

July 26, 1960

L. T. SKEGGS
METHOD AND MEANS FOR THE DETERMINATION OF
CARBON DIOXIDE IN BIOLOGICAL FLUIDS

2,946,665

Filed Sept. 26, 1956

2 Sheets-Sheet 1

FIG. 1

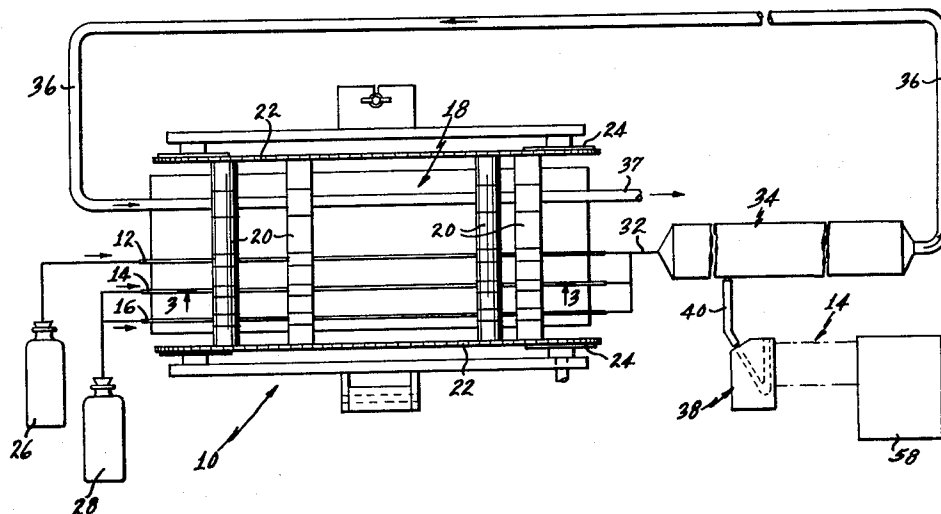
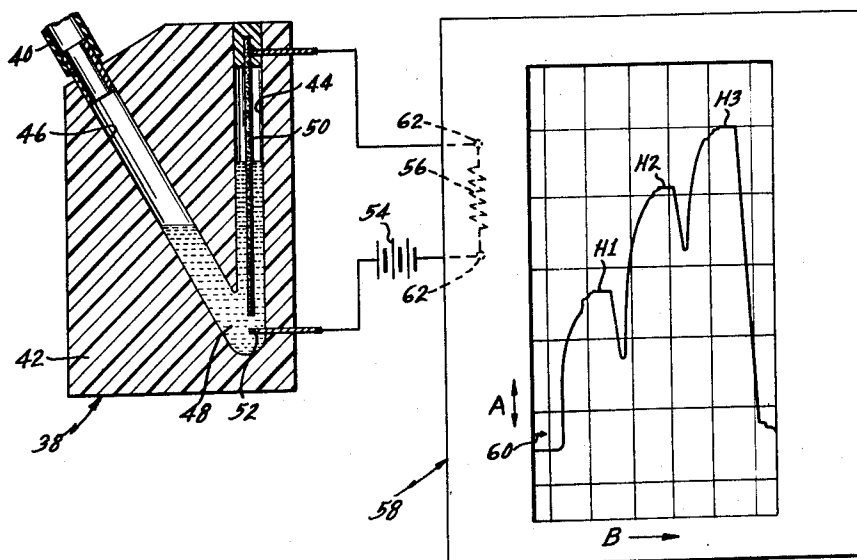


