contact your local Oxoid subsidiary or distributor.

If multiple strips are used it is recommended that an 8-channel pipette is used for addition of Conjugate, Substrate and Stop Solution.

10.2.1 Specimen and Control Addition

Locate the required number of microwell strips into the microwell holder. Add 100µL of diluted serum to the appropriate microwells. Add 100µL of Positive Control, Cut-off Control and Sample Diluent to separate microwells. (At least 3 Cut-off Control microwells and 2 Positive Control microwells and 1 Sample Diluent microwell should be included with each batch of specimens tested).

10.2.2 Specimen Incubation

Cover and incubate the microwells at room temperature (**20-25°C**) with **shaking** for **60 minutes**.

10.2.3 Washing the Microwells

The microwells should be washed using working strength Wash Buffer (see Section 5.2.3).

The washing technique is critical to the test performance (see Section 8.2.10) and should be carried out so as to ensure complete filling (with a minimum of **350µL** of working strength Wash Buffer) and emptying of the microwells.

Four wash cycles are essential, by either automated or manual washing techniques, which should include a 2 minute soak period during the second wash or a total of a 2 minute soak period during the complete cycle.

Manual Washing
If washing microwells manually, aspirate or shake out the contents of the microwells and using freshly prepared Wash Buffer, ensure complete filling and emptying of microwells. Between each wash step remove all remaining Wash Buffer by tapping the inverted microwells on to clean absorbent paper. Manual washing efficiency is further ensured if the Wash Buffer is delivered at an angle so as to produce a vortex in the microwells. After the final wash, the plate should be inverted and tapped on absorbent paper to remove the last traces of Wash Buffer.

Automated Washing
Automated washers should be programmed to complete 4 wash cycles and to incorporate the equivalent of 2 minutes soaking time during the complete washing cycle. Washers must be correctly calibrated to ensure complete filling and emptying of microwells between each wash. After the final wash, the plate should be inverted and tapped on absorbent paper to remove the last traces of Wash Buffer.

10.2.4 Anti-IgG Conjugate Addition

Add 100µL of anti-IgG Conjugate to each microwell.

10.2.5 Conjugate Incubation

Cover and incubate the microwells at room temperature (**20-25°C**) **with shaking** for **60 minutes**.

10.2.6 Washing the Microwells

The microwells should be washed as in Section 10.2.3.

10.2.7 Substrate Addition and Incubation

Add 100µL of Substrate to each microwell.

Cover and incubate the microwells at room temperature **(20–25°C) without shaking** for **10 minutes**.

10.2.8 Stopping the Reaction

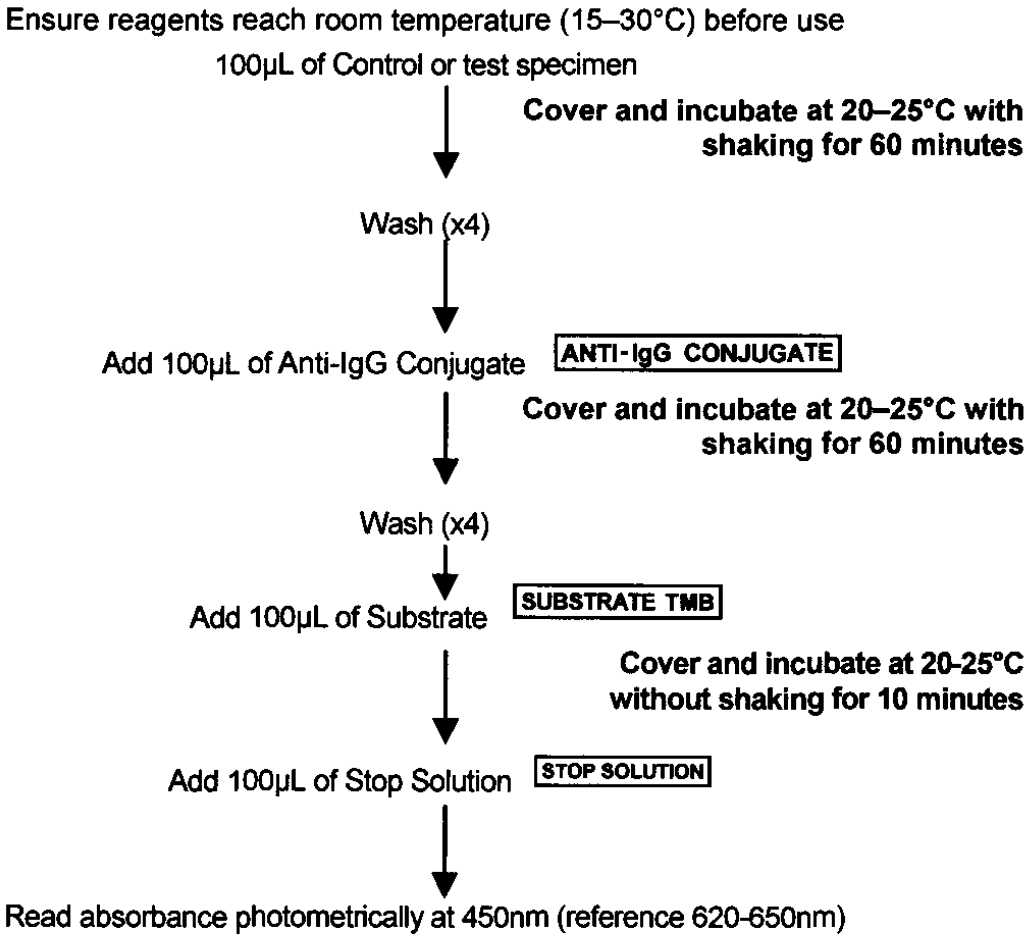
Add 100µL of Stop Solution to each microwell. Ensure thorough mixing in the microwells. The coloured product is stable for 30 minutes. **Do not expose to direct sunlight** as photobleaching of the coloured product may occur.

