United States Patent

[11] 3,627,424

[72] Inventors **Horton E. Dorman; Charles Soodak, both of Silver Spring; Herbert M. Cullis, College Park; C. David Miller, Greenbelt; Rodolfo Ramiro Rodriguez; Eric W. Youngquist, both of Silver Spring, all of Md.**

[21] Appl. No. **771,911**
[22] Filed **Oct. 30, 1968**
[45] Patented **Dec. 14, 1971**

[73] Assignee **Baxter Laboratories, Inc. Morton Grove, Ill.**

[54] **BACTERIA COUNTER**
21 Claims, 6 Drawing Figs.

[52] **U.S. Cl.** **356/103, 356/167**
[51] **Int. Cl.** **G01n 21/00, G01b 11/00**
[50] **Field of Search** 356/103, 39, 244, 246, 203, 158, 208

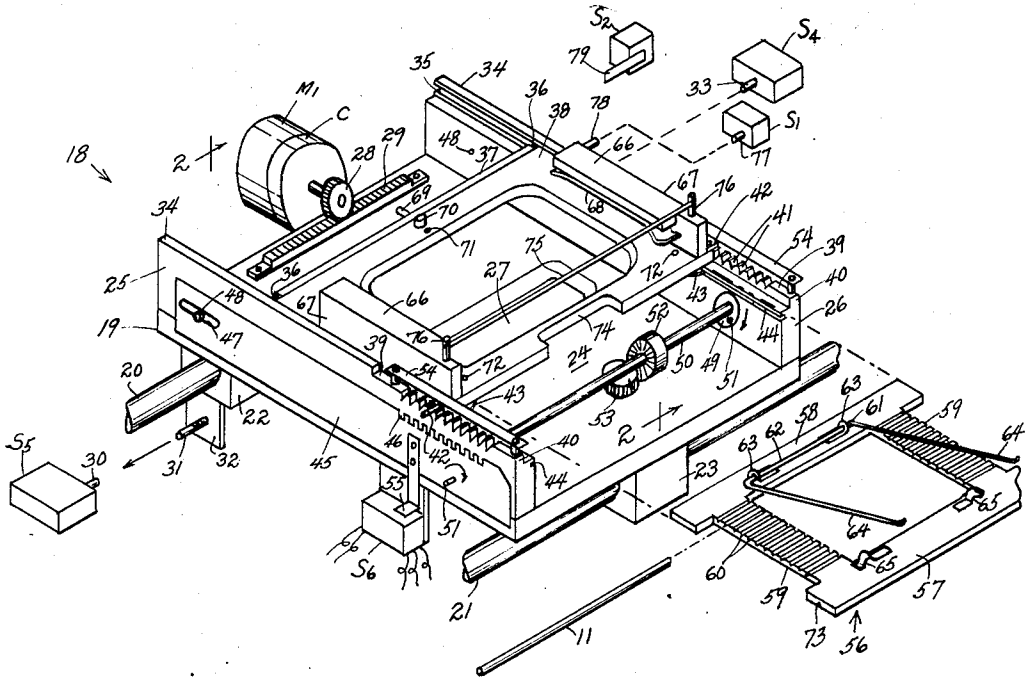
[56] **References Cited**

UNITED STATES PATENTS

2,982,170	5/1961	Wyss	256/39 X
2,825,261	3/1958	Jones	356/244
3,461,280	8/1969	Vitt et al.	356/102 X

Primary Examiner—Ronald L. Wibert
Assistant Examiner—Conrad Clark
Attorney—Herman L. Gordon

ABSTRACT: An automated device for detecting measuring and counting growing microcolonies of bacteria by their light-scattering property. The growing bacteria are contained in glass capillary tubes filled with nutrient agar and are counted by passing the capillary tubes through a narrow beam of light. The tubes are mounted in a holder which is, in turn, detachably secured on a carriage. The carriage is translated in a direction lengthwise of the tubes and then sequentially moved transversely in a step equal to the distance between adjacent tubes. Transverse stepping takes place at the end of the longitudinal travel of each tube. A fixed light source and associated stationary optical means projects a narrow light beam which intersects the tubes as they translate. The presence of growing bacterial microcolonies causes light scattering which is detected in the form of pulses of light by a photomultiplier tube located opposite the path of translation of the tubes relative to the light source. The corresponding signal pulses produced by the photomultiplier tube are counted by an electronic counting circuit which provides a fast count of the particles scanned.



