

SALMONELLA TYPHI H MICRO

Determination of antibodies associated with Salmonella Typhi H infections by coloured bacterial suspension in microplate

TEST SUMMARY

The Antibodies associated with Salmonella Typhi H infections cause agglutination of inactive bacteria present in suspension. The intravital colouring permits an easier reading of agglutination formation.

SAMPLES

Serum. Stability 6 days at 4°C.

REAGENTS

Suspension: Coloured intravital inactive bacterial suspension; conservative and stabilizer.

Salmonella
Positive control: Solution of rabbit antisera that gives a clear agglutination with Salmonella Suspension; conservative and stabilizer.

Negative control: Proteic bovine solution that doesn't react with suspension; conservative and stabilizer.

REAGENTS PREPARATION

The bacterial suspension must be resuspended with much care, shaking many times by inversion.

The Positive Control be diluted 1:10 with physiologic solution (100 µl + 900 µl).

Stability: the components of this kit will remain stable until the expiration date stated on the label, when stored at 2-8°C protected from direct light. Do not freeze.

MATERIAL REQUIRED BUT NOT SUPPLIED

Physiologic solution. Automatic micropipette. Normal laboratory equipment.

PRECAUTIONS

Reagent may contain not reactive and conservative components. It is opportune to avoid contacts with the skin and do not swallow.

Perform the test according to the general "Good Laboratory Practice" (GLP) guidelines.

SAMPLES PREPARATION

The serum must be diluted 1:10 with physiologic solution (100 µl of serum with 900 µl of physiologic).

PROCEDURE

In a microplate with "U" wells dilute the serum with physiologic solution as indicated in the following table.

Using the same pipette (inspiring and discharging many times) mix carefully content of the second well and transfer 100 µl in the following well etc.

Discharge 100 µl from last well (well n°9).

Well	1	2	3	//	9	Susp. Contr.	Contr. -	Contr. +
Physiolo.	--	100 µl	100 µl	//	100 µl	100 µl	--	--
Diluted serum	100 µl	100 µl	100 µl from 2	//	100 µl from 8	--	--	--
Discharge 100 µl from well n°9								
Diluted Positive control	--	--	--	-	--	--	--	100 µl
Negative control	--	--	--	-	--	--	100 µl	--
Bacterial suspens.	100 µl	100 µl	100 µl	//	100 µl	100 µl	100 µl	100 µl
Titre	1:20	1:40	1:80	//	1:5120	--	--	--

Shake the plate by slow rotations for 20-30 sec.
Incube at 37 °C for 16-18 h or at 22°C for 2 days, to improve bottoms formation it is advisable put the plate in the fridge after the incubation for 2 hours.

RESULTS INTERPRETATION

A coloured bottom with a clear point shape, on the well bottom, indicates negativity.

An agglutinate that cover all the well bottom indicates a clear positivity, while, a no uniform agglutinate with a bottom in the centre, on the well bottom, indicate a feeble positivity.

The serum titre is given by a high dilution in which there is a feeble positivity.

DIAGNOSTIC VALUES

Titres until 1:40 are considered negative; from 1:80 to 1:160 are suspect, and from 1:320 are positive.

It is a distinctive sign for the infection diagnosis the significant increase of titre between examined samples after some days.

NOTE

- If the results are incompatible with clinical presentation, they have to be evaluated within a total clinical study.

CALIBRATION/QUALITY CONTROL

Positive and Negative control sera should be always used to distinguish an eventual background's agglutination of reactive.

TEST PERFORMANCE

Sensitivity

In presence of high antibodies titres, phenomenon of prozone can happen, therefore positivity is absent for low dilutions also being present for higher dilutions.

Specificity

A comparison with an available commercial method gave following results on 50 samples compared, giving a specificity = 100%.

		LTA srl		TOT.
		+	-	
COMPETITORS	+	17	0	17
	-	0	33	33
	TOT.	17	33	50

WASTE DISPOSAL

Product is intended for professional laboratories. Waste products must be handled as per relevant security cards and local regulations.

PACKAGING

CODE BM00100/P

S. typhi H micro suspension 5 x 10 ml
Salmonella Positive control 1 x 0.5 ml
Negative control 1 x 0.5 ml
"U" bottom plate with 96 well 1

CODE BM00105/P

S. typhi H micro suspension 3 x 10 ml
Salmonella Positive control 1 x 0.5 ml
Negative control 1 x 0.5 ml
"U" bottom plate with 96 well 1









REFERENCES

Widal F. – Bull. Men. Soc. Med. Hop de Paris – 6; 26 (1986) Bergey's Manual of Determinative Bacteriology 8 Th Ed. Williams and Wilkins Co (1974) Weil E., Felix A.-Wein.Klin.Woch 29; 974 (1916) Guaitney J.B. e coll. – Microagglutination procedures for febrile agglutination tests-Applied microbiology-4; 635-640 Vol.22 (1971) Rose N.R., Friedman H.-Manual of clinical Immunology-American Society for Microbiology, II ed.

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SYMBOLS

-  Only for IVD use
-  Lot of manufacturing
-  Code number
-  Storage temperature interval
-  Expiration date
-  Warning, read enclosed documents
-  Read the directions
-  Biological risk

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