

# QuickGel® SPE Procedure

CAT. NO. 3405, 3505, 3505T

## INTENDED USE

The QuickGel SPE System is intended for the separation of serum, cerebrospinal fluid (CSF) or urine proteins by agarose gel electrophoresis using the SPIFE 3000 system or the QuickGel Chamber.

Cat. No. 3405 - for use with SPIFE 3000

Cat. No. 3505 - for use with QuickGel Chamber (Blade application)

Cat. No. 3505T - for use with QuickGel Chamber (Template application)

For In Vitro Diagnostic use.

Rx Only

## SUMMARY

Serum contains over one hundred individual proteins, each with a specific set of functions and subject to specific variation in concentration under different pathologic conditions.<sup>1</sup> Since the introduction of moving-boundary electrophoresis by Tiselius<sup>2</sup> and the subsequent use of zone electrophoresis, serum proteins have been fractionated on the basis of their electrical charge at a particular pH into five classical fractions: albumin, alpha<sub>1</sub>, alpha<sub>2</sub>, beta and gamma proteins. Each of these classical electrophoretic zones, with the exception of albumin, normally contains two or more components. The relative proportions of these fractions have proven to be useful aids in the diagnosis and prognosis of certain disease states.<sup>3-5</sup>

## PRINCIPLE

Proteins are large molecules composed of covalently linked amino acids. Depending on electron distributions resulting from covalent or ionic bonding of structural subgroups, proteins can be either polar or nonpolar at a given pH. In the QuickGel SPE procedure, proteins are separated according to their respective electrical charges on agarose gel using both the electrophoretic and electroosmotic forces present in the system. The proteins are then stained with a visible stain.

## REAGENT

### 1. QuickGel SPE Gel

**Ingredients:** Each gel contains agarose in a tris/sodium barbital/MOPS buffer with salicylic acid, citric acid, stabilizers and a preservative.

**WARNING: FOR IN-VITRO DIAGNOSTIC USE ONLY.** The gel contains barbital which, in sufficient quantity, can be toxic.

**Preparation for Use:** The gels are ready for use as packaged.

**Storage and Stability:** The gels should be stored at room temperature (15 to 30°C) and are stable until the expiration date indicated on the package. The gels must be stored horizontally in the protective packaging in which they are shipped. **DO NOT REFRIGERATE OR FREEZE THE GELS.**

**Signs of Deterioration:** Any of the following conditions may indicate deterioration of the gel: (1) crystalline appearance indicating the agarose has been frozen, (2) cracking and peeling indicating drying of the agarose, (3) bacterial growth indicating contamination, (4) thinning of the gel blocks.

### 2. Acid Blue Stain

**Ingredients:** When dissolved as directed, the stain contains 0.5% (w/v) acid blue stain.

**WARNING: FOR IN-VITRO DIAGNOSTIC USE ONLY. DO NOT INGEST.**

**Preparation for Use:** Dissolve the dry stain (entire contents of vial) in 1 L of 5% acetic acid. Mix thoroughly for 30 minutes.

**Storage and Stability:** The dry stain should be stored at 15 to 30°C and is stable until the expiration date indicated on the package. The diluted stain is stable six months when stored at 15 to 30°C.

**Signs of Deterioration:** The diluted stain should be a homogeneous mixture free of precipitate. Discard if precipitate forms.

### 3. Citric Acid Destain

**Ingredients:** After dissolution, the destain contains 0.3% (w/v) citric acid.

**WARNING: FOR IN-VITRO DIAGNOSTIC USE. DO NOT INGEST - IRRITANT.**

**Preparation for Use:** Pour 11 L of deionized water into the Destain vat. Add the entire package of Destain. Mix well until completely dissolved.

**Storage and Stability:** Store the Destain at 15 to 30°C. It is stable until the expiration date on the package.

**Signs of Deterioration:** Discard if solution becomes cloudy.

### 4. Acid Violet Stain (Optional Urine Stain)

**Ingredients:** The stain is comprised of Acid Violet stain.

**WARNING: FOR IN-VITRO DIAGNOSTIC USE ONLY. DO NOT INGEST.**

**Preparation for Use:** Dissolve the dry stain in 1 liter of 10% acetic acid and mix thoroughly. Fill the SPIFE stain vat.

