Revised September 2009
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Anti-Human Globulin
Anti-IgG (Rabbit) (Green)
Ortho BioVue® System
(IgG Cassette)

REF

400 cassettes 707400
100 cassettes 707450

INTENDED USE

Qualitative procedure for the detection of IgG bound to red blood cells

FOR IN VITRO DIAGNOSTIC USE

SUMMARY AND EXPLANATION

Anti-IgG is an important diagnostic aid in determining the presence or absence of IgG on human red blood cells. This reagent will detect cells sensitized with IgG and will not react with cells sensitized with complement. Anti-IgG is a suitable reagent for direct and indirect antiglobulin testing.

PRINCIPLE OF PROCEDURE

The Ortho BioVue System utilizes column agglutination technology comprised of glass beads and reagent contained in a column which, upon centrifugation of the cassette, trap agglutinated red blood cells and allow nonagglutinated red blood cells to travel to the bottom of the column. Red cells are separated from serum proteins prior to exposure to the Anti-IgG reagent. The density of the reagent allows the red blood cells to pass through the column, while the less dense neutralizing serum proteins remain above the glass bead/reagent interface.

REAGENTS

Ortho BioVue System IgG cassettes are comprised of 6 columns containing a buffered solution with bovine albumin and macromolecular potentiators, as well as the preservatives 0.1% (w/v) sodium azide and 0.01M ethylenediaminetetraacetic acid (EDTA).

Product Codes 707400 and 707450	**Component Description**
Columns 1-6: Anti-Human Globulin Anti-IgG (Rabbit) (Green)	Anti-IgG (rabbit) FD&C Blue No. 1 FD&C Yellow No. 5

STORAGE REQUIREMENT

Store cassettes upright at 2 to 25 °C.

Do not store the cassettes in a self-defrosting refrigerator/freezer.

Do not store cassettes near any heat source (e.g., heat block, radiator, large instrumentation, refrigerator, freezer, etc., or any area receiving direct sunlight).

PRECAUTIONS

1. Handle all blood and materials in contact with blood as if capable of transmitting infectious agents. It is recommended that blood and materials in contact with blood be handled using established good laboratory practices.[1]
2. All blood products should be treated as potentially infectious. Source material from which this product was derived was found negative for hepatitis B surface antigen (HBsAg) and antibodies to hepatitis C virus (HCV) and human immunodeficiency virus type 1 (HIV-1) and type 2 (HIV-2) when tested in accordance with current FDA required tests. No known test methods can offer assurance that products derived from human blood will not transmit infectious agents.
3. Some cassette components may be considered as hazardous or potentially infectious waste. Dispose of all materials according to applicable guidelines and regulations.[2]
4. Do not use reagents beyond their labeled expiration date.
5. Freezing of the cassettes or evaporation of the liquid due to heat may interfere with free passage of unagglutinated red blood cells through the glass bead column.
6. Do not use cassettes that appear damaged (i.e., break in foil seal or break, crack or bubble in the column) or exhibit drying (i.e., liquid level is at or below the top of the glass beads) or exhibit discoloration (due to bacterial contamination which can cause false reactions).
7. Use the Ortho BioVue System Centrifuge to provide the required centrifugation parameters for this system. Proper calibration of the centrifuge is essential to achieve accurate test results.
8. Erroneous results may be obtained due to improper technique in performing any diagnostic test. The most common sources of such results are:
   - Use of red blood cell concentrations other than those described under Specimen Collection and Preparation section
   - Microbial contamination of supplementary materials used in the procedure
   - Use of specimens containing particulate matter (impedes the free flow of red blood cells through the column)
   - Use of severely hemolyzed samples (may interfere with reading reactions in the column)

9. In order to minimize the presence of bubbles with your Ortho BioVue cassettes, we recommend that if you normally store your cassettes in the refrigerator at 2 to 8°C you should equilibrate your cassettes at room temperature (20 to 25°C) for at least 96 hours prior to use.
10. If testing is to be performed by methods other than those identified in the package insert, standardization of procedures must be undertaken to assure accuracy of test results.

SPECIMEN COLLECTION AND PREPARATION

No special preparation of the patient/donor is required prior to specimen collection. Blood should be collected by approved medical techniques. Samples should be tested as soon as possible following collection. If a delay in testing occurs, samples should be stored at 2 to 8 °C.

