

## Peer Review File

Article information: <https://dx.doi.org/10.21037/jlpm-21-37>

### Reviewer A

The authors present a narrative review which deals with diagnostic and predictive biomarkers” in T2DM and prediabetes.

The manuscript is very tedious to read because the English is poor.

Aside from this limitation my impression is that the review does not live up to its title.

The authors present literature including many reviews which mostly deal with biomarkers in heart failure (HF) in general. The specific role, if any, of biomarkers to diagnose HF and/or predict future HF or cardiovascular events in patients with diabetes or prediabetes remains obscure. On page 10 the authors report that “the Dallas Heart Study has revealed that levels of galectin-3 correlated well with incident T2DM, metabolic syndrome, and body fat composition. While this is correct, it has little to do with the title.

Some articles are also not cited correctly. Two examples:

Page 12, lines 345 ff: The authors of reference 85 state in the abstract that “concentrations of adiponectin were not associated with (incident) heart failure” while it is cited as showing “that a risk of HF occurrence was accompanied by increased circulating levels of several adipokines, such as adiponectin and resistin...”.

**Reply: We thank you for your concise comment. We checked the reference #85 and have modified our text according to the authors’ findings: “The results of the Framingham Offspring Study have shown that an incident HF might accompany by increased circulating levels of several adipokines, such as resistin, and that this relationship was changed after adjustment for prevalent CAD, abdominal obesity, insulin resistance and inflammation”**

Page 12, lines 355 ff: Here ref. 89 is cited to indicate that adiponectin showed a J-shaped association with cardiovascular disease and/or HF risk. Again in their abstract the authors of this article indicate that adjustment for additional potential confounders “led to the attenuation of the estimate of effect...across consecutive quintiles of adiponectin”. In other words the initially observed significant differences in relative risk between quintiles were lost.

**Reply: We thank you for your concise comment. We checked the reference #89 and have modified our text accordingly. “The relationships between serum levels of the majority of pro-inflammatory adipokines and CV risk and a risk of HF development were not linear, but were J-shaped (adiponectin) or U-shaped (chemerin-1, omentin) associations, which were diminished for adiponectin after adjustment for additional potential confounders”**

These examples of an area I am quite familiar with raise the possibility that more citations may not be correct and it is not the task of reviewers to check every citation. An additional issue here is that the reference list contains many reviews. In my opinion reviews should rely on original literature rather than other reviews. This does not apply only when review articles are cited to briefly present the state of knowledge before a certain date. However, this does not appear to be the case here.

**Reply: We thank you for your proposal. We checked the review articles that were cited and minimized their number when relevant.**

Minor points:

page 7, line 178: EASD – European Association for the Study (not society) of Diabetes

page 9, line 257: a protein can be overexpressed in cells but not in extracellular matrix (no protein expression in the extracellular space).

**Reply: We thank you for your concise comment. We have modified our text as advised. The abbreviation of EASD was correctly explained (page 7, line 178). The sentence (p 9, line 257) was modified accordingly: Galectin-3 is multiphase protein, which belongs to the  $\beta$ -galactoside-binding lectin family [58], and is overexpressed in different types of cells due to tissue injury or stress.**

## **Reviewer B**

I read with interest and I congratulate the authors for hard work. The review is balanced and of clinical value. My suggestions to further improve the quality of the review are listed below:

I would like to advise the authors to re-structure the abstract to give more space the use of these biomarkers for diagnosis and risk stratification of patients with heart failure and diabetes and not to concentration on limitations (primarily) as shown in the following 2 statements:

1. “NPs demonstrated their optimal ability to diagnose and predict HF with reduced (HFrEF) and preserved (HFpEF) ejection fraction, but their utility to stratify individuals at risk and manage patients with HFpEF exerts strict limitations due to a high variability in CV and metabolic comorbidities, such as DM and pre-DM.”

**Reply: We thank the reviewer for comprehensive comment. We have re-written the sentence and it is now provided in the following manner: “Natriuretic peptides (NPs) and high sensitive cardiac troponins remain to be powerful tool to stratify, diagnose and manage patients at risk of HF and with established HF”**

2. Although natriuretic peptides (NPs) and high sensitive cardiac troponins are suggested to use as powerful tool to stratify, diagnose and manage patients at risk of HF and with established HF, NPs remain the only biomarkers that are incorporated in the majority of International HF clinical guidelines.”

