US

Blood Grouping Reagent

Erytype S Rh + K Type

FOR IN-VITRO DIAGNOSTIC USE
Microplate for Tango® optimo
MEETS FDA POTENCY REQUIREMENTS
U.S. License Number: 1798

Package size
REF 806195100 VOL 10 plates Erytype S Rh + K Type

Intended Use
Each microplate is used for the determination of the presence or absence of the D, C, c̄, E, e and K antigens on human red blood cells with the TANGO® optimo.

Summary
Landsteiner and Wiener first described the Rhesus blood group system in 1940.[1]The antigen discovered is now known as the D antigen.
The D antigen is the most important antigen outside of the ABO blood group system. Most D negative and D "category" (i.e. DVI) individuals will make anti-D when exposed to the D antigen, through transfusion or during pregnancy as a result of a feto-maternal hemorrhage.
More than 50 antigens belong to the Rhesus blood group system. The antigens C, c̄, E and e, along with D, are the principle antigens of the Rh system. Although many other antigens have been identified, the antibodies associated with these 5 antigens are responsible for the majority of hemolytic transfusion reactions and cases of hemolytic disease of the fetus and newborn associated with the Rh system[2].
For the determination of Rh phenotypes the C, c̄, E and e antigens on the red blood cells are tested with Biotest Anti-C (RH2), Anti-c̄ (RH4), Anti-E (RH3) and Anti-e (RH5). If the red blood cells carry only big C or little c̄ (or E or e), the individual is treated as being homozygous for that particular antigen (allele). The most probable genotype can be presumed by determining the phenotype. The ethnic origin influences the genotype, which can be seen in the table.

Incidence of the More Common Genotypes in D+ Persons[2]

Antigens Present	Genotype DCE	Mod. Rh-hr	Incidence (%) Whites	Blacks
D,C,c,e	DCe/ce	R_1r	31.1	8.8
	DCe/Dce	R_1R_0	3.4	15.0
	Dce/Ce	R_0r'	0.2	1.8
D,C,e	DCe/DCe	R_1R_1	17.6	2.9
	DCe/Ce	R_1r'	1.7	0.7
D,c,E,e	DcE/ce	R_2r	10.4	5.7
	DcE/Dce	R_2R_0	1.1	9.7
D,c,E	DcE/DcE	R_2R_2	2.0	1.3
	DcE/cE	R_2r''	0.3	<0.1
D,C,c,E,e	DCe/DcE	R_1R_2	11.8	3.7
	DCe/cE	R_1r''	0.8	<0.1
	DcE/Ce	R_2r'	0.6	0.4
D,c,e	Dce/ce	R_0r	3.0	22.9
	Dce/Dce	R_0R_0	0.2	19.4

The Kell antigen was first identified in 1946 when the corresponding antibody was found to cause hemolytic disease of the fetus and newborn (HDFN). Anti-K antibody has also been shown to cause hemolytic transfusion reactions (HTR). Although in low density on the red blood cell membrane the Kell antigen is strongly immunogenic.[2]
The frequencies of the common phenotypes of the Kell System are shown in the table.

Phenotypes and Frequencies in the Kell System[1]		
Phenotype	Whites	Blacks
K+k-	0.2	Rare
K+k+	8.8	2
K-k+	91.0	98
Kp (a+b-)	Rare	0
Kp (a+b+)	2.3	Rare
Kp (a-b+)	97.7	100
Js (a+b-)	0.0	1
Js (a+b+)	Rare	19
Js (a-b+)	100.0	80
K_0	Exceedingly rare	

Biotest Erytype S Rh+K Type blood group reagents are used to test for the presence or absence of the D, C, c, E, e and K antigen. Routine pretransfusion studies always include tests for the D antigen. The other Rhesus reagents like Anti-C (RH2), Anti-c̄ (RH4), Anti-E (RH3), Anti-e (RH5) and Anti-K (KEL1) are used principally in the resolution of antibody problems or in family studies.

Principle of the Test
The test principle is hemagglutination. The antigens on the red blood cells being tested react with the respective reagent in the strip wells. Agglutinates form if the antigen is present on the red blood cells for the corresponding antibody being tested.

Reagent
Each strip on the Erytype S Rh+K Type contains the following configuration for the performance of Rh and K typing. Reagents are dried on the strips in the order depicted below.

The following antibodies are produced using intermediate products produced for Biotest Medical Diagnostics in a shared manufacturing agreement with Millipore, Ltd., 9 Fleming Road, Kirkton Campus, EH547BN, Livingston, UK; License Number 1721.
MS24, MS33, MS260, MS212, MS16, MS21, MS63 and MS 56.

The following antibody is produced using intermediate products produced for Biotest Medical Diagnostics GmbH in a shared manufacturing agreement with DIAGAST, Parc Eurasante, 251 av. Eugene Avinee-BP9, 59374 Loos Cedex France; License Number 1744.
P3X25513G8

Preservative: 0.1% sodium azide

Well No.	Reagent	Source	Antibody Class	Clone	Manuf.
A	Anti-D	Human Monoclonal	IgM	BS226	Biotest / Sifin
B	Anti-D	Human Monoclonal	IgM	BS232	Biotest / Sifin
C	Anti-C	Human Monoclonal	IgM IgM	MS24 P3X25513 G8	Millipore/ Diagast
D	Anti-c̄	Human Monoclonal	IgM	MS33	Millipore
E	Anti-E	Human Monoclonal	IgM IgM	MS260 MS12	Millipore
F	Anti-e	Human Monoclonal	IgM IgM IgM	MS16 MS21 MS63	Millipore
G	Anti-K	Human Monoclonal	IgM	MS56	Millipore
H	Neg. Control	Casein diluent + preservative	/	/	Biotest

