Peer Review File
Article information: https://dx.doi.org/10.21037/jlpm-23-40

Reviewer Comments

Reviewer A

Comment 1: In my opinion, however, I sometimes miss references that clarify or underline particular statements. It will improve readability when they are included in the first sentence belonging to e.g. line 78, 97, 101 (!), 201, 204, 252, 266, but this is the case throughout the manuscript.

Reply 1: All references have been updated both in the first line and end of the corresponding paragraph.

Comment 2: Abstract, Introduction

- Line 46, the authors state that acute myocardial injury and type 2 MI account for more than 50% of measurable cardiac troponin elevation in hospitalized patients. To me, this seems somewhat exaggerated. This number is also not really discussed later on in the manuscript. I think this is important to include in the manuscript to put things better into perspective.

Reply 2: We accept that the incidence of acute myocardial injury and type 2 myocardial infarction differs greatly in different study populations, owing to patient selection and the troponin assay used. We have referenced a range of studies in the manuscript but in the abstract have focused on unselected populations of suspected acute coronary syndrome as we believe this gives the most generalizable representation of the population. Thus, we have amended the comment to say

"Acute non-ischaemic myocardial injury and type 2 myocardial infarction are responsible for around half of measurable cardiac troponin elevation in unselected hospitalized patients presenting with suspected acute coronary syndromes."

Comment 3: Diagnosis of myocardial infarction and myocardial injury

- This paragraph should refer to Figure 1 since this figure is about the identification of the subtypes of MI and myocardial injury using cardiac troponin.

Reply 3: This has been updated

Comment 4: - Figure 1 suggestion: For acute and chronic myocardial injury, this figure describes some information on how to use the troponin concentration for identifying myocardial injury, while for the types of MI only the cause is mentioned.
Mechanisms of troponin release in type 2 myocardial infarction and non-ischaemic myocardial injury

Reply 4: We agree with the reviewer that we should define the clinical mechanisms of injury in both type 2 myocardial infarction and non-ischaemic myocardial injury. We have adapted Figure 1 so that a cause is also present for each subtype of injury/infarction.

Comment 5: - Line 117, this paragraph refers to figure 2 before figure 1 is referred to, also figure 2 does not provide information that fits with this paragraph.
Reply 5: Thank you for recognizing this oversight, which has been corrected

Comment 6: Using cardiac troponin to differentiate myocardial injury or infarction subtype
- For Highs-STEACS, perhaps mention that cardiac troponin I was measured, instead of generalizing it to "troponin".
Reply 6: We have updated the text to specify troponin I.

Comment 7: Using cardiac troponin to risk stratify type 2 myocardial infarction and acute myocardial injury
- N.A.

Randomized control trials in type 2 myocardial infarction and acute myocardial injury
- N.A.

Novel cardiac biomarkers
- The paragraph (Line 225 – 233) of troponin release into the circulation misses references. The 10-fold higher troponin I concentrations will definitely be assay-dependent and is no common knowledge.
Reply 7: We apologize for this oversight which occurred in error. This paragraph is now appropriately referenced.

Comment 8: - With the statement (Line 227-230), "following cardiomyocyte necrosis… phagocytosis and degradation", you indicate that cTnI is only degraded in the circulation and that cTnT is only degraded inside the cardiomyocyte. However, cTnT is also degraded in the circulation, in particular by thrombin. So both for cTnI and for cTnT fragments are in the circulation that are detectable by current clinical immunoassays, since antibodies are directed against the central part of the troponins both detect intact and fragmented proteins. Line 231 is thus incorrect. Please rephrase this statement.
Reply 8: We apologize for our overly simplistic description. We agree fragments of both cardiac troponin I and cardiac troponin T are released and detectable within the circulation. Prior studies demonstrate most cardiac troponin T appears bound to insoluble filaments, therefore the concentration of circulating T available for detection is relatively lower than I and may explain differences in observed concentrations. This paragraph has been rewritten for clarity.

