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Vinogradov et al.

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(54) **QUANTITATIVE ANALYSIS OF A BIOLOGICAL SAMPLE OF UNKNOWN QUANTITY**

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(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 148 days.

This patent is subject to a terminal disclaimer.

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Related U.S. Application Data

(60) Division of application No. 10/706,321, filed on Nov. 12, 2003, now Pat. No. 7,611,670, which is a continuation-in-part of application No. 10/421,086, filed on Apr. 23, 2003, now Pat. No. 7,479,392.

(60) Provisional application No. 60/374,629, filed on Apr. 23, 2002.

(51) **Int. Cl.**
B01L 3/00 (2006.01)
G01N 33/555 (2006.01)

(52) **U.S. Cl.** **422/430**; 436/520

(58) **Field of Classification Search** 422/430; 436/520

See application file for complete search history.

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(57) **ABSTRACT**

Disclosed is a method for testing a modified specimen such as a dried blood spot, plasma or serum specimen, for an analyte of interest, such as cholesterol. In accordance with the disclosed subject matter, the level of the analyte of interest in the medium from which the modified specimen was obtained (e.g., from a patient's blood) is determined based on the level of an analyte in a solution formed from the modified specimen and on the level of at least one normalizing analyte. The analyte and normalizing analyte each may be an ion, compound, biochemical entity, or property of the specimen. Also disclosed are a fluid collector and a fluid collection device.

14 Claims, 11 Drawing Sheets

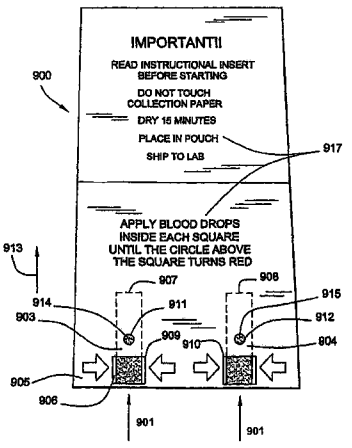

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FIG. 1

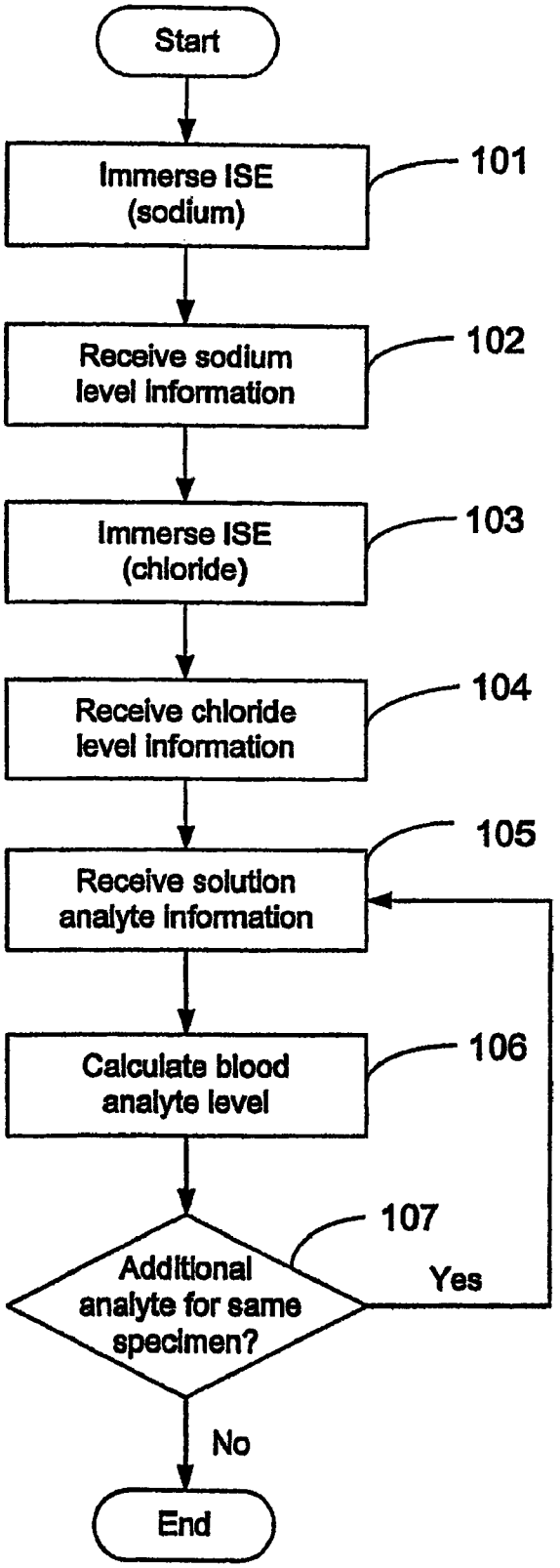

Start
Immerse ISE (sodium)
101
Receive sodium level information
102
Immerse ISE (chloride)
103
Receive chloride level information
104
Receive solution analyte information
105
Calculate blood analyte level
106
Additional analyte for same specimen?
107
Yes
No
End

FIG. 2

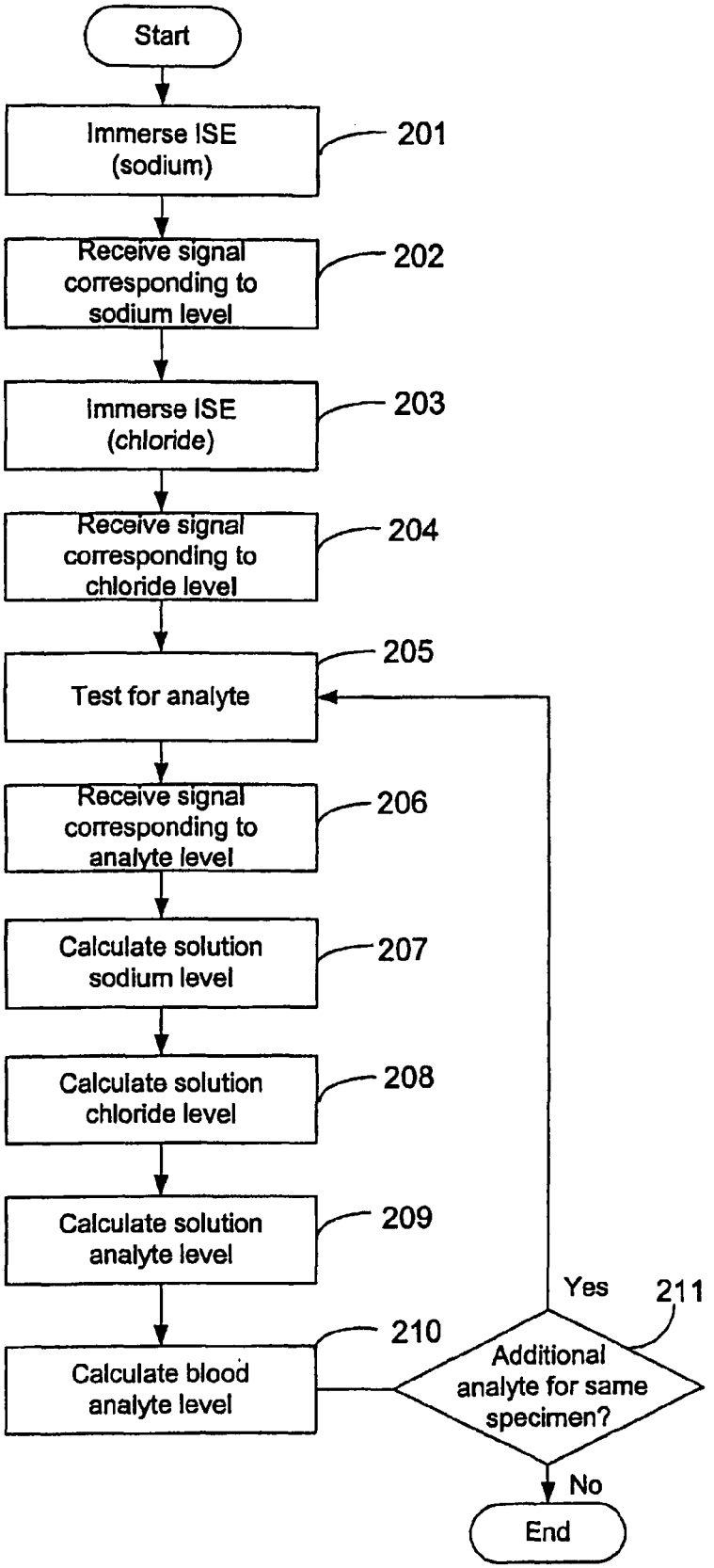

Start
Immerse ISE (sodium)
201
Receive signal corresponding to sodium level
202
Immerse ISE (chloride)
203
Receive signal corresponding to chloride level
204
Test for analyte
205
Receive signal corresponding to analyte level
206
Calculate solution sodium level
207
Calculate solution chloride level
208
Calculate solution analyte level
209
Calculate blood analyte level
210
Additional analyte for same specimen?
211
Yes
No
End

FIG. 3

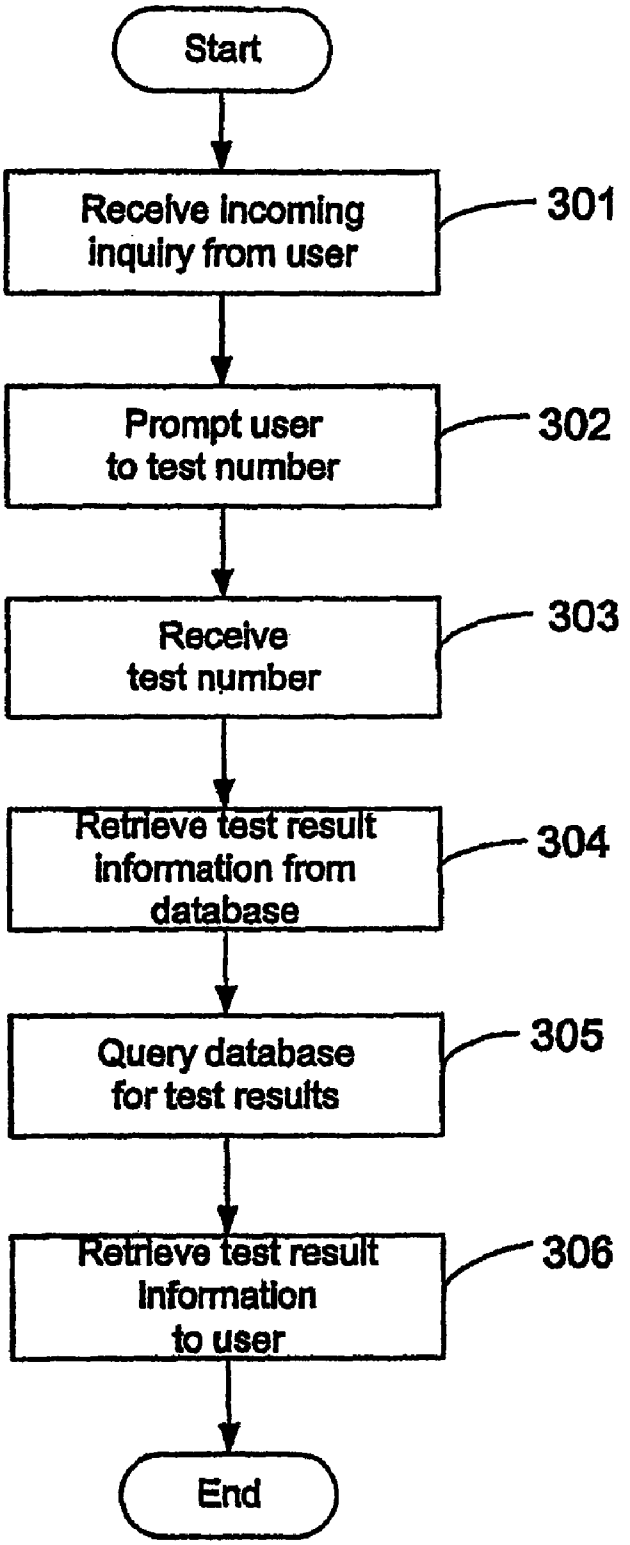

Start
Receive incoming inquiry from user
301
Prompt user to test number
302
Receive test number
303
Retrieve test result information from database
304
Query database for test results
305
Retrieve test result information to user
306
End