NOTE: Red blood cells obtained from umbilical cord samples should be washed one time in isotonic saline.

DIRECT ANTIGLOBULIN TEST (DAT)
Blood drawn into EDTA is preferred for the direct antiglobulin test. Blood drawn into heparin or with no anticoagulant (clotted specimens) should be tested within three days. Blood drawn into EDTA or sodium citrate should be tested within seven days. Donor blood may be tested up to the date of expiration.

For the direct antiglobulin test, red blood cell suspensions can be prepared in the acceptable range of 3% to 5% using the following combinations of saline and packed red blood cells:

Saline Volume	Packed Red Blood Cell Volume[a]	Red Blood Cell Concentration
1 mL	40 µL	3%
1 mL	50 µL	4%
1 mL	65 µL	5%

[a] Blood samples centrifuged at 900 to 1000 x g for 5 minutes will result in a packed red blood cell concentration of approximately 80%. These specifications for centrifugation eliminate over-packing of red cells which may result in false positive results. Data on file at Ortho-Clinical Diagnostics, Inc.

INDIRECT ANTIGLOBULIN TEST
Either serum or plasma may be used when performing the indirect antiglobulin test in the Ortho BioVue System. This test is designed to be performed using a low ionic strength environment. Ortho 0.8% Reagent Red Blood Cells are supplied in a low ionic strength diluent suitable for this method. Red cell suspensions for crossmatch, autocontrol or antigen testing should be prepared using Ortho® 0.8% Red Cell Diluent according to the instructions for use.

For LISS-additive methods, refer to ORTHO BLISS instructions for use.

REAGENT PREPARATION

The Ortho BioVue System cassette is provided ready to use. Each column contains a single specificity of reagent suitable for one test. The cassette is heat-sealed with aluminum foil to preserve the integrity of the reagents. Upon opening of the foil seal, the cassettes should be used within one hour. Do not use the cassette if the liquid level in the column is at or below the top of the glass beads.

Reverse Diluent may be used as a control since it contains all components used in the IgG cassette except the rabbit anti-human IgG.

PROCEDURE

The procedure identified below is for manual BioVue cassette testing only. When using automated instruments, follow the procedures that are contained in the operator's manual provided by the device manufacturer. Laboratories must follow their approved validation procedures to demonstrate compatibility of this product on automated systems.

For LISS-additive methods, refer to ORTHO BLISS instructions for use.

Materials Provided

400 cassettes (Product Code 707400)
100 cassettes (Product Code 707450)
(See Reagents section for component description)

Materials Required But Not Provided

Direct Antiglobulin Test

1. Ortho BioVue System Centrifuge
2. Isotonic saline
3. Micropipetter for delivery of 10 µL
4. Disposable pipette tips
5. Ortho BioVue System Work Rack (Product Code 707830)

Indirect Antiglobulin Test

1. Ortho BioVue System Centrifuge
2. Ortho BioVue System Heat Block, 37 °C (Product Code 707820)
3. Micropipetters for delivery of 40 µL and 50 µL
4. Disposable pipette tips
5. Ortho BioVue System Work Rack (Product Code 707830)
6. Ortho 0.8% Reagent Red Blood Cells
7. Ortho 0.8% Red Cell Diluent

Test Procedure for Direct Antiglobulin Test

1. Prepare red cell suspensions according to Specimen Collection and Preparation section.
2. Allow the cassette and test sample to come to room temperature before use. Orient the cassette with the back label (bar code side) facing you. Label the cassette appropriately with sample identification.
3. Peel off the foil strip on the top of the cassette only exposing the reaction chambers needed for the test(s) being performed. Visually inspect the cassette(s) to ensure that residual film does not block the opening of any wells after removing the foil.
   NOTE: The cassette should be used within one hour after removal of the foil. Cassettes with covered reaction chambers can be saved and these columns used for additional tests. Do not use the cassette if the liquid level is at or below the top of the glass beads.
4. Add 10 µL of the 3% to 5% saline red blood cell suspension to the appropriate reaction chamber(s) of the cassette.
5. Centrifuge the cassette using the Ortho BioVue System Centrifuge.
   NOTE: Centrifugation should occur within 30 minutes of addition of the samples to the reaction chamber.
6. Read the front and back of the individual columns for agglutination upon test completion.
7. Record the reaction strength from the side with the stronger positive result.

