

[54] **ARRANGEMENT FOR THE OPTICAL
EVALUATION OF SEVERAL
SIMULTANEOUSLY OBTAINED
SPECIMENS**

[75] Inventors: **Nikolaus Seiler; Josef Thobe**, both of
Frankfurt; **Gottfried Werner**,
Heusenstamm, all of Germany

[73] Assignee: **Max-Planck Gesellschaft zur
Foerderung der Wissenschaften e.V.**,
Goettingen, Germany

[22] Filed: **Apr. 26, 1972**

[21] Appl. No.: **247,830**

[30] **Foreign Application Priority Data**

Apr. 27, 1971 Germany..... 2120597

[52] U.S. Cl. **356/244, 356/201**

[51] Int. Cl. **G01n 1/18**

[58] Field of Search **356/105, 201, 205, 239,
356/240, 244, 246; 204/299; 73/423**

[56] **References Cited**

UNITED STATES PATENTS

3,478,598	11/1969	Nielsen	356/246
3,503,683	3/1970	Isreeli et al.	356/244
3,551,062	12/1970	Brown	356/244
3,584,964	6/1971	Nejame, Jr.	356/244
3,609,040	9/1971	Kuzel	356/246

Primary Examiner—William L. Sikes

Attorney, Agent, or Firm—Stanley J. Price, Jr.

[57]

ABSTRACT

The apparatus is arranged to collect specimens or components of a mixture that were separated by electrophoresis and includes a plurality of small tubes or cuvettes mounted on a preferably rotatable cuvette holder. The cuvettes are connected to the separate outlet openings of the separating apparatus by flexible tubes and other flexible tubes are provided to convey the measured material from the cuvettes to a collection unit. The cuvette holder has a plurality of spaced wedge-shaped members extending radially from the periphery and the cuvettes are supported between adjacent wedge members. Openings or slots are provided in the cuvette holder between the top and bottom of the cuvettes for the light beam to pass through the cuvettes onto photodetector apparatus. The cuvette holder has spaced peripheral recesses for resilient ring-like securing members that maintain the cuvettes on the cuvette holder. The cuvette holder is arranged to be positioned with the cuvettes in the path of a light beam of photometric apparatus and the cuvette holder is arranged to rotate about a fixed axis through a pre-selected arc so that the cuvettes are successively subjected to the light beam and optically evaluated by measuring the absorbency of the component passing through the cuvette. The cuvette holder may have the wedge-shaped members arranged radially or at various angles to the axis of the rotary cuvette holder.

