

[54] **ELECTROPHORESIS KIT AND SYSTEM**

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[51] Int. Cl. **B01k 5/00**

[58] Field of Search **204/180 C, 180 S, 204/299**

[56]

References Cited

UNITED STATES PATENTS

3,407,133	10/1968	Oliva et al.	204/299
3,482,943	12/1969	Csizmas et al.	204/299 X
3,494,846	2/1970	Arquembourg.	204/180 G

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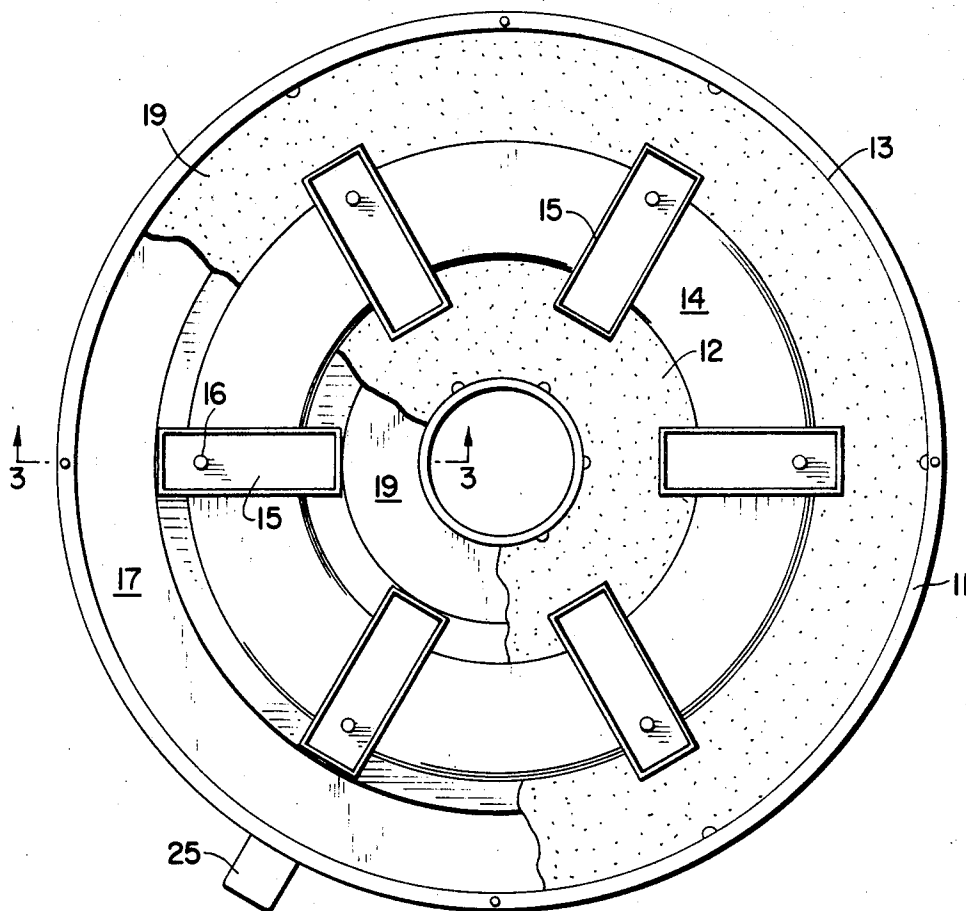
Attorney—Richard G. Wynne, John A. Finken et al.

[57]

ABSTRACT

An electrophoresis kit and system is provided wherein a base unit containing the electrodes and spaced apart buffer chambers and the test samples are contained in the bridges in a corresponding lid in contact with the buffer chambers. The test samples are poured into molding bridge chambers in the base unit, the lid placed thereon, transferring the test sample to the lid, and the lid replaced to make electrical contact with the spaced bridge chambers in the base. Registration means are provided for correctly positioning the lid with respect to the base for operation of the electrophoresis apparatus. The kit may assume a variety of shapes, such as rectangular or circular, and the entire assembly is preferably made of molded consumable plastic material so as to be disposable. The invention includes the method of assembly and operation of the electrophoresis unit.

