CHAPTER 11

Cardiac markers: state of the art and future perspectives.

Current state of the art

Conventionally, the cardiac markers play an important role in the (sub)acute detection of myocardial damage. For diagnostic purposes, they are commonly used, especially, in patients with acute ischemic syndromes. However, the 'gold standard' CKMB-isoenzyme, whether measured as activity or mass, has appeared to be non-heart specific. Subsequently, an acute myocardial infarction (AMI) may be "under-" or "over-" diagnosed.

By the end 1980's the highly cardiac specific troponin T has become available as a biochemical utility to detect myocardial tissue damage. However, it took several years before cardiac troponin T was accepted as a specific cardiac marker. First, because it was a new parameter, whereas CKMB had been widely accepted and used already for several decades ('unknowned means unbeloved'). Second, the first generation cardiac troponin T reagent was not completely heart specific, as the label-antibody used in the ELISA methodology showed 20% cross reactivity with skeletal muscle tissue. Several years after the introduction of cardiac troponin T, cardiac troponin I was introduced as a possibility for the detection of cardiac damage, because the use of cardiac troponin T was patented.

The interpretation of cardiac troponin I levels from different reagent manufacturers is complicated, because the various manufacturers of this marker use both different antibodies as well as different calibration standards. The standards vary in their compositions regarding the various troponin I-C-T related complexes. Furthermore, uniformity of troponin I standards is complicated, because, after cell necrosis, troponin I exists of several forms in blood circulation (most of them are the complexes troponin I-C and troponin I-C-T). Moreover, the troponin I molecule is not stable in circulation as it is susceptible to proteolysis, resulting in degradation of the molecule. To overcome the standardisation problem, an AACC-subcommittee was installed to make recommendations for uniformity of the cardiac troponin I analysis.

During the first part of the last decade attention was focussed on the performance of cardiac troponin T and cardiac troponin I as marker for the detection of AMI. Although both markers have a relatively low molecular weight, and, subsequently, may fast released from damaged cells, it turned out that both cardiac troponin T and cardiac troponin I were not the best 'early' biochemical markers for the detection of AMI. The reason is that both troponins are myofibril bound, and, therefore, it takes more time than for the cytosol linked parameters (such as myoglobin) to be elevated in circulation after myocardial cell necrosis. As is reported in chapters 4 and 5 the CKMB2/CKMB1 isoform ratio is more suitable as cardiac marker for early detection of AMI. However, the analytical procedure is more complex than it is for troponin analysis.

Besides the assessment as marker for the detection of myocardial cell damage, also studies were carried out to investigate the performance of cardiac troponin T and cardiac troponin I as markers for prognostic purposes in patients with ischemic syndromes including (un)stable angina pectoris ('risk stratification'). As the first reported results look very promising more attention was focussed on this subject. Moreover, more or less at the same time, the cardiac troponin I and cardiac troponin T methodologies were improved. The troponin T methodology was improved, because, first, the label-antibody was further developed towards a 100% heart-specificity, and, second, calibration standards of human instead of bovine origin were introduced. Furthermore, the sensitivities of the cardiac troponin T and cardiac troponin I assays, especially in the low detection region, were enhanced. Consequently, this resulted in both a better accuracy and precision, which is, concerning the low concentration area, the most important for risk stratification.

