

<b>Name:</b>	<b>C1q/Factor D-Dpl</b>
<b>Catalog Number:</b>	<b>A399</b>
<b>Sizes Available:</b>	1.0 ml/vial
<b>Concentration:</b>	>50 mg/ml (see Certificate of Analysis for exact conc.)
<b>Form:</b>	Frozen liquid
<b>Activity:</b>	>70% versus normal human serum standard
<b>Purity:</b>	No C1q and factor D detectable by immunodiffusion
<b>Buffer:</b>	5 mM Sodium phosphate, 145 mM NaCl, pH 7.3
<b>Preseervative:</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

C1q/Factor D-depleted serum is normal human serum in which complement proteins C1q and factor D have been depleted by ion exchange and immunoaffinity chromatography. The product is tested for C1q and factor D proteins by double immunodiffusion and the absence of C1q and factor D is measured by testing classical and alternative pathway function. The product is certified to exhibit less than 5% classical and alternative pathway activity. After reconstitution with purified C1q protein (70 µg/mL), C1q/Factor D-depleted serum is certified to possess a functional classical pathway for complement activation (Morgan, B.P. (2000); Dodds, A.W. and Sim, R.B. (1997)). Similarly, a functional alternative pathway can be reconstituted by addition of purified factor D (1.4 µg/mL) indicating that all other complement components necessary for alternative pathway activation are present. Lectin pathway activity is not tested, but it would be expected to be active.

### Physical Characteristics & Structure

C1q/Factor D-Dpl is supplied as a clear, straw-colored liquid containing all proteins of normal human serum except complement components C1q and factor D.

### Function

C1q/FD-Dpl is tested for 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 70 µg/mL C1q (CompTech #A099) and 1.4 µg/mL factor D (CompTech #A136) and retested to verify that a functional classical pathway and alternative 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 and the unit of alternative pathway activity is the AP50. A CH50 unit is the amount of complement needed to lyse 50% of  $1 \times 10^8$  EA cells (antibody-sensitized sheep erythrocytes (CompTech #B200))

when that amount of serum is incubated with EA cells in GVB<sup>++</sup> (CompTech #B100) in a total volume of 1.5 mL for 60 min at 37°C. See the Certificate of Analysis for lot specific values. An AP50 unit is defined as the amount of complement yielding 50% lysis of  $1.5 \times 10^7$  rabbit erythrocytes (Er, CompTech #B300) when incubated for 30 min at 37°C in a total reaction volume of 100 µL of GVB<sup>o</sup> containing a final MgEGTA (CompTech #B106) concentration of 5 mM. Lectin pathway activity of C1q/Factor D-Dpl is not tested.

### **Applications**

C1q/Factor D-Dpl serum is used as a source with minimal ability to activate the classical pathway and alternative pathway. Although the lectin pathway activity is not tested, lectin pathway complexes of the lectins and MASPs can be activated leading to activation of C2 and C4.

### **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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