PATENTED DEC 14 1971

3,627,424

SHEET 1 OF 4

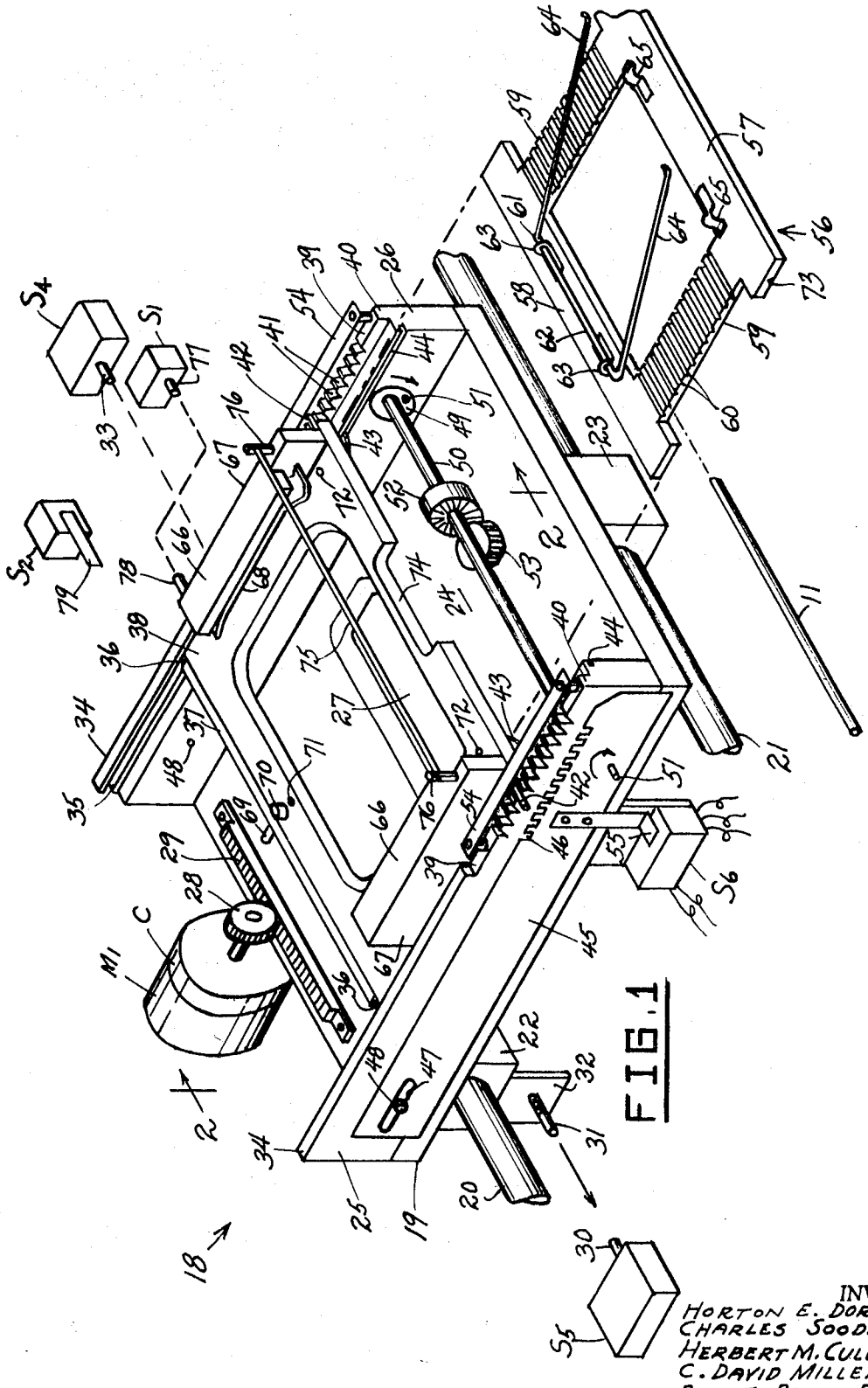
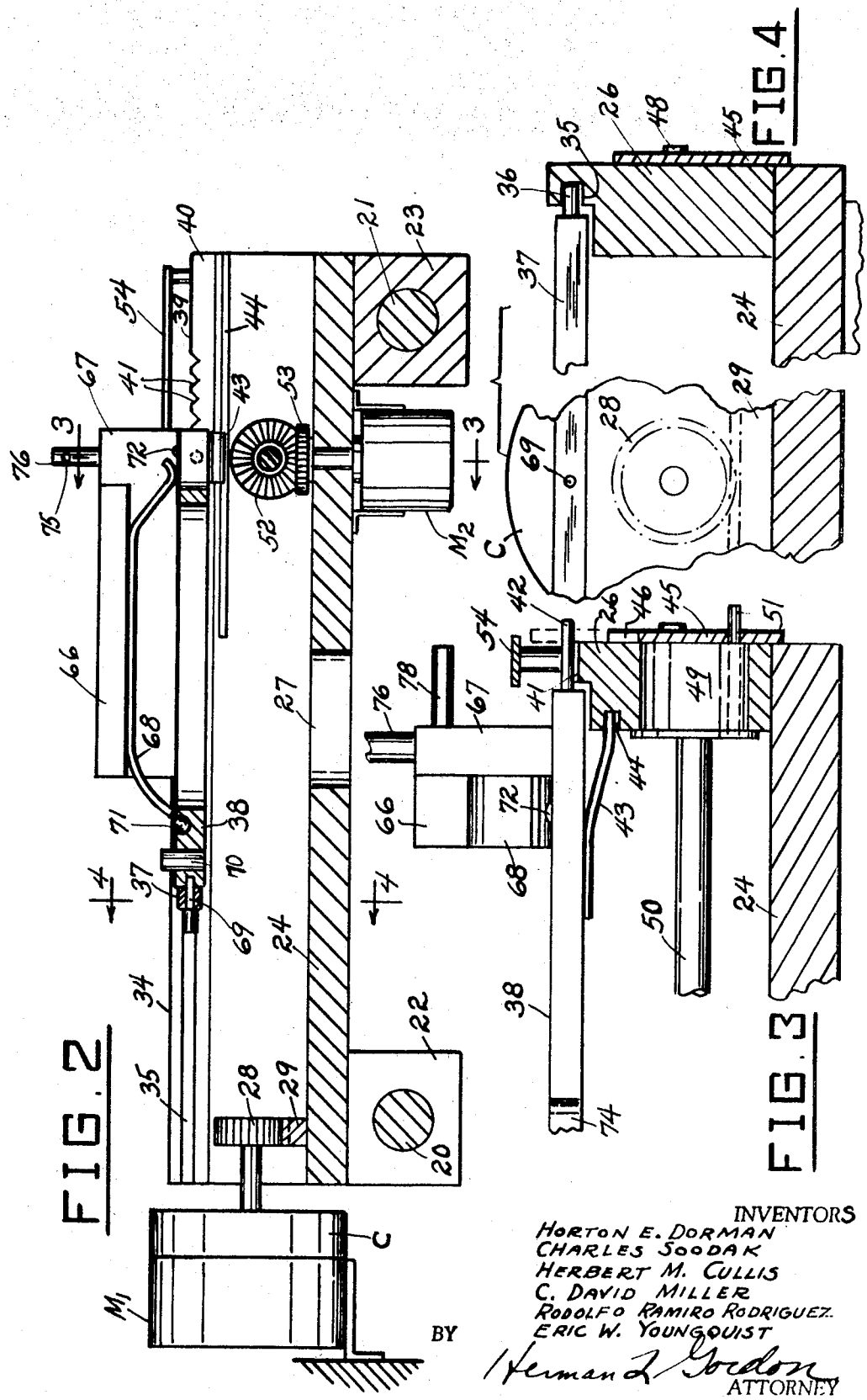