**Storage and Stability:** The dry stain should be stored at 15 to 30°C and is stable until the expiration date indicated on the package. The stain solution is stable six months when stored at 15 to 30°C in a closed container.

**Signs of Deterioration:** The diluted stain should be a homogeneous mixture free of precipitate.

## INSTRUMENT

A SPIFE 3000 or the QuickGel Chamber must be used to electrophorese, stain, destain, and then dry the gels. The gels may be scanned on the QuickScan Touch Plus (Cat. No. 1640) or a separate densitometer. Refer to the Operator's Manual for detailed instructions.

## SPECIMEN COLLECTION AND HANDLING

**Specimen:** Fresh serum, CSF or urine is the specimen of choice. Use of plasma will cause a fibrinogen band to appear as a distinct narrow band between the beta and gamma fractions.

**Storage and Stability:** If storage of serum is necessary, samples may be stored covered at 15 to 30°C for 4 days, 2 to 8°C for 2 weeks, or -20°C for 6 months.<sup>6</sup> Urine or CSF samples may be stored covered at 2 to 8°C for up to 72 hours or at -20°C for 1 month.

**Urine Sample Preparation:** Urine samples may be run diluted, neat or concentrated. Shake samples to homogenize. Centrifuge desired volume at 2000 x g for 5 minutes. Remove supernatant and concentrate as follows:

Total Protein (mg/dL)	Conc. Factor
<50	100x
100-100	50x
300-300	25x
600-600	10x
>600	5x

**CSF Sample Preparation:** CSF samples may be used after proper concentration (10-50X).

## Interfering Factors:

1. Hemolysis may cause false elevation in the alpha<sub>2</sub> and beta fractions.
2. Inaccurate results may be obtained on specimens left uncovered, due to evaporation.

## PROCEDURE FOR SPIFE 3000

**Materials provided:** The following materials needed for the procedure are contained in the QuickGel SPE Kit (Cat. No. 3405). Individual items are not available.

- QuickGel SPE Gels (10)
- Acid Blue Stain (1 vial)
- QuickGel Blotter C (10)
- Citric Acid Destain (1 pkg)
- Modified Applicator Blade Assembly (10)

## Material provided but not contained in the kit:

ITEM	CAT. NO.
SPIFE 3000 Analyzer	1088
QuickScan Touch Plus	1640
Applicator Blade Weights	3387
SPIFE Dispo Sample Cups (deep well)	3360
Modified Applicator Blades	3451
Gel Block Remover	1115
SPE Normal Control	3424
SPE Abnormal Control	3425
REP Prep	3100
SPIFE Applicator Blades (for Urine & CSF)	3450
Disposable Sample Cups (for Urine and CSF)	3369
QuickGel Dispo Cup Tray	3353
SPIFE QuickGel Electrode	1111
SPIFE QuickGel Holder	3358
SPIFE QuickGel Chamber Alignment Guide	86541003

**Materials needed but not provided:**

- 5% acetic acid
- 0.85% saline

**STEP-BY-STEP METHOD**

**I. Chamber Preparation**

1. The SPIFE QuickGel Chamber Alignment Guide must be used to mark the location for gel placement. It is recommended that the markings be placed directly on the copper floor under the contact sheet.
2. Remove the contact sheet and clean the chamber floor according to instructions in the Operator's Manual.
3. Place the round hole in the guide over the left chamber pin and the obround hole over the right pin.
4. Using an indelible marker, outline the square open area onto the copper floor. Allow marking to dry, and apply another contact sheet.

**II. Sample Blade Application Method**

1. Remove one Disposable Applicator Blade from the packaging. Remove the protective guard from the blades by gently bending the protective piece back and forth until it breaks free.
2. Place the Applicator Blade into the vertical slot numbered 6 in the Applicator Assembly. If using two Applicator Blades, place them into the vertical slots numbered 6 and 12. When testing serum with urine or CSF samples, serum application is made after the second urine or CSF application. Therefore the blade for serum application is not added until after the second urine/CSF application.

**NOTE: The blade assembly will only fit into the slots in the Applicator Assembly one way; do not try to force the Applicator Blades into the slots.**

3. Place an Applicator Blade Weight on top of each blade assembly.
4. Slide the Disposable Sample Cups into the top row numbered 1 to 10 of the appropriate cup tray. If testing more than 10 samples, place cups into both rows.
5. Pipette the following amount of sample into cups 1 to 5 and 6 to 10. If testing more than 10 samples, pipette sample into cups 11 to 15 and 16 to 20. Cover the tray until ready to use.

