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(54) TESTING DEVICE FOR IDENTIFYING ANTIGENS AND ANTIBODIES IN BIOFLUIDS

TESTVORRICHTUNG ZUR IDENTIFIKATION VON ANTIGENEN UND ANTIKÖRPERN IN BIOFLÜSSIGKEITEN

DISPOSITIF D'ANALYSE DESTINÉ À IDENTIFIER DES ANTIGÈNES ET DES ANTICORPS DANS DES BIOFLUIDES

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Description

FIELD OF THE INVENTION

[0001] The present invention is directed to the identification of antigens and antibodies within a biofluid. While the invention will be described with specific reference to its use in determining a person's blood type, it is to be appreciated that other applications of the invention are also envisaged.

BACKGROUND TO THE INVENTION

[0002] Blood is essential for sustaining living tissue, with the most important roles of supplying oxygen and other soluble nutrients, immune protection and metabolic turnover. While it is a tissue in its own right, blood in a chemical sense can be considered as a stable, highly packed colloid suspension made of red blood cells (erythrocytes, 4-6 million/mL, 6-8 μm), white cells (leukocytes, 4000-6000 /mL, 10-21 μm), platelets (150,000-400,000 /mL, 2-5 μm) carried within a fluid solution (serum) containing a host of biomolecules (eg albumins, fatty acids, hormones), metabolites and electrolytes. A subset of these biomolecules, such as the binding proteins responsible for tissue immunity (antigens) and blood type, are directly adsorbed onto the surface of blood cells. Common portable testing methods for blood include analysis of glucose content, cholesterol, metabolic panel (sodium, potassium, bicarbonate, blood urea nitrogen, magnesium, creatine, calcium, triglycerides), microbial and disease markers and protein molecular profile (liver, prostate). Surprisingly, and in spite of its vital importance, there are no convenient low cost disposable tests available for "on the spot" analysis of blood type. Blood samples are typically outsourced to an analytical laboratory. Reliable low cost tests which are able to instantaneously and reliably provide critical blood analysis without the requirement of sophisticated laboratory analytical instrumentation such as chromatographic and spectroscopic methods, would be invaluable for improving human health in developing countries, where economic resources are limited. Blood analysis is also important in non-human applications, such as veterinary medicine, where there is a demand for low cost and versatile devices suitable for field use.

[0003] Document WO 87/07304 A1 discloses a cell detection system and method, wherein a membrane uses capillary action for detection particles.

[0004] The article "Analytical Techniques for Cell Fractions" by N.G. Anderson, Analytical Biochemistry 38, p. 175-189 (1970) discloses an experimental method of blood typing with strips of filter paper.

DESCRIPTION OF INVENTION

[0005] With this in mind, the invention is directed to a method for identifying an antigen according to claim 1. Further specific aspects of the present invention are discussed below:

According to one aspect of the present invention, there is provided a testing device for identifying an antigen or antibody within a biofluid sample including;
a substrate having a hydrophilic surface thereon;
the surface including a collection zone, and at least one detection zone extending therefrom;
wherein the biofluid sample can be mixed with a specific antigen or antibody, and deposited on the collection zone and transferred by capillary action to the detection zone;
the antigen or antibody in the biofluid sample reacting with an appropriate said antibody or antigen thereby resulting in a visual indication within the detection zone.

[0006] According to another aspect of the present invention there is provided a testing device for identifying an antigen or antibody within a biofluid sample including;
a substrate having a hydrophilic surface thereon;
the surface including a collection zone, and at least one detection zone extending therefrom;
the detection zone having an antibody or antigen immobilised therein;
wherein the biofluid sample can be deposited on the collection zone and transferred by capillary action to the detection zone;
the antigen or antibody in the biofluid sample reacting with an appropriate said antibody or antigen within the detection zone thereby resulting in a visual indication therein.

[0007] The substrate may be formed from paper or other cellulosic materials. Alternatively, the substrate may include a chromographic layer thereon or may be a wettable porous medium.
The biofluid sample being tested may preferably be blood, and the visual indication may be due to an agglutination of the blood upon reaction with a specific antibody resulting in reduced wicking and/or separation of the blood in the detection zone.

[0008] The substrate surface may have a hydrophilic microfluidic channel pattern thereon defining the collection zone and the detection zone. Preferably, a plurality of detection zones may extend from the collection zone.

[0009] It has been found that red blood cell agglutination, triggered by specific antigen interaction, drastically decreases blood wicking and dispersion on paper or chromatographic media. The agglutination process also considerably enhances the chromatographic separation (elution) of the individual blood components, especially the red blood cells from the serum. The testing device according to the present invention can allow direct analysis of blood cells because of this visual indication. This can be performed instantaneously, either with a detection/reporting system built-in to the device or in conjunc-

tion with other off-line analytical equipment. The testing device may also allow for the identification and quantification of specific biomolecules (eg antigens and antibodies) based on induced coagulation, followed by the wicking and elution (separation) of the soluble protein fraction from the blood sample onto the porous substrate. The blood colloids, whose coagulation directly affects their wicking/ separation, can either be present in the fluid of interest, such as the red blood cells of blood, or introduced as nanoparticles (gold, silver, micro-silica, zeolite, titanium dioxide and the like). In the latter case, the nanoparticle is typically covered with the specific counter-biomolecule or molecule of interest used as sensitive reporter component. The colloid particles may be of a size ranging from 1 nm to 100 μm and may be introduced into the biofluid being analysed.

[0010] In the application of the present invention for determining the type of blood group, the present invention may determine the antigens present within a blood sample, the antigens determining whether the blood type is type A, B, O, AB and Rhesus +/-. Antibodies A, B and D (Rhesus) are deposited into separate detection zones. It may also be preferable to include an untreated control zone in one of the detection zones. A drop of blood is then deposited on the central collection zone, the blood sample being transferred by capillary action to each of the detection zones. When the blood sample contacts an appropriate antibody, the reaction of the red blood cells antigen with its corresponding antibody results in agglutination or coagulation of the red cells. This agglutination results in a drop in velocity of the movement of the blood sample along the microfluidic channel providing the detection zone and separation of the red blood cells from the serum. The velocity of the blood samples travelling along other detection zones with non-specific antibodies is unaffected. This visual contrast facilitates easy and rapid identification of the blood type of the blood sample.