FIG. 1

INVENTORS
HORTON E. DORMAN
CHARLES SOODAK
HERBERT M. CULLIS
C. DAVID MILLER
RODOLFO RAMIRO RODRIGUEZ
ERIC W. YOUNGQUIST

BY Herman L. Gordon
ATTORNEY



INVENTORS
HORTON E. DORMAN
CHARLES SOODAK
HERBERT M. CULLIS
C. DAVID MILLER
RODOLFO RAMIRO RODRIGUEZ
ERIC W. YOUNGQUIST

BY Herman L. Gordon
ATTORNEY

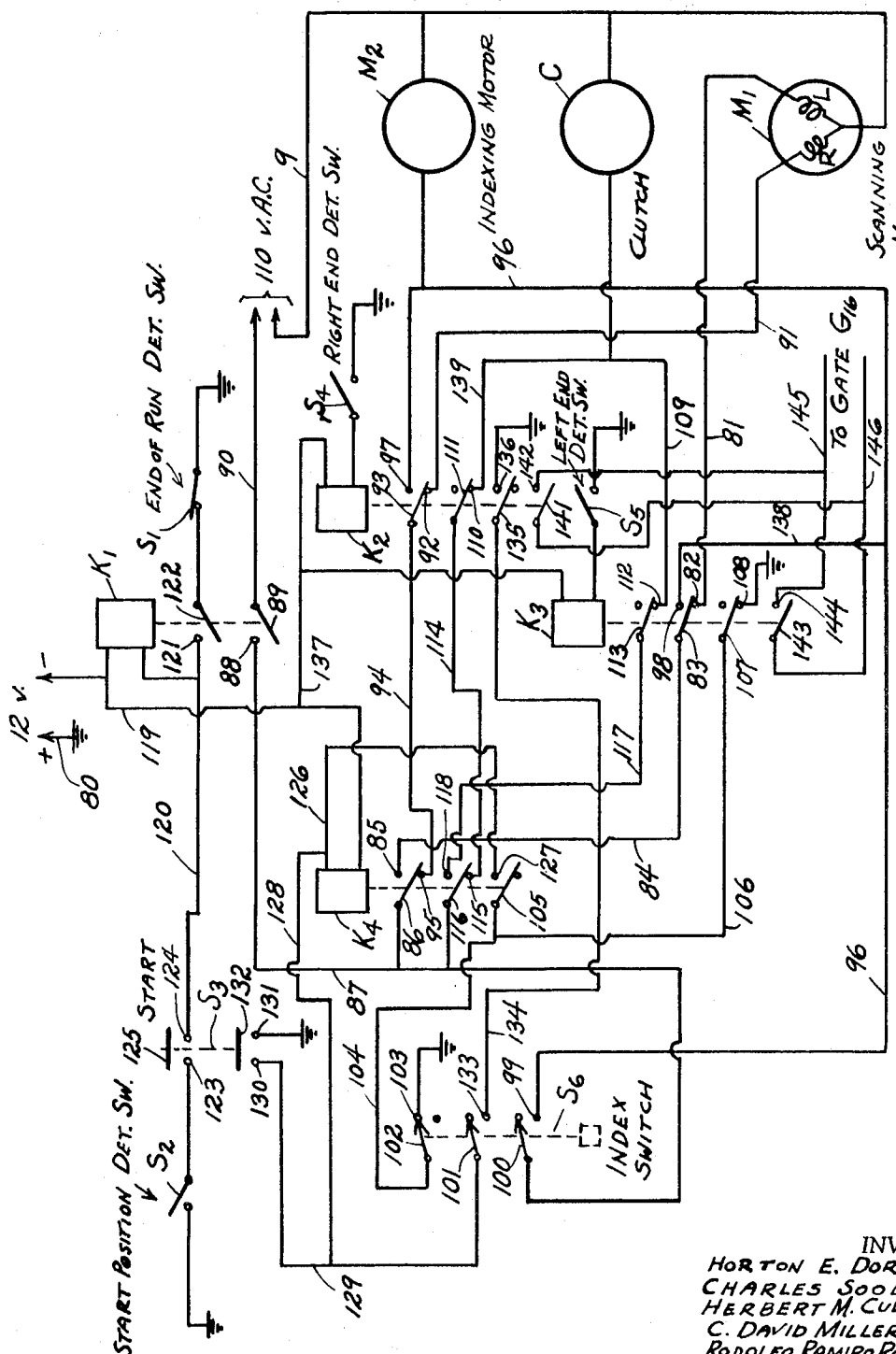


FIG. 5

INVENTORS
HORTON E. DORMAN
CHARLES SOODAK
HERBERT M. CULLIS
C. DAVID MILLER
RODOLFO RAMIRO RODRIGUEZ
ERIC W. YOUNGQUIST

BY Herman L Gordon
ATTORNEY

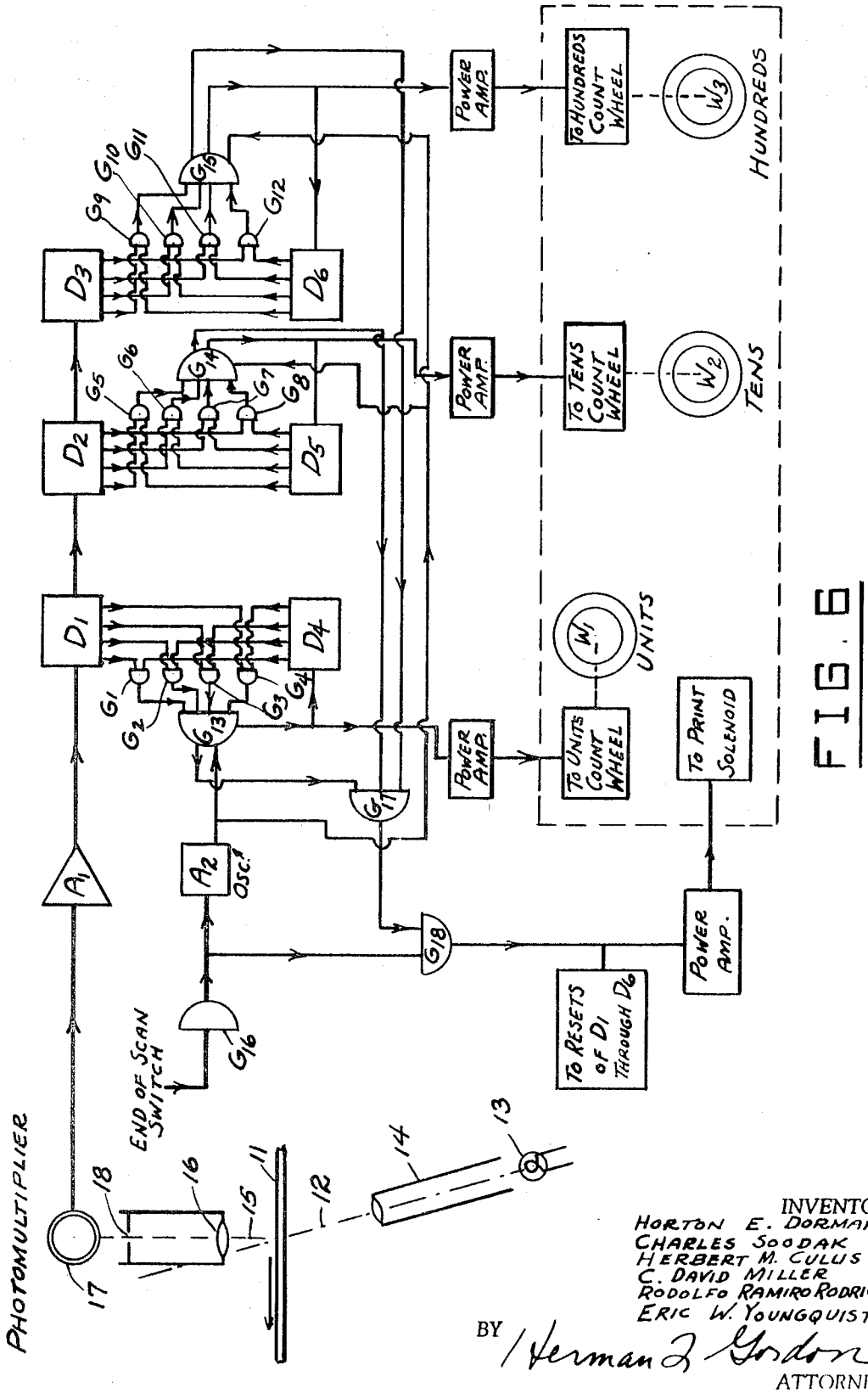


FIG. 6

INVENTORS
HORTON E. DORMAN
CHARLES SOODAK
HERBERT M. CULUS
C. DAVID MILLER
RODOLFO RAMIRO RODRIGUEZ
ERIC W. YOUNGQUIST

BY Herman J. Gordon
ATTORNEY

BACTERIA COUNTER

This invention relates to bacteria counting devices, and more particularly to an apparatus for scanning and counting bacterial microcolonies contained in capillary tubes or equivalent elongated containers.

