Quality control

To control the quality of each electrophoretic run you should run one lane with a control serum. The corresponding values to reach are given in the products information leavelet.

Interpretation or results

It is recommended that any evaluation of the gels is performed against normal values produced for this method in each individual laboratory.

1. **Qualitative check:** Electrophoresis pattern has to be aproved for additional or missing bands.
2. **Quantitative check:** The calculated values by densitometer have to be aproved against normal values.

In all cases for values outside the normal range must be run further laboratory and analysis tests.

Limitation

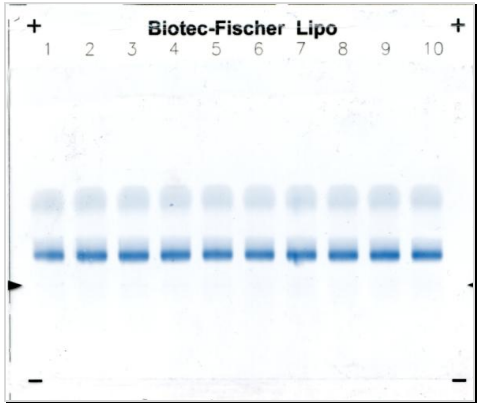
Since all electrophoresis procedures are non-linear, it is important to follow these instructions for use closely to ensure optimal resolution and reproducible results. Failure to follow these instructions for use may affect the results obtained.

Reference values

See type definition in specific literature

Literature / Support

1. Mills, G.L. Patricia A. Lane and P.K. Weech. A guidebook to lipoproteine technique (Laboratory techniques in biochemistry and molecular biology; v. 14)
2. Lena A. Lewis, Jan J. Opplt, Handbook of electrophoresis Volume I, CRC Press inc. 1980



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Manual

Lipoprotein Electrophoresis on Agarosegel

for product code

371601-001 / 371601-002
371601-101 / 371601-102

Intend purpose

The folllowing named reagents are only used to migrate lipo proteines on agarose gels. Human serum contains more than 100 single proteines which every single proteine has its specific function which depends on different physiological conditions. Since the introduction of moving boundary electrophoresis by Tiselius, and the subsequent use of zone electrophoresis, serum proteines have been fractionated on tha basis of their charge at a particular pH. The BIOTEC-FISCHER Lipo-kit enables the Standard separation and the quantification of the four zones of lipo-proteins in human serum or plasm.They have an isoeleciric point at about pH 5,5 above which they are negatively charged. Due to their electrically charge, they'll) migrate in an electric field. A buffered agarose gel is used for the electrophoretic Separation of lipo proteins into four main populations of particles, namely; Chylomicrons (particles that are too large to migrate into the pories oft he stabilising medium, they form a band at the origin), alpha-lipoproteines (their mobility is that of the alpha1-globulins), pre-ß-lipoproteins (their mobllity is close to that of the alpha 2-globulins), ß-lipoproteins (they migrate at the same rate as the beta-globuluis). All these classes are on their turn polydisperse.

Warning

All reagents are for in vitro diagnostic use only. Don not ingest or pipette by mouth any component. Refer to the product safety sheet for component risk and safety phrases in disposal information.

Buffer contains sodiumazide. Do not mix acid solution with sodiumazide to avoid toxic vapours. Always rinse abundantly with water to avoid the formation of metal-azides, because of the potential risk of explosion when piled up in metal conduct-pipes..

Necessary Equipment

Reagents

Prod. No.:	Name
371601-xxx	**Kit for xxx LPE Electrophoresis on Agarosegel** Content: agarose gels for 10 tests per gel thin sheet drying paper for agarose gels sample templates drying paper for sample templates ATX buffer concentrate *Fill up one bottle to 1000 ml with Aqua dest.* 1 x 100 ml Sudan Black staining concentrate *Dilute as discribed on bottle*
Prod. no.:	Name
	For kits with 10 Tests, buffer solution is ready to use

Equipment

Prod. no.:	Name
321001	Electrophoresis tank model 1000
321000	*Electrophoresis tank model BFA (altern.)*
321101	*Electrophoresis tank FILIPO*
341225	Staining vessel kit for agarose gels
332010	Drying oven KB19
313001	MAESTRO 101 (no further equipment necessary)
331000	Densitometer TurboScan

Reagents which are not part of kit

Name
Fixativ
dist. Water : Acetic acid (9 : 1)
Destaining
50% Ethanol solution

Shelf live

Unopened agarose gels will be usable until shelf live marking on the envelope

Sample preparation

Freshly collected serum or plasma is the specimen of choice.

Interfering factors:

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

Sample preparation:
Use undiluted sample material only

Step by step procedure

1. Fill first vessel with fixative, second vessel with staining solution and the last two vessels with destaining solution.
2. Fill one compartment of tank model 1000 with buffer solution until it flows into the second compartment.
3. Tilt the tank so both compartments will be leveled (For Filipo and MAESTRO 101: fill both electrode compatments with 35ml buffer each. For tank model BFA 100 ml for each compartment).
4. Dry middle separator with paper towel
5. Carefully cut gel envelope on opposite small side of label with scissor and carefully remove the gel. Don't press to hard not to damage the gel. Position the gel in front of you so you can read the gel information
6. Carefully remove blue protection foil
7. Place one thin paperwick onto the gel for approx. 1-2 seconds and remove it afterwards carefully
8. Position the sample template with small marking holes to the markers (▶ and ◀).
9. Place small paperwick in the middle of template and move your finger over the wick to remove air bubbles between template and gel.
10. Applicate 3 - 5µl sample to each application slid
11. Wait for 5 minutes until the sample intruded into the gel.
12. Place the small paper wick again onto the template to remove remaining sample and remove template together with paper wick and give it to the waste.
13. Place gel into the gel bridge with gelside down. Care about correct electrical positioning ("-" = black; "+" = red).
14. Move gel bridge into the tank. Care about correct electrical positioning!
15. Close tank lid
16. Run migration at 90-100 Volts for 30 minutes
17. Remove after migration and give it for 5 minutes in first vessel with fixation solution
18. Dry gel at 80°C in drying oven KB19 or use hair dryer.
19. After gel is completely dry give it for 10 minutes to staining solution
20. Move gel to remaining destaining vessel for 5 minutes each at last until there is no more visible background.
21. Again dry the gel like mentioned under point 18.
22. Calculate Gel with TurboScan densitometer