[0011] The applicant has developed a low cost paper based microfluidic system which is described in International patent application no. PCT/AU2009/000889 details of which are incorporated herein by reference. The microfluidic systems described in this application utilise a paper based substrate, with the described fabrication methods producing hydrophilic microfluidic channels on the paper based substrates. It should be noted that the term paper is used in this application to refer to all cellulosic materials including woven fabrics and non-woven cellulosic material as well as paper. The microfluidic systems described in these applications can be readily adapted for the purpose of the present invention.

[0012] The testing device according to the present invention may also be used to detect illness as a result of blood cell malfunction on the blood cells being of abnormal shape as is the case with malaria. Alternatively, the testing device according to the present invention may be used to detect illness by identifying the presence of an antigen, antibody, virus (such as HIV, influenza) or protein.

[0013] In the International application, the microfluidic channels are fabricated by printing a hydrophobic agent on the substrate surface to define a peripheral edge of the microfluidic channels. According to the present invention, the antibody or antigen may also be printed within the microfluidic channels. The technology used, namely ink jet printing technology, may also be used to print the antigen or antibody within the microfluidic channels.

[0014] According to a further aspect of the present invention, there is provided a method for identifying an antigen or antibody within a biofluid sample, including:

mixing the biofluid sample with a specific antigen or antibody,
depositing the mixed biofluid sample on a collection zone of a testing device including a substrate having a hydrophilic surface thereon, the surface including said collection zone and at least one detection zone extending therefrom, the biofluid sample being transferred by capillary action to the detection zone; and
identifying the antigen or antibody by a resultant visual indication within the detection zone arising where the antigen or antibody in the biofluid sample reacts with an appropriate said antibody or antigen.

[0015] According to yet another aspect of the present invention, there is provided a method for identifying an antigen or antibody within a biofluid sample, including:

depositing the biofluid sample on a collection zone of a testing device including a substrate having a hydrophilic surface thereon, the surface including said collection zone and at least one detection zone extending therefrom, the detection zone having an antibody or antigen immobilised therein, the biofluid sample being transferred by capillary action to the detection zone; and
identifying the antigen or antibody by a resultant visual indication arising where the antigen or antibody in the biofluid sample reacts with an appropriate said antibody or antigen within the detection zone.

BRIEF DESCRIPTION OF THE DRAWINGS

[0016] It will be convenient to further describe the invention with respect to the accompanying drawings which illustrate preferred embodiments of the testing device according to the present invention. Other embodiments of the invention are possible, and consequently, the particularity of the accompanying drawings is not to be understood as superseding the preceding description of the invention.

[0017] In the drawings:

Figure 1 shows a testing device according to the present invention used to determine B+ blood;
Figure 2 shows a testing device according to the present invention used to determine O+ blood;

Figure 3 shows the testing device according to the present invention using AB+ and B+ blood;
Figure 4 is the testing device according to the present invention showing blood wicking and blood separation as a function of time;
Figure 5 shows a testing device according to the present invention incorporating a valve;
Figure 6 A-F shows the operation of a testing device according to the present invention incorporating valves and switches;
Figure 7 are schematic representations of testing devices according to the present invention adapted for testing different blood types; and
Figure 8 shows the testing device of Figure 7 showing the separation of red blood cells from the blood serum.

DETAILED DESCRIPTION OF THE INVENTION

[0018] The applicants have discovered that blood agglutination mediated by specific antibody-antigen interactions drastically affects its separation behaviour on contact with paper or any thin layer chromatographic surface. The invention relies on this biochemical phenomenon to control the rate of wicking and separation, which enables (i) identification and quantitative assessment of a specific antibody/antigen, (ii) blood typing, and potentially (iii) identification of blood-borne pathogens as a disease diagnostic. The present invention is intended predominantly for applications in human and veterinary medicine and biotechnology.

[0019] Standard techniques for detection of blood agglutination are traditionally manual, involve dispensing of antibodies on a glass slide and microscopic visualization. However, the visualisation of agglutination is often subjective, and its automation requires a bank of sophisticated analytical equipment. The present invention provides a single-step blood test that simplifies and circumvents these difficulties.

[0020] One application of the invention involves a two step process in which the blood sample is first coagulated/agglutinated by combining it with the specific antibody/antigen of interest, followed by its deposition on the analytical substrate (eg Non-woven paper or porous mesh) on which the sample wicking and separation by elution/chromatography is measured, directly or indirectly. These two mixing steps can be enhanced and more accurately performed by mixing on paper substrates using built-in valving and channelling control features.

[0021] A second application of the invention involves a single-step process in which the biofluid/sample is deposited directly on the substrate/device which has previously been treated with the specific antigen/antibody. For this process, the analyte samples simultaneously coagulate and elude on the same substrate. The measured elution velocity and the extent of sample separation are directly related to the extent of coagulation, enabling the concentration of the biomolecule to be detected and quantified at the same time.

[0022] Both applications of the invention can be applied to a test device made of paper or any non-woven chromatographic surface, which are relatively cost effective. These substrates are also able to be modified with the use of advanced printing techniques to create microfluidic features composed of hydrophobic materials, as previously described in the applicant's International application no. PCT/AU2009/000889. Combined with methods of direct antibody deposition using printing, manufacture and placement of antibody reagent can enable very accurate spatial control of blood flow within the paper substrate.

EXAMPLES

Example 1: Sequential agglutination/coagulation of blood followed by wicking on paper: B+ (two step process) (See Figure 1)

[0023] Antibody A and B (Epiclone™ Anti-A, Anti-B, and Anti-D; CSL, Australia) solutions were used. Anti-A and Anti-B come as blue and yellow colour reagents, respectively. 'B+' blood was used in this study. The blood sample was supplied into plastic vials with anti-coagulant. 'B+' blood was separately mixed with pure Anti-A and Anti-B (as received) to prepare 100μL solution. Paper strips (70mmx2mm) were made from Whatman#4 filter paper on which 2 mm unit marks were printed. The paper strips were soaked into phosphate buffer saline (PBS). Excess PBS was removed from the paper strips using standard blotting papers (Drink Coster Blotting, 280 GSM). The paper strips were then placed on Reflex Paper (80 GSM). 20 μL of every mixed solution was dispensed at the centre of paper strip using a calibrated micro-pipette. Pictures were taken after 4 minutes wicking.

[0024] It can be seen that:

B+ blood mixed with the solution of antibody A wicked and did not separate upon mixing and paper elution/wicking.
B+ blood mixed with the solution of antibody B wicked and STRONGLY separated (red cells from serum) and showed wicking.
B+ blood mixed with the solution of antibody D (Rhesus +) wicked and STRONGLY separated (red cells from serum).

[0025] A blood sample agglutinated/coagulated upon contact with its specific antibodies separated/eluded upon contact with paper (here Blood B+ with Anti-B and Anti-D antibodies).

