Biomaghreb

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STREPTOLYSIN-O

Determination of anti streptolysin - O

IN VITRO DIAGNOSTIC USE

IVD

REF	**48011**	6 x 3 doses (50 Tests)
REF	**48028**	10 x 3 doses (100 Tests)

PRINCIPLE

A,C,G groups of haemolytic b streptococcus secreted an enzyme : Streptolysin- O. wich present an haemolytic activity in its reduced form. This streptolysin-O causes antibody formation revealed by a neutralization reaction of its enzymatic haemolytic activity towards rabbit red blood cells.

REAGENT COMPOSITION

- Streptolysin lyophilisate
- Streptolysin buffer in 10 times concentrated form: It is to be diluted for use: 1 part buffer to 9 parts distilled water, in case of crystallization, reheat to 37°C to put the crystals back into solution.

A 1% rabbit red blood cell suspension in streptolysin buffer 1x is conventionally used. However, the high titre of the streptolysin used makes it possible to obtain identical results with very carefully washed human red blood cells of group O: 3 washes in physiological water. The last base is taken up in the streptolysin 1x buffer.

Reconstitution of the titrated Streptolysin: Insert a hollow needle through the rubber stopper so that the vial is filled with air without risk of losing any of the lyophilisate.

Uncap and uncap the vial to introduce exactly 8 ml of isotonic buffered solution. Recap and promote solution by shaking moderately. The streptolysin is then in its active form.

SAFETY CAUTIONS

Biomaghreb reagents are intended for qualified personnel, for in vitro use (do not pipette with the mouth).

- Consult the current MSDS available on request or on www.biomaghreb.com;
- Check the integrity of the reagents before use; and
- Disposal of waste: comply with the legislation in force.

For safety reasons, treat any specimen or reagent of biological origin as potentially infectious. Respect the legislation in force.

MACROMETHOD SAMPLES

The sera to be examined must not show haemolysis. They must be inactivated by heating at 56°C for 30 minutes (If sera which have been inactivated for more than 24 hours are used, it is recommended that they be placed again in a water bath at 56°C for 10 minutes).

PRESERVATION AND STABILITY

- **Lyophilisate :** our streptolysin has a shelf life of about 3 years at + 4° C.
- **Reconstituted:** it must be used within 5 hours.

PROCEDURE

(See Table 1)

Primary dilutions :

In 2 haemolysis tubes A and B, introduce 2 ml of buffered isotonic solution, add 0.5 ml of serum to be studied in tube A (dilution made 1/5). Mix and transfer 1 ml from tube A to tube B (dilution carried out at 1:15).

Preparation of the titration dilutions of the sera to be examined.

For each serum, prepare 2 sets of tubes numbered 1-2-4-6-8-10 for serum stock dilution A (at 1/5) and 3-5-7-9 for serum stock dilution B (at 1/15) and two tubes 11 and 12 for the red blood cells and streptolysin controls.

Proceed according to Table 1 in geometric progression. Distribute the volumes indicated in ml.

Table 1 :

Usual values are less than 200 ASU samples with value equal or greater than 200 ASU are considered pathological.

Tubes n°	1	2	4	6	8	10	3	5	7	9	11	12
Dilutions	1/5	1/10	1/20	1/40	1/80	1/160	1/15	1/30	1/60	1/120		
Isotonic solution Buffer		0,25	0,25	0,25	0,25	0,25		0,25	0,25	0,25	0,5	0,25
Dilution A (serum 1/5)	0,25	0,25	0,25		0,25	0,25						
Dilution B (serum 1/15)							0,25	0,25	0,25	0,25		
Strepto reconstituée	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25		0,25
Shake and leave for 10 minutes at laboratory temperature												
1% Erythrocytes Suspension	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25
Leave for 10 minutes at laboratory temperature, then 20 minutes at 37°C, centrifuge for 3 minutes at 1500 rpm.												
Aslo amount according to the last non-haemolyzed tube	50	100	200	400	800	1600	150	300	600	1200	No lysis	Total hemolysis

MICROMETHOD

Prepare the dilutions of the test sera (See Table 2):

- Dilution 1/5: 0.1 ml of serum + 0.4 ml of buffer
- Dilution 1/15: 0.1 ml serum + 1.4 ml buffer

Spread on microplate (in ml)

Table 2 : MICROMETHOD

Usual values are less than 200 ASU. Samples with value equal or greater than 200 ASU are considered pathological.

	1st RANGE						2nd RANGE						
CUP NUMBER	**1**	**2**	**3**	**4**	**5**	**6**	**1**	**2**	**3**	**4**	**5**	**Controls**	
Serum Dilution	1/5	1/10	1/20	1/40	1/80	1/60	1/15	1/30	1/60	1/120	1/240	Erythrocy	Strepto
Isotonic Buffer Solution		0,05	0,05	0,05	0,05	0,05		0,05	0,05	0,05	0,05	0,10	0,05
1/5 Serum Dilution	0,05	0,05											
1/15 Serum Dilution							0,05	0,05					
Previous mix		-->	0,05- >	0,05- >	0,05- >	0,05- >		-->	0,05->	0,05- >	0,05- >		
Reconstituted Streptolysin	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05		0,05
	Shake well and leave for 20 minutes at laboratory temperature												
1% Erythrocytes suspension Introduce from right to left	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05	0,05
	Allow 10 minutes at laboratory temperature then 20 minutes at 37°. C Centrifuge the plate for 2 minutes at 500 g or allow to settle at 18-25°												
Antistreptolysin amount of the serum studied according to the last non-haemolyzed tube	50	100	200	400	800	1600	150	300	600	1200	2400		


Manufacturer


Use by

In Vitro Diagnostic


Temperature Limitation

Catalogue number


See insert


Keep away from light


Sufficient for < n > essais

Batch number