# United States Patent [19]

**Tiffany et al.**

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[54] **MULTIPLE-SAMPLE ROTOR ASSEMBLY FOR BLOOD FRACTION PREPARATION**


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[52] **U.S. Cl.** ..... **23/258.5,** 23/259, 233/26, 233/28

[51] **Int. Cl.** ..... **B04b 9/12,** B04b 11/02

[58] **Field of Search** ..... 23/258.5, 259, 253 R; 233/26, 28



[56] **References Cited**

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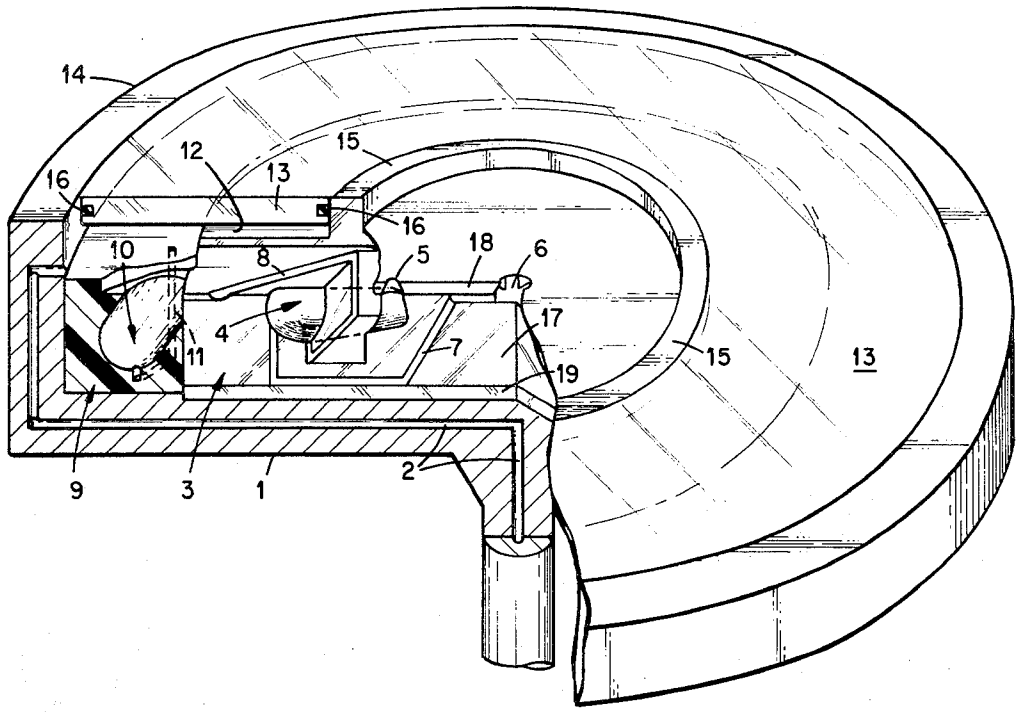
[57] **ABSTRACT**

A multiple-sample centrifugal rotor for blood fraction preparation is described. The rotor assembly includes an inner disk-shaped portion defining a circular array of whole blood sample-receiving chambers. Blood samples are statically loaded into the respective chambers through static loading ports. Liquids for washing and hemolyzing the cell fractions are introduced to the chambers following recovery of the plasma fraction by means of a central dynamic distribution port and a multiplicity of distribution passageways extending between the dynamic distribution port and the centrifugal ends of respective chambers. Unloading of blood fractions and washing liquid is accomplished through transfer passageways extending from a point intermediate the centrifugal and centripetal ends of the chambers radially inward and then outward to the periphery of the inner disk-shaped rotor assembly portion. A removable outer rotor portion defining at least one collection chamber for receiving materials discharged from the transfer passageways is nested concentrically about the inner rotor assembly portion.