Precautions
- For In-vitro diagnostic use.
- Store at 2 to 8°C.
- Do not use beyond the expiration date.
- Handle and dispose of reagents as potentially infectious
- Caution: All Blood Products should be treated as potentially infectious. Source material from which this product was derived 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 derived from human blood will not transmit infectious agents.
- Warning: Contains sodium azide (NaN_3), which may react with lead or copper plumbing to form explosive azides. If discarded in the sink, flush with large amounts of water to prevent the build-up of explosive metal azides.
- Do not use beyond seven days when loaded on the TANGO® optimo.
- Opened plates that are not loaded onto the TANGO® optimo can be used for up to 24 hours if stored in a dry area at room temperature (20 to 24°C).
- Do not reuse test strips.
- Let plate come to room temperature before opening the foil packet to limit condensation.
- Do not use samples collected with gel separators of any kind.

Specimen Collection
EDTA or citrate 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 specimens should be stored at 2 to 8°C. Citrated specimens (donor segments) at 1 to 6°C.
Use of EDTA anticoagulated samples older than seven days should be avoided. Donor blood stored in citrate anticoagulant at 1 to 6°C may be tested until the expiration date of the donor unit. These red blood cells to be tested must be prepared prior to testing. Refer to instructions in the

TANGO® optimo Users Guide. Stored samples should be allowed to reach room temperature prior to testing. Blood specimens exhibiting gross hemolysis or contamination should not be used.

Materials
Material provided
- Erytype S Rh + K Type

Material required but not provided
- TANGO® optimo (e.g. Biotest REF 848900010)
- Bromelin for Erytype (e.g. Biotest REF 806210100)

Test Procedure
TANGO® optimo (Erytype S)
Please refer to the instructions for use of the TANGO® optimo.

Stability of the Reactions
For the TANGO® optimo the results are read immediately and a digital image is stored for review by the operator. Image algorithms contained in the software evaluate and provide an interpretation (positive or negative) of 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.

Additionally, 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.

Control samples should be selected to verify positive and negative reactions with every reagent.

Controls may be selected from previously tested blood samples. Controls should be selected from samples that are less than seven days old. Clotted, grossly hemolyzed, or grossly lipemic specimens should not be used as quality control samples. Control samples should be selected to verify positive and negative reactions with every reagent.
To confirm the reactivity or specificity of Biotest Erytype S Rh + K Type Blood Grouping Reagents, each should be tested with antigen-positive (preferably from heterozygous individuals) and antigen-negative red blood cells, respectively. Each reagent is satisfactory for use if it reacts only with antigen-positive red blood cells.
A negative control is performed on each samples tested with Erytype S Rh + K Type.

Interpretation of QC
The tests are considered valid if an appropriate positive and negative result exists for each reagent tested. A positive result is not expected for the negative control. If the controls do not give the expected results, you must determine the cause for the failed QC.
Frequencies in the population are listed in the "Summary" section.

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

Interpretation of results
Negative result: a diffuse suspension of red blood cells throughout the well.
Positive result: an aggregate of red blood cell clumps at the bottom of the well.
For the TANGO® optimo the results are read immediately and a digital image is stored for review by the operator. Image algorithms contained in the software evaluate and provide an interpretation (positive or negative) of the well. The operator performs verification of the final results.

Reagent sera with patient red blood cells				
Anti-D	Control	D^{weak} Test	DAT**	Interpretation
+	0	/	/	Rh positive
0	0	0	0	Rh negative
0	0	+	0	* Rh positive
0	0	+	+	Invalid Test
+	+	/	/	Invalid Test

+ = agglutination 0 = no agglutination

*A test for weak D may be performed on samples that test negative with Anti-D to determine the Rh status. A reagent containing an IgG anti-D must be used. Certain groups of patients may require testing for weak D. Follow facility specific policies guidance for determining which samples require weak D testing.

**Testing is not valid unless the sample can be shown to react negatively with an appropriate Rh control or exhibits a negative direct antiglobulin test.

Limitations
- Category VI D antigen will not be detected with the anti-D on this strip. Category VII and very weak expressions of the D antigen (D weak with very few receptors) will give weakened or negative reactions with the anti-D on this strip.
- Contaminated materials, sample condition (excessive lipemia or hemolysis) improper centrifugation or pipetting may cause false test results.
- False positives may occur if the TANGO® optimo is stopped during processing for too long a period of time. The orbital shaker on the analyzer keeps red blood cells in suspension while the CCD camera is reading strips. Stopping this process may allow red blood cells to settle in the center of the well on the strip, thus leading to a false interpretation of the well.
- Some conditions that may cause false positive results are:
  - Contamination of sample or reagents
  - Autoantibodies
  - Improper storage or preparation of red blood cells
  - Antibodies to antibiotics or other reagents in the TANGO® optimo
  - Cold 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 Erytype S Rh + K Type Blood Grouping Reagents is tested in the Quality control by package insert method against a panel of antigen positive red blood cells (heterozygous antigen expression and if possible weakened antigen expression) to insure suitable reactivity. The products meet FDA potency requirements. The specificity testing for the presence of contaminating antibodies is performed according to the product specific SOPs.
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
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. Issitt, Peter D., Issitt, Charla H. Applied Blood Group Serology, Oxnard, CA:Spectra Biologicals, 1979.
2. Mark E. Brecher, MD et al. Technical Manual 15th Edition, Bethesda, MA: AABB, 2005.

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