[0026] A blood sample upon contact with non-specific antibody (here Blood B+ with Anti-A) does not agglutinate and does not separate/elute upon contact with paper.

[0027] This dramatic difference in elution/separation of blood/antibody mixing can be used to communicate

specific agglutination and therefore can be used to identify blood typing.

Example 2: Sequential agglutination/coagulation of blood followed by wicking on paper: O+ (two step process) (See Figure 2)

[0028] Antibody A and B (Epiclone™ Anti-A, Anti-B and Anti-D; CSL, Australia) solutions were used. Anti-A and Anti-B come as blue and yellow colour reagents, respectively. 'O+ blood was used in this study. The blood sample was supplied into plastic vials with anti-coagulant. 'O+' blood was separately mixed with Anti-A and Anti-B to prepare 100μL solution. Paper strips (70mmx2mm) were made from Whatman#4 filter paper on which 2 mm unit marks were printed. The paper strips were soaked into phosphate buffer saline (PBS). Excess PBS was removed from the paper strips using standard blotting papers (Drink Coster Blotting, 280 GSM). The paper strips were then placed on Reflex Paper (80 GSM). 20 μL of every mixed solution was dispensed at the centre of paper strip using a calibrated micro-pipette. Pictures were taken after 4 minutes wicking.

[0029] It can be seen that:

- O+ blood mixed with the solution of antibody A wicked and did not separate upon mixing and paper elution/wicking.
- O+ blood mixed with the solution of antibody B wicked and did not separate (red cells from serum) and showed wicking.
- O+ blood mixed with the solution of antibody D (Rhesus +) wicked and STRONGLY separated (red cells from serum).

[0030] A blood sample agglutinated/coagulated upon contact with its specific antibodies separated/eluded upon on contact with paper (here Blood O+ with Anti-D antibodies).

[0031] A blood sample upon contact with non-specific antibody (here Blood O+ with Anti-A and Anti-B) does not agglutinate and does not separate/elute upon contact with paper.

[0032] This dramatic difference in elution/separation of blood/antibody mixing can be used to communicate specific agglutination and therefore can be used to identify blood typing.

Example 3: Simultaneous agglutination/coagulation of blood followed by wicking on paper: Effect of antigen concentration (one step process) (See Figure 3)

[0033] In another embodiment of the invention, the paper is first treated with specific antibodies, dried or conditioned before been exposed to a sample of pure blood. This example provides a single step treatment in which the only requirement is to deposit a drop of blood on the paper. This example also illustrates the effect of diluting the antibody solution on the wicking and separation performance of blood on paper. Antibody dilution affects the ratio blood (with its antigen) antibody.

[0034] Antibody A and B (Epiclone™ Anti-A and Anti-B; CSL, Australia) solutions were used. Anti-A and Anti-B come as blue and yellow colour reagents, respectively. "AB+" and 'B+' blood were used in this study. The blood sample was supplied into plastic vials with anti-coagulant. Paper strips (70mmx2mm) were made from Whatman#4 filter paper on which 2 mm unit marks were printed. Paper strips were soaked into antibody solutions of different concentrations (Anti-A@1.0x, 0.8x, 0.6x, 0.4x, 0.2x and 0.0x); phosphate buffer saline (PBS) was used as diluent. Excess antibody was removed from the paper strips with blotting papers. The antibody (Anti-A) active paper strips were then placed on Reflex Paper. Blood drops of 20μL were dispensed at the centre of paper strip using a calibrated micro-pipette. The wicking distance was measured from centre to either direction. Pictures were taken after 10 minutes.

[0035] The results are shown in Figure 3. It can be seen that:

- Blood separates upon wicking with its specific antibody treated paper.

[0036] Blood separation is a non-linear function of the antibody concentration on the treated paper. The higher the antibody concentration, the more abrupt is the cell separation from the serum.

[0037] There is an optimum concentration to maximize wicking/separation/visualization.

[0038] Coagulation of red cell upon contact with its specific antibody drastically reduces its wicking/diffusion speed on the chromatographic surface, which promotes separation of cells from the serum. This drastic reduction and differentiation of elution speeds can serve as direct indicator of the type of blood.

Example 4: Effect of time on the wicking/separation of blood on bioactive antibody paper (see Figure 4)

[0039] In another embodiment of the invention, the paper is first treated with specific antibodies, dried or conditioned before been exposed to a sample of pure blood. This example illustrates the effect of contact time blood-antibody treated paper on the wicking and separation performance of blood on paper.

[0040] Antibody A and B (Epiclone™ Anti-A; CSL, Australia) solutions were used. Anti-A comes as a blue colour reagent. "AB+" blood was used in this study. The blood sample was supplied into plastic vials with anti-coagulant. Paper strips (70mmx2mm) were made from Whatman#4 filter paper on which 2 mm unit marks were printed. Paper strips were soaked into antibody solutions (Anti-A@); phosphate buffer saline (PBS) was used as diluent. Excess antibody was removed from the paper strips with blotting papers. The antibody (Anti-A) active paper

strips were then placed on Reflex Paper. Blood drops of 20μL were dispensed at the centre of paper strip using a calibrated micro-pipette. The wicking distance was measured from centre to either direction. Pictures were taken after different intervals of time.

[0041] It can be seen that:

blood wicking/separation levels off after about 4 minutes.

[0042] There is a minimum time of contact of antibody-blood required to allow proper blood coagulation/agglutination and wicking/separation.

[0043] There is an optimum time of contact of blood-antibody-paper. Too short, the blood does not properly coagulate; too long, the separation of red cell and serum can loose some of its sharpness.

Example 5: Paper microfluidic system to control flow, reaction and dilution (see Figure 5)

[0044] In the embodiment of the invention, paper-based microfluidic reactors can be used to conduct blood type tests. Specific antibodies are printed into the reactor designed on paper. Then blood cell suspension is introduced into the same reactor. The required period of time is allowed so that the antibodies and cell suspension can contact and mix. After a preset period of time, the valve of the reactor is closed to facilitate penetration of blood across the valve. If only the penetration of serum is observed, the test is positive because of agglutination of blood during the mixing time. If the penetration of blood is observed, the test is negative. Thus paper-based microfluidic reactor can provide a rapid visual test of blood type.