FIG. 4

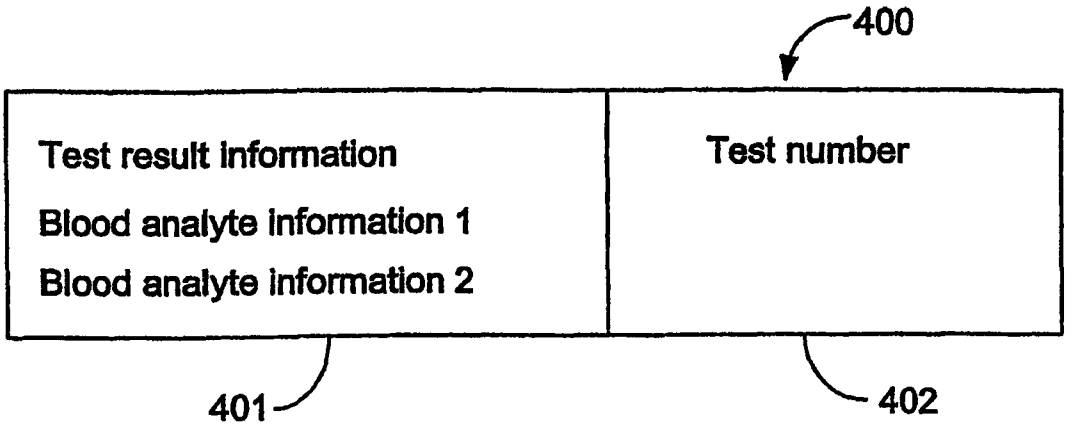

400
Test result information
Blood analyte information 1
Blood analyte information 2
Test number
401
402

FIG. 5

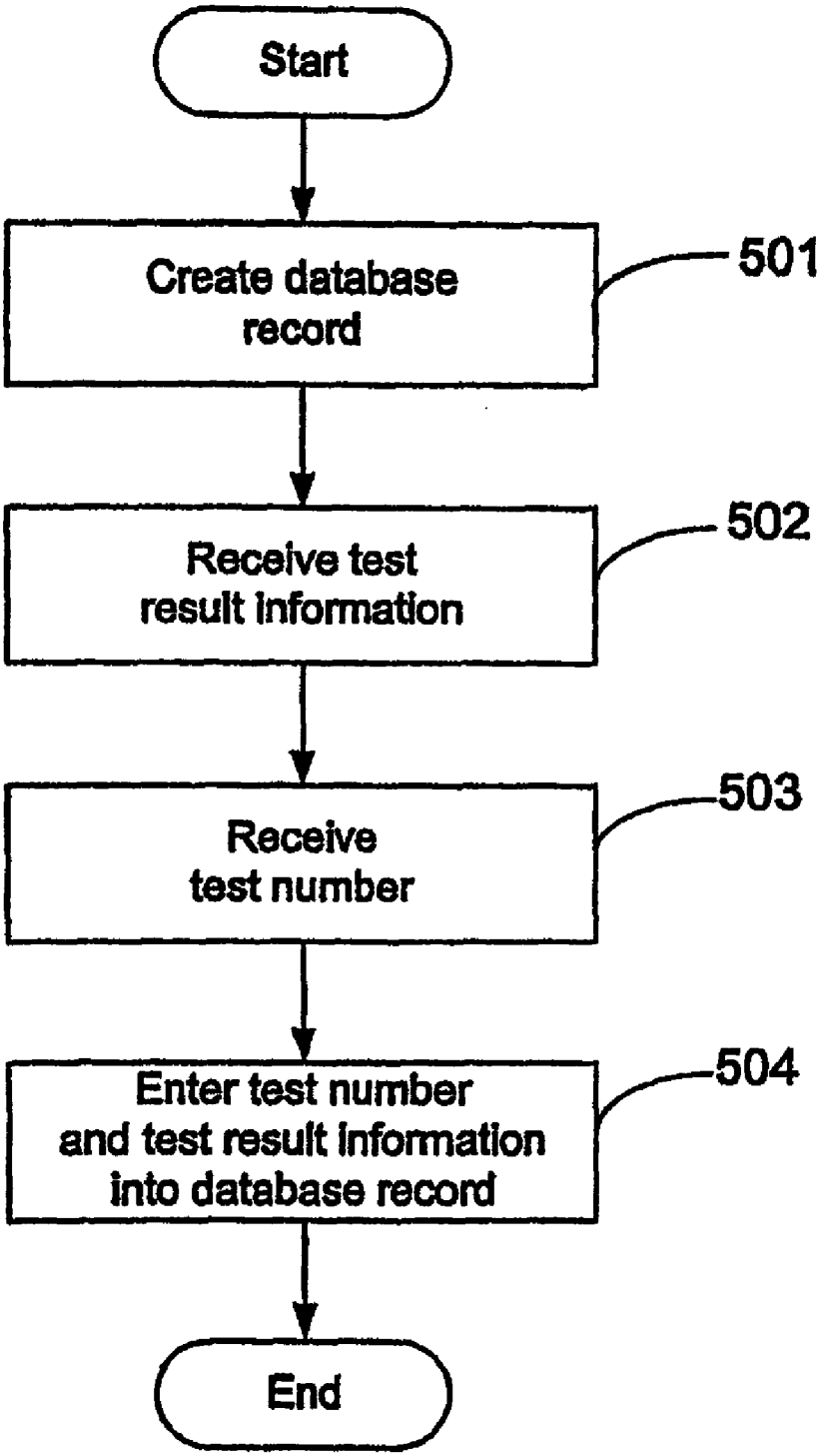

Start
Create database record
501
Receive test result information
502
Receive test number
503
Enter test number and test result information into database record
504
End

FIG. 6

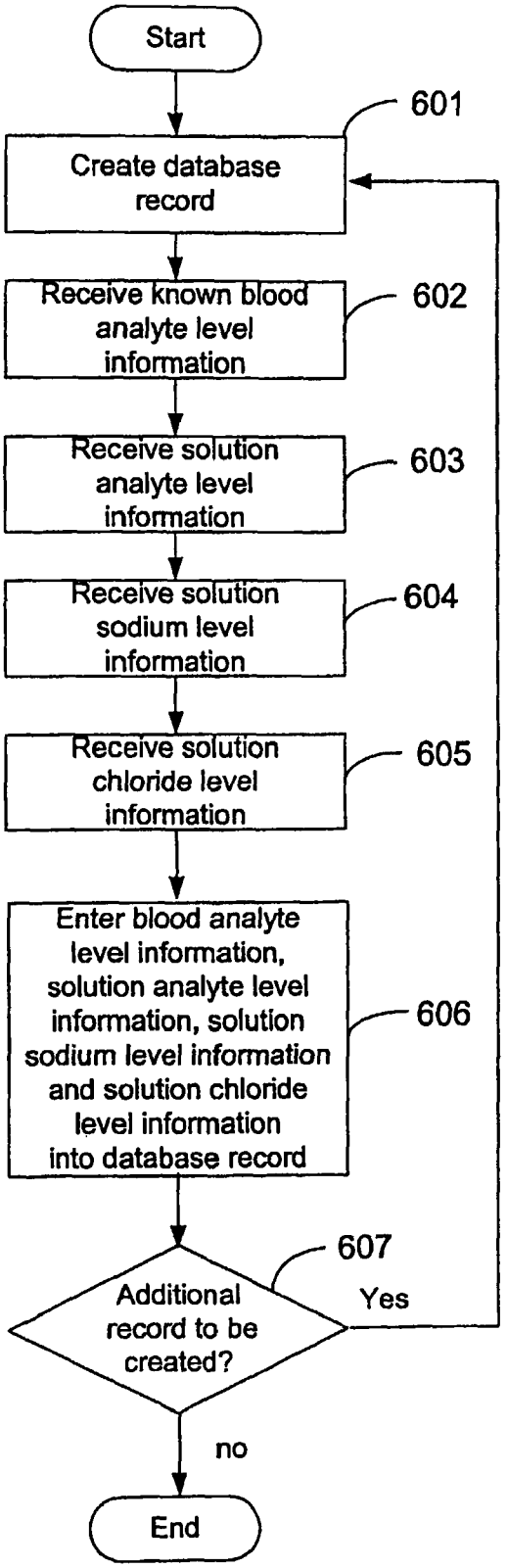

Start
601
Create database record
Receive known blood analyte level information
602
Receive solution analyte level information
603
Receive solution sodium level information
604
Receive solution chloride level information
605
Enter blood analyte level information, solution analyte level information, solution sodium level information and solution chloride level information into database record
606
607
Additional record to be created?
Yes
no
End

