Noninvasive Assessment of Reperfusion and Reocclusion After Thrombolysis in Acute Myocardial Infarction

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The clinical significance of ST-segment changes and of the time course of appearance in serum of different cardiac proteins has been reviewed for the diagnosis of coronary reperfusion and reocclusion after thrombolysis. In particular, the value of serial 12-lead electrocardiographic (ECG) studies, of Holter monitoring, and of continuous multilead computer-assisted ECG monitoring is compared. Regarding the serum proteins, the clinical significance of reperfusion indices described so far for serum creatine kinase (CK), its isoenzyme serum creatine kinase MB, the CK isoforms, and myoglobin is reviewed. Emphasis is placed on (1) the calculation method used for deriving the reperfusion indices; (2) the sensitivity and the specificity of the reperfusion indices; (3) the minimum turn-around time needed to produce the reperfusion indices (depending on the practicability of the analytical and calculation methods and their applicability in an emergency laboratory); (4) the ability of the indices to produce reliable estimates of reperfusion efficacy of the thrombolytic agents under study; and (5) the ability of the marker proteins to detect reinfarction as well as the suitability of the markers to detect real-time necrosis.

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Early reperfusion and sustained patency of the infarct-related coronary artery are important determinants of survival.[1] Thus, more aggressive therapy may be indicated if thrombolytic therapy fails to open the occluded vessel or if reocclusion of an initially reperfused coronary artery occurs. In contrast, administration of thrombolytic agents may be discontinued to minimize bleeding risk in patients with a rapidly reperfused artery. In this manner, therapy of acute myocardial infarction can be tailored to the status of the infarct-related vessel. In order to guide therapy, continuous monitoring of the vessel status is mandatory. Coronary angiography is the "gold standard" to assess patency and occlusion or reocclusion. However, angiography supplies only very momentary information on the status of the infarct-related vessel, and this invasive technique is not useful for continuous monitoring.

Noninvasive methods for monitoring reperfusion of the infarct-related vessel include clinical markers (such as resolution of chest pain), electrocardiographic (ECG) findings (such as the occurrence of accelerated idioventricular rhythm and normalization of the ST segment), and monitoring of specific cardiac proteins in plasma.[2–5] Resolution of chest pain is very subjective and may frequently be related to analgesic medication. Certain arrhythmias suggesting reperfusion are specific, but not sufficiently sensitive, actually to detect reperfusion. Thus, neither of these 2 methods can be used to predict reliably coronary vessel status. In this overview, use of continuous multilead ECG monitoring and of monitoring of myocardial proteins in serum is discussed.

ROLE OF THE ELECTROCARDIOGRAM FOR DETECTION OF REPERFUSION AND PATENCY

Following reperfusion, as documented by coronary angiography, resolution of ST-segment elevation occurs earlier, with an almost 5-fold faster time course than the changes associated with the

TABLE I Serial 12 Lead Electrocardiographic Recording to Predict Coronary Reperfusion After Thrombolysis Therapy*

Study	Criteria	No. ECGs/ Assessment Interval	Interval Thrombolysis Angiography	No. Pt.	Pt. with Criteria (%)	Sensitivity (%)	Specificity (%)	Positive/ Negative Predictive Value (%)	Comment on Study
Kircher et al[3]	? ↓ average ST elevation, 2 worst leads	2 ECGs/90 min	90 min	56	34	52	88	88/46	Correct study design but criteria not defined
Clemmensen et al[11]	≥20% ↓ Σ ST elevations	2 ECGs/180 min	180 min	53	62	88	80	88/80	Correct study design, best serial ECG study
Saran et al[12]	>25% ↓ ST elevation, worst lead	2 ECGs/180 min	60–90 min	45	84	97	43	79/86	Angiography before final ECG assessment
Hogg et al[13]	≥50% ↓ ST elevation, worst lead	2 ECGs/300 min	45–90 min	17	82	93	67	93/67	Angiography before final ECG assessment
Nicolau et al[14]	≥50% ↓ ST elevation, worst lead within 4 hr after thrombolysis	8 ECGs/48 hr	<72 hr	101	49	58	83	92/37	Not suitable for early patency assessment
Barbash et al[15]	≥50% ↓ Σ ST elevations	2 ECGs/60 min	72 hr	286	66	87	76	87/76	Not suitable for early patency assessment
Richardson et al[16]	≥2 mm ↓ ST elevation, worst lead or resolution within 30 min	9 ECGs/180 min	6 days	188	56	67	80	92/40	Not suitable for early patency assessment

*Sensitivity and specificity given for patency assessment. Positive predictive value: predictive value for patency. Negative predictive value: predictive value for occlusion. ECG = electrocardiogram; Pt. = patients; ? ↓ = unspecified decrease.

natural evolution of an acute myocardial infarction.[6–10] Thus, rapid resolution of ST-segment elevation is a marker of reperfusion. However, more recent studies, using serial 12-lead ECG, have questioned the reliability of ST-segment changes to predict reperfusion.[2,3] Only recently, when newer techniques, such as continuous multilead ECG monitoring, became available could distinct patterns of ST-segment behavior be recognized as predicting reperfusion and patency after thrombolytic therapy.