A main object of the invention is to provide a novel and improved bacteria counting device which is relatively simple in construction, which is reliable in operation, and which makes it possible to assay viable micro-organisms rapidly and to determine their sensitivity to specific antibiotics or other materials.

A further object of the invention is to provide an improved automatic scanning, counting and memory system which can be employed to identify and measure proliferation of bacteria after relatively brief periods of incubation, the system involving relatively inexpensive components, being relatively compact in size, and eliminating many of the time-consuming, repetitious and tedious steps involved in conventional clinical laboratory assay procedures.

A still further object of the invention is to provide an improved automated bacteria counting apparatus wherein micro-organisms are narrowly arrayed in agar culture mediums in transparent capillary containers and are exposed to a beam of light which produces light scatter pulses corresponding to growing colonies of the micro-organisms, the apparatus being arranged to sequentially rapidly translate the containers in paths intersecting the light beam, to detect the light scatter pulses, and to accurately count the pulses electronically.

Further objects and advantages of the invention will become apparent from the following description and claims, and from the accompanying drawings, wherein:

FIG. 1 is a perspective view, partly diagrammatic, of a transport mechanism for moving capillary tubes containing bacteria colonies, as employed in an apparatus constructed in accordance with the present invention.

FIG. 2 is an enlarged vertical cross-sectional view taken substantially on line 2—2 of FIG. 1.

FIG. 3 is a fragmentary vertical cross-sectional view taken substantially on line 3—3 of FIG. 2.

FIG. 4 is a fragmentary vertical cross-sectional view taken substantially on line 4—4 of FIG. 2.

FIG. 5 is an electrical wiring diagram showing the motor control circuit of the transport mechanism of FIGS. 1 to 4.

FIG. 6 is a block diagram of a counting circuit employed with the transport mechanism of FIGS. 1 to 5 and forming part of an improved bacteria counting apparatus according to the present invention.

A photometric method of assaying bacteria by counting light scatter pulses from developing colonies in a capillary tube is described in a recent article by Robert L. Bowman, Philip Blume and Gerald G. Vurek appearing in "Science," Vol. 158, Oct. 6, 1967, entitled "Capillary-Tube Scanner for Mechanized Microbiology." This article describes a generalized procedure, performed manually, wherein a capillary tube 11 (see FIG. 6) containing developing bacterial colonies in agar along with various nutrients is translated in the path of a narrow light beam 12 from a light source 13. The light is focused by a suitable optical system 14. The presence of proliferating bacterial colonies in the capillary tube causes scattering of the beam, and scattered rays in a beam 15 pass through a microscope objective 16 and are directed thereby to a photomultiplier tube 17 through a slit 18 located at the image plane of the microscope objective 16. Thus, the photomultiplier tube 17 "sees" the microcolonies in the form of pulses of scattered light as the capillary tube 11 translates longitudinally through the incident beam 12. This general technique is employed in the apparatus of the present invention.

The above-mentioned article points out the desirability of an automated mechanism for scanning the capillary tubes and for efficiently counting the photomultiplier signal pulses when their amplitude exceeds a predetermined threshold. Said article suggests the use of a synchronous motor-driven carriage to translate the capillary tube through the incident light beam

and the recording of the scatter pulses on a chart, whereby the recorded charts or accumulated counts of successive scans of a capillary tube could be compared directly. The article mentions the sue of a plurality of holders supporting respective capillary tubes, the holders being designed to be replaced (manually) precisely on the carriage without disturbing the orientation of the capillary tubes in their holders.

It is an aim of the present invention to provide a means of automatically shifting the next capillary tube into scanning position as scanning of a preceding tube is completed and to provide a cycle wherein a relatively large number of capillary tubes are successively scanned. It is a further important aim of the present invention to provide a means of automatically digitally recording the total count of the scatter pulses.

The above-described technique and the apparatus of the present invention facilitates early and more successful therapy in cases of acute bacterial infection by making it possible to assay viable micro-organisms rapidly and to determine their sensitivity to specific antibiotics. The apparatus can be used to identify proliferation of bacteria after relatively brief periods of incubation without the heretofore employed time-consuming, repetitious and tedious steps, such as those involved where petri dishes are employed as culture vessels and the microcolonies are developed in planar array on the surfaces of the petri dishes.

The above technique and the apparatus of the present invention offer a way to greatly reduce the time previously required for physicians to obtain biological data necessary to determine and begin optimal treatment for individual patients. For example, specimens for assay are diluted with measured amounts of melted agar and introduced into capillary tubes, which are then sealed, placed in a holder, mounted on a scanning carriage, and then moved sequentially through the intersecting light beam. All colonies or particulate debris within the capillary tubes act as scattering points to produce light scatter pulses. These pulses are processed through a suitable amplifier system which delivers the accepted pulses corresponding to colony counts to an electronic pulse counter.

Since individual bacteria colonies retain their positions in the agar, only those organisms that show growth will produce increased light scatter pluses when rescanned after incubation. By adding antibiotics to the agar in different samples, proliferation or nonproliferation of organisms similarly will show resistance or susceptibility to the respective preparations.

Once an initial light scatter record is obtained, the specimen is incubated and a new recording is taken. Repeat scanning demonstrates additional pulses due to proliferated bacteria, and makes it possible to identify and subtract nonincreased pulses caused by inert particles.

Referring to FIGS. 1 to 4 of the drawings, 18 generally designates a capillary tube transport mechanism constructed in accordance with the present invention. The mechanism 18 comprises a rectangular carriage 19 slidably mounted on a pair of parallel stationary rails 20 and 21 fixed to a suitable supporting structure, not shown. Thus, the carriage 19 may be provided with a pair of depending rear corner blocks 22 which have aligned bores slidably receiving the rear rail 20 and with a depending front intermediate block 23 which has a bore slidably receiving the front rail 21. The carriage 19 is thus slidable in either direction on the parallel rails 20, 21.

The carriage 19 comprises the horizontal rectangular main wall 24 and the upstanding opposite sidewalls 25 and 26. Main wall 24 is formed with an aperture 27 located above the lamp 13 and the optical assembly 14 (not shown in FIGS. 1 to 4) to allow passage therethrough of the scanning beam 12 of FIG. 6.

Designated at M_1 is a conventional reversible electric motor having the respective "right" and "left" operating windings shown diagrammatically at R and L in FIG. 5. Motor M_1 is mounted on a suitable stationary support, not shown, and is coupled through a conventional electrically controlled clutch C to a pinion gear 28 which is drivingly engaged with a rack

bar 29 secured on the rear marginal portion of main wall 24 and which extends parallel to rails 20, 21. When either the R or L motor winding and the clutch C are simultaneously energized, the carriage 19 is driven either to the right or left on the rails 20,21, depending on which of the motor windings R or L is energized. As will be presently described, the translation of the carriage is limited by the provision of respective right and left stationary limit switches S_4 and S_5 which close at the ends of the respective right and left excursions of the carriage and which act to cause reversal of movement of the carriage. Thus, the normally open limit switch S_5 is mounted on an adjacent stationary portion of the associated supporting frame, the operating plunger 30 of said switch being axially aligned with and in the path of movement of a pin 31 secured to a lug 32 depending from the left corner block 22. A similar pin is provided at the right side of the carriage 19, axially aligned with and engageable with the operating plunger 33 of the stationary normally open limit switch S_4.

