BECKMAN COULTER

IMMAGE Immunochemistry Systems

Instructions For Use

RF

Rheumatoid Factor

REF 447070 (300 tests)

For *In Vitro* Diagnostic Use

FOR PROFESSIONAL USE ONLY

Rx Only

PRINCIPLE

INTENDED USE

RF reagent, when used in conjunction with IMMAGE Immunochemistry Systems and Calibrator 5 Plus, is intended for the quantitative determination of rheumatoid factor (RF) in human serum or plasma by rate nephelometry.

CLINICAL SIGNIFICANCE

Rheumatoid Factors are antibodies that react with the individual's own immunoglobulin. The antibodies are usually directed against the Fc region of the IgG molecule.[1] Rheumatoid Factors can be detected in the serum of the majority of patients with rheumatoid arthritis and is important for the diagnosis and prognosis of those patients with higher concentrations. These patients tend to suffer a more severe form of the illness and develop extra-joint complications more readily.[2,3] Rheumatoid Factors are not disease specific and can occur in lower frequencies in several other autoimmune disorders, chronic inflammation and normal individuals.[4,5]

METHODOLOGY

The RF test measures the rate of increase in light scattered from particles suspended in solution as a result of complexes formed during an antigen-antibody reaction.

CHEMICAL REACTION SCHEME

Rheumatoid Factor(sample) + Particle bound-human IgG(antigen) ⟶ [Rheumatoid Factor(sample)-Antigen Complex]

E011333L.EPS

SPECIMEN

TYPE OF SPECIMEN

Serum samples are recommended. Plasma samples (Lithium Heparin and Sodium Heparin) can be used. EDTA is an unacceptable anticoagulant.

Serum or plasma samples should be collected in the manner routinely used for any clinical laboratory test.[6] Freshly drawn serum or plasma from a fasting individual is preferred. Anticoagulants tested are listed in the PROCEDURAL NOTES section of this instructions for use.

SPECIMEN STORAGE AND STABILITY

1. Tubes of blood are to be kept closed at all times and in a vertical position. It is recommended that the serum or plasma be physically separated from contact with cells within two hours from the time of collection.[7]

2. If serum and plasma samples are not assayed within 8 hours, samples may be stored at +2°C to +8°C. If serum and plasma samples are not assayed within 48 hours, samples should be stored frozen at -15°C to -20°C for up to 3 months. Frozen samples should be thawed only once. Analyte deterioration may occur in samples that are repeatedly frozen and thawed.[7,8]

Additional specimen storage and stability conditions as designated by this laboratory:

SAMPLE VOLUME

For sample volumes refer to the Sampling Template.

CRITERIA FOR UNACCEPTABLE SPECIMENS

Refer to the PROCEDURAL NOTES section of this instructions for use.

Criteria for sample rejection as designated by this laboratory:

PATIENT PREPARATION

Special instructions for patient preparation as designated by this laboratory:

SPECIMEN HANDLING

Special instructions for specimen handling as designated by this laboratory:

REAGENTS

CONTENTS

Each kit contains the following items:

KIT COMPONENTS	QUANTITY
RF Cartridge	1
Antigen	
Evaporation Caps	2
RF Reagent Bar Code Card	1

INITIAL VOLUMES OF SAMPLE AND REAGENTS IN THE CUVETTE

Sample Volume	5 µL
Total Reagent Volume	346 µL
Antigen	21 µL
Buffer 2	300 µL
Diluent 1	25 µL

REACTIVE INGREDIENTS

REAGENT CARTRIDGE CONSTITUENTS	VOLUME
RF Antigen (particle bound human immunoglobulin G)	7.6 mL
Sodium Azide (used as a preservative)	< 0.1% (w/w)

Also bovine serum albumin and non-reactive chemicals necessary for optimal system performance.

⚠ **CAUTION**

Sodium azide preservative may form explosive compounds in metal drain lines. See NIOSH Bulletin: Explosive Azide Hazard (8/16/76).
To avoid the possible build-up of azide compounds, flush wastepipes with water after the disposal of undiluted reagent. Sodium azide disposal must be in accordance with appropriate local regulations.