FIG. 7

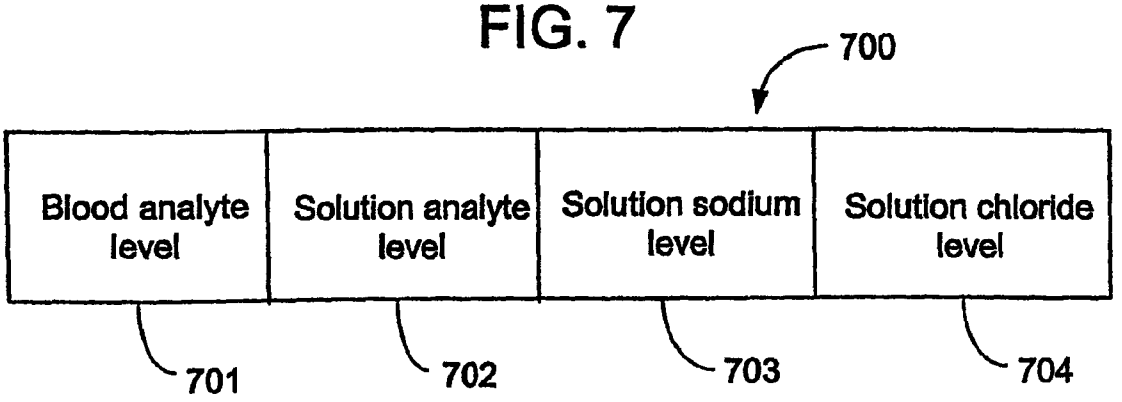

700
Blood analyte level
Solution analyte level
Solution sodium level
Solution chloride level
701
702
703
704

FIG. 8

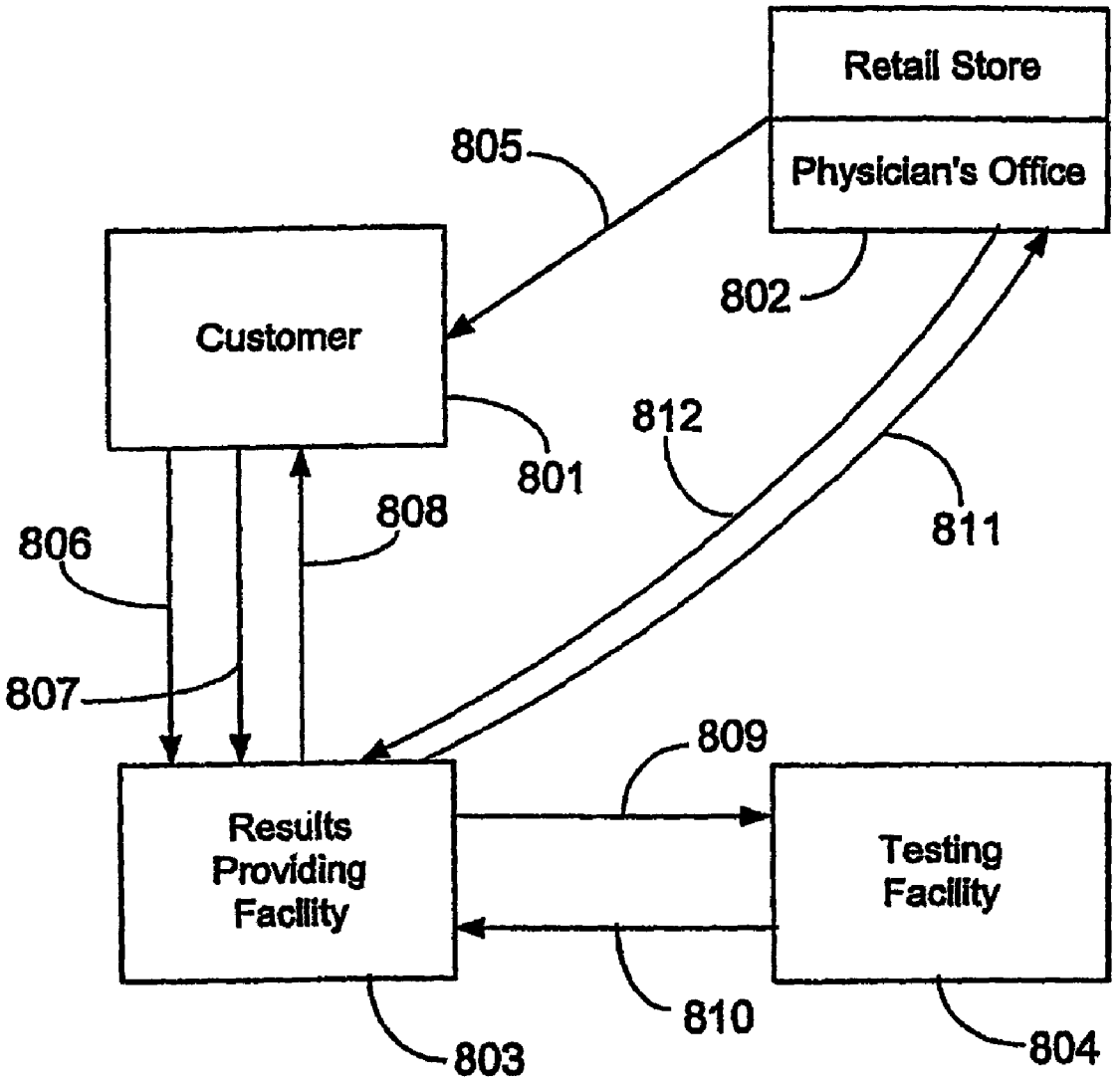

Retail Store
Physician's Office
805
802
Customer
801
812
811
808
806
807
809
Results Providing Facility
Testing Facility
810
803
804

FIG. 9

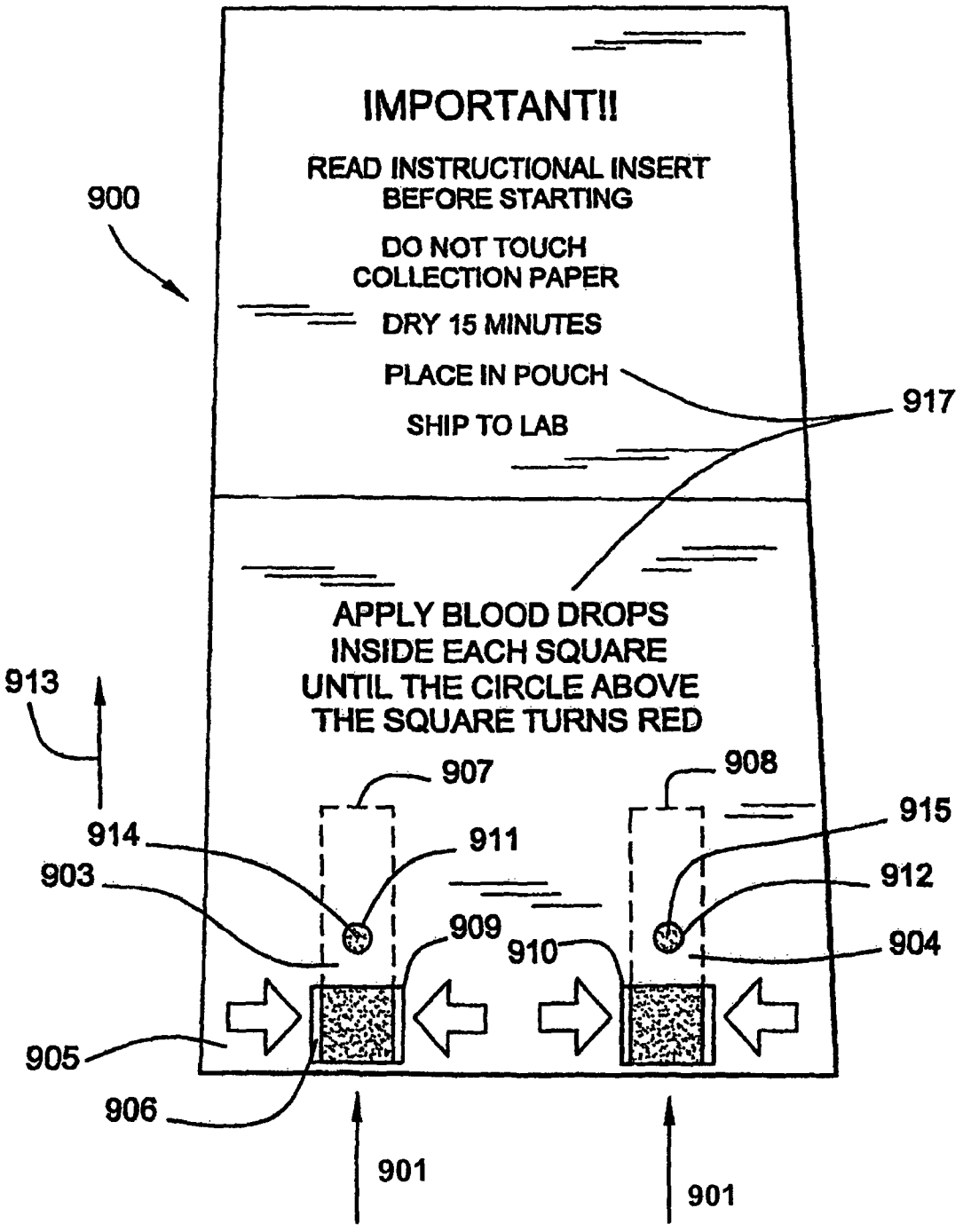

IMPORTANT!!
READ INSTRUCTIONAL INSERT BEFORE STARTING
DO NOT TOUCH COLLECTION PAPER
DRY 15 MINUTES
PLACE IN POUCH
SHIP TO LAB
APPLY BLOOD DROPS INSIDE EACH SQUARE UNTIL THE CIRCLE ABOVE THE SQUARE TURNS RED
900
913
914
903
905
906
907
908
911
909
910
915
912
904
917
901
901

FIG. 10

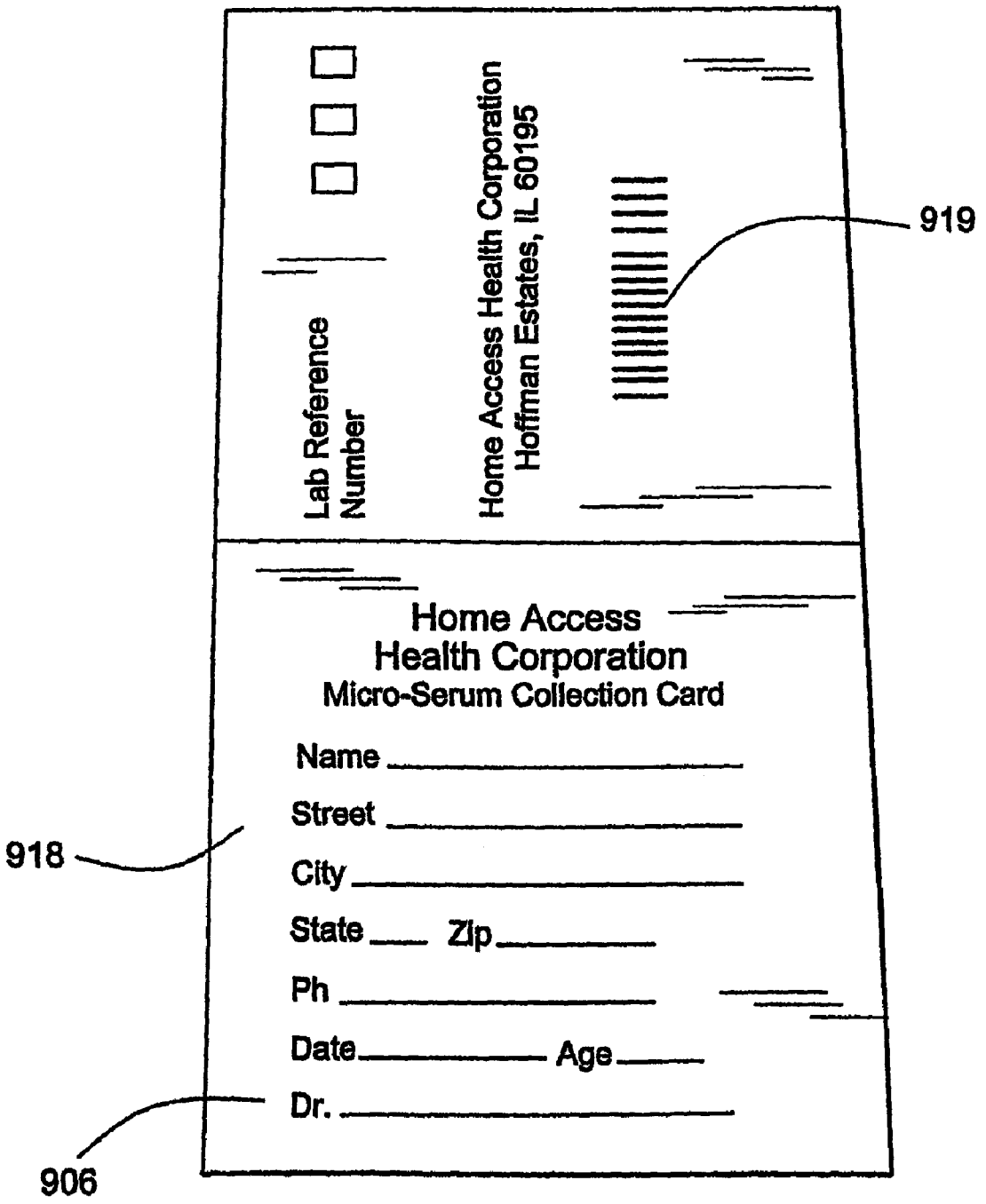
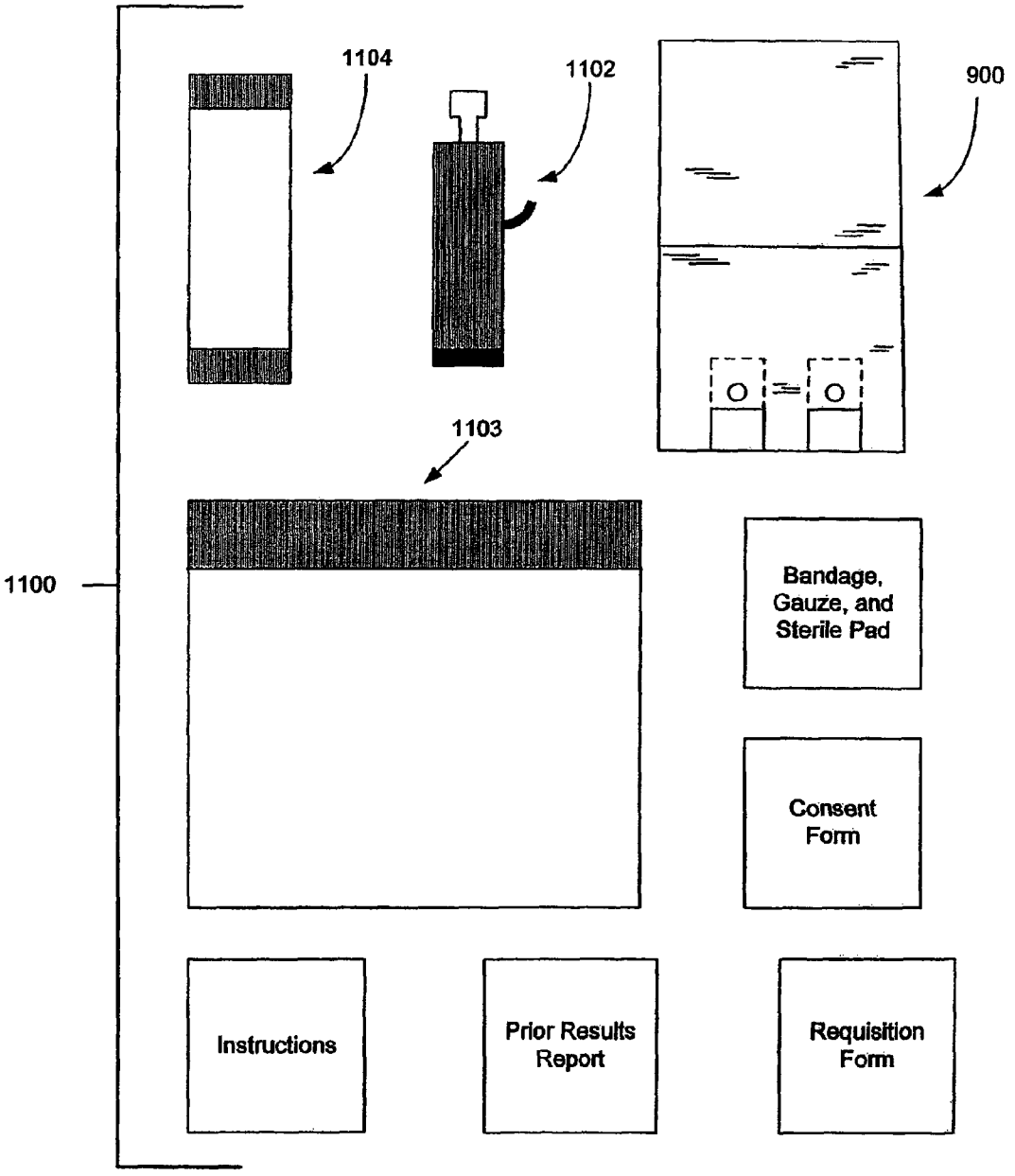