10 Claims, 15 Drawing Figures

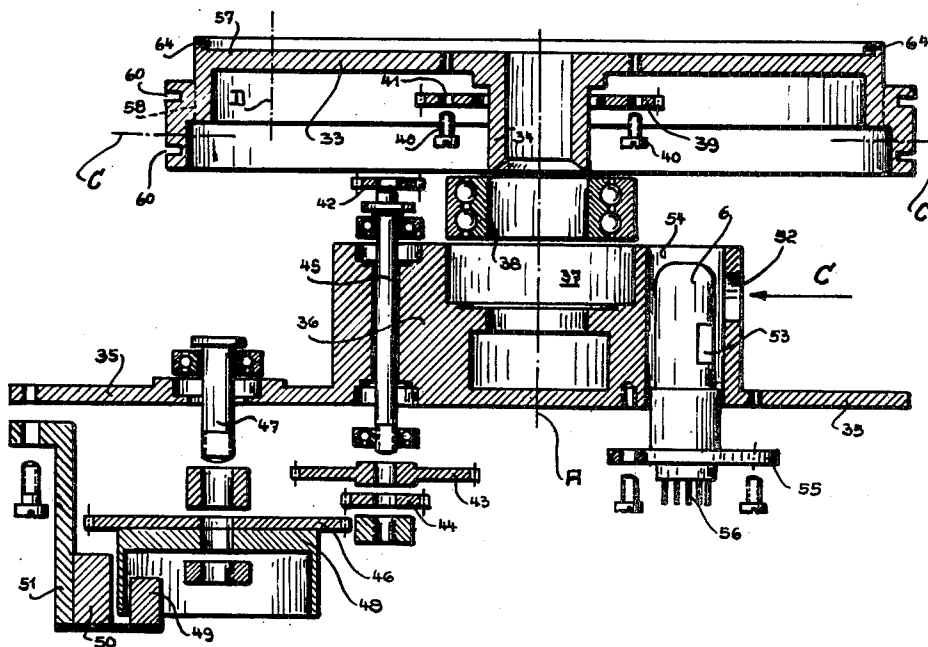


Fig. 1

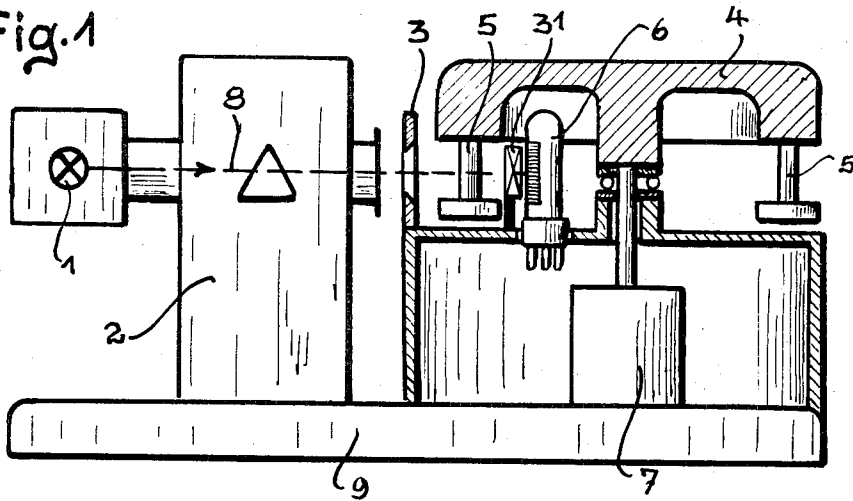


Fig. 2

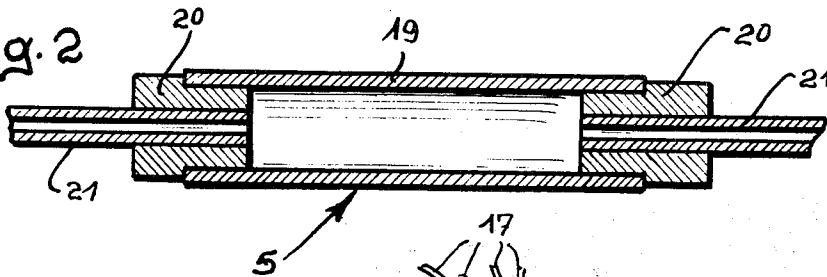
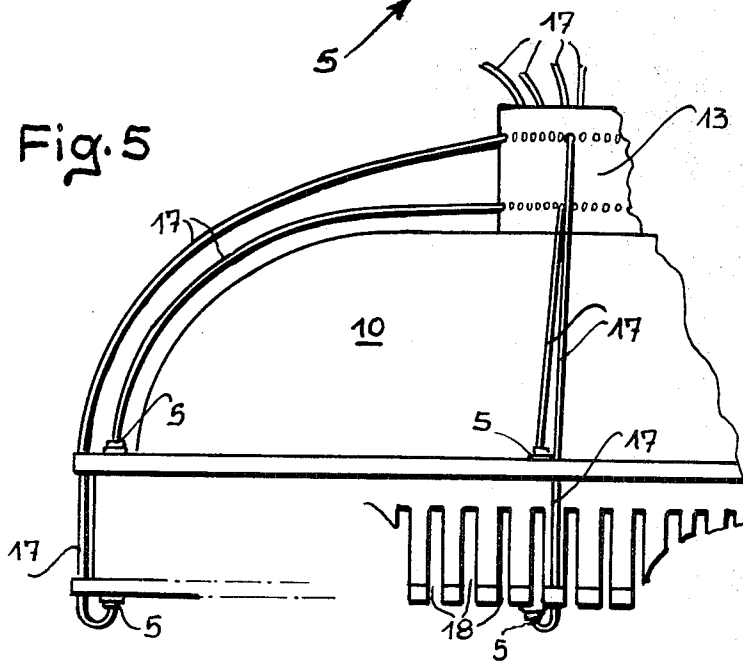


Fig. 5



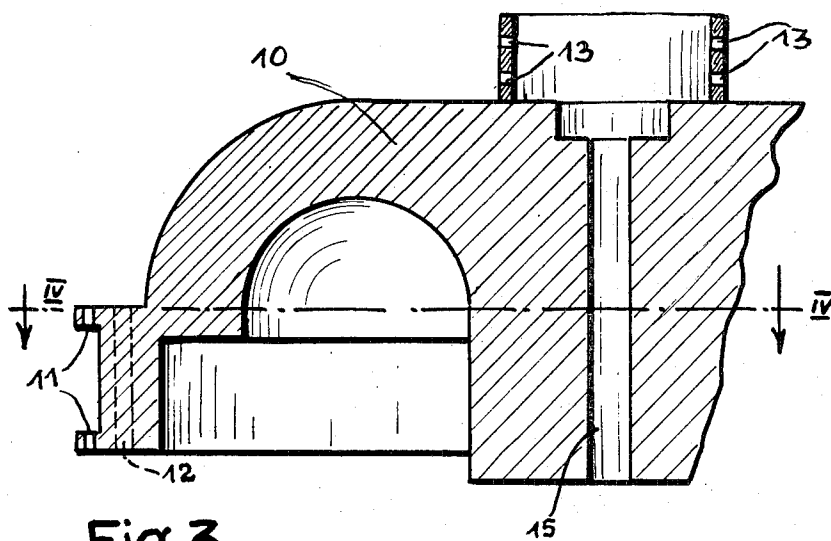


Fig. 3

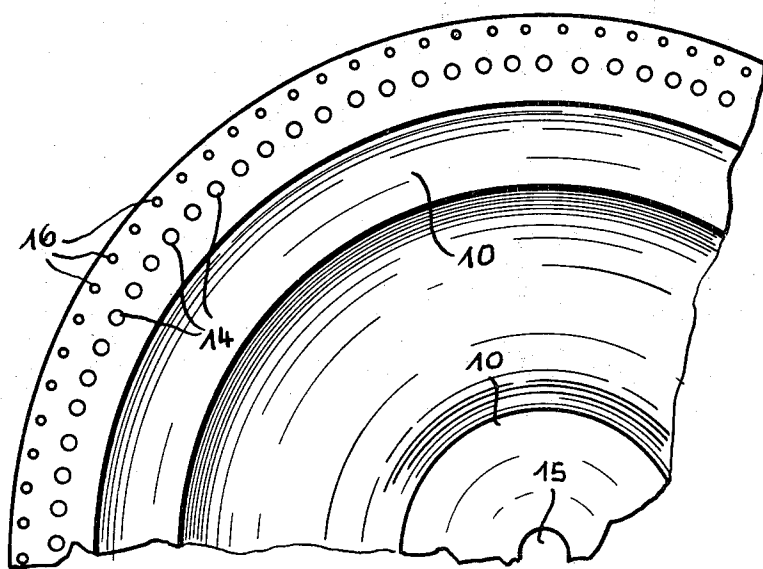
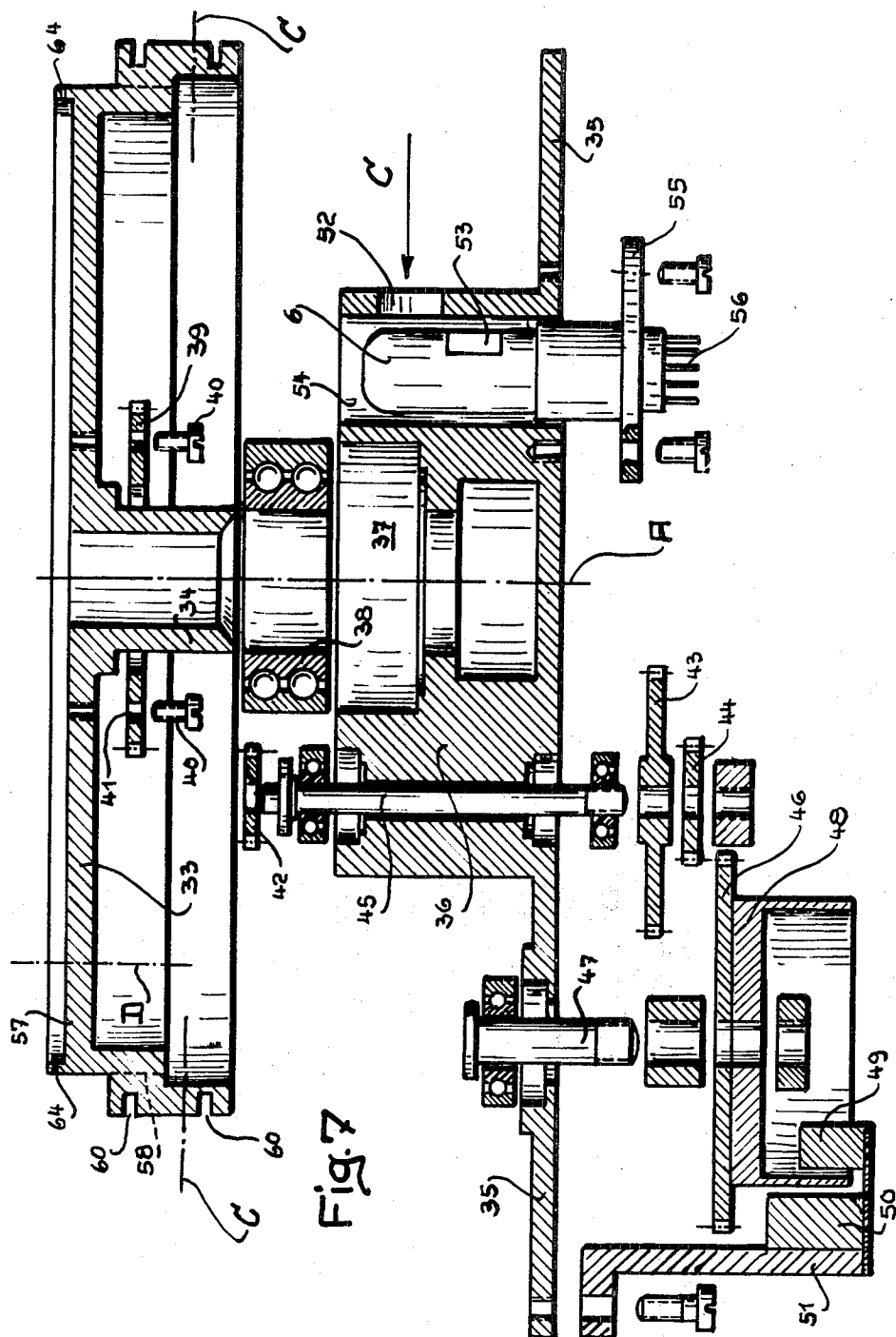