<div style="border: 1px solid black; padding: 10px;">

⚠ CAUTION

Because this product is of human origin, it should be handled as though capable of transmitting infectious diseases. Each serum or plasma donor unit used in the preparation of this material was tested by United States Food and Drug Administration (FDA) approved methods and found to be negative for antibodies to HIV and HCV and nonreactive for HbsAg. Because no test method can offer complete assurance that HIV, hepatitis B virus, and hepatitis C virus or other infectious agents are absent, this material should be handled as though capable of transmitting infectious diseases. This product may also contain other human source material for which there is no approved test. The FDA recommends such samples to be handled as specified in Centers for Disease Control's Biosafety Level 2 guidelines.[9]

</div>

GHS HAZARD CLASSIFICATION

Not classified as hazardous

SDS	Safety Data Sheet is available at beckmancoulter.com/techdocs

MATERIALS NEEDED BUT NOT SUPPLIED WITH REAGENT KIT

IMMAGE Immunochemistry Systems Wash Solution
IMMAGE Immunochemistry Systems Buffer 2
IMMAGE Immunochemistry Systems Diluent 1
Calibrator 5 Plus
At least two levels of control material

REAGENT PREPARATION

1. Invert cartridge gently before removing screw caps.

2. Remove screw caps from reagent cartridges. Check each cartridge for bubbles and remove any bubbles present.

3. Place evaporation caps on both reagent cartridge compartments before loading the cartridge on the instrument.

4. Reagent cartridges should be stored upright and can be removed from the refrigerator and used immediately.

5. Mix all buffers and diluents thoroughly by inversion. Remove screw cap from container. Check each container for bubbles and remove any bubbles present. Place evaporation cap on container before loading the container on the instrument.

ACCEPTABLE REAGENT PERFORMANCE

Acceptability of a reagent is determined from the successful performance of quality control testing, as defined in the QUALITY CONTROL section of this instructions for use.

REAGENT STORAGE AND STABILITY

Storage conditions other than those recommended may cause erroneous results.

Reagent Cartridges

1. Return all reagent cartridges to the refrigerator (+2°C to +8°C) upon completion of the daily workload.

2. The RF reagents are stable for 30 days with the evaporation caps in place.

3. The RF reagents are stable until the expiration date on the label if the reagents are stored at +2°C to +8°C with the screw caps in place.

Diluent 1 and Buffer 2

1. Diluent 1 and Buffer 2 are stable on the system for 30 days with the evaporation caps in place.

2. Diluent 1 and Buffer 2 are stable until the expiration date on the label if they are stored at room temperature with the screw caps in place.

Reagent storage location:

CALIBRATION

CALIBRATOR REQUIRED

Calibrator 5 Plus

CALIBRATOR PREPARATION

No preparation is required.

CALIBRATOR STORAGE AND STABILITY

The calibrator is stable until the expiration date printed on the calibrator bottle if stored capped in the original container at +2°C to +8°C.

Calibrator storage location:

⚠ CAUTION

Because this product is of human origin, it should be handled as though capable of transmitting infectious diseases. Each serum or plasma donor unit used in the preparation of this material was tested by United States Food and Drug Administration (FDA) approved methods and found to be negative for antibodies to HIV and HCV and nonreactive for HbsAg. Because no test method can offer complete assurance that HIV, hepatitis B virus, and hepatitis C virus or other infectious agents are absent, this material should be handled as though capable of transmitting infectious diseases. This product may also contain other human source material for which there is no approved test. The FDA recommends such samples to be handled as specified in Centers for Disease Control's Biosafety Level 2 guidelines.[9]

IMMAGE IMMUNOCHEMISTRY SYSTEM CALIBRATION INFORMATION

1. The IMMAGE Immunochemistry Systems calibration is reagent lot specific.

2. The RF reagent lot should be recalibrated when changing the Buffer 2 lot or following specific part replacements or maintenance procedures as defined in the IMMAGE *Operations Manual* .

3. The IMMAGE Immunochemistry System is designed for minimum calibration. Calibrations retained in system memory should be monitored by the performance of quality control procedures on each day of testing.

4. Calibration for RF is stable for 30 days.

5. The system will automatically perform a verification check during calibration and produce a calibration report. The system will alert the operator of a failed calibration. An explanation of any accompanying error message can be found in the TROUBLESHOOTING Section of the IMMAGE Immunochemistry Systems *Operations Manual*.

TRACEABILITY

For Traceability information refer to the Calibrator instructions for use.

QUALITY CONTROL

It is recommended that at least two levels of control material, normal and abnormal, be analyzed daily. Controls should also be run with each new calibration, with a new lot of reagent or buffer, and after specific maintenance or troubleshooting as detailed in the IMMAGE Immunochemistry Systems *Operations Manual*. More frequent use of controls or the use of additional controls is left to the discretion of the user based on work load and work flow.

The following controls should be prepared and used in accordance with the package inserts. Discrepant quality control results should be evaluated by your facility.