**NOTE: Application of Urine and CSF samples cannot be done with the Applicator Blades or Cups packaged in the kit. Another Blade (Cat. No. 3450) and Cup (Cat. No. 3369) must be purchased.**

Sample	Volume	Blades	Cups
Serum or control	45 µL	3451	3360
Urine/Concentrated CSF	20 µL	3450	3369

Specimens with insufficient volumes may be run using the QuickGel Accessory Kit (Cat. No. 3426).

6. Carefully cut open the end of the gel pouch. Remove one gel from the protective packaging. Fold and tape the open end of the pouch to prevent drying of the gel. Remove the gel from the plastic mold and discard the mold.
7. Place a QuickGel Blotter C on the gel with the longer edge parallel with the gel blocks. Gently blot the entire gel using slight fingertip pressure on the blotter and remove the blotter.
8. Dispense approximately 1 mL of REP Prep onto the left side of the electrophoresis chamber.
9. Place the gel over the REP Prep inside the rectangle on the chamber floor. Gently lay the gel down on the REP Prep, starting from the left side and ending on the right side. Use 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 the gel remains in place and that no bubbles remain under the gel.
10. Clean the QuickGel Electrodes with deionized water before and after each use. Wipe with a lint-free tissue.
11. Place a QuickGel Electrode on the outside ledge of each gel block inside the magnetic posts. Close the chamber lid. Proceed to Step IV.

**III. Sample Template Application Method**

Template application may be used for testing CSF or urine specimens which have insufficient volumes for blade application.

1. Carefully open one end of the pouch and remove one gel from the protective packaging. Reseal the pouch with tape to prevent

drying of the gel. Remove the gel from the plastic mold and discard the mold.

2. Place a QuickGel Blotter C on the gel with the longer edge parallel with the gel blocks. Gently blot the entire gel using slight fingertip pressure on the blotter and remove the blotter.
3. Dispense about 1 mL of REP Prep onto the left side of the marked area of the chamber.
4. Place the gel over the REP Prep inside the rectangle on the chamber floor. Gently lay the gel down on the REP Prep, starting from the left side and ending on the right side. Use lint-free tissue to wipe around the edges of the gel backing to remove excess REP Prep. Make sure the gel remains in place and that no bubbles remain under the gel.
5. Depending on the number of samples tested, place one or two templates across the gel aligning the slits with the marks on the gel backing.
6. Apply fingertip pressure to the template, making sure there are no bubbles under it. **NOTE: If wearing rubber gloves to perform this step, place a QuickGel Blotter A over the template and then apply fingertip pressure to the template. Remove the blotter. Powder from the gloves can produce gel artifacts.**
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 inside the magnetic posts. Close the chamber lid. Proceed to Step V. (pg.3) for the SPIFE 3000 instructions.

**IV. Electrophoresis with Blade Application**

Using the instructions provided in the appropriate Operator's Manual, set up the parameters as follows for the SPIFE 3000. If testing only serum samples, follow the instructions marked "• Serum". If testing serum with urine or CSF, follow instructions marked "• Serum and CSF or Urine". The blade used for serum application will be added after the second application of urine or CSF.

**NOTE: A "Dry 1" time of 10 minutes is recommended. However, due to variations in environmental conditions, the following ranges are acceptable.**

**\*Dry 1 = 10 to 15 minutes.**

**• Serum**

**Electrophoresis Unit**

- 1) No Prompt  
Load Sample 1    00:30    21°C    SPD1
- 2) No Prompt  
Apply Sample 1    00:30    21°C    SPD1    LOC1
- 3) No Prompt  
Electrophoresis 1    6:30    21°C    375V    80 mA
- 4) Remove gel blocks, (continue)  
Dry 1    \*10:00    54°C
- 5) No prompt  
END OF TEST

**• Serum and CSF or Urine**

**Electrophoresis Unit**

- 1) No Prompt  
Load Sample 1    00:30    21°C    SPD1
- 2) No Prompt  
Apply Sample 1    00:30    21°C    SPD1    LOC1
- 3) No Prompt  
Load Sample 2    00:30    21°C    SPD1
- 4) No Prompt  
Apply Sample 2    00:30    21°C    SPD1    LOC1
- 5) To Continue, (continue)  
Load Sample 3    00:30    21°C    SPD1
- 6) No Prompt  
Apply Sample 3    1:00    21°C    SPD1    LOC1
- 7) No Prompt  
Absorb 1    1:00    21°C
- 8) No Prompt  
Electrophoresis 1    6:30    21°C    375V    80 mA
- 9) Remove gel blocks, (continue)  
Dry 1    \*10:00    54°C

