SPIFE Urine/CSF Protein Procedure

Cat. No. 3427 Helena Laboratories

This procedure is intended to be used in conjunction with the appropriate SPIFE Serum Protein procedures provided in the gel kit. It utilizes a template application method for low volumes of concentrated urine or cerebrospinal fluid (CSF). The urine or CSF must be concentrated according to the instructions provided in the kit.

I. Gel Preparation

1. Remove the gel from the protective packaging and discard the overlay.
2. Using a SPIFE Blotter C, gently blot the entire gel using slight fingertip pressure on the blotter. Remove the blotter.
3. Carefully place the gel on the SPIFE Urine IFE Alignment Tray (Cat. No. 3380).
4. The templates have been marked with a hole in one corner. One to three templates can be placed on the gel. Hold the template so that the marked corner is in the lower left position. Align the application slits with the pins on the sides of the Alignment Tray. Place the template on the gel and apply slight fingertip pressure to each template, making sure there are no air bubbles under them. Carefully remove the gel from the Alignment Tray.
5. Dispense approximately 2 mL of REP Prep onto the left side of the electrophoresis chamber.
6. Place the left edge of the gel over the REP Prep aligning the round hole on the left pin of the chamber. Gently lay the gel down on the REP Prep, starting from the left side and ending on the right side, fitting the obround hole over the right pin. Use a lint-free tissue to wipe around the edges of the plastic gel backing, especially next to electrode posts, to remove excess REP Prep. Make sure no bubbles remain under the gel.
7. Clean the electrodes with deionized water before and after each use. Wipe with a lint-free tissue.
8. Place a carbon electrode on the outside ledge of each gel block outside the magnetic posts.

II. Electrophoresis

Using the instructions provided in the appropriate Operator's Manual, set up the electrophoresis parameters as follows for the SPIFE, SPIFE 2000, or SPIFE 3000.

NOTE: A "Dry 1" time and an "Absorb 1" time is recommended below. However, due to variations in environmental conditions, the following ranges are acceptable.

* Dry 1 = 10-15 minutes.

** Absorb 1 = 7-10 minutes

Refer to the options below for the specific instrument type and test kit configuration.

A. SPIFE 3000 - Split Beta SPE 20/40/60

1) Apply Sample to Template, (continue)
   Absorb 1 **10:00 21°C
2) Blot and Remove Template, (continue)
   Electrophoresis 1 8:00 21°C 650V 130mA
3) Remove Gel Blocks, (continue)
   Dry 1 *10:00 54°C

4) No Prompt
END OF TEST

B. SPIFE 3000 - Split Beta SPE 80/100 and SPE 20/40/60

1) Apply Sample to Template, (continue)
Absorb 1 **10:00 21°C
2) Blot and Remove Template, (continue)
Electrophoresis 1 6:00 21°C 650V 130mA
3) Remove Gel Blocks, (continue)
Dry 1 *10:00 54°C
4) No Prompt
END OF TEST

C. SPIFE 2000 - Split Beta SPE 80/100 and SPE 20/40/60

1) Apply Sample to Template, (continue)
Absorb 1 **10:00 21°C
2) Blot and Remove Template, (continue)
Electrophoresis 1 6:00 21°C 650V
3) Remove Gel Blocks, (continue)
Dry 1 *10:00 54°C
4) No Prompt
END OF TEST

D. SPIFE-SPE 20/40/60

1) Apply
No Prompt **10:00 21°C
2) Electrophoresis
Prompt 6:00 21°C 650V
3) Dry
Prompt *10:00 54°C
4) No Prompt
END OF TEST

III. Sample Application

Urine and/or CSF Application

1) Press the **TEST SELECT/CONTINUE** button located on the electrophoresis chamber side of the instrument until the appropriate serum protein option appears on the display.
2) Apply urine and/or CSF by placing 3 µL of each sample onto one of the twenty available slits on the Urine/CSF Template.
3) Close the chamber lid, and press the **START/STOP** button for the electrophoresis chamber. Sample application will be timed for 10 minutes.
4) Gently blot the template with a Blotter A Plus and carefully remove the template.
5) Close the chamber lid, and press the **TEST SELECT/CONTINUE** button to start electrophoresis. SPIFE will beep when electrophoresis is complete.
6) Remove the electrodes, then use the Gel Block Remover to remove the gel blocks. Replace the electrodes on each end of the gel to prevent curling during drying.
7) Close the chamber lid and press the **TEST SELECT/CONTINUE** button to dry the gel.
8) Refer to the appropriate procedure to stain and destain the gels.