10.3 READING THE TEST RESULTS

10.3.1 Photometric Reading

The microwells should be read photometrically within 30 minutes after addition of the Stop Solution. Mix the contents of the microwells and read the absorbance of each microwell using a suitable spectrophotometer or EIA plate reader set at 450nm. Ensure that the bottoms of the microwells are clean before reading and check that no foreign matter is present in the microwells. The reader should be blanked on air (ie with no plate in the carriage) before the plate is scanned. Alternatively, if the spectrophotometer or EIA plate reader allows for the use of a reference wavelength (at 620 to 650nm), dual wavelength reading should be performed as this eliminates any potential interference caused by aberrations, such as dirt or marks, on the optical surface of the microwells.

10.4 SUMMARY OF IDEIA™ *BORRELIA BURGDORFERI* IgG ASSAY PROCEDURE



11 QUALITY CONTROL AND INTERPRETATION OF THE TEST RESULTS

11.1 SAMPLE DILUENT

The OD value of the Sample Diluent well must be less than 0.100, but greater than 0.000 (dual wavelength). If the value is above 0.100, inadequate washing or contamination of the Substrate may be the cause. If the value is less than 0.000, the plate reader should be re-blanked on air and the microwells re-read.

If the quality control requirements are not satisfied, test results are invalid and the assay should be repeated.

11.2 IgG CUT-OFF CONTROL AND IgG POSITIVE CONTROL

Calculate the mean OD values for the 3 IgG Cut-Off Control microwells ($OD_{IgG\ Cut\text{-}Off}$) and for the 2 IgG Positive Control microwells ($OD_{IgGPositive}$). Individual OD values should not differ more than 25% from the mean OD value. If one of the OD values of the IgG Cut-Off Control differs by more than 25% from the mean OD value, it should be omitted from the calculation and the mean re-calculated.

The difference between the OD value of IgG Cut-Off Control and IgG Positive Control must be at least 0.500. If this value is less than 0.500 it may be caused by inadequate washing, inadequate shaking during incubations, or low ambient temperature, particularly during incubation with the Substrate.

If the quality control requirements are not satisfied, test results are invalid and the assay should be repeated.

11.3 PATIENT SPECIMENS

Calculate the mean OD value for each patient specimen ($OD_{Specimen}$). Individual OD values should not differ more than 25% from the mean. Any such specimens should be retested. However, if both test microwells show a negative result a difference of more than 25% may be accepted without retesting because low OD values are measured with less precision.

11.4 INTERPRETATION OF RESULTS

11.4.1 Qualitative Interpretation

Compare the mean OD values of specimens ($OD_{Specimen}$) with the mean OD value of the IgG Cut-Off Control ($OD_{IgG\ Cut\text{-}Off}$).

