Poster
M1026

Comparison of a Laboratory Developed Test with a CE Marked Commercial Kit for the Analysis of Complement Function and Inhibition

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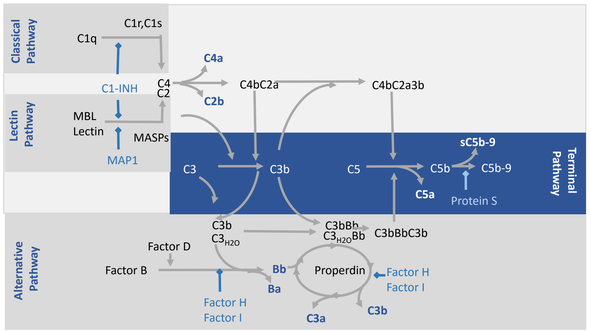
Purpose:

With the advent of complement targeting therapeutics, it has become increasingly important that robust complement function assays be available. Therefore we conducted a comparison of the two lead methods for measuring complement function: the historic hemolytic assays and the newer ELISA style deposition assays.

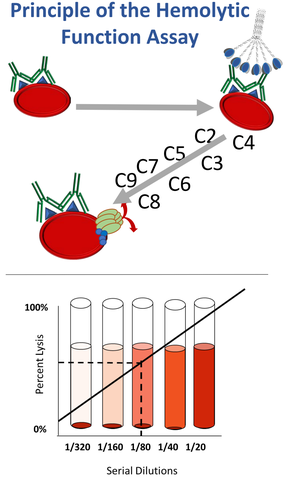
Background:

Complement is part of the innate immune system responsible for 1) killing some invading microbes; 2) directing the larger immune system to the site of infection; 3) tagging debris for clearance; 4) directing removal of host cellular debris and excess immune complexes. Because of these strong pro-inflammatory properties of complement, it is important that it is properly controlled.

Complement Pathway:

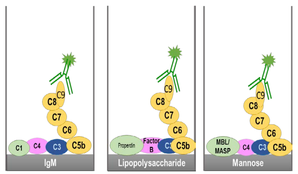


Principle of the Hemolytic Function Assay



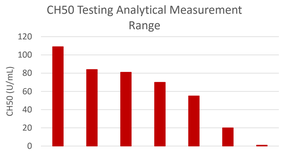
This assay is a laboratory developed test

Principle of the ELISA Style Function Assay

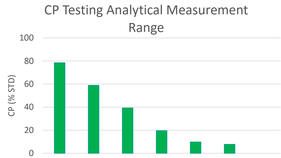


This assay is CE marked but RUO marked in the USA

Relative Sensitivity: Classical Pathway Test



CH50 (U/mL)
109
84
81
70
55
20
1



CP (% STD)
78.5
58.9
39.3
19.6
9.8
7.8
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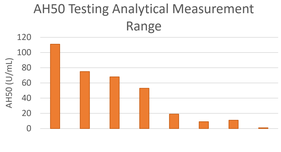
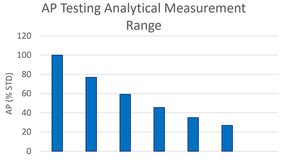
Relative Sensitivity: Alternative Pathway Test



AH50 (U/mL)
111
75
68
53
19
9
11
1



AP (% STD)
100
76.9
59.2
45.5
35
26.9
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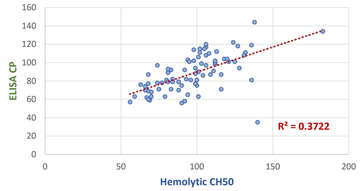
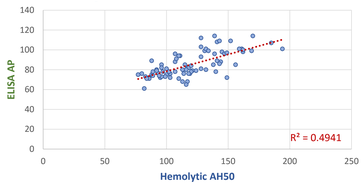
Classical Pathway: Repeat Measurements



Only 4% discordance on diagnostic category

80 Normals (40 males & 40 Females) were measured by both methods

Alternative Pathway: Repeat Measurements



Only 6% discordance on diagnostic category

80 Normals (40 males & 40 Females) were measured by both methods

Intra-Assay & Inter-Assay Precision

Assay Precision, %CV			
Pathway	Level	Intra-Assay	Inter-Assay
CH50 Hemolytic	High	7.8%	NT
	Med	1.6%	6.0%
	Low	8.1%	NT
CP ELISA	High	6.2%	10.1%
	Med	2.6%	15.1%
	Low	2.2%	15.6%

Assay Precision, %CV			
Pathway	Level	Intra-Assay	Inter-Assay
AH50 Hemolytic	High	4.6%	NT
	Med	3.7%	6.0%
	Low	5.9%	NT
AP ELISA	High	6.0%	11.4%
	Med	1.8%	6.2%
	Low	1.7%	8.3%

A minimum of 15 repeat measurements were made by a minimum of two technologists. For inter-assay, measurements were made over at least 2 months.

Conclusions:

These two methods for measuring complement function performed comparably for assay precision. The hemolytic versions did demonstrate lower limits of quantitation. There was a disturbing lack of correlation between the two methods at the raw data level. However, this resulted in only a 4% and 6% discordance in the diagnostic category.

References:

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