Lab Reference Number
Home Access Health Corporation
Hoffman Estates, IL 60195
919
Home Access
Health Corporation
Micro-Serum Collection Card
Name
Street
City
State Zip
Ph
Date Age
Dr.
918
906

FIG. 11


1104
1102
900
1103
1100
Bandage, Gauze, and Sterile Pad
Consent Form
Instructions
Prior Results Report
Requisition Form

QUANTITATIVE ANALYSIS OF A BIOLOGICAL SAMPLE OF UNKNOWN QUANTITY

CROSS REFERENCE TO RELATED APPLICATIONS

This application is a divisional of co-pending U.S. patent application Ser. No. 10/706,321, filed Nov. 12, 2003, which is a continuation-in-part of U.S. patent application Ser. No. 10/421,086, filed Apr. 23, 2003, now U.S. Pat. No. 7,479,392, which claims priority to Provisional Application No. 60/374,629 filed Apr. 23, 2002, now expired, which prior applications are hereby incorporated herein by reference.

TECHNICAL FIELD OF THE INVENTION

The invention is in the field of testing, in particular quantitative testing, and in preferred embodiments medical testing. In highly preferred embodiments, the invention is directed towards the testing of body fluid specimens, in particular blood or serum specimens.

BACKGROUND OF THE INVENTION

Modern medical and wellness practices increasingly make use of self-administered tests and self-collection of test specimens. For instance, U.S. Pat. Nos. 5,978,466; 6,014,438; 6,016,345; and 6,226,378, issued to Richard Quattrocchi and assigned to Home Access Health Corporation of Hoffman Estates, Ill., all disclose a method of anonymously testing for a human malady. In accordance with certain embodiments of the subject matter disclosed in the foregoing patents, a patient obtains a blood specimen, typically by pricking his or her finger, and allows the blood to wick onto a blood spot card. After the card has dried, the user then sends the blood spot card to a medical testing facility, where it is tested to determine whether the patient is afflicted with a specific malady. The user may contact the facility anonymously to receive the test result.

The subject matter of the foregoing patents is usable in connection with testing for the presence of human antibodies directed against viral antigens in the blood, for instance, in determining whether a patient is infected with HIV (human immuno-deficiency virus) or with a hepatitis virus. Another document, U.S. Pat. No. 5,435,970, issued to Mamenta et al. and assigned to Environmental Diagnostics, Inc. of Burlington, N.C., discloses a device for separating blood cells from biological fluids, for instance, for separating serum from whole blood. The device disclosed in the '970 patent purports to enable the shipment and testing of a serum sample.

The blood spot and serum specimen cards known in the art are suitable for use in the collection of specimens for qualitative testing, i.e., testing for the presence or absence of a given compound in blood or a given medical condition. Heretofore, however, such blood spot and serum cards have been somewhat unsatisfactory in the quantitative testing of blood and serum specimens.

For instance, general wellness protocol indicates the measurements of a patient's total cholesterol value, which is the number of milligrams of total cholesterol in a deciliter of blood. The value is often used in conjunction with a full lipid profile, which provides levels of triglycerides, HDL (high density lipoprotein) cholesterol, and LDL (low density lipoprotein) cholesterol in a patient's blood. It can be very difficult to gauge the amount of blood or serum that is present in the blood or serum spot card. Particularly when the blood or serum spot card has been self-prepared by a person without medical training, it is difficult to know to certainty whether the spot card has been "underfilled" with less than the intended quantity of blood or serum or "overfilled" with more than the intended quantity. If the amount of blood and serum varies by even a small amount over or under the expected level, the usefulness of the quantitative test can be severely diminished. For instance, it is generally thought that a person's total cholesterol number should be under 200 mg/dl, with cholesterol numbers above 240 mg/dl being considered high and with intermediate cholesterol number being deemed borderline. A 10% margin of error in a cholesterol determination of 220 mg/dl provides no information as to whether the person's cholesterol level is low, intermediate, or high.

In recognition of these problems, the prior art has provided attempts to provide a quantitative determination of analyte levels in a blood specimen. For instance, U.S. Pat. No. 6,040,135, issued to Steven Tyrell and assigned to Biosafe Laboratories, Inc., Chicago, Ill., purports to disclose a method for correcting for blood volume in a serum analyte determination. The method that is purportedly disclosed by this document is limited and is believed generally to be somewhat unsatisfactory.

The invention seeks to improve upon prior art testing methods, and to provide a method for quantitative testing of modified specimens such as dried blood spot and dried serum specimens.

THE INVENTION

The invention provides multiple embodiments in the field of testing, in particular medical testing. In accordance with the invention, a modified specimen, preferably a dried blood fluid sample, such as a dried serum or dried whole blood specimen of unknown quantity, is eluted (re-solubilized) and then tested for an analyte. The level of analyte in the blood from which the modified blood specimen was obtained is determined from the level of analyte in a solution formed from the blood specimen. A normalizing analyte, which in the preferred embodiment is sodium ion, chloride ion, and/or osmolality, is measured and is used in conjunction with the solution level of analyte to determine the level of analyte in the blood from which the modified specimen was obtained. The invention is not limited to the field of medical testing but, to the contrary, is useful in connection with other forms of testing. The invention further provides methods for preparing a database of test results, for preparing a regression using a database of test results, and for providing test results to a user.

In alternative embodiments the invention further encompasses a fluid collector that includes an absorbent substrate coated with a saccharide. A device that includes the collector (as described hereinbelow) also is encompassed by these embodiments.

Other features of preferred embodiments of the invention are set forth hereinbelow.

BRIEF DESCRIPTION OF THE FIGURES

FIG. **1** is a flowchart representing steps in a method for calculating the level of an analyte in blood from which a blood specimen was obtained.

FIG. **2** is a flowchart representing steps in an alternative method for calculating the level of an analyte in blood from which a blood specimen was obtained.

FIG. **3** is a flowchart representing steps in a method for providing test result information to a user.

FIG. **4** is a representation of a database record correlating test result information with a test number.

FIG. **5** is a flowchart representing steps in a method for preparing a database of test results and test numbers.

FIG. **6** is a flowchart representing steps in a method for preparing a database of blood analyte levels, solution analyte levels, and solution normalizing analyte levels.

FIG. **7** is a representation of a database record for a database containing blood analyte level information, solution analyte level information and solution normalizing analyte level information.

FIG. **8** is a schematic illustration showing various communications between a customer, a results providing facility, and others in connection with a testing protocol.

FIG. **9** is a perspective view of the obverse side of a blood collection device useful in conjunction with the invention.

FIG. **10** is a perspective view of the reverse side of the device shown in FIG. **9**.

FIG. **11** is a representation of a kit useful in conjunction with the invention.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

The invention is applicable to the testing of any specimen that is modified from its original form prior to testing. Most commonly, the specimen is a dried specimen, which has been dried to facilitate storage or transport of the specimen or for other purposes. In preferred embodiments of the invention, the specimen is a medical specimen, and in highly preferred embodiments of the invention, the specimen is a blood fluid specimen, by which is contemplated a dried blood spot, a dried serum spot (for instance, as obtained from the device disclosed in U.S. Pat. No. 5,435,970 or that shown in U.S. Pat. No. 4,839,296 issued to Kennedy, et al. and assigned to Chem-Elec, Inc. of North Webster, Ind.), or another blood fluid specimen. The invention is applicable to the testing of the modified specimen for any suitable purpose, and in particular to testing for any analyte in the specimen. For instance, when the specimen is a blood fluid specimen, the test may be a test for prostate specific antigen (PSA), alanineamino transferase (ALT), lipids, such as triglycerides, high density lipoprotein (HDL), low density lipoprotein (LDL), or any other analyte of interest. The invention is applicable to the determination of the level of analyte in the original specimen, for instance, the level of total cholesterol in the blood from which a blood fluid specimen has been obtained. The "level" of the analyte can be expressed in any suitable units, such as molar concentration, weight concentration, or the like. Blood serum is particularly preferred, but it is contemplated that other fractions such as cells, platelets, gamma globulins, plasma or the like may be employed. For instance, it may be designed to test blood cells in connection with a fasting plasma glucose test. More generally, any body fluid is susceptible to analysis in conjunction with the invention. In light of the foregoing, the preferred embodiments of the invention will be further described with respect to the determination of the lipid profile in a blood sample, but it should be understood that the invention is not limited thereto.

The facility or other entity that performs the test of the blood fluid specimen may or may not be the same entity that calculates the level of the analyte in the blood fluid specimen or the entity that receives an inquiry from a user and reports the test results to the user. To test the blood fluid specimen, the specimen is first received by the testing entity and is eluted with a liquid, preferably deionized water. It is contemplated that the liquid may be a non-aqueous liquid or may be an aqueous solution, preferably a solution that is free or essentially free of sodium ions or any other normalizing analyte. Alternatively, the solution may have a known amount of the normalizing analyte that can be taken into account during normalization. Preferably, when the testing entity is a testing facility that is intended to test numerous specimens, the eluant is added in a standard amount, which typically is 600 μl (0.6 ml). The eluant in some embodiments may be a buffered electrolyte solution.

After eluting the specimen, preferably the specimen first is tested for the content of a normalizing analyte, such as sodium and chloride content, and in some embodiments osmolality, which generally represents total content of sodium, glucose, and blood urea nitrogen (BUN). To test for sodium and chloride, an ion specific electrode (ISE), such as that sold by Orion may be employed. Preferably, information concerning both the sodium and the chloride content of the solution are obtained, the information being, for instance, analog information such as an electrical signal or digital information such as a printout representing the sodium or chloride content or a digital signal containing information concerning the sodium or chloride content. Most preferably, osmolality also is measured. It should be noted that the invention is not limited to the use of sodium or chloride as normalizing analytes, but to the contrary, any other analyte (which includes a property such as osmolality) may be measured. It is contemplated in preferred embodiments that the sodium, chloride, and osmolality levels are measured against a predetermined range to determine whether the amount of serum is sufficient to perform an adequate test. For instance, it is contemplated that for a cholesterol test, there ideally should be at least approximately 15-17 μl of serum available for testing. If the sodium content of the eluted solution demonstrates that the serum level is far outside this range, the specimen may be rejected as unsuitable for testing. Generally, the specimen may be rejected if there is insufficient serum in the solution, although it is contemplated that in some cases excess serum may be grounds for rejection. Persons skilled in the art may determine how far outside of the desired range the content of normalizing analyte may be allowed to vary without triggering rejection of the specimen.