Serial 12-lead ECG recordings: Studies that evaluated the usefulness of ST-segment recovery from serial 12-lead ECGs as a marker of reperfusion are presented in Table I.[3,11–16] For proper comparison, angiographic assessment should be performed immediately after, and not before, the final serial ECG assessment. Further, assessment of patency is most important during the first few hours following thrombolytic therapy. Unfortunately, 3 studies appear invalid because of late angiography,[14–16] and in 2, angiography was planned before final ECG assessment.[12,13] Finally, another study[3] did not clearly define the criteria for ST recovery. Thus, the study by Clemmensen et al[11] appears to be the most valid. This study comprised 53 patients with an acute myocardial infarction treated with thrombolytic therapy. Two serial ECGs were recorded, one on admittance and the other approximately 180 minutes after the initiation of thrombolytic therapy. Directly following the second ECG, angiographic patency was assessed, using the classification of the Thrombolysis in Myocardial Infarction (TIMI) trial at first injection of contrast material.[17] Reduction of the summed ST-segment elevation by ≥20% within 180 minutes rendered sensitivity and specificity values of 88% and 80% for ECG versus angiographic patency assessment, respectively.

On average, looking at the results of the studies on early patency assessment using serial 12-lead ECGs, it appears that a rapid reduction of ST-segment elevation or depression by ≥20–50% of the highest previous ST value, occurring within 3 hours from start of thrombolytic therapy, is a reasonably accurate predictor of reperfusion, whereas lack of rapid ST-segment recovery suggests occlusion. Depending on the severity of ST elevation on the initial ECG, the use of either the single lead with maximal ST deviation ("worst lead") or the summed lead ST changes may be of importance. If a large ST deviation is present on the initial ECG, the ST reduction read at the single worst lead is preferable, although the reduction of the summed ST deviation may be more useful in cases of mild ST deviation.

The conflicting results of these studies reflect the limitations of serial 12-lead ECG recording for prediction of reperfusion and patency. Coronary reperfusion is a dynamic, rapidly changing process that is often accompanied by intermittent or sustained reocclusion and cyclic flow changes.[18,19] One third of episodes of recurrent ST elevation are silent and may remain unrecognized unless continu-

ous ECG monitoring is performed (Figure 1). These changes may remain unrecognized if serial ECG recording is used for monitoring. The time interval of the serial ECGs may be crucial for accurate detection of reperfusion and for interpretation of the coronary status. For example, if an undetected delayed ST (re)elevation peak precedes the moment of ST-segment elevation recovery, serial ECG recording may suggest an occluded artery if the serial measurement is taken just before the rapid decline. On the other hand, if a serial ECG is taken at the moment of a reelevation episode that followed the first period of ST recovery, initial patency following thrombolysis will be missed (Figure 2). Because treatment strategies should nowadays be based on the presence or absence of initial reperfusion, this time resolution problem may lead to erroneous decision making. Therefore, it is more convenient to use continuous ECG monitoring techniques to avoid less accurate assessments of reperfusion and patency.

Continuous ECG monitoring techniques: Several continuous ECG monitoring techniques have been evaluated for the assessment of reperfusion and patency. Holter ST-segment recording is the easiest technique to monitor the ST segment continuously. However, it has some major limitations. Firstly, only a restricted number of leads can be monitored, which may not always reflect the area with maximal ST deviation. Further, Holter ST monitoring allows retrospective analysis only. This makes immediate feedback impossible and thus the technique cannot be used for on-line ST-segment monitoring and tailoring of thrombolytic therapy. This may explain why only a few studies have evaluated this technique for early patency assessment. The only 2 representative studies are those from Hohnloser et al[4] and Krucoff et al,[20] mentioned in Table II. Both studies demonstrate that Holter ST-segment recording may be used for ischemia monitoring. However, apart from the possibility to study reperfusion and (re)occlusion patterns retrospectively, the technique offers no major advantages above serial ECG monitoring.

More recently, computer-assisted continuous multilead ECG monitoring techniques have become available for real-time noninvasive patency assessment. These techniques use continuous ECG sampling and averaging techniques, offering a continuous real-time accurate measurement of the QRS complex and ST segments.

