Introduction

Because no bedside method is currently available to evaluate myocardial contractility independent of loading conditions, a biological marker that could detect myocardial dysfunction in the early stage of severe sepsis would be a helpful tool in the management of septic patients *(Raphael and Remi., 2006).*

In 2000, the Joint European Society of Cardiology/American College of Cardiology Committee proposed a new definition of myocardial infarction based predominantly on the detection of cardio-specific biomarkers troponin T and troponin I in the appropriate clinical setting.

Given that cardiac troponin is highly sensitive for detecting even minimal myocardial-cell necrosis, these markers may become 'positive' even in the absence of thrombotic acute coronary syndromes *(Jeremias and Gibson., 2005).*

The major advantage of cardiac troponin I (cTnI) is its ability to detect myocardial cell damage that is undetectable by conventional enzyme methods, with high sensitivity and specificity. Abnormal cardiac troponin-I levels in patients with sepsis and septic shock have been reported *(Ammann et al., 2001).*

Sepsis and other systemic inflammatory processes may lead to myocardial depression and cellular injury, greatly increased oxygen consumption, reduced micro-vascular circulation, and decreased oxygen delivery to the heart, ultimately resulting in the release of troponin into the systemic circulation *(Spies et al., 1998).*

Abnormalities of cardiac function are quite common in patients with sepsis. The prevalence of this transient phenomenon critically depends on the population studied, the definition applied, and the time point during the course of the disease. Approximately 50% of patients with severe sepsis and septic shock seem to have some form of impairment of left ventricular systolic function *(Ver Elst et al. 2000).*

Recently, plasma cardiac troponin has been proposed as a biomarker that accurately detects myocardial dysfunction and provides prognostic information in septic patients. *(Raphael and Remi., 2006).*

Aim of the Work

The aim of this study is to determine myocardial injury in patient with sepsis by measuring serum cardiac Troponin I (cTnI), to evaluate relationship between elevated cTnI and sepsis induced myocardial dysfunction and to determine if cTnI is a predictor of outcome in these patients.

Historical Perspective

The word sepsis is derived from the Greek language. Pepsis was good, embodying the natural processes of maturation and fermentation. Sepsis, however, was bad and synonymous with putrefaction as characterized by bad smell *(Majno, 1991).*

It was thousands of years later before Pasteur conclusively linked putrefaction to bacterial cause. The word *shock* has its derivation from the French root "choquer," meaning "to collide with." Based on our current understanding of the pathophysiology of septic shock, the collision of the body's defenses with the invading organism, this seems to be particularly appropriate terminology *(Phillip, 2003).*

Myocardial dysfunction in the setting of acute organ dysfunction in severe sepsis and septic shock has been known for many years. This cardiac depression, mainly characterized by left ventricular failure, is estimated by echocardiogram-derived left ventricular ejection fraction (LVEF). In addition to the cardiac effects of the inflammatory responses in septic patients, cardiac tissue histopathology studies have also revealed cellular tissue necrosis, as a consequence

of hypotension, the action of circulating myocardial depressant substances or the use of catecholamine. *(Parker,1999).*

Myocardial cell injury accompanies causes or results from the decreased cardiac performance in sepsis. High levels of cardiac troponins have been reported in many critically ill adult patients, including sepsis, without acute coronary syndromes *(Ammann et al., 2003).*

Sepsis, Severe Sepsis & and Septic Shock:

Sepsis is a complex disease and is the manifestation of the immune and inflammatory response to infection. Severe sepsis is defined as sepsis with organ dysfunction, while septic shock is sepsis with hypotension that persists despite resuscitation with i.v. fluids. Recent definitions recognize the importance of myocardial depression and include a low cardiac index (CI) or echocardiographic evidence of cardiac dysfunction as one of the criteria for diagnosis of severe sepsis *(Annane et al., 2005).*

Heart and Sepsis (Patho-physiological view)

Clinical manifestation of cardiovascular dysfunction:

Our understanding of the cardiovascular manifestations in severe septic shock has evolved over

the years, as new techniques to assess cardiovascular performance have become available. Before the introduction of the pulmonary catheter (PAC), two distinct cardiovascular clinical presentations of septic shock were described: a high cardiac output (CO) state, associated with warm, dry skin and a bounding pulse despite hypotension (warm shock); and a low CO state associated with hypotension, cold , clammy skin and thread pulse (cold shock) *(MacLean et al., 1967).*