7 Claims, 5 Drawing Figures



2 Sheets-Sheet 1

FIG. 2

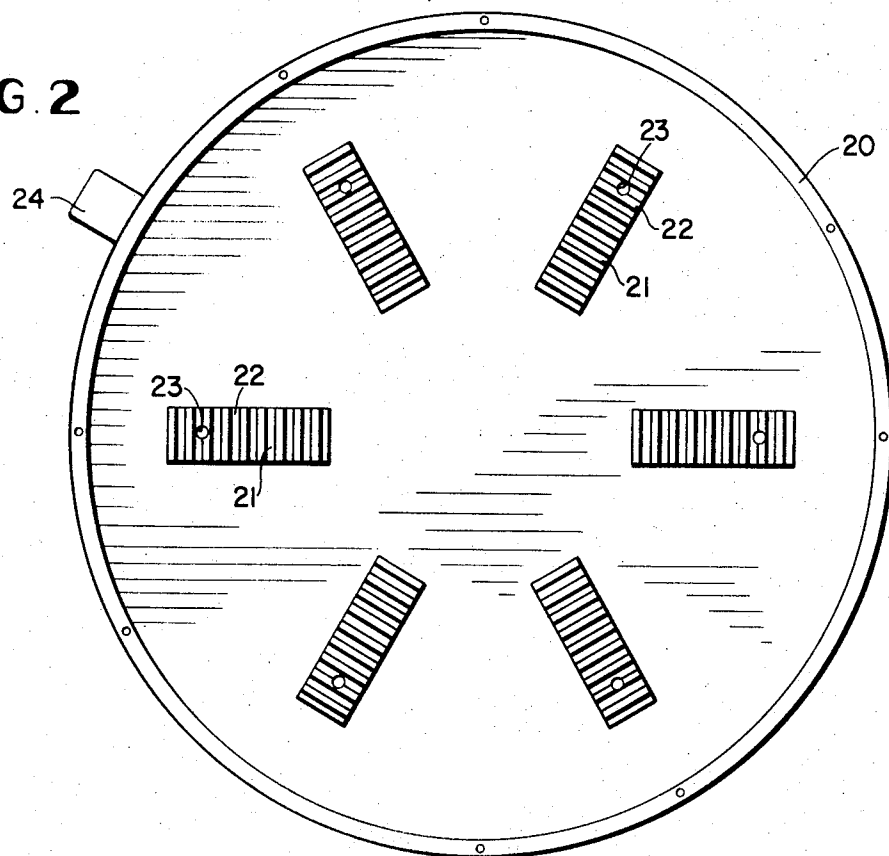
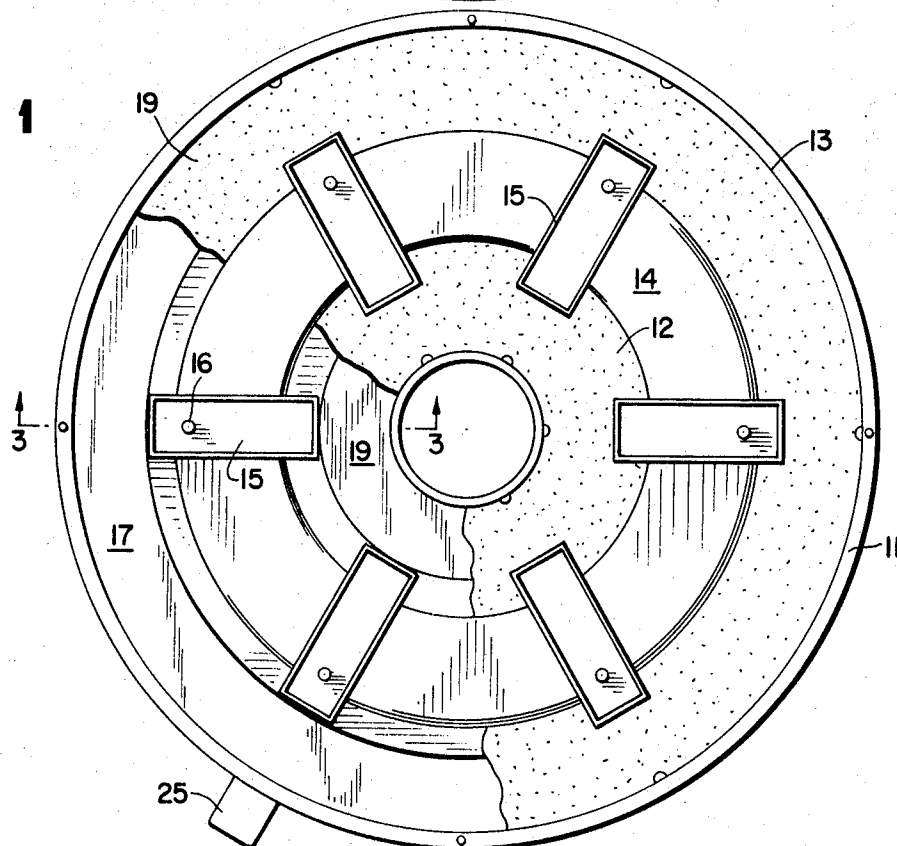