Two different approaches for computer-assisted ECG analysis have been developed: continuously updated multilead ST monitoring, based on on-linc

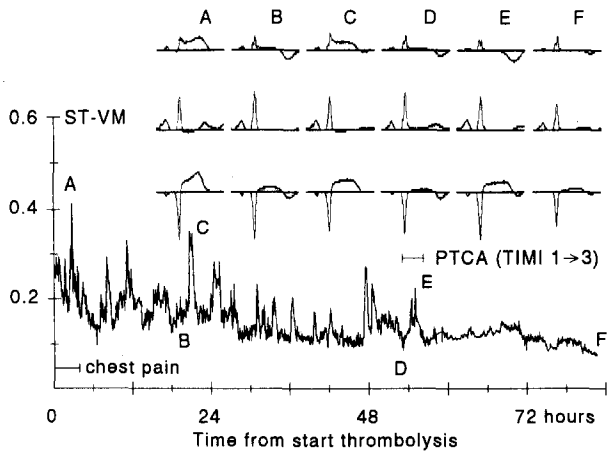

FIGURE 1. Continuous electrocardiographic (ECG) recording (vector ECG leads X, Y, Z) of 84 hours' duration in a patient with an anterior wall infarction. On admittance to the hospital, the patient was having chest pain, and severe ST-segment elevation was present in leads X and Z (A). Over the first 48 hours, frequent episodes of recurrent ST elevation and recovery were present without chest pain (B, C, D). Because of these recurrent silent ischemic episodes, angiography was performed 54 hours after admittance to the hospital. The anterior descending artery only showed minimal flow (Thrombolysis in Myocardial Infarction [TIMI] grade 1). After percutaneous transluminal coronary angioplasty (E), coronary flow was restored (TIMI grade 3) and ischemic episodes remained absent. VM = vector magnitude.

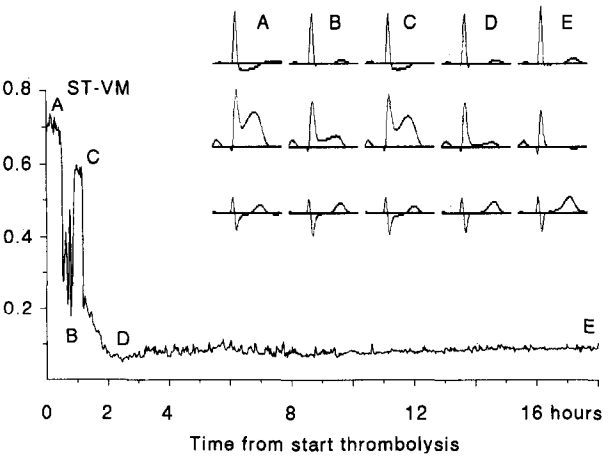

FIGURE 2. Continuous electrocardiographic (ECG) recording (vector ECG leads X, Y, Z) in a patient with an inferior wall infarction. Leads X, Y, Z more or less resemble V5, II, and V2. On admittance, considerable ST elevation is present in lead Y (A). The ST vector magnitude (ST-VM, sum of the ST deviation in X, Y, Z) decreases as a result of thrombolytic therapy (B), followed by recurrent ST elevation (C). Thereafter, the ST level stabilizes (D–E). Most probably, reperfusion (B) and reocclusion (C) occurred, followed by permanent reperfusion of the infarcted area (D–E).

analysis of the conventional 12-lead ECG, using either the single lead or the summed ST level (ELI, Mortara Instrument, Milwaukee, WI)[21] and continuous vectorcardiographic monitoring. The latter technique offers the possibility of both studying on-line vectorcardiographic QRS-complex and ST-

TABLE II Continuous Electrocardiographic Monitoring to Predict Coronary Reperfusion After Thrombolysis Therapy

Study	Technique	Criteria	Interval Thrombolysis to Angiography	No Pt.	Pt. with Criteria (%)	Sensitivity (%)	Specificity (%)	Positive/ Negative Predictive Value (%)	Comment on Study
Hohnloser et al[4]	Holter	≥50% ↓ ST elevation 1 of 2 leads within 90 min after thrombolysis	60–90 min	82	48	60	95	97/42	Only 2 leads, short assessment interval
Krucoff et al[20]	Holter	Achievement of ST steady state within 100 min after thrombolysis	100 min	36*	56	89	82	85/87	Less confined criteria, selected group of patients
Krucoff et al[19]	Continuous 12 lead	<50% ↓ or reelevation single lead or sum of ST at contrast medium injection	<6 hr	22	?	90†	92†	?	Large, not defined assessment interval, small study
Dellborg et al[22]	Vectorcardiography	QRSVD and STVM qualitative evaluation	At least 15 min	21	76	94	80	94/80	Pilot study, assessment interval not defined
		QRSVD increase ≥0.1 μV/min, plateau <2 hr		21	86	94	40	83/67	
Dellborg et al[24]	Vectorcardiography	QRSVD increase ≥0.1 μV/min, plateau <2 hr STVM decrease ≥0.83 μV/min qualitative evaluation	90 min	96	65	83	73	89/61	Correct study design, short assessment interval

