CK-MB Mass Test in Ischemic Myocardial Injury. Comparison of Two Tests: Biomerieux Vidas and Sanofi Access Immunoassays

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The analytical and clinical performances of the new fluorescent immunoassay (CK-MB mass Vidas-BioMerieux) were examined and compared to the chemiluminescent test (CK-MB mass Access-Sanofi-Pasteur). Assay precisions of the CK-MB Vidas test within-assay or between-assay were less than 5.4 and 5.3%, respectively. Linearity was tested up to 214 µg/L. The CK-MB Vidas test was free of interference with CK-BB, CK-MM, and macro-CK. One hundred nineteen blood samples from patients with ischemic myocardial injury (IMI): acute myocardial infarction (AMI), suspected myocar-

dial contusion (SMC), and unstable angina pectoris (UA), were tested using both immunoassays. In AMI, a good correlation was found ($Y [\text{CK-MB Access}] = 1.1372 \times [\text{CK-MB Vidas}] - 6.3902$; $r^2 = 0.96$). In UA and SMC, low values were observed and both methods were well correlated ($Y [\text{CK-MB Access}] = 1.3662 \times [\text{CK-MB Vidas}] + 0.0671$; $r^2 = 0.97$). Clinical data were in good agreement with both immunoassays. ROC analysis performed in AMI demonstrated that the clinical performances of the two assays were similar. J. Clin. Lab. Anal. 14:43–47, 2000. © 2000 Wiley-Liss, Inc.

Key words: CK-MB mass; immunoassays; ischemic myocardial injury

INTRODUCTION

CK-MB is a biochemical marker currently used in emergency care unit. CK-MB enzymatic activity has been widely used to diagnose acute ischemic syndrome. This enzymatic assay is rapid, automated, and easy to use. Its major problem is the appearance of false positive results due to the presence of CK-BB, atypical forms of CK (macro-CK), and adenylate kinase activities (1). Electrophoresis was used as a reference method to separate the three CK isoenzymes (MM, MB, and BB), but this assay takes too long to be used in emergency situations. Recently, new and rapid immunoassays have been proposed to measure CK-MB mass concentration. These methods are known to be more sensitive and specific, using various monoclonal anti-CK-MB antibodies and none of the previous interferences (CK-BB, macro-CK, and adenylate kinase) has been reported (2).

The aims of the present study were: (1) to evaluate the analytical performances of the CK-MB mass test using the Vidas assay; (2) to compare CK-MB mass values with the Vidas procedure to those obtained with the Access method previously published (3); and (3) to evaluate the clinical performances of these two automated procedures.

MATERIALS AND METHODS

Patients

One hundred nineteen serum samples were analyzed; 42 patients (27 men and 15 women) ranging from 29 to 96 years old, were evaluated for acute myocardial infarction (AMI), suspected myocardial contusion (SMC), and unstable angina (UA). Patients with UA or SMC gave informed consent for extra blood samples. In AMI, an aliquot of blood samples from cardiac enzyme requests, was frozen. Thirty-six patients with AMI (84 serum values), 4 patients with SMC (21 serum values), and 2 patients with UA (14 serum values) were included in this study. Serial CK-MB samples were collected on the admission, and during the course of hospitalization.

A healthy control group of 50 patients without cardiac disorders was tested with the two assays. Results were used to perform ROC analysis.

The diagnosis of AMI was done according to World Health

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TABLE 1. Within-assay and between-assay precision of the CK-MB Vidas test

	Within assay (n = 12)			Between assay (n = 10)		
	Low pool	Medium pool	High pool	Low pool	Medium pool	High pool
Mean (µg/L)	6.3	54.9	168	7.45	66.9	134.7
SD	0.33	1.09	7.2	0.38	3.1	2.6
CV (%)	5.3	1.9	4.3	5.1	4.7	1.95

Organization criteria (4), without knowing CK-MB mass results. Diagnosis of SMC was based on echocardiographic or ECG abnormalities. Diagnosis of UA was based on Braunwald's classification (5).

