

DAVIES-HINTON FLOCCULATION TEST
for
SPINAL FLUID

REAGENTS.

1. Glycerinated Hinton Indicator (See Page ____, PREPARATION OF GLYCERINATED HINTON INDICATOR).

2. Hinton negative human serum.

- a. Select one or more clear Hinton negative serums and retest in accordance with the RAPID HINTON TEST technic, (See page ____), employing the following two indicated quantities.

Tube 1.-0.5 ml. serum and 0.5 ml. glycerinated Hinton indicator
Tube 2.-0.1 ml. serum and 0.5 ml. glycerinated Hinton indicator

(NOTE: When large numbers of spinal fluids are tested it is convenient to pool, Seitz filter, merthiolate (1-10,000), and perform Rapid Hinton test as above. Store at 8 - 10°C. for not more than 3 weeks. Avoid the use of cloudy serum.)

3. Gum acacia, 20 percent solution.

- a. Put in a 4-oz. bottle* 20 gms. of white powdered gum acacia (USP).
- b. Add 100 ml. of 3 percent salt solution in distilled water.
- c. Place bakelitecap on bottle, but do not screw on.
- d. Place bottle in the autoclave and maintain for 15 minutes at 15 lbs. of pressure.
- e. Remove from autoclave, screw cap on bottle, shake well to completely dissolve the acacia and maintain in a sterile condition.

*Flint glass bottles with bakelite screw caps with vinylite liners are very satisfactory for the preparation of the acacia solution.

PRELIMINARY TEST OF HINTON NEGATIVE SERUM-GUM ACACIA SOLUTION MIXTURE.

1. For every ten spinal fluids to be tested mix 5 ml. Hinton negative serum with 5 ml. 20 percent gum acacia.
2. Perform a rapid test as follows:
 - a. Into ^{a tube} tube No. 1 pipette 0.6 ml. of 0.85 percent sodium chloride solution, 0.2 ml. of freshly mixed acacia-serum mixture, 0.2 ml. glycerinated Hinton indicator and mix well.
 - b. Place tube in a 37°C. water bath for 30 minutes.
 - c. Centrifuge tube at 2000 r.p.m. for 5 minutes.
 - d. A satisfactory acacia-serum mixture yields a negative reaction.

PREPARATION OF SPINAL FLUID.

Centrifuge and decant spinal fluid. Fluids which are visibly contaminated with bacteria are unsatisfactory for testing. A centrifuged bloody spinal fluid should be reported as unsatisfactory if it yields a positive reaction.

SPINAL FLUID FLOCCULATION TEST.

1. Arrange test tubes (100 x 11½ mm. O.D.) in 4 rows so that there are 4 tubes (one behind the other) for each spinal fluid to be tested and for positive and negative spinal fluid controls. Number tubes to correspond to the identifying number of each fluid.
2. For each specimen pipette 0.6 ml. of spinal fluid into correspondingly numbered tube in first row, 0.4 ml. into tube

in second row, 0.2 ml. into tube in third row, and 0.1 ml. into tube in last row.

3. Add 0.2 ml. of acacia-serum mixture to every tube.
4. Add 0.2 ml. of glycerinated Hinton indicator to every tube.
5. Shake racks of tubes vigorously until contents become completely homogenous.
6. Place racks of tubes in a 37°C. water bath for 16 hours.
7. Remove all tubes from water bath and centrifuge at 2000 r.p.m. for 5 minutes.

READING AND REPORTING.

1. Remove tubes gently from centrifuge without disturbing contents.
2. Before a suitable artificial light, (see READING AND REPORTING of Hinton tests), tap each tube gently at the base while holding it near the top.
3. Report as POSITIVE all reactions that show definite floccules dispersing downward from the meniscus in either tube.
4. Recentrifuge all other tubes at 2000 r.p.m. for 5 minutes.
5. Remove tubes from the centrifuge and examine with tapping as previously described.
6. Report as follows:

POSITIVE: Definite floccules dispersed downward from the meniscus in either tube.

DOUBTFUL: Questionable flocculation in either tube.

NEGATIVE: Absence of flocculation with a ground glass appearance.