Example 6: Microfluidic system with valves (see Figure 6)

[0045] Paper microfluidic devices can be designed to increase the ratio of blood/antibody and to provide the required time delay to allow blood and antibody interactions before the test. This example shows that all these steps can be performed using a paper device. Figure 6 shows the design of the paper device. (A) A filter paper sheet is printed and cut as shown, and specific antibodies are either printed or deposited in the circled region. A paper switch is made on the right hand side of the device. (B) Blood sample is introduced onto the indicated region. (C) The cut paper is folded towards the blood sample as shown. (D) Blood sample is allowed to stay in contact with the antibody loaded paper for a set time. (E) After a short period of contact time, the switch is closed as shown. If the test is positive, blood will agglomerate and only serum will wick out along the switch. (F) After a short period of contact, the switch is closed as shown. If the test is negative, blood will not agglomerate and will wick out along the switch.

Example 7: Paper microfluidic system for blood typing (See Figure 7)

[0046] In another embodiment of the invention, a microfluidic system is printed on paper or a chromatographic medium and antibodies A, B and D (Rhesus) are printed into each of the 3 detection arms. Blood typing is analysed by placing a blood droplet in the middle reservoir and reading the results. All the different combinations of blood type and their representations are represented in Figure 7.

Example 8: Chromatographic Separation of RBC/Blood Serum on Paper (See Figure 8)

[0047] Figure 8 illustrates blood group detection using chromatographic separation of red blood cells (RBC) and blood serum on antibody active paper surface; (a) schematic of chromatographic separation on paper bioassay; (b) and (c)(I) are trial 1 and 2 using A+ blood sample, respectively; (b)(II), (c)(II) are the converted images of (b)(I), (c)(I) (RGB colour to BRG colour), respectively, for better resolution.

[0048] Modifications and variations as would be deemed obvious to the person skilled in the art are included within the ambit of the present invention as claimed in the appended claims.

Claims

1. A method for identifying an antigen that causes agglutination or coagulation of the red cells within a blood sample using a testing device having a cellulosic material excluding nitrocellulose substrate having a hydrophilic surface thereon, the surface including a collection zone and at least one detection zone extending therefrom, **characterized in that** an antigen or antibody being non covalently pre coated on the extent of the detection zone immobilized along the extent of the detection zone; the method including the steps of:

   depositing the blood sample directly on the collection zone of the testing device;
   the blood sample being transferred by capillary action to the detection zone;
   allowing the blood sample to react for a predefined optimal time; and
   identifying the antigen by a resultant visual indication arising where the antigen in the blood sample reacts with an appropriate antibody within the detection zone, wherein the resultant visual indication results from reduced wicking and/or a differentiation of eluation speeds.

2. A method according to claim 1, including mixing nanoparticles within the blood sample to facilitate said

reaction in the detection zone.

3. A method according to any one of claims 1 or 2, wherein the visual indication is due to an agglutination of the blood upon reaction with a specific said antibody resulting in reduced wicking and/or separation of the blood in the detection zone.

4. A method according to claim 1, including detecting blood type from the visual indication.

5. A method according to claim 1, including detecting illness from the visual indication.

Patentansprüche

1. Verfahren zum Identifizieren eines Antigens, das Agglutination oder Koagulation der roten Blutkörperchen in einer Blutprobe herbeiführt, unter Verwendung einer Testvorrichtung mit einem Cellulosematerial unter Ausschluss von Nitrocellulosesubstrat mit einer hydrophilen Oberfläche darauf, wobei die Oberfläche eine Sammelzone und mindestens eine sich daraus ausdehnende Nachweiszone einschießt, **dadurch gekennzeichnet, dass** ein Antigen oder Antikörper nicht-kovalent als Vorbeschichtung auf die Ausdehnung der Nachweiszone aufgebracht und entlang der Ausdehnung der Nachweiszone immobilisiert ist; wobei das Verfahren die folgenden Schritte einschließt:

   direktes Absetzen der Blutprobe auf der Sammelzone der Testvorrichtung;
   wobei die Blutprobe durch Kapillarwirkung auf die Nachweiszone übertragen wird;
   Reagierenlassen der Blutprobe für eine vordefinierte optimale Zeit und
   Identifizieren des Antigens durch eine resultierende visuelle Anzeige, die zum Vorschein kommt, wenn das Antigen in der Blutprobe mit einem geeigneten Antikörper innerhalb der Nachweiszone reagiert, wobei die resultierende visuelle Anzeige aus reduzierter Dochtwirkung und/oder einer Differenzierung der Eluierungsgeschwindigkeiten resultiert.

2. verfahren nach Anspruch 1, das Mischen von Nanopartikeln in die Blutprobe einschließt, um die Reaktion in der Nachweiszone zu erleichtern.

3. Verfahren nach einem der Ansprüche 1 oder 2, wobei die visuelle Anzeige auf eine Agglutinierung des Blutes bei Reaktion mit einem bestimmten Antikörper zurückzuführen ist, was zu reduzierter Dochtwirkung und/oder Trennung des Blutes in der Nachweiszone führt.

4. Verfahren nach Anspruch 1, welches das Nachweisen des Bluttyps aus der visuellen Anzeige einschließt.

5. Verfahren nach Anspruch 1, welches das Nachweisen von Krankheit aus der visuellen Anzeige einschließt.

Revendications

1. Procédé d'identification d'un antigène qui cause une agglutination ou une coagulation des globules rouges dans un échantillon de sang au moyen d'un dispositif d'essai ayant un matériau cellulosique à l'exclusion d'un substrat de nitrocellulose ayant une surface hygrophile sur celui-ci, la surface comprenant une zone de collecte et au moins une zone de détection s'étendant depuis celle-ci, **caractérisé en ce qu'**un antigène ou anticorps étant pré-enduit de façon non covalente sur l'étendue de la zone de détection immobilisée le long de l'étendue de la zone de détection ; le procédé comprenant les étapes de :

   dépôt de l'échantillon de sang directement sur la zone de collecte du dispositif d'essai ;
   transfert de l'échantillon de sang par action capillaire vers la zone de détection ;
   réaction de l'échantillon de sang pendant un temps optimal prédéfini ; et
   identification de l'antigène par une indication visuelle résultante survenant à l'emplacement où l'antigène dans l'échantillon de sang réagit avec un anticorps approprié dans la zone de détection, l'indication visuelle résultante étant due à une capillarité et/ou une différenciation réduites des vitesses d'élution.

2. Procédé selon la revendication 1, comprenant le mélange de nanoparticules dans l'échantillon de sang afin de permettre ladite réaction dans la zone de détection.

3. Procédé selon l'une quelconque des revendications 1 ou 2, dans lequel l'indication visuelle est due à une agglutination du sang après réaction avec ledit anticorps spécifique conduisant à une capillarité et/ou séparation réduite du sang dans la zone de détection.