**Reply: We thank the reviewer for expressive comment. The sentence has been changed accordingly. “NPs demonstrated their optimal ability to diagnose and predict HF with reduced (HFrEF), mildly reduced (HFmrEF) and preserved (HFpEF) ejection fraction regardless of presentation of different metabolic comorbidities, such as DM and pre-DM.”**

I also went throughout the material and found several defective sentences and I gave my proposals to improve them (please see below)

**Reply: We thank the reviewer for a hard work with the aim of improving the quality of the paper. We agree with all proposals given below and all correction were made by point-to-point accordingly**

Line 64 ...a leading cause... not “a leading causes”

**Reply: we have modified our text as advised**

Sentence in lines 66-69, problematic, incomplete, not understandable.

**Reply: we have modified this sentence: Due to dramatic number of new cases of DM worldwide HF is suggested to be steadily growing life-threatening complications of DM**

Sentence in lines 72-73 not understandable.

**Reply: we have modified this sentence: Additionally, economic burden of this strategy appears to be challenging and requires more attention to an optimal choice of the biomarkers in routine clinical practice**

Sentence in lines 75-77. Please explain for what strategy are you speaking; term “practical matter” is inadequate.

**Reply: We changed this sentence completely and refused from the term “practical matter”: Additionally, economic burden of this strategy appears to be challenging and requires more attention to an optimal choice of the biomarkers in routine clinical practice**

Sentence in lines 82-84 is problematic; needs restructuring.

**Reply: We changed this sentence: there is no biomarker-guided strategy for DM patients at higher risk of HF and also for HF patients with at risk or overt DM.**

Line 107-108: what does the “decline the first admission and rehospitalizations due to HF progression” mean?

**Reply: We changed the sentence as “...decrease in a risk of the admissions, ...”**

Lines 131-132: “while their potency in this manner”. Does it mean: “their performance appears to be better for HFrEF and HFmrEF than HFpEF”?

**Reply: We changed the sentence as proposed “while their performance appears to be better for HFrEF and HFmrEF than HFpEF”**

Line 132: “their role” not “roles”.

**Reply: we have modified our text as advised**

Line 133: please avoid the term “diabetics”; use instead “patients with diabetes”. A friendly suggestion.

**Reply: we have modified our text as advised**

Sentence in lines 132-135, not understandable; needs restructuring.

**Reply: We changed this sentence: “NPs are considered to be predictors for atrial fibrillation / flutter and HF manifestation in patients with DM and obesity, however, the serum levels of these biomarkers are remarkably variable for these patients and those who have chronic kidney disease”**

Sentences in lines 136-142, not understandable; need restructuring.

**Reply: We changed this sentence: Adipose tissue accumulation was found to be associated with near normal plasma NP levels, but chronic kidney disease was, on contrary, a frequent cause of dramatic rise of NP levels due to lowered kidney clearance**

Opening word “Despite” and “Regardless” are used inappropriately.

**Reply: Opening word have been deleted**

Lines 42-142; please substitute “ simultaneously having risks of” with “at risk of”.

**Reply: we have modified our text as advised**

Line 147: please correct “overt the disease”.

**Reply: We substituted the term to “established HF”**

Line 151: “similar predictive value”; similar to what?

**Reply: We substituted “similar predictive value” to “high predictive value”**

Line 154: correct “diagnose rule-in”

**Reply: We substituted “diagnose rule-in” to “diagnostic utility”**

Line 155: please substitute “is needed” with “needs”.

**Reply: we have modified our text as advised**

Lines 159-161: this part of the sentence is problematic. “the elevated levels of NT-proBNP by itself have revealed discriminatory ability similar to a multivariable model? This part of the sentence is not understandable.

**Reply: The sentence was changed: the elevated levels of NT-proBNP predicted both CV mortality and composite CV events (CV death, resuscitated cardiac arrest, nonfatal myocardial infarction, stroke, or HF hospitalization)**

Line 168: substitute “declined” with “reduced”.

**Reply: we have modified our text as advised**

Line 169: delete “therefore”.

**Reply: we have modified our text as advised**

Lines 171-172: please substitute “dynamic of NTproBNP levels” with “dynamic NTproBNP levels”.

**Reply: we have modified our text as advised**

Line 173: If the authors here start a new paragraph, please delete “therefore” at the beginning of the sentence.

**Reply: we have modified our text as advised**

Sentence in lines 173-176 too complex; not understandable; needs restructuring.

**Reply: the sentence was changed “Implementation of BNP biosensing platforms based on optical and electrochemical immunosensor methodology allowed sufficiently reducing total expenditures on BNP level monitoring in follow-up, and also this test demonstrated unsurpassed sensitivity, selectivity, and reproducibility”**

Line 178: delete “when”.