The introduction of PAC (which could measure pulmonary artery wedge pressure as a more accurate estimate of left ventricular preload) has allowed for better definition of the cardiovascular dysfunction in septic shock and has improved volume resuscitation *(Winslow et al., 1973).*

Despite the strong evidence characterizing sepsis as a hyperdynamic state, studies that examined myocardial performance still showed left ventricular dysfunction (illustrated by decreased left ventricular stroke work index) in properly resuscitated septic patients *(Weisel et al.,1977).*

Left ventricular function in sepsis:

Left ventricular diastolic dysfunction in septic shock is not as clearly defined, the dilation of the left ventricle,

and the lack of discordance between pulmonary artery wedge pressure (PAWP) and left ventricular end-diastolic volume. Both argue against significant diastolic dysfunction in sepsis *(Munt et al., 1998).*

More recent studies using echocardiography, however, have demonstrated slower left ventricular filling and aberrant left ventricular relaxation in septic patients, suggesting that impaired compliance may significantly contribute to myocardial depression in sepsis *(Charpentier et al., 2004).*

Right ventricular function in sepsis:

Low peripheral vascular resistance in sepsis leads to decreased left ventricular afterload. However, the right ventricular afterload is frequently elevated due to increased pulmonary vascular resistance from acute lung injury, these different physiologic conditions mean that right ventricle can't be expected to behave like the left ventricle in septic patients, yet there is also evidence of right ventricular diastolic dysfunction in septic patients *(Parker et al., 1990).*

The Cardiac Troponins

Cardiac troponins are regulatory proteins that control the calcium-mediated interaction of actin and myosin. The troponin complex consists of three subunits:

- Troponin T, which binds to tropomyosin and facilitates contraction;
- Troponin I, which binds to actin and inhibits actin-myosin interactions; and
- Troponin C, which binds to calcium ions *(Jeremias and Gibson., 2005).*

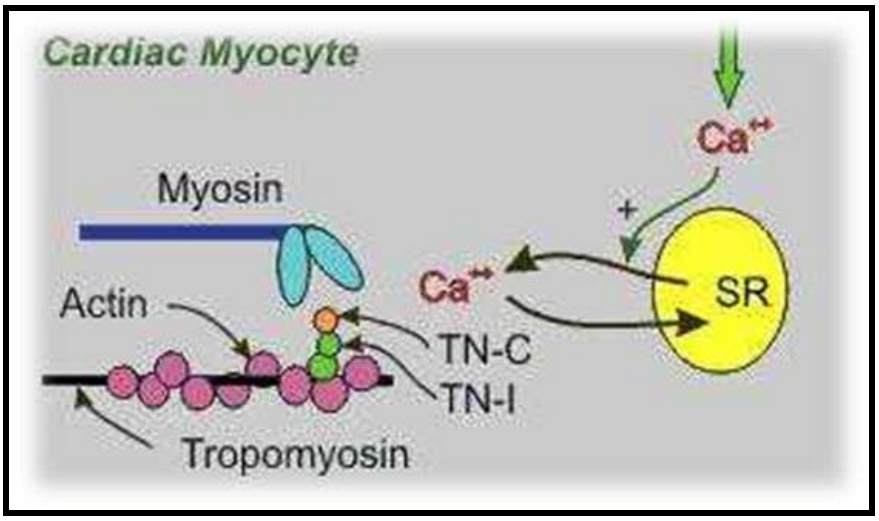

Fig. (1): Cardiac myofilaments. Myosing (thick filament) contains two heads having ATPase activity. Thin filament is made of actin , tropomyosin, and tropoinin (TN), TN-C binds Ca released by the sarcoplasmic reticulum (SR). TN-I inhibits actin-myosin binding until Ca binds to TN-C

The amino acid sequences of the skeletal and cardiac isoforms of cardiac troponin T and troponin I are sufficiently dissimilar and, therefore, differentially detectable by monoclonal antibody based assays. Troponin C is not used clinically because both the

cardiac and skeletal muscle share troponin C isoforms. Cardiac troponin I is 13 times more abundant in the heart than creatine kinase MB iso-enzyme, so the signal-to-noise ratio associated with troponin I is much more favorable for the detection of minor amounts of cardiac damage *(Wu and Jialal. 2000)*.