FIG. 2



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FIG. 3

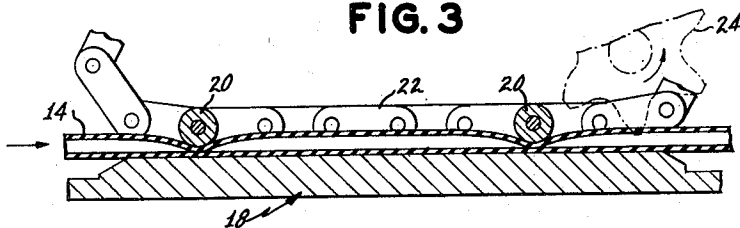


FIG. 4

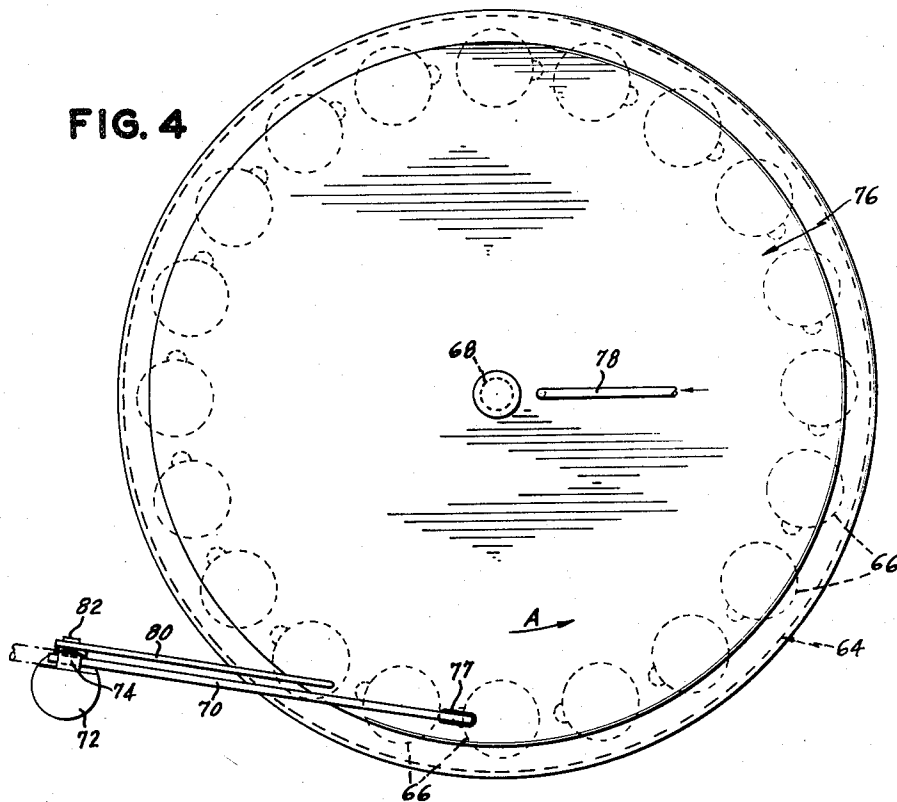
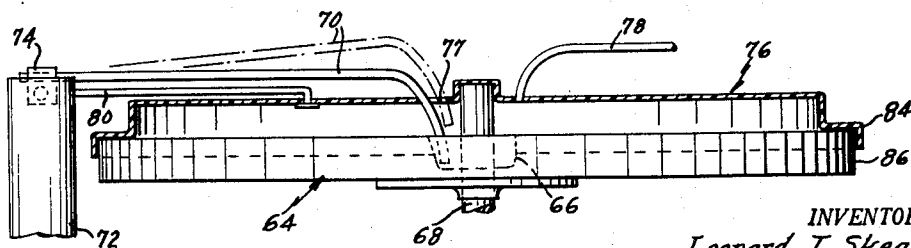


FIG. 5



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1

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METHOD AND MEANS FOR THE DETERMINATION OF CARBON DIOXIDE IN BIOLOGICAL FLUIDS

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11 Claims. (Cl. 23—230)

This invention relates to a method and apparatus for the determination of carbon dioxide in fluid, especially biological fluids.

The primary object of this invention is to provide a method and apparatus by which the quantity of a substance which is present in a liquid and which can be liberated therefrom in gaseous form, for example CO₂ in human blood plasma can be readily and quickly determined.

Another object of the invention is to enable the quantitative analysis of blood in respect to the CO₂ content thereof to be carried out under conditions appropriate to the demands of a busy hospital or biological laboratory.

A further object of the invention is to provide apparatus operable automatically to determine and record automatically in succession the quantity of CO₂ in each of a series of different specimens.

The above and other objects, features and advantages of this invention will be apparent from or further explained in the following detailed description of the presently preferred mode of carrying out the present invention, reference being had to the accompanying illustrative drawings, in which:

Fig. 1 is a more or less diagrammatic view of apparatus embodying the present invention, and is illustrative of the method employed pursuant to this invention;

Fig. 2 is a view, partly in section, showing part of the apparatus;

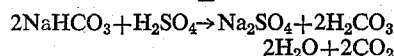
Fig. 3 is a sectional view, on a larger scale, on the line 3—3 of Fig. 1;

Fig. 4 is a top plan view of apparatus for supplying a plurality of plasma samples successively to the apparatus illustrated in Fig. 1; and

Fig. 5 is a side view partly in section, of the supply device illustrated in Fig. 4.