FIG. 1



2 Sheets-Sheet 2

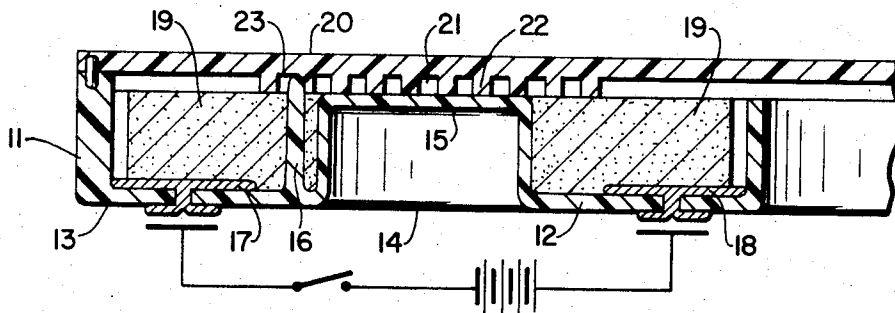


FIG. 3

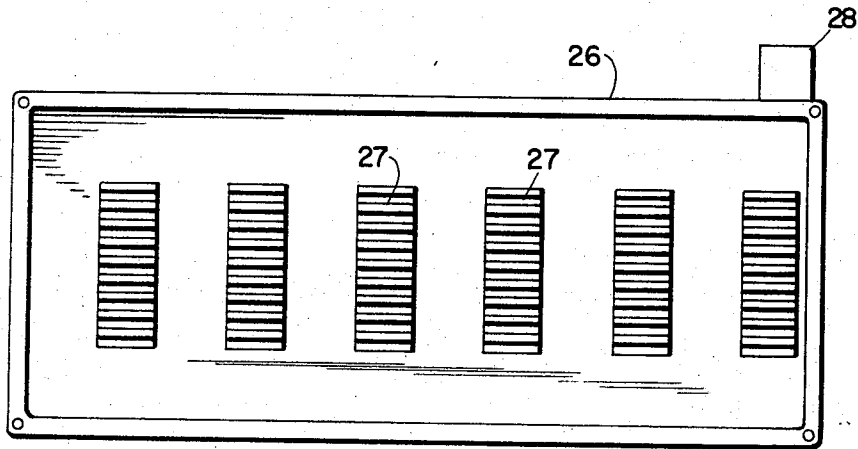


FIG. 4

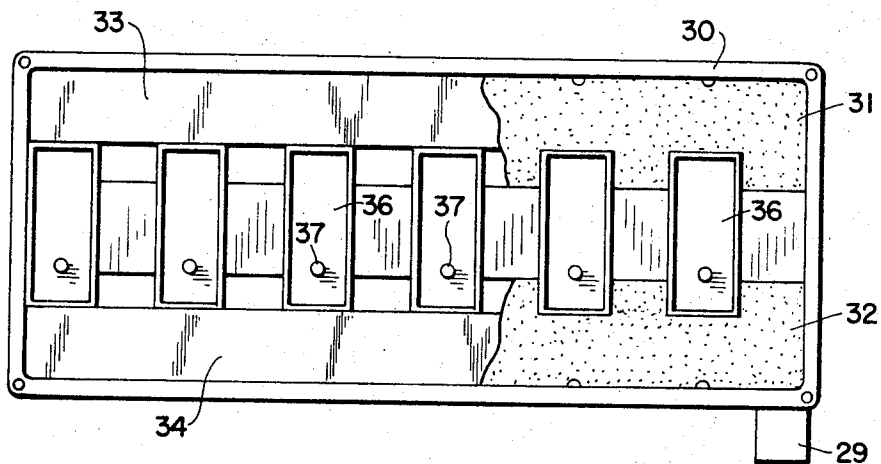


FIG. 5

ELECTROPHORESIS KIT AND SYSTEM

BACKGROUND OF THE INVENTION

Electrophoresis apparatus for testing of fluids such as blood sera and similar fluids is well known and a wide variety of electrophoresis kits and systems are known in the prior art. Similar disposable electrophoresis apparatus wherein the entire unit may be incinerated or otherwise disposed of after use is known, for example, U.S. Pat. No. 3,407,133. While such systems are generally effective, they require skilled personnel in order to assure reproducibility and generally require much skill in the assembly and preparation of the test samples for use.

SUMMARY OF THE INVENTION

Electrophoresis is the migration of particles under the influence of an electrical current. The electrical charges produce ionization, ion absorption, and ion dissolution. These changes influence the distribution of ions in the support media. Ions of opposite charge are attracted to each other. Ions of like charge are repelled, both lead to the formation of a double layer (made up of charged surface and neutralizing excess of counter ions in the support media).

When an electrical field is applied over the support media, a force is exerted on the electrical double layer. The charged surface tends to move in one direction, while ions in the mobile part of the double layer, show a net migration in the opposite direction, causing it to flow.

Four electrokinetic phenomena are:

1. Electrophoresis - the movement of charged particles relative to a stationary liquid layer by an applied electrical field.