*Totally occluded vessel at initial angiography.
†Sensitivity and specificity for assessment of occlusion.
QRSVD = QRS vector difference; STVM = ST vector magnitude. ? = unspecified. For other explanations and abbreviations, see Table I.

segment changes simultaneously (Mida1000 and Coronet, Ortivus Medical, Täby, Sweden).[22]

Using these continuous digital monitoring techniques, 5 distinct patterns of ST-segment behavior following thrombolysis have been observed[19,22]: (1) rapid ST recovery without reelevation; (2) rapid ST recovery following a delayed ST-elevation peak; (3) persistent ST elevation without a recovery pattern; (4) rapid ST recovery followed by recurrent ST elevation; and (5) a delayed ST-elevation peak followed by a rapid ST recovery and recurrent ST elevation. The first 3 patterns may point directly to the status of the infarct-related vessel. Rapid ST recovery without reelevation or a rapid ST recovery following a delayed ST-elevation peak both appear to be highly suggestive of reperfusion of the infarct-related artery, whereas persistent ST elevation without a recovery pattern suggests persistent occlusion, provided that significant ST elevation is present at the start of the monitoring period. Rapid ST recovery followed by recurrent ST elevation or a delayed ST-elevation peak followed by a rapid ST recovery and recurrent ST elevation may be less specific and may suggest unstable reperfusion. Patency assessment may then be difficult.

In Table II the 3 studies using continuous multilead and vectorcardiographic monitoring are listed. Krucoff et al[19] recently reported on either the presence of 1 episode of ≥50% ST recovery (worst single lead or the summed ST deviation of all 12 leads), its absence, or recurrent ST elevation, using continuous updated 12-lead computer-assisted ST monitoring. Absence of ST recovery or reelevation at the moment of angiographic assessment predicted occlusion of the infarct-related vessel with sensitivity of 90% and specificity of 92%. However, the time interval between hookup and final ECG and angiographic assessment was rather long, up to 6 hours, making assessment easier and only 22 patients were studied. Dellborg et al[22–24] reported on vectorcardiographic assessment of reperfusion and patency. All of their studies used both QRS-vector difference and ST-vector magnitude changes for prediction of patency. In a pilot study of 21 patients, sensitivity and specificity for patency at angiography were 94% and 80%, respectively, if a rapid change of both the QRS-vector difference and the ST-vector magnitude was observed, ending in a steady state. If only the QRS-vector difference changes were used, sensitivity remained 94%, but specificity dropped to 40%.[23]

In a recent study, preliminarily reported in an abstract form and recently submitted for publication, Dellborg et al[24] studied 96 patients using computer-assisted vectorcardiographic monitoring. More refined criteria were used and the angiographic assessment interval was fixed at 90 minutes as much as possible. Sensitivity and specificity for identification of patency were 81% and 70%, respectively.

How should the ECG be used to detect reperfusion in clinical practice? On the basis of the

overall findings in the literature, it is suggested that a baseline ECG should be recorded or the patient should be hooked up to a continuous ECG monitoring system immediately on admittance to the hospital. Obviously, hookup should not delay the start of therapy but should preferably be done before thrombolytic therapy has been initiated. The lead with maximal ST deviation should be used for ST monitoring. If less pronounced ST deviation is present, the sum of the ST deviations should be monitored. If only serial ECG recordings are used, short time intervals, <10 minutes, should be chosen for ECG recordings until ST recovery has occurred and has stabilized (Figures 1 and 2). If no ST recovery is observed within 3 hours after the start of thrombolytic therapy, this will be highly suggestive of persistent occlusion. If continuous on-line ECG recording techniques are used, the detection of ST-segment recovery will be easier and also reelevation episodes may be observed more accurately, offering the possibility of applying early change of treatment strategy, if necessary.

From the few studies on early patency assessment using ECG monitoring techniques, it may be deduced that rapid $\geq 50\%$ ST recovery may prove to be the most valid criterion. However, ST-vector magnitude and QRS-vector difference changes may comprise complementary information on the coronary vessel status that are not yet studied in detail. The upcoming results of the Global Utilization of Streptokinase and t-PA for Occluded Arteries (GUSTO) trial, in which an ST monitoring substudy using both Holter, multilead, and vectorcardiographic ECG monitoring is included, will provide more information on which criteria and time intervals should be applied in order to predict more accurately reperfusion, patency, and (re)occlusion.

ROLE OF CARDIAC SERUM PROTEINS

It is generally accepted[25–27] that, in case of successful reperfusion, cardiac enzymes appear earlier in the peripheral circulation, compared with unsuccessful reperfusion. Most studies on serum markers of reperfusion have been performed using serial measurements of creatine kinase (CK), its isoenzyme CK-MB, and myoglobin (Figures 3–5).