**5 Claims, 4 Drawing Figures**



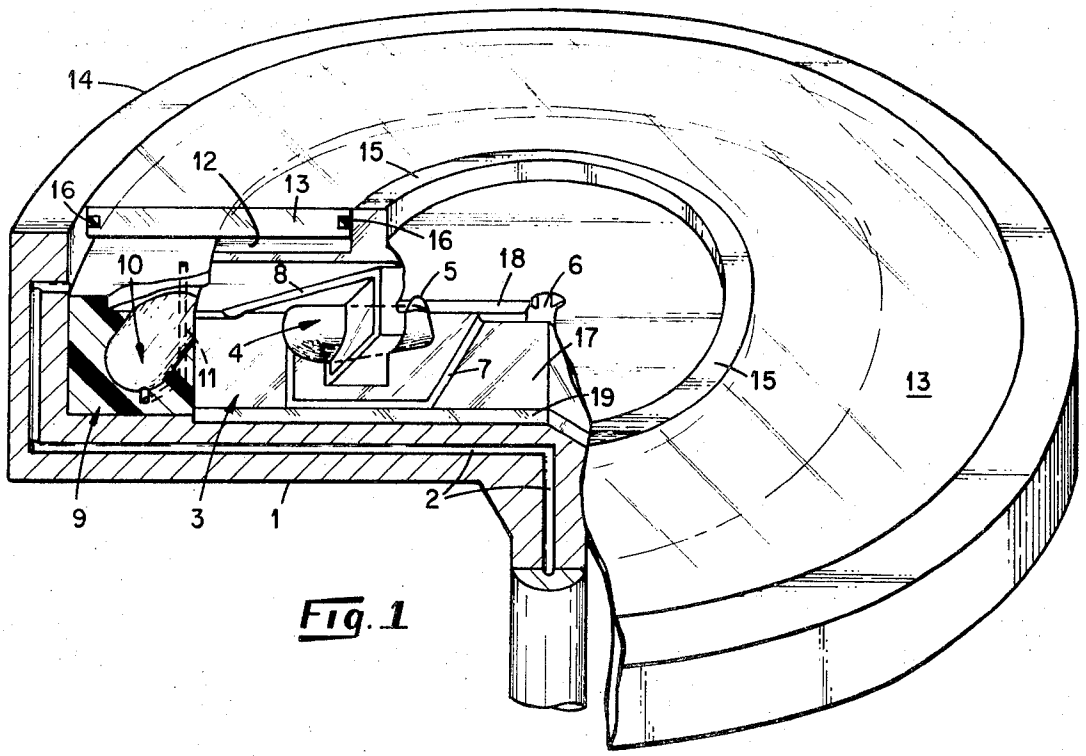



Fig. 1

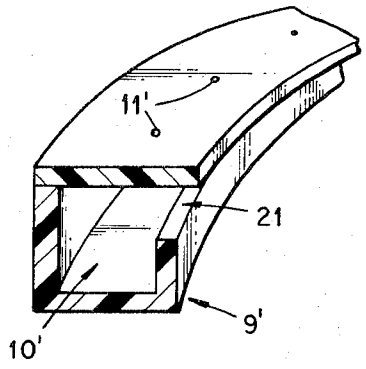



Fig. 3

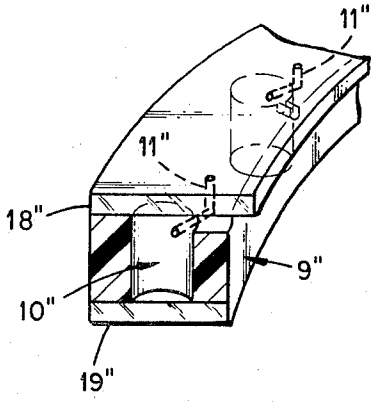



Fig. 4

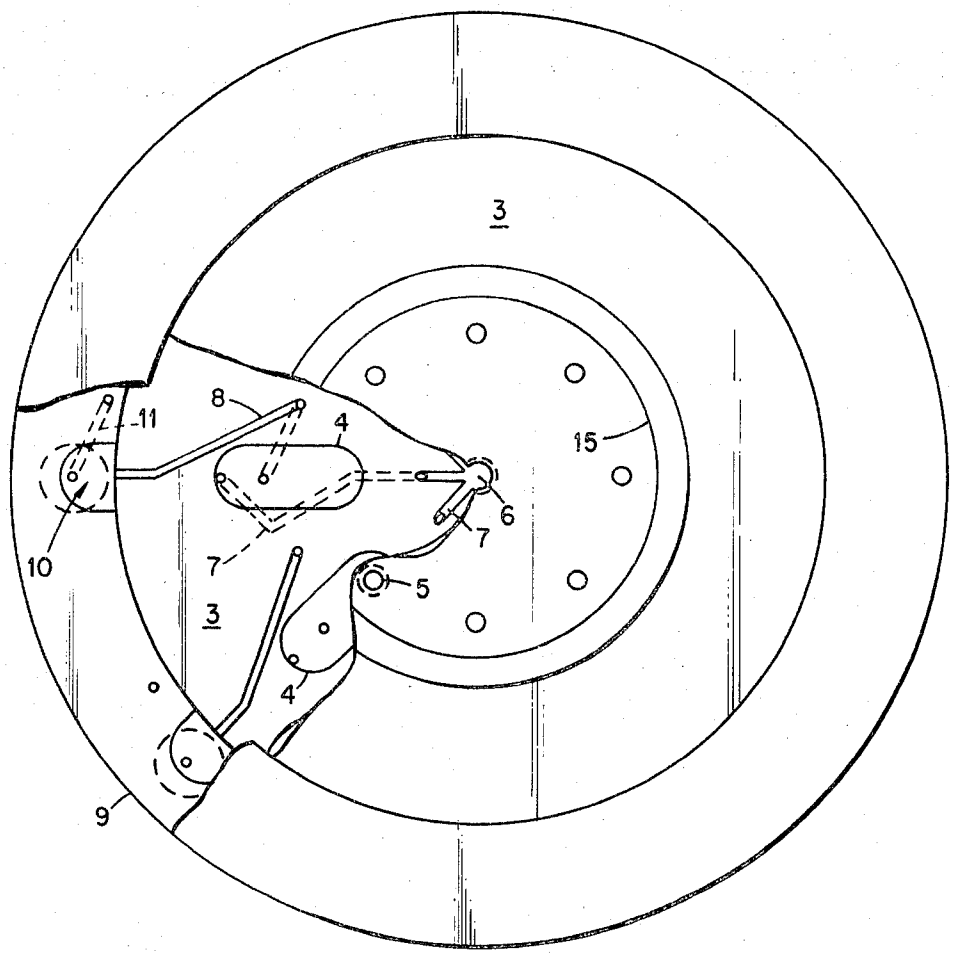



Fig. 2

# MULTIPLE-SAMPLE ROTOR ASSEMBLY FOR BLOOD FRACTION PREPARATION

## BACKGROUND OF THE INVENTION

The invention described herein relates generally to blood fraction preparation systems and more particularly to an improved multi-sample rotor assembly suitable for separating blood into plasma and cell fractions, washing and hemolyzing the cell fraction, and for separately recovering the plasma, hemolysate, and washed cells. It was made in the course of, or under, a contract with the U.S. Atomic Energy Commission.

In clinical blood work, it is necessary to separate stabilized blood samples into plasma and washed cell fractions before many biochemical tests of interest can be performed. For example, photometric analysis may be performed on the plasma fraction only since the presence of red blood cells interferes with the desired absorption measurement.

Genetic monitoring programs to determine mutations in man caused by environmental conditions such as the presence of ionizing radiation, chemical pollutants, etc., as well as other natural causes require the taking, preparation, and analysis of very large numbers of blood samples due to low mutation rates presently postulated. Present clinical laboratory blood fraction preparation techniques involve tedious and time-consuming operations which would make an effective genetic monitoring program impractical, however.

It is, accordingly, a general object of the invention to provide a rotor assembly which is suitable for simultaneously preparing blood fractions from a multiplicity of whole blood samples.

Another, more particular object of the invention is to provide a rotor assembly suitable for separating a multiplicity of blood samples into plasma and cell fractions, washing and hemolyzing the cell fractions, and separately recovering the plasma, hemolysate, and washed cells.

Other objects of the invention will be apparent upon examination of the following written specification and appended drawings.