2. Electroosmosis - the movement of liquid relative to a stationary charged surface by an applied electrical field.

3. Streaming potential - the electrical field created where liquid is made to flow along a stationary surface. (The opposite of electroosmosis).

4. Sedimentation potential - the electrical field created when charged particles move relative to a stationary liquid. (The opposite of electrophoresis).

In alkaline pH protons (H^+) will be dissociated and proteins lose positive charges (NH_3^+) and become more negatively charged. In an electrical field, it will move to the positive pole (anode) for every protein there is a pH at which the positive and negative charges are equal. At this pH protein will not move. This is the isoelectric point (IP) of a protein.

Application of these facts can be used to design methods for separating particles in an electrical field making use of optimum parameters of (buffer) pH — support media and location of the sample wells, the thickness of the agar, the configuration and location of sample wells, and the potency, species, and specificity of antisera as well as endosmotic flow. Endosmotic flow is the migration of water in the opposite direction of the protein ions.

Endosmosis generates water flow and may move sluggish proteins, for example, gamma globulin towards the cathode. This can be exploited to demonstrate Australian antigen (HAA) by the method of Immuno-electro-osmophoresis.

Another technique is the solution of specific antibody in the electrolyte buffered gel, and to electro-

phoretically migrate the soluble antigen into the agar producing an antigen antibody complex (precipitin reaction) which is visible as an opaque white band. This is called electro-immuno diffusion.