As shown in FIG. 5, the limit switches S_4 and S_5 are connected in the energizing circuits of respective relays K_2 and K_3.

The top rear edge portions of sidewalls 25 and 26 are formed to define reduced upstanding ribs 34,34 which are grooved at 35 to define opposing inwardly facing channels. Pivotally and slidably received in said channels are respective pins 36,36 fixed to and projecting from the opposite ends of a bar 37 which is, in turn, pivotally secured to the rear edge of a rectangular platelike frame member 38. The sidewalls 25 and 26 are recessed at their forward top edge portions, as shown at 39,39 and are formed thereat with reduced ribs 40,40. Said ribs are formed at their intermediate portions with aligned serrations 41 defining seats for at times receiving oppositely projecting front corner pins 42,42 provided on the platelike frame member 38.

As shown in FIG. 2, bar 37 is pivotally connected at its midpoint to the midpoint of the rear edge of member 38 by a pivot pin 69.

Pins 42,42 are rigidly secured in and project from the forward portions of the respective side edges of member 38, and can be successively engaged in aligned pairs of V-shaped notches of the serrations 41, thereby providing positive positioning of member 38 on carriage 19. As will be presently explained, this provides positive identical steps of transverse movement of the capillary tubes, whereby to insure that the incident beam 12 will always intersect a capillary tube for proper scanning thereof.

The forward portion of frame member 38 is biased downwardly by laterally projecting leaf springs 43,43 secured to the bottom surface of its forward corner portions, the outer ends of the leaf springs being slidably received in horizontal guide grooves 44 formed in the sidewalls 25 and 26.

The transverse stepping mechanism for the frame member 38 comprises respective sideplates 45,45 slidably and pivotally mounted on the sidewalls 25 and 26 and having notches 46 at the forward portions of their top edges engageable with the pins 42 to lift them out of one pair of aligned serrations 41 and move them into an adjacent pair of aligned serrations 41. The rear portions of the sideplates 45 are formed with longitudinal slots 47 through which extend headed pins 48 secured respectively in the sidewalls 25 and 26, whereby the plates 45 are slidably and pivotally connected to said sidewalls. Respective cylindrical crank members 49,49 are rotatably mounted in the forward portions of the sidewalls 25 and 26 and are rigidly secured to the opposite ends of a common indexing drive shaft 50. The crank members 49 have pins 51 engaged in apertures in the respective sideplates 45,45, whereby said sideplates are simultaneously translated and oscillated responsive to the rotation of shaft 50. The amplitude of such translation and oscillation is sufficient to elevate the pins 42,42 out of one aligned pair of serrations 41 and step them into the next pair of aligned serrations 41 with each revolution of shaft 50. As shown in FIG. 1, indexing shaft 50 drives pins 51 in a clockwise direction. Said shaft is provided at its intermediate portion with a radial-toothed gear 52 which is meshingly engaged by a pinion gear 53 carried on the shaft of an indexing motor M_2 mounted beneath and secured to the main wall 24 of carriage 19.

Respective horizontal retaining bars 54,54 are secured in the recesses 39,39 overlying the serrations 41 but spaced sufficiently above said serrations to provide adequate clearance for the stepping of pins 42,42 from one pair of aligned serrations to the next pair as above described.

A limit switch S_6 is mounted on the associated carriage plate structure in a position such that its operating element will be engaged by a lug 55 secured to the sideplate member 45 of sidewall 25 and will operate the switch after approximately 200° of clockwise rotation of its pin 51 from its lowermost position shown in FIG. 1. The switch S_6 will be released shortly after the pin 51 completes 360° of clockwise rotation.

Designated generally at 56 is a capillary tube holder arranged to be releasably mounted on the frame member 38 in a precisely established position on said frame member. The holder 56 comprises a flat frame having front and rear arms 57 and 58 and sidearms 59,59 formed with aligned pairs of serrations or grooves 60 adapted to receive capillary tubes 11 and to position said tubes parallel to each other and to the front and rear arms 57 and 58. A U-shaped resilient wire bail 61 has its bight portion 62 pivotally connected by apertured hinge lugs 63,63 to the rear arm 58. The sidearms 64 are clampingly engageable with the capillary tubes 11 positioned in the grooves 60, and are lockingly retained in spring clips 65,65 mounted on front arm 57.

The holder 56 is engageable on the member 38 with the ends of the front and rear arms 57 and 58 received beneath inwardly projecting flanges 66,66 provided on upstanding sideplates 67,67 secured on the side marginal portions of member 38. Bowed leaf springs 68,68 are respectively secured to the undersides of flanges 66,66 with their depending ends clampingly engageable with the ends of the front and rear arms 57 and 58 of the holder 56 when said holder is inserted. The rear arm of member 38 is provided with an upstanding stop pin 70 and with a positioning ball 71 spaced forwardly thereof. Similar positioning balls 72,72 are provided in the forward corner portions of member 38. Grooves 73 are provided in the holder 56 to receive the positioning balls 71 and 72, with the rear edge of the holder in abutment with stop pin 70, whereby to constrain the holder 56 to occupy a fixed precise location on the member 38.

The grooves 73 may comprise respective V-grooves extending perpendicular to and intersecting their adjacent edges. Thus, then the positioning balls 71 and 72 are seated in their corresponding V-grooves, the holder 56 is positively positioned in an accurately reproducible position on member 38. This is necessary in order to insure that the scanning beam 12 will properly impinge on each of the tubes 11 carried by any holder inserted on the member 38 without the necessity of otherwise aligning or adjusting the holder when it is inserted.

The front edge of member 38 is notched away at its intermediate portion, as shown at 74, to facilitate grasping the rear arm 57 of holder 56 in inserting or removing the holder.

A cross rod 75 is secured between upstanding studs 76,76 mounted on the forward end portions of sideplates 67,67, said cross rod serving as a handle for at times manually positioning the transversely movable carriage member 38.

It will be noted that the pivot pin 69 in cooperation with bar 37, and the engagement of the springs 43 in the grooves 44, provide a three-point suspension of the member 38 on the main body of the carriage assembly 19.

A normally closed limit switch S_1 is mounted on the associated adjacent fixed structure, not shown, with its operating element 77 in the path of movement of a pin 78 projecting from the upstanding plate 67 carried on the margin of member 38 adjacent sidewall 26. As will be presently described, pin 78 engages element 77 at the end of a run, and thereby opens switch S_1, which deactivates the mechanism.

Another limit switch S_2 is likewise suitably mounted with its operating lever 79 in a position to be engaged by a portion of

member 35, for example, the rear end of the right flange 66, when the transport mechanism 18 is in starting position. In starting position the switch S_2 is closed and the mechanism can be started, as will be presently described. Starting cannot take place unless switch S_2 is closed.