## SUMMARY OF THE INVENTION

In accordance with the invention, a rotor assembly is provided for preparing blood fractions from a multiplicity of whole blood samples. The rotor assembly includes an inner rotor portion defining a circular array of radially extending whole blood sample-receiving chambers. Static loading ports communicate between the top end surface of the inner rotor portion and respective chambers in the circular array of chambers to facilitate the static loading of whole blood samples therein. A central dynamic distribution port communicates, by way of a multiplicity of radially extending distribution passageways, with the centrifugal ends of each of the sample receiving chambers. Transfer passageways for unloading of blood fractions and washing liquid from the chambers extend from a point intermediate the centrifugal and centripetal ends of the chambers radially inward and then outward to the periphery of the inner rotor portion. A removable outer rotor portion, defining at least one collection chamber for receiving materials discharged from the transfer passageways, is nested concentrically about the inner rotor portion. Rotor assemblies made in accordance with the invention are suitable for separating blood into plasma and cell fractions, washing and hemolyzing the cell fraction, and for separately recovering the plasma, hemolysate, and washed cells.



## BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 is a perspective view, vertically sectioned, showing a rotor assembly made in accordance with the invention mounted within a turntable.

FIG. 2 is a top plan view, partially cut away, of the rotor assembly of FIG. 1.

FIG. 3 is a perspective view of a removable outer rotor portion designed for collecting cell fraction wash liquid.

FIG. 4 is a perspective view of a removable outer rotor portion defining an array of sample analysis cuvettes suitable for use with the invention and in a fast analyzer of the rotary cuvette type.

## DESCRIPTION OF THE PREFERRED EMBODIMENT

Referring now to the drawings, initially to FIG. 1, a rotor assembly made in accordance with the invention is shown nested within a motor driven turntable 1. As shown, turntable 1 is provided with passageways 2 extending from the turntable axis to several points about its periphery. Passageways 2 communicate with a suitable vacuum source for reasons explained below in connection with operation of the subject rotor assembly.

The rotor assembly includes an inner disk-shaped rotor portion 3 defining a circular array (only one shown in FIG. 1) of whole blood sample-receiving chambers 4. Static loading ports 5, extending through the top surface of disk-shaped rotor portion 3, facilitate the direct loading of individual whole blood samples into respective sample-receiving chambers 4 under static conditions. Ports 5 are disposed near the centripetal ends of chambers 4 to avoid overflow of chamber contents through the ports during rotation. Other liquids such as washing or hemolyzing liquids may be dynamically distributed to the entire array of chambers 4 by means of a central dynamic distribution port 6 and a multiplicity of distribution passageways 7 communicating between that port and the centrifugal ends of respective chambers 4. Distribution passageways 7 intersect at the periphery of dynamic distribution port 6 to create a saw-tooth or serrated-edge effect which provides a substantially equal distribution of liquid into passageways 7 when the rotor assembly is rotating and liquid is injected into port 6. Transfer passageways 8 extend from a radially intermediate point along the bottom of each chamber 4, radially inward, upward, and then radially outward to the periphery of inner disk-shaped rotor portion 3. FIG. 3 is cut away to illustrate a passageway 8.

Nested concentrically about inner disk-shaped rotor portion 3 is a removable outer rotor portion 9 defining a plurality of collection chambers 10 for receiving liquids discharged from respective sample-receiving chambers 4. As shown, chambers 10 open in register with the radial extremities of respective passageways 8. Vents 11 extend radially inward and upward from each collection chamber 10 to the top surface of rotor portion 9.

A vacuum annulus 12 is formed above outer rotor portion 9 and the adjoining area of inner rotor portion 3 by means of an annular sealing disk 13 positioned be-

tween upstanding rim 14 of turntable 1 and a raised flange 15 formed on the top surface of inner rotor portion 3. O-rings 16 provide the necessary vacuum seal while permitting removal of disk 13 for replacement of outer rotor portion 9 or removal of the entire rotor assembly from the turntable. As shown, passageways 2 open at the side of annulus 12.

Rotor assemblies made in accordance with the invention are conveniently fabricated by machining cavities and channels into a central plastic disk 17 which is sandwiched between and attached, by cementing for example, to top and bottom cover disks 18 and 19, to form chambers and interconnecting channels. Although the sandwich construction is specifically illustrated with reference to the inner rotor portion 3, outer rotor portion 9 may be constructed in a like manner. All or part of the disks may conveniently be made of transparent plastic to facilitate the observation, using a strobe light for example, of the rotor contents.

