Ref: 0514

IMMUNOQUICK® TETANUS

Rapid test for the detection of anti tetanus toxin antibodies in serum, plasma or human whole blood.

CE

INTENDED USE

IMMUNOQUICK TETANUS is an immunochromatographic rapid test for semi-quantitative detection of anti tetanus toxin antibodies in serum, plasma or human whole blood.
IMMUNOQUICK TETANUS is intended for professional use within the framework of laboratory analysis or point of care analysis to detect immunity to tetanus toxin.

INTRODUCTION

Clostridium tetani is a bacterium that causes tetanus in humans. Clostridium tetani are Gram-positive, spore-forming rods that are anaerobic. If they enter the body through a wound, they can multiply and produce a toxin that affects the nerves and controls the activity of muscles. Toxin of Clostridium tetani binds to membrans of peripheric nervous cells and inhibits the release of neurotransmitters.

Antibodies to tetanus toxin are produced in the human by the injection of chemically inactivated tetanus toxin (tetanus toxoid). Immunization is the best way to prevent C. tetani infections in children and adults. Moreover, injection of specific and purified anti tetanus toxin IgG is used in order to refrain toxin action during an acute infection.
It is sometimes better to know the level of anti tetanus toxin antibodies in a patient, to evaluate their immune status, in order to determine the necessity of a complementary vaccination which would assure an immunity towards tetanus toxin.

In emergency situations, it is important for the clinician to know the immune status in order to decide on the correct antitetanus prophylaxis for high risk patients (deep wounds).

TEST PRINCIPLE

IMMUNOQUICK TETANUS is an immunochromatographic test for the detection of anti tetanus toxin antibodies in human blood, plasma or serum. The test uses purified tetanus toxoid (a non-pathogenic derivative of tetanus toxin) coated onto a membrane and a latex-tetanus toxoid conjugate in the sample pad. When human blood, plasma or serum is applied with dilution buffer on the membrane, the tetanus antibodies in the specimen bind to the conjugate at the sample pad level. Then, the conjugate sample flows through the membrane. If tetanus antibodies are present, the sample-conjugate will bind to the immobilized tetanus toxoid on the membrane to form one grey line at the "T" position. Appearance of a purple line at the "C" (control) position is indicative of proper flow characteristics and acceptable test results.

MATERIAL PROVIDED

- 20 aluminium pouches: each one includes one cartridge with a dessicant.
- 20 disposable plastic pipettes
- One vial of dilution buffer
- One instruction of use
- One card with barcode to identify the test lot and to check the compatibility of the SD card when used with the BIOSYNEX Reader.

MATERIAL REQUIRED BUT NOT PROVIDED

- Timer
- 10 µl laboratory pipette (for use with serum or plasma)

STORAGE AND STABILITY

The kit can be stored between 2°C to 30°C. Avoid exposing the kit to hot and cold. DO NOT FREEZE. Do not use beyond the expiration date indicated on the box.

PRECAUTIONS

1. For *in vitro* use only
2. Samples and reagents should be brought to room temperature before running the test.
3. Do not use the test beyond the expiry date. Use of the test beyond the expiry date may lead to incorrect results.
4. Do not pipet samples or reagents by mouth.
5. Follow the product insert instructions carefully.
6. Remove the test cartridge from its sealed pouch just before running the test.
7. Do not eat, drink or smoke when operating samples or reagents.
8. Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
9. Avoid splashes and aerosol formation
10. Humidity and temperature can adversely affect results.
11. IMMUNOQUICK TETANUS contains sodium azide as a preservative which may be toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive salts. On disposal, flush with large quantities of water.
12. Handle all specimens as if they contained infectious agents. Observe established precautions against microbiological hazards throughout all procedures and follow the standard procedures for proper disposal of specimens.

SPECIMEN COLLECTION AND HANDLING

It is recommended that patient samples should be tested immediately.
Specimens may be refrigerated for up to 3 days at 2-8°C following sample collection.
Serum specimens may be frozen for 6 months at -20° C.

TEST PROCEDURE

1. Bring samples and reagents to room temperature before running the test.
2. Open the pouch and remove the test cartridge from its sealed pouch. Put the test on a flat surface.
3. Collection and preparation of sample :

   a. Capillary whole blood
   1. Clean the finger. Puncture the finger with a single use lancet. .
   2. Rub down the finger in order to collect a big drop of blood.
   3. Use the plastic pipette to pipette the drop from the finger.
   4. Add exactly one drop of blood (20 µL), into the sample well of the cartdrige.

   b. Serum, plasma or venous whole blood (EDTA or heparin)
   1. Collect a sample of serum on dry tube, plasma, or whole blood on EDTA or heparin tube.
   2. Add exactly one drop of blood (20 µL) with the plastic pipette or 10µl of serum or plasma with a laboratory pipette into the sample well of the cartridge.
5. Wait till the drop of serum, plasma or whole blood is fully absorbed into the sample well and then add 3 drops of the dilution buffer in the sample well of the cartridge.
6. Read the results at ten minutes.
7. After reading, discard the IMMUNOQUICK TETANUS cartridge according to potential biohazard wastes. Final disposal must be in accordance with local legislation.

RESULTS

The test result can be read visually or with the help of the ***BIOSYNEX Reader***.