4. Procédé selon la revendication 1, comprenant la détection de type sanguin à partir de l'indication visuelle.

5. Procédé selon la revendication 1, comprenant la détection d'une maladie à partir de l'indication visuelle.

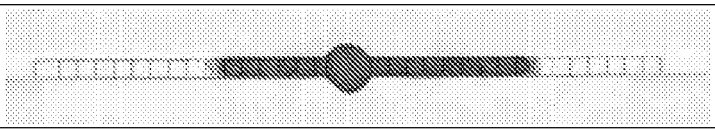
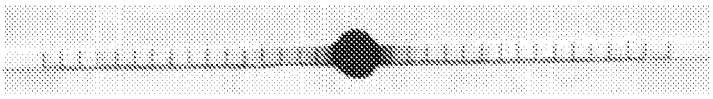
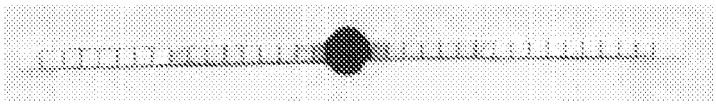
Blood sample mixed with antibody solutions	Blood Sample –B+
Anti-A	
Anti-B	
Anti-D	

Figure 1

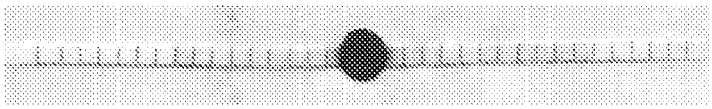
Blood sample mixed with antibody solutions	Blood Sample O+
Anti-A	
Anti-B	
Anti-D	

Figure 2

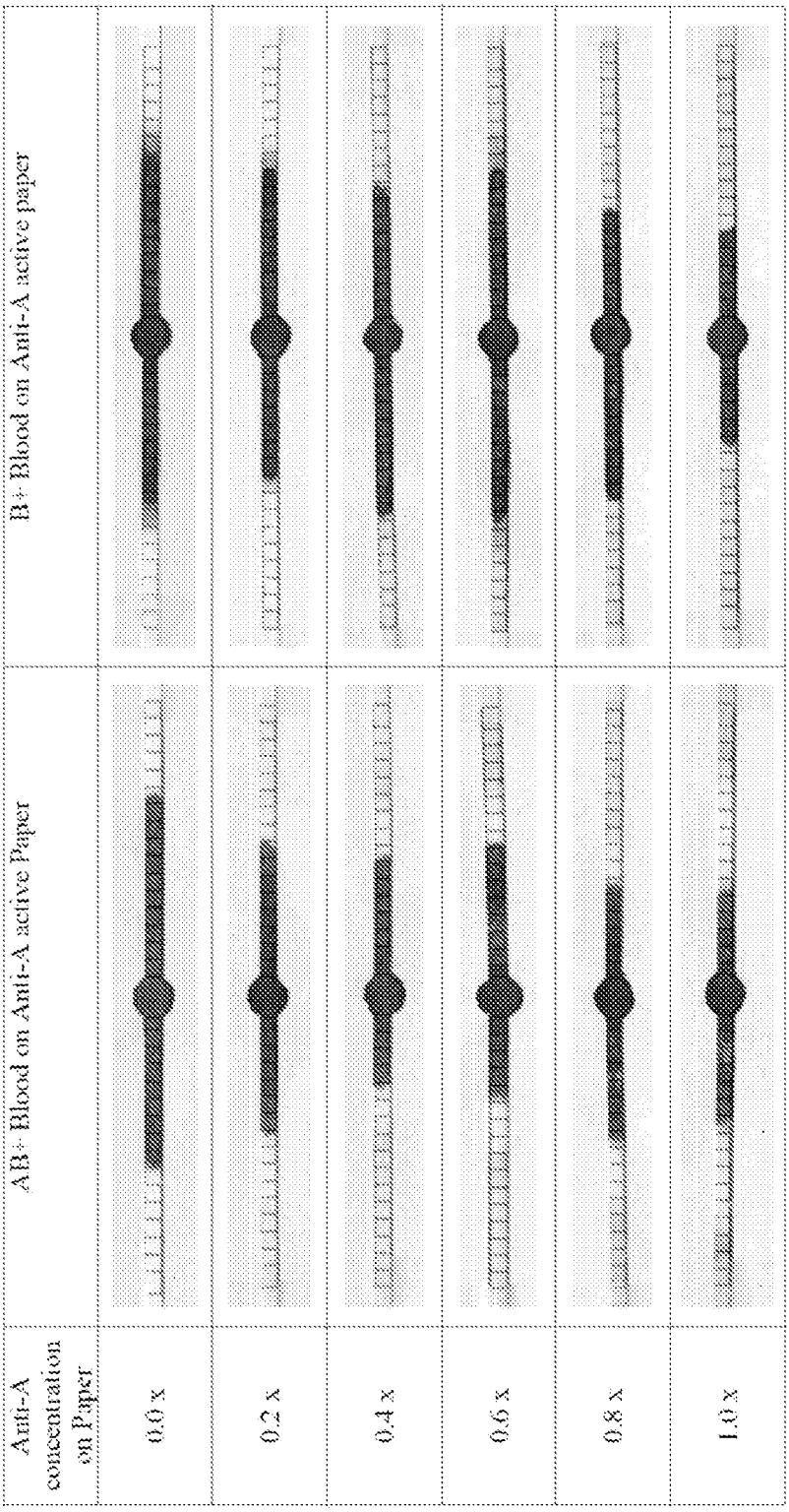

Anti-A concentration on Paper
AB+ Blood on Anti-A active Paper
B+ Blood on Anti-A active paper
0.0 x
0.2 x
0.4 x
0.6 x
0.8 x
1.0 x