Fig. 4



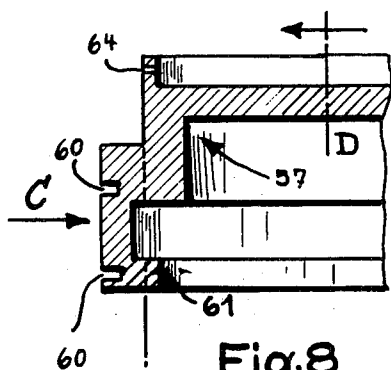


Fig. 8

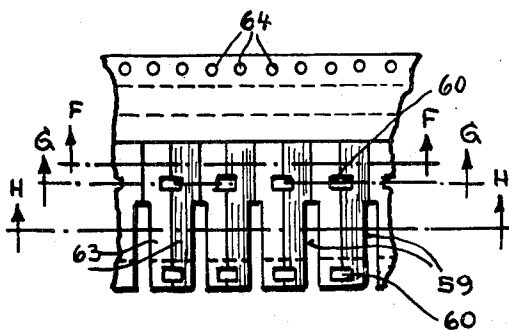


Fig. 9



Fig. 10a



Fig. 10b

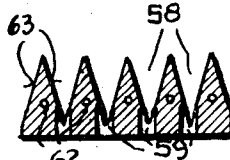


Fig. 10c

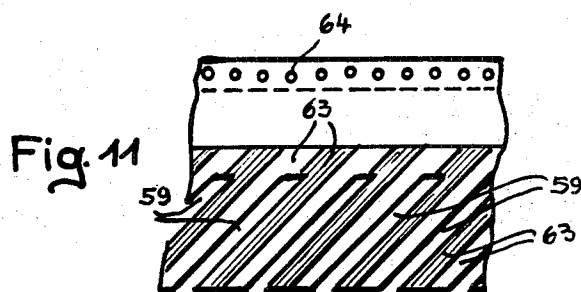


Fig. 11

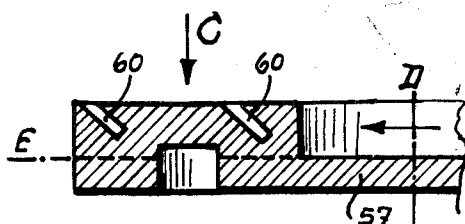


Fig. 12

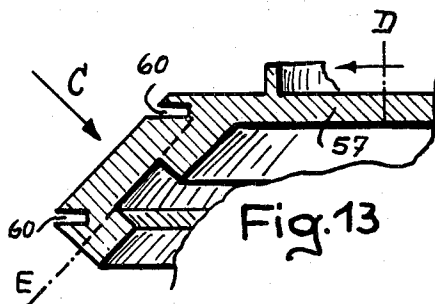


Fig. 13

ARRANGEMENT FOR THE OPTICAL EVALUATION OF SEVERAL SIMULTANEOUSLY OBTAINED SPECIMENS

BACKGROUND OF THE INVENTION

1. Field of the Invention

This invention relates to an arrangement for the optical evaluation of several simultaneously obtained specimens and more particularly to apparatus for the optical evaluation of several simultaneously obtained specimens flowing through a transparent measuring chamber.

2. Description of the Prior Art

In electrophoresis it is known to move a carrier free buffer stream downwardly between two plane parallel plates which have a small space, as for example a space of 0.5 millimeters thickness, therebetween. The buffer stream in curtain form has the same thickness as the interspace and moves downwardly therein. The mixture to be separated is introduced into the buffer stream at one location and moves vertically downward as a band. Under the influence of an electric current field, whose lines of force run at right angles to the vertical longitudinal borders of the plates, the components of the mixture are deflected laterally according to their charge. This lateral deviation is a measure of the chemical properties of the individual components of the mixture which according to this procedure can be analyzed. A description of electrophoretic separation may be found in the Journal of the Electrochemical Society entitled "Electrochemical Science" dated July, 1970 beginning on Page 874 in which an article by G. Brouwer and G. A. Postema discusses the theory of separation in displacement electrophoresis.