Methods

Blood collection

Serum specimens were collected and centrifuged at 2,000g for 15 min and frozen at -20°C until CK-MB analysis was performed.

CK-MB mass tests

CK-MB mass assays were performed with two immunochemical methods using the MiniVidas instrument (BioMerieux, Marcy-L'Etoile, France) and the Access instrument (Sanofi Diagnostics Pasteur, Chaska, MN). The CK-MB Vidas test is an enzyme-linked fluorescent immunoassay (ELFA) carried out on the MiniVidas instrument. The upper limit of the reference interval of the CK-MB Vidas test determined by the manufacturer is less than 6.8 µg/L.

The CK-MB Access assay is a two-site immunoenzymatic "sandwich" assay. This method uses mouse monoclonal anti-human CK-MB antibody-alkaline phosphatase conjugate and paramagnetic particles coated with mouse monoclonal anti-human CK-BB antibody. A chemiluminescent substrate is added, and the light generated during the reaction is measured with a luminometer. The upper reference limit of the CK-MB Access chemiluminescent test is 4 µg/L.

Specificity

One sample and one control were overloaded with 10,000 µg/L of CK-BB and CK-MM. CK-MB mass was determined before and after overload with the Vidas test. Two serum samples with macro-CK were analyzed by both electrophoresis and immunoenzymofluorimetry (Vidas).

TABLE 2. Linearity of the CK-MB Vidas assay

Dilution	Expected value (µg/L)	Experimental value (µg/L)	Experimental value/expected value *100 (%)
Nondiluted		214	
2/3	142.6	136.2	95
1/2	107	102.4	95
1/3	71.3	67.5	94
1/4	53.5	53	99
1/8	26.7	27.9	104
1/6	13.3	14	105

Statistical analysis

ROC curves were performed for the two assays in AMI patients, and the areas under the curve (AUC) were calculated and compared, using the nonparametric Hanley test. ROC curves were done for 3 periods: 0–8 hours, 9–24 hours, and > 24 hours after the hospital admission. Percentiles 5, 10, 25, 50, 75, 90, and 95 were used for ROC curves thresholds.

RESULTS

Analytical Characteristics of the Two Immunoassays

Within- and between-assay variation

Three pooled serum samples (low, medium, high) were tested 12 times in within-assay variations. Results obtained are reported in Table 1. Coefficients of variation varied from 1.9 to 5.3%. For between-assay evaluation, another series of three pooled samples (low, medium, high) was tested: two series per day for 5 days (Table 1). Coefficients of variation varied from 1.95 to 5.1%.

Linearity

One elevated serum sample (214 µg/L) was diluted in a low level CK-MB serum sample (< 1 µg/L). Six dilutions were assayed and run in duplicate. The percentage of recovery varied from 94 to 105%. This assay was linear from 0 to 214 µg/L (Table 2).

Cross-reactivity

No cross-reactivity was observed with CK-BB and CK-MM up to 10,000 µg/L (Table 3). Two samples enriched with macro-CK1 (51% of the total CK) and macro-CK2 (42% of the total CK) did not interfere in the CK-MB Vidas test (data not shown).

TABLE 3. Cross-reaction test between CK-MB mass and CK-MM and CK-BB

	Serum 1	Serum 1 + CK-MM (10,000 µg/L)	Serum 1 + CK-BB (10,000 µg/L)	Control 2 (20 µg/L)	Control 2 (20 µg/L) + CK-MM (10,000 µg/L)	Control 2 (20 µg/L) + CK-BB (10,000 µg/L)
Vidas CK-MB mass value (µg/L)	0.98	0.53	0.93	18.92	19.37	18.83

Fig. 1. Linear regression analysis of CK-MB values (AMI patients) determined by the Vidas and the Access instruments. Eighty-four serum samples were tested with the MiniVidas and the Access instruments and CK-MB values were analyzed with the Cricket Graph 1.3.2 (Macintosh). AMI, acute myocardial infarction.

