Name: C3 Protein (Rat)

Catalog Number: R113 Sizes Available: 100 µg/vial

Concentration: 1.0 mg/mL (see Certificate of Analysis for actual concentration)

Form: Frozen liquid

Purity: > 85% by SDS PAGE

Buffer: 10 mM sodium phosphate, 145 mM NaCl, pH 7.2

Extinction Coeff. $A_{280 \text{ nm}} = 10.16 \text{ at } 1.0 \text{ mg/ml for pure C3}$

Molecular weight: 187,000 Da (2 chains)
Preservative: None, 0.22 μm filtered.

Storage: -70°C or below. Avoid freeze/thaw.

Source: Normal rat serum from healthy animals of mixed gender **Precautions:** Use normal precautions for handling animal blood products.

Origin: Manufactured in the USA.

General Description

Rat C3 is purified from pooled normal rat serum. C3 is central to the activation of all three pathways of complement activation (Law, S.K.A. and Reid, K.B.M. (1995)). Initiation of each pathway generates proteolytic enzyme complexes (C3 convertases) which are bound to the target surface. These enzymes cleave a peptide bond in C3 releasing the anaphylatoxin C3a and activating C3b. For a brief time (~60 µs) this nascent C3b is capable of reacting with and covalently coupling to hydroxyl groups on the target surface. Carbohydrates are the favored target, but protein hydroxyls and amino groups also react. This process of tagging the target surface with C3b is called opsonization. The reactive site in nascent C3b is a thioester (Tack B.J., et al. (1980); Pangburn M.K. and MüllerEberhard H.J. (1980)) and C3b is linked to the target through a covalent ester bond (an amide bond is formed if C3b is attached to amino groups). Most of the C3 activated during complement activation never attaches to the surface because its thioester reacts with water forming fluid phase C3b which is rapidly inactivated by factors H and I forming iC3b. Surface-bound C3b is necessary in all three pathways for efficient activation of C5 and formation of C5b-9 complexes that lyse the target cell membrane. Surface-bound C3b and its breakdown products iC3b and C3d are recognized by numerous receptors on lymphoid and phagocytic cells which use the C3b ligand to stimulate antigen presentation to cells of the adaptive immune system. The end result is an expansion of target-specific B-cell and T-cell populations.

Physical Characteristics & Structure

The calculated molecular weight of rat C3 based on its amino acid sequence is 184,111 daltons (without the signal peptide) and is similar to that of human C3 (185,000 daltons). The molecular weight of rat C3 as determined by SDS/polyacrylamide gel electrophoresis has been reported by Daha, M.R. et al., (1979) to be 187,000 daltons composed of two disulfide linked chains, alpha chain (123,000 daltons) and beta chain (76,000 daltons). The extinction coefficient of rat C3 (E^{1%}/₂₈₀nm = 10.16) is calculated based on its amino acid sequence using ProtParam and assumes all pairs of Cys residues form cystines (i.e. a pair of cysteine molecules are joined by a disulfide bond). The theoretical pI of rat C3 is 6.12. The normal plasma concentration of C3 in Wistar rats has been reported to be 0.581mg/ml (Daha, M.R. et al., (1979)).

Function

The biological functions of C3 are described above in the General Description section.

Genetics

Rat C3 chromosome location 9. The NCBI Gene ID number for rat C3 is 24232 and UniProt accession number is P01026.

Precautions/Toxicity/Hazards

This protein is purified from animal plasma/serum and therefore precautions appropriate for handling any animal blood-derived product must be used.

References

Law, S.K.A. and Reid, K.B.M. (1995) Complement 2nd Edition (ISBN 0199633568) Oxford University Press, Oxford.

Tack BF, Harrison RA, Janatova J, Thomas ML, Prahl JW. (1980) Evidence for presence of an internal thiolester bond in third component of human complement. Proc Natl Acad Sci U S A. 77:5764-8.

Pangburn M.K. and Müller-Eberhard H.J. (1980) Relation of putative thioester bond in C3 to activation of the alternative pathway and the binding of C3b to biological targets of complement. J Exp Med. 152:1102-14.

Daha MR, Stuffers-Heiman M, Kijlstra A and Van ES LA. (1979) Isolation and characterization of the third component of rat complement. Immunology 36:63-70.

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