Test Procedure for Indirect Antiglobulin Test

1. Prepare red cell suspensions according to Specimen Collection and Preparation section.
2. Allow the cassette and test sample to come to room temperature before use. Orient the cassette with the back label (bar code side) facing you. Label the cassette appropriately with sample identification.
3. Peel off the foil strip on the top of the cassette only exposing the reaction chambers needed for the test(s) being performed. Visually inspect the cassette(s) to ensure that residual film does not block the opening of any wells after removing the foil.
   NOTE: The cassette should be used within one hour after removal of the foil. Cassettes with covered reaction chambers can be saved and these columns used for additional tests. Do not use the cassette if the liquid level is at or below the top of the glass beads.
4. Add 50 µL of 0.8% red cell suspension to appropriate reaction chamber(s) of the cassette.
5. Add 40 µL of the patient's serum or plasma to the appropriate reaction chamber(s) of the cassette. **Do not touch the pipette tip to the side of the reaction chamber. If this occurs, change pipette tip before proceeding to the next chamber.**
6. Observe that the contents of the reaction chamber(s) are combined. If necessary, tap gently.
   NOTE: Assure that the reagents remain in the reaction chamber during incubation. There should be no mixing of reactants with reagents in the column prior to centrifugation. (See LIMITATIONS OF PROCEDURE.)
7. Incubate at 37 °C for a minimum of 10 minutes to a maximum of 30 minutes.
8. Centrifuge the cassette using the Ortho BioVue System Centrifuge.
   NOTE: Centrifugation should occur within 30 minutes of addition of the samples to the reaction chamber.
9. Read the front and back of the individual columns for agglutination and/or hemolysis upon test completion.
10. Record the reaction strength from the side with the stronger positive result.

Quality Control Procedures

Serological testing is necessary to recognize reagent deterioration. It is recommended each lot of reagents be tested on each day of use with appropriate positive and negative controls according to approved standard operating procedures.

Positive Control – Use human serum or plasma containing an IgG antibody and antigen-positive red blood cells to test for the anti-IgG activity of the reagent. Follow the indirect antiglobulin test procedure.

Negative Control – Use human serum or plasma free of unexpected red cell antibodies and unsensitized red blood cells. Follow the indirect antiglobulin test procedure.

Antiglobulin control cells are not required with the Ortho BioVue System and should not be used to control each individual test.

INTERPRETATION OF RESULTS

Positive Result (+): Agglutination of the red blood cells is a positive test result and indicates the presence of the corresponding antigen. The presence of hemolysis with or without agglutination is considered a positive test result.

Negative Result (-): No agglutination or no hemolysis of the red blood cells is a negative test result and indicates the corresponding antigen is not demonstrable.

Hemolysis will result in a slight pink to red appearance in the reagent above the bead column. In cases of partial hemolysis, agglutination may or may not be present.

4+ Reaction	Agglutinated red blood cells form a band at the top of the bead column.
3+ Reaction	Most agglutinated red blood cells are retained or trapped in the upper half of the bead column.
2+ Reaction	Agglutinated red blood cells are observed throughout the length of the bead column. A small button of cells may also be visible at the bottom of the bead column.
1+ Reaction	Most agglutinated red blood cells are retained or trapped in the lower half of the bead column. A button of cells will also be visible at the bottom of the bead column.
0.5+ Reaction	Most agglutinated red blood cells pass through and form a disrupted (not smooth) button at the bottom of the bead column. Small agglutinates are visible above the button.
0 Negative	All red blood cells pass through and form a smooth button at the bottom of the bead column.

