US

Reagent Red Blood Cells

Biotestcell® -I 8

Biotestcell® -I 11

3.0 to 3.4%

FOR USE IN IDENTIFICATION OF UNEXPECTED ANTIBODIES WITH TANGO® OPTIMO AND BY TUBE TEST

FOR IN-VITRO DIAGNOSTIC USE
NO U.S. STANDARD OF POTENCY
U.S. License Number: 1798

Package size

REF 816020100 VOL 8 x 4 mL Biotestcell®-I 8
REF 816021100 VOL 11 x 4 mL Biotestcell®-I 11

Intended Use
Biotestcell®-I 8 and Biotestcell®-I 11 are used for the identification of unexpected antibodies which are found in an antibody screening test using Reagent Red Blood Cells such as Biotestcell® 1 & 2, Biotestcell® 3 and/or Biotestcell® Pool. Biotestcell®-I 8 and Biotestcell®-I 11 are used in tube test and solid phase test Solidscreen II with the TANGO® optimo and is suited for all routine testing.

Summary
Antibody identification is used to determine specificity and possible clinical significance of antibodies against red blood cell antigens detected in the method used for detection of unexpected antibodies. This determination is important for the selection of products for transfusion as well as for pre-natal care.
Biotest Reagent Red Blood Cells Biotestcell®-I 8 and Biotestcell®-I 11 are used to determine the specificity of an antibody detected by the previous antibody screening or crossmatch. Routine pretransfusion studies always include tests for the antibody identification of detected antibodies to red blood cells.

Principle of the Test
Antigens of the Reagent Red Blood Cells react with the corresponding antibodies in the serum or plasma directly or after addition of Anti-Human Globulin. In a tube test agglutination will occur. In solid phase test Solidscreen II a carpeting of red blood cells on the microtest plate wells will occur.

Reagent
Biotestcell®-I 8 and Biotestcell®-I 11 are Reagent Red Blood Cells with polyvalent antigens.
Biotestcell®-I8 and Biotestcell®-I 11 contain the following antigens: D, C, C^w, E, c, e, K, k, Fy^a, Fy^b, Lu^a, Lu^b, Jk^a, Jk^b, Js^b, M, N, S, s, Le^a, Le^b, P_1, Xg^a, Co^a.
Biotestcell®-I 11 contains the additional antigens: Js^a, Di^a, and Kp^a. For the antigen content of each production lot, please refer to the enclosed table.
Biotestcell®-I8 and Biotestcell®-I 11 are also suited for use with enzymes (papain, ficin, bromelin, trypsin) or enhancement reagents (albumin, LISS). The life span of enzyme treated Reagent Red Blood Cells is listed in the instructions for use of the respective enzymes.
Biotestcell®-I8 and Biotestcell®-I 11 are suspended 3.0 to 3.4% in a modified Alsevers solution and can be used immediately after careful resuspension.
Biotestcell®-I 8 and Biotestcell®-I 11 are produced every 4 weeks.

Preservative:
0.01% Neomycin, 0.033% Chloramphenicol, 5 ppm Amphotericin B

Precautions
- For In-vitro diagnostic use.
- Store at 2 to 8°C.
- Do not use beyond the expiration date.
- Do not use damaged vials.
- Do not use if markedly hemolyzed or discolored
- Handle and dispose of reagents as potentially infectious
- Caution: Do not pipette by mouth. The absence of all viruses has not been determined.
- Caution: ALL BLOOD PRODUCTS SHOULD BE TREATED AS POTENTIALLY INFECTIOUS. SOURCE MATERIAL FROM WHICH THIS PRODUCT WAS DEFIVED WAS FOUND NEGATIVE WHEN TESTED WITH FDA LICENSED EIA/ELISA TESTS. NAT TESTING WAS NOT PERFORMED. NO KNOWN TEST METHOD CAN OFFER ASSURANCE THAT PRODUCTS DERIED FROM HUMAN BLOOD WILL NOT TRANSMIT INFECTIOUS AGENTS.
- Caution: This product contains Natural Rubber Latex Which May Cause Allergic Reactions.
- Do not use beyond seven days when loaded on the TANGO® optimo.
- Do not use samples collected with gel separators of any kind.

Specimen Collection
Tube Test
Fresh samples of clotted or EDTA anticoagulated whole blood collected following general blood sampling guidelines are acceptable.
The specimen should be tested as soon as possible after collection. If testing is delayed, EDTA and clotted specimens should be stored at 2 to 8°C.
Blood specimens exhibiting gross hemolysis or contamination should not be used. Do not use specimens collected with gel separators.
Clotted samples or those collected in EDTA older than ten days can be tested, however antibody reactivity has been shown to decrease in older samples. Stored samples should be allowed to reach room temperature (18 to 26°C) prior to testing.