10) No Prompt  
END OF TEST

• **Serum and CSF or Urine**

**Stainer Unit**

1) No Prompt Stain 1	4:00	REC = OFF	VALVE = 3
2) No Prompt Destain 1	1:00	REC = ON	VALVE = 2
3) No Prompt Destain 2	1:00	REC = ON	VALVE = 2
4) No Prompt Destain 3	1:00	REC = ON	VALVE = 2
5) No Prompt Dry 1	*10:00	63°C	
6) No Prompt			
END OF TEST			

1. Press the **TEST SELECT/CONTINUE** button located on the electrophoresis side of the instrument until the **QG-SERUM/URINE PROTEIN** option appears on the display. Open the chamber lid.
2. Place the Cup Tray with samples on the SPIFE 3000. Align the holes in the tray with the pins on the instrument. Close the chamber lid.
3. With **QG-SERUM/URINE PROTEIN** on the display, press the **START/STOP** button. An option to either begin the test or skip the operation will be presented. Press **START/STOP** again to begin. The SPIFE 3000 will apply the samples, electrophorese and beep when completed.
4. If testing serum and CSF or urine, open the chamber lid and place the Modified Blade in place for serum application. Press **TEST SELECT/CONTINUE**. Proceed to Step VI.

**V. Electrophoresis with Template Application**

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

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

\*Dry 1 = 10 to 15 minutes

\*\*Absorb 1 = 7 to 10 minutes

1) Apply Sample to Template, (continue)				
Absorb 1	**10:00	21°C		
2) Blot and Remove Template, (continue)				
Electrophoresis 1	6:30	21°C	375V	80 mA
3) Remove Gel Blocks, (continue)				
Dry 1	*10:00	54°C		
4) No Prompt				
END OF TEST				

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. Press **START/STOP** twice.
2. Apply urine and/or CSF by placing 3 µL of each sample onto one of the ten available slits on the Sample Template(s).
3. Close the chamber lid, and press the **TEST SELECT/CONTINUE** button for the electrophoresis chamber. Sample application will be timed for \*\*10 minutes.
4. Open the chamber lid and gently blot the template with a QuickGel Blotter A and carefully remove the blotter and template. Dispose of templates as biohazardous waste.
5. Close the chamber lid, and press the **TEST SELECT/CONTINUE** button to start electrophoresis. SPIFE will beep when electrophoresis is complete.

**VI. Visualization**

1. After electrophoresis is complete, open the chamber lid and use the Gel Block Remover to remove the gel blocks. Dispose of blades and cups as biohazardous waste. Replace the electrodes on each end of the gel to prevent curling during drying.
2. Close the chamber lid and press the **TEST SELECT/CONTINUE** button to dry the gel.
3. After the gel has been dried, carefully remove the gel from the electrophoresis chamber.

4. Remove the SPIFE QuickGel Holder from the stainer chamber. While holding the gel agarose side down, slide one side of the gel backing under one of the metal bars. Bend the gel backing so that the gel is bowed, and slip the other side under the other metal bar. The two small notches in the backing must fit over the small pins to secure the gel to the holder.

5. Place the SPIFE QuickGel Holder with the attached gel facing backwards into the stainer chamber.

6. With the appropriate test name on the display, press the **START/STOP** button. An option to either begin the test or skip the operation will be presented. Press **START/STOP** to begin. The instrument will stain, destain, and dry the gel.

7. When the process is completed, the instrument will beep. Carefully remove the SPIFE QuickGel Holder from the stainer because the metal piece on the holder will be hot. Scan the bands in a densitometer or the QuickScan Touch Plus.

**PROCEDURE FOR QUICKGEL® CHAMBER**

The following instructions are for using the QuickGel Chamber (Cat. No. 1284) for electrophoresis.

**Materials Provided:** The following materials needed for the procedure are contained in the QuickGel SPE Kit (Cat. No. 3505 or 3505T). Individual items are not available.