A variation is to place the soluble antigen in the agar and migrate the antibody with reactions as above. Either method can be used to quantitate the antigen or antibody by plotting curves with the use of standard control.

Some of the material tested can be highly infectious and dangerous viruses can migrate into the buffer-electrode vessels creating a health hazard to the user. In this method a number of novel effects of the design are achieved.

1. The entire system is self contained and disposable.

2. The user need only apply the test sample and connect the power supply.

3. After use the test is read and may be autoclaved or burned to destroy dangerous test samples.

4. Different antigen antibody reactions may be run simultaneously with the same test sample, for example, anti-tetanus, anti-diphtheria, anti-botulism.

In a typical electrophoresis unit, the unit is designed for a large variety of test procedures, therefore, the base unit consists of; electrodes, electrode vessels, container lid (to prevent evaporation); support bridge, support media, buffer supply and wicks.

The unit of the present invention is economical, permits the use of prepreparation antigen or antibody, permits simultaneous testing of different test samples, and the unit may be used for many test parameters as follows:

a. Immuno-electroendosmophoresis.

b. Electro immuno diffusion.

c. Standard electrophoresis defined for a given test

d. Immuno diffusion

e. Multi-specific antigen antibody test run simultaneously on a single test surface.

f. Discontinuous buffer systems where:

1. There is a variation of ionic strength of the buffer and buffer-wick package.

2. There is a variation of the type of buffer, for example, in the wick-buffer package, barbital buffer, and tris EDTA borate buffer in gel.

3. Where there is a variation of pH in the gel and the buffer-wick package.

4. Where gel thickness and well-configurations are ideally located to provide for optimum test performance.

Among the test parameters which may be assayed are: Tetanus toxin; Diphtheria toxin; Rubella virus; Mumps virus; Penicillin sensitivity; H.A.A.; Polyvalent botulism; Polyvalent Shigella toxin; Polyvalent salmonella; Staphylococcus enterotoxin; Poliomyelitis virus; Echo virus; Coxsackie virus; Coxsackie A. and B. virus, and 150 other species; Insect venom; Reptilian venom; Rickettsia; Encephalitis; Atoperc; Quantitation of hereditary disphenemias; Psitt. LG.V. Common Antigen; Tissue specific antibodies, as Hashimoto's thyroid disease; Transplant antibodies; lymphocyte antibodies and Lupus erythematosus.

DESCRIPTION OF THE DRAWINGS

FIG. 1 is a top plan view of a base unit of the electrophoresis apparatus of the present invention.

FIG. 2 is a bottom plan view of the lid for use with the base unit of FIG. 1.

FIG. 3 is a cross-section taken along lines 3—3 of FIG. 2 showing the assembled unit ready for use in electro-phoretic analysis, showing the electrical connections in schematic form.

FIG. 4 is a bottom plan view of a lid of an alternatively embodiment of the electrophoresis apparatus.

FIG. 5 is a top plan view of the base unit of an electrophoresis apparatus usable with the lid of FIG. 4.

DETAILED DESCRIPTION OF THE INVENTION

Referring to FIGS. 1, 2 and 3, the base unit 11, preferably molded of expendable plastic material or the like, is provided with concentric moat or buffer chambers 12 and 13 respectively. The annular area 14 between the concentric moats is bridged by a plurality of electrophoretic bridges 15 integrally formed with the base unit 11. Within each bridge 15 an upwardly extending pin 16 is provided. The base unit 11 is provided with electrodes 17 and 18 within the buffer chambers 12 and 13 which contain a conductive buffer material 19. Such buffer material 19 is in electrical contact with the electrodes 17 and 18 and may consist of agar or similar gel, or fibrous or sponge-like materials soaked in a suitable buffer solution.

A registration tab 25 is provided on the base unit 11, the purposes of which will be described hereinafter.