The goal of measuring cardiac troponin T and cardiac troponin I has been extended from a particular marker for AMI to a marker of prognostic value for patients with acute coronary syndromes presenting with acute chest pain. Typically for this category of patients is the introduction of the so-called 'Observation Unit Department'. The purpose of this department is to improve and shorten the Triage-process of patients who are presenting with chest pain complaints at the Emergency Department, but for whom, at presentation to the hospital, a reliable diagnosis can not be made according to established criteria. Conventionally these patients were directly admitted to the Coronary Care Unit (CCU), where they were observed for at least 24 hours. About 75% of these patients did not experienced AMI and, therefore, they might unnecessarily be admitted to the CCU. This resulted not only in an unnecessarily emotional experience for the patient, but also in an increase in costs. With the introduction of the Observation Unit, it becomes possible to postpone the decision about admission to the CCU for maximally several hours until the final diagnosis has been established. In daily medical practice this includes, that, after arrival at the hospital, the patient will stay in the Observation Unit. Here, the ECG of the patient is continuously registered and during this several hours period also the troponin level is measured. At least 8 hours after the onset of anginal complaints an AMI is ruled out when the ECG has not been changed and the troponin concentration is still below the upper limit of the reference range or has not changed significantly. Subsequently, the patient does not have to be admitted to the CCU and may be discharged. This procedure will yield to a more efficient use of the CCU. In this respect it is important that the turn around times (TATs) of cardiac marker results are within one hour after blood collection. If the central laboratory cannot fulfil this requirement, point-of-care (POC) testing might be implemented according to recommendations of the National Academy for Clinical Biochemistry (NACB). If POC-testing is implemented it should be equipped to report cardiac marker results quantitatively. This is a necessity, as, most of the time, serial measurements will be performed, so that trend analysis may be used for interpretation purposes. As is reported in chapter 6, the Stratus CS and Triage Cardiac Panel are such POC-testing devices. After completing the study described in chapter 6, the Stratus CS was installed at the CCU for daily clinical practice. It turned out that non-analytical educated personnel can reliably perform the cardiac marker tests. Moreover, the troponin I results were more useful for classifying patients with acute chest pain than the "conventional" CKMB results.

Besides the application of measuring cardiac troponin in patients with myocardial ischemia, it appeared that both cardiac troponin I and cardiac troponin T were more sensitive and more specific markers than CKMB for the detection of myocardial damage in patients with accompanying skeletal muscle damage. This category of patients includes patients after (blunt) trauma, after surgery, and after other invasive procedures causing myocardial tissue damage.

In chapter 7 the results are reported of a study concerning patients experiencing blunt trauma. From this study it was concluded that the cardiac troponin I and cardiac troponin T are more reliable for the detection of myocardial damage than the conventional CKMB parameter. In particular, the conventional CKMB was more frequently elevated than the cardiac troponins were. This may be explained, because, on the one hand, CKMB is also part of skeletal muscle, whereas cardiac troponin I and cardiac troponin T are unique for myocardial muscle tissue. On the other hand, both new markers are more sensitive for minor myocardial damage, because per gram heart tissue the content of troponin is higher than that of CKMB. Moreover, cardiac troponin is, in contrast to CKMB, in healthy human beings hardly detectable in circulation. This means that minimal detectable concentrations of cardiac troponins in circulation are suspicious for myocardial tissue damage.

Therefore, it can be concluded that the role of CKMB as "gold standard" for myocardial damage has to be replaced by cardiac troponin I or cardiac troponin T. In this respect, the role of CKMB may have been become more or less obsolete and, consequently, this parameter should be removed from the request forms for blood investigation in routine (biochemical) patient care.

Future perspectives

Patients with an increased risk on cardiac events may be identified by (slightly) elevated concentrations of cardiac troponin I or cardiac troponin T. Therefore, efforts should be taken to prevent that they experience an irreversible ischemic event. This strategy may be executed either by treatment with drugs or by making use of invasive techniques.

A promising marker for reversible myocardial ischemia may be glycogen phosphorylase BB (GP-BB). This non-heart specific enzyme plays a role in the glycolytic pathway. In contrast to all other biochemical markers, GP-BB may be already released into the circulation during periods of reversible ischemia. In addition, like the other markers, GPBB can also be measured in increased levels in circulation following cell necrosis.

Another line of investigation may be the role and use of cardiac markers together with coagulation parameters for patients with acute coronary syndromes. However, it is crucial, that all indices can reliably and quantitatively be measured 24 hours a day, 7 days a week, with a turn-a-round time of preferably less than 30 minutes, but definitely less than 60 minutes. In this respect, it will probably turn out that the best way to fulfill this requirement will be the use of point-of-care equipment.

Another issue to investigate may be the content of cardiac markers within the human heart in relation to heart failure. The marker brain natri-uretic peptide (BNP) may, in respect to heart failure, also be a promising parameter for diagnostic purposes.