Table 1.0 Quality Control Material

CONTROL NAME	SAMPLE TYPE	STORAGE

TESTING PROCEDURE(S)

1. After setup, load reagents onto the system as directed in the IMMAGE *Operations Manual*.

2. Select chemistries to be calibrated, if necessary. Load bar coded calibrators, controls, and samples or program and load non-bar coded controls and samples for analysis as directed in the IMMAGE *Operations Manual* .

3. Follow the protocols for system operation as directed in the IMMAGE *Operations Manual*.

CALCULATIONS

The IMMAGE Immunochemistry System will automatically calculate results.

REPORTING RESULTS

REFERENCE INTERVALS

Greater than 95% of a group of 108 apparently healthy adults in Southern California showed no detectable RF (<20 IU/mL) with this assay. Published "upper limits of normal" vary widely among different sources.[10,11,12] Each laboratory must establish its own reference interval based on its patient population. The presence of RF in patient serum should be considered as one criterion of rheumatoid disease, and not as a definitive diagnosis.[13]

Table 2.0 Reference intervals

	REFERENCE INTERVALS
Beckman Coulter	< 20 IU/mL (in 95% of the population tested)

	REFERENCE INTERVALS
Laboratory	

Refer to References (14,15) for guidelines on establishing laboratory-specific reference intervals.

Additional reporting information as designated by this laboratory:

UNITS AND CONVERSION FACTOR

Results for the RF test are reported in default units of IU/mL. Metric conversion within the same unit category will occur automatically if a new unit is selected. A conversion factor must be entered when selecting a unit category different from the default.

Refer to the System Setup section of the IMMAGE *Operations Manual* for more detailed information on units and conversion factors.

PROCEDURAL NOTES

ANTICOAGULANT TEST RESULTS

The following anticoagulants were tested. The results of 20 serum and 20 plasma samples, containing the same level of RF, were tested and averaged.

Table 3.0 Anticoagulant Test Results

ANTICOAGULANT	LEVEL OF ANTICOAGULANT TESTED[a]	AVERAGE SERUM RESULT (IU/mL)	AVERAGE PLASMA RESULT (IU/mL)	% DIFFERENCE
Lithium Heparin	14 Units/mL	164	160	-2.44
Sodium Heparin	14 Units/mL	154	152	-1.30

a   Based on full tube collection.

LIMITATIONS

1. Rheumatoid factor is not a finding isolated to rheumatoid arthritis and may be present in a number of connective tissue and inflammatory diseases including infectious mononucleosis, systemic lupus erythematosus, scleroderma, and hepatitis.[16]

2. Extremely high RF concentrations (> 6,000 IU/mL) in serum may result in an antigen excess condition producing a low RF value on the IMMAGE System. If the patient`s clinical condition does not correlate with the reported RF concentration, dilute the sample and repeat the analysis.

INTERFERENCES

1. The following substances were tested in serum for interference with this methodology at the initial dilution:

Table 4.0 Interferences

SUBSTANCE	SOURCE	LEVEL TESTED	OBSERVED EFFECT
Bilirubin	Porcine	5 – 30 mg/dL	None
Lipid	Human Triglyceride	50 – 300 mg/dL	None[a]
Hemoglobin	Human	200 – 1,000 mg/dL	None

a   Quantitation of specific proteins by nephelometry may not be possible in lipemic samples due to the extreme light scattering properties of the sample.

2. Samples stored refrigerated or frozen for extended periods may exhibit aberrant results.

3. Serum samples may yield different concentration values for rheumatoid factor when analyzed at various dilutions. This phenomenon may be caused by the presence of rheumatoid factor complexes with inhibitors or interfering substances present in the serum. Sample dilution will reduce the effective concentration of the interfering substances so that the final result obtained for rheumatoid factor in the diluted sample will be increased over the result obtained for the undiluted (neat) or lesser diluted sample.

4. Dust particles or other particulate matter (i.e. debris and bacteria) in the reaction solution may result in extraneous light-scattering signals, resulting in variable sample analysis.

5. Quality control materials prepared from serum pools are not recommended.

PERFORMANCE CHARACTERISTICS

NOTE ON PERFORMANCE CHARACTERISTICS

Performance characteristics are based on the initial analytical range. The instrument automatically dilutes samples to be within the initial analytical range. Refer to IMMAGE Immunochemistry Systems Operations Manual for more information.

ANALYTIC RANGE

The RF test is designed to detect concentrations of this analyte using an initial 1:6 serum sample dilution.