Creatine kinase and creatine kinase MB: Cytoplasmic CK is a dimer (M_r 86,000–89,000) consisting of 2 subunits: an M subunit (M_r 43,000) and a B subunit (M_r 44,500), each encoded by a separate gene.[28] The subunits combine to give 3 isoenzymes: CK-MM, CK-MB, and CK-BB.[29] In

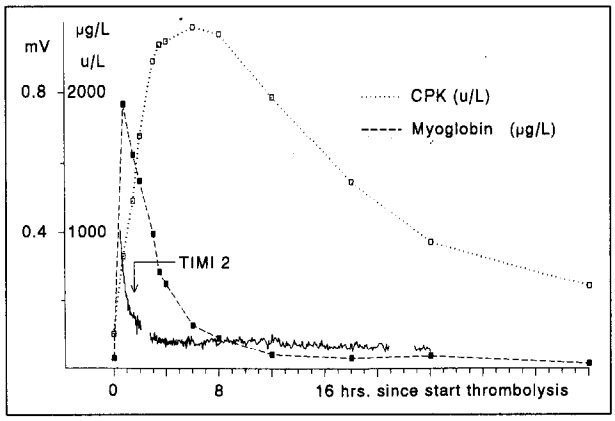

FIGURE 3. Combined monitoring of ST-segment changes (continuous line) and serial assessment of myoglobin and creatine kinase (CPK), in case of successful recanalization. The drop of ST segment elevation is rapid, and the slope serum rise of myoglobin is more rapid than that of creatine kinase.

patients with an acute myocardial infarction, blood CK and CK-MB levels become abnormal 6–8 hours after infarction, peak in 12–24 hours, and become normal approximately 48 hours after infarction.[30]

CK and/or CK-MB determinations are usually available on a stat basis in emergency laboratories. Most laboratories make use of an enzymatic method for total CK. The CK assay is a European Standardized Method that can easily be adapted on automated clinical chemistry analyzers. Results are available in 15–20 minutes. CK-MB activity is frequently measured after polyclonal immunoinhibition of the M subunit. This method is also easily applicable on routine clinical chemistry analyzers and results are produced in 15–20 minutes. False high CK-MB activities are produced in case of hemolysis because of adenylate kinase interference and in case of presence of macro-CK. Consequently, immunologic CK-MB mass assays (e.g.,

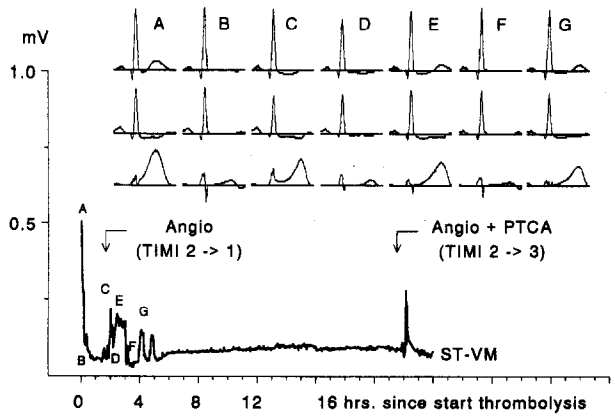

FIGURE 4. Monitoring of ST vector magnitude (VM) changes in a case of a patient with several recurrent reocclusions, with angiographic (Angio) documentation. PTCA = percutaneous transluminal coronary angioplasty; TIMI = Thrombolysis in Myocardial Infarction.

CK-MB on IMx, Abbott; CK-MB on Stratus, Baxter) gain interest nowadays because these methods are specific and do not suffer from macro-CK nor from adenylate kinase interference. Results can be produced in 20–45 minutes, depending on the type of immunoassay.

Lewis et al[31] described a rapid initial absolute increase in plasma CK and CK-MB activities (units per liter per hour) following reperfusion. The authors calculated a mean relative first-hour increase of plasma CK activity of 34 ± 18% (mean ± SD) of the peak increase (range, 13–67%) and a relative first-hour increase in CK-MB activity of 27 ± 13% of the peak increase (range, 13–57%) in patients that evolved from total occlusion to TIMI flow grade 3 of the infarct-related artery. When reperfusion was incomplete or not achieved, the relative increase for CK and CK-MB was <6% of the peak rise in the last hour of the 2.5-hour sampling period. The authors claim that 0.5 hourly determinations of CK and CK-MB during 2.5 hours following start of thrombolysis provide a useful tool for recognition of reperfusion. Shortcomings of these indices are that (1) absolute rates of increase of CK and CK-MB activities are dependent on the assay temperatures used, and (2) measuring the relative rates of increase requires knowledge of the peak increase, which becomes available only several hours later. Also, (3) the rapid and augmented release of CK with reperfusion has been associated with large, hemorrhagic infarcts in some dog models.[32]