Origin of cardiac troponin release:

Normally, cardiac troponins T and I are not detectable in the blood of healthy persons. Release of these troponins can occur when myocytes are damaged by a variety of conditions, such as trauma, exposure to toxins, inflammation, and necrosis due to occlusion of a portion of the coronary vasculature (*Roongsritong et al., 2004).*

The majority of cardiac troponin T and cardiac troponin I is bound to myofilaments, and the remainder is free in the cytosol. When myocytes damage occurs, the cytosolic pool is released first, followed by a more protracted release from stores bound to deteriorating myofilaments *(Wu, 2001)*.

Abnormal values have been described in various conditions not related to acute coronary disease, like: myocarditis, pulmonary embolism, acute heart failure,

septic shock, and as a result of cardiotoxic drugs as well as after therapeutic procedures, such as coronary angioplasty, electrophysiological ablation, or electrical cardioversion *(Hamm et al., 2002).*

The mechanisms of release and clearance of cardiac troponins T and I are complex and incompletely understood in these pathological conditions. Although both are structural proteins, it has been suggested that cytosolic pools of these proteins are released into the circulation after cell injury. The cytosolic pool for cardiac troponin T was estimated at 6% to 8% of total cardiac troponin and that for soluble cardiac troponin I at 2.8% of total cardiac troponin. Release of cardiac troponin T may be related to transient leakage from the cytosolic component due to loss of sarcolemmal integrity during reversible ischemia or from its continuous release when ischemic injury is irreversible *(Giannitsis and Katus, 2004).*

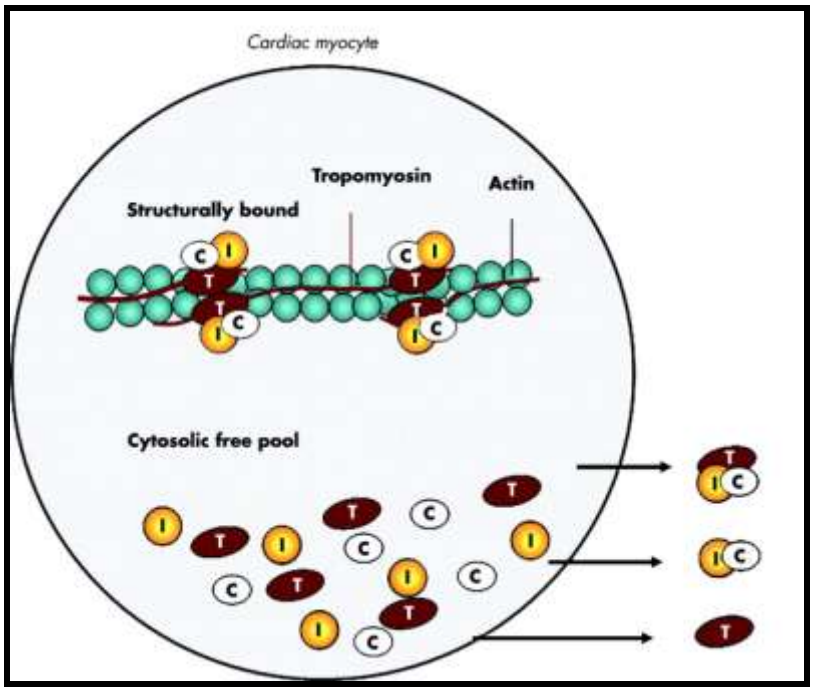

Fig. (2): Mechanism of cardiac troponin release.

Mechanism of heart tissue damage in sepsis:

The high prevalence of elevated serum levels of cardiac troponins in septic shock raises the question of *what mechanism results in troponin release in septic shock.* Proposed mechanisms include focal ischemia, and direct cardiac myocytotoxic effects of endotoxins, cytokines or reactive oxygen radicals ***(Van Bockel et al., 2005).***

In addition, activation of many intracellular pathways may cause degradation of free troponin to lower molecular weight fragments, which are released

into the systemic circulation because of increased membrane permeability *(Wu, 2001)*.

