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**SUBJECT** **bioelisa HAV IgM:  
Product evaluation**

Please find attached a product evaluation on bioelisa HAV IgM conducted in a reference hospital in Barcelona, Spain.

A panel of 28 positive and 40 negative samples characterized by the Johnson & Johnson *Vitros* anti-HAV IgM assay were used.

Only one discrepant sample appeared: *Vitros* positive and bioelisa HAV IgM negative. The sample was tested with the Abbott AxSYM HAVAB-M as a third technique. The Abbott AxSYM result on this sample was also negative.

In this evaluation, bioelisa HAV IgM showed 100% for both sensitivity and specificity.



**STUDY OF THE BIOKIT BIOELISA HAV IgM KIT IN  
COMPARISON TO THE JOHNSON & JOHNSON VITROS ANTI-  
HAV IgM TECHNIQUE USING THE VITROS  
IMMUNODIAGNOSTIC SYSTEM.**

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Barcelona, March 2004

## **STUDY OF THE BIOKIT BIOELISA HAV IgM KIT IN COMPARISON TO THE JOHNSON & JOHNSON VITROS ANTI-HAV IgM.**

### **GENERAL OBJECTIVE**

To analyse the specificity and sensitivity of the Biokit bioelisa HAV IgM kit in comparison to the Johnson & Johnson *Vitros* Anti-HAV IgM technique.

### **SPECIFIC OBJECTIVES**

To assess the degree of sensitivity of the Biokit bioelisa HAV IgM kit in comparison to the Johnson & Johnson *Vitros* Anti-HAV IgM technique in the detection of Anti-HAV IgM.

To analyse the specificity of the Biokit bioelisa HAV IgM kit for the detection of IgM antibodies in comparison to the *Vitros* Anti-HAV IgM technique.

To determine the positive predictive value (PPV) and the negative predictive value (NPV).

## BIOELISA HAV IgM. (Biokit)

The study was performed in the Microbiology Laboratory of the Santa Creu i Sant Pau Hospital in Barcelona.

### **MATERIAL**

#### **Samples:**

Determinations were made on a total of 68 samples. All the serum samples were preserved at  $-20^{\circ}\text{C}$  and distributed as follows:

- 28 sera positive for IgM specific antibodies to the Hepatitis A Virus as determined by the *Vitros* Immunodiagnostic System.
- 40 sera negative for IgM specific antibodies to the Hepatitis A Virus as determined by the *Vitros* Immunodiagnostic system.

### **METHOD**

#### **Commercial Technique**

The study was carried out using the commercial kit for the detection of anti-HAV IgM antibodies in human serum or plasma, BIOELISA HAV IgM CODE 3000-1098 from Biokit, identified as batch A-4204 and with expiration date March 7, 2005. The technique is an immunoenzymatic method based on the capture of the IgM antibodies present in the sample by anti-human IgM fixed to the solid phase. The addition of inactivated Hepatitis A Virus and an Anti-HAV marked with peroxidase will form a complex that is evidenced by the appearance of a blue colour after addition of the substrate.

The test and interpretation of the results were both carried out in accordance with the manufacturer's instructions.

Any discrepant results were retested in duplicate using the study techniques and a third technique, the Abbott AxSYM System (kit HAVAB-M<sup>TM</sup>). A result was considered as true when it coincided with two of the three test techniques.

## RESULTS

### Sensitivity

Of the 28 IgM sera positive by *Vitros*, 27 were also positive by BIOELISA HAV IgM. (Table 1). The discordant serum, positive for *Vitros* and negative for Biokit, was retested in duplicate with each of the techniques. The values obtained in the first determination were confirmed. This serum was finally considered as negative as it gave a negative result when using the third test technique (Abbott) (Table 2).

The discrepant serum belonged to a male patient of 76 years of age, admitted to the Intensive Care Ward for intestinal obstruction and with cardiomyopathy as the base pathology. He was submitted to a surgical operation in which there was a transfusion of blood components. The results of biochemical analysis always showed normal hepatic enzyme values. The patient died a few days after surgery from lung and heart complications and sepsis.

As the test result was negative using the third HAVAB-M<sup>TM</sup> technique, this means that the positive result for IgM by *Vitros* can be considered a false positive (on occasions post-transfusion sera may give non-specific results that later disappear). This lack of specificity was not detected by the Biokit technique and so the specificity of the technique is good. Unfortunately, it was not possible to make a new determination to confirm the negative result for Anti-HAV IgM. Therefore the sensitivity of the BIOELISA HAV IgM kit was calculated over 27 sera, giving a result of 100%.

## Specificity

All the 40 sera negative by *Vitros* Anti-HAV IgM were also negative by BIOELISA HAV IgM, corresponding to a specificity of 100%.

**Table 1.** Comparison of Biokit BIOELISA HAV IgM with the *Vitros* Anti-HAV IgM technique by Johnson & Johnson.

VITROS	BIOKIT		TOTAL
	NEGATIVE	POSITIVE	
NEGATIVE	40	0	40
POSITIVE	1	27	28
TOTAL	41	27	68

**Table 2.** Result of the discrepant serum.

Sample No.	<i>Vitros</i> Anti-HAV IgM (Pos >1.2)		BIOELISA HAV IgM. (Biokit) (Cut-off)		HAVAB-M™ (Abbott) (Pos >1.2)
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	
02-231700	4.8	4.7	0,152 (0,330)	0,198 (0,360)	Negative (0.09)

## Positive Predictive Value (PPV)

The positive predictive value of the Biokit BIOELISA HAV IgM technique was 100% compared to 96.4% for *Vitros* Anti-HAV IgM.

$$PPV = \frac{TP}{TP + FP} \times 100$$

**PPV:** Positive predictive value. **TP:** true positive. **FP:** false positive

### **Negative Predictive Value (NPV)**

The negative predictive value of the Biokit BIOELISA HAV IgM technique was 100%, the same as the value for the *Vitros* Anti-HAV IgM technique.

$$\text{NPV} = \frac{\text{TN}}{\text{TN} + \text{FN}} \times 100$$

**NPV:** Negative Predictive Value. **TN:** true negative. **FN:** false negative.

## CONCLUSIONS

The evaluation of the ELISA technique for the detection of IgM specific antibodies to the Hepatitis A Virus, **BIOELISA HAV IgM** by Biokit compared to the *Vitros* Anti-HAV IgM technique by Johnson & Johnson results in the following conclusions:

1. The **sensitivity and specificity** of the BIOELISA HAV IgM technique was 100%. This was also true for the **positive and negative predictive value**.
2. The technique is simple to use and only requires small volumes of serum or plasma (5-10  $\mu$ l). The only consideration is that if the reagents were coloured then, when performing the technique manually, this would simplify knowing whether they have been dispensed or not and so avoid possible errors.
3. Its handling characteristics as well as the sensitivity and specificity make it an excellent technique to confirm the diagnosis of acute infection by the Hepatitis A Virus.