Before or after the levels of the normalizing analytes are determined (but preferably after), the solution can be split into four aliquots, or "channels." Each channel is then respectively tested for triglyceride level, HDL level, LDL level, and in a preferred embodiment, ALT level (which may be of interest in informing a physician whether the patient has an abnormal liver which would contraindicate the use of certain drugs). The analyte levels are measured using any technique known in the art or otherwise found to be suitable. For instance, a cholesterol test is disclosed in Allain, C. C., Poon, L. S., Chan, G. S. G., Richmond, W., and Fu, P. C., *Clin. Chem.* 20:474-75 (1974); see also Roeschlau, P. Brent, E. and Gruber, W A., *Clin. Chem. Clin. Biochem.* 12:226 (1974). A test for HDL is disclosed in RiFai, N., Wamick, G. R., Ed., *Laboratory Measurement of Lipids Lipoproteins and Apolioproteins* (1994). A test for triglycerides is disclosed in McGowan, M. W., Artiss, J. D., Strandbergh, D. R., Zak, B. *Clin. Chem.* 29:583 (1983). A test for the liver enzyme ALT is disclosed in Wroblewski, F., LaDue, J. S., *Proc. Sec. Exp. Biol. Med.* 34:381 (1956). The invention is not limited to the foregoing tests or analytes, but to the contrary is applicable to other tests for these or other analytes.

After the analyte levels have been measured, the level of at least one analyte (and preferably all analytes) in the blood from which the blood fluid specimen was obtained is calculated or otherwise determined based on the solution level of

the analyte and on the solution level of at least one normalizing analyte. It is contemplated that the calculation of a blood analyte level may be as simple as multiplying the solution analyte level by the ratio of the blood normalizing analyte level to the solution normalizing analyte level, the blood normalizing analyte level being estimated based on the mean of a normal population distribution. For instance, it is believed that the normal blood sodium level in humans ranges from 136 to 142 mEq/L with a mean of 139 mEq/L and the normal chloride level ranges from 95 to 103 mEq/L with a mean of 99 mEq/L. It is contemplated that through the use of two normalizing analytes, the blood analyte level may be determined by calculating the blood analyte level based on the first normalizing analyte level, calculating the blood analyte level based on the second normalizing analyte level, and then calculating the mean average of the blood analyte levels thus determined.

If additional normalizing analytes are evaluated, the mean average of all blood level analytes thus determined may be calculated; if desired, where there are at least two normalizing analytes, the average may be weighted towards a specific normalizing analyte. For instance, it is contemplated that Bayesian statistical methods may be used to assign a relative weight to the blood analyte levels determined with reference to each analyte. Such statistical techniques may take into account not only the absolute magnitude of the level of the normalizing analyte level but also the difference between the actual level and the magnitude expected based on the expected amount of serum, and the standard deviation of the normal population distribution of the analyte. These techniques, sometimes referred to as "maximum likelihood" or "prior probability analysis" techniques, may be used to provide an approximation of the blood analyte level. Further testing concerning such statistical techniques may be found in Casella, G., Berger, R. L., *Statistical Inference* (1990) and Carlin, B. P., Louis, T. A., *Bayes and Empirical Baves Methods for Data Analysis* (2d Ed. 2000).

Further details concerning the distribution of sodium, chloride, and osmolality in the normal human population may be found in Ravel, *Clinical Laboratory Medicine* (6th Ed. 1995); see also Penney, M. D. and Walters, G., *Ann. Clin. Biochem.* 24:566-71 (1987) and Fraser, C. G., Cummings, S. T. Wilkinsen, S. O. et al., *Clin Chem.* 35:783-86 (1985). It is further contemplated that a more complicated function of solution analyte level and the levels of one or more normalizing analytes may be employed to calculate the blood analyte levels.

With reference now to FIG. **1**, the generalized method shown therein is applicable where the same entity performs the test and calculates the blood analyte level. Thus in steps **101** and **102** respectively the ISE (e.g., sodium) is immersed into the solution, and sodium level information is obtained. The steps are repeated for the receipt of chloride information, as shown in steps **103** and **104**. Information concerning the analyte of interest is received in step **105**, and the blood analyte level is calculated in step **106**. If, in step **107**, it is desired to test an additional analyte for the same specimen, control passes to step **105** where the solution analyte information is received for the new analyte. It is contemplated that the steps of testing for the analytes of interest and the normalizing analytes may be performed by one entity and that the calculation of the blood analyte level may be performed by a separate entity. Thus, for instance, in FIG. **1**, steps **101** and **103** may be omitted if the entity calculating the blood analyte level is not the same entity as the entity that performs the test. The method outlined in FIG. **1** is very general, and other steps may be added, steps may be omitted or performed in a different order, and more generally the method may be otherwise performed. For instance, steps of elution and verifying proper serum level are not shown, but are preferably employed.

It is contemplated that the analyte level, first normalizing analyte level, and second normalizing analyte level may be independently determined and these values used to calculate the blood level of the analyte. For instance, the cholesterol tests hereinbefore discussed typically are performed via enzymatic techniques in which the optical density of a solution is measured. The "cholesterol value" of the solution then may be expressed as:

$$CV_s=f(OD)$$

wherein CVs, the solution cholesterol concentration, is calculated as a function of the optical density, OD, when analytical reagents are added to the sample in accordance with testing techniques known or otherwise found to be suitable. The solution sodium concentration, or Na_s, may be used to calculate the blood cholesterol level, CV_b, in the following manner:

$$CV_b=f(CV_s,Na_s)$$

Numerous other forms of such calculations are possible. For instance, a correction factor (CF) may be determined as a function of the solution's sodium level, wherein:

$$CV_b=f(CV_s,CF)$$

and

$$CF=f(Na_s)$$

It is alternatively contemplated that a single apparatus or system may be designed for the calculation of blood analyte levels, wherein an analog or digital electrical signal is generated corresponding to the levels of analyte and normalizing analyte in the solution. For instance, the blood cholesterol number may be calculated as a function of the magnitude of two electrical signals:

$$CV_b=f(E_1,E_2)$$

wherein E_1 represents the magnitude of an electrical signal received from a spectrophotometer in measuring optical density for purposes of evaluating total solution cholesterol level and E_2 represents the magnitude of an electrical signal received from an electrode specific to sodium.

In actual practice, it is contemplated that numerous variables will affect the results obtained for a given set of specimens. For instance, the readings obtained from an ISE may "wander" from day to day, and the device used to collect the blood or other fluid specimen may contain impurities (such as sodium) that have the potential to introduce errors into the test. For this reason, from time to time a "tare" procedure may be employed. Periodically, a plurality of specimens having a known or measurable analyte level is provided, and from these specimens are prepared modified specimens, the modified specimens being specimens as modified in the manner expected of the unknown specimens. For instance, some number (e.g., six) blood specimens may be periodically placed onto a blood spot collection device similar to those used in the field and dried, followed by elution of the dried samples to form solutions. The solutions are then tested for the level of the analyte and one or more normalizing analytes. From these tests, an algorithm for determining the original fluid analyte level as a function of the measured analyte level and the levels of the normalizing analyte or analytes may be derived. Using this algorithm, modified fluid specimens may be analyzed, wherein the levels of analyte and normalizing analyte may be measured, and the level of analyte in the original specimen may be determined as a function thereof.

Errors introduced by impurities (such as sodium) in the collection device will be resolved by this methodology, and errors introduced by factors such as machine calibration will be resolvable with periodic re-calculation of the algorithm. The tare procedure may be performed occasionally or regularly at predetermined intervals (e.g., every day, week, month, or year).

The foregoing exemplary equations are not meant to be exhaustive but, to the contrary, are intended to illustrate that innumerable variants of the methods for calculating the blood analyte level are included within the scope of the invention. For instance, with respect to FIG. **2**, in one such variant, an ISE (sodium) is immersed into an eluted sample at step **201**, and a signal corresponding to the sodium level is received at step **202**. The signal may be a digital signal, or may be an analog signal, the level of which is recorded. At steps **203** and **204**, the same steps are repeated for chloride level, and at steps **205** and **206** respectively, a test for the analyte is performed and a signal is received corresponding to the analyte level. At step **207**, the solution sodium level is calculated; at step **208**, the chloride level is calculated, and at step **209**, the solution analyte level is calculated. At step **210**, the blood analyte level is calculated, in this instance based on the magnitude of the solution sodium level, the solution chloride level, and the solution analyte level. If, at step **211**, it is desired to test for an additional analyte for the same specimen, control passes to step **205**. In such case, if the solution sodium and chloride level have been stored, steps **207** and **208** may be omitted after a signal is received corresponding to the second analyte level. The process may be controlled by any suitable microprocessor or microcontroller (not shown).

As stated hereinabove, it is contemplated that the entity who provides test results to a user, who may or may not be the health care professional who has ordered the test, in turn may be the same or different entity from the entity which performs the calculation of the blood analyte level, which in turn may be the same or different entity from the entity which tests the specimen and generates information corresponding to the analyte level or levels and the normalizing analyte level or levels. A very general protocol for a results providing facility is set forth in FIG. **3**, wherein an inquiry is received from a user at step **301**, and the user is prompted for his or her test number at step **302**. At step **303**, the test number is received, and at step **304**, a test result database is queried for test result information. The information is received at step **305** and is provided to the user at step **306**.

With further reference to FIG. **4**, the test result database described above may be structured in any suitable manner. With respect to, for instance, database record **400**, the test result information **401**, which in the illustrated embodiment includes two items of information, blood analyte information 1 and blood analyte information 2, is correlated with the test number **402**. The test number may be an anonymous test number or may be a test number that is associated with a user, for instance, elsewhere in the database record **400** (not shown) or in a different database.

With reference to FIG. **5**, the database may be prepared by creating a database record (shown in step **501**), receiving test result information and a test number (shown in steps **502** and **503** respectively) and, as shown in step **504**, entering the test number and test result information into the database record. More information concerning the role of a results providing facility in a medical or wellness testing protocol can be found in the aforementioned Quattrocchi patents and in copending application Ser. No. 09/709,884.

The invention additionally contemplates a method for preparing a database for use in calculating blood analyte levels. The blood analyte level may be calculated with specific reference to the database, or alternatively the database may be used in conjunction with the preparation of an algorithm for enabling blood level calculation. The database preferably is prepared with reference to blood having a known level of cholesterol or other analyte of interest. Plural specimens of blood having different levels of the analyte are then reduced to an modified specimen, such as a blood spot or serum specimen, and each specimen is analyzed for the analyte of interest and for a normalizing analyte. For instance, with respect to FIG. **6**, a database record is created at step **601**, and known blood analyte level information is received at step **602**. Information as to the solution analyte level and the level of two normalizing analytes, sodium and chloride, for example, are received at steps **603-605**, and at step **606**, the information received is entered into the database record. If, at step **607**, an additional database record is to be created, control passes to step **601**, wherein a new database record is created for the new specimen. It should be noted that the order of the steps is not critical, and indeed the database may be prepared sequentially with respect to each blood specimen (i.e., each specimen is reduced to an modified specimen, tested, and the results entered into a database record prior to altering the next specimen of blood), sequentially with respect to database record (wherein all of the blood specimens are reduced to modified specimens prior to entering the first database records) or by any other suitable methodology. A database record **700** as shown in FIG. **7** is thus prepared, with entries **701** through **704** representing respectively blood analyte level, solution analyte, solution sodium level, and solution chloride level.

As discussed above, rather than being calculated, the blood analyte level in a blood fluid specimen may be determined with reference to the database, for instance, by finding the solution analyte level and solution normalizing analyte level or levels in the database that are closest to those of the specimen. Alternatively, any suitable statistical or mathematical technique may be used to derive an algorithm for calculating the blood analyte level from the solution analyte level and at least one normalizing analyte level. In some embodiments, the algorithm is first order with respect at least to the solution analyte level, and may be first order with respect to the solution analyte level and one or both normalizing analyte levels.