Figure 3

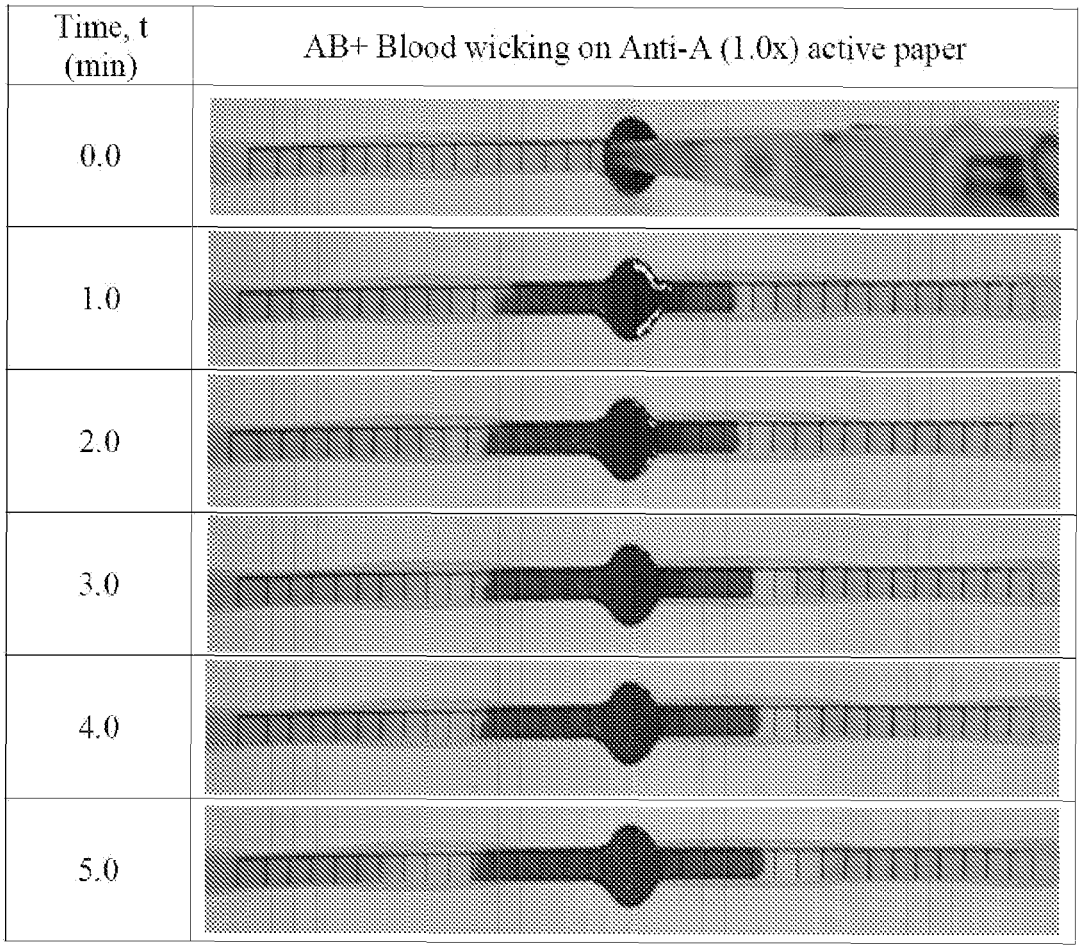


Figure 4

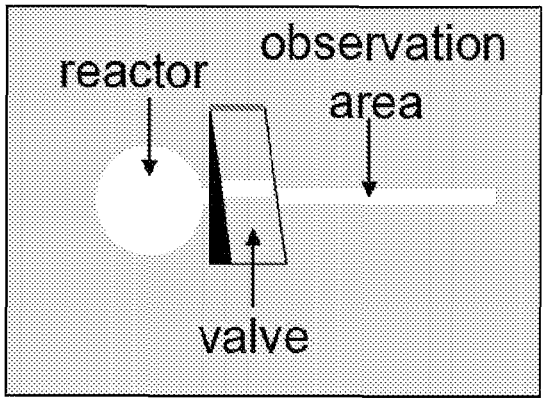


Figure 5

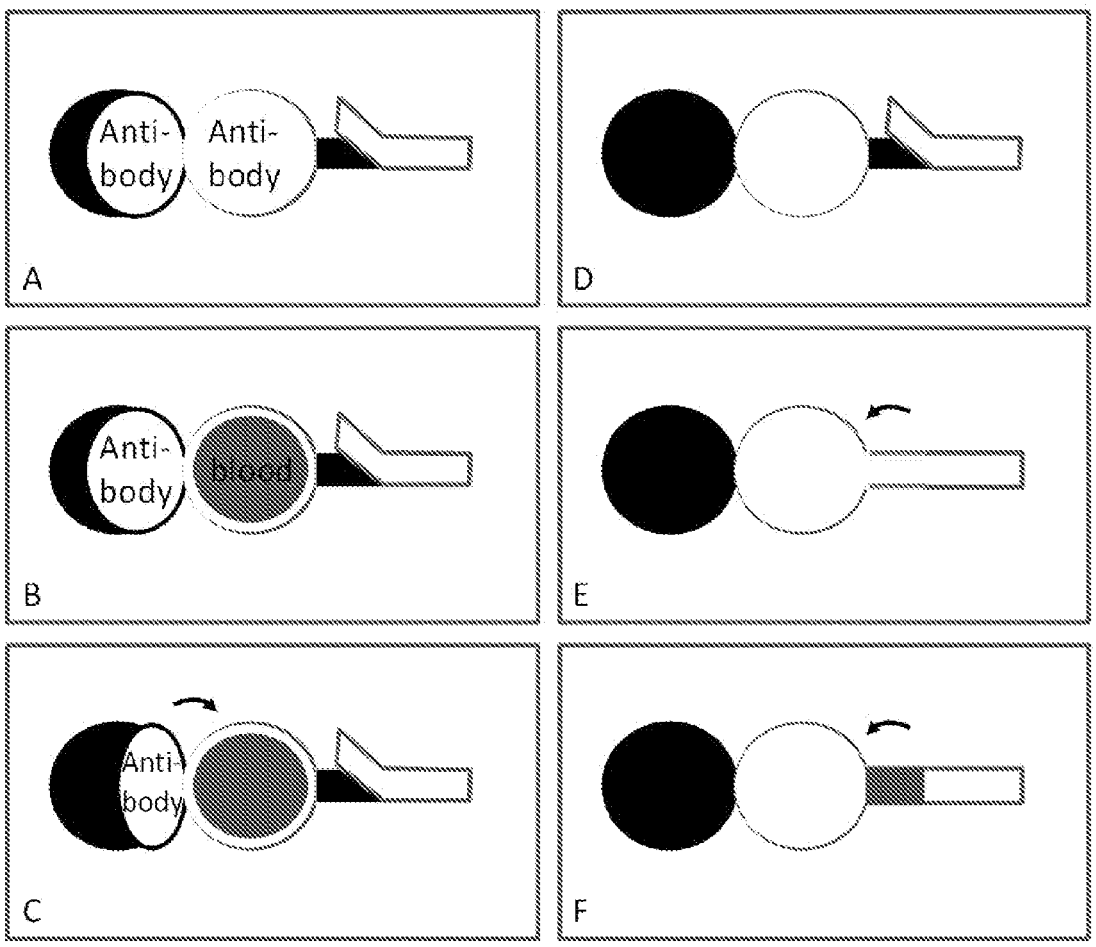
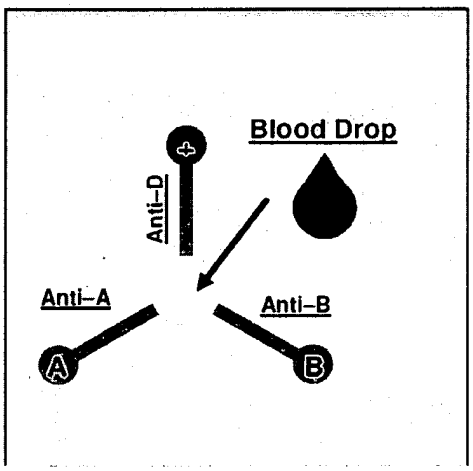
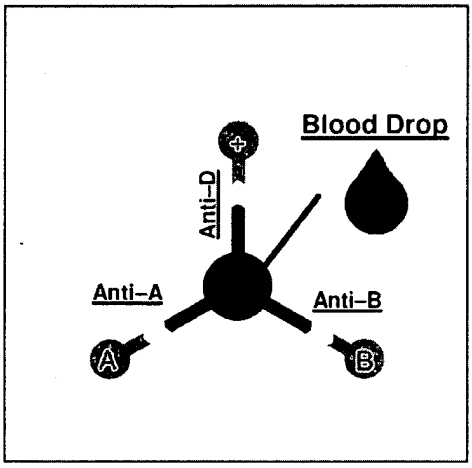


