Pariksha Biotech
A game changer in IVD

Command on Quality

Complement C3 Estimation Kit

High-Q C3 Factor

Turbidimetric Immuno Assay (TIA)

Intended Use:

For the Quantitative Determination of C3 Factor in Human Serum

Clinical Significance:

All complement proteins are acute phase reactants and rise rapidly in concentrations during inflammatory episodes. Conversely, the rates of complement protein catabolism may greatly increase in various autoimmune diseases. Because complement component determinations represent a static measurement of the net concentrations that result from a dynamic balance between component synthesis and catabolism, serial sample quantitations are more clinically useful. In most disease states, complement functions "normally" in producing inflammation and tissue damage. When complement plays a role in the development of a disease, it is often due to activation by an "abnormal" antibody, immune complex, or foreign material. Increased C3 levels are associated with acute phase reaction, rheumatic disease, viral hepatitis, myocardial infarction, cancer, diabetes, pregnancy, sarcoidosis, amyloidosis, thyroiditis, inflammatory bowel disease, typhoid fever, and pneumococcal pneumonia. The magnitude of C3 increase is rarely more than two-fold and may mask decreases in levels due to concurrent consumption. Decreased levels of C3 occur in individuals with congenital deficiency or immunologic diseases (where complement is consumed at an increased rate). C3 and / or complement C4 (C4) levels may be decreased in cases of: systemic lupus erythematosus (SLE) (especially with lupus nephritis), acute and chronic hypocomplementemic nephritis, infective endocarditis, disseminated intravascular coagulation (DIC) (especially with hemolytic uremic syndrome form), and partial lipodystrophy (with associated nephritis-like activity in serum).Cases of hereditary C3 deficiency, while rare, are characterized clinically by recurrent infection and by immune complex disease, in particular, membranoproliferative glomerulonephritis. The central role of C3 in both classical and alternate pathways, results in C3 deficient patients being at risk for especially severe infections by encapsulated bacteria such as S. pneumoniae, H. influenzae, and N. meningitidis. Bacteremia, sinopulmonary infections, meningitis, paronychia, and impetigo may occur. Deficient C3 levels have also been found in cases of uremia, chronic liver diseases, anorexia nervosa, and celiac disease.

Refer to the following table for a general guide to evaluation of C3 and C4 protein levels in the presence of decreased hemolytic complement activity.

	Normal C4	Decreased C4
Normal C3	1) Alterations in vitro (e.g., improper specimen handling) 2) Coagulation-associated complement consumption 3) Inborn errors (other than C3 or C4)	1) Immune Complex Disease 2) Hypergammaglobulinemic states 3) Cryoglobulinemia 4) Hereditary angioedema 5) Inborn C4 deficiency
Decreased C3	1) Acute glomerulonephritis 2) Membranoproliferative glomerulonephritis 3) Active SLE 4) Inborn C3 deficiency	1) Active SLE 2) Serum sickness 3) Autoimmune/chronic active hepatitis 4) Infective endocarditis 5) Immune complex disease

Principle:

Quantitative determination of C3 may be done by an immunoturbidimetric method, by automatic analyzers or in manual. Mixing a sample with a precise Antigen to a solution having the corresponding anti-serum (Antibody) in a well-defined ratio, it is possible to have turbidity; the use of undiluted sample may require bichromatism.

Using our calibrator which is traceable to the CRM 470 International Standard it is possible to determine the concentration of human serum sample

Precautions for use:

1. This product has been formulated for in vitro diagnostic use.
2. A proportional variation of the reaction volumes does not change the result.
3. DO NOT mix Reagents from different Production lots.

Kit Components:

Reagent 1:

(R1) 4-Hydroxyethyl Piperazine Ethanesulfonic acid 50mmol/L

Reagent 2:

(R2) Goat anti-human complement 3 antibody

C3 Calibrators :

High-Q Complement C3 is provided with 4 Levels of Lyophilized Calibrators. **Reconstitute each level with 0.5 ml of Distilled Water and keep it for 20 Minutes.** Mix gently and make a uniform suspension. Reconstituted Calibrators are stable for 60 Days once stored properly at 2-8°C. Aliquot it in to small volumes and store at 2-8°C for the contamination free use and for good reconstitution stability. Calibrators are stable for 6 Months when frozen at -20°C if the repeated freeze and thaw cycles are avoided.

Complement C3 Calibrators are validated with a traceability. Calibrators are calibrated to the Reference Material CRM 470/RPPHS (Institute for Reference Materials and Measurements).

Stability

The Reagents are stable up to the expiry date mentioned on the labels when properly stored at 2-8°C.

Preparation of the reagent:

Reagents are supplied as ready to use reagents and do not requires reconstitution and working reagent preparation.

Samples

• Not haemolysed and non lipemic fresh serum.
Samples collection in compliance with CLSI (NCCLS)
The sample can be stored at 2-8°C up to 6 days.

