

<b>Name:</b>	<b>C2-Dpl</b>
<b>Catalog Number:</b>	<b>A312</b>
<b>Sizes Available:</b>	1.0 mL/vial
<b>Concentration:</b>	>50 mg protein/mL (see Certificate of Analysis for actual conc.)
<b>Form:</b>	Frozen liquid
<b>Activity:</b>	>80% versus normal human serum standard when reconstituted
<b>Purity:</b>	No C2 detectable by immunodiffusion
<b>Buffer:</b>	10 mM sodium phosphate, 145 mM NaCl, pH 7.3
<b>Preservative:</b>	None, 0.22 µm filtered
<b>Storage:</b>	-70°C or below. Minimize freeze/thaw cycles.
<b>Source:</b>	Normal human serum (shown by certified tests to be negative for HBsAg and for antibodies to HCV, HIV-1 and HIV-II).
<b>Precautions:</b>	Use normal precautions for handling human blood products.
<b>Origin:</b>	Manufactured in the USA.

### General Description

Normal human serum depleted of complement C2 protein by immunoaffinity chromatography. The product is tested for the absence of C2 by functional assays for classical pathway activity and for C2 protein by double immunodiffusion. C2-Dpl is certified to possess a functional alternative pathway for complement activation (Morgan, B.P. (2000); Dodds, A.W. and Sim, R.B. (1997)). A functional classical pathway can be reconstituted by addition of purified C2 protein (25 µg/mL) indicating that all other complement components necessary for classical pathway activation are present. The absence of C2 also prevents complement activation by the lectin pathway but the alternative pathway will still activate on appropriate surfaces.

### Physical Characteristics

C2-Dpl is supplied as a clear, straw-colored liquid containing all proteins of normal human serum except complement component C2.

### Function

The depleted serum is tested for remaining classical pathway activity by hemolytic assays using antibody-sensitized sheep erythrocytes (CompTech #B200) and for alternative pathway function using rabbit erythrocytes (CompTech #B300). The depleted serum is reconstituted with 25 µg/mL C2 (CompTech #A112) and retested to verify that a functional classical pathway is restored. The Certificate of Analysis provided with each lot gives a description of the assays and specific titers for the depleted and reconstituted sera compared to normal human serum.

### Assays

The unit of classical pathway activity is the CH50. A similar unit, the C2H50, is used to quantitate the activity of C2 and C2-Dpl. A C2H50 unit is the amount of functional C2 needed to lyse 50% of  $3 \times 10^7$  EA cells (antibody-sensitized sheep erythrocytes (CompTech #B200)) when that amount of C2 (CompTech #A112) is incubated with the recommended volume of C2-Dpl in GVB<sup>++</sup> (CompTech #B100) in a total volume of 500 µL for 30 min at 37°C. This amount of C2 indicates the sensitivity of

the assay for C2 which is typically about 1 ng C2 with 5 µL C2-Dpl. See the Certificate of Analysis for lot specific values.

Alternative pathway titers are performed to document that this pathway of complement activation is fully functional in C2-Dpl. Lectin pathway activity is not tested, but it would be expected to be inactive due to the absence of C2.

### **Applications**

C2-Dpl is used to assay C2 hemolytic activity in samples and to supply an alternative pathway activating system that is incapable of activating the classical or lectin pathways of complement.

### **Precautions/Toxicity/Hazards**

The source is human serum, therefore precautions appropriate for handling any blood-derived product must be used even though the source was shown by certified tests to be negative for HBsAg and for antibodies to HCV, HIV-1 and HIV-II.

Hazard Code: B      WGK Germany 3

MSDS available upon request.

### **References**

Dodds, A.W. and Sim, R.B. editors (1997) Complement. A Practical Approach (ISBN 019963539) Oxford University Press, Oxford.

Morgan, B.P. ed. (2000) Complement Methods and Protocols. (ISBN 0-89603-654-5) Humana Press, Inc., Totowa, New Jersey.

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NOT FOR HUMAN OR DRUG USE.**

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