Gore et al[33] described early peak CK as a reperfusion index in patients with acute myocardial infarction. In the nonreperfused group (TIMI flow grade 0 or 1) the mean time-to-peak (TTP) from onset of symptoms was 20.1 hours. In patients who achieved reperfusion (TIMI 2 or 3) the mean TTP was 14.3 hours. In the nonreperfused group, the mean TTP from onset of pain was 16.1 hours and this did not differ significantly from the TTP of the reperfused group. Peaking of plasma CK within 4 hours after starting thrombolysis is highly suggestive of reperfusion. Late peaking, >16 hours after starting thrombolysis, rarely indicated reperfusion. A major drawback of this reperfusion index is the considerable overlap of the TTP ranges in the reperfused and occluded patient groups between 4 and 16 hours. Another limitation of TTP CK as a reperfusion index is the overestimation of drug-induced reperfusion by including patients who had a subtotal occlusion at the time of treatment.

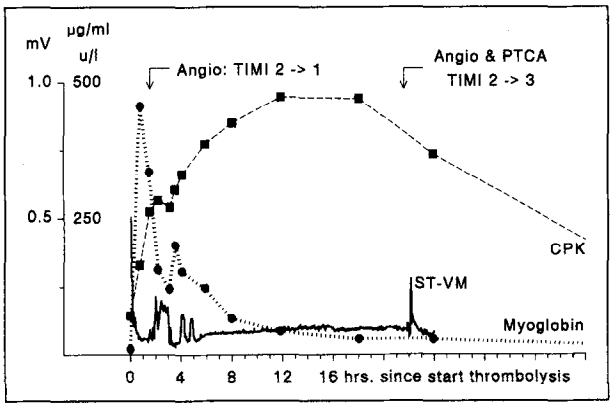

FIGURE 5. Serial assessment of serum creatine kinase (CPK) and myoglobin in the same case reported in Figure 2. The early rise of myoglobin is more rapid than that of creatine kinase. A second small rise of myoglobin paralleled a reelevation of the ST segment, secondary to a reocclusion of the infarct-related vessel. Angio = angiography; PTCA = percutaneous transluminal coronary angioplasty; TIMI = Thrombolysis in Myocardial Infarction; VM = vector magnitude.

Garabedian et al[34] documented that a >2.5-fold increase in CK-MB levels at the end of a 90-minute recombinant tissue-type plasminogen activator (rt-PA) infusion provided evidence for reperfusion of the left anterior descending coronary artery (sensitivity of 93% and specificity of 83%) and that a >2.2-fold increase in CK-MB levels could identify reperfusion of the right coronary artery (sensitivity of 89% and specificity of 100%). Slightly different cutoff points were proposed because of the greater prevalence of well-developed collateral circulation in the right coronary artery circulation bed. Failure of this CK-MB increase ratio to detect reperfusion may result from initial presence of subtotal occlusion, well-developed collateral circulation, or cyclic reperfusion and reocclusion.

Creatine kinase isoforms: Zonal electrophoresis is widely used for separation of CK-MM, CK-MB, and CK-BB in the routine clinical chemistry laboratory.[29] Prolonging the CK isoenzyme electrophoresis time, increasing the voltage, or using isoelectric focusing revealed at least 3 isoforms of CK-MM (denoted $CK\text{-}MM_{1\text{-}3}$ or $CK\text{-}3_{1\text{-}3}$) and at least 2 isoforms of CK-MB (denoted $CK\text{-}MB_{1\text{-}2}$ or $CK\text{-}2_{1\text{-}2}$).[35–40] The CK isoforms are no true isoenzymes but result from post-translational modification in serum of the CK-M subunit by a CK conversion factor, i.e., carboxypeptidase N.[41] In case of $CK\text{-}MM_3$, the tissue isoform, hydrolytic cleavage of the carboxy-terminal lysine gives rise sequentially to $CK\text{-}MM_2$ and $CK\text{-}MM_1$ (most anodal fraction). $CK\text{-}MB_2$ is converted to $CK\text{-}MB_1$. The discovery of probably 3 isoforms for CK-MB is recent and yet no commercialized methods exist that recognize these 3 CK-MB isoforms.[42,43]

The interpretation of electrophoresis data of serum CK following acute myocardial infarction has been complicated because of contradictions in

nomenclature. The International Union of Pure and Applied Chemistry–International Union of Biochemistry (IUPAC–IUB) Commission on Nomenclature prescribes that the most anodal isoenzyme should be identified with the lowest arabic numerical, i.e., CK-BB = CK-1, CK-MB = CK-2, and CK-MM = CK-3. In practice, letter subscripts have been replaced by numbers: CK-MB = CK-2 and the anodal conversion is represented by $CK\text{-}2_2 \rightarrow CK\text{-}2_1$.[29,44]

