

NAZAR

Serum Proteins Kit 260 Tests

Electrophoresis Procedure

REF SPE261NZ

Intended use

The Serum Protein Electrophoresis (SPE) kit is intended for the separation of proteins in human serum by electrophoresis on agarose gel strips. Human serum proteins are separated into five zones or bands which are composed of many individuals proteins. The patterns are examined visually for abnormalities. The kit is used with the automated instruments in Agarose Gel Nazar.

Summary

Human body fluids contains a varied mixture of proteins and protein complexes. Each of these protein entities apparently fulfills a specific function within the life process; furthermore, it is well known that the levels of various proteins in blood serum bear a close relationship to states of health and disease. In fact, the concentration and compositions of the over one hundred proteins contained in the serum may vary due to physiological conditions. Electrophoresis is a well established and versatile technique, routinely used in clinical laboratories. Serum protein electrophoresis performed at pH 8.8 yields five bands: Albumin and four globulins (alpha 1, alpha 2, beta and gamma). About sixteen of the known proteins contribute to the formation of the five bands in the electrophoretic pattern. Evaluation of single bands by visual inspection provides valuable diagnostic support as it offers a display of the major proteins involved in functional and pathological processes.

Principle

Electrophoresis separates serum proteins based on the premise that the individual protein species have different mobilities when subjected to an electric field. Every molecule possesses an electrical charge due to the presence of positively charged groups and negatively charged groups. The net charge dictates the migration characteristics of the species at a given pH. With the NAZAR Serum protein procedure, proteins are separated at an alkaline pH using the principle of Zone Electrophoresis on a suitable support medium: Agarose Gel. When the migration is complete, the proteins are stained and fixed with Amido Black solution and then washed with a specific destaining solution.

Warning: This kit is for in vitro diagnostic use only.

Specimen Collection and Handling

Serum samples should be collected using the laboratory's procedures and in accordance with Good Laboratory Practice (GLP) Guidelines. Fresh serum samples without hemolysis or lipemia are the optimal choice for testing. Due to interference of fibrinogen, plasma is not recommended. Serum samples may be stored covered at 15° to 30° C for 4 days or 2° to 6° C for two weeks, or -20° C for 6 months.

Reagents

Reagents are supplied in concentrated solutions and ready to use. Please reference the operator's manual for the correct addition of reagents to the Instrument reagent rack.

Storage and stability: Store all reagents at room temperature (15° to 30°C). All reagents are stable until the expiration date indicated on the label.

Items Provided

Kit code: SPE261NZ For NAZAR Instruments 260 TESTS

Ref	Description	Q.ty
GA85SP10	Plate in Agarose Gel SPE Dimensions 85x100 mm	10
SAB2GA5	Staining 250 mL Amido Black (Ready to use)	1
DES2GA5	Destaining Solution 250 mL Concentrated 20 X Warning: Irritant for eyes and skin	1
SBP01NZ	SET 10 Blotter Papers	1
MBP01NZ	SET sponges buffer	10

Reagents and Materials Required But Not Provided

- Serological pipettes. Preferably pipetting devices 10-100µL
- Distilled Water.
- Serum Protein Control

Test Procedure

Reagents Preparation

1. insert 250 ml of staining (ready to use) in the External tank Staining 500ml
2. insert 250 ml of destaining Solution 20 X in the external Tank Washing Solution. Dilute the destaining solution: Bring it to 5000 ml with Deionized water.
3. Insert deionized water in the external tank 1000 ml
Warning: do not insert distilled water otherwise the external probe does not feel the presence of the liquid.
4. Make sure that the External Tank of Waste 5000 ml is empty

Buffer Paper

Remove the sponges from aluminium container and put them in the external slot of cover of migration chamber.

Samples

Dispense about 30 µL of sample into each well in the first line of the sample Holder. Avoid introducing air bubbles. Bubbles and foam can interfere with results.

Blotting of the agarose gel strip

Blotting of the agarose gel strip
Remove the strip from the plastic blister taking care not to touch the gel surface with your fingers. Blott the paper for about 10 seconds (Ref.SBP01NZ) in order to remove the excess buffer on the strip.