Comment 9: Grammar/layout:
- Line 105, the word "as" is missing between classified and chronic.
- Line 161, check double "odds ratio"
- Line 236, 878 (408+46+424=478) patients had suspected ACS. Please correct.

- Line 297, typing error “understudied”
- Figure 2: these mechanisms are all hypothesized, it has not been proven that troponin is released in that manner.

Reply 9:
All errors have been corrected.

Reviewer B

Comment 1: The review is concise, however precise in reporting relevant data from recent clinical trials.
Since troponin enriched in circulating extracellular vesicles was detected in patients with unstable angina this observation must be commented and considered as a future biomarker tool to stratify patients (see PMID: 34111564, PMID: 34638611).

Reply 1: Thank you for your helpful and constructive review, and for highlighting two recent and important papers. We have now included both in our updated manuscript, please see lines 136-139

“Recently troponin enriched in circulating extracellular vesicles was detected in patients with documented unstable angina. This may serve as a future biomarker to risk stratify patients with both acute and chronic coronary syndromes for intensification of cardioprotective therapies [23,24]”

Reviewer C

Comment 1: General comment:
It's an interesting paper, pleasant to read, well-written and concise.

However, it would be useful to add a sentence on how the literature review was carried out. It would also be important to add an introductory sentence on the reason for carrying out this literature review.

Reply 1: We thank the reviewer for their constructive appraisal of our manuscript and are glad it was of interest. This was not a formal systematic review, rather a synthesis of the available evidence based on current novel areas of cardiac biomarker technology and areas of clinical uncertainty. It would be disingenuous for us to suggest we have undertaken a systematic literature review and therefore we have not added this to our manuscript.

Comment 2: Abstract:
Line 47-49: reformulate, please. The message is not clear.

Reply 2: In line with the recommendations of this and other reviewers, our abstract has been rewritten for clarity.

Comment 3: Introduction:
It could be more appropriate to write: “Increasingly, we recognize/ highlight cardiac troponin concentration above the 99th upper reference limit (URL)) across a spectrum of both cardiac and non-cardiac pathologies” instead of “Increasingly, we recognize

both acute and chronic non-ischaemic myocardial injury (cardiac troponin concentration above the 99th upper reference limit (URL)) across a spectrum of both cardiac and non-cardiac pathologies".
Reply 3: Thank you, we agree and have updated the text.

Comment 4: Diagnosis of myocardial infarction and myocardial injury:
Add references for the paragraph from line 94-101
Add references for line 103-105.
Reply 4: Thank you, these have been updated.

Comment 5: - Mechanisms of troponin release in type 2 myocardial infarction and non112 ischaemic myocardial injury
Reply 5: Thank you, these have been updated.

Comment 6: Line 128: Cardiomyocytes

Conclusion:
You're just repeating the same sentence as in the introduction. Then you come up with a conclusion that is unsatisfactory in the sense that you don't propose anything. It would be interesting to flesh it out and make some suggestions.
Reply 6: Thank you for your feedback we have updated the conclusion see lines 304-313

Following the implementation of high-sensitivity cardiac troponin assays, both acute non-ischaemic myocardial injury and type 2 myocardial infarction are increasingly recognised in practice. In an increasingly elderly and co-morbid population, the prevalence of these conditions is likely to increase. Current treatment strategies focus on correcting the mechanism of supply or demand imbalance, but evidence is emerging that important unaddressed cardiovascular disease exists. Targeted investigation for coronary disease and left ventricular impairment followed by appropriate secondary prevention therapy may provide the best opportunity to modify future cardiovascular risk. It is now time that such strategies are evaluated prospectively in a randomized controlled trial.

Reviewer D
Comment 1: - L45-47: not clear where "over 50%" comes from.
- L91: not clear where "one in five events" comes from. My understanding is that the % of type 2 events has been very variable depending on the study and patient population
- L100-101
- L225-233
- L285-286 ("one in six at one year")
- Table 1: "A Study of Microcirculatory Function in Type 2 Myocardial Infarction" needs a reference
Reply 1: Thank you for your review. We have clarified this point in line with your own

and previous reviewers' comments. All references and typographical errors are now corrected.