

[54] **ELECTROPHORESIS DEVICE**

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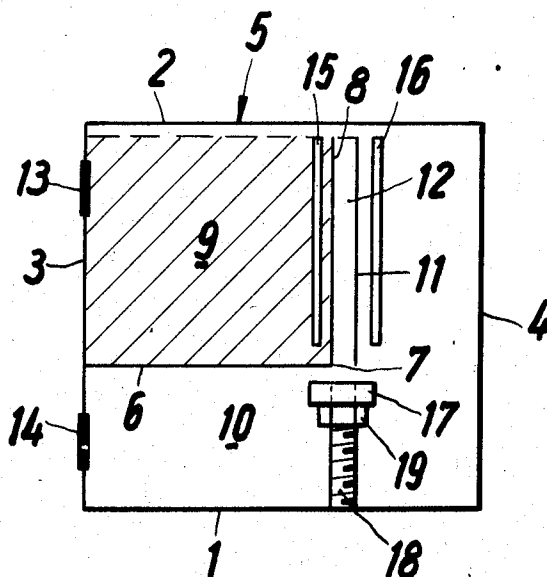
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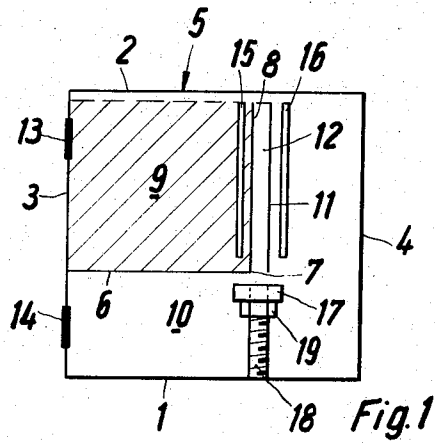
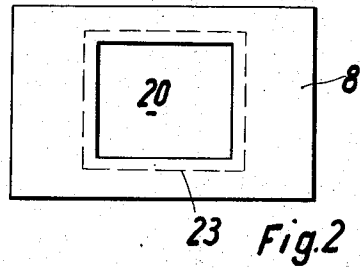
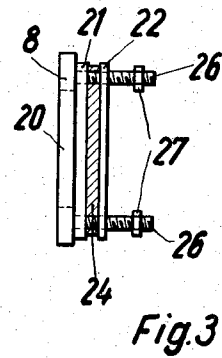
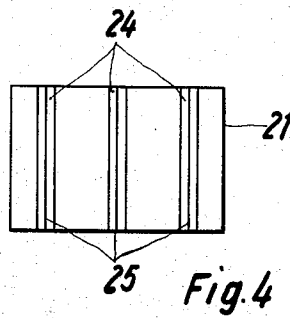
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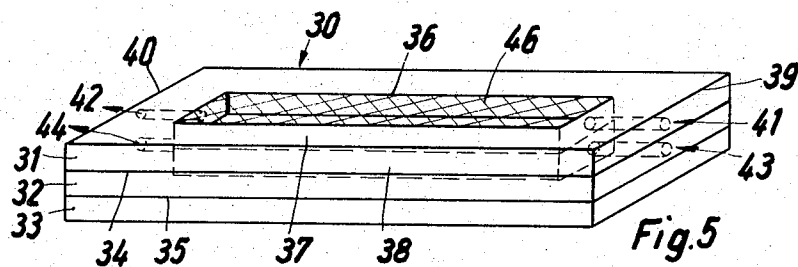
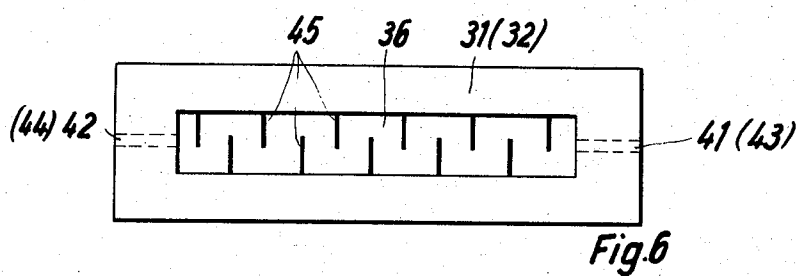
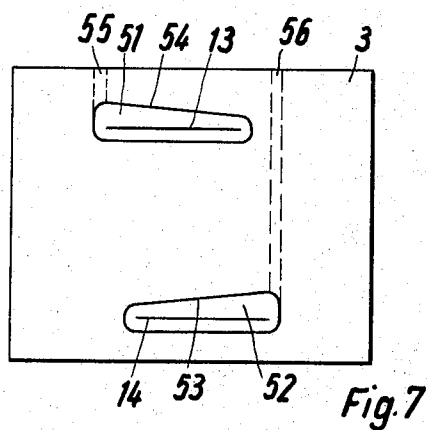
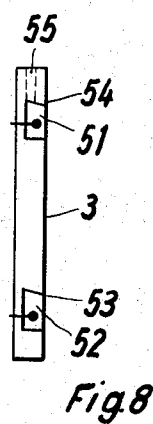
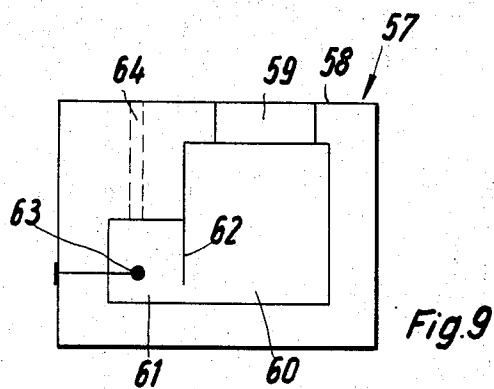
ABSTRACT