The reperfusion results after induced acute myocardial infarction in dogs suggested that prompt detection of reperfusion is possibly based on analysis of sequential changes in the plasma CK-MM isoform activities.[45,46] CK isoforms are not usually routinely determined in most clinical laboratories.[25] So far, different technologies have been applied to separate and quantitate the CK isoforms.[25,47–57] Analytical performance of these methods is poorly documented,[48] yet, electrophoresis is reported to be the most cost-effective, practical, reproducible, and sensitive method. However, differences in method specificity have also been reported, isoelectric focusing and high-voltage electrophoresis techniques being denaturating and producing artifacts (extra bands).[48]

Although several of these methods allow for 45-minute turn-around times, none is suitable for stat analysis because they are too laborious and specialized. Future potential for the clinical application of CK-MM and CK-MB isoforms depends on the development of simple, rapid, and automated systems. Recently, a sufficiently automated high-voltage electrophoresis analyzer (REP, Helena Laboratories, Beaumont, Texas)[58] that fulfills these requirements has been introduced. Also, monoclonal antibodies specific to certain CK isoforms became available.[59–61] The future development of rapid, automated immunoassays that quantitate specific isoforms may provide the basis for a new generation of immunoassay tests that distinguish tissue isoforms from serum isoforms.

CK-MM ISOFORMS: Seacord et al[62] found a marked increase of the percent $CK\text{-}MM_3$ activity over the first hour postdosing in all patients with successful reperfusion. Apple et al[40] found that the $CK\text{-}MM_3/CK\text{-}MM_1$ ratio peaked significantly earlier than total CK and CK-MB in both reperfused and occluded individuals. Morelli et al[63] confirmed these findings and documented that the TTP of the $CK\text{-}MM_3/CK\text{-}MM_1$ ratio in the first 3 hours postdosing differentiated reperfused from nonreperfused individuals ($p < 0.01$). On the other hand, the serum appearance constant of $CK\text{-}MM_3$ also differentiated successfully versus unsuccessfully reperfused patients with acute myocardial infarction.[63,64] Puleo et al[65] studied the rate of decline of plasma $CK\text{-}MM_3$ levels over 18 hours after onset of infarction. They found a sensitivity of 87% and a specificity of 74% when a minimum rate of decline of 3.1% hr^{-1} was exceeded.

The group of Abendschein et al[66] reported that conjoint analysis of percent $CK\text{-}MM_3$ and myoglobin increases over the first hour following initiation of thrombolysis, when these increases exceeded 0.18% min^{-1} and 2.6 ng/mL min^{-1}, respectively, provide robust noninvasive criteria for reperfusion evaluation.

In summary, (1) the determination of the rate of decrease of percent $CK\text{-}MM_3$ is less suitable for reperfusion evaluation because of the requisite time delay (preventing additional interventions to salvage myocardium at a time when this can be initiated effectively), and because it is influenced by the residual stenosis grade and the amount of collateral blood flow. (2) Determination of the TTP of the percent $CK\text{-}MM_3$ and the appearance constant (K_a) of $CK\text{-}MM_3$ are impractical as reperfusion indices because of the requisite time delay and repeated blood sampling necessary to establish the peak or to calculate K_a. (3) The rate of increase of the percent $CK\text{-}MM_3$ or $CK\text{-}MM_3/CK\text{-}MM_1$ ratio is superior for reperfusion evaluation. The rate of increase of percent $CK\text{-}MM_3$ is the earliest available reperfusion index because only 2–3 plasma specimens, sampled before and within 1 hour after starting thrombolysis, are required.

CK-MB ISOFORMS: CK-MM and its isoforms lack cardiac specificity; in contrast, CK-MB isoforms are more cardiac specific.[67,68] Until recently, available MB isoform assays lacked the necessary analytical sensitivity to detect CK-MB isoforms in normal plasma,[47,48] yet sensitive assays with good analytical performance are available.[49,69] Christenson et al[68] found that the $CK\text{-}MB_2/CK\text{-}MB_1$ ratio peaked 90 minutes after starting thrombolysis, compared with the 2-hour time to peak for the $CK\text{-}MM_3/CK\text{-}MM_1$ ratio in the study of Morelli et al.[63] Puleo et al[70] concluded that the $CK\text{-}MB_2/CK\text{-}MB_1$ ratio provided the best discrimination between reperfused and nonreperfused individuals within 1 hour after onset of pain. The limited body of clinical data on the utility of CK-MB isoform indices for reperfusion evaluation needs to be extended in order to understand their future potential. Especially, the existence of perhaps 3 CK-MB isoforms must be clarified.[42,43]

Myoglobin: Myoglobin is a cytosolic hemoprotein in cardiac and skeletal muscles (M_r 17,700) necessary for the last step in the oxygen transport

to the mitochondria.[71,72] Normal serum levels of myoglobin are reported to be in the range of 6–85 ng/mL and 4–60 ng/mL for men and women, respectively.[30] After acute myocardial infarction, the serum concentration of myoglobin becomes abnormal in about 2 hours, peaks in about 6–9 hours, and becomes normal in 24–36 hours. Myoglobin increases up to 10 times the baseline level. Renal impairment, vigorous exercise, and many other conditions can influence the serum myoglobin levels.[30] Myoglobinemia has also been described to be useful for evaluation of skeletal muscle damage.[73,74] The advantage offered by myoglobin as a marker for myocardial injury is that it appears earlier in the peripheral blood circulation compared with CK, CK-MB, and the CK isoforms.[75–80].