The invention preferably is conducted in accordance with the general schematic set forth in FIG. **8**. Generally the customer **801** purchases a test kit from a physician or retail store **802** (transfer of the kit is shown via transfer communication **805**) or in other embodiments a patient is provided with a test kit by or at the direction of a health care provider. The test kit (not shown in FIG. **8**) preferably includes instrumentalities for allowing the customer to obtain a blood, serum or serum spot specimen. For instance, as discussed more fully in the aforementioned Quattrocchi patents, the test kit may include a lancet for pricking the user's finger, a blood spot card, or serum spot card, (or the device shown in FIGS. **9** and **10** hereinafter discussed) an informed consent form, and a test number. After preparing the blood, serum or serum spot card, the customer sends the dried blood specimen to a results providing facility **803** as shown via transfer communication **806**. In the illustrated embodiment, the results providing facility **803** sends the specimen to a separate testing facility **804**, as shown via transfer communication **809**. As shown via communication **810**, the testing facility provides the test results to the results providing facility. The results may be "raw" results, i.e., results in which the level of the analyte in the blood has not been determined or obtained, or alternatively the testing facility may calculate the blood analyte level and report that result to the results providing facility. As

shown at communication **807**, the customer contacts the results providing facility, and at communication **808**, the results providing facility provides the test results to the customer. Optionally, the results providing facility may be equipped to communicate directly with the physician's office, as shown at communications **811** and **812**. Except where transfer of a physical specimen is required, the communication may be made via any means or method now known or hereinafter discovered, for instance, via telephone, wireless communication, electronic mail or "chat" or other electronic communication, or other form of communication.

With reference now to FIGS. **9** and **10**, the illustrated fluid collection device **900** includes two gangs **901**, **901**, each comprising a fluid collector **903**, **904** that is disposed between a superstrate sheet **905** and a substrate sheet **906** and that is generally fixed with respect to the superstrate sheet **905**. The fluid collector is ordinarily connected to the substrate sheet **906** (a portion of which is visible) at one end **907**, **908**, although the collector may be flexible and thus not entirely fixed with respect to the substrate sheet **905**. The substrate is provided with at least one aperture (two shown as **909**, **910**) by which a user may fluidically transfer blood to the collector. In the illustrated embodiments, secondary apertures **911**, **912** are provided. To use the device, a user dispenses blood onto the collector, whereby some or all of the blood wicks in the direction shown by arrow **913** until the portions **914**, **915** of the collectors **903**, **904** visible through the secondary apertures **914**, **915** become tinted, whereupon the user is provided with an indication that sufficient blood has been collected. As illustrated, the device preferably is disposed horizontally relative to the ground during the wicking of blood. Any other suitable indicator that an amount of blood predetermined to be adequate may be provided. In the illustrated embodiments, instructions **917** are provided on the substrate sheet **905** and identification information spaces **918** (shown in FIG. **10**) are provided on the substrate sheet **906**. The device may be provided with non-textual machine-readable indicia (such as barcode **919**) or textual indicia that indicates, for instance, a test number, code number, lot number, or other desired information.

With reference to FIG. **11**, the illustrated kit **1100** includes the specimen collection device **900** illustrated in FIG. **9**, and numerous other components, some or none or all of which in practice may be included in a kit. The kit includes a barrier pouch **1103**, a dessicant pouch **1104**, a lancet **1102**, and instruction sheet separate from the kit, a results form from a previous test, and a requisition form as specifically shown in the figure. In preferred embodiments, the kit includes a mailing device, most preferably a preaddressed envelope with postage prepaid for sending the collection device to a testing facility or other appropriate facility. In practice, the kit may further include a bandage, gauze pad, and alcohol pad for use with drawing blood from the patient (not shown) and a form for providing informal consent, which informed consent may be provided anonymously as described in the heretofore mentioned Quattrocchi patents. The barrier pouch should be a pouch that is effective in protecting the dried blood sample during shipping. One suitable barrier material is sold by Caltex Plastics of Vernon, Calif. and comprises a multi-layer barrier film consisting of 25 bleach MG Paper, 48 GA polyester film, 0.0005 aluminum foil, and 0.003 EVA co-polymer, the layers being adhesively bonded together. The pouch preferably is formed with at least one self-sealing device, such as a "zipper" disposed at least one end of the pouch. A pouch that includes two self-sealing devices, one at each end of the pouch, alternatively may be provided.

The dessicant pouch should be a porous container that includes suitable dessicant effective to provide a dessicating protective effect on a blood fluid specimen, and to some extent to protect the integrity of the collection device during transport to the physician or patient. Any suitable dessicant material may be used in conjunction with the invention. One suitable dessicant is made by SudChemie of Balen, N.M. under part number 4286. This material comprises silica and clay disposed in admixture in a 5 gram pouch. Any other suitable dessicant may be used in conjunction with the invention.

Likewise, any suitable lancet may be employed in conjunction with the invention. The illustrated lancet **1102** preferably comprises a blood-obtaining lancet such as that presently available from Palco Labs of Santa Cruz, Calif. as the EZ-LETS II. This device includes a single-use lancet that is spring-loaded to enable the lancet to sharply pierce a user's skin. Any other suitable lancet may be used in conjunction with the invention. The barrier film pouch is sized to receive the fluid collection device. Preferably, the pouch is sized to receive the dessicant pouch and the fluid collection device.

An instruction set may be included as a separate sheet within the kit, or alternatively the instructions may be integral with (for example, imprinted on) the fluid collection device. The kit may further include results from a previous test. Such is useful, for example, in the case of patients who require periodic testing, for instance, of blood cholesterol. The invention encompasses in some embodiments a method of providing a test kit and test results to a health care provider and/or a patient, the test results being results from a previous test and the test kit being a kit as heretofore described. In some embodiments, the patient responds to an indication in the results form as to whether or when to obtain a subsequent blood sample or other type of sample.

The invention contemplates methods wherein a physician is provided with a test kit as heretofore described and wherein the patient's blood is drawn at the direction of the physician or other health care provider, either at the premises of the health care provider or elsewhere without the healthcare provider being present. In keeping with these embodiments, the kit may include a requisition form, the requisition form permitting indication of the type of test or tests to be conducted on the fluid to be collected by the device. In some embodiments, the requisition form lists a plurality of test types, and the healthcare provider need only indicate (such as with a check mark) the type of test desired. On any such form, space may be indicated for the health care provider to indicate any other sort of test desired to be conducted.

In a highly preferred embodiment of the invention, the fluid collector is an absorbent paper or glass fiber substrate that is coated with a saccharide, preferably a mono or di-saccharide and most preferably xylose. The saccharide should be present in contact with the substrate in an amount effective to inhibit triglycerides ordinarily present in the expected blood sample from binding to the fiber matrix. The substrate should be one that permits at least substantial separation of the red blood cell component of blood cells from other portions of the blood (i.e., serum). It is believed that the saccharide component permits more effective recovery of the serum components from the substrate sheet. The substrate may be coated only at the surface on one or both sides with the saccharide, but preferably the substrate is coated on internal surfaces as well as on the exterior surface. In one embodiment, 180 μl of a 5% solution of xylose is applied to the internal surface of the 0.8×7 cm substrate (such that substantially all of the substrate is wetted) and allowed to air dry. If the fluid collector is used in the device shown in FIGS. **9** and **10**, the blood cells will

remain near the end of the collection device (opposite the direction of arrow **913**) while the serum will wick toward the other end of the card. Upon receipt by a testing laboratory, a portion of the fluid collector may be excised and eluted. Preferably, the excised portion includes a portion of the collector "above" the terminal wicking point of the serum. One commercial product (Whatman GF/AVA paper) contains sodium, and it is believed that by excising filter paper above the terminal wicking point a consistent amount of sodium will be introduced into the eluted fluid. The device may be prepared by applying a solution of the saccharide to the substrate.

The glass fiber paper heretofore described comprises a mat of glass fibers that are at least substantially coated with polyvinyl alcohol. The fibers define a plurality of pores that have a pore size that, in preferred embodiments of the invention, is effective to at least substantially prevent lysing of red blood cells while permitting at least substantial separation of serum from red blood cells via differential wicking. Any suitable substrate that provides such a pore size and that permits such substantial separation in the absence of blood cell lysing may be used in conjunction with the invention. Preferably, the average pore size defines a fluid removal rating, as this term is used in conjunction with filtration technology, of 1.7 micron.

The invention enables venous blood analyte levels to be determined from capillary blood specimens. It is contemplated that in most embodiments the solution analyte level will be normalized to the venous blood level of the analyte, but it is also contemplated that the solution value may be normalized to capillary blood level (or for that matter a different blood level).

The databases discussed herein may be created and stored as computer files on a computer readable medium, such as a diskette, hard disk, CD-ROM, DVD-ROM, ROM chip or EPROM chip, or any other suitable medium as may be now known or hereinafter discovered. The tests for the analyte and normalizing analytes may be performed by any conventional or otherwise suitable technique now or hereinafter found to be suitable, and likewise the analyte and normalizing analyte (which may be discrete atoms, ions, compounds, biochemical materials, or properties) may be those specifically described herein or others as may be found suitable for use in conjunction with the invention.

The following examples are provided to illustrate the invention, but should not be construed as limiting the invention in scope unless otherwise indicated. Unless otherwise indicated in these examples, the measured analyte level was corrected using sodium as the sole normalizing analyte. The correction was made using a simple linear regression. It should be understood that more complex single variable and multivariate regressions may be used in conjunction with the invention, and thus the statistical techniques employed in these examples should be viewed as non-limiting.

Example 1

This example demonstrates the performance of the invention in the measurement of total cholesterol.

Fifteen patients were used to obtain blood specimens (micro-serum specimens) via venal puncture. Serum from each specimen was spotted and dried on filter paper with applied volumes ranging from approximately 8 to 16 μl. The number of spots for each blood specimen is listed in the column "No." in the table below. Each spot was eluted and measured for cholesterol and sodium. For each specimen for each patient, the normalized cholesterol level was calculated based on the level of a measured analyte in the fluid (cholesterol) and a normalizing analyte (sodium). The normalized cholesterol level was obtained according to the present invention using linear regression techniques to yield the following function: Normalized Cholesterol=Measured Cholesterol/((−0.003306)+0.9781×(Measured Sodium/13)), where 139 (mEq/L) is the population mean for sodium. The regression was calculated based on five direct measurements of the cholesterol level from the same blood sample, as listed in the column "Mean Serum Cholesterol." The mean average of the normalized cholesterol values for each patient is given in the column "mean normalized cholesterol" and the coefficient of variation of the normalized cholesterol levels obtained for each patient is listed in the column designated "Normalized Cholesterol CV %."

Patient	No.	Mean Serum Cholesterol	Mean Normalized Cholesterol	Normalize Cholesterol CV %
A	11	152.35	153.68	3.85
Ja	12	165.79	162.50	1.42
Il	14	180.93	180.47	4.61
C_a	12	186.20	182.28	0.70
Br	10	187.06	185.35	2.93
Mi	12	187.14	186.21	1.85
Gr	12	187.42	189.14	1.65
Ed	12	200.38	197.18	1.36
Tr	11	220.83	221.89	2.00
Bb	11	232.65	233.06	1.89
Ma	11	236.73	245.53	1.02
Jo	11	237.37	237.24	1.95
11	14	262.41	259.24	1.75
Kt	12	264.30	268.23	1.86
TT	13	269.36	273.53	2.79

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

$$\text{Mean Normalized Cholesterol} = -7.97 + 1.04 \times \text{Mean Serum Cholesterol},$$

with the correlation coefficient, expressed as R^2, being greater than 0.99.

Example 2

This example demonstrates the performance of the invention in the measurement of HDL.

The same dried spots from the same fifteen patients in Example 1 were used to obtain a measured value for HDL. The normalized HDL:level was obtained according to the present invention using linear regression techniques yielding the following function:

$$\text{Normalized HDL} = \text{HDL}/(0.0158 + 1.060 \times (\text{Sodium}/139)).$$

The following data was measured or calculated in the same manner as in Example 1.