An electrophoresis device having a separating chamber, and two buffer containers one of which is attached to one end of said separating chamber and the other one is connected to the other end, and an electrode in each in said buffer containers, said device essentially having a box-shaped housing and a box area which is divided up into two partial areas serving as buffer containers by means of an essentially horizontal intermediate bottom of lesser depth than the box together with an intermediate wall which extends essentially vertically from the intermediate bottom.

36 Claims, 9 Drawing Figures







ELECTROPHORESIS DEVICE

BACKGROUND OF THE INVENTION

This invention relates to an electrophoresis device with two buffer working tanks one of which is connected to one end of a separating chamber and the other one of which is connected to the other end of a separating chamber, and with one electrode each in the buffer tanks.

Prior art electrophoresis devices are built in such a way that a vertical separating chamber follows a first box-shaped container for the buffer solution. A second buffer container, which can also be box-shaped, is located on the vertical separating chamber. The production of these components, which are preferably circular in shape, is expensive. Furthermore, because of the upper buffer tank, the overall design of the device is very unstable so that the danger of damage is quite great. In these prior art devices good accessibility to the upper and lower chamber limitation, which is necessary in various electrophoresis methods, is not present.

Also prior art preparative electrophoresis devices are essentially constructed of axisymmetric parts they consequently have a cylindrically shaped separating chamber, bowl-shaped buffer containers and an elution chamber which is also circularly shaped in the cross section. The parts of the prior art devices are balanced to one another in such a way that preparative electrophoreses can be carried out. For the carrying out of analytical tasks, however, these devices are not suitable.

The yields of known preparative electrophoresis devices are also relatively low even in more modern devices. Also the selectivity which is achieved with known devices is still not optimum so that substances with practically the same electrophoretic rate of travel cannot be securely separated.

OBJECTS OF THE INVENTION

One object of this invention to create an electrophoresis device in which these prior art disadvantages are not present, in which therefore a compact form of construction is available, simple parts can be used, the danger of damage from a low degree of stability and difficult construction is excluded and a good accessibility of the upper and lower separating chambers is provided for. It is a further object of this invention to provide a simple and efficient cooling of the separating chambers, as well as a hydrostatic pressure equalization in the two buffer containers without special means, such as lifting pipes or similar, which are required for this purpose.

It is a still further object of this invention to eliminate the disadvantages of known preparative electrophoresis devices so that a device is created with which higher yields and greater selectivities can be achieved.

A still further object of the invention is to provide a device which makes it possible, without problems, to carry out not only preparative electrophoreses but also analytical electrophoreses.

A yet still further object of this invention is to permit an operation according to the most varied known electrophoresis methods so that the range of application of the proposed device is expanded.

SUMMARY OF THE INVENTION

The objects of this invention are achieved by means of an essentially box-shaped design of the electropho-

resis device. The device of this invention is provided with a box-shaped area by means of an essentially horizontal intermediate bottom which is of smaller depth than the box, can be split up together with an intermediate wall which goes out from the intermediate bottom and extends essentially vertically to it into two partial areas serving as a buffer container. It is particularly advantageous to have the intermediate wall extending vertically upwards into the box because when both partial areas are open upwardly the buffer fluids can be brought practically to the same level whereby the hydrostatic pressure equalization is brought about in the simplest manner. The design according to this invention permits an arrangement of the separating chamber essentially in the plane of the intermediate wall. The thickness of the separating chamber may be varied in a particularly simple manner when parallel to the intermediate wall and in the region of the greater angle formed by the intermediate bottom and the intermediate wall. A plate can be applied which can be adjustable in its distance and which, together with the intermediate wall, forms the separating chamber.

The accessibility to the buffer containers and the separating chamber is particularly simplified in that the front side wall of the box-shaped housing and the lid of the housing are removable. According to a further embodiment of this invention the device can be used for the application of the most varied electrophoresis methods, for example the continuous electrophoresis method, the discontinuous electrophoresis method, the isoelectric focusing and the iso-tachoelectrophoresis when means for pressing in inserts (such as elution insert, isoelectric focusing insert, and the like) in the partial space located below the separating chamber to the lower limitation of the separating chamber. These pressing means are achieved preferably by means of a pressing device which operates in the manner of a lifting stand.