Myoglobin kinetics after coronary artery ligation and subsequent reperfusion in dogs show a rapidly evolving time pattern, with peak concentrations at 45 minutes after vessel reopening.[81] Despite its interesting kinetics, the myoglobin measurement has not been used extensively in the past. Reasons were the lack of reliable, rapid, and automated myoglobin assays,[47] as well as the poor specificity of the protein (60–95%), with large amounts being present in skeletal muscle. In the past a complement fixation test, numerous radioimmunoassays, and a semiquantitative latex test have been developed. Recently, however, stat immunoassays have been introduced on the European market.[82,83] Behring Diagnostics (Behringwerke, Marburg, Germany) commercialized 2 myoglobin immunoassays: (1) an automated nephelometric fixed-time method with NA-latex myoglobin reagent on a BN100 or BNA nephelometer (Behring Diagnostica) that produces results within 20 minutes (12 minutes analysis time); and (2) a manual turbidimetric immunoassay on the precalibrated Behring Turbitimer with Turbiquant myoglobin reagent that produces results within 180 seconds. The latter reagent can also be applied on routine clinical chemistry analyzers that are capable of measuring optical turbidity at 340 nm. Also, a 1-step 2-site particle concentration fluorescence immunoassay using monoclonal antibodies has been described.[84]

Kinetics of myoglobin appearance depend on infarct reperfusion and TIMI flow grade scores. Differences in kinetics between reperfused and nonreperfused arteries in patients with acute myocardial infarction will be reflected by different TTP values, different peak levels, and different rates of increase.

Ellis et al[80] found a mean TTP of 111 minutes and 360 minutes in the reperfused and nonreperfused groups, respectively. They described a mean T_{25-100} (i.e. the time required for myoglobin to increase from 25% to 100% of the peak level) of 71 and 341 minutes in the reperfused and nonreperfused groups, respectively. A >4.6-fold increase the first 2 hours following initiation of thrombolysis correctly identified 85% of the reperfused patients and 100% of the nonreperfused patients.

Katus and coworkers[85] determined the predictive power of the TTP values of myoglobin, CK, and CK-MB in their recanalization study. In case of recanalization <3.5 hours after onset of chest pain, the probability of correct classification varied between 90–100%. In case of late reperfusion, the probability of correct classification ranged between 5–99%. Myoglobin allowed the earliest and best discrimination between reperfused and nonreperfused individuals. These findings fit with the data of McCullough et al,[86] who also concluded that TTP analysis of myoglobin is not useful as a reperfusion index because of the large overlap of TTP values for reperfused and nonreperfused patients.

Clemmensen et al[87] also evaluated TTP of myoglobin as a reperfusion index and concluded that peak myoglobin levels reached within 4 hours after starting thrombolysis accurately predict coronary reperfusion with 91% sensitivity and 88% specificity, and this within the time limit in which rescue angioplasty is still effective.

Ishii et al[88] calculated a myoglobin and CK-MB ratio (defined as the ratio of the level at 15, 30, or 60 minutes after reperfusion to the level before reperfusion). The predictive accuracy of a myoglobin ratio >2.4 at 15 minutes was 96%; for CK-MB the predictive accuracy of a CK-MB ratio >2 at 15 minutes was only 68%. Consequently, the myoglobin ratio at 15 minutes was found to be a reliable marker of reperfusion and more useful than CK-MB for detection of reperfusion within 30 minutes.

Dillon et al[89] calculated and estimated myoglobin appearance rates (estimated K_a = rate of myoglobin increase over the first hour to myoglobin at the time of application of therapy). A significantly higher mean K_a was found in the reperfused group ($0.122 \pm 0.182\ min^{-1}$ [mean ± SD]) versus the nonreperfused group ($0.005 \pm 0.003\ min^{-1}$).

CONCLUSION

Continuous multilead ST monitoring techniques are of clinical use for prediction of vessel status early after myocardial infarction. In addi-

tion, with the introduction of myoglobin as a biochemical parameter, a real-time relation with electrical events during acute myocardial infarction and subsequent reperfusion can be observed. Consequently, myoglobin seems to be the monitoring biochemical parameter of choice for the evaluation of coronary reperfusion within the time limits during which additional therapeutical interventions are feasible and effective.

Future studies are needed to prove if the combined evaluation of clinical, electrocardiographic, and enzymatic monitoring will provide the optimal information for assessing coronary patency after thrombolysis.

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