In the starting position, namely, with switch S_2 closed, the beam 12 will impinge on the left end portion of the foremost capillary tube carried on the holder 56 seated on member 38. At the end of the run, namely with switch S_1 open, the beam 12 will impinge on the left end portion of the rearmost capillary tube 11 mounted on the holder.

The various switch-operating pins 78, 31, etc., are threadedly engaged with their supports, so that they can be readily adjusted to provide proper timing of the switching actions performed thereby.

As shown in FIG. 5, the control circuit for the motors M_1, M_2 and clutch C includes the various limit switches S_1, S_2, S_4, S_5 and S_6, a two-pole manually operated pushbutton switch S_3, and respective relays K_1, K_2, K_3 and K_4.

Indexing limit switch S_6 is of the three-pole, double-throw type, whose poles normally engage their associated upper contacts, as shown.

Again referring to FIG. 5, it will be seen that the R and L windings of the scanning motor M_1 have a common junction, and said common junction is connected to the AC supply wire 9. The remaining terminal of the L winding is connected by a wire 81, a bottom contact 82 and the associated pole 83 of the manually deenergized relay K_3, and a wire 84 to an upper contact 85 of the normally deenergized, relay K_4. The associated pole 86 is connected to a wire 87, which is in turn connected through the normally open contacts 88–89 of the normally deenergized relay K_1 to the other supply wire 90 connected to a 110-volt AC source. Thus, the L winding will be energized responsive to the simultaneous energization of relays K_1 and K_4.

The remaining terminal of the R winding is connected by a wire 91, a bottom contact 92 and the associated pole 93 of the normally deenergized relay K_2, and a wire 94 to the bottom contact 95 of relay K_4 normally engaged by pole 86. Thus, the R winding will be energized responsive to the energization only of relay K_1, with relays K_2 and K_4 deenergized.

One terminal of indexing motor M_2 is connected to wire 9. The other terminal thereof is connected to a wire 96. Wire 96 is connected to the upper contact 97 of relay K_2 associated with pole 93, to the upper contact 98 of relay K_3 associated with pole 83, and to the lower contact 99 associated with a pole 100 of the three-pole indexing limit switch S_6. In their normal positions, the respective poles 100, 101 and 102 of switch S_6 engage their upper contacts, as shown. The upper contact 103 associated with pole 102 is grounded. Pole 102 is connected by a wire 104 to a pole 105 of relay K_4 and by a further wire 106 to a pole 107 of relay K_3, normally engaging a grounded lower contact 108.

Clutch C has one terminal connected to wire 9. The other terminal thereof is connected to a wire 109, which in turn is connected to a lower contact 110 of relay K_2, normally engaged by a pole 111, and to a lower contact 112 of relay K_3 normally engaged by a pole 113. Pole 111 of relay K_2 is connected by a wire 114 to a lower contact 115 of relay K_4 normally engaged by a pole 116, which is connected to wire 87. Pole 113 of relay K_3 is connected by a wire 117 to the upper contact 118 of relay K_4 associated with pole 116.

One terminal of the winding of each of relays K_1, K_2, K_3 and K_4 is connected to the negative 12-volt DC supply wire 119. The remaining terminal of the winding of relay K_1 is connected to a wire 120. Wire 120 is connected through the upper contacts 121–122 of relay K_1 and normally closed limit switch S_1 to ground. Also, wire 120 is connected through the upper contacts 123–124 and pole 125 of a two-pole pushbutton operated switch S_3 and normally open limit switch S_2 to ground.

The remaining terminal of the winding of relay K_2 is connected to ground terminal the normally open right-end limit switch S_4. The remaining terminal of the winding relay K_3 is connected to ground through the left-end limit switch S_5.

The remaining terminal of the winding of relay K_4 is connected by a wire 126 to the upper contact 127 of relay K_4 associated with pole 105, and said wire 126 is connected by a further wire 128 to a wire 129, which is connected to ground through the lower contacts 130–131 and pole 132 of pushbutton switch S_3. Wire 129 is also connected to a pole 101 of indexing switch S_6. Pole 101 is engageable with a contact 133, which is connected by a wire 134 to the lower pole 135 of relay K_2. When relay K_2 is energized, pole 135 is engageable with a grounded upper contact 136.

In operation of the circuit of FIG. 5, let it be assumed that a holder 56 carrying 12 capillary tubes 11 has been loaded on the transport mechanism 18 and that the carriage 19 is in its starting position and that the transversely movable transport member 38 is in its rearmost position, namely, with the pins 42,42 in the rearmost pair of aligned serrations 41. Under these conditions, switch S_2 is closed. When pushbutton switch S_3 is actuated, relay K_1 becomes energized by a circuit comprising negative supply wire 119, the winding of relay K_1, wire 120, switch contacts 124–125–123, switch S_2 and ground. Relay K_1 closes its holding contacts 121–122, thereby connecting wire 120 to ground through normally closed limit switch S_1. Relay K_1 thus remains energized until the end of the run.

The closure of contacts 130–132–131 of switch S_3 energizes relay K_4 through a circuit comprising negative supply wire 119, the winding of relay K_4, wire 128, wire 129, contacts 130–132–31 and ground. The holding contacts 105–127 of relay K_4 close and keep wire 126 grounded through wire 106 and contacts 107–108 of relay K_3 as long as said relay K_3 is deenergized.

Clutch C is energized through a circuit comprising line wire 90, contacts 88–89 of relay K_1, wire 87, contacts 116–118 of relay K_4, wire 117, contacts 113–112 of relay K_3, wire 109, the clutch winding, and wire 9.

The L winding of motor M_1 is energized through a circuit comprising line wire 90, contacts 88–89, wire 87, contacts 86–85 of relay K_4, wire 84, contacts 83–82 of relay K_3, wire 81, winding L and wire 9. The carriage 19 is then moved to the left by motor M_1, releasing switch S_2.

At the end of the leftward scan of the first (foremost) capillary tube 11, limit switch S_5 closes and energizes relay K_3 by a circuit comprising negative supply wire 119, a wire 137, the winding of relay K_3, switch S_5 and ground. Clutch C is deenergized by the opening of contacts 113–112 and the L motor winding is deenergized by the opening of contacts 83–82. The holding circuit of relay K_4 is retained through wire 104 and contacts 102–103 of switch S_6.

Indexing motor M_2 is energized through a circuit comprising line wire 90, relay contacts 88–89, wire 87, contacts 86–85 of relay K_4, wire 84, contacts 83–98 of relay K_3, a wire 138, wire 96, the winding of motor M_2 and wire 9.

Motor M_2 rotates shaft 50 clockwise, as above described, and after approximately 200° of rotation of pins 51 (by which time the pins 42,42 have been transferred forwardly to the next pair of aligned serrations 41,41) lug 55 actuates limit switch S_6. This deenergizes relay K_4 by the opening of contacts 102–103, but motor M_2 continues to run, since contacts 100–99 remain closed, until shortly after 360° of rotation of shaft 50, whereupon contacts 100–99 open and deenergize motor M_2.