Patient	No.	Mean Serum HDL	Mean Normalized HDL	Normalized HDL CV %
Il	14	45.77	47.03	2.35
A	11	46.05	47.77	2.17
Jo	11	47.40	48.50	2.12
Ja	12	48.87	53.22	2.23
11	14	49.07	48.15	1.68
Gr	12	49.64	52.45	1.62

-continued

Patient	No.	Mean Serum HDL	Mean Normalized HDL	Normalized HDL CV %
Mi	12	59.96	58.95	1.69
Br	10	57.20	55.83	2.66
Ed	12	71.00	71.09	0.92
Kt	12	73.08	72.46	1.53
TT	13	76.16	75.77	2.27
Ca	12	78.01	75.93	1.50
Bb	11	78.77	73.35	1.99
Ma	11	87.84	84.75	0.94
Tr	11	91.15	86.42	1.46

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

$$\text{Mean Normalized HDL}=8.15+0.87\pm\text{Mean Serum HDL},$$

with the correlation coefficient, expressed as R^2, being greater than 0.99.

Example 3

This example demonstrates the performance of the invention in the measurement of triglycerides (TG).

The same dried spots from the same fifteen patients in Example 1 were used to obtain a measured value for TG. The normalized TG level was obtained according to the present invention using linear regression techniques yielding the following function:

$$\text{Normalized TG}=\text{TG}/((-0.0136)+0.9307\times(\text{Sodium}/139)).$$

The following data was measured or calculated in the same manner as in Example 1.

Patient	No.	Mean Serum TG	Mean Normalized TG	Normalized TG CV %
Ca	12	37.63	38.76	1.95
Bb	11	46.86	48.55	1.75
A	11	48.75	50.16	2.73
Ja	12	49.68	49.94	3.31
Kt	12	52.15	48.19	1.32
Br	10	55.00	56.56	4.14
Ma	11	56.05	56.40	2.03
111	14	59.09	60.88	6.22
Ed	12	62.91	61.65	1.25
Tr	11	66.69	67.66	1.63
TT	13	68.76	72.14	13.37
Mi	12	71.84	72.63	1.62
Jo	11	109.28	107.10	2.27
11	14	117.31	112.24	5.03
Gr	12	139.47	136.74	2.13

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

$$\text{Mean Normalized TG}=3.36+0.95\times\text{Mean Serum TG},$$

with the coefficient, expressed as R^2, being greater than 0.995.

Example 4

This example demonstrates the performance of the invention in the measurement of LDL. The same observations from the same fifteen patients in Example 1, 2 and 3 were used to calculate a value for LDL in serum and a value for LDL in MSS according to the Friedewald formula:

$$\text{Mean Serum LDL}=\text{Mean Serum Cholesterol}-\text{Mean Serum HDL}-\text{Mean Serum TG}/5$$

$$\text{Mean Normalized LDL}=\text{Mean Normalized Cholesterol}-\text{Mean Normalized HDL}-\text{Mean Normalized TG}/5,\text{ respectively.}$$

The following data was calculated (mean serum LDL was calculated from the mean values reported in Examples 1-3)

Patient	No.	Mean Serum LDL	Mean Normalized LDL	Normalized LDL CV %
A	11	96.55	95.30	5.36
Ca	12	100.66	98.82	1.17
Ja	12	106.98	99.22	2.32
Gr	12	109.88	109.00	2.19
Mi	12	115.81	112.90	2.80
Tr	11	116.35	121.21	3.11
Ed	12	116.80	113.76	1.98
Br	10	118.86	118.21	3.23
11	14	123.34	119.75	5.72
Ma	11	137.68	149.45	1.72
Bb	11	144.51	150.01	2.12
Jo	11	168.11	167.55	2.66
Tt	13	179.45	183.33	3.33
Kt	12	180.78	186.13	2.19
11	14	189.88	189.54	1.89

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

$$\text{Mean Normalized LDL}=-8.16+1.07\times\text{Mean Serum LDL},$$

with the correlation, expressed as R^2, being equal to 0.98.

Example 5

This example demonstrates the performance of the invention in the measurement of total cholesterol.

One hundred thirty-two patients were used to obtain blood via venal puncture (venous blood specimens) and by pricking their fingers (capillary blood specimens). Capillary blood was spotted on xylose-coated Whatman GF/AVA filter paper, using a device similar to that shown in FIG. **9**. Capillary blood specimens were dried and the portion of the filter paper which contained separated serum was cut out and eluted. Eluate from each specimen was measured for cholesterol and sodium. The normalized cholesterol level was obtained according to the present invention using a variable formula: Normalized Cholesterol=Measured Cholesterol/(A+B×(Measured Sodium/139)). In this equation, A and B were scalar values that were periodically recalculated based on the "tare" procedure heretofore described, whereby a regression for six patients was calculated and the A and B values from this regression were used to calculate normalized cholesterol values for specimens analyzed before the next tare period.

Actual (directly measured in venous blood) and calculated normalized cholesterol valves for these patients are given below.

Patient	Serum Cholesterol	Normalized Cholesterol
1	172.68	157.54
2	149.25	154.61
3	176.81	175.60
4	189.78	187.41
5	170.38	173.03
6	189.67	188.80
7	130.52	128.80
8	266.76	276.31
9	151.29	152.49
10	219.86	211.23
11	242.00	251.07
12	232.41	230.66
13	173.09	176.48
14	190.89	190.86
15	264.47	260.46
16	236.18	244.49
17	272.58	279.76
18	240.29	228.83
19	169.32	166.57
20	192.02	195.03
21	239.83	235.33
22	225.13	225.13
23	169.40	156.05
24	197.93	183.67
25	151.59	146.26
26	235.43	247.88
27	178.84	170.79
28	196.40	191.34
29	240.99	230.52
30	171.53	173.95
31	229.43	229.43
32	217.54	223.84
33	187.23	183.58
34	175.68	173.95
35	174.69	172.34
36	251.23	249.20
37	203.70	185.98
38	123.30	114.96
39	136.04	127.97
40	251.33	243.27
41	216.14	218.02
42	145.14	156.86
43	208.58	203.43
44	250.25	245.07
45	235.76	250.40
46	193.19	187.83
47	211.75	223.38
48	221.15	226.04
49	199.41	196.35
50	249.35	259.44
51	166.46	165.63
52	154.64	151.56
53	187.36	190.37
54	256.78	260.40
55	230.59	222.39
56	208.57	224.14
57	183.92	181.28
58	159.73	156.20
59	155.31	153.59
60	205.29	197.61
61	204.49	198.97
62	219.21	221.45
63	122.83	114.88
64	175.13	176.48
65	201.35	211.70
66	216.66	209.09
67	227.50	231.96
68	151.28	153.23
69	130.10	128.40
70	175.95	173.45
71	182.38	183.21
72	201.03	195.89
73	175.86	189.73
74	146.10	149.88
75	116.17	103.88
76	193.58	197.59
77	291.91	296.11
78	184.93	185.49
79	145.82	141.34
80	182.73	180.78
81	175.84	170.03
82	148.99	151.67
83	212.79	213.40
84	228.82	225.39
85	218.44	229.26
86	169.43	173.84
87	151.43	157.96
88	217.96	218.63
89	239.39	244.11
90	148.62	152.86
91	136.81	132.60
92	119.13	113.31
93	121.10	119.61
94	165.31	163.34
95	111.65	132.34
96	190.25	184.44
91	201.78	206.49
98	133.26	137.69
99	225.84	221.61
100	244.66	230.25
101	164.72	168.10
102	150.75	146.82
103	163.51	110.41
104	196.06	198.89
105	213.32	206.01
106	186.62	183.13
107	163.46	162.71
108	244.58	250.24
109	231.82	231.32
110	171.94	172.27
111	201.12	209.36
112	205.41	209.00
113	157.54	156.02
114	191.41	190.59
115	192.20	197.31
116	193.52	183.12
117	257.83	248.49
118	178.32	171.44
119	203.64	209.32
120	210.36	230.25
121	207.14	220.04
122	200.05	205.38
123	216.34	219.09
124	190.10	179.14
125	293.34	272.48
126	228.57	226.02
127	111.60	174.88
128	142.80	148.94
129	197.16	205.05
130	220.50	218.43
131	220.32	231.50
132	255.18	255.23

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

$$\text{Normalized Cholesterol} = -1.16 + 1.00 \times \text{Serum Cholesterol},$$

with the correlation coefficient, expressed as R2, being 0.966.

Example 6

This example demonstrates the performance of the invention in the measurement of HDL. The dried spots and venous blood specimens from the same one hundred thirty-two

patients in Example 5 were used to measure HDL in capillary blood and compare it to a measured value for HDL in venous blood. The normalized HDL level in capillary blood was obtained according to the present invention using a formula: Normalized HDL=Measured HDL/(A+B×(Measured Sodium/139)), where A and B were obtained as previously described. The following results were observed.

Patient	Serum HDL	Normalized HDL
1	58.90	61.12
2	41.28	42.33
3	38.54	39.15
4	48.84	46.19
5	61.56	54.98
6	52.68	48.79
7	47.69	45.15
8	34.69	39.49
9	57.45	56.32
10	38.00	36.33
11	47.53	42.14
12	60.04	58.94
13	36.08	37.35
14	46.09	48.37
15	42.22	42.82
16	34.70	38.98
17	55.76	55.79
18	21.16	24.53
19	55.33	55.69
20	44.66	42.65
21	83.26	81.00
22	44.33	46.14
23	40.71	40.69
24	47.24	43.98
25	49.46	47.71
26	44.37	43.30
27	50.16	48.34
28	55.49	61.30
29	58.90	61.12
30	41.28	42.33
31	38.54	39.15
32	48.84	46.19
33	61.56	54.98
34	52.68	48.79
35	47.69	45.15
36	34.69	39.49
37	57.45	56.32
38	38.00	36.33
39	47.53	42.14
40	60.04	58.94
41	36.08	37.35
42	46.09	48.37
43	42.22	42.82
44	34.70	38.98
45	55.76	55.79
46	21.16	24.53
47	55.33	55.69
48	44.66	42.65
49	83.26	81.00
50	44.33	46.14
51	40.71	40.69
52	47.24	43.98
53	49.46	47.71
54	44.37	43.30
55	50.16	48.34
56	55.49	61.30
57	49.27	45.94
58	51.73	51.78
59	38.07	36.98
60	38.22	38.49
61	43.57	45.05
62	54.16	51.46
63	38.66	34.13
64	50.14	48.65
65	57.94	54.11
66	46.02	44.67
67	49.21	52.36

-continued

Patient	Serum HDL	Normalized HDL
68	43.15	45.31
69	37.20	38.42
70	49.66	50.00
71	63.00	65.28
72	79.92	79.17
73	37.12	44.57
74	59.14	60.35
75	32.49	28.57
76	56.08	59.37
77	64.22	70.04
78	46.54	48.66
79	37.68	37.28
80	75.41	74.70
81	44.06	44.73
82	40.65	40.88
83	93.40	91.97
84	40.97	47.04
85	69.63	75.17
86	36.13	38.81
87	34.88	36.42
88	43.90	49.40
89	63.29	66.41
90	49.21	49.65
91	29.54	31.27
92	49.30	49.87
93	35.82	34.39
94	49.66	51.20
95	39.01	39.79
96	36.92	34.49
97	43.40	43.45
98	48.70	45.97
99	42.15	41.04
100	59.09	55.11
101	49.46	47.04
102	33.36	29.81
103	49.36	47.93
104	43.02	39.12
105	39.81	41.06
106	60.29	56.62
107	59.84	55.33
108	84.77	82.31
109	55.20	55.72
110	54.77	56.06
111	69.16	67.30
112	38.18	40.50
113	37.11	36.49
114	51.31	49.24
115	39.69	42.54
116	61.17	56.56
117	29.94	30.25
118	75.50	77.62
119	56.94	57.49
120	68.89	71.30
121	37.89	40.82
122	73.57	72.14
123	78.31	78.16
124	48.88	47.45
125	83.96	79.26
126	95.12	92.48
127	51.44	52.50
128	38.88	38.10
129	41.70	44.58
130	47.80	46.24
131	56.42	59.35
132	55.14	56.98

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

$$\text{Normalized HDL}=2.47+0.953\times\text{Serum HDL},$$

with the correlation coefficient, expressed as R^2, being greater than 0.96.

Example 7

This example demonstrates the performance of the invention in the measurement of triglycerides (TG). The dried spots and venous blood specimens from the same one hundred thirty-two patients in Example 5 were used to measure TG in capillary blood and compare it to a measured value for TG in venous blood. The normalized TG level in capillary blood was obtained according to the present invention using the formula: Normalized TG=Measured TG/(A+B×(Measured Sodium/139)), where A and B were obtained as previously described. The following results were observed.