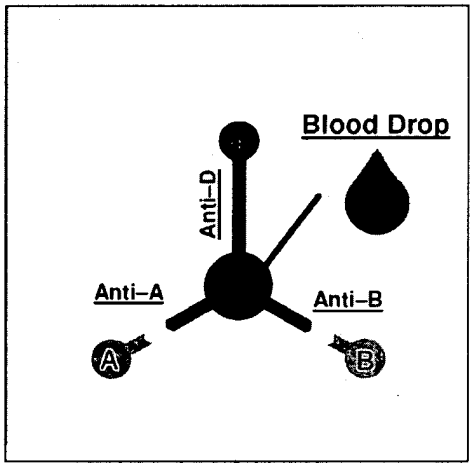
Figure 6



(a) Paper based (ABO) blood type detection device



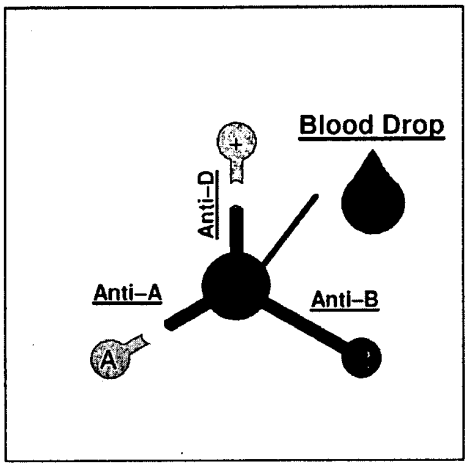
(b) Detection of AB+ blood



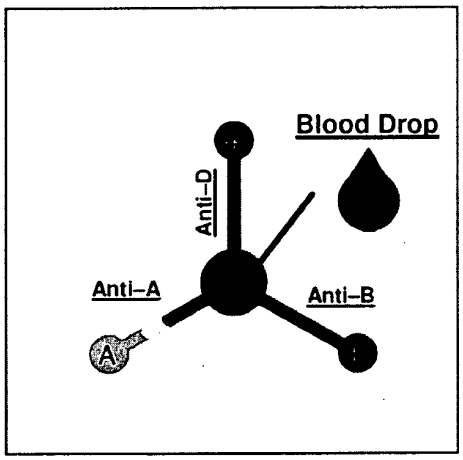
(c) Detection of AB- blood

Figure 7

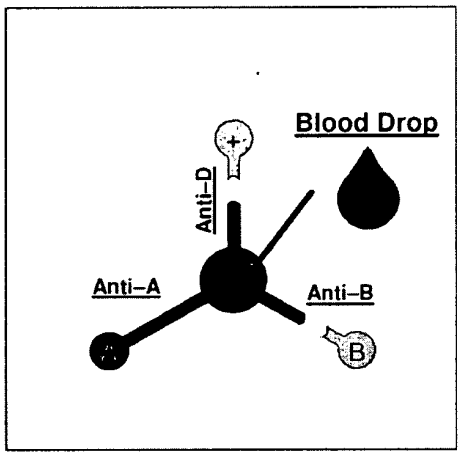
Figure 7 (cont.)