In the presently known apparatus for analyzing the electrophoretically separated components of a mixture a measuring chamber is connected to the lower portion of the parallel plates of the separation chamber and forms a continuation of the upper separation chamber. This measuring or evaluation chamber is also formed by two plane parallel plates which are maintained at a given distance from each other. The plates are sealed on their sides in a manner similar to the parallel plates of the separation chamber. A plurality of bores or openings are formed in the underside of the measuring or evaluation chamber through which the components of the mixture to be analyzed are separately drawn off through a corresponding number of single tubes for further analysis. A known continuous flow electrophoretic separator is illustrated in FIG. 19 on Page 458 of a publication entitled "Electrophoresis Theory, Method And Apparatus" edited by Milan Bier, Volume II, published by the Academic Press in 1967.

In order to determine the chemical composition of a given stream of the mixture which is conducted to a specific collection tube, it is known to illuminate the measuring chamber with a monochromatic beam so that the absorbance of the fluid can be measured in a connected photomultiplier. This known arrangement has a disadvantage in that the effective path of light in the measuring chamber is equal to the thickness of the film in the chamber, therefore, only 0.5 millimeters. This known arrangement provides evidence of the separation result within the separation chamber. It is, however, impossible to measure directly at the exit of the separation chamber. Therefore, the measured values

inside the separation chamber cannot be correlated to the measured values of the single fractions. Also, forming the measuring chamber integral with the separation apparatus weakens their mechanical stability. It is further necessary to dismantle the entire system with even a small contamination of the separation chamber or the measuring chamber. There is a need for apparatus that will efficiently and rapidly evaluate optically several simultaneously obtained specimens.

SUMMARY OF THE INVENTION

The present invention is directed to apparatus for eliminating the foregoing disadvantages and its primary object is to further develop a procedure so that exact measured results can be obtained within a short period of time. In accordance with the invention, there are provided a plurality of single chambers that are separate from each other as the measuring or evaluation chamber. These single chambers can be directly connected to the separation chamber so that the previously mentioned errors in the separation limits can be avoided. Further the diameters of the single chamber can be enlarged more than the distance between the plane parallel plates of the separation chamber so that the effective path of light for optical evaluation and the corresponding accuracy of the evaluation is increased.

In the preferred embodiment of the invention the single chambers are formed as small tubes or cuvettes. Tubes of this type are easily made from a single long tube and have substantially equal optical qualities. Also, it is possible to connect the tubes to hoses, stoppers and the like in a simple manner. With the separate tubes an arrangement can be provided whereby there is relative movement between the separate tubes or single chambers and the optical measuring beam.

Several arrangements of the single chambers are possible. The single chambers or cuvettes can be arranged linearly. It is preferred, however, to arrange the cuvettes in a circular fashion. With either arrangement the cuvette holder for holding the single chambers and the photometric system or apparatus can be moved relative to each other.

The preferred apparatus includes mounting the single separate tubes or cuvettes on a driven rotatable disc with the tubes or cuvettes arranged in spaced relation to each other. Such apparatus is easily connected to an already existing photometric system. Further, rotary movement of the apparatus is accomplished more easily with this arrangement.

For making single tubes in quantity a quartz tube can be cut into many small tubes of preselected dimension and both ends of the tubes can be closed by stoppers which have a bore therethrough and connecting pipes, preferably flexible tubes, are introduced into the bores of the stoppers. The stoppers are preferably made from silicone and the connecting pipe from flexible Teflon tubular material. The small tubes or cuvettes on the cuvette holder should be exchangeably mounted in such a manner that tubes of various thicknesses can be attached on wedge shaped notches of the cuvette holder. Because of the wedge shape of the notches holding the tubes, the thickness or diameter of the tubes no longer has to be exactly the thickness or diameter of the bores. Furthermore, the tubes can be easily exchanged as they can be removed from the front of the open notches. The arrangement with the wedge shaped notches fur-

ther provides for the formation of a slot or slit for limiting the measuring beam. This is provided by the notches partially entering the material of the cuvette holder and thereby creating the slots or slits for limiting the measuring beam.

In one form of this invention the vertical axis of the notches are parallel to the axis of the cuvette holder that is fashioned as a rotary platen or plate. If the notches are at an angle to the axis of the cuvette holder, then other arrangements are possible. If the angle is 90° whereby the notches are in a plane that is slanted to the axis of the cuvette holder, the axis of the cuvette holder can be arranged horizontally. This results in a particularly compact arrangement with a small height profile. Other angles between 0 and 90° are possible. With an inclination of about 45° the path of the measuring beam can be extended by this angle.

The notches can also be inclined to the vertical plane which extends through the cuvette holder axis. In this form the measuring beam which works as a band and runs horizontally can scan each tube one after the other and each tube can be scanned along its entire length. With adequate adjustment of the tube length relative to the length of the slot or slit through which the tubes are observed, a continuing process of the display between the single tubes can be achieved because the measuring beam always exactly illuminates one portion of the tube.

A preferred method for fastening the tubes to the wedge shaped notches in the cuvette holder so that they may be easily removed from the cuvette holder is to provide the cuvette holder with at least one peripheral recess extending therearound in which a clamping ring is positioned. The clamping ring fastens the tubes to the cuvette holder. Preferably, there are two peripheral recesses; one on the upper end and one on the lower end of the tube or cuvette and also generally outside of width of the slit. A rubber ring is adequate in many cases for the clamping ring. It is advantageous in other instances to singly attach the tubes of the group to the cuvette holder. For this purpose single elements are then provided in the form of clamps with which the tubes are positioned and held in the notches and retained therein.

It is important that the flexible tubes through which the material to be analyzed is conveyed to the small tubes or cuvettes and is again conveyed from the small cuvette are kept outside of the area of the slit or slot to be illuminated. It is therefore preferable to have the bores for these connection tubes run parallel to the wedge shaped slot through the cuvette holder at locations between the slots. The connecting tubes are led through the bores and care must be exercised to see that they do not extend into the area of the light beam.

The drive for the cuvette holder can be aligned with the axis of axial shaft of the cuvette holder. There are, however, advantages to arranging the drive in spaced relation to the axis of the cuvette holder. Where the drive is spaced from the axis of the cuvette holder space is provided in the area of the axis of the cuvette holder for the conveying tubes and the tubes can be directed from this space to any desired location.