This invention has a further advantage by permitting the cooling means which are provided for cooling the separating chamber to engage the buffer solutions in both buffer containers. It then suffices for the buffer solutions to be cooled in the region of the separating chamber since the buffer solutions themselves can act as a cooling medium. Within the framework of the invention it is advantageous to arrange a laminar cooling plate or hose parallel to the separating chamber walls in order to be able to achieve in this way the greatest possible cooling area.

For this purpose the invention is based on the fact that with the known construction principle of preparative electrophoresis devices neither the selectivity nor the yields can be increased to any considerable degree. This is due to the fact that in the amplification of the separating chamber diameter for increasing the yields the cooling of the carrier substance becomes problematical and also the elution of greater circular-shaped or circular-ring shaped cross sections can no longer occur in a satisfactory manner. Inadequate cooling of the carrier substance in the separating chamber results namely in the fact that a temperature gradient occurs between the inside, the center of the cylindrical separating chamber and its cooled outside wall.

Since the rate of travel in the separating chamber depends upon the temperature and the flushing occurs with different flow velocities it is not possible to in-

crease the quality of the electrophoresis devices when maintaining the known principal design.

According to this invention the separating chamber has an essential rectangular cross section and the elution chamber is attachable to one end of the separating chamber. By means of the steps according to this invention it becomes possible to cool large separating chamber volumes effectively owing to their essentially greater surfaces and also to carry out the flushing on the elution side of the separating chamber with uniform flow velocities. With this, however, the preconditions for a considerable improvement in quality of the known preparative electrophoresis devices are given. The rectangular separating chamber construction can be considerably simplified and made more economical in comparison to most of the blown cylindrical separating chambers by means of the utilization of simple glass plates. The rectangular separating chamber design also permits a compact design of the entire device including the buffer container. In addition, by removing the elution chamber the device can easily be used immediately for analytical electrophoresis. An interchanging of the elution chamber against other attachments to be placed on the underside of the separating chamber makes it possible to operate according to various electrophoresis methods and simplifies the work in that, for example, by means of an attachment formed as a fastener, the separating chamber can be filled with carrier substance without having to be removed from the device and the electrophoresis can immediately be started in the device after the polymerization and the removal of the fastener attachment without the disturbing transport of the carrier substance and without any difficulties of installation into the device.

Within the framework of this invention it is appropriate to construct the elution chamber in such a way that its form essentially corresponds to the lower separating chamber opening, i.e., so that the elution chamber is rectangular. It is advantageous to construct the elution chamber out of a plate which has a recess which corresponds to the lower separating chamber opening so that the lower separating chamber limitation should be tightly attachable to the upper side of the plate over the recess. When the recess corresponds to the diameter of the plate, the plate is slit, and means are provided for closing the lower side of the slit opposite the separating chamber. Connections for the feeding and removal of the elution fluid are provided in the region of the small sides of the recess or of the slit. This procedure permits uniform flow velocity over the entire cross section of the chamber, therefore uniform washing out of the electrophoretically separated substances and consequently a high degree of selectivity on the part of the device. The feeding and removal of the elution fluid on the narrow sides of the rectangular elution chamber also causes a high concentration of the eluted substance.

The slit may be provided with a semipermeable membrane as a means for closing the separating chamber of the plate. This membrane is preferably squeezed, by means of a second plate, between the second plate and the first plate. In this manner the second plate can have a corresponding recess as in the first plate. Preferably, in the region of the narrow sides of the recess of the second plate, attachments are provided for the feeding and removal of a buffer fluid which is particularly highly concentrated within the framework of the inven-

tion. The recess in the second plate can also be formed as a slit, whereby the slit on the side opposite that of the elution chamber is closed by means of a second membrane. In a similar manner the second membrane is also squeezed, by means of a third plate which is also slit, between that plate and the second plate. There is thus obtained a plate stack composed of three plates with a slit which passes through and which corresponds in size to the separating chamber outlet, whereby a continuous semipermeable membrane is located between the plates. Such a plate stack is produced in the simplest manner and combined with the elution chamber according to the invention and also inserted into the device. For this purpose, within the framework of the invention, means are provided for the compact pressing together of the stack and for the tight pressing of the stack against the bottom side of the separating chamber.

In the framework of the device, the elution chamber and the means for bearing pressure upon the elution chamber are preferably provided for in the lower located region serving as the buffer container in the case of a compact box-shaped construction of the electrophoresis devices in which, by means of an intermediate bottom and an intermediate wall, the housing is divided up into two areas forming the buffer container between which regions the separating chamber is located. The means for pressing against the elution chamber has a lifting stand which is preferably formed of threaded bolts. These bolts are located on the bottom of the lower-situated region, lie vertically and the stand plate can move to and fro on these bolts by means of threaded nuts. In this manner, the elution chamber can be pressed against the lower side of the separating chamber.

It is particularly advantageous, for the cooling of the separating chamber which is rectangular in cross section, to arrange the cooling agents parallel to the large sides of the separating chamber which can be composed, for example, of a cooling house proceeding in a plane with immersion frames.