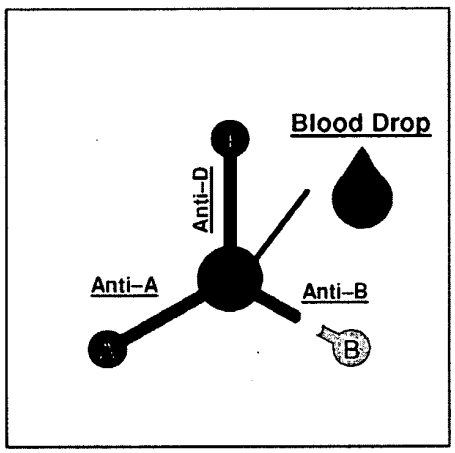
(d) Detection of A+ blood



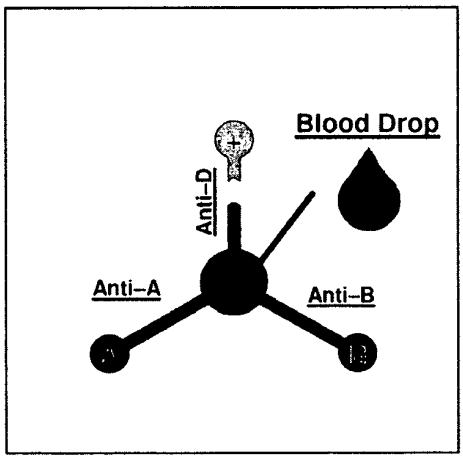
(e) Detection of A- blood



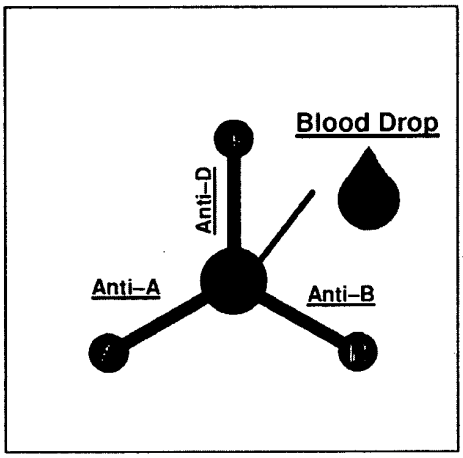
(f) Detection of B+ blood



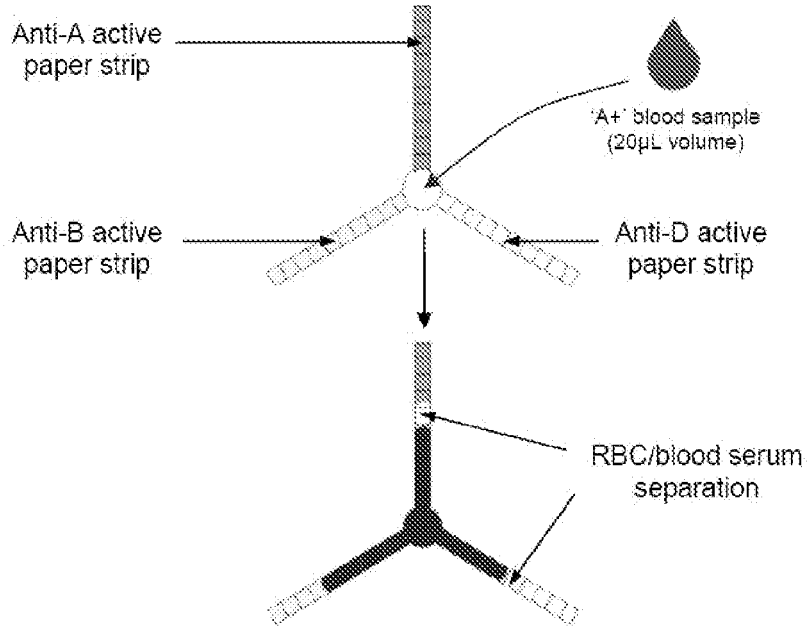
(g) Detection of B- blood



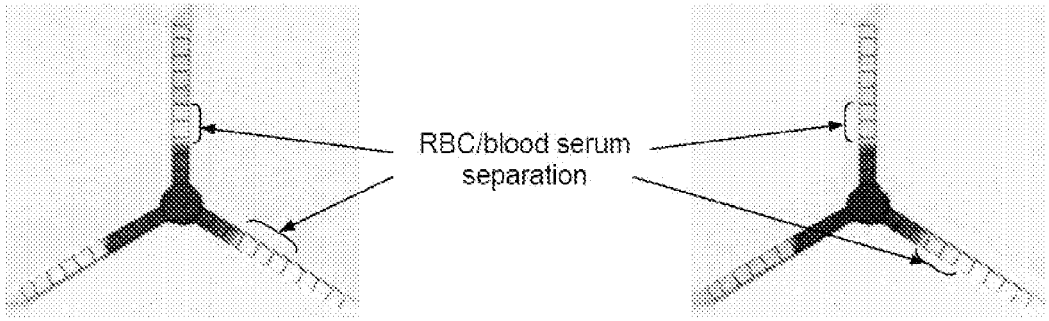
(h) Detection of O+ blood



(i) Detection of O- blood

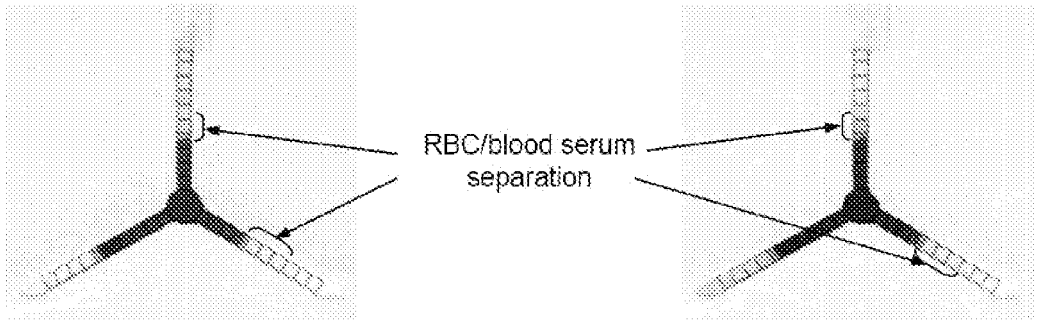


(a)



(I) (II)

(b)



(I) (II)

(c)

Figure 8

REFERENCES CITED IN THE DESCRIPTION

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Patent documents cited in the description

- WO 8707304 A1 **[0003]**
- AU 2009000889 W **[0011] [0022]**

Non-patent literature cited in the description

- **N.G. ANDERSON.** Analytical Techniques for Cell Fractions. *Analytical Biochemistry,* 1970, vol. 38, 175-189 **[0004]**

专利名称(译)	用于鉴定生物流体中的抗原和抗体的测试装置		
公开(公告)号	EP2480885A1	公开(公告)日	2012-08-01
申请号	EP2010818158	申请日	2010-09-24
申请(专利权)人(译)	莫纳什大学		
当前申请(专利权)人(译)	莫纳什大学		
[标]发明人	GARNIER GIL SHEN WEI KHAN MOHIDUS SAMAD LI XU THOUAS GEORGE		
发明人	GARNIER, GIL SHEN, WEI KHAN, MOHIDUS SAMAD LI, XU THOUAS, GEORGE		
IPC分类号	G01N33/53 D21H25/00 G01N33/80 A61B5/00 G01N30/91		
CPC分类号	G01N30/90 G01N33/558 Y02A50/58 G01N21/82 G01N33/80 G01N2021/752 G01N2021/757 G01N2021/825		
优先权	2009904643 2009-09-24 AU		
其他公开文献	EP2480885A4 EP2480885B1		
外部链接	Espacenet		

摘要(译)

一种用于鉴定生物流体样品中的抗原或抗体的测试装置，包括：其上具有亲水表面的基底;所述表面包括收集区，以及至少一个从其延伸的检测区;其中生物流体样品可以与特定抗原或抗体混合，并沉积在收集区上并通过毛细管作用转移到检测区;生物流体样品中的抗原或抗体与适当的所述抗体或抗原反应，从而在检测区内产生视觉指示。