Patient	Serum TG	Normalized TG
1	73.24	55.65
2	97.89	97.31
3	45.26	38.38
4	70.31	60.30
5	119.71	119.33
6	105.97	100.56
7	77.47	73.30
8	220.18	236.94
9	191.79	203.18
10	177.10	177.03
11	112.19	116.71
12	73.24	55.65
13	97.89	97.31
14	45.26	38.38
15	70.31	60.30
16	119.71	119.33
17	157.70	164.69
18	122.09	124.56
19	66.86	63.24
20	138.31	151.08
21	146.08	137.36
22	95.85	97.05
23	77.27	60.69
24	85.44	82.87
25	86.25	77.32
26	112.51	110.68
27	176.25	184.16
28	190.63	189.57
29	95.17	98.92
30	98.52	98.76
31	102.13	97.07
32	117.77	128.91
33	123.08	125.56
34	135.72	132.69
35	76.46	71.14
36	230.90	210.77
37	80.41	67.66
38	99.43	85.63
39	86.87	91.07
40	125.01	120.98
41	362.90	322.04
42	132.98	118.47
43	83.21	75.43
44	52.45	53.34
45	53.91	50.52
46	349.76	357.87
47	135.25	139.57
48	209.20	208.33
49	374.36	386.86
50	74.90	79.81
51	395.31	399.34
52	56.38	54.87
53	217.08	258.78
54	52.83	71.35
55	136.53	144.81
56	115.98	118.45
57	78.41	62.45
58	70.38	65.13
59	91.00	68.59
60	180.98	179.72
61	163.32	188.88
62	72.16	65.05
63	102.89	101.45
64	50.24	49.05
65	184.45	195.42
66	183.07	194.25
67	65.28	65.04
68	111.40	109.43
69	67.25	87.27
70	74.92	72.25
71	100.19	105.33
72	136.82	132.52
73	119.29	129.90
74	119.76	119.83
75	121.90	125.90
76	75.55	80.65
77	74.44	89.06
78	226.78	243.05
79	71.19	78.23
80	98.89	93.66
81	127.93	135.56
82	333.65	352.31
83	97.18	91.96
84	139.77	133.20
85	73.23	72.05
86	160.00	148.64
87	131.69	133.49
88	69.07	66.79
89	271.22	248.43
90	91.86	98.00
91	231.14	224.76
92	153.65	171.85
93	115.95	107.16
94	263.50	257.68
95	95.38	92.85
96	143.96	125.21
97	110.10	131.36
98	97.72	93.75
99	158.22	151.23
100	123.80	127.26
101	279.56	271.61
102	192.26	176.02
103	59.41	59.23
104	197.04	186.32
105	182.29	170.98
106	96.16	91.53
107	80.46	72.56
108	65.55	68.16
109	215.37	210.92
110	186.09	191.14
111	96.41	96.52
112	78.68	80.54
113	83.96	73.13
114	207.32	208.03
115	37.41	37.32
116	103.17	93.38
117	193.21	210.21
118	119.46	103.27
119	67.57	58.99
120	119.56	117.34
121	75.42	52.90
122	311.18	315.01
123	67.72	68.28
124	127.36	129.28
125	59.82	64.57
126	85.54	83.90
127	43.24	41.49
128	85.09	78.05

-continued

Patient	Serum TG	Normalized TG
129	95.15	99.45
130	92.21	75.05
131	72.46	88.51
132	56.52	57.13

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

Normalized TG=−2.5+1.01×Serum TG,

with the coefficient, expressed as R^2, being 0.98.

Example 8

This example demonstrates the performance of the invention in the measurement of LDL. The same observations from the same one hundred thirty-two patients in Example 5, 6 and 7 were used to calculate a value for LDL in serum and a value for LDL in MSS according to the Friedewald formula:

Serum LDL=Serum Cholesterol−Serum HDL−Serum TG/5

Normalized LDL=Normalized Cholesterol−Normalized HDL−Normalized TG/5.

The following results were calculated:

Patient	Serum LDL	Normalized LDL
1	110.85	101.31
2	97.93	101.60
3	109.82	103.89
4	110.51	112.89
5	108.21	107.74
6	126.07	121.49
7	49.76	54.13
8	173.19	173.53
9	78.72	77.71
10	129.52	119.61
11	174.74	180.58
12	149.04	140.09
13	98.72	101.13
14	115.22	115.25
15	187.28	180.56
16	146.29	158.21
17	195.20	200.00
18	167.30	161.22
19	101.87	99.92
20	122.26	127.45
21	168.27	164.79
22	149.27	148.28
23	100.92	85.06
24	129.40	115.93
25	92.09	88.71
26	165.02	175.92
27	114.88	104.61
28	94.16	90.62
29	154.94	142.87
30	114.95	117.39
31	144.71	148.13
32	152.62	164.12
33	105.78	111.48
34	105.62	106.95
35	101.99	102.99
36	143.96	145.31
37	119.65	105.96
38	68.65	63.55
39	78.02	75.16
40	180.50	174.23

-continued

Patient	Serum LDL	Normalized LDL
41	110.10	109.10
42	72.00	80.58
43	124.52	118.94
44	140.68	128.72
45	165.02	178.66
46	92.96	83.43
47	145.15	156.72
48	133.07	131.63
49	105.59	101.08
50	177.71	184.34
51	102.56	101.25
52	91.72	95.09
53	123.82	129.64
54	194.21	203.38
55	144.23	138.11
56	120.42	125.06
57	120.22	122.32
58	87.42	84.13
59	107.19	106.80
60	130.18	120.03
61	124.30	115.07
62	151.99	156.98
63	61.89	58.86
64	111.54	110.38
65	128.43	143.14
66	150.60	143.35
67	150.93	153.10
68	84.28	81.94
69	68.95	66.01
70	101.92	98.27
71	104.27	101.79
72	106.22	98.91
73	93.38	96.56
74	72.72	73.88
75	63.90	56.58
76	111.92	111.11
77	160.96	155.60
78	118.95	118.43
79	80.19	77.42
80	92.68	91.67
81	99.78	95.56
82	82.00	84.09
83	105.58	108.07
84	133.61	128.66
85	130.44	134.49
86	87.08	90.07
87	85.82	87.16
88	150.87	147.80
89	123.40	126.22
90	80.33	84.64
91	78.47	76.28
92	107.81	97.16
93	65.74	66.47
94	84.06	81.90
95	53.88	67.10
96	97.42	95.63
97	119.93	127.83
98	72.69	79.87
99	144.28	143.36
100	149.12	140.94
101	96.03	102.76
102	101.29	102.50
103	101.09	108.85
104	109.96	117.58
105	136.29	126.72
106	107.05	107.20
107	87.89	91.28
108	143.01	153.30
109	135.15	134.00
110	109.68	108.74
111	117.33	123.38
112	128.59	126.45
113	96.53	98.88
114	126.58	129.56
115	128.60	131.36
116	117.26	116.58
117	165.65	155.24

-continued

Patient	Serum LDL	Normalized LDL
118	89.27	80.15
119	121.23	125.97
120	129.51	146.03
121	152.74	162.44
122	117.83	124.94
123	121.01	125.32
124	122.19	111.80
125	190.94	178.21
126	118.95	115.83
127	108.85	110.95
128	76.17	84.41
129	116.38	120.15
130	116.39	113.82
131	134.85	142.75
132	173.09	172.40

A comparative linear regression was generated for the data points collected in this Example. The linear fit followed the following equation:

$$\text{Normalized LDL} = -0.25 + 1.00 \times \text{Serum LDL},$$

with the correlation, expressed as R^2, being equal to 0.96.

It is thus seen that the invention provides a method for determining the level of an analyte in a specimen.

While particular embodiments to the invention have been described herein, the invention is not limited thereto, but to the contrary should be deemed defined by the full scope of the appended claims. All references and prior and co-pending applications cited herein are hereby incorporated by reference in their entireties.

What is claimed is:

1. A kit comprising: a fluid collector with an absorbent substrate coated with a saccharide, said substrate comprising a mat of glass fibers at least substantially coated with polyvinyl alcohol, said fibers defining a plurality of pores, the pores in said mat having a pore size effective to at least substantially prevent lysing of red blood cells while permitting at least substantial separation of serum from red blood cells via differential wicking and a superstrate, said fluid collector being generally fixed with respect to said superstrate, said superstrate having an aperture defining a blood receiving opening and permitting access to said fluid collector, and further comprising a desiccant, said desiccant being present in an amount effective to provide a desiccating protective effect on a blood fluid specimen.

2. A method for collecting a specimen from a patient, comprising: providing a fluid collection device with a fluid collector, said fluid collector comprising an absorbent substrate coated with a saccharide, said substrate comprising a mat of glass fibers at least substantially coated with polyvinyl alcohol, said fibers defining a plurality of pores, the pores in said mat having a pore size effective to at least substantially prevent lysing of red blood cells while permitting at least substantial separation of serum from red blood cells via differential wicking; and a superstrate fixed with respect to the fluid collector; and allowing said patient to bleed onto said collector until at least a predetermined adequate amount of blood has been deposited onto said collector, further comprising sending the fluid collection device to a remote location for testing.

3. The method according to claim **2**, comprising sealing the collector in a barrier film pouch.

4. The method according to claim **3**, said barrier film pouch comprising a laminar structure that includes a polyester film and an aluminum foil film.

5. The method according to claim **3**, further comprising adding a desiccant to said barrier film pouch, said desiccant being present in an amount effective to provide a desiccating protective effect on a blood fluid specimen.

6. The method according to claim **5**, said desiccant comprising silica.

7. The method according to claim **2**, further comprising receiving a results reporting form after sending said fluid collection device to a remote location for testing.

8. The method according to claim **2**, further comprising receiving a results reporting form after sending said fluid collection device to a remote location for testing.

9. A method for collecting a specimen from a patient, comprising: providing a fluid collection device with a fluid collector, said fluid collector comprising an absorbent substrate coated with a saccharide, said substrate comprising a mat of glass fibers at least substantially coated with polyvinyl alcohol, said fibers defining a plurality of pores, the pores in said mat having a pore size effective to at least substantially prevent lysing of red blood cells while permitting at least substantial separation of serum from red blood cells via differential wicking and a superstrate fixed with respect to the fluid collector; and allowing said patient to bleed onto said collector until at least a predetermined adequate amount of blood has been deposited onto said collector, further comprising indicating a type of test desired on a requisition form.

10. A method for collecting a specimen, comprising: receiving a fluid collection device with a fluid collector, and a superstrate fixed with respect to the fluid collector, said fluid collector comprising a mat of glass fibers at least substantially coated with polyvinyl alcohol, said fibers defining a plurality of pores, the pores in said mat having a pore size effective to at least substantially prevent lysing of red blood cells while permitting at least substantial separation of serum from red blood cells via differential wicking; and bleeding onto said collector until at least a predetermined adequate amount of blood has been deposited onto said fluid collection device, further comprising sending the fluid collection device to a remote location for testing, comprising sealing the fluid collection device in a barrier film pouch.

11. The method according to claim **10**, said barrier film pouch comprising a laminar structure that includes a polyester film and an aluminum foil film.

12. The method according to claim **10**, further comprising adding a desiccant to said barrier film pouch, said desiccant said desiccant being present in an amount effective to provide a desiccating protective effect on a blood fluid specimen.

13. The method according to claim **12**, said desiccant comprising silica.

14. A method for providing a test and test results to a patient, comprising: providing a kit, said kit comprising: a fluid collection device comprising; a fluid collector with an absorbent substrate coated with a saccharide, said substrate comprising a mat of glass fibers at least substantially coated with polyvinyl alcohol, said fibers defining a plurality of pores, the pores in said mat having a pore size effective to at least substantially prevent lysing of red blood cells while permitting at least substantial separation of serum from red blood cells via differential wicking and a superstrate, said fluid collector being generally fixed with respect to said superstrate, said superstrate having an aperture defining a blood receiving opening and permitting access to said fluid collector; a lancet; a desiccant, said desiccant being present in an amount effective to provide a desiccating protective effect on a blood fluid specimen collected in said device; and a barrier film pouch sized for receiving said fluid collection device and said desiccant; and results from a previous test of the patient.

* * * * *