In the case of the prior art electrophoresis devices, in the upper and lower buffer container, locking elements are provided for a frame in which an electrode is housed. These frames are placed immediately above and below the separating chamber. The placement below the separating chamber has the disadvantage that gas bubbles rising up on the electrode reach the lower side of the separating chamber and in this way cause a disruption in the electrophoresis. In order to eliminate this disadvantage it is necessary to provide, between the lower side of the separating chamber and the electrode in the buffer container, means which prevent the gas bubbles from reaching the lower side of the separating chamber. This is achieved by holding in another frame a deflecting means such as an absorptive paper which is shaped into an acute-angled groove in such a manner that the point points downwards and the upper edges of the groove lie laterally outside of the lower limit of the separating chamber. Gas bubbles rising up are pushed laterally upwards from the lower side of the groove and cannot reach the separating chamber.

It can be seen that in this kind of construction the structure and assembly of the device are complicated and that there is the danger that the absorptive paper

is mistakenly not inserted and that impurities can reach the buffer solution through the paper.

In accordance with a further embodiment of this invention at least the buffer container, which is in contact with the lower end of the separating chamber, has a recess in the inner container wall, preferably the container backwall, for housing the electrode.

This feature prevents the rising gas bubbles from reaching the bottom side of the separating chamber. There is furthermore achieved that no special frame and no special holding means in the buffer container are needed for the electrode so that the construction and erection of this part of the electrophoresis device are considerably simplified.

Prior art devices also have a considerable disadvantage in that the separating chamber is not easily accessible from above when the upper electrode attachment is placed in the buffer container. The subsequent insertion of the electrode attachment into the buffer container is disadvantageous, however, since the buffer fluid is placed into motion through the insertion and thus the fluid substances which are piled up on the upper side of the separating chamber can easily be mixed. Bearing this situation in mind, this invention makes it possible for the accessibility to the upper limit of the separating chamber to be unhampered and also makes it possible to do away with a subsequent insertion of the electrodes into the buffer containers.

This is achieved by also providing a recess for the electrode in the buffer container which comes into contact with the upper end of the separating chamber. This thereby provides the advantage of good accessibility and the elimination of the necessity of having to insert the upper electrode subsequently.

Examples of construction of the invention are described in what follows based on the drawing.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 shows a side view of the electrophoresis device of this invention in which the side wall has been removed;

FIG. 2 shows an alternate form of a separating chamber wall for use with this invention;

FIG. 3 shows the assembly of a separating chamber for use with this invention;

FIG. 4 shows a special design of a separating chamber part for use in this invention;

FIG. 5 shows the elution attachment of the device of this invention;

FIG. 6 shows a special form of an elution chamber part for use with this invention;

FIG. 7 is a view of the inside of the backwall of the electrophoresis device of this invention;

FIG. 8 is a cross section view through the backwall of the device according to FIG. 7, and

FIG. 9 is a cross section view through an addition for working according to the method of iso-electric focusing.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

In FIG. 1 there is shown an electrophoresis device having a bottom 1, lid 2, backwall 3 and front side wall 4 of a box-shaped housing 5 with a side wall removed. Inside of the housing 5 an intermediate bottom 6 has been inserted into about one-third of the overall height of the housing. It extends beyond half of the depth of

the fronting to the front side wall 4. An intermediate wall 8 extends vertically upwards from its free end 7. The intermediate bottom 6 and the intermediate wall 8 provide the boundary, together with the backwall 3 and the side wall, not shown, of the housing 5, for a buffer container 9, and bound it tightly against the remaining area which forms a second buffer container 10. Parallel to the intermediate wall 8 in the direction of the front side wall 4 of the housing 5 with a distance, there is provided a plate 11 which forms the separating chamber 12 of the electrophoresis device together with the intermediate wall 8. The plate 11 is somewhat higher than the intermediate wall 8 so that the hatched-line-indicated buffer solution in the buffer container 9 can reach the upper end of the separating chamber. The lower end of the separating chamber 12 is surrounded by the fluid which is located in the buffer container 10. An electrode 13 or 14 is provided in the backwall 3 in the region of the two buffer containers 9 and 10.

By means of the described construction of the device, in which according to the invention the two buffer containers 9 and 10 are located essentially laterally, preferably on both sides of the separating chamber 12, a pressed construction form is given, only simple parts are used, and there is no danger that the device will be exposed to the risk of damage because of inadequate stability or complicated construction. When the lid 2 is removed, the device is easily accessible from above so that the extra shifting or layering of substances, which is often to be undertaken with great sensitivity, is easily possible. Also the introduction of the buffer solution from above into both buffer containers is easily possible. Complicated and expensive pressure equalization devices are therefore not necessary.

The front side wall 4 is detachable from the two side walls. The detachability of the front side wall provides good access to the lower end of the separating chamber 12 so that it can be filled with carrier substance without having to be removed from the device, when the easily accessibly lower separating chamber opening is closed. The preparation of the carrier substance can therefore take place inside of the device, whereas in the case of previously known devices this had to be done outside of the device. The loaded separating chamber can also remain in the device during the time of the polymerization of the carrier substance.