Table 5.0 Analytical Range

SAMPLE TYPE	BECKMAN COULTER ANALYTICAL RANGE
Serum	Initial: 20 – 800 IU/mL
	Extended: 20 – 28,800 IU/mL

REPORTABLE RANGE (AS DETERMINED ON SITE):

Table 6.0 Reportable Range

SAMPLE TYPE	LABORATORY REPORTABLE RANGE

SENSITIVITY

Sensitivity is defined as the lowest measurable concentration which can be distinguished from zero with 95% confidence. Sensitivity for RF determination is 20 IU/mL.

EQUIVALENCY

Equivalency was assessed by Deming regression analysis of samples to an accepted clinical method. Values obtained for human rheumatoid factor using the IMMAGE RF Test were compared to the values obtained using a commercially available automated nephelometric assay (NIA) method. Both normal and abnormal rheumatoid factor serum samples were included in the analysis.

Table 7.0 Equivalency Values

	NIA METHOD
N	143
Slope	1.055
Intercept	0.41
Mean (IMMAGE)	154
Mean (NIA)	146
Correlation Coefficient (r)	0.940

The equivalency values were determined using patient samples ranging from 20.0 to 758 IU/mL. Refer to References (16,17) at the end of this instructions for use for guidelines on performing equivalency testing.

PRECISION

A properly operating IMMAGE Immunochemistry Systems should exhibit imprecision values less than or equal to the maximum performance limits listed below. Maximum performance limits were derived by an examination of the precision of various methods, proficiency test summaries, and literature sources.

Table 8.0 Maximum Performance Limits

TYPE OF PRECISION	SAMPLE TYPE	SD (IU/mL)	% CV	CHANGEOVER VALUE (IU/mL)[a]
Within-run	Serum	5.0	4.0	125
Total	Serum	7.5	6.0	125

a   When the mean of the test precision data is less than or equal to the changeover value, compare the test SD to the SD guideline given above to determine the acceptability of the precision testing. When the mean of the test precision data is greater than the changeover value, compare the test % CV to the guideline given above to determine acceptability. Changeover value = (SD guideline/CV guideline) x 100.

Comparative performance data for the IMMAGE Immunochemistry Systems evaluated using the NCCLS Proposed Guideline EP5-T2 appears in the table below.[18] Each laboratory should characterize their own instrument performance for comparison purposes.

Table 9.0 Typical Imprecision Values

TYPE OF PRECISION	SAMPLE	Data Points[a]	Test Mean Value (IU/mL)	SD (IU/mL)	% CV
Within-run	Serum Level 1	80	124	2.5	2.1
	Serum Level 2	80	299	4.5	1.5
	Serum Level 3	80	637	12.4	1.9
Total	Serum Level 1	80	124	3.5	2.8
	Serum Level 2	80	299	8.8	2.9
	Serum Level 3	80	637	20.7	3.2

a   The point estimate is based on the data from 1 system, run for 20 days, 2 runs per day, 2 observations per run on an instrument operated and maintained according to the manufacturer`s instructions.

Refer to References (16,18) for guidelines on performing precision testing.

NOTICE

These degrees of precision were obtained in typical testing procedures and are not intended to represent performance specifications for this test procedure.

ADDITIONAL INFORMATION

For more information, refer to the IMMAGE Immunochemistry Systems *Operations Manual*.

Beckman Coulter, the stylized logo, and the Beckman Coulter product and service marks mentioned herein are trademarks or registered trademarks of Beckman Coulter, Inc. in the United States and other countries.

May be covered by one or more pat. -see www.beckmancoulter.com/patents.

SHIPPING DAMAGE

If damaged product is received, notify your Beckman Coulter Clinical Support Center.

REVISION HISTORY

Revision AE

Updated references to manual sections.

Revision AF

Updated corporate address.

Revision AG

Added Revision History

Revision AH

Added new language requirement: Czech, and Korean.

Revision AJ

Revised Reagent Cartridge Constituents Volume.

Revision AK

Added GHS classification and Trademark statement.

Revision AL

Added new language requirement: Romanian

Revision AM

Updates to comply with requirements per Beckman Coulter Global Labeling Policy.

Revision AN

Added new language requirement: Brazilian Portuguese.

Revision AP

Added new language requirement: Dutch, and Slovak. Additional changes to comply with requirements per Beckman Coulter Global Labeling Policy.

Revision AR

Removed references to "Chemistry Reference Manual", CE mark and EC Rep name and address.

Revision of sample stability information, minor instructional clarification and corrections throughout, and updates to comply with requirements per Beckman Coulter Global Labeling Policy.

