Troponin Release—Reversible or Irreversible Injury? Should We Care?

Allan S. Jaffe* and Alan H.B. Wu

Controversies concerning whether cardiac biomarkers can be released in the absence of cell death have existed for years. The initial observations were made with lactate dehydrogenase (LDH), the 134-kDa enzyme that was found to be released from cells or organs in response to tissue injury in the absence of overt cell death [1]. Similar issues arose with creatine kinase (CK) and CK isoenzyme MB (CK-MB). Heyndrickx et al. [2], who found increases in CK in response to brief (5–15 min) coronary occlusions thought to be insufficient to induce cardiac injury, argued that the release was due to ischemia and not cell death; however, pathologic confirmation of this finding was not demonstrated. Subsequently, Ishikawa et al. attempted to cause CK release in animals in the absence of cell death by creating small, graded coronary occlusions [3]. Whenever they found CK release in the circulation, they observed cells that appeared severely damaged and necrotic, according to electron microscopy results and extensive evaluations of the potentially ischemic area. These findings were supported by the absence of mitochondrial CK in the blood, a biomarker that would provide clear evidence of irreversible cell death. For these reasons, this group argued that the release of CK (86 kDa) was due to cell death. The discussion has been extensive regarding this controversial issue of whether biomarkers can be released from cells in the absence of cell death. Initially, it was thought that regeneration of cardiac myocytes did not occur. That led to a real concern that if the release of biomarkers was due to cell death, one could in essence run out of heart tissue over time. That possibility is no longer a concern, because we now know that cardiomyocytes can regenerate and repair the heart [4].

This controversy became topical again with the advent of cardiac troponin assays. Increases in cardiac troponins are far more sensitive as biomarkers of cell damage than CK-MB, with larger -fold increases produced relative to the upper reference limit. This fact has led to speculation that all of the cardiac troponin found in blood might not be due to cell death [5]. This speculation has been supported by cardiac troponin release kinetics in certain clinical situations, which has led to the argument that perhaps cardiac troponin is released after reversible injury. The argument goes as follows: An early-releasable cytosolic pool of cardiac troponin (although whether this pool resides exclusively in the cytosol is not clear) appears to be responsible for the early release of cardiac troponin. Cardiac troponin concentrations are similar to those of CK-MB if one includes only this pool. Thus, the greater clinical sensitivity of cardiac troponin reflects the fact that a greater percentage of this pool reaches the blood after cell injury. Subsequent cardiac troponin release from a structural pool over time explains the prolonged persistence of cardiac troponin in the circulation despite the quite short half-life of the protein (observed in animal studies in which purified cardiac troponin was injected). Release from the structural pool was postulated to be synonymous with cell death, and perhaps release from the cytosolic pool could be due to either reversible or irreversible injury. Initially, prolonged cardiac troponin release was not observed in patients with pulmonary embolism, a finding that led to the idea that pulmonary embolism was a pathology in which release came only from the releasable pool and therefore could be indicative of reversible injury and not cell death [5]. Unfortunately, we now know that most of those studies were done with inadequately sensitive cardiac troponin assays. Indeed, resolving the question of whether sustained release occurs in patients with pulmonary embolism or in individuals after extreme exercise will require a study to determine whether sustained minor increases in cardiac troponins occur. On the basis of one published study in this area conducted with a high-sensitivity cardiac troponin T (hs-cTnT) assay, it appears that 72 h are required for all values to return to normal [6]. Because high-sensitivity assays are now available, new studies are needed to sort out the data that originally led to the speculation that cardiac troponin was released in response to reversible injury.

Mechanistic studies have shown that necrosis is not essential for cardiac troponin release, but, in fact,
there are other causes of cell death. Preload (7) and integrin stimulation (8) both have been shown to cause proteolysis and cardiac troponin release. For preload increases, the cells die because of apoptosis; therefore, that does not appear to be a circumstance in which reversible injury occurs. In this situation, although not due to necrosis, cell death is present. The circumstance involving integrin stimulation is more complex. With LDH used as a biomarker of cell death, the argument was that because LDH was not seen in the media, the cells were viable. There are a variety of experimental issues having to do with the time courses of such studies, however, when one needs to look for different proteins and whether the cells need to be stimulated. In addition, the finding of blebs on cardiac cells has led to the speculation that cardiac troponins might be released via blebs (9). Mechanisms might be invoked to allow for this possibility (9, 10), but direct proof outside of hepatocytes is lacking. Data from experimental studies suggest that bleb formation can occur in cultured myocytes (9) and perfused rat hearts (9). At present, the controversy regarding whether these cells are fated to die has not been resolved.