The described construction also makes it possible to use the buffer fluid in the containers 9 and 10 as a cooling agent, in which a cooling device, for example cooling plates or cooling hoses 15, 16 are placed parallel to the surfaces 8 and 11 limiting the separating chamber 12 at a distance from the side walls of the housing. This arrangement is particularly simple in the utilization of an immersion frame which can be introduced into the device from above.

Advantageously, the intermediate wall 8 forms at least partially a part of the separating chamber 12. Thus the erection of the device becomes particularly easy.

The plate 11, which is placed parallel to the intermediate wall 8 and in the region of the larger angle which is formed by the intermediate bottom 6 and the intermediate wall 8, and which, together with the intermediate wall, forms the separating chamber 12, is adjustable with respect to the intermediate wall in which corresponding means, such as studs, are provided on the in-

intermediate wall and return-leading elements, such as boreholes, which are suitable for this are provided on the plate. These have not been represented for purposes of simplicity. In this manner there is achieved in the simplest manner the variation of thickness of the carrier substance in the separating chamber 12 according to the particular requirements.

The electrodes 13 and 14 which are provided on the inside housing backwall 3, preferably in a recess for each one, are provided with connecting elements (not shown) which project through the backwall of the housing. As will be described in greater detail later on, the recesses in which the electrodes are placed are connected to the outside air preferably by means of a canal which runs upwards in the housing backwall so that air bubbles rising on the electrode can escape. The removability of the lid 2 and the front sidewall 4 of the housing 5 is solved by any known manner. The tightness of the removable parts with respect to the rest of the housing is also achieved by any known means.

The described arrangement makes it possible to provide means for pressing attachments onto the lower limit of the separating chamber in the partial space located below the separating chamber 12. These means can be used to close the bottom side of the separating chamber by means of a special attachment in order to make possible the already-described introduction of the carrier substance inside of the device. Preferably, the pressing means are shaped in the manner of a lifting stand 17 which can be easily activated from the front with the front side wall 4 removed. A motorized height adjustability of the lifting stand is possible, but it is hardly necessary in practice since the manual activation, for example, by means of one or more threaded spindles 18 and spindle nuts 19 can be carried out with few movements of the hand.

The cooling of the separating chamber 12 is particularly easy and efficient, in the case of this invention, in comparison to the already known preparative electrophoresis devices which operate basically with cylinder-shaped separating chambers. Because of the large side surfaces of the separating chamber which is rectangular in shape in the cross section, provision is made for a good heat exchange and it is achieved in that no significant temperature gradient occurs in the separating chamber. It is thus provided that inside of the separating chamber no temperature-caused differences in rate of travel occur so that the individual bands do not undergo any distortions. It thus also becomes possible to cleanly separate from one another even the bands which are located close to one another.

Within the framework of the invention another form of the intermediate wall 8 and of the separating chamber 12 is also possible. This possibility is described in further detail based on FIG. 2.

The intermediate wall 8 is provided with a window 20, which is closed or closeable from the front side 4 of the box-shaped housing 5 through the attachment of a plate 21 and which, together with a plate 22 which corresponds to the plate 11, forms the separating chamber. Between the plate 21 which closes the window 20 and the window frame (intermediate wall 8), packing means are provided (shown by dashes in FIG. 2) so that no buffer fluid can enter from the container 9 into the remaining area of the housing 5. Preferably the side walls of the housing 5 are not used as side limit of the separating chamber 12, but between the two

plates 21 and 22 which form the separating chamber there are provided laterally limiting packing means which may be composed of glass rods 24. The glass rods 24 are quadrangular or rectangular shaped in the cross section (see FIG. 4) and carry compressible packing means 25 running in corresponding nuts on their two sides opposite the two plates 21 and 22. When the two plates, which are also preferably made of glass, are pressed together, then the separating chamber 12 is practically of the same material on all of its limiting walls, so that, for example, expansion phenomena or other disturbances do not cause the carrier substance to detach from the chamber walls and creeping currents to occur which could disrupt the electrophoresis. As already shown in FIG. 1, the plate 8 or 12 which closes the buffer container 9 towards the front is arranged in such a way that a distance is present between its upper edge and the housing lid 2, whereas the second plate 11 or 22 is closer to the housing lid 2 than the upper edge of the previously mentioned plate 8 or 21. This thus causes the buffer fluid in the buffer container 9 to only be able to reach the upper end of the separating chamber 12 but not the other buffer container 10.

It is also possible within the scope of this invention to make the two plates which form the separating chamber 12 of the same height and to see to it that the buffer container 9, limited by the housing backwall 3 and the plate 21 which closes the window 20, together with the upper opening of the separating chamber 12 is made tight in comparison with the other buffer container 10 through the housing lid 2. Naturally, instead of the housing lid an intermediate lid can also be used.

It can be seen from FIG. 3 that on the window frames (intermediate wall 8) there are provided threaded bolts 26 projecting towards the front side 4 of the housing 5 and onto which there are placed nuts 27. When these bolts are fastened securely the outside plate 22 is pressed against the packing means 24, 25 so that the plate 21 which closes the window 20 is also pressed against the window frame. There thus occurs a tight separating chamber in the housing 5 having a thickness which corresponds to the thickness of the glass rods 24. The thickness of the separating chamber can be adjusted in a simple manner by varying the thickness of the glass rods. For use in connection with this invention it may be advantageous in analytical devices to provide for a number of packing means (24, 25) running parallel to the lateral packings in order to have several separating chamber stretches available under otherwise equal iontophoresis conditions.