➢ **Visual reading:**

Expected values :

According to WHO, a concentration of 0.1 IU/ml in serum is considered as protective against tetanus.
Absence of T band corresponds to absence of protection which can be found in non vaccinated or old vaccinated patients.
Note: There is no relationship between the intensity of the T grey line and the concentration anti tetanus antibodies in the sample.

POSITIVE RESULT:

One grey band appears at the lowest part of test "T" zone and a second purple control band appears at the control "C" zone.
Sample is considered positive with an antibody level above 0.1 IU/ml.

NEGATIVE RESULT :

Only one purple band appears at the level of control "C" zone. The sample is considered negative (with anti tetanus antibody level below 0.1 IU/ml).

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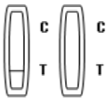
INVALID RESULT:



If no control band « C » appears, the result is invalid and a new test should be performed with a new cartridge, even if T band appears.

➢ **Reading with the reader**

- The IMMUNOQUICK TETANUS test is compatible with the BIOSYNEX Reader, in combination with the SD card 'IMMUNOQUICK TETANUS'.
  To read results with the reader, please refer to the reader Instructions for use.
- The barcode printed on the supplied card should be scanned to identify the test lot and to check the compatibility of the SD card.

QUALITY CONTROL

A procedural control is included in the test. A purple line appearing in the control region (C) is the internal procedural control. It confirms sufficient specimen volume and correct procedural technique. A clear background is also required.
It is also recommended to use positive and negative controls to check the test performances for each new batch. Add 20 µl (1 drop) of positive control or one drop of a negative control on the sample well of the cassette. Add 3 drops of buffer in the sample well. **If positive control is needed, it can be requested at BIOSYNEX.**

TEST LIMITATIONS

IMMUNOQUICK TETANUS gives a semi-quantitative result of IgG anti tetanus antibodies on whole blood, plasma or serum.
Absence of Test band after 5 minutes is indicative of a low and non protective titre of antibodies.
Choice of prophylaxis should also rely on clinical criteria (deepness and dirtiness of the wound).

PERFORMANCES

Sensibilty

Detection limit : Determination of detection limit of IMMUNOQUICK TETANUS was performed with NIBSC standard 76/589. Detection limit of anti toxoid tetanus was assessed on serum. This study indicates a detection limit of IMMUNOQUICK TETANUS at 0.1 UI/ml on serum for a sample of 10µl of serum.
The detection limit in whole blood is determined by transposition of the detection limit in serum. A detection limit of 0,1 IU/ml for a 10µl serum sample applies to whole blood provided that the sample volume is doubled i.e. 20µl blood

Hook effect: The NIBSC standard # 02/232 (lyophilised anti tetanus toxin antibodies) has been tested, on 3 lots, to high concentration up to 120 IU/mL. No hook effect was observed. Samples with high antibodies concentration can be tested.

Specificity

Interferences : Samples containing Bilirubin, triolein and albumin to different concentration has been tested on triplicate. For the three components, test results comply with expected results: no interference has been detected.

Anticoagulants : Whole blood has been sampled with EDTA tube and with Heparin tube to obtain plasma samples. Both kinds of sample have been tested to compare test results. No difference has been observed between plasma EDTA and heparin. Whole blood can be sampled with EDTA or heparin without differences.

Precision

Reproductibility : Reproductibility has been tested and validated on several batches. Samples (negative, low and medium / strong positive sample) have been tested several times, on different days and places by several persons. For all the tests, the obtained result complies with expected result.

Repeatability : Samples (negative, low and medium / strong positive sample) have been tested 10 times on one batch. For all the tests, the obtained results comply with the expected ones.

Clinical study

- A clinical study was performed in emergency room on capillary blood. Same patients were also tested on serum and venous blood in the laboratory. Corresponding sera were also measured with ELISA kit (Genzyme Virotech ref EC 124.00). Results are summarized hereunder :

Capillary blood (reading in emergency room)		ELISA titer			
		Positive	Limit (0.1)	Negative	Total
IQ Tetanus	Positive	139	1	0	140
	Negative	29	8	1	38
	Total	168	9	1	178

Negative if = 0,1	
Specificity	90,00%
Sensibility	82,74%

Positive if = 0,1	
Specificity	100,00%
Sensibility	79,10%

Venous blood (reading in the laboratory)		ELISA titer			
		Positive	Limit (0.1)	Negative	Total
IQ Tetanus	Positive	156	0	0	156
	Negative	12	9	1	22
	Total	168	9	1	178

Negative if = 0,1	
Specificity	100,00%
Sensibility	92,86%

Positive if = 0,1	
Specificity	100,00%
Sensibility	88,14%

Serum (reading in the laboratory)		ELISA titer			
		Positive	Limit (0.1)	Negative	Total
IQ Tetanus	Positive	154	0	0	154
	Negative	14	9	1	24
	Total	168	9	1	178

Negative if = 0,1	
Specificity	100,00%
Sensibility	91,67%

Positive if = 0,1	
Specificity	100,00%
Sensibility	87,01%

- Specificity of IMMUNOQUICK TETANUS was further assessed on whole blood preparations.

		ELISA	
		Positive samples	Negative samples
IMMUNOQUICK TETANUS (Whole blood)	Positive	1	0
	Negative	1	12

Literature

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SYMBOLS

	Attention, see instructions for use	LOT	Lot number
IVD	For *in vitro* diagnostic use only		Manufacturer
2°C 30°C	Store between 2-30°C		Do not reuse
Σ	Tests per kit	REF	Catalog number
	Expiry		

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