TANGO® optimo
Fresh samples of clotted or EDTA anticoagulated whole blood collected following general blood sampling guidelines are acceptable.
The specimen should be tested as soon as possible after collection. If testing is delayed, EDTA and clotted specimens should be stored at 2 to 8°C.
Blood specimens exhibiting gross hemolysis or contamination should not be used. Do not use specimens collected with gel separators.
Samples older than seven days can be tested, however antibody reactivity has been shown to decrease in older samples. A distinct separation between plasma and red blood cells must be visible for testing. Samples may be centrifuged or allowed to settle. Stored samples should be allowed to reach room temperature prior to testing.

Materials
Material provided
- Biotestcell® -I 8 or Biotestcell®-I 11

Material required but not provided
A. 3-phase-tube test
- Pipettes (drop volume 40 to 50 µl)
- Isotonic saline solution
- Anti-Human Globulin Anti-IgG (e.g. Biotest REF 804175100)
- Anti-Human Globulin, Anti-IgG, -C3d Polyspecific (e.g. Biotest REF 804115100)
- IgG coated red blood cells (e.g. Biotest Coombscell-E REF 816030100)
- Biotest MLB2 (Modified LISS Biotest 2) REF 805200100
- Glass tubes 10 x 75mm or 12 x 75mm
- Serological centrifuge
- Interval Timer
- Markers
- Optical aid (optional). The use of an optical aid for agglutination reading must be vaildated by the user.

Resuspend Reagent Red Blood Cells prior to use and allow to reach room temperature.

Material required but not provided
B. Tango® optimo
- TANGO® optimo Biotest REF 848900010
- Solidscreen II Biotest REF 806521100
- Anti-Human Globulin, Anti-IgG Solidscreen II REF 806516100
- Biotest MLB2 (Modified LISS Biotest 2) REF 805200100
- Solidscreen II Control REF 806514100
- Solidscreen II Control B REF 806519100
- Solidscreen II Negative Control REF 806509100
- Alsevers Solution REF 806510100
- Cell mixers

Test Procedure
Resuspend Reagent Red Blood Cells prior to use and allow to reach room temperature.

A. 3-phase-test
If an enzyme or enhancement reagents (albumin, LISS) is used, please refer to the respective instructions for use. If an autocontrol was not performed with antibody detection testing, it should be performed with antibody identification testing.

Phase 1: Immediate Spin
1. Place two drops of serum/plasma to be tested into each tube labelled for a selected red blood cell.
2. Add 1 drop of corresponding Biotestcell® Reagent Red Blood Cell suspension to tube labelled for it and mix.
3. Centrifuge for 20 seconds at 800 -1000 x g.
4. Gently dislodge the red blood cell button and observe for agglutination.
5. Record results

Often the immediate centrifugation test shows expression of anti-M, -N, -P and cold reactive antibodies.

Phase 2: Incubation

Refer to instructions for the enhancement reagent being used.

6. Incubate at 37°C for 30 to 60 minutes or as appropriate to the enhancement reagent used.
7. Centrifuge for 20 seconds at 800 -1000 x g.

8. Gently dislodge the red blood cell button and observe for agglutination.
9. Record results

With the centrifugation test after incubation mainly Rh-antibodies as well as some incomplete antibodies are detected.

Phase 3: Indirect Antiglobulin-Test

10. Wash the red blood cells 3 times with isotonic saline. Decant supernatant saline completely.
11. Follow the directions of the Anti-Human Globulin manufacturer.
12. Centrifuge for 20 seconds at 800 -1000 x g.
13. Gently dislodge the red blood cell button and observe for agglutination.
14. Record results

This test shows IgG antibodies such as anti-Duffy, anti-Kidd, all Rh-antibodies.

B. Solidscreen® II and TANGO® optimo

Detailed test procedure instructions as well as details for the evaluation of test results are given in the instructions for use of Solidscreen II and TANGO® optimo, User's Guide.

Stability of the Reaction

Tube Test

Following centrifugation, all tube tests should be read immediately and results interpreted without delay. Time delays may cause a dissociation of the antigen-antibody complexes resulting to false negative or more often weak positive reactions.

TANGO® optimo

The results are read immediately and a digital image is stored for review by the operator. Image algorithms contained in the TANGO® optimo evaluate, and provide an interpretation (positive or negative) for the well. The operator performs verification of the final results.