Correlation Between the Two Assays

One hundred nineteen patient samples were tested (range: 0.1–977 µg/L). Figure 1 represents the linear regression analysis line obtained in AMI patients ($n = 84$): $Y (\text{Access}) = 1.1372 \times (\text{Vidas}) - 6.3902$, $r^2 = 0.96$. Figure 2 illustrates the regression line observed in UA and SMC patients ($n = 35$).

Clinical Performances of the Assays

In AMI, AUC were not significantly different for both assays, at 0–8 hours (Table 4), 9–24 hours, and > 24 hours (data not shown) after the hospital admission. ROC curves between 0 and 8 hours are illustrated in Figures 3 and 4.

DISCUSSION

CK-MB mass values performed on automated instruments were compared in this study. Coefficients of variation of the Vidas assay were less than 5.3 %, a value similar to that published with the Access instrument (3). The linearity of the MiniVidas instrument was accurate up to 214 µg/L. As previously reported (2), we did not note any interference of macro-CK, CK-BB, and CK-MM. Comparison of the values between

Fig. 2. Linear regression analysis of CK-MB values (SMC and UA patients) determined by the Vidas and the Access instruments. Thirty-five serum samples were analysed as described in Figure 1. SMC, suspected myocardial contusion; UA, unstable angina.

CK-MB Vidas assay and CK-MB Access test has shown good concordance whatever the range of values (Figs. 1, 2).

Patients with IMI represent a heterogeneous population, composed of AMI, SMC, and UA patients. In the present study, high CK-MB mass concentrations were found in patients classified as having AMI. These concentrations were correlated with both methods ($r^2 = 0.96$). Using serial patterns of CK-MB mass variations in AMI, we noted a good correlation between the two methods during patients' time courses. Peak values were obtained at the same time with both immunoassays, and values returned simultaneously to the normal range. AUC calculated at 0–8 hours, 9–24 hours, and > 24 hours after admission were similar with both assays, demonstrating that the two methods may be accurately used for the diagnosis of AMI. Sensitivity and specificity of the two assays were quite similar. As previously reported (6–10), we confirmed that CK-MB mass assay was an accurate biological marker for the diagnosis of AMI.

Moderated CK-MB mass concentrations were observed in patients with SMC. Figure 2 indicated that values obtained in SMC were similar with both methods. SMC was clinically confirmed in only one patient. This patient had no elevation

TABLE 4. Compared diagnostic sensitivity and specificity of CK-MB assays in AMI patients¹

	Time after admission	Threshold value (mg/L)	Sensitivity (%)	Specificity (%)	AUC	AUC variance	P value
Vidas	0–8 hr	2.13	95.2	72	0.9581	0.00068	NS
		4.82	81	100			
Access	0–8 hr	2.4	90	72	0.9429	0.000107	NS
		5.2	81	100			

¹NS, Not significant; AUC, Area under curve.

Fig. 3. Vidas ROC curve: comparison of control group patients ($n = 50$) and AMI patients ($n = 21$), determination of threshold CK-MB Vidas values.

of CK-MB. The three other patients had no myocardial contusion but some CK-MB values were above the normal range. These findings are consistent with prior studies demonstrating the low specificity of the CK-MB test to detect myocardial damages in patients with concomitant skeletal muscle injury (11–13).

In patients with UA, we reported low CK-MB mass concentrations. All values were under the normal reference value, ranging from 0.8 to 2.6 $\mu\text{g/L}$. However, similar time courses were observed for the two immunoassays. As previously reported (14,15), our results confirmed the poor sensitivity of the CK-MB test to detect minor myocardial injury.

In conclusion, the analytical performances of the CK-MB Vidas test results were correlated with those obtained with the

CK-MB Access test. Clinical performances were also in good concordance with the two assays in ischemic myocardial injury.

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