Quality Control:

Quality Control sera are recommended to monitor the performance of manual and automated assay procedures. High-Q Specific Proteins Controls are available optionally
Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

Test Procedure:

System Parameters:

Reaction type : **End Point-Bichromatic- Non Linear- Multi Cal- Spline**

Reaction Direction	: Increasing
Primary Wave Length	: 340
Secondary Wave Length	: 700 (600-700)
Flow cell Temp.	: 37°C
Sample volume	: 5 µl
Reagent volume	: R1 350 µl + R2 70 µl
Calibrators Conc: 1,2,3,4	: Lot Specific (Check the labels)
Units	: mg/dL
Blanking with	: Reagent
Low normal	: 90
High normal	: 180
Linearity	: 350

Reagent	Reagent Blank	C	S
R1	350 µL	350 µL	350 µL
Calibrators (1,2,3,4)	----	5 µL	----
Sample	----	----	5 µL
Incubate 5 Minutes at 37°C			
R2	70 µL	70 µL	70 µL

Mix carefully and wait for about 5 minutes. Measure the absorbance of calibrators and of the samples against blank reagent blank.

Calculations:

The Multipoint Non Linear /Semi logarithmic calibration model was used , and the Spline function was used as the calculation model. The dose / response curve was made based on the value of the calibrator and the change of absorbance. The concentration of C3 in the sample could be calculated on the dose/ response curve based on the change of absorbances

Within-run Precision: Determined on 20 replications of 2 samples.
The results obtained are following:

Run-to-run Precision: Determined for 5 days with 20 replications for each days, for two samples. The results obtained are the following:

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Accuracy: A group of 20 sera has been tested using this procedure and using a similar reagent available on the market. The comparison gave these results:

Sample	Mean (mg/dL) 2s	CV %
Human 1	68.3 1.4	1.1
Human 2	126.8 2.3	0.9

Sample	Mean (mg/dL) 2s	CV %
Human 1	67.7 2.8	2.1
Human 2	126.7 4.8	1.9

Linear regression equation y = 1.0341 x - 14
Correlation coefficient r = 0.9721 n = 20

Attention:

A) Applications on routine analyzers may be totally different from what developed as manual determination; in addition the procedures are specific for each analyzer.

B) Very deep attention must be given to interfering substances: certain drugs and other substances are able to influence levels of C3

C) The calibration curve has to be always repeated at each change of the lot of the Reagent and/or calibrator.
2. A proportional variation of the reaction volumes does not change the result.

D. For concentration of C3 higher than the maximum value of the Calibrator dilute the sample 1:5 with saline solution repeat the determination and multiply the result by 5.

Reference Values:

Neonates: Between 70 - 196 mg/dL.

Adults: Between 90 – 180 mg/dL.

Each laboratory should establish its own reference range

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Analytical Performances: (Validated on MINDRAY Bs300)

The performances of the Reagent **C3 FACTOR** has been tested with a MINDRAY BS300 analyzer. The data, while representing the characteristics of the product, could be different for each laboratory and for different analyzers.

Method Linearity: The test is linear up to 350 mg/dL.
However, for C3 concentrations higher than the maximum value of the calibrator, it is recommended to dilute the sample 1:5 with saline solution, and test again and multiply the result x 5.

Method Sensitivity (LoD): the sensitivity limit, that is the minimum concentration that can be distinguished by zero, is 4.2 mg/dL.

Interferences:

Interference test criterion: Recovery ± 30% of initial value. No interference found on samples with:

-Total bilirubin up to 20 mg/dL;
-Haemoglobin up to 150 mg/dL;
-Acorbic acid up to 50 mg/dL.

Bibliography:

1. Clinical Guide to Laboratory Tests, Edited by NW Tietz W B Saunders Co., Phipladelphia, 483, 1983.
2. Carrol MC. Annual Review of Immunology 1998; 16: 545-568.
3. Lambris JD. Cruse JM Lewis RE Jr (eds): Complement Today. Complement Profiles. Basel, Karger, 1993; Vol1: 16-45.

Ordering information

Ref./Cat.	Pack Size	Presentation
P-C3-24	24 ml	Two Reagents with 4 level Calibrators

Product Features

- **Turbidimetric Immuno Assay (TIA)**
- **Liquid Stable Two Reagents**
- **4 Level Lyophilized Calibrators Provided**
- **5 Minutes End Point Bichromatic Reaction**
- **Measurement at 340 nms**
- **Test Procedure time 5 minutes at 37°C**
- **Linearity : 350 mg/dL**
- **Adaptable to Semi and Automated Analyzers**

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Symbols used with IVD devices

	Date of manufacture		Manufactured by
IVD	In vitro diagnostic device		Keep away from sunlight
	Do not freeze		This way up
	Use by (yyyy-mm-dd or mm/yyyy)	R	Reagent
CAL	Calibrator Material	LOT	Batch code
2°C 8°C	Temperature limitation (store at)	CONTROL	Control
	Consult instructions for use		Keep dry Keep away from rain
REF	Catalog Number		

eIFU Indicator

Pariksha's world inside
SCAN TO EXPLORE MORE

Manufactured in India by :
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AN ISO 13485 Certified Company

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