Which cardiac troponin parameter (I or T) should be used in clinical practice? So far, only in patients with end-stage renal disease different results have been reported for cardiac troponin I compared with cardiac troponin T. The levels of cardiac troponin T are more frequently elevated compared with cardiac troponin I. To date, however, there is no consensus which parameter is more reliable in this respect. Several explanations have been reported to explain the difference between the troponin I and troponin T results in patients with end-stage renal failure, varying from real myocardial tissue damage to reexpression of cardiac troponin T in skeletal muscle. In this respect, important evidence has been provided by Apple et al. (Clin Chem December 1999). They concluded that cardiac troponin T mRNA was reexpressed in skeletal muscle of end stage renal disease patients.

As mentioned previously, the use of cardiac troponin T has been patented by Roche™ (the former Boehringer Mannheim™). So, this marker can only be measured with equipment of this particular manufacturer. In contrast, cardiac troponin I can be measured on (binding-)analysers from most of the other equipment manufacturers. But the compatibility among cardiac troponin I results from the different manufacturers is complicated by the lack of cardiac troponin I standardisation.

In conclusion, clinically (except for end-stage renal disease patients) there is no preference for cardiac troponin I or cardiac troponin T. Therefore, whether cardiac troponin I or cardiac troponin T should be performed, depends usually on the manufacturer of the equipment used in the laboratory.

Finally, it should be taken into consideration that the role of CKMB as “gold standard” for myocardial tissue damage has been become more or less obsolete with the acceptance of the troponins as 100% specific markers for myocardial tissue damage. The use of cardiac troponin I or cardiac troponin T in combination with CKMB should be avoided, because this procedure may induce confusion, especially, if only one of the markers is beyond the upper limit of reference range. Moreover, it is more expensive to measure CKMB in conjunction with cardiac troponin I or cardiac troponin T.

Epilogue

This thesis could not have been completed without the cooperation of many persons. First of all, I want to thank Prof.dr. S. Poppema, Prof.dr. H.J.G.M. Crijns and Prof. F. S. Apple for being my promotores. Dear Fred Apple, a special word for you. Besides I am very honoured you are one of my promotores, I want to thank you for the quick responses after reviewing the various manuscripts of the thesis, and I am very pleased that you come to Groningen to be present during the public defence of the thesis.
The referent Mike de Jongste deserves special attention. He was one of the persons, who was very constructive and enthusiastic from the beginning. Moreover, more than any other person, he was able to stimulate and to inhibit, whenever it was necessary.
Prof.dr. M.P. van Dieijen-Visser, Prof.dr. T. Ebels, and Prof. D.B. Sacks are gratefully acknowledged for being member of the commission for judging the thesis.
Marja van Luyn is gratefully acknowledged for her cooperation in the design of the cover.
The skilful technical assistance of the stageairs Katrijn Straatman, Sandra Oomkes, Bertine Grootemarsink and Erik Nijboer is also gratefully acknowledged. Besides the cooperation of many laboratory technicians, especially, the technical assistance of Michiel Weening (for performing the CK-isoform analyses) and of Petra Visser-van Brummen (for performing the work on the heart tissue specimens obtained after autopsy and for performing most of the troponin I and troponin T measurements) is gratefully acknowledged.
Furthermore, I want to thank Marcel Volmer. For answering the statistical problems I could always rely on him. Also he manufactured most of the figures shown in the various chapters of the thesis.
Of great importance was also the pleasant and skilful cooperation with many physicians in order to investigate the clinical assessment of the cardiac markers in different patient populations. From these persons I want to mention T. Tiebosch (chapters 2 and 3), S. van den Broek (chapter 6), J. Klaase (chapters 7 and 10.1), H.J. ten Duis (chapter 7), H. Sonneveld, M. Wierda and A. Girbes (chapter 8), A. Epema, B. Loef and J. Grandjean (chapter 9), and W. Meijer and I Kema (chapter 10.2).
Financial support from Abbott™, Beckman/Sanofi™, Dade Behring™, DPC™, Helena Laboratories™, Merck™, Ortho Clinical Diagnostics™, and Roche™ (former Boehringer Mannheim™) is also gratefully acknowledged for making it possible to carry out the various biochemical analyses for most of the reported studies and for completion of the manuscript.
Of course, I also want to thank Eddy Ligeon and Ido Kema for being my paranimfs.
But most of all I want to thank all patients for their approval to cooperate with the various studies.