In order to automatically record and store the results of the tests and the optical evaluations, an important improvement is seen in installations that are provided with apparatus to produce an analog or digital signal

proportional to the relative movement of the cuvette holder and measuring beam. This signal can trigger other apparatus for individual changes in the measured signals received from the tubes by the photometric apparatus. For example, a zero point or hundred point correction can be made of the measured values or of their accumulated or stored values.

The extent of change can automatically be derived from the size of the measuring system. For example, a filter can be automatically brought into the path of the beam as soon as all of the tubes or cuvettes are measured. Then the difference of two signals which emanate from one or the same tube are automatically made; one with and one without the filter. Other arrangements for manipulating the measuring beam can also be made and the extent of change can be obtained manually. The drive for the rotor or cuvette holder can also be used to bring the filter or other apparatus into the path of the measuring beam, for manipulating this measuring beam, limiting the slot, mirror etc. Synchronization is easily achieved by a common drive for these elements and for rotation of the rotary cuvette holder. This provides a particularly simple arrangement for manipulating the measuring beam that is positioned centrally to the axis of the cuvette holder.

The invention will be further explained by examples of methods for electrophoresis in a carrier free buffer stream. It should, however, be understood that this is only one of many possible areas of uses pertaining to this invention. The advantages of the invention will be seen from the following description and the examples.

DESCRIPTION OF THE DRAWINGS

FIG. 1 is a schematic view illustrating the rotary cuvette holder and photometric apparatus for optically evaluating the material within the separate cuvettes mounted on the cuvette holder.

FIG. 2 is a view in longitudinal section of a cuvette with the material conveying tubes connected thereto.

FIG. 3 is a more detailed view of the cuvette holder rotor body illustrated in FIG. 1.

FIG. 4 is a view in section taken along the line IV—IV of FIG. 3 illustrating the vertical bores in the cuvette holder for the cuvettes and the material conveying tubes.

FIG. 5 is a diagrammatic illustration of the cuvettes positioned on the cuvette holder rotor body and the flexible tubes for conveying the material to and from the cuvettes.

FIG. 6 is a circuit diagram of the electrical controls for rotating the rotor in both directions.

FIG. 7 is an exploded sectional view in elevation of another embodiment of the rotor.

FIG. 8 is a fragmentary view in section of the cuvette holder portion of the rotor illustrated in FIG. 7.

FIG. 9 is a developed view in side elevation of the cuvette holder portion of the rotor illustrated in FIG. 8.

FIG. 10a is a fragmentary view in section taken along the line F—F in FIG. 9.

FIG. 10b is a fragmentary view in section taken along the line G—G in FIG. 9.

FIG. 10c is a fragmentary view in section taken along the line H—H in FIG. 9.

FIG. 11 is a view similar to FIG. 9 illustrating the wedge-shaped recesses in an angle or slanted position.

FIG. 12 is a view similar to FIG. 8 illustrating an embodiment where the measuring beam of light enters from above.

FIG. 13 is a view similar to FIGS. 8 and 12 illustrating another embodiment where the beam of light enters at an angular or slanted position to the axis of the rotor.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

Referring to the drawings, FIG. 1 schematically illustrates the arrangement of one embodiment of this invention associated with photometric apparatus. The beam of light 8 from the lamp 1 is spectrally split in a monochromator 2 and passes through and is limited by a slit 3. The light beam 8 then passes through the cuvettes 5 mounted on the rotor 4. The cuvette holder which will also be referred to as rotor 4 has a plurality of cuvettes or small tubes 5, later described in detail, mounted around the periphery of the rotor 4. The light beam 8, after passing through the cuvette 5, strikes a photodetector tube 6 whose signal is amplified or multiplied and recorded by a suitable recording instrument. A filter 31 can be inserted before the photodetector 6. The above described apparatus is mounted on an optical stand 9. The rotor 4 is driven by a drive mechanism 7 around its axis whereby the individual cuvettes 5, which are arranged in this embodiment axially on the periphery of rotor 4, are successively passed through the measuring light beam 8. The drive mechanism 7 is arranged to rotate the rotor 4 so that the respective spaced cuvettes pass successively through the light beam 8.

FIGS. 3-5 illustrate details of the rotor 4. In FIG. 3 the rotor body designated by the numeral 10 has vertical bores or holes 11 in the periphery through which the flexible tubes extend. The rotor body 10 has adjacent vertical bores 12 in which the respective cuvettes 5 are positioned. A cylindrical member 13 extends upwardly from the rotor body 10 and is arranged as a gathering device for the flexible conveying tubes. In this embodiment the connecting ring 13 is positioned concentrically with the axis 15 of rotor 4 on the upper surface of the rotor body 10.

FIG. 4 illustrates in top plan the radially extending disc portion of the rotor body 10 that includes the vertical apertures 14 which form the bores 12 previously described. The bores 12 have vertical openings therein extending radially outwardly on the periphery of the rotor and parallel to the rotor axis 15 to form vertical slots or slits in the rotor body 10 and provide separate slots for each bore 12 so that the beam of light passes through the cuvette and is focused on a photodetector similar to the photodetector 6 positioned within the periphery of the rotor body 10. The space in the rotor body 10 between adjacent slots has apertures 16 therein which form the bores or holes 11 previously described for the conveying tubes.

FIG. 5 illustrates the rotor body 10 with the flexible tubes 17 connected to the top and bottom of the vertically positioned cuvettes 5. FIG. 5 further illustrates the manner in which the cuvettes 5 are supported on the rotor body 10 and the spaced slots 18 in the cuvette

holder for passage of the light beam 8 through the cuvettes and onto the photodetector 6.

The rotor 4 is preferably anodized or oxidized in a nonreflective black to avoid disturbing reflections of the light beam and corrosion of the metal surface. The drive mechanism 7 includes a DC motor with a suitable gear reducing unit. By varying the voltage to the drive motor of drive mechanism 7 the rotation of the rotor through a preselected arc can be regulated to between 15 seconds and 5 minutes as will be explained in reference to FIG. 6. contact switches provide for a polarity reversal of the drive motor to rotate the drive motor to an original position after the rotor 4 has rotated to about 350°.