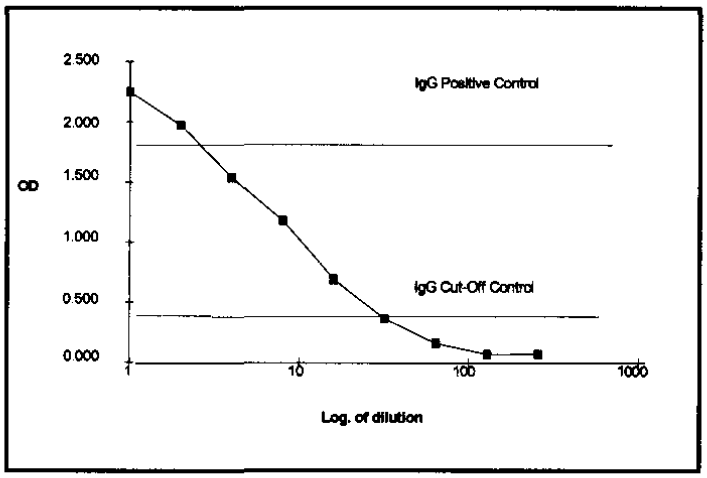
Interpret as follows:

Negative for IgG antibodies to *B. burgdorferi*: $OD_{Specimen} < OD_{IgG\ Cut\text{-}Off}$
Positive for IgG antibodies to *B. burgdorferi*: $OD_{Specimen} \geq OD_{IgG\ Cut\text{-}Off}$

11.4.2 Semiquantitative Interpretation (arbitrary units)

In the range of OD values between $OD_{IgG\ Cut\text{-}Off}$ and $OD_{IgG\ Positive}$, OD values correspond directly to the logarithm of arbitrary units of specific antibody in the specimen. This is illustrated in Figure 1 showing results from an IgG anti-*B. burgdorferi* positive serum serially diluted in negative serum.

Figure 1 Result of a serial 2-fold dilution of a serum positive for IgG antibodies to *B. burgdorferi* in negative serum. In addition, the OD values of the IgG Cut-Off Control and the IgG Positive Control are shown.



The level of specific antibody in the IgG Cut-Off Control has been defined as 1 ($U_{IgG\ Cut\text{-}Off}$ = 1 unit). The level of specific antibody in the IgG Positive Control has been adjusted to 8 x $U_{IgG\ Cut\text{-}Off}$ ($U_{IgG\ Positive}$ = 8 units).

For a specimen the arbitrary units of specific antibody ($U_{Specimen}$) can be calculated utilizing the formula:

$$U_{Specimen} = 10^{a},\ a = \frac{OD_{Specimen} - OD_{IgG\ Cut\text{-}off}}{OD_{IgG\ positive} - OD_{IgG\ Cut\text{-}off}} \times 0.9^{*}$$

$$^{*}\ logU_{IgG\ Positive} - logU_{IgG\ Cut\text{-}off} = log8 - log1 = 0.9$$

Specimens with less than 1 unit of specific antibody are interpreted as negative for IgG antibodies to *B. burgdorferi*. Specimens with 1 or more units of specific antibody are interpreted as positive for IgG antibodies to *B. burgdorferi*.

For specimens with OD values above $OD_{IgG\ Positive}$, the units of specific antibodies to *B. burgdorferi* should be reported as greater than 8.

A change in a patient's specific antibody level can be considered significant when the arbitrary units in a subsequent specimen are either doubled or halved. These are guidelines only.

11.4.3 Comments on Interpretation of Results

Negative results
A negative result does not exclude exposure to *B. burgdorferi*. If Lyme borreliosis is still suspected, an additional specimen should be collected at a later date.

Positive results
A positive result is indicative of a recent exposure to *B. burgdorferi*.

Equivocal results
Any result within ±20% of OD IgG Cut-off should be considered equivocal and interpreted cautiously. A repeat test of such specimens is recommended.

An equivocal result should lead to collection, within 2 weeks, of a second specimen to be tested. If both (or further sequential) specimens yield equivocal results, the patient may be regarded as being IgG negative.

12 PERFORMANCE LIMITATIONS

12.1 A negative result does not exclude the possibility of *B. burgdorferi* infection in the patient. Failure to detect *B. burgdorferi* may be a result of factors such as collection of specimen at an improper time prior to appearance of detectable antibodies, improper sampling, handling of the specimen. Early antibiotic therapy may suppress the antibody response, and some individuals may not produce antibodies at a detectable level.