The deenergization of relay K_4 energizes the R winding of motor M_1 by a circuit comprising line wire 90, relay contacts 88–89, wire 87, contacts 86–95 of relay K_4, wire 94, contacts 93–92 of relay K_2, wire 91, the R winding, and wire 9. Clutch C is likewise energized by the closure of contacts 115–116 of relay K_4 by a circuit comprising wire 90, contacts 88–89, wire 87, contacts 116–115, wire 114, contacts 111–110 of relay K_2, a wire 139, the clutch winding, and wire 9. Motor M_1 thus moves carriage 19 rightwards, with the second capillary tube 11 in the path of scanning beam 12.

At the right end of the scanning travel forward step, during carriage **19**, limit switch S_4 is closed, energizing relay K_2 through a circuit comprising negative supply wire **119**, wire **137**, the winding of relay K_2, switch S_4 and ground. This opens contacts **111–110** and **92–93**, deenergizing motor M_1. Indexing motor M_2 is again energized by the closure of relay contacts **93–97**, by a circuit comprising line wire **90**, contacts **88–89**, wire **87**, contacts **86–95** of relay K_4, wire **94**, contacts **93–97** of relay K_2, wire **96**, the winding of motor M_2, and wire **9**. Another indexing cycle occurs, moving the member **38** through a second forward step, during which relay K_4 becomes energized through a circuit comprising negative supply wire **119**, the winding of relay K_4, wire **128**, wire **129**, contacts **101–133** of switch S_6, wire **134**, contacts **135–136** of relay K_2 and ground. Relay K_4 is held energized by the holding circuit including wire **126**, contacts **105–127**, wire **106**, contacts **107–108** of relay K_3, and ground.

Clutch C is then energized through contacts **116–118** of relay K_4 and the L winding of motor M_1 is likewise energized through contacts **86–85** of relay K_4, repeating the leftward scan for the third capillary tube **11**, as above described for the first capillary tube. The action continues until the completion of the twelfth scan, wherein the carriage **19** completes the last rightward scan, whereby pin **78** opens switch S_1 and releases relay K_1. This opens contacts **88–89** and deactivates the mechanism.

It is thus seen that after a holder **56** carrying the full complement of 12 capillary tubes has been placed in the carriage member **38** as above described, with the parts in starting position, actuation of pushbutton switch S_3 will begin a cycle of operation wherein the 12 capillary tubes **11** are individually linearly scanned at the same rate, and that the cycle will terminate after the 12th tube has been scanned, with the holder **56** in its foremost position, namely, with the pins **42,42** resting in the foremost pair of aligned serrations **41,41**. The holder **56** may then be removed, a new holder may be inserted, the member **38** may be manually returned to its starting position, and the capillary tubes **11** of the new holder may then be scanned by again actuating pushbutton switch S_3.

The light scatter pulses produced during the scanning of each tube **11** cause a corresponding series of signal pulses to be produced by the photomultiplier tube **17** (FIG. **6**). The pulses of each scan are counted and recorded by the counting circuit diagrammatically shown in FIG. **6**. To initiate counting action at the end of each scan, respective pairs of contacts **141–142** and **143–144** of relays K_2 and K_3 close, these contacts being connected in parallel in a switching circuit controlling a gate G_{16} which, in turn, controls the action of the printer and oscillator of FIG. **6**, as will be presently described. Thus, as shown in FIG. **5**, the respective pairs of contacts **141–142** and **143–144** are connected in parallel to the control wires **145** and **146** leading to gate G_{16}, whereby said gate is turned on responsive to the closure of either left-end limit switch S_5 or right-end limit switch S_4, which occurs at the end of each tube scan.

The signals from the photomultiplier tube **17** are delivered (a conventional band-pass amplifier A_1) and emerge as pulses corresponding to counts. The count pulses are tallied from one to 999 by a decade counter D_1–D_2–D_3. This decade counter is of the BCD type, and thus there are four outputs for each decade. Each output is individually compared to the corresponding output of decade counter D_4–D_5–D_6 by gates G_1, G_2...G_{12}. If any output is different from the corresponding output with which it is compared, then the gate G_1 and G_2 or...G_{12}, which compares them, places a signal into gate G_{13} or G_{14} or G_{15} (as is appropriate), allowing a signal from the oscillator A_2 to enter the appropriate decade counter D_4 or D_5 or D_6, and simultaneously advancing count wheel W_1 or W_2 or W_3 (as is appropriate) within the printer. Thus, the count wheel will advance to the numeral appropriate for its decade, at which time oscillator pulses will become blocked by the action of gate G_{13} or G_{14} or G_{15} (as is appropriate), each of which will turn off when each of its four inputs are off.

Oscillator A_2 is a multivibrator producing square waves of frequency appropriate to the speed capabilities of the print wheels. A_2 is furthermore activated by gate G_{16}, which allows oscillations only at the end of the scan of each tube, while no further counts are being generated by amplifier A_1.

When all three count wheels have reached their appropriate numerals, and therefore gates G_{13}, G_{14} and G_{15} are all off (in the zero state), then gate G_{17} turns on, and if gate G_{16} is also on, this causes gate G_{18} to turn on and to cause the printer to "print" and to reset decade counters D_1, D_2...D_6 to zero.

Thus, assume that in normal operation amplifier A_1 delivers, for example, 238 pulses for the scan of some given tube. Then D_1 has the output 1,000, D_2 has the output 0011, and D_3 has the output 0010 in binary counting. At the end of the scan, gate G_{16} turns on, starting oscillator A_2, producing a square wave at, say 30 c.p.s. Gate G_{13} allows this signal to enter gate D_4 and the "units" count wheel system, until D_4 reads 1,000, at which time the "units" wheel reads "eight." At this point, gate G_{13} turns off. The "10's" and "100's" wheels operate similarly. When gates G_{13}, G_{14} and G_{15} are all "off," and gate G_{16} is "on," gate G_{18} causes the printer to print and also causes all the "D" units to clear to zero.

While a specific embodiment of an automatic bacteria counter has been disclosed in the foregoing description, it will be understood that various modifications within the spirit of the invention may occur to those skilled in the art. Therefore it is intended that no limitations be placed on the invention except as defined by the scope of the appended claims.

What is claimed is:

1. In a particle counting apparatus comprising a fixed light source providing a directed scanning beam; a carriage member; means on said carriage member defining a plurality of substantially parallel spaced elongated transparent sample receptacles located so that one of said receptacles is in the path of said scanning beam; means for moving said carriage member longitudinally along the axes of said receptacles thereby to scan each one of said receptacles in one direction to a predetermined carriage position; said moving means comprising a support movably mounting said carriage, fixed guide means extending parallel to said receptacles, means mounting said support on said guide means so as to be constrained to move there along, and means to move said support on said fixed guide means; light-sensitive means located to receive light scattered from particles in said one receptacle as a result of said scanning action; means to move said carriage member transversely through a distance substantially equal to the spacing between adjacent sample receptacles responsive to said predetermined longitudinal positioning of said carriage member thereby to dispose the receptacle next adjacent to said one receptacle to the path of said scanning beam; and means to then move said carriage member longitudinally in a reverse direction whereby said next adjacent receptacle is scanned in said reverse direction.