FIG. 2 is a detailed illustration of the cuvettes 5 and the connecting conveying tubes. The cuvette 5 is preferably fabricated from a quartz tube 19 into which a short piece of silicone tube 20 is inserted at each end. The silicone tubes have axial bores therethrough and flexible Teflon tubes 21 are provided as the conveying conduits 17 to convey the separated components to and from the cuvette tubes 19.

In FIG. 6 there is illustrated a power pack 22 that is connected to a 220 volt source of alternating current. Secondary DC 8 volt and 15 volt direct current is supplied from the power pack 22. A potentiometer 23, relay 24 and a keying device 25 are arranged in the circuit to actuate the relay 24 into a holding position to thereby energize and rotate the motor 26 in a forward direction. A keying device 27 is arranged to terminate the forward rotation of the rotor at any desired location and automatically switches the rotor for return reverse movement. On the rotor shaft of motor 26 there is a rod 28 provided for activating contacts 29 and 30. The contact 29 is a micro switch for stopping the forward rotation of the measuring rotor. Contact 30 is a micro switch which stops the rearward rotation of the measuring rotor.

The following apparatus and materials were employed for the optical evaluation of specimens. An optical stand, monochromator (M4Q11), an indicator of the spectral photometer (PMQ11 Carl Zeiss Oberkochen), a secondary electron multiplier (RCA 1P28) served as a photodetector, and the multiplier was provided with a suitable rheostat for connection to an amplifier. For recording the measuring signal, a potentiometric recorder (RIKADENKI KOGYO B24S) with an adjustment speed of 0.3 seconds was employed. The quartz tubes or cuvettes (2.4 millimeters inner diameter and 3.0 millimeters exterior diameter, and 1,000 millimeters in length) were furnished by Haereus-Schott, Hanau. Teflon tubes were used for the tube connections 20 and had an inner diameter of 0.5 millimeters and an outer diameter of 1 millimeter. The flexible tubes 21 were silicone tubes having an 0.5 millimeter inner diameter and 2.5 millimeter outer diameter. A 6 volt direct current drive motor was used to rotate the rotor 4. The rotor 4 was provided with 50 cuvette tubes for testing or performing the optical evaluation. The tubes or cuvettes were cut from two almost identical quartz tubes and the sequence of the cuvettes or cut tubes was statistical in that no attempt was made to arrange adjacent segments of the long tube in the rotor and no distinction was made for cuvettes cut from the two different tubes. Distilled water, hemoglobin and ovalbumin in a physiological saline solution and a solution of amido black in water were circulated through

selected rows of cuvettes. The absorbencies of these solutions were determined in repeated tests with various light wave lengths (see table). In these tests groups of 10 cuvettes having the same composition were used.

The above described apparatus is intended to be used as an accessory for high voltage electrophoresis apparatus. The apparatus is, however, also useful to evaluate optically a large number of simultaneous specimens within a relatively short time.

In spite of the low cost in manufacturing the flow through cuvettes, the dispersion in their transmissivity or transparency values is remarkably small (see values for uncorrected standard deviations and maximum deviations in the following table). For each single small tube the difference was determined in the measured absorbency of the solutions and pure solution media so that a considerable reduction of dispersion resulted (see corrected value in the table). This primarily illustrates the difference in the transparency qualities of the small tubes by variable wall thickness, material and surface qualities and not by the variable lumina of the cuvette tubes.

For testing the transparencies greater than 80% this method to select cuvettes is for certain applications not sufficiently accurate. There are many ways to reduce the deviations between the individual tubes, as for example:

1. A large number of cuvettes are measured and a group of cuvettes are selected for their similar values.
2. The cuvettes are arranged in the cuvette holder according to increasing transparency values. This results in a reference line which is taken into account in the correction of the test values.
3. Mechanical correction by a device which changes the effective slot or slit height.
4. Electronic compensation:
 - a. A contact disc is attached to the cuvette holder which by a potentiometer, for example, switches on an adjustable compensation voltage.
 - b. The cuvette holder drive also moves a recording tape on which the compensation values, i.e., the transparency values for pure solution media, are stored for each individual cuvette.

There is very little necessity for uniform rotation of the rotor or cuvette holder since the measured transparencies and the correlation of the transparency values to certain fractions or specimens are independent of the measuring rotor rotational speed. In practice a maximum scanning speed is determined in the above described apparatus by the speed of the potentiometric recorder and amounts to about 50 cuvettes per minute. An increased scanning speed can be achieved with this apparatus by a stepwise drive of the measuring system and by illumination with a homogeneous light beam whose width is somewhat less or equal to the distance between two cuvettes, i.e., the slot, or slit (in a round system the light beam must converge so that its focal point is in the center of the measuring rotor). In both methods the reversal of the rotor is avoided for the value $E = \infty$ between two cuvettes or at least is replaced by a small reversal. A far greater recorder speed can be achieved by selecting suitable amplifiers and by recording the measuring signals with a very low adjustment time or by an oscilloscope.

Arranging the cuvettes 5 in the rotor bores 12 that have the slots or slits 18 is advantageous in that the spaced cuvettes 5 do not influence each other and in principle fluorescence measurements are possible. It was ascertained in separate tests that the absorbency measurements of non-fluorescing, non-scattering solutions do not require shielding. Tubes or cuvettes positioned directly adjacent to each other do not show any mutual influences.

- 10 There are, as previously stated, several alternatives for the erection of the measuring apparatus. With an evaluation photometer the arrangement can be similar to the above mentioned circular arrangement or with fewer tubes a linear arrangement of the cuvettes appears most favorable. The above described system can thus be developed as an addition to the photometer. A rotating filter photometer can be of advantage in the installation with fast, succeeding changes of the transparency values in the cuvettes or with integration of the measuring system with the apparatus furnishing the specimens or with fewer demands on the spectral purity of the measuring beam.

If one dispenses with the measuring of very high absorbencies or very little differences in absorbencies the area of use of this measuring system in the present state of development will be limited mainly by the scanning speed. This amounts to, as previously stated, one cuvette per second and if you consider the return time, one cuvette every 2 seconds. With 50 parallel tests, as perhaps with previously described separation chamber measurements can be obtained after two minutes for each flow through cuvette. This measuring frequency is acceptable for the control of preparatory separations by carrier-free high voltage electrophoresis since with these separations only a slow drift of the separation parameter must be followed. At the same time analysis of the content is obtained for the fraction collected in the various vessels.