**Cat. No. 3505**

- QuickGel SPE Gels (10)
- Acid Blue Stain (1 vial)
- QuickGel Blotter C (10)
- Citric Acid Destain (1 pkg)
- QuickGel Modified Applicator Blades (10)
- QuickGel Dispo Sample Cups (10)

**Cat. No. 3505T**

- QuickGel SPE Gels (10)
- Acid Blue Stain (1 vial)
- QuickGel Blotter C (10)
- Citric Acid Destain (1 pkg)
- QuickGel Blotter A (10)
- QuickGel Templates (10)

**Materials provided but not contained in the kit:**

Item	Cat. No.
QuickGel Chamber	1284
QuickScan Touch Plus	1640
REP Prep	3100
QuickGel Gel Block Remover	1262
SPE Normal Control	3424
SPE Abnormal Control	3425
QuickGel Accessory Kit	3426
Acid Violet Stain	552351
QuickGel Applicator	1265
QuickGel Applicator Base	1266
QuickGel Applicator Weights	1267
QuickGel Dispo Cup Tray	1268
QuickGel Dispo Sample Cups (Deep well)	1259
QuickGel Dispo Sample Cups (Shallow well)	1269
QuickGel Modified Applicator Blades	1271
QuickGel Applicator Blades (for urine and CSF)	1270
QuickGel Applicator Kit (includes Applicator, Applicator Base, Weights, and Cup Tray)	1274

**Materials needed but not provided:**

- 5% acetic acid
- 0.85% saline
- Power Supply capable of providing at least 400 Volts.

**STEP BY STEP METHOD**

**NOTE: The use of templates for sample application is offered as an option instead of the Applicator Blade. Instructions are provided for both methods in Section II.**

**I. Chamber Preparation**

1. The QuickGel Chamber must be plugged into a power supply. Set a timer for \*5:45 minutes and the power at 400 Volts. \*An electrophoresis time of 5:15 to 6:15 minutes is acceptable.
2. Snap the Electrophoresis Lid into place on the chamber.
3. Ensure that the chamber floor is cool (room temperature) before starting the test.

**II. Sample Application**

**A. QuickGel Applicator Blade**

**NOTE: Application of Urine and CSF samples cannot be done with the Applicator Blades or Cups packaged in the kit. Another Blade (Cat. No. 1270) and Cup (Cat. No. 1269) must be purchased.**

Sample	Volume	Blades	Cups
Serum or control	45 µL	1271	1259
Urine/Concentrated CSF	20 µL	1270	1269



Specimens with insufficient volumes may be run using the QuickGel Accessory Kit (Cat. No. 3426).

1. Remove one QuickGel Applicator Blade from the packaging. If testing more than 10 samples, remove two Applicator Blades from the packaging.
2. Urine and CSF samples must be applied on the gel three times. When testing serum with urine or CSF samples, serum application is made after the second urine or CSF application. Therefore the blade for serum application is not added until after the second urine/CSF application.
3. Place an Applicator Weight on top of each Applicator Blade. If using two Applicator Blades, place them into the vertical slots A and C of the Applicator. One blade should be placed into the slot corresponding to cup placement.
4. Place the appropriate number of QuickGel Disposable Sample Cups into either or both Row A and Row C of the Dispo Cup Tray. Pipette the appropriate amount of specimen into the Sample Cups and cover tray until ready to use. When ready, place the Dispo Cup Tray into the Applicator Base.
5. Carefully cut open one end of the gel pouch. Remove one gel from the protective packaging. Fold and tape the open end of the pouch to prevent drying of the remaining gel. Remove the gel to be used from the plastic mold and discard the mold.
6. Dispense approximately 1 mL of REP Prep onto the left side of the electrophoresis chamber.
7. Place the left notch of the gel so that it fits the left pin of the chamber floor and gently roll the gel to the right side fitting the right notch to the right pin of the chamber floor. Use a lint free tissue to wipe around the edges of the gel backing to remove any excess REP Prep. Make sure that no bubbles remain under the gel.
8. Place a QuickGel Blotter C on the gel with the longer edge parallel with the gel blocks. Gently blot the entire gel using slight fingertip pressure on the blotter and remove the blotter.
9. If testing only serum samples, use the following steps 10 through 15. If testing serum and urine or CSF on the same gel, perform steps 10 through 14 twice for the urine and CSF samples. Place the Modified Blade for serum samples into the applicator, and repeat steps 10 through 14 for a total of 3 applications for urine and CSF samples and 1 application for serum samples.
10. While holding the white Applicator knob up, place the Applicator into the designated slot on the Applicator Base aligning the small red dots on the Applicator with those on the Base.
11. Slowly lower the Applicator Knob allowing the blades to enter the sample cups, and immediately start a timer for 30 seconds.
12. After 30 seconds, lift the Applicator Knob. Immediately place the Applicator into the slot on the chamber floor, aligning the red dots.
13. Slowly lower the Applicator Knob to apply sample to the gel. Set a timer for 30 seconds.
14. After the 30 seconds application, raise the Applicator Knob and remove the Applicator from the chamber.
15. Close the lid, press the power switch to turn on the chamber and start the power supply. Proceed to Step III.