With the already-mentioned lifting stand 17 it is also possible to attach to the bottom side of the separating chamber 12 a so-called elution attachment which is described in detail below. The yields in the device according to this invention are considerably greater than those of known devices which leads to the result that a considerably greater surface can be elutriated, whereby consequently the amounts of elution fluid which are used can be kept small. The elution takes place from the narrow side wall of the separating chamber 12 or the elution attachment and on the other narrow side there occurs the removal of the elution fluid. The yields can be increased to a still greater extent by means of meander-shaped path. Not only is the yield in the device according to this invention considerably increased in comparison to that of known electrophoresis devices but the selectivity of bands closely following one an-

other is also increased. This is also a result of the design according to this invention since, in comparison with devices with cylindrical separating chambers, a uniform elution current can be provided. In the case of cylindrical separating chambers the elution does not take place with uniform rate of flow since the feeding and removal of the elution current through a circular shaped or circular ring shaped cross section is not possible with prior art means and consequently irregular flushings occur which have an adverse effect upon the selectivity of the separation. In the design example which is represented, the plate of the lifting stand 17 is composed of a frame into which the elution attachment 30 shown in FIG. 5 can be inserted. The frame is rectangular and extends over the entire separating chamber cross section. It is open from below so that the buffer fluid which is present in the container 10 can reach the elution attachment from below. With the aid of the lifting stand arrangement, the attachment 30 can be pressed tightly against the lower separating chamber opening.

The elution attachment 30 is composed of a first plate 31, a second plate 32, a third plate 33, a first semipermeable membrane 34 and a second semipermeable membrane 35. The plates 31, 32, 33 are rectangular and are provided in the center of the plates with a slit 36 which goes through the thickness of the plate and the dimensions of which correspond to those of the bottom side of the separating chamber. The slit 36 need not extend over the entire length of the plate.

On the narrow sides 39, 40 of the plates 31 and 32 there are leads 41, 42, 43, 44 to the slit ends 36. At 41 elution fluid is fed to the elution chamber 37 formed in the plate 31 and at 43 concentrated buffer solution is fed and removed at 44. The concentrated buffer solution serves the purpose of greatly diminishing the electrical field intensity and in this way achieves a braking effect for the substances which are to be elutriated and results in an increase in the effective flushing. An absorption of the proteins which are to be elutriated on the membrane 34 and their electrophoresis through the membrane is thus effectively prevented. By means of the flowing of the highly concentrated buffer fluid through the buffer chamber 38 formed in the plate 32 provision is made to keep the concentration of the buffer fluid uniform at the membrane 34.

The plate 31 in the vicinity of its upper side above the slit 36 has a net 46 which serves for supporting the gel which is located in the separating chamber 12. Baffle rods 45 may be provided diagonally to the flow direction in the chambers 37 and 38 interchangeably from both sides of the slit 36 in order to force a meander-shaped flow. Thus, at a relatively high rate of flow only a slight drive is necessary and the obtention of highly-concentrated eluates is possible. The baffle rods 45 also serve to keep the membranes 34 and 35 tight (see FIG. 6).

It can be seen from FIG. 5 that the plates 31 and 32 must possess a thickness which corresponds at least to the strength of the connections 41 to 44. Thereby there is realized a relatively large elution chamber. It is, however, worthwhile to keep the elution chamber as small as possible so that high elution speeds can be achieved with a low elution fluid weight rate of flow in order to provide for a good dissolution and quality of the device. According to a further development of the invention a reduction in the elution chamber volume is achieved by

providing the connections for the feeding and removal of the elution fluid on the ends of the separating chamber 12 which are adjacent to the elution chamber. The connections are achieved by means of boreholes (from outside of the separating chamber 12 to the elution chamber inside). In this way the plate 31 can be kept very thin and consequently the elution chamber volume can be kept very small so that the desired quality improvement (better dissolving power) is achieved.

FIG. 7 shows a view of the inside of the backwall 3. It can be seen that the electrodes 13 and 14, which are shaped in the form of a wire here, are housed in recesses 51, 52 which are milled in the backwall 3. The recesses are somewhat wedge-shaped so that their upper edges 53 and 54 rise upwards obliquely. Gas bubbles rising to the electrodes 13, 14 travel along these upper edges 53, 54 which run obliquely upwards and gather at the highest point. For diverting the existing gas, therefore, at the highest points of the wedge-shaped recesses 51, 52 a channel 55 or 56 is bored upwards through the backwall 3 through which the gas can escape into the open air. From FIG. 8, which shows a cross section through the backwall 3 it can be seen that the upper edges 53 and 54 are also designed obliquely in the direction of the outside of the backwall 3. In this way the recesses 51, 52 are given a defined highest position at which the gas collects and at which point the canals 55, 56 engage into the recesses. For purposes of simplicity only the canal 55 of the upper recess 51 is shown in FIG. 8.