If diverse components are injected into the separation chambers for a length of 5 minutes with a successive pause of 5 minutes, then for each specimen two absorbency measuring determinations of all 50 cuvettes can be obtained. Analytical use under these assumptions is also possible in the carrier-free high voltage electrophoresis apparatus in combination with the system of measurement according to the above described invention.

For the quasi continuous column type chromatography processes the measuring apparatus, according to the invention, is only then suitable if with 50 columns a quick, timely analysis is not expected. If, however, only a part of the measuring rotor is scanned, meaning the evaluation or scanning of perhaps 10 columns, then a measure value can be registered for each of the cuvettes in 20 to 25 seconds. This frequency is sufficient in most cases for elutriation (elution) processes.

Other embodiments of the rotary cuvette holder and embodiments of the manner of supporting the cuvettes on the cuvette holder are illustrated in FIGS. 7-13. The cuvette holder illustrated in FIG. 7 has a measuring rotor 33 which is driven by drive apparatus (not shown) around its axis A. The rotor 33 has a depending bearing sleeve 34 that extends into a bearing 38. A carrier block 36 extends upwardly from a base plate 35 and has a recessed bore 37 therein. The bearing 38 is positioned in the recessed bore 37 with the rotor bearing sleeve 34 positioned in the bearing 38. On the un-

derside of the rotor 33 a gear 39 is secured thereto by screws 40 extending upwardly through the apertures 41. The gear 39 is arranged to mesh with gear 42 that is secured to the shaft 45 for rotation therewith. Thus, rotation of the shaft 45 is transmitted through gear 42 to the rotor 33 through gear 39. The shaft 45 has gears 43 and 44 secured to its lower portion in spaced relation to the gear 42 and both gears are positioned beneath the base plate 35. The gear 43 is arranged to be drivingly connected to a drive mechanism (not shown) so that rotation of gear 43 by the drive mechanism is imparted to the shaft 45 and through gears 42 and 43 to the rotor 33. The gear 44 meshes with a gear 46 that is secured to a coding device 48 by means of a shaft 47. Heads 49 and 50 are arranged to scan the coding device 48 and are suitably supported with an annular end portion of the coding device therebetween by a mounting support 51. The drive from the drive mechanism (not shown) is transmitted through gear 43 to shaft 45 and from shaft 45 through gears 44 and 46 to rotate the coding device 48 in timed relation to the rotor 33. Through the above described gears the rotor 33 and coding device 48 are rotated in synchronization. By coding system 48, 49 and 50 an analog or digital signal can be produced in accordance with the relative position of the rotor 33.

The rotation of rotor 33 will move the peripheral ring (shown in FIG. 7 through the area to the left of Line D) through the measuring beam C. FIG. 7, being an exploded view of the elements, illustrates the measuring beam C in two parallel locations; in one location by the directional arrow and in the second location thereabove by the -- line. However, when the apparatus illustrated in FIG. 7 is assembled, i.e. the rotor sleeve 34 positioned within the bearing 38 and the bearing 38 positioned within recess 37 of carrier block 36, the -- line indicates the measuring beam C is in overlying relation with the indicating arrow for the measuring beam C.

The rotation of rotor 33 moves successive portions of the periphery of rotor 33 through the measuring beam C. The beam enters through an opening 52 in the carrier block 36 and impinges on the window 53 of photodetector 6. This detector 6 is inserted in bore 54 from below with its mounting plate 55 secured to the underside of base plate 35 by screws. The signals from the photodetector 6 are transmitted through the connections 56.

In FIG. 7 the peripheral ring designated by the numeral 57 is illustrated in section through the axis of the rotor 33 and is integral with the rotor 33. The peripheral ring 57 is arranged to receive, hold and position the cuvettes 5 and is provided with elements for guiding the conveying tubes connected to the respective cuvettes. FIGS. 7, 8 and 10A illustrate the wedge shaped recesses 58 in the periphery of ring 57 in which the tubes or cuvettes 5 are inserted. These recesses extend along a peripheral line E and also form the measuring slot 59 illustrated in FIGS. 9 and 10C.

The upper and lower portions of the wedge shaped members between the wedge shaped recesses have peripheral or circular recesses 60 which extend around the outer circular surface of rotor 33 and in which resilient or rubber rings are positioned to hold the tubes or cuvettes in the recesses 58 from above and below. The embodiment illustrated in FIGS. 7 and 8 differs from FIG. 1 in that the embodiment in FIGS. 7 and 8 provides a lower height limit of the measuring beam by the

extension on the periphery designated by the numeral 61.

FIGS. 10A, 10B and 10C illustrate other bores 62 for accepting the conveying flexible tubes which convey the fluids to the cuvettes and convey the fluids from the cuvettes.

In FIG. 11 the slots 59 are slanted relative to the rotor axis A at an angle of about 45°. This arrangement permits when using a beam that is parallel to the axis A and at a suitable height to receive a continuously produced signal from tube to tube.

FIG. 10A is taken along the Line F—F in FIG. 9 and illustrates again in greater detail the wedge shaped grooves 58 whose depth of penetration (wedge cut E) into the peripheral ring 57 is so dimensioned that openings 59 are provided to serve as the measuring slot 59. The tubes or cuvettes are laid in the grooves 58 and urged against the flanges 63 of the wedges by the circular resilient tension bands positioned in the circular recesses 60 to maintain the cuvettes in a fixed position in front of the measuring slot 59 as illustrated in FIG. 9.

All flexible tubing leading to and away from the cuvettes extends through the bores 64 in the circular upwardly extending flange of rotor 33. The flexible tubes extending through the bores 64 can be combined in the area of the rotor axis A and led upwardly away therefrom. The combined group of flexible tubes can also extend through the center of the measure rotor ring 34 and downwardly through the bore 37 into the opening therebelow and extend away from the rotor in a plane normal to the plane of the drawing.

