

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 four 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.

Tube 3.-0.1 ml. serum and 1.0 ml. glycerinated Hinton Indicator.

Tube 4.-0.1 ml. serum and 2.0 ml. glycerinated Hinton Indicator.

(NOTE: When large numbers of spinal fluid are tested it is convenient to pool, Seitz filter, merthiolate (1-10,000), and retest Hinton negative serums. Store at 8-10°C. for not more than 3 weeks. Avoid the use of cloudy serum and serum more than 3 weeks old.)

3. Gum acacia, 20 percent solution.

- a. Dilute 2 parts of 30 percent gum acacia solution containing 4.5 percent sodium chloride, (obtainable from Eli Lilly Co., Indianapolis, Indiana), with one part of 0.85 percent sodium chloride solution. Store in refrigerator and discard when cloudy. Small quantities may be autoclaved at 15 pounds pressure for 15 minutes, and maintained under sterile conditions until used.

b. Perform preliminary tests on each lot of acacia solution.



- d. Centrifuge both tubes at 2000 r.p.m. for 5 minutes.
- e. A satisfactory acacia-serum mixture yields a negative reaction.

#### PREPARATION OF SPINAL FLUID

Centrifuge and decant spinal fluid. Fluids which are visibly contaminated or contain gross blood are unsatisfactory for testing.

#### SPINAL FLUID FLOCCULATION TEST

1. Arrange test tubes (100 x 11 $\frac{1}{4}$  mm. O.D.) in 2 rows in suitable racks so that there are 2 tubes (one behind the other) for each spinal fluid to be tested and for positive and negative spinal fluid and saline controls. Number tubes to correspond to the identifying number of each fluid.
2. Pipette 0.6 ml. of spinal fluid into each of two correspondingly numbered tubes.
3. Add 0.2 ml. of acacia-serum mixture to every tube.
4. Add 0.2 ml. of glycerinated Hinton Indicator to each tube of the first row.
5. Add 0.6 ml. of glycerinated Hinton Indicator to each tube of the second row.
6. Shake racks of tubes vigorously until contents become completely homogenous.
7. Place racks of tubes in a 37°C. water bath for 16 hours.
8. 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.