K944438

510(k) Summary

FEB 20 1997

1 ***Submitter's Name/Contact Person***

Joseph M. Califano, Regulatory Affairs Manager

Address

Hemagen Diagnostics, Inc.
34-40 Bear Hill Road
Waltham, MA. 02154

Phone: (617) 890-3766
Fax: (617) 890-3748
e-mail: jcalifano@hemagen.com

Date Prepared

9 September 1994

Date Amended

13 Feb 1997

2 ***Name of Device***

Anti-cytomegalovirus test kit

Proprietary name:

Hemagen CMV IgG Kit

3 ***Comparative Device***

Gull Laboratories, Inc. CMV IgG ELISA Test
Product No. CME 100 (96 determinations)

4 *Description of Device*

An enzyme-linked immunosorbent assay (ELISA) designed for the qualitative or semi- quantitative detection of circulating IgG antibodies to cytomegalovirus in human serum.

The ELISA methodology is commonly used for serum antibody evaluations. Purified antigens from *Cytomegalovirus* have been attached to the inner surfaces of the microwell plate. During the initial incubation step, antibodies in patient serum bind specifically to the immobilized antigen and remain in place after a wash step.

A second antibody, which is conjugated to the enzyme horseradish peroxidase, is used to recognize the gamma-chain region of the bound anti-cytomegalovirus antibodies. In the wells where the second antibody remains bound, the enzyme catalyzes a color change in the substrate, tetramethylbenzidine (TMB). After the reaction is stopped, the color is read in an EIA plate reader.

5 *Intended Use of Device*

This enzyme-linked immunosorbent assay (ELISA) is intended to determine an individual's serologic status with respect to IgG antibodies to CMV. When the assay is used in the qualitative mode, a reactive result may indicate current or past infection with CMV. When used in the semi-quantitative mode, this test can detect significant antibody rises associated with seroconversion, reinfection, or reactivation of latent disease. This product is not FDA-cleared for use in screening blood or plasma donors. The performance of this assay has not been established for neonates and pregnant women. Results from Immunocompromised patients should be interpreted with caution.

6.*(A) Technological Characteristics*

The Hemagen CMV IgG Kit and the Gull Laboratories CMV IgG ELISA Test are both based on similar technologies: they are both enzyme-linked immunosorbent assays. The proposed device and the predicate device utilize optical density as a measure of antibody presence, with an established cutoff point between a positive and a negative reaction.

The cutoff for the proposed device is based upon the comparative performance with the predicate device. The optimal cutoff value was selected utilizing receiver operating characteristic (ROC) methods. The cutoff is subject to an equivocal zone of +/- 10 %.

The low calibrator supplied with the proposed device is set at the cutoff activity level. The activity of patient specimens is determined by using the standard curve to find the corresponding activity level for the optical density measured.

The proposed device is supplied with three calibrators (low, medium, and high) that have been standardized to a characterized material. A standard curve is generated by plotting the optical densities obtained for each of the calibrators as a function of the calibrator activity level, AU/mL. The activity of patient specimens is determined directly from this standard curve.

6.(B) *Performance Data*

I. ***Comparison Testing***

The Hemagen kit and the comparative device were used to test serum specimens from normal blood donors. A total of 252 samples were evaluated. The comparison data for these specimens are summarized in Table 1:

Table 1: Summary of all specimens. N=252

		COMPARATIVE DEVICE			
		POS	NEG	IND	TOTAL
PROPOSED	POS	188	1	0	189
	NEG	0	63	0	63
	IND	0	0	0	0
	Total	188	64	0	252

The relative sensitivity was found to be 100 % (189/188). $_{0.95}$Confidence interval of 98.0 to 100 %
The relative specificity was found to be 98.4 % (63/64). $_{0.95}$Confidence interval of 91.7 to 99.7 %

II. ***Alternate Site Evaluations***

A panel consisting of nine "blind" serum samples was evaluated by three independent laboratories following a formal protocol:

i. Four CMV IgG negatives
ii. Five CMV IgG positives

The samples were evaluated in parallel with both the predicate and proposed devices in triplicate on three different days at each laboratory.