This controversy has continued unabated with the emergence of several other lines of clinical evidence indicating that reversible injury can be the mechanism of cardiac troponin release. The first is the consistent finding that cardiac troponin increases can occur after extreme exercise. The idea that exercise might be associated with myocardial necrosis has led to discomfort and concern among physicians and participants alike. Studies have suggested that such increases are not associated with short-term cardiovascular risk. The results of long-term studies, which are now just beginning to appear, are suggestive—but are not definitive—that a hazard may be associated with consistent long-term severe exertional exercise with regard to the development of cardiomyopathy and sudden cardiac death (11). Eventually, an answer is needed for this critical question.

As high-sensitivity assays have been developed, a variety of other lines of evidence have developed to argue in favor of cardiac troponin release in the absence of myocardial necrosis. Increases of 1 to 2 ng/L in the concentration of cTnI measured with an hs-cTnI assay have been observed in ischemia induced during stress testing (12). The same signal has not been detected with hs-cTnT, suggesting that the hs-cTnT assay has lower sensitivity and precision or that a different mechanism may be responsible for the egress of cTnT out of the myocyte. It is unclear whether the cells involved are viable and persist or whether they exhibit minor degrees of irreversible injury leading to cell death and replacement. Similar observations based on coronary sinus sampling suggest that rapid pacing that causes supply-demand imbalance induces modest increases detected in high-sensitivity cardiac troponin assays, not only in the coronary sinus but eventually in the systemic circulation (13). These increases were observed with and without increases in lactate, which is the traditional marker of myocardial ischemia. The increases could be due to reversibly injured myocytes and the release of cardiac troponin from such cells. An alternative explanation is that cardiac muscle is more like skeletal muscle than previously believed. Extreme exercise produces skeletal muscle damage, which then leads to replenishment of the muscle, presumably from primitive skeletal muscle satellite cells that exist within the muscle (14). Biopsy studies have clearly shown necrosis of the skeletal muscle to be followed by the reestablishment of viable myocytes in response to the injury. One wonders whether the same paradigm might apply to the heart as well. We may have been biased over these many years by the concept that such reparative processes were not available in myocardium.

This issue seems to be important from a basic science perspective; these questions are interesting ones that have relevance to cell biology. The more important clinical question is whether one can continue to use cardiac troponin as a biomarker of cell death. First, if answering this question is problematic with cardiac troponins, it is very likely also problematic with all of the biomarkers, including CK-MB and LDH, although cardiac troponin is a smaller protein and thus might be more prone to release with reversible injury. The idea that release related to reversible cell injury is a phenomenon unique to cardiac troponin may not be correct. The second issue is that it is quite clear from pathologic studies that the vast majority of the increases in cardiac troponin are associated with myocardial cell death of one sort or another (not always necrosis but at least cell death) (15). Even if some of the release is due to reversible injury, the transition between reversible and irreversible injury is not easy to detect. From the clinical perspective, equating cardiac troponin increases with cardiac injury still makes a good deal of clinical sense. In almost all circumstances, increases in cardiac troponins are associated with an adverse prognosis over time, but not necessarily in the short term. That suggests that perhaps there is some degree of injury and repair but that repetitive injury may be of more clinical importance. This proposition is at present only speculation, but at least it fits the scientific data that are available. This view would then allow cardiac troponin to continue to be used as we use it today, because it does not entail a paradigm shift or some attempt to distinguish the types of lesions that can lead to its release. One needs to acknowledge that
arguments in both directions can be made at present, and there is scientific support for both points of view. Clinically, however, there is no information currently to suggest that one can distinguish one type of injury from another (even if both types exist) or to answer why, clinically, one needs to make a distinction between these release mechanisms. In the short term, the lack of such information should not affect the use of cardiac troponin for routine clinical care.

Author Contributions: All authors confirmed they have contributed to the intellectual content of this paper and have met the following 3 requirements: (a) significant contributions to the conception and design, acquisition of data, or analysis and interpretation of data; (b) drafting or revising the article for intellectual content; and (c) final approval of the published article.

References