### B. Template Application

1. Serum specimens and controls are diluted 1:5 (1 part serum with 4 parts 0.85% saline). Urine samples may be run diluted, neat or concentrated. CSF samples must be concentrated as instructed in **SPECIMEN COLLECTION AND HANDLING**.
2. Carefully cut open one end of the gel pouch. Remove one gel from the protective packaging. Fold and tape the open end of the pouch to prevent drying of the remaining gel. Remove the gel to be used from the plastic mold and discard the mold.
3. Dispense approximately 1 mL of REP Prep onto the left side of the electrophoresis chamber.
4. Place the left notch of the gel so that it fits the left pin of the chamber floor and gently roll the gel to the right side fitting the right notch to the right pin of the chamber floor. Use a lint free tissue to wipe around the edges of the gel backing to remove any excess REP Prep. Make sure that no bubbles remain under the gel.
5. Place a QuickGel Blotter C on the gel with the longer edge parallel with the gel blocks. Gently blot the entire gel using slight fingertip pressure on the blotter and remove the blotter.
6. Remove one QuickGel Sample Template from the package if only one row of samples is tested. Remove two templates if two rows are tested. Hold the template so that the small hole in the corner is toward the front right side of the chamber.

7. Carefully place the template(s) on the gel aligning the template slits with the marks on each side of the gel backing. The center hole in the template(s) should align with the indentation in the center of the gel.
8. Apply slight fingertip pressure to the template making sure there are no air bubbles under it. **NOTE: If wearing rubber gloves to perform this step place a QuickGel Blotter A over the template and then apply fingertip pressure to the template. Remove the blotter. Powder from the gloves can produce gel artifacts.**
9. Use the following chart to apply the appropriate serum dilution or urine/CSF concentration to the template slits. After the last sample application, allow time for the proper absorption.

	Sample Volume	Absorption Time
Serum or Control	1 µL	3 minutes
Urine/CSF	3 µL	7 to 10 minutes

10. Use the QuickGel Blotter A to gently blot the excess sample from the template. Carefully remove the blotter and the template(s). Dispose of templates as biohazardous waste.
11. Close the lid, press the power switch to turn on the chamber and start the power supply.

### III. Electrophoresis and Staining

1. Electrophorese the gel for \*5:45 minutes at 400 Volts.
2. Turn off the Power Supply and the QuickGel Chamber.
3. Using the Gel Block Remover, remove the two gel blocks from the gel. Again, use a lint free tissue to wipe around the edges of the gel backing to remove any excess moisture.
4. Replace the Electrophoresis Lid with the Drying Lid. Clean the two electrodes on the Electrophoresis Lid with deionized water after each use. Wipe with a lint-free tissue. Close the lid.
5. Dry the gel for 15 minutes or until dry by turning on only the QuickGel Chamber. After drying is complete, turn the chamber off and remove the gel.
6. Fill a container with prepared stain. Fill another container with Destain solution.
7. Place the gel into the Staining Dish containing the prepared stain for 4 minutes. Remove the gel from the stain and allow it to drain on a blotter.
8. Destain the gel in three consecutive washes of Destain solution. Use a gentle alternately rocking and swirling technique. Allow the gel to remain in each wash for 1 minute. The gel background should be completely clear. Tap the gel to remove the excess destain solution.
9. Ensure that the chamber floor is clean. Place the gel onto the QuickGel Chamber floor, close the Drying Lid and turn the QuickGel Chamber on. Dry the gel for 10 minutes or until dry. Turn off the QuickGel Chamber and remove the gel.