Strip Positioning

Placement of the gel strip in the Strip Holder
Pour 0.5 ml of H₂O into the center of the migration chamber so that the strip adheres perfectly to the surface. put the strip in the elastic slot of the strip holder and insert it in the correct position on the migration chamber. taking care to lift the seal block first as shown in the operator's manual.

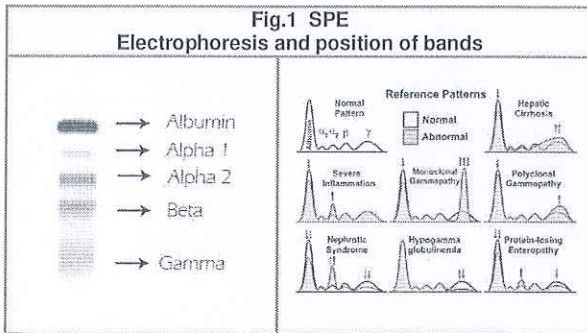
Electrophoresis Conditions

Check on the instrument that the Serum Proteins migration conditions are correct for the specific test. Consult the Operator's Manual for further information on Gemini Software setting



Test Results

Fig. 1. Normal five fraction serum protein pattern on cellulose acetate. From the start point (cathodic edge) of the cellulose acetate strip to the anode, the bands are: Gamma Globulin, Beta Globulin, Alpha 2 Globulin, Alpha 1 Globulin and Albumin. It should be specified that, with conventional serum proteins dyes, a slight or mild staining of the background between the major fractions may occur due to the presence of lipoproteins, whose protein moieties can bind to the dye. In a normal pattern, Albumin, Alpha 1 and Beta fractions appear to be homogeneous, well shaped bands. Alpha 2 and Gamma fractions are diffuse bands with the Gamma fraction showing a more intensely stained zone in its central part.



Reference Values

In the following table, the serum reference ranges were established with SPE kit on NAZAR instruments. These values are presented as a guideline. Each laboratory should establish its own reference values.

Fraction	Reference range (%)
Albumin	52.0 - 68.0
Alpha 1	2.0 - 5.0
Alpha 2	6.5 - 13.5
Beta	8.0 - 14.0
Gamma	11.0 - 21.0

Quality control

It is recommended to include controls with the patient sample runs in accordance with the guidelines or requirements of local, state, and/or federal regulations or accrediting organizations. The laboratory should establish acceptance parameters for each lot of control material. If the controls are not within these parameters, patient test results are suspect and the analysis should be repeated.

Bibliography

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3. Aguzzi F, Jayakar SD, Merlini G., Petrini C. Electrophoresis: cellulose acetate vs agarose gel, visual inspection vs densitometry. Clin. Chem. 1981; 27.
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5. Savoia M., Palmieri D., Guarini R., Oriani G., La Rocca S., Rivisitazione critica del tracciato sieroproteico: contributo sperimentale per la metodologia e le implicazioni fisiopatologiche. Giorn.It.Chim.Clin. Vol. 14, N.1, 1989.
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The compliance of the kit with the directive 98/79/CE and related performance described in the procedure, are guaranteed only if the products are used according to our instructions of use

SAFETY CLAIMS

1. Before starting the assay, read the instructions completely and carefully. Be sure that everything is understood.
2. Don't mix reagents of different lots. Don't use expired reagents.
3. Follow good laboratory practice and safety guidelines. Wear lab coats, disposable latex gloves and protective glasses.
4. If the reagents contained in the kit have a hazardous label, it may cause eye and skin irritations. MSDS for this product are available upon written request to the manufacturer.
5. Chemicals and prepared or used reagents have to be treated as hazardous waste according to national biohazard and safety guidelines or regulations.

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SYMBOLS AND MEANING



PRODUCT CODE



LOT NUMBER



READ THE INSTRUCTIONS



EXPIRATION DATE



STORAGE TEMPERATURE



MANUFACTURER



DO NOT RE-USE



IN VITRO DIAGNOSTIC



CE MARK

CONF
CONFECTION



GIÁM ĐỐC
Lãng Đỗ Dũng