The lid 20, preferably molded of transparent plastic material, is provided with a plurality of wells 21, corresponding to the support bridges 16 of the base unit 11. Such wells 21 are preferably provided with a plurality of transverse elements 22 integrally molded to the lid 20. An aperture 23 is provided in each of the wells for mating with the pins 16 in the base unit 11 for the purposes to be described hereinafter. A registration tab 24 is provided on the lid 20.

In operation, the bridges 16 of the base unit 11 with the lid 20 removed, are filled with a gel of antibody in a suitable buffer material. Alternatively, the antibody gel may be supplied with the test unit and the entire unit sealed for shipment and storage. The antibody test material is poured into the support bridges 15, preferably with an overfill to form a gel bridge. The lid 20 is then positioned on the base unit 11 such that the wells 21 are in registration with the support bridges 15 of the base unit 11 and the lid pushed firmly into position. The lid 20 is then removed from the base unit 11, thereby transferring the gelled antibody from the base unit 11 to the lid 20. It will be apparent that a hole or well will be left in the antibody gel by virtue of the pin 16 and the base and the mating aperture 23 in the lid. The test sample material is then deposited into such wells in the antibody gel bridge, as by a pipette or the like. The lid 20 containing the test samples and antibody materials is then replaced over the base 11 with the registration tabs 24 and 25 adjacent each other. It will be apparent that the test wells 21 containing the samples and antibody gel will then be in electrical contact with the buffer materials 18 and 19 as shown in FIG. 3 and will be remote from the storage wells 15 of the base unit 11. The unit is then ready for connection to an electrical power source for the desired test to be performed. The results are read by conventional analytical procedures, such as colorimetry and may be determined both qualitatively and quantitatively. It is apparent that the presence of more than one antibody

may be detected in the course of a simple test. The use of test wells of predetermined size assures uniformity of thickness of the test material, and hence, reproducibility.

FIGS. 4 and 5 show an alternative form of the invention employing a rectangular test kit. The lid member is shown at 26 provided with the test wells 27 as described in connection with the circular modifications described above. The lid 26 is provided with a registration tab 28 for registration with a corresponding tab 29 on the base unit 30. The base unit 30 has spaced-apart buffer chambers 31 and 32 containing suitable buffer material and is provided with electrodes 33 and 34. A corresponding number of support bridges 36 are provided, each having an upstanding pin element 37 for producing the chamber well. Mating wells or apertures are provided in the support bridges 27 of the lid 26, not shown. The manner of operation is as described in connection with FIGS. 1—3, namely, transferring the antibody gel from the base to the lid and thereafter positioning the lid with the tabs 28 and 29 in registration to complete the electrical circuit.

It is apparent that the geometrical configuration of the unit may be varied to include other variations other than the circular and rectangular configurations shown and described.

I claim:

1. An electrophoresis test kit comprising a base member having a pair of remotely spaced electrodes therein and a pair of chambers associated therewith for receiving a buffer, a plurality of storage wells extending between said buffer chambers for receiving test samples, a lid conforming to said base chamber and having a plurality of electrophoretic support bridges corresponding in one position to said storage wells, registration means carried by said base member and said lid member for positioning said support bridges in electrical contact with said buffer chambers in a second position, said lid member sealing said base chamber.

2. The test kit set forth in claim 1 wherein each of said storage wells is provided with a pin member for creating a sample well in the test sample.

3. The test kit set forth in claim 1 wherein said base member and said lid member are circular and said buffer chambers and associated electrodes comprise concentric areas within said base chamber.

4. The test kit set forth in claim 1 wherein said base member and said lid member are of rectangular configuration and said buffer chambers are positioned on opposite sides of said base chamber.

5. The test kit set forth in claim 1 wherein said registration means comprises tabs on each said base member and said lid member.

6. The test kit set forth in claim 1 wherein said base member and lid member assembly is composed of combustible plastic material.

7. The method of preparing a test kit unit for electrophoretic analysis comprising pouring a test sample into a storage well positioned between spaced buffer chambers in a base member, transferring said sample to a lid member mating with said base member and repositioning said lid member containing said sample on said base member in electrical contact with said buffer chambers.

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