Agreement between sites

All 3 sites reported 100 % agreement for all 9 of the samples with both the proposed and predicate devices.

III. ***Interfering Substances***

Lipemic and hemolytic samples were evaluated with the **Hemagen CMV IgG Kit** following NCCLS Proposed Guideline, "Interference Testing in Clinical Chemistry" Document EP7-P ISSN 0273-3099. Samples with hemoglobin concentrations of ≤ 500 mg/dL and lipid concentrations of ≤ 3,000 mg/dL did not have any significant effect on the assay results.

IV. *Prozone and "Hook Effect"*

The **Hemagen CMV IgG Kit** was used to assay a high titered serum sample to determine if the kit would return unexpectedly low values. The results of this evaluation indicate that the kit gives appropriately high positive results with high titered sera.

V. *Cross reactivity*

Five samples which were positive for cytomegalovirus antibodies and five samples which were negative for cytomegalovirus antibodies with another commercially available assay were evaluated for the presence of other viral antibodies. IgG antibodies to rubella virus, varicella-zoster virus, and herpes simplex virus did not affect the specificity of the **Hemagen CMV IgG Kit.**

VI. *Evaluation of the CDC CMV/HSV serum panel*

The CMV/HSV Evaluation Panel consists of 100 {50 pairs} blind coded serum samples from clinically evaluated patients. All of the 100 samples were evaluated and characterized with the **Hemagen CMV IgG Kit** in accordance with the draft package insert. The results were submitted to CDC for scoring. The CDC reported that the **Hemagen CMV IgG Kit** had correctly characterized 66 of 66 samples as "CMV positives," and 34 of 34 samples as negatives.

VII. Semi-Quantitation Evaluations

A. Paired sera precision studies

Four fold dilutions of CMV reactive serum samples were utilized to evaluate within-run and between-run precision of the **Hemagen CMV IgG Kit**.

Intra-assay precision

Four positive samples were selected. A 1:4 dilution of each sample was prepared. Each neat and 1:4 Dilution was assayed 10 consecutive times for each of the five samples.

	Dilution	Mean AU/mL	Std. Dev	% CV[1]	AU Ratio
Sample 1	Neat	175	12.0	6.9	
Sample 1	1:4	45	4.6	10.4	3.90
Sample 2	Neat	147	5.6	3.8	
Sample 2	1:4	42	1.9	4.6	3.50
Sample 3	Neat	129	6.8	5.3	
Sample 3	1:4	30	0.9	2.9	4.30
Sample 4	Neat	118	5.2	4.4	
Sample 4	1:4	30	2.1	7.0	3.93

Inter-assay precision

Five positive samples were selected. A 1:4 dilution of each sample was prepared. Each neat and 1:4 dilution was assayed twice a day for 5 different days.

	Dilution	Mean AU/mL	Std. Dev	% CV[1]	AU Ratio
Sample 1	Neat	156	10.8	6.9	
Sample 1	1:4	39	4.1	10.6	4.00
Sample 2	Neat	133	15.4	11.6	
Sample 2	1:4	37	3.9	10.5	3.59
Sample 3	Neat	144	15.1	10.5	
Sample 3	1:4	38	5.0	13.0	3.79
Sample 4	Neat	122	6.7	5.5	
Sample 4	1:4	29	3.7	12.8	4.21
Sample 5	Neat	133	13.9	10.5	
Sample 5	1:4	33	4.3	13.1	4.03

1. These values include the well-to-well variation inherent in the plastic strips, which can range up to 5 %, according to the plastic strip supplier.

B. AU Ratio Establishment

A total of 43 high titered serum samples were selected. Multiple 2 and 4 fold dilutions were performed. Based on the results, it was determined that AU ratios of > 3.2 are indicative of a significant rise in antibody titer.

7 *Conclusions*

The results of the comparative studies and the fact that both the Hemagen kit and the predicate kit utilize an enzyme-linked immunosorbent assay method support the claim that the Hemagen kit is a safe and effective *in vitro* diagnostic test and is substantially equivalent to the comparative device.