### EVALUATION OF THE PROTEIN BANDS

**Qualitative evaluation:** The urine and CSF samples run on the QuickGel SPE Gel can only be visually inspected for the presence of the bands.

**Quantitative evaluation:** If a QuickScan Touch Plus is used, scan on the acid blue setting.

**Stability of End Product:** The completed, dried QuickGel SPE Gel is stable for an indefinite period of time.

### Quality Control

SPE Normal Control (Cat. No. 3424) and SPE Abnormal Control (Cat. No. 3425) may be used to verify all phases of the procedure and should be used on each gel run. If desired, a control or patient sample may be diluted 1:7 with 0.85% saline (1 part sample + 6 parts saline) and run with urines and CSFs for qualitative comparison. Refer to the package insert provided with the control for assay values.

### REFERENCE VALUES

The reference range presented was established on 50 normal specimens using the QuickGel SPE System on the SPIFE 3000 Analyzer and the QuickGel Chamber. These values are presented as a guideline. Each laboratory should perform its own normal range study.

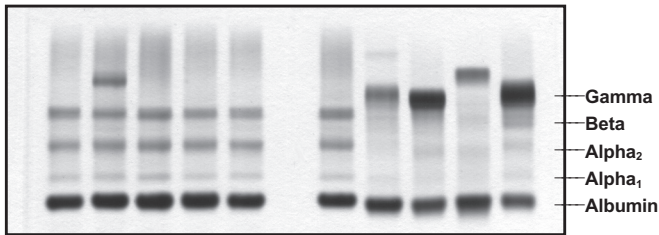
Protein Fraction	% of Total Protein $\bar{X} \pm 2 \text{ S.D.}$
Albumin	46.5 - 58.8
Alpha <sub>1</sub>	1.8 - 4.6
Alpha <sub>2</sub>	9.2 - 16.5
Beta	11.2 - 18.8
Gamma	9.1 - 23.5

**Variations of Expected Values<sup>5</sup>**

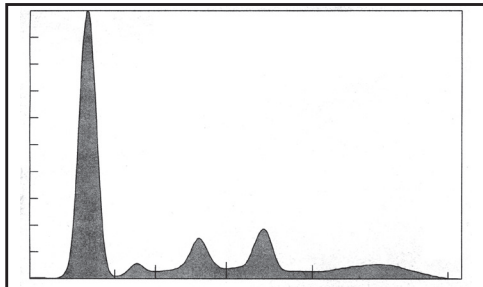
Studies show that values are the same for both males and nonpregnant females. (Some differences are seen in pregnant females at term and in women on oral contraceptives.) Age has some effect on normal levels. Cord blood has decreased total protein, albumin, alpha<sub>2</sub>, and beta fractions and slightly increased alpha<sub>1</sub> and normal or increased gamma fractions (largely of maternal origin). The gamma globulins drop rapidly until about three months of age, while the other fractions have reached adult levels by this time. Adult levels of the gamma globulins are not reached until 16 years of age. The albumin decreases and beta globulin increases after the age of 40.

**RESULTS**

Figure 1 illustrates the electrophoretic mobilities of the albumin, alpha<sub>1</sub>, alpha<sub>2</sub>, beta and gamma protein bands on QuickGel SPE Gel. The fastest moving band, and normally the most prominent, is the albumin band found closest to the anodic edge of the gel. The faint band next to this is alpha<sub>1</sub>, followed by alpha<sub>2</sub>, beta and gamma globulins.



**Figure 1:** A QuickGel SPE Gel showing relative position of the bands.



**Figure 2:** A scan of a QuickGel SPE pattern.

**Calculations of the Unknown**

The Helena QuickScan Touch Plus will automatically calculate and print the relative percent and the absolute value of each band when the total protein is entered. Refer to the Operator's Manual provided with the instrument.

**INTERPRETATION OF RESULTS<sup>5</sup>**

Results on normal individuals will cover age and sex-related variations and day-to-day biologic variations. Disease states in which abnormal patterns are observed include inflammatory response, rheumatic disease, liver diseases, protein-loss disorders, plasma cell dyscrasias, pregnancy and genetic deficiencies.

**Further Testing Required**

The serum protein electropherogram or densitometric tracing should be evaluated for abnormalities. If abnormalities are observed, appropriate follow-up studies should be initiated. These may include immunoelectrophoresis, immunofixation, quantitation of immunoglobulins, bone marrow examination and other appropriate tests.