The aforementioned type of arrangement and design of the electrodes 13, 14, as well as their housing into recesses 51, 52 in the backwall 3 is recommended, for example, in disc electrophoresis.

A further advantage of this invention is that the device can also be used for the method of iso-electric focusing in which particularly small buffer container chambers are used. It is possible with the aid of the lifting stand 17 to press a corresponding attachment 57 onto the lower end of the separating chamber 12 which only has a small volume and contains an electrode 63. In this way it is possible to use one and the same device for all known electrophoresis methods and for analytical and preparative purposes only by using individual attachments.

When using the method of iso-electric focusing the electrodes should not be far from the separating chamber so that the potential gradient lies essentially within the separating chamber which is filled with the carrier means. The attachment of the electrode to the lower separating chamber opening entails, however, the danger that the gas bubbles rising from the electrode can reach the underside of the separating chamber and disturb the electrophoresis.

This danger is eliminated by means of the iso-electric focusing attachment according to this invention. FIG. 9 shows sectional view through the attachment 57 which is essentially composed of a square-shaped container the length of which is at least the length of the lower opening of the separating chamber 12. At its upper side the container has the opening 59 which creates a connection of the lower opening of the separating chamber 12 and the inside 60 of the container. By means of a closeable opening (not shown) the container or the inside 60 of the container can be filled with buffer fluid. The attachment 57 has an adjacent chamber 61 towering above the lower separating cham-

ber limit laterally and which is partially delimited from the remaining part of the buffer container by means of a diaphragm 62 which is essentially vertical and ending over the bottom of the buffer container. The electrode 63 is housed in the adjacent chamber 61 and the gas bubbles which go out from it rise to the upper limit of the adjacent chamber 61 where they can escape through a canal 64. The opening 59 at the upper side 58 of the attachment 57 is surrounded by packing material which is arranged in such a way that it can be pressed against the lower limit of the separating chamber 12. FIG. 1 schematically shows how the pressing can occur with the aid of the lifting stand 17. For this purpose, there can be used vertically standing threaded bolts located at the housing bottom 1, therefore inside of the large buffer container 10 which is not filled during an iso-electric focusing procedure. The bolts can be moved back and forth on the threaded bolts with the aid of which the lifting stand 17 can move back and forth on the bolts.

In the case of iso-electric focusing a correspondingly smaller buffer container with electrode can be attached on the upper end of the separating chamber 12. The advantage of the small volume is thus obtained and also the danger of the gas bubble contact with the upper end of the separating chamber 12 does not occur. The elimination of the developed gas by means of a ventilation canal provided over the electrode in its adjacent chamber according to the invention is nevertheless useful so that no disturbing excess pressure occurs in the buffer container. The adjacent chamber can be kept very small, by the way, so that it is only insignificantly different from the volume of the iso-electric focusing attachment.

Obviously, the embodiments shown are exemplary only and a wide variety of embodiments may be devised without departing from the spirit and scope thereof.

What is claimed is:

1. A gel-electrophoresis device comprising a vertical separating chamber, the cross section of which is elongated and has a substantially rectangular shape, two buffer containers, one being in communication with the upper end of said separating chamber, elution means interconnected between the lower end of said separating chamber and the other buffer container, said elution means comprising an elongated elution chamber, means at each of the opposed ends of said elongated chamber for supplying and removing an elution fluid to and from said elution chamber, an electrode in each of said buffer containers, and means for connecting the electrodes to a source of direct current.

2. A device as claimed in claim 1 wherein said elution means further comprises a second elongated chamber beneath said elongated elution chamber for receiving a concentrated buffer fluid.

3. A device as claimed in claim 1 wherein the lower wall of said elongated elution chamber is composed of semipermeable membrane material.

4. A device as claimed in claim 2 wherein said second elongated chamber has an upper and a lower wall each composed of semipermeable membrane material, said lower wall separating said second chamber from said other buffer container and said upper wall constituting the lower wall of said elution chamber.

5. A device as claimed in claim 2 and further comprising means at each of the opposed ends of said sec-

ond elongated chamber for supplying and removing the concentrated buffer fluid.

6. A device as claimed in claim 1 wherein said elution means comprises a plate having a recess therein which constitutes said elongated elution chamber, the shape of said recess conforming with the shape of the lower end of said separating chamber and means for tightly securing the upper side of said plate to the lower end of said separating chamber.

7. A device as claimed in claim 6 wherein said recess extends through said plate and further comprising means for closing the lower side of said recess.

8. A device as claimed in claim 6 wherein said elution means further includes a second plate beneath said first mentioned plate and having a recess therein for receiving a concentrated buffer fluid and a semipermeable membrane firmly gripped between said first and second plates and constituting the lower wall of said elongated elution chamber.

9. A device as claimed in claim 8 wherein the recess of said second mentioned plate corresponds in shape to the recess of said first plate.

10. A device as claimed in claim 8 wherein said elution means further includes a third plate and a second semipermeable membrane gripped between said third plate and said second plate to form the lower wall of the recess in said second plate.