The general procedure heretofore employed for blood analysis is well described in "Practical Physiological Chemistry," by Philip B. Hawk, Bernard L. Oser, and William H. Summerson, twelfth edition, published by The Blakiston Company, Philadelphia, Pa. Briefly and specifically in respect to the analysis for CO₂, as there described the blood sample is centrifuged for separating the plasma and the latter is treated with an anti-coagulant, usually potassium oxalate, and the oxalated plasma sample to be analyzed for CO₂ determination, is subjected to the action of an acid, such as heretofore used in known methods, for example lactic acid, sulphuric acid, etc., as a result of which the CO₂, which is normally present in blood containing the same in a combined form, mainly, i.e., about 95% of the CO₂ in blood plasma, NaHCO₃, is liberated in the form of a gas according to the following equation when sulphuric acid is employed.

2



In accordance with this present invention, gaseous pressure changes in accordance with the quantity of liberated CO₂ in the sample under analysis are utilized for obtaining the quantitative CO₂ indications. The manner in which this is accomplished pursuant to the present invention in its presently preferred form will now be described.

Referring to the drawings, there is shown a pump 10 of the type which comprises resiliently compressible tubing acted upon by pressure rollers or the like to cause fluids to travel through the tubing in the direction of the length of the latter from a source of supply to a point of delivery. In Fig. 1, the resiliently compressible tubing, preferably "Tygon" tubing, which automatically regains its shape or expanded condition upon release of external pressure thereon, is designated by the numerals 12, 14 and 16. This tubing, forming the fluid conduits of the pump 10, are disposed on a bed or platen 18 against which they are pressed progressively along their lengths in the direction of the arrows by rollers 20, carried by and extending between the endless sprocket chains 22 operated by the sprocket wheels 24. The particular pump here shown is more fully described in the copending application of Andres Ferrari, Jr., and Jack Isreeli, Serial No. 463,860, filed October 22, 1954, now Patent No. 2,865,303. It will be understood, however, that any suitable proportioning pump may be used.

The tube 12 is here shown connected to a flask 26 which may be considered as containing the sample of plasma of the blood to be analyzed while the tubing 14 and 16 are shown connected to the flask 28 which may be considered as containing the acid employed for liberating the CO₂, as described above. As here shown, the acid is supplied by two tubes 14 and 16 from flasks 28, but only one tube of the proper internal diameter is necessary, the two tubes being here shown only because the tubing employed is of the same diameter as the tubing 12 for the blood plasma, and it was considered convenient to use such tubing of a single size in the pump illustrated by Fig. 1. The tubings 12, 14 and 16 are joined at the junction 32 where the fluids pumped through said tubes meet and are delivered to a conduit chamber 34. Conduit 34 is preferably incompressible and non-expandable, and may be formed of any suitable, comparatively rigid plastic or other tubing material inert to the fluids flowing therethrough. The latter has a larger cross sectional area than the combined cross sectional areas of the tubings 12, 14 and 16. The discharge end of conduit 34 is connected to the inlet end of the resiliently flexible "Tygon" or other tube 36 of the pump 10. The fluids are discharged from the outlet end 37 of tube 36 and are discarded. Tube 36 must have a greater volumetric pumping capacity and have a greater diameter than the combined diameters of tubes 12, 14 and 16 so that a vacuum is present in conduit chamber 34 during the operation of the apparatus. A partial vacuum or sub-atmospheric pressure in chamber 34 is necessary to achieve a rapid release of the carbon dioxide for increasing the gaseous pressure in said chamber according to the quantity of CO₂ in the plasma. Preferably the vacuum should be about 95%.