FIG. 2 is a plan view of a rotor assembly identical to that illustrated in the perspective view of FIG. 1. As shown in FIG. 2, a multiplicity of sample handling systems comprising sample-receiving chambers 4, associated passageways 7 and 8, and collection chambers 10 are contained in a single rotor assembly, thereby facilitating the simultaneous preparation of separate blood fractions from multiple samples. More or fewer than the eight sample handling systems illustrated may be provided in a single rotor depending on the size of the rotor and the respective chambers used therein.

FIG. 3 illustrates part of a removable outer rotor portion 9' designed for collecting cell fraction wash liquid. As shown, a single annular collection chamber 10' is provided for collecting wash liquid from all of the sample handling systems in a rotor assembly. A single annular opening 21 facilitates the discharge of wash liquid from the transfer passageways into collection chamber 10'. Intermingling of wash liquid from the respective systems is permitted in chamber 10' since the wash liquid is discarded without additional analysis except as needed to determine the need, if any, for additional washing. Vents 11' extend from chamber 10' to the top surface of rotor portion 9'.

FIG. 4 illustrates a removable outer rotor portion 9'' defining an array of cuvettes 10'' suitable for use in a fast photometric analyzer of the rotary cuvette type such as described in U.S. Pat. No. 3,744,974 issued to common assignee on July 10, 1973, in the name of W. L. Maddox et al. Transparent top and bottom plates 18'' and 19'' permit light passage through the cuvettes for photometric analysis in accordance with the teachings of that patent. Vents 11'' extend radially inward and upward from each cuvette to the top of plate 18''.

## ROTOR OPERATION

Using a rotor assembly and turntable substantially as described in reference to FIG. 1, stabilized whole blood samples are first loaded through ports 5 into respective sample-receiving chambers 4 with the rotor at rest using automatic or manual pipetting techniques. Following loading, the turntable and rotor assembly are rotated at about 2,500 rpm until the cells and plasma are well separated. At this point the rotor is slowed to about 1,000 rpm and vacuum applied through passageways 2 to provide a reduced pressure in annulus 12. This causes the plasma in chambers 4 to pass through passageways 8 to respective collection chambers 10. Passageways 8 open within chambers 4 at a radially intermediate position which is calculated to be slightly centripetal to the blood cell-plasma interface for normal blood and specific sample volumes in order to remove most of the plasma fraction without disturbing the blood cell fraction. The rotor is then stopped and the outer rotor portion 9 removed to permit recovery and testing of the respective plasma fractions.

Following recovery of the plasma fractions, outer rotor portion 9 is replaced with an outer rotor portion 9' such as that shown in FIG. 3 and the reassembled rotor rotated at about 2,000 rpm. A selected volume of physiological saline wash solution is then injected into dynamic distribution port 6 causing it to pass in essentially equal volumes through passageways 7 to respective chambers 4 where it mixes with and washes the blood cells remaining in those chambers. Mixing of the saline solution and cells is enhanced by rapid braking and acceleration of the rotor. The cells are then centrifugally resedimented and the wash liquid drawn off into collection chamber 10' by applying vacuum through passageways 2. Vents 11' provide communication between vacuum nnulus 12 and collecton chamber 10', thereby causing reduced pressure in that chamber and the resultant transfer of the saline wash solution from chambers 4 in a manner similar to that used to transfer the plasma fraction to collection chambers 10. The washing step is repeated as needed to achieve the desired cleansing action.