**LIMITATIONS**

Since all electrophoretic procedures are nonlinear, it is critical to fill the wells with the recommended volume of serum to obtain optimal resolution and reproducible results. Noncompliance with the recommended procedure may affect the results.

**PERFORMANCE CHARACTERISTICS FOR SPIFE 3000**

**PRECISION**

**Within Run:** A normal and an abnormal control were run alternately on a single gel with the following results: N = 7

Normal Control			
Protein Fraction	Mean %	SD	CV
Albumin	54.9	0.8	1.4%
Alpha <sub>1</sub>	3.6	0.2	5.9%
Alpha <sub>2</sub>	12.0	0.3	2.6%
Beta	16.6	0.3	1.9%
Gamma	12.8	0.4	3.2%

Abnormal Control			
Protein Fraction	Mean %	SD	CV
Albumin	45.6	0.6	1.3%
Alpha <sub>1</sub>	3.4	0.3	7.6%
Alpha <sub>2</sub>	11.5	0.3	3.0%
Beta	12.7	0.2	1.3%
Gamma	26.7	0.2	0.6%

**Between-Run:** A normal control and an abnormal control were run in replicate on three lots of gels with the following results: N = 84

Normal Control			
Protein Fraction	Mean %	SD	CV
Albumin	55.1	1.4	2.5%
Alpha <sub>1</sub>	3.5	0.2	6.7%
Alpha <sub>2</sub>	12.1	0.3	2.7%
Beta	16.0	0.8	4.9%
Gamma	13.2	0.7	5.1%

Abnormal Control			
Protein Fraction	Mean %	SD	CV
Albumin	45.3	1.1	2.4%
Alpha <sub>1</sub>	3.4	0.2	5.7%
Alpha <sub>2</sub>	11.6	0.3	2.5%
Beta	13.0	0.3	2.5%
Gamma	26.7	0.6	2.1%

**CORRELATION**

Fifty normal and forty abnormal serum samples, as well as normal and abnormal controls, were analyzed using the SPIFE SPE system and the QuickGel SPE system.

N = 92  
 $Y = 0.999X + 0.054$   
 R = 0.999  
 X = SPIFE SPE  
 Y = QuickGel SPE Gel on SPIFE

**PERFORMANCE CHARACTERISTICS FOR QUICKGEL CHAMBER**

**PRECISION**

**Within Run:** A normal and an abnormal control were run alternately on a single gel with the following results: N = 7

Normal Control			
Protein Fraction	Mean %	SD	CV
Albumin	54.1	0.8	1.5%
Alpha <sub>1</sub>	3.6	0.1	3.1%
Alpha <sub>2</sub>	13.0	0.3	2.5%
Beta	16.2	0.5	3.0%
Gamma	13.1	0.6	4.7%

Abnormal Control			
Protein Fraction	Mean %	SD	CV
Albumin	46.0	1.1	2.4%
Alpha <sub>1</sub>	3.4	0.1	4.3%
Alpha <sub>2</sub>	11.6	0.3	2.6%
Beta	12.7	0.2	2.0%
Gamma	26.3	0.6	2.2%

**Between-Run:** A normal control and an abnormal control were run in replicate on three lots of gels with the following results: N = 84

Normal Control			
Protein Fraction	Mean %	SD	CV
Albumin	55.3	1.4	2.5%
Alpha <sub>1</sub>	3.6	0.3	7.2%
Alpha <sub>2</sub>	12.4	0.4	3.5%
Beta	15.6	0.7	4.7%
Gamma	13.0	0.5	4.0%

Abnormal Control			
Protein Fraction	Mean %	SD	CV
Albumin	46.7	1.3	2.7%
Alpha <sub>1</sub>	3.3	0.2	5.5%
Alpha <sub>2</sub>	11.4	0.4	3.5%
Beta	12.5	0.6	4.5%
Gamma	26.1	0.6	2.4%

## CORRELATION

Fifty normal and forty abnormal serum samples, as well as normal and abnormal controls, were analyzed on the QuickGel Chamber and on the SPIFE SPE System.

N = 92

Y = 0.992X + 0.155

R = 0.998

X = SPIFE SPE

Y = QuickGel SPE on the QuickGel Chamber

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