A manometer 38 is connected to conduit chamber 34 by a tube 40 which is non-compressible and non-expandable so that, like conduit 34, it remains constant in cross-sectional area during the flow of the fluids through the

apparatus. As illustrated in Fig. 2, the manometer comprises a plastic block 42 having bores 44 and 46 drilled therein providing in effect a U-tube. Manometer liquid, for example mercury, is indicated at 48. An electrode 50 having a higher resistance than mercury is disposed in one arm of the manometer U-tube and extends vertically thereof, terminating in spaced relation to a companion electrode 52. Electrodes 50 and 52 are in series with a source of electric voltage, here shown as a battery 54, and with a resistance 56 across which a voltage drop is developed for operating an electronic control device (not shown) of the recorder schematically illustrated at 58. As a recorder of the indicated type is well known and does not per se form part of the present invention, further description thereof is unnecessary. The stylus or pen of the recorder is schematically illustrated at 60. It will be understood that the stylus or pen 60 is caused to move in opposite directions as indicated by the arrow A, while the chart paper is moved in a transverse path as indicated by the arrow B, the chart paper being moved at a constant rate by a suitable motor conventionally included in a recorder while the excursion of the stylus 60 is determined by the voltage variations impressed on the terminals 62 under the control of the fluid pressure-electric manometer 38.

It is to be observed that the manometer is illustrated as of the closed end capillary mercury type with a substantially complete Torricellian vacuum above the liquid in arm 44 so that normally the mercury tends to rise in the arm 44 of the manometer tube. When the flow of fluids through conduit chamber 34 is induced by the operation of pump 10, the pressure communicated to the arm 46 of the manometer U-tube varies according to the quantity of CO₂ gas liberated from the blood when the acid pumped through the tubes 14 and 16 encounter the blood at the junction point 32. The fluctuation in pressure of the resultant fluid containing liquid and gaseous CO₂ is proportional to the CO₂ content of the blood specimen and induces corresponding variations in pressure which are communicated to the manometer 38. The rise and fall of the mercury in the arm 44 of the manometer U-tube results in corresponding variations in the electric resistance of the electric circuit which includes the battery 54, so that corresponding voltage variations are impressed on the terminals 62 of the control apparatus of the recorder, and of course a record is obtained of these variations. Any other suitable pressure-responsive device may be used in lieu of manometer 38 for controlling an electrical signalling device for providing the signals to the recorder.

Fig. 2 is illustrative of the response of the apparatus. The three high points *h1*, *h2*, and *h3* of the chart illustrated by way of example in Fig. 2 are representative of the quantitative response of the recorder according to three runs of two minutes duration each with three different NaHCO₃ solutions of gradually increasing strength, with an interspaced run of distilled water in lieu of NaHCO₃ of a duration of 1 or 2 minutes between successive two runs of NaHCO₃. Of course, these values are merely illustrative for explanatory purposes in describing the present invention.

The recorder may be calibrated in any suitable way by the use of solutions of sodium bicarbonate in a series of known different concentrations.

It will be understood that a series of blood tests may be made in a very simple manner and at a comparatively rapid rate by intermittently pumping different blood samples and the acid reagent to the junction 32 of the conduit 34, with intervening rinsing operations of the apparatus by applying distilled water to the tube 12, and if necessary, through the acid supply tubes 12 and 14, or through the single acid supply tube when the pump is made to operate with one such tube.

It will be observed that it is not necessary to separately measure the quantity of blood plasma and the

quantity of acid, as the pump is automatically operative to supply the correct amount of acid in accordance with the quantity of blood pumped through the tube 12, said pump being inherently a proportioning pump according to the selected internal diameter of the tubing through which the blood plasma and acid flow, respectively.

In my U.S. application Serial No. 547,087, filed November 16, 1955, for Automatic Analyzing Apparatus, now Patent No. 2,879,141, there is shown a liquid feeding device for supplying a plurality of different samples automatically to an analyzer; such feeding device may be provided in lieu of the flask 26 so that tube 12 will be supplied automatically with different samples of blood in succession and in spaced time relation when pump 10 is operated. Distilled water or other suitable fluid may be introduced into tube 12 automatically by said feed device to space the samples from each other as they flow through tube 12, and the recorder will automatically record the CO₂ contents of the different samples on the chart paper in clearly distinguishable relation on the latter. Thus, as illustrated by Figs. 4 and 5 the supply device comprises a turntable 64 provided with a plurality of wells 66 disposed laterally of each other in a circular row. The turntable 64 is rotated at a suitable speed by a shaft 68 in the direction of the arrow A (Fig. 4) and a sample pick-up tube 70 is pivotally mounted on a suitable stationary post 72 as indicated at 74 so that as table 64 turns the lower end of pick-up tube 70 moves in succession into and out of the several wells 66 to supply the samples seriatim to the tube of pump 10, taking the place of the single sample flask 26 shown in Fig. 1. It will be understood that the shaft 68 may be intermittently rotated by any suitable mechanism (not shown).