The above described arrangement is capable of several possible variations. The driving axis, i.e. the axis for rotating the rotor, and the rotor axis A (see FIG. 7) are separate and displaced from each other to provide a means to lead part or all of the connecting flexible tubes through the center of the measure rotor 33. The guideway, as shown in FIG. 7, in the lower area of bore 37 normal to the plane of the drawing can be replaced with a guideway down through the base 35. The axis of the coding system, which is the axis of shaft 47, was selected to be separate from the drive axis, i.e. the axis of shaft 45, in order to select dimensions and rotation speed for the coding device 48 and further to simplify the assembly and disassembly of the apparatus. It is, however, also possible to position the coding device axis coaxially with the drive axis, i.e. the axis of shaft 45, or coaxially with the measure rotor 33. Coding and scanning can be of the analog type by a suitable potentiometer and also digital (over perforated strips, discs, electro-magnetic or electro-optical scanning). The form of the peripheral ring 57, as shown in FIGS. 7 and 8, can be further modified as illustrated in FIGS. 12 and 13. The grooves, the boundary of which is designated by the letter E, can be arranged perpendicular to the rotor axis A or at an angle thereto. FIG. 12 illustrates an arrangement where the wedge shaped groove 58 with the edge E is perpendicular, i.e. slanted around 90° in the direction of the radius opposite measuring rotor axis A, with the light beam C being substantially parallel to the axis of the rotor 33. In FIG. 13 another embodiment is illustrated where the grooves are arranged at a divergent angle of about 45° to the axis of the rotor 33. With this arrangement the light beam is at a 45° angle to the axis of the rotor 33. The advantage of the embodiments illustrated in FIGS. 12 and 13 is greater freedom in ar-

ranging the elements for controlling the measuring beam C.

TABLE

TESTS FOR DETERMINING THE DEVIATION OR STRAYING OF THE TRANSPARENT QUALITIES OF THE FLOW-THROUGH CUVETTES

Groups of n cuvettes had solutions amido-black B in water, hemoglobin and ovalbumin in physiological sodium chloride solution flowing therethrough and absorbence values were determined. The absorbences of the solutions were first determined in plane parallel cuvettes of 10 millimeter thickness. E_{10} — the measured absorbence in 10 millimeter cuvettes; λ — the wave length of light used for measurements; n — number of cuvettes included in the measurement; E_D — mean value of the absorbences measured in n flow-through cuvettes; s_1 — standard deviation from the mean value E_D — in absorbence units; s_2 — standard deviation from mean values E_D in percent; F — maximum deviation from the mean value E_D ; those with index K values of s and F are calculated from values which for each single tube were obtained by the subtraction of absorbences of the pure solvent from the solution's absorbences (corrected values). E_D/E_{10} gives the ratio of the absorbence values which on one hand were realized with the flow-through cuvettes and on the other hand with the 10 millimeter cuvettes. It is a measure for the effective thickness of the flow-through cuvettes and for the proportionality of the absorbence values which were obtained by both methods. For water and for physiological NaCl, $n = 100$ $s_1 \pm 0.0052$ and $F \pm 0.01$ with all wave lengths.

TABLE

Solution	Amido-black $\lambda=600$ nm.				Hemoglobin $\lambda=420$ nm.			Ovalbumin $\lambda=280$ nm.	
	I	II	III	IV	I	II	III	I	II
E_{10}	0.085	0.155	0.33	0.70	0.21	0.54	1.1	-----	-----
n	50	50	50	50	20	20	20	50	50
E_D	0.021	0.036	0.082	0.163	0.050	0.13	0.26	0.046	0.138
s_1	± 0.0051	± 0.0051	± 0.0051	± 0.0045	± 0.0055	± 0.0056	± 0.0065	± 0.0055	± 0.005
s_2	± 24	± 14	± 6	± 2.8	± 11	± 4.3	± 2.5	± 12	± 3.6
F	± 0.01	± 0.01	± 0.01	± 0.01	± 0.008	± 0.01	± 0.012	± 0.008	± 0.014
s_{1K}	± 0.00086	± 0.001	± 0.0015	± 0.0022	± 0.0014	± 0.0021	± 0.0042	± 0.0014	± 0.0025
s_{2K}	± 4	± 2.8	± 1.8	± 1.4	± 2.8	± 1.6	± 1.6	± 3	± 1.8
F_K	± 0.0015	± 0.002	± 0.003	± 0.005	± 0.003	± 0.005	± 0.007	± 0.003	± 0.004
E_D/E_{10}	0.247	0.233	0.248	0.233	0.238	0.240	0.236	-----	-----

According to the provisions of the patent statutes, the principle, preferred construction and mode of operation of the invention have been explained and what is considered to represent its best embodiment has been illustrated and described. However, it should be understood that, within the scope of the appended claims, the invention may be practiced otherwise than as specifically illustrated and described.

We claim:

1. Apparatus for the optical evaluation of several simultaneously obtained specimens comprising, a plurality of separate transparent chambers, conduit means connecting said chambers to sources of the separated specimens to introduce said separated specimens into said separate transparent chambers for optical evaluation of each of said separated specimens,

a rotor having a vertical axis and a peripheral portion, means to support said separate transparent chambers along said peripheral portion of said rotor in spaced relation to each other,

5 photometric apparatus having a single light beam emanating therefrom and a single photodetector tube, said light beam focused on said photodetector tube with one of said transparent chambers therebetween,

10 drive means to rotate said rotor in a first direction to successively pass said separate transparent chambers through said light beam for optical evaluation of each of said specimens in each of said transparent chambers, and

15 means to reverse said drive means and rotate said rotor in an opposite direction.

2. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 1 in which,

20 said separate transparent chambers have a tubular configuration.

3. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 2 in which,

25 said separate tubular transparent chambers are formed from a quartz tube, stoppers positioned in each end of said tube, said stoppers have longitudinal bores therethrough, connecting tubes extending into said longitudinal bores for circulating fluid specimens from said sources of separated specimens through said tubular transparent chambers.

4. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 1 in which said separate transparent chambers are posi-

tioned in a circular arc in spaced relation to each other.

5. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 1 which includes wedge shaped notches in the periphery of said rotor,

55 said separate transparent chambers having a tubular configuration, said separate transparent chambers positioned in said wedge shaped recesses in said rotor periphery.

6. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 5 in which said wedge shaped recesses are arranged parallel to the axis of said rotor.

65 7. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 5 in which said wedge shaped recesses are arranged perpendicular to the axis of said rotor.

13

8. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 5 in which said wedge shaped recesses are at an angle to the plane extending through the axis of said rotor.

9. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 5 in which said rotor includes a pair of spaced circular recesses extending around said rotor periphery, resilient ring members positioned in said circular re-

14

cessed portions and frictionally engaging said separate transparent chambers in said wedge shaped recesses.

10. Apparatus for the optical evaluation of several simultaneously obtained specimens as set forth in claim 5 in which said rotor drive means is arranged in lateral spaced relation to said rotor axis.

* * * * *

10

15

20

25

30

35

40

45

50

55

60

65