LIMITATIONS OF THE PROCEDURE

1. The Test Procedure and Interpretation of Results must be followed closely to assure the accuracy of the test results. A laboratory that institutes the Ortho BioVue System should have a program that will train personnel on the proper use and handling of the product.
2. Some literature reports indicate that Anti-IgG may occasionally fail to detect antibodies that are demonstrable only by the use of an Anti-Human Globulin reagent containing anti-complement. Antibodies not detected by Anti-IgG may be clinically significant in some cases.
3. Low ionic strength solutions (LISS) have been shown to enhance many antigen-antibody reactions. However, samples may be encountered that contain antibody specificities, such as Anti-K (K1), which are not optimally reactive in LISS test systems.
4. Enzyme-treated red blood cells will nonspecifically agglutinate in the Anti-IgG reagent in the IgG cassette.
5. Plasma expanders have been shown to interfere with some blood bank tests. Data are not available concerning interference using the Ortho BioVue System. Problem-solving techniques should be used if interference is observed.
6. Anomalous results may be caused by the following:
   - fibrin or particulate matter, such as clots, in serum or plasma
   - incompletely washed specimen red blood cells
   - mixing of reactants with reagents in the column (indirect antiglobulin test)
   - red blood cells that stick to the sides of the reaction chamber
   - bubble(s) that impede the passage of unagglutinated red blood cells

   An example of an anomalous result may be a line of red blood cells on top of the bead column. These may be prevented by removing fibrin or particulate matter from the sample by centrifugation before testing, washing red blood cells at least one time to remove residual plasma and/or platelets, and by reading results upon test completion. Tests with these or other anomalous results should be repeated.

EXPECTED RESULTS*

Antibody Detection (Indirect Antiglobulin Test)

Specificity analysis for the IgG cassette using 0.8% SURGISCREEN® Reagent Red Blood Cells was performed on the antibody screening results of 225 random population samples. The estimated specificity of 0.8% SURGISCREEN relative to 3% SURGISCREEN is 100% with a 95% confidence interval of 98.4% to 100.0%.

Sensitivity analysis for the IgG cassette using 0.8% SURGISCREEN was performed on the antibody screening results of 316 antibody-positive population samples. The distribution contained representative samples from the Rh-hr (32.59%), Kell (22.15%), Kidd (7.59%), Duffy (9.18%), MNS (7.59%), Lewis (6.33%) and P (1.27%) systems. Samples containing multiple antibodies (13.29%) were also included.

The estimated sensitivity of 0.8% SURGISCREEN relative to 3% SURGISCREEN is 100% with a 95% confidence interval from 98.8% to 100.0%.

Direct Antiglobulin Test (DAT)

In clinical studies using 3% to 5% red cell suspensions in saline with the IgG cassette, the results obtained for DAT by the BioVue method gave 99.6% (248/249) agreement when compared to the licensed tube test. There was 100% (237/237) agreement between tube and BioVue methods for direct antiglobulin testing of cord blood samples. Percent agreement indicates concordance between the two assays only and does not indicate which method gave the correct result.

*Data on file at Ortho-Clinical Diagnostics, Inc.

SPECIFIC PERFORMANCE CHARACTERISTICS[3-6]

The immunogen used to produce rabbit anti-human IgG is a gamma globulin fraction of human plasma. Testing for antibodies to immunoglobulin mu (μ) chains and light (κ and λ) chains is not performed but such antibodies may be present in this reagent.

Each lot of IgG cassettes is tested and shown to agglutinate red cells weakly sensitized with IgG. Unsensitized red cells and red cells sensitized in vitro with C3b and C4 are negative. This reagent may agglutinate IgM sensitized red cells.

SUMMARY OF REVISIONS

Section	Revision
REAGENTS	(French version only) Changed “l’acide de sodium” to “l’azide de sodium.”

BIBLIOGRAPHY / BIBLIOGRAPHIE / LITERATUR / BIBLIOGRAFIA / BIBLIOGRAFI / BIBLIOGRAFIE / LITTERATUR / ΒΙΒΛΙΟΓΡΑΦΙΑ / LITTERATURFÖRTECKNING / 参考文献

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KEY TO SYMBOLS / LÉGENDE DES SYMBOLES / SYMBOL-LEGENDE / CLAVE DE LOS SÍMBOLOS / LEGENDA DEI SIMBOLI / SIMBOLOGIA / FORKLARING TIL SYMBOLER / ΕΠΕΞΗΓΗΣΗ ΣΥΜΒΟΛΩΝ / FÖRKLARING TILL SYMBOLERNA /符号标志