The current understanding of myocardial dysfunction in sepsis is that there is no evidence of global coronary hypoperfusion. Tools for assessing tissue and heart dysfunction have, however, evolved and enable us to reconsider the above assumption as a universal concept. In this respect, microvascular dysfunction is now considered an intrinsic aspect of sepsis sequelae. Indeed, evidence suggests that sepsis may induce perturbations in regional coronary blood flow and microvascular failure leading to myocardial ischemia *(Goddard et al., 1998).*

Endotoxins:

Endotoxin is released by lysis of gram-negative bacteria. They cause the release of other mediators such as cytokines with myocardial depressant properties. It is likely that Toll-like receptor-4 plays a pivotal role in endotoxin-induced myocyte dysfunction *(Tavener and Kubes, 2005).*

These receptors provide critical links between immune stimulants produced by micro-organisms and the initiation of host defenses. Activation causes the

release of various cytokines and propagation of the inflammatory response *(Tavener et al., 2004).*

Myocardial depressant substance:

The concept of a circulating myocardial depressant factor in sepsis was first proposed in the 1970s. In 1985, it was shown that serum obtained from patients with septic shock caused a significant depression in an in-vitro model of mammalian myocardial cell performance. This study concluded that a circulating myocardial depressant substance was a cause of the myocardial depression frequently accompanying human septic shock *(Parrillo et al., 1985).*

It is unlikely that a single factor is responsible for myocardial depression. In a study of neonatal rat cardiac myocyte cultures exposed to an ultrafiltrate obtained from either healthy volunteers or those with severe sepsis, the ultrafiltrate from septic patients caused significant toxic effects, while the serum from the control group had no effect. Analysis of the ultrafiltrate from the septic patients revealed significantly higher amounts of IL-1, IL-8, and complement component 3a when compared with controls, suggesting that a number of circulating factors are involved in sepsis-induced myocardial depression *(Hoffmann et al., 1999).*

The phenomenon of myocardial depression can be mediated by circulating depressant substances, which until now have been incompletely characterized. Among those possible candidates, tumor necrosis factor (TNFα), IL-1β and IL-6 play a central role in septic myocardial dysfunction *(Prabhu, 2004).*

TNF-α, alone or in association with IL-1β, has been implicated in the pathophysiology of septic myocardial dysfunction. Proposed mechanisms of TNF-α-induced myocardial depression include the activation of the neutral sphingomyelinase, and suppression of the calcium transient and nitric oxide pathways. TNF-α can also modulate tissue destruction and biosynthesis/activation of intracellular proteases *(Prabhu.2004).*

For example, TNF-α may induce activation of calpains and caspases that could participate in the degradation of crucial cardiac contractile proteins, including troponins. Upon activation by calcium, active calpain is released by calpastatin and cleaves cardiac troponin I at the carboxyl terminus to produce the cardiac troponin I degradation fragment. Caspases, the executioners of apoptotic cell death, also induce sarcomere disarray and are involved in the cleavage of α-actin, α-actinin and troponin T *(Communal et al., 2002).*

Alternatively, TNF-α may have an important role in cardiac injury through a variety of mechanisms, including: second messenger pathways, arachidonate metabolism, protein kinases, oxygen free radicals, nitric oxide, transcription of variety of cytotoxic genes, regulation of nuclear regulatory factors, ADP-ribosylation and, potentially, DNA fragmentation *(Prabhu.2004).*

Leukocyte and reactive oxygen species

Reactive oxygen species may play a role in the induction of several types of organ failure, including that of the heart, following the development of sepsis. Leukocyte derived superoxide and its daughter molecules are thought to be a major cause of heart injury in sepsis *(Granton et al., 1997).*

In addition, activated NADPH oxidase complexes and mitochondria are also potential sources of free radicals in the septic heart, which may have multiple potential sites of action *(Supinski and Callahan, 2006).*

Under patho-physiological conditions, dramatically elevated levels of reactive oxygen species may cause significant damage to cellular proteins and membranes as well as to nucleic acids, leading to single strand breaks and chromosomal alterations, which are all likely to