Quality Control

A series of quality control samples must be run each day before testing or according to local requirements to ensure that the reagents and analyzer are functioning properly.

Refer to TANGO® optimo instructions for recommended instrument quality control.

Controls should be run whenever:

- Lot numbers change (plate, reagent).
- A new bottle or preparation is placed on the system.
- After service/repair of the analyzer.

Negative results in a tube antiglobulin test should be verified with IgG coated red blood cells in a tube test: Add 1 drop of IgG coated red blood cells, mix and centrifuge for 20 seconds at 800 - 1000 x g. Positive result: The negative reaction in the indirect antiglobulin test is valid, reactive Anti-Human-Globulin is present. Negative result: A technical error was made and the test must be repeated.

Interpretation of QC

The tests are considered valid if the expected results for the controls are obtained. If the controls do not give the expected results, you must determine the cause for the failed QC.

Follow institutional SOP for repeat testing of QC samples, repeat testing of patient/donor samples and documentation of QC results and corrective action if required.

Please contact Biotest Diagnostics Corporation (800-522-0090) if controls repeatedly fail to give expected results.

Interpretation of results

Tube Test

Agglutination of the red blood cells is a positive result and indicates the presence of an unexpected antibody(ies). No agglutination is a negative result and indicates that no unexpected antibody was detected.

An agglutination viewer may facilitate the reading of tube tests (as recommended by the AABB Technial Manual, 15th edition).

The positive and negative reactions are compared to the Biotestcell®-I8 & I 11 antigen pattern and are read accordingly. For verification an additional antigen test of the red blood cells may be performed.

TANGO® optimo

When used for testing with Solidscreen II and the TANGO® optimo, a smooth monolayer of red blood cells is indicative of a positive reaction. A compact button of red blood cells in the middle of the well is indicative of a negative reaction.

The results are read immediately and a digital image is stored for review by the operator. Image algorithms contained in the TANGO® optimo evaluate, and provide an interpretation (positive/negative) for the well. The operator performs verification of the final results.

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The positive and negative reactions are compared to the Biotestcell®-I8 and Biotestcell®-I 11 antigen pattern and are read accordingly. For verification an additional antigen test of the red blood cells may be performed.

Limitations

- Negative reactions and subsequent positive reactions with IgG coated red blood cells indicate that the serum contains no detectable antibodies against one of the antigens present on the Reagent Red Blood Cells (enclosed antigen pattern).
- Because some antibodies show dosage effect, the antigen density on the Reagent Red Blood Cells needs to be considered when evaluating the test results (homozygous or heterozygous hereditary disposition). A heterozygous expression of the antigen may result in non-detection of weak antibodies depending on the used test method.
- In very rare cases HLA-antigens within the product may lead to false positive reactions.
- The reactivity of the product may decrease during the dating period and therefore should not be used after the expiration date. The rate of decrease in reactivity is partially dependant on individual donor characteristics that are neither controlled nor predicted by the manufacturer.
- Negative reactions will be obtained if the sample contains antibodies present in concentrations too low to be detected by the test method employed. No test method is capable of detecting all red cell antibodies.

Specific Performance Characteristics

Testing is performed in accordance with FDA recommended methods.

The final release testing is performed according to the product specific SOPs. Each lot of Biotest Reagent Red Blood Cells is tested in the Quality control by package insert method against a panel of blood grouping reagents to insure suitable reactivity. To exclude a mixup of the Reagent Red Blood Cells with identical Rh phenotype at least one differential antigen has to be tested. The result must react appropriately positive or negative.

No FDA Standard of potency. For the product performance it is necessary to adhere to the recommended method in the instructions for use.

For Technical Support or further product information, contact Biotest Diagnostics Corporation at 800-522-0090.

Note

Each facility should verify the optimum spin time for the specific centrifuge in use.

Manual techniques are to be performed according to the manufacturer's instructions. Each deviation from these instructions is the sole responsibility of the user. Used test material must be discarded as hazardous material. Manage waste according to local, state and national regulations.

Glossary of Symbols

Symbol	Definition	Symbol	Definition
LOT	Batch Code	IVD	*In vitro* diagnostic medical device
⚠	Caution, consult accompanying documents		Consult instructions for use.
	Manufacturer		Use by YYYY-MM-DD
	Contains sufficient quantity for <n> tests.	REF	Catalog number
	Temperature limitation	VOL	Volume

Bibliography

1. Mark E. Brecher, MD et al. Technical Manual 15th Edition, Bethesda, MA: AABB, 2005.

Key: Underline = Addition of changes ◄ = Deletion of text

Biotest

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