

SER: Seminal fluid – Semenogelin Rapid Stain Identification (RSID) Semen Test

Principle The Rapid Stain Identification (RSID)-Semen Test is a test for semenogelin, a protein found in seminal fluid, a component of semen.

The RSID™-Semen system consists of a test cassette containing two monoclonal antibodies specific to human semenogelin.

Equipment The following equipment is used for this procedure:

- microcentrifuge tubes
 - sterile cotton swab
 - pipettes
 - barrier pipette tips.
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Reagents The following prepared reagents are used in this procedure:

- RSID™ test cassettes
 - RSID™ extraction buffer
 - RSID™ running buffer
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Controls The following controls must be run daily before casework samples:

- positive control (semen or seminal fluid stain)
 - negative control (saline or RSID Extraction Buffer)
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Continued on next page

SER: Seminal fluid – Semenogelin Rapid Stain Identification (RSID) Semen Test, Continued

Quality control Each new lot of cassettes must be tested with a positive and negative control to evaluate the *Test Line* band. If problems are noted, that lot will not be used for casework.

A record of the quality check for new lot numbers including a photograph documenting the results is kept in the *Quality Control Log Book* located in the Biology Laboratory.

NOTE: Refer to the log book for previous band intensities observed for the positive control to determine whether a new lot of cassettes is acceptable for casework.

Positive control preparation The positive control is prepared using the following procedure:

Step	Action
1	Add 50 µl of known semen or seminal fluid to a sterile cotton swab and let air dry.
2	Extract the entire swab in 1mL of saline for 1-2 hours.
3	Add 1 µl of extract to 99 µl of RSID™ semen running buffer. This entire volume will be used as the positive control.

Prepare frozen pre-aliquoted positive controls using the following procedure:

Step	Action
1	Perform the Steps 1 and 2 above.
2	Place 1 µl of the extract and 19 µl of deionized water or saline into small tubes and freeze. Prior to freezing, test one of the tubes in the batch to ensure that it is working properly.
3	When preparing the positive control, thaw a 20 µl aliquot.

Continued on next page

SER: Seminal fluid – Semenogelin Rapid Stain Identification (RSID) Semen Test, Continued

Sample preparation

Samples are prepared for analysis using the following procedure:

Step	Action
1	Extract the stain cutting (approximately 5 mm x 5 mm) or swab (approximately one quarter) in 50 µl of saline or RSID Extraction Buffer for approximately 30 minutes.
2	Centrifuge the sample.
3	Pipette 20 µl of the supernatant into a labeled microcentrifuge tube.

Continued on next page

SER: Seminal fluid – Semenogelin Rapid Stain Identification (RSID) Semen Test, Continued

Procedure The RSID test is performed as follows.

Note: Each sample will require a separate cassette.

Step	Action
1	Prepare the positive control as stated above. Prepare the negative control by pipetting 20 µl of saline or extraction buffer into a labeled microcentrifuge tube.
2	Add 80 µl of the running buffer to each of the controls and sample tubes. NOTE: If using freshly prepared positive control from above, do not add additional buffer.
3	Add one of the 100 µl samples to each sample window of the cassettes.
4	After ten minutes, evaluate the cassettes. Record the results.

NOTE: Due to *High Dose Hook Effect* (a false negative reaction when very high levels are present in the sample), negative results for samples that are AP positive with rapid color development must be re-tested as follows:

Step	Action
1	Add 1 µl of the original sample extract to 99 µl of running buffer.
2	Add the 100 µl sample to the sample well of the cassette and record the result at 10 minutes.

Continued on next page

SER: Seminal fluid – Semenogelin Rapid Stain Identification (RSID) Semen Test, Continued

Interpretation A visible red band at the Control (C) position only is a negative (-) result for semenogelin.

A visible red band at both the Control (C) and Test (T) positions is a positive (+) result for semenogelin which indicates the presence of seminal fluid, a component of semen.

If no control (C) band appears, the test is invalid and should be repeated.