11. A device as claimed in claim 10 wherein the recess of said third plate corresponds in shape to the recess in said first plate.

12. A device as claimed in claim 6 wherein the means for supplying and removing an elution fluid comprises channels leading to and from the opposed ends of the recess in said plate.

13. A device as claimed in claim 8 and further comprising channel means in said second plate for supplying and removing the concentrated buffer fluid to and from the opposed ends of the recess in said second plate.

14. A device as claimed in claim 10 wherein said first, second and third plates are superposed with semipermeable membranes disposed therebetween, said recesses being in register and conforming with the shape of the lower end of said separating chamber and further comprising means for pressing the superposed plates together and for pressing the assembled plates against the bottom of said separating chamber.

15. A device as claimed in claim 1 and further comprising baffle rods disposed within said elongated elution chamber, arranged alternately on opposite sides thereof and extending diagonally to the direction of flow of the elution fluid passing therethrough.

16. A device as claimed in claim 1 and further comprising vertical walls defining said separating chamber, said walls being provided with channels for supplying and removing the elution fluid to and from said elongated elution chamber.

17. A device as claimed in claim 1 and further comprising adjustable means within the other buffer container for supporting said elution means and pressing it against the lower end of said separating chamber.

18. A gel electrophoresis device comprising two laterally spaced parallel, vertical walls forming a vertical separating chamber therebetween, two buffer containers on opposed sides of said separating chamber, one being only in communication with the upper end of said separating chamber and the other only in communica-

tion with the lower end of said separating chamber, whereby each container may be easily filled with buffer fluids to the same level in each container, an electrode in each of said containers and means for connecting said electrodes to a source of direct current.

19. A device as claimed in claim 18 wherein each of the laterally spaced walls form one wall of each of the buffer containers.

20. A device as claimed in claim 18, wherein one of said laterally spaced walls extends upwardly to a higher point than the other of said walls.

21. A device as claimed in claim 18 and further comprising a lid member disposed above said separating chamber, and wherein said laterally spaced walls extend upwardly to the same level, said lid member forming a fluid tight seal with one of said laterally spaced walls.

22. A device as claimed in claim 18 and further comprising a substantially rectangular box-like housing having a bottom and side elements and including an intermediate bottom element above the bottom of said housing and extending partially but not less than one half of the distance across said housing, the bottom of one of said laterally spaced walls being connected to the inner end of said intermediate bottom, whereby the intermediate bottom element and said one laterally spaced wall together with side portions of said housing form one of said buffer containers, the other of said buffer containers being defined by the other of said laterally spaced walls, the bottom and side elements of said housing, and the intermediate bottom element.

23. A device as claimed in claim 22 wherein the space between said laterally spaced walls is adjustable.

24. A device as claimed in claim 22 wherein a portion of said other buffer container is disposed beneath the lower end of said separating chamber.

25. A device as claimed in claim 22 and further comprising a detachable lid for said housing and wherein the side element of said housing spaced from said intermediate bottom element is detachable.

26. A device as claimed in claim 22 wherein the one laterally spaced wall forming a part of said one buffer container is provided with a window therethrough, and further comprising a cover plate for covering said window on that side facing said separating chamber and means for securing said cover plate over said window in said one spaced wall, said means also securing said other spaced wall in spaced relation to said cover plate to form said separating chamber.

27. A device as claimed in claim 26 and further comprising packing means between said cover plate and said other laterally spaced wall.

28. A device as claimed in claim 27 wherein said packing means comprises at least two groups of vertically extending parallel members, said groups being spaced apart to divide said separating chamber into at least two parallel, vertical chambers, said packing means being composed of the same material as the material facing said separating chamber.

29. A device as claimed in claim 26 and further comprising a lid member disposed above said separating chamber, said lid member forming a fluid tight seal with the top of said other-spaced wall to prevent communication between said other buffer container and said one buffer container.

30. A device as claimed in claim 26 wherein said securing means comprises bolts extending outwardly from said one laterally spaced wall and through said cover plate and said other laterally spaced wall and nuts for securing the cover plate and spaced walls together.

31. A device as claimed in claim 22 and further comprising a recess on the inside of one wall of each of said two buffer containers for receiving each of said electrodes.

32. A device as claimed in claim 18 wherein a wall of the other buffer container in communication with the lower end of said separation chamber is provided with a recess for receiving one of said electrodes.

33. A device as claimed in claim 32 wherein the wall of the one buffer container in communication with the upper end of said separation chamber is provided with a recess for receiving the other of said electrodes.

34. A device as claimed in claim 31 wherein each of said recesses in the walls is provided with an upwardly inclined upper surface and said walls are provided with channels extending from the uppermost part of said recesses to the top of said walls for removing gas bubbles formed at said electrodes.

35. A device as claimed in claim and further comprising detachable cooling means disposed in each of said buffer containers adjacent said laterally spaced walls for cooling said separation chamber.

36. A device as claimed in claim 35 wherein said cooling means comprises cooling plates arranged parallel to said laterally spaced walls.

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