THYMOL TURBIDITY TEST 300 (TTT 300)

Cat. No. 10003149

Store at (+2 to +25) °C

Reagent Set for the preparation of 1000 ml of working solution used for the performance of thymol turbidity test with serum. The Set is sufficient for at least 300 analyses.

 Principle

Serum beta-globulins, gama-globulins and lipoproteins are precipitated by thymol reagent at pH 7.55. Resulting turbidity corresponds to the concentration of individual protein fractions in serum and its intensity is measured using turbidimetry.

References

Mac Lagan, N. F.: J. Exper. Path. 23, 234 (1944)

Reagents

Concentrated thymol solution (17 ml)
Tris buffer 11 mmol/l, maleic acid 3.36 mmol/l, thymol 6.66 mmol/l
Calibration solution I (11 ml)
sulphuric acid 2.5 mol/l
Calibration solution II (5 ml)
barium chloride 48 mmol/l

Composition of reaction mixture

Tris buffer, pH 7.55 (25 °C) 0.160 mmol/l
Thymol 0.098 mmol/l
Volume ratio of serum reaction mixture 1/61

Reference values

s S-TTT (Shank-Hoagland Units)
The upper limits (S-H U.) 4 – 5
Pathological values (S-H U.) over 5

The range of reference values is only approximate, it is recommended to all laboratories to verify the extention of reference interval for their concrete examined population.

Reproducibility

Approx. ± 8 %

Auxiliary solution (not included in the Set)
Physiological solution

Prepare by dissolving of 0.9 g of sodium chloride in 100 ml of distilled water.

Working solutions

Solution 1 Into 1000 ml volumetric flask pour approx. 900 ml of distilled water and under continuous stirring on magnetic stirrer pipette slowly 15.0 ml of Reagent 1. The pipette tip must be immersed into water in the volumetric flask. Refill with distilled water up to the mark and stir for another 10 min.

Solution 2 Into 250 ml volumetric flask pipette 10 ml of Reagent 2, refill up to the mark with distilled water cooled to approx. +8 °C and stir.

Solution 3 Into 50 ml volumetric flask pipette 1.50 ml of Reagent 3 and refill up to the mark with Solution 2 cooled exactly to +10 °C and stir thoroughly.

Stability: several months when stored at (+15 to +25) °C.

Procedure

Wavelength (620–660) nm, 1 cm cuvette, temperature (+15 to +25) °C

In two test tubes mix Solution 1 in the ratio 60:1 with serum (sample), or with physiological solution (control solution 1) – for example, mix 3.0 ml of Solution 1 and 0.05 ml of serum, or physiological solution.

In another test tube, mix physiological solution in ratio 60+1 with serum (control solution 2) – for example, mix 3.0 ml of physiological solution and 0.05 ml of serum. Stir and let stand for exactly 30 min.

Then stir again and read the absorbance of sample (A) against control solution 1. With chylous serum, read the absorbance of the sample against control solution 2.

<table>
<thead>
<tr>
<th>Pipette (ml)</th>
<th>Solution 2 (ml)</th>
<th>Solution 3 (ml)</th>
<th>Turbidity Units</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
<td>0.05</td>
<td>–</td>
<td>0.05</td>
</tr>
<tr>
<td>Solution 1</td>
<td>3.00</td>
<td>3.00</td>
<td>–</td>
</tr>
<tr>
<td>Physiol. sol.</td>
<td>–</td>
<td>0.05</td>
<td>3.00</td>
</tr>
</tbody>
</table>

Stir and let stand for exactly 30 min. Then stir again and read the absorbance of the sample against control solution 2.

Calibration

By dilution of Solutions 2 and 3 prepare turbidity standards corresponding to (5–20) Shank-Hoagland Units (S-H U.).

<table>
<thead>
<tr>
<th>Solution No.</th>
<th>Solution 2 (ml)</th>
<th>Solution 3 (ml)</th>
<th>Turbidity Units</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4.50</td>
<td>1.50</td>
<td>5</td>
</tr>
<tr>
<td>2</td>
<td>3.00</td>
<td>3.00</td>
<td>10</td>
</tr>
<tr>
<td>3</td>
<td>1.50</td>
<td>4.50</td>
<td>15</td>
</tr>
<tr>
<td>4</td>
<td>–</td>
<td>6.00</td>
<td>20</td>
</tr>
</tbody>
</table>

In test tubes, mix Solutions 2 and 3 and after exactly 30 min stir thoroughly and read the absorbance against dist. water.

The determination is carried out at the same wavelength as the sample.

Notes

If the turbidity of the sample exceeds 20 S-H Units, repeat the determination with a sample diluted with physiological solution in the ratio 1 + 1 (result x2).

Health protection

For in vitro diagnostic use. To be handled by entitled and professionally educated person.

Reagents of the set contain sulphuric acid (R2, 25%) – corrosive substance and thymol in low concentration (R1, 6,7%) – corrosive substance, very flammable methanol (R1, 3%) – toxic substance, very flammable ethanol (R1, 66%) and very flammable methanol (R1, 3%) – toxic substance, in low concentration. Reagent 3 does not contain dangerous substances.

While working it is especially significant to observe sanitary regulations, not to eat, drink, or smoke and to use self protectives.

First aid

At an accidental ingestion drink approx. 0.5 l of water. At an eye contact flush immediately and thoroughly with large quantity of water. At a skin contact wash the affected spot with soap and warm water. In all serious cases of health damage consult the physician.

Waste disposal

All tested samples should be treated as potentially infectious and with the contingent rest of the reagents should be liquidated in accordance with any other local and national regulations relating to the safe handling of such materials.

Put packaging paper waste and rinsed containers to recycling.

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