When the turntable is to be used it is advisable to make provision to compensate for loss of CO₂. For this reason it is desirable to maintain an atmosphere containing a quantity of carbon dioxide comparable with the quantity thereof in alveolar air, i.e., a 5.5 percent carbon dioxide-air mixture from a suitable tank. For this purpose the supply device is provided with a hood 76 preferably formed of a suitable transparent plastic material. An opening 77 is provided in said hood to accommodate the movement of the pick-up tube 70 during the turning of the turntable 64. The carbon dioxide-air mixture may be supplied to the hood by a tube 78 from a supply tank (not shown). Said hood is supported in stationary position over the turntable 64 in any suitable way as by the bracket arm 80 pivotally mounted on the post 72 as indicated at 82. The carbon dioxide air mixture may be allowed to flow continuously at a low rate into and out of the hood, and it will be understood that the flow of this mixture from the hood may occur without the provision of a special outlet opening inasmuch as a slight clearance will ordinarily exist between the skirt 84 of the hood and the peripheral edge 86 of turntable 64.

Although the invention has been described in detail with reference to the presently preferred mode of practicing the invention, it will be understood by those skilled in the art, in view of the present disclosure, that various changes and modifications may be made without departing from the spirit of the invention and the scope of the appended claims.

Having thus described my invention, what I claim and desire to secure by Letters Patent, is:

1. Apparatus for analyzing a liquid for determining the presence therein of a substance which is capable of being liberated in gaseous form by the addition of a reagent to said liquid, comprising a conduit, pump means for effecting a flow of the liquid and the reagent in predetermined proportions to a junction point at which reaction of said reagent with said substance occurs and which is in fluid-flow communication with said conduit and a flow of the resulting fluid through said conduit oc-

5

curs, pump means connected to said conduit at said reaction point for providing sub-atmospheric pressure at said reaction point, said conduit having an enlarged part through which the fluids are transmitted by said first and last mentioned pump means, and pressure responsive means connected to said enlarged part of said conduit for indicating pressure changes therein during the flow of said fluid through said conduit and thereby indicating the quantity of the liberated gaseous substance.

2. Apparatus for analyzing a liquid for determining the presence therein of a substance which is capable of being liberated in gaseous form by the addition of a reagent to said liquid, comprising a conduit, a proportioning pump for effecting a flow of the liquid and the reagent in predetermined proportions to a junction point at which reaction of said reagent with said substance occurs and which is in fluid-flow communication with said conduit and a flow of the resulting fluid through said conduit occurs, pump means connected to said conduit at said reaction point for providing sub-atmospheric pressure at said reaction point, said conduit having an enlarged part through which the fluids are transmitted by said first and last mentioned pump means, pressure responsive means connected to said enlarged part of said conduit, electrical means operable under the control of said pressure responsive means for producing corresponding electric signals, and means operable under the control of said signals for indicating the pressure in said conduit and thereby indicating the quantity of the liberated gaseous substance.

3. Apparatus for analyzing a liquid for determining the presence therein of a substance which is capable of being liberated in gaseous form by the addition of a reagent to said liquid, comprising a conduit, a proportioning pump for effecting a flow of the liquid and the reagent in predetermined proportions to a junction point at which reaction of said reagent with said substance occurs and which is in fluid-flow communication with said conduit and a flow of the resulting fluid through said conduit occurs, pump means connected to said conduit at said reaction point for providing sub-atmospheric pressure at said reaction point, said conduit having an enlarged part through which the fluids are transmitted by said proportioning pump, a manometer having one arm thereof connected to said enlarged part of said conduit, an electrically conductive liquid in said manometer, a resistance member extending longitudinally of the other arm of said manometer in contact with said manometer liquid, a terminal in contact with said manometer liquid and spaced from said resistance, a terminal connected to said resistance member at a point remote from said first mentioned terminal member so that the resistance between said terminals varies in accordance with the level of liquid in said other arm of the manometer, and means operable in accordance with variations in said resistance for indicating change of pressure in said conduit and thereby indicating the quantity of the liberated gaseous substance.