SYMBOLS KEY

Table 10.0

REF	Catalogue Number	IVD	In Vitro Diagnostic
CONTENTS	Contents		Temperature limit
	Manufacturer		Expiration Date
LOT	Batch code	SDS	Safety Data Sheet
CE	CE Mark		Consult Instructions for Use
EC REP	Authorized Representative in the European Community		Date of Manufacture
⚠	Caution		Biological risks
REAGENT CART	Reagent Cartridges	CAPS	Caps
BAR CODE CARD	Bar Code Card	SN	Serial Number
Made in USA of US and Foreign Components		Made in USA of US and Foreign Components	

REFERENCES

1. Johnson, P. M., Faulk, W. P., "Rheumatoid Factor: Its Nature, Specificity and Production in Rheumatoid Arthritis", Clin. *Immunol. Immunopathol.*, 6:414 440 (1976).

2. Zrein M., De Marcillac, G., Van Regenmortel M. H. V., "Quantitation of Rheumatoid Factor by Enzyme Immunoassay Using Biotinylated IgG", *J. Immunol. Methods*, 87:229 237 (1986).

3. Msai, A. T., Maldonado-Cocco, J. A., Kaplan, S. B., Frigenbaum, S. L., Chandler, R. W., "Prospective Study of the Early Course of Rheumatoid Arthritis in Young Adults: Comparison of Patients With and Without Rheumatoid Factor Positively at Entry and Identification Variables Correlating with Outcome", *Semin. Arthritis Rheum.*, 5:229 326 (1976).

4. Wagner, O., *Ann Clin Res.*, 7:168 182 (1975).

5. Waller, M., Toone, E. C., *Arthritis Rheu.*, 11:50 55 (1968).

6. Tietz, N. W., "Specimen Collection and Processing; Sources of Biological Variation", *Textbook of Clinical Chemistry*, pp 478 518, W. B. Saunders, Philadelphia, PA (1986).

7. Clinical and Laboratory Standards Institute (CLSI). *Procedures for the Handling and Processing of Blood Specimens for Common Laboratory Tests* CLSI document GP44-A4. CLSI. (2010).

8. World Health Organization (WHO). Use of Anticoagulants in Diagnostic Laboratory Investigations and Stability of Blood, Plasma and Serum Samples, WHO/DIL/LAB/99.1-Rev.2. Geneva: WHO, (2002).

9. CDC-NIH, *Biosafety in Microbiological and Biomedical Laboratories,* 5th Edition, (Washington, D.C.: U.S. Government Printing Office, 2009). (CDC 21-1112)

10. Painter, P., Cope, J., Smith, J., "Reference Information for the Clinical Laboratory", Chapter 50, Tietz, *Textbook of Clinical Chemistry*, Burtis, C., Ashwood, E., eds. W.B. Saunders, New York (1999).

11. Tietz, N. W., ed.,*Clinical Guide to Laboratory Tests*, W.B. Saunders, New York (1995).

12. Mayo Medical Laboratories, *Interpretive Handbook: Interpretive Data for Diagnostic Laboratory Tests*, Mayo Clinic, Rochester, MN (1997).

13. Arnett, F., Edworthy, S., Bloch, D., McShane, D., Fries, J., Cooper, N., et al., The American Rheumatism Association 1987, " Revised Criteria for the Classification of Rheumatoid Arthritis", *Arthritis Rheum.*, 31:315 324 (1998).

14. Gary Horowitz and Graham R.D. Jones In TIETZ TEXTBOOK OF CLINICAL CHEMISTRY AND MOLECULAR DIAGNOSTICS, SIXTH EDITION. Ch 8 Establishment and Use of Reference Intervals. ISBN: 978-0-323-35921-4. Copyright © by Elsevier, Inc. All rights reserved (2018).

15. Clinical and Laboratory Standards Institute (CLSI), *Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline*, Third Edition, October 2010. EP28-A3c Vol. 28 No. 30 ISBN 1-56238-682-4, Formerly C28-A3c ISSN 0273-3099 Vol. 28 No. 30. (2010).

16. Tietz, N. W., ed., *Fundamentals of Clinical Chemistry*, 6th Edition, W. B. Saunders, Philadelphia, PA (2007).

17. National Committee for Clinical Laboratory Standards, *Method Comparison and Bias Estimation Using Patient Samples*, Tentative Guideline, NCCLS publication EP9-T, Villanova, PA (1993).

18. National Committee for Clinical Laboratory Standards, *Precision Performance of Clinical Chemistry Devices* Tentative Guideline, 2nd Edition, NCCLS publication EP5-T2, Villanova, PA (1992).

Beckman Coulter, Inc., 250 S. Kraemer Blvd., Brea, CA 92821 U.S.A.
+(1) 800-854-3633
www.beckmancoulter.com