Following cell washing, the rotor assembly is stopped and outer rotor portion 9' replaced with an outer rotor portion having individual collection chambers 10 identical to that used in the collection of plasma fractions. The rotor assembly is then accelerated and lyzing liquid such as distilled water distributed to respective chambers 4 by injecting it into dynamic distribution port 6 in the same manner as the aforementioned wash solution. Hemolysate is recovered by (1) dynamically introducing carbon tetrachloride into port 6 to settle cell debris against the centrifugal end of the chambers 4 and to centripetally displace the hemolysate and (2) applying vacuum through passageways 2 so as to cause the lysate to pass through passageways 8 to respective collection chambers 10. Sufficient carbon tetrachloride can be used to displace the hemolysate to a point where the carbon tetrachloride-hemolysate interface is just centrifugal to the opening of passageways 8 in chambers 4. Alternatively, recovery can be effected by (1) centrifugally compacting cell debris against the centrifugal end of chambers 4, (2) bringing the rotor assembly to a standstill, and (3) applying vacuum through passageways 2.

Where it is desired to photometrically analyze the plasma fractions of the blood samples, an outer rotor portion defining sample analysis cuvettes as shown in FIG. 4 can be used to collect those fractions. The cuvettes can be preloaded with reactants and the entire rotor assembly disposed in a fast analyzer system as referenced above or the outer rotor portion containing the cuvettes transferred to a fast analyzer where both reagent addition and photometric analysis functions are performed. Subsequent washing and lyzing of the cell fraction can be carried out in the manner described above.

Following the above-described washing step, all or part of the washed blood cell fractions may be recovered for testing or storage for future comparison. Such

recovery is effected with the rotor assembly at rest or at low speed by applying vacuum to passageways **2**, thereby causing cells filling the bottoms of chambers **4** to pass through passageways **8**. The cells are collected in respective collection chambers **10** in an outer rotor portion identical to that used to collect plasma fractions. Where only part of the cell fractions is recovered, the remaining cell fractions can be lyzed and the lysate recovered in the manner previously described.

The above description of one embodiment of the invention should not be interpreted in a limiting sense. For example, the exact configuration of chambers **4** and associated passageways **7** and **8** may vary from that illustrated without departing from the invention. Passageways **8** may extend from the sides rather than the bottoms of chambers **4** if transfer of chamber contents under dynamic conditions only is contemplated. It is necessary, however, that passageway **8** extend radially inward to a point centripetal to the maximum centripetal level of sample liquid in chamber **4** to avoid overflow of the sample during rotation. Likewise, passageways **7** and **8** should extend upward to a level sufficient to prevent overflow of sample liquid from chambers **4** under static conditions. It is intended, rather, that the invention be limited only by the scope of the appended claims.

What is claimed is:

**1.** A multiple-sample centrifugal rotor assembly for blood fraction preparation comprising:

a. an inner disk-shaped rotor portion having a top end surface, said inner rotor portion defining:

i. a multiplicity of radially oriented sample-receiving chambers having centripetal and centrifugal ends, said sample-receiving chambers being disposed in a circular array;

ii. a multiplicity of static sample-loading ports, each of said ports communicating between said top end surface and respective sample-receiving chambers;

iii. a centrally located dynamic distribution port open to said top end surface;

iv. a multiplicity of distribution passageways, each of said distribution passageways communicating between said dynamic distribution port and the centrifugal end of a respective sample-receiving chamber; and

v. a multiplicity of transfer passageways, each of said transfer passageways communicating between one of said sample-receiving chambers at a point intermedite its centrifugal and centripetal ends and the radial periphery of said inner rotor portion, each of said transfer passageways extending radially inward from its point of communication with said sample-receiving chamber and then generally radially outward to said radial periphery of said inner rotor portion; and

b. a removable outer rotor portion having an annular configuration nested concentrically about said inner rotor portion, said outer rotor portion defining at least one collection chamber in fluid communication with said transfer passageways in said inner rotor portion for receiving material discharaged from said transfer passageways.

**2.** The rotor assembly of claim **1** wherein said removable outer rotor portion defines a multiplicity of collection chambers, each of said collection chambers having an opening in register with the radial extremity of a respective transfer passageway so as to receive liquids discharged from said transfer passageways.

**3.** The rotor assembly of claim **2** wherein each of said collection chambers is vented through the top surface of said outer rotor portion.

**4.** The rotor assembly of claim **1** wherein said static sample loading ports communicate with the centripetal ends of said sample-receiving chambers.

**5.** The rotor assembly of claim **1** wherein said transfer passageways communicate with the bottom ends of said sample-receiving chambers.

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