2. The particle counting apparatus of claim **1** wherein said predetermined longitudinal carriage member position is defined by a location of said carriage member wherein the path of said scanning beam intersects an end portion of said one receptacle whereby upon transverse movement of said carriage member said next adjacent receptacle starting at its corresponding end portion is scanned to its other end.

3. The particle counting apparatus of claim **2**, wherein means are provided to again move the carriage member transversely through an additional distance substantially equal to the spacing between adjacent receptacles responsive to the reverse movement of the carriage member to a location wherein the scanning beam intersects an opposite end portion of said next adjacent receptacle, and means to then again move the carriage member in the first-named longitudinal direction.

4. The particle counting apparatus of claim **3** comprising means to deactivate the carriage-moving means responsive to the completion of the longitudinal scanning movement of the last of the receptacles through the scanning beam.

5. The particle counting apparatus of claim 1 wherein said light-sensitive means includes an electronic device generating an electrical signal responsive to light received thereby, and means counting the electrical signals generated by the light scattered from the respective particles during said scanning action.

6. The particle counting apparatus of claim 5 wherein said counting means includes means to tally said count for said scanning action in digital form.

7. The particle counting apparatus of claim 6, wherein said counting means further includes means to print out said count and means to reset said counting means to zero, and means to simultaneously energize said printout means and said reset means responsive to said predetermined longitudinal positioning of the carriage member.

8. The particle counting apparatus of claim 1 wherein said carriage member is provided with means defining a seat, said receptacle-defining means comprising a holder removably mounted on said seat, said sample receptacles comprising individual transparent tubes, said holder having positioning means engaged with the tubes to lock them in spaced parallel fixed longitudinal positions on said holder, and interengaging positioning means on said holder and said seat constraining said holder to occupy a fixed position on said carriage member.

9. The particle counting apparatus of claim 1 wherein said support includes transversely extending guide means, and means on the carriage member engaged with said transversely extending guide means and constraining the carriage member to move therealong.

10. The particle counting apparatus of claim 9 comprising means providing a sliding and pivotal connection of said carriage member with said transversely extending guide means, said pivotal connection being on an axis parallel to said receptacles.

11. The particle counting apparatus of claim 1 wherein said means to move said carriage member transversely through a distance substantially equal to the spacing between adjacent receptacles comprises crank means on said support rotating on an axis parallel to said receptacles, and interlocking means on said crank means and said carriage member for stepping said carriage member through said distance between adjacent receptacles responsive to each revolution of said crank means.

12. The particle counting apparatus of claim 11 wherein said interlocking means comprises a transverse arm, means slidably and pivotally connecting said arm to said support, means drivingly and pivotally connecting said arm to said crank means whereby said arm is translated and oscillated responsive to rotation of said crank means, a lateral projection on said carriage member, and means on said arm drivingly engageable with said projection during a portion of the translation and oscillation of said arm.

13. The particle counting apparatus of claim 12, wherein said arm is provided at its top edge with a series of drive notches in which said projection is sequentially receivable responsive to successive revolutions of said crank means.

14. The particle counting apparatus of claim 13, wherein said support has a transverse wall element underlying said projection, the top edge of said wall element being formed with a series of serrations spaced apart substantially the same distance as said receptacles, said projection being receivable successively between adjacent serrations to provide registry of a receptacle with the scanning beam when said receptacle is stepped into scanning position.

15. The particle counting apparatus of claim 11, wherein said crank means is provided with a driving shaft and an indexing electric motor drivingly connected to said driving shaft.

16. The particle counting apparatus of claim 15, wherein said indexing motor has an energizing circuit including switch means closing responsive to said predetermined longitudinal positioning of said carriage member.

17. The particle counting apparatus of claim 16, wherein said means to move said support on said fixed guide means comprises a reversible electric motor drivingly coupled to said support, said motor having respective forward and reverse windings, circuit means energizing one of said windings during the scanning movement in one direction of said support, circuit means deenergizing said one of the windings during the operation of said indexing motor, circuit means terminating operation of said indexing motor after the carriage member has been stepped, and circuit means energizing the other winding upon termination of the operation of said indexing motor.

18. A holder for use with a scanning and particle counting apparatus including a carriage member movable in a plurality of directions, said holder comprising a substantially planar body, a multiplicity of elongated transparent sample tubes disposed substantially adjacent one another in side-by-side parallel relation on said body, means on said body surface opposed from said sample tubes adapted for removable mounting cooperation with said carriage member whereby said body is predeterminately positioned thereon so that said tubes are individually and sequentially scanned during carriage movement.

19. The holder of claim 18 wherein said sample tubes are removably received by said holder, and means on said holder cooperating with said sample tubes to constrain movement of the same in directions both along and normal to their axes when received on said holder.

20. The holder of claim 19 wherein said holder body provides a central opening bounded on opposite sides by a pair of arms, said cooperating means including a multiplicity of equidistantly spaced grooves formed in said pair of arms, one groove in each arm providing support for opposite ends of said sample tubes on an upper surface of said holder.

21. The holder of claim 20 wherein said cooperating means additionally comprises a resilient clamp element, said clamp element being substantially C-shaped and pivotally mounted at its bight portion to a rear arm of said holder, the sides of said clamp element positioned to overlie said grooves, and a pair of lugs supported by a front arm of said holder for releasably receiving said clamp element sidearms.

* * * * *

UNITED STATES PATENT OFFICE
CERTIFICATE OF CORRECTION

Patent No. 3,627,424 Dated December 14, 1971

Inventor(s) Horton E. Dorman, et al.

It is certified that error appears in the above-identified patent and that said Letters Patent are hereby corrected as shown below:

Abstract, line 1, after "detecting" include --, --.
Column 1, line 9, change "micro-organisms" to --microorganisms--.
line 22, change "micro-organisms" to --microorganisms--.
line 25, change "micro-organisms" to --microorganisms--.
line 50, change "light scatter" to --light-scatter--.
Column 2, line 4, change "sue" to --use--.
line 18, change "micro-organisms" to --microorganisms--.
line 52, change "nonincreased" to --non-increased--.
Column 3, line 28, change "platelike" to --plate-like--.
line 33, change "platelike" to --plate-like--.
Column 5, line 29, change "manually" to --normally--.
Column 6, line 31, change "130-132-31" to --130-132-131--.
Column 7, line 1, delete "forward step, during" and substitute --of--.
line 59, delete "(" and ")" and before "a" insert --to--.
line 66, change "and" to --or--.

Signed and sealed this 26th day of September 1972.

(SEAL)
Attest:

EDWARD M.FLETCHER,JR.
Attesting Officer

ROBERT GOTTSCHALK
Commissioner of Patents

FORM PO-1050 (10-69)

USCOMM-DC 60376-P69
* U.S. GOVERNMENT PRINTING OFFICE : 1969 O—366-334