The following symbols may have been used in the labeling of this product. / Les symboles suivants ont pu être utilisés pour étiqueter ce produit. / Es ist möglich, dass die folgenden Symbole bei der Etikettierung dieses Produktes verwendet wurden. / Los siguientes símbolos pueden haber sido empleados en el etiquetado de este producto. / Nelle etichette di questo prodotto possono essere stati utilizzati i simboli seguenti. / Os seguintes símbolos podem ter sido utilizados no rótulo deste produto. / Følgende symboler ka være anvendt ved mærkningen af dette produkt. / Τα ακόλουθα σύμβολα ενδέχεται να έχουν χρησιμοποιηθεί στη σήμανση αυτού του. / Följande symboler kan ha använts vid märkningen av denna produkt. / 以下符号标签可能贴于本产品的相应位置。

Attention: See instructions for use /
Attention : Se référer aux instructions d'utilisation/
Wichtig: Siehe Gebrauchsanweisung /
Atención: Ver las instrucciones de uso /
Attenzione: Vedi le istruzioni per l'uso /
Atenção: Consulte as instruções de utilização /
Obs.: Se brugsanvisning /
Προσοχή: Βλέπετε οδηγίες χρήσης /
Obs! Se bruksanvisningen /
注意: 参阅《使用说明》

LOT

Lot Number / Numéro de lot / Loscode /
Número de lote / Lotto numero / Número de lote /
Lot-nummer / Αριθμός παρτίδας / Lot-nummer /
批号

Use by/expiration date (CCYY-MM-DD) /
Utiliser avant/date d'expiration (AAAA-MM-JJ) /
Benutzen vor/Verfalldatum (JJJJ-MM-TT) /
Úsese antes de/Fecha de caducidad (SSAA-MM-DD) /
Data di scadenza (AAAA-MM-GG) /
Utilizar até/data de validade (SSAA-MM-DD) /
Anvendes senest/udløbsdato (ÅÅÅÅ-MM-DD) /
/ημερομηνία λήξης (AAEE-MM-HH) /
Används före/Utgångsdatum (ÅÅÅÅ-MM-DD) /
失效日期（年-月-日）

Store between / Conserver entre / Lagern zwischen /
Almacenar entre / Conservare ad una temperatura tra /
Armazenar entre / Opbevares mellem / Φύλαξη μεταξύ /
Förvaras mellan / 贮存温度范围

IVD

For In Vitro Diagnostic Use / Pour Diagnostic in Vitro /
In-vitro-Diagnostikum / Para uso diagnóstico in vitro /
Per uso diagnostico in vitro / Para diagnósticos In Vitro /
Til diagnostisk brug in vitro / Για διαγνωστική χρήση
In Vitro / För diagnostisering in vitro / 用于体外诊断

EC REP

Authorized Representative / Mandataire / Bevollmächtigeter /
Representante autorizado / Rappresentante autorizzato /
Representante Autorizado / Autoriseret repræsentant /
Εξουσιοδοτημένος Αντιπρόσωπος */ Auktoriserad representant /*
授权代表

REF

Product Code / Code produit / Artikelcode / Código del producto /
Codice prodotto / Código do Produto / Produktkode /
Κωδικός Προϊόντος / Produktkod /产品编号

This end up / Haut / Diese Seite nach oben / Este extremo hacia arriba / Questa estremità in alto / Este Lado Para Cima /
Denne side op / Η Συσκευασία Πρέπει να Είναι Όρθια από Αυτήν την Πλευρά / Denna sida upp /此端向上

Keep Dry / Conserver dans un endroit sec / Trocken aufbewahren /
Mantener seco / Conservare in luogo asciutto /
Mantenha em Ambiente Seco / Undgå fugt /
Διατηρήστε το Στεγνό / Förvaras torrt /保持干燥

Fragile, handle with care / Fragile, à manipuler avec précaution /
Zerbrechlich, mit Vorsicht behandeln / Frágil, manipular con precaución / Fragile, maneggiare con cura / Frágil, tratar com cuidado / Fragil, håndteres med forsigtighed /
Εύθραυστο, χειριστείτε με προσοχή / Ömtålig, hanteras varsamt /
易碎，小心搬运

Cassette / Cassette / Kassette / Cassette / Cassetta / Cassete /
Kassette / Κασέτα / Kassett /试剂卡

EC REP

Ortho-Clinical Diagnostics
Johnson & Johnson
50-100 Holmers Farm Way
High Wycombe
Buckinghamshire
HP12 4DP
United Kingdom

Ortho Clinical Diagnostics
a Johnson & Johnson company
1001 US Highway 202, Raritan, NJ 08869 USA

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