12.2 A positive result indicates previous immunological exposure and is not proof of active infection.

12.3 All positive results must be interpreted in conjunction with patient related clinical information, epidemiological data and do not justify treatment of a patient. The possibility of exposure to tick bite should always be taken into consideration.

12.4 Test performance will be adversely affected if reagents are modified or stored under conditions other than those detailed in Section 5.2.

13 EXPECTED VALUES

The level of the IgG Cut-Off Control has been adjusted to give a specificity of 98% for normal sera. The antibody response to *B. burgdorferi* flagellum depends on the clinical manifestation of the infection and the duration of the disease. In some of the most common clinical manifestations of a *B. burgdorferi* infection, the diagnostic sensitivity of an indirect IgG assay using purified, native *B. burgdorferi* flagellum as test antigen was reported as follows[4,5]:

Clinical Manifestation	Diagnostic Sensitivity
Erythema migrans	36%
Lymphocytic meningoradiculitis	77%
Acrodermatitis chronica atrophicans	100%

14 SPECIFIC PERFORMANCE CHARACTERISTICS

14.1 SPECIFICITY

The specificity of IDEIA™ *Borrelia burgdorferi*, IgG was evaluated at an independent routine diagnostic laboratory in Sweden. The study was conducted on a panel of 200 serum specimens taken from presumably healthy blood donors living in an area endemic for Lyme borreliosis.

Specificity	
Expected	**Evaluation***
98%	98.5%

*Equivocal results are interpreted as negative.

14.2 DIAGNOSTIC SENSITIVITY

The diagnostic sensitivity of IDEIA™ *Borrelia burgdorferi*, IgG was evaluated at an independent routine diagnostic laboratory in Sweden. The study was conducted on three panels of serum specimens from patients with some of the most common clinical manifestations of a *B. burgdorferi* infection: 45 serum specimens from patients with erythema migrans, 38 serum specimens from patients with lymphocytic meningoradiculitis, and 20 serum specimens from patients with acrodermatitis chronica atrophicans. Results are compared to previously reported expected values[a].

	Diagnostic Sensitivity	
Clinical Manifestation	**Expected**	**Evaluation***
Erythema migrans	36%	38%
Lymphocytic meningoradiculitis	77%	79%
Acrodermatitis chronica atrophicans	100%	100%

* Equivocal results are interpreted as negative.

14.3 CROSS REACTIVITY

Sera from patients with syphilis and inflammatory diseases (rheumatoid factor (RF) positive) were tested in IDEIA™ *Borrelia burgdorferi* IgG with the following results:

Patient sera	Number of sera	Number of positive*
Syphilis	25	0
RF	18	1

* Equivocal results are interpreted as negative.

15 REFERENCES/REFERENZEN

1. **Burgdorferi W, Barbour AG, Hayes SF, Benach JL, Grunwaldt E, Davis JP. (1982)**
Lyme disease - a tick-borne spirochetosis?
Science **216**: 1317-9.

2. **Steere AC, Grodzicki RL, Kornblatt AN, Craft JE, Barbour AG, Burgdorferi W, et al. (1983)**
The spirochetal etiology of Lyme disease.
N Engl J Med **308**: 733-40.

3. **Steere AC. (1989)**
Lyme disease.
N Engl J Med **321**: 586-96.

4. **Craft JE, Duncan KF, Shimamoto GT, Steere AC. (1986)**
Antigens of *Borrelia burgdorferi* recognized during Lyme disease. Appearance of a new immunoglobulin M response and expansion of the immunoglobulin G response late in the illness.
J Clin Invest **78**: 934-9.

5. **Zöller L, Burkard S, Schäfer H. (1991)**
Validity of Western immunoblot band patterns in the serodiagnosis of Lyme borreliosis.
J Clin Microbiol **29**: 174-82.

6. **Hansen K, Pii K, Lebech A-M. (1991)**
Improved immunoglobulin M serodiagnosis in Lyme borreliosis by using a μ-capture enzyme-linked immunosorbent assay with biotinylated *Borrelia burgdorferi* flagella.
J Clin Microbiol **29**: 166-73.

7. **Hansen K, Hindersson P, Pedersen NS. (1988)**
Measurement of antibodies to the *Borrelia Burgdorferi* flagellum improves serodiagnosis in Lyme disease.
J Clin Microbiol **26**: 338-46.

8. **Karlsson M. (1990)**
Western immunoblot and flagellum enzyme-linked immunosorbent assay for serodiagnosis of Lyme borreliosis.
J Clin Microbiol **28**: 2148-50.