4. A method of quantitatively analyzing a liquid for the presence therein of substance capable of being liberated therefrom in gaseous form, comprising forming a stream of said liquid in a conduit, adding a reagent thereto in a part of said conduit and providing sub-atmospheric pressure in said part of the conduit for causing said substance to be evolved from the liquid in gaseous form, and measuring the fluid pressure in said part of the conduit during the flow of said fluid therein.

5. A method of analyzing blood for the quantitative determination of carbon dioxide therein, comprising forming a stream of the plasma of said blood in a conduit, adding an acid thereto capable of liberating the carbon dioxide from the blood plasma in gaseous form in a part of said conduit and providing sub-atmospheric pressure in said part of the conduit, causing the resultant fluid comprising the blood plasma and the carbon dioxide in gaseous form to flow through said part of the conduit, and

6

measuring the fluid pressure in said part of the conduit.

6. A method of analyzing blood for the quantitative determination of carbon dioxide therein, comprising forming a stream of the plasma of said blood in a conduit, adding an acid thereto capable of liberating the carbon dioxide from the blood plasma in gaseous form in a part of said conduit and providing sub-atmospheric pressure in said part of the conduit, causing the resultant fluid comprising the blood plasma and the carbon dioxide in gaseous form to flow through said part of the conduit, subjecting a fluid-pressure responsive device to the fluid pressure in said part of the conduit, and operating a recorder under the control of said fluid-pressure responsive device.

7. A method of quantitatively analyzing a liquid for the presence therein of substance capable of being liberated therefrom in gaseous form, comprising forming a stream of said liquid in a conduit, adding a reagent thereto in a part of said conduit and providing sub-atmospheric pressure in said part of the conduit for causing said substance to be evolved from the liquid in gaseous form, and disposing the resultant fluid in fluid pressure relation to a fluid pressure responsive device for measuring the fluid pressure of said resultant fluid.

8. A method of quantitatively analyzing a liquid for the presence therein of substance capable of being liberated therefrom in gaseous form, comprising forming a stream of said liquid in a conduit, adding a reagent thereto in a part of said conduit and providing sub-atmospheric pressure in said part of the conduit for causing said substance to be evolved from the liquid in gaseous form, and disposing the resultant fluid in fluid pressure relation to a fluid-pressure responsive device, and operating a recorder under the control of said fluid-pressure responsive device.

9. A method of analyzing a series of blood plasma specimens for the quantitative determination of carbon dioxide in said specimens, respectively, comprising flowing each specimen through a conduit, one after another in time spaced relation, adding an acid to each specimen in a part of said conduit and providing sub-atmospheric pressure in said part of the conduit while it flows so as to liberate the carbon dioxide therefrom in gaseous form without interrupting the flow of the resultant fluid in the conduit, and subject a fluid-pressure responsive means to the pressure of said resultant fluid of each specimen for measuring the latter as an indication of the carbon dioxide therein.

10. Apparatus for analyzing a liquid for determining the presence therein of a substance which is capable of being liberated in gaseous form by the addition of a reagent to said liquid, comprising a reaction conduit for said liquid and reagent, pump means for transmitting said liquid and said reagent through said conduit, said conduit having a part of enlarged cross section, pressure responsive means connected to said enlarged part of said conduit for indicating change in pressure therein in response to the effect of the addition of said reagent to the liquid, means including a plurality of wells for supplying a plurality of samples automatically in succession to said conduit, means for maintaining a gaseous mixture of predetermined ingredients over the samples in said wells and means for providing sub-atmospheric pressure in said enlarged part of said conduit.

11. Apparatus for analyzing a liquid for determining the presence therein of a substance which is capable of being liberated in gaseous form by the addition of a reagent to said liquid, comprising a conduit for the flow of said liquid and reagent, pump means for transmitting said liquid and said reagent through said conduit, said conduit having a part of enlarged cross section, pressure responsive means connected to said enlarged part of said conduit for indicating change in pressure therein in response to the effect of the addition of said reagent to the liquid, means including a plurality of wells for supplying

a plurality of samples automatically in succession to said conduit, said supplying means comprising a turntable provided with said wells, and means for providing sub-atmospheric pressure in said enlarged part of said conduit.

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