**Reply: we have modified our text as advised**

Line 180: please substitute “issue” with “finding”.

**Reply: we have modified our text as advised**

Sentence in lines 185-186: Please change as follows: “Finally, NPs are the most accurate test for HF diagnosis and progression of the disease as well as for predicting adverse outcomes and guiding HF therapy.”

**Reply: we have modified our text as advised**

Sentence in lines 187-188 may be deleted.

**Reply: we have modified our text as advised**

Line 191 please substitute “structure” with “structural”.

**Reply: we have modified our text as advised**

Line 192: troponin is structural protein; “hs-cTn” is an assay for troponin measurement and not structural protein.

**Reply: We substituted “hs-cTn” with “troponins”**

Sentence in lines 198-200: Please change as follows: “In the last 2 decades, attempts are made to identify minor asymptomatic myocardial injury and interpret its clinical relevance in patients beyond acute myocardial infarction and acute coronary syndromes”.

**Reply: we have modified our text as advised**

Lines 200-201: Please change as follows: “They were enabled by the development and use of new analytical approaches to detect cardiac troponins with higher accuracy and reproducibility.”

**Reply: we have modified our text as advised**

Line 201: “measurement” instead of “measure”.

**Reply: we have modified our text as advised**

Line 202: these words “shifted previously shaped opinion” are not understandable.

**Reply: we have modified our text as the following: “measurement of the cardiac troponin levels using commercially available second generation high-sensitive tests allowed turning previous opinion of them as only indicators of myocardial injury to biomarkers of biochemical stress?”**

Line 203: “mitochondrial” or “myocardial”?

**Reply: biochemical stress sounds better**

Line 206: please change “mildly upper of normal level” to “mildly above the normal level” or simply “mildly elevated”.

**Reply: We changed the sentence: “were mildly above the normal level”**

Line 207: delete “a” before “poor”.

**Reply: we have modified our text as advised**

Line 209: “individuals at risk”; at risk for what?

**Reply: we have modified our text the following: “at risk of HF / established HF”**

Line 212: please substitute “has been revealed” with “showed”.

**Reply: we have modified our text as advised**

Line 218: please substitute “has yielded” with “showed”.

**Reply: we have modified our text as advised**

Line 221. please substitute “has yielded” with “showed”.

**Reply: we have modified our text as advised**

Line 223: please substitute “among” with “compared with”.

**Reply: we have modified our text as advised**

Line 225: please substitute “global” with “general”.

**Reply: we have modified our text as advised**

Line 230: please substitute. “but not in both version (2016 and 2021) ESC HF” with “but not in 2016 and 2021 ESC guidelines for HF”.

**Reply: we have modified our text as advised**

Line 232: please substitute “require being clearer elucidated” with “need further elucidation”.

**Reply: we have modified our text as advised**

Line 235: please substitute “adds predictive value to” with improved predictive value of”.

**Reply: we have modified our text as advised**

Line 243: please deleted “provided” before “by”.

**Reply: we have modified our text as advised**

Line 252: please substitute “while respectively” with “but”.

**Reply: we have modified our text as advised**

Line 257: please substitute “and” with “that”.

**Reply: we have modified our text as advised**

Line 258-259: please change the sentence to “There has been a continuously rising interest in galectin-3 due to extensive studies involving the molecule in adverse cardiac remodeling, atherosclerosis and T2DM.”

**Reply: we have modified our text as advised**

Line 268: “has yielded” with “showed that”.

**Reply: we have modified our text as advised**

Line 273: please substitute “at the baseline” with “compared with baseline values”.

**Reply: we have modified our text as advised**

Line 280: please substitute “Suggesting” with “Considering that”.

**Reply: we have modified our text as advised**

Line 281: please substitute “correspond) with “lead”.

**Reply: we have modified our text as advised**

Line 282: please substitute “has revealed” with “showed”.

**Reply: we have modified our text as advised**

Line 287: “galectin-2” or “galectin-3”?

**Reply: We changed this typos from galectin-2 to galectin-3**

Line 290: please add “but” before “it”.

**Reply: we have modified our text as advised**

Line 293: please substitute “based on manly” with “based mainly on”.

**Reply: we have modified our text as advised**

Line 297: please substitute “substantially” with “markedly”.

**Reply: we have modified our text as advised**

Lines 299-300: please substitute “frequently reserved to exert improved prediction for” with “are increasingly being used to assess”.

**Reply: we have modified our text as advised**

Line 302: please substitute “informative of them” “important biomarkers”.

**Reply: we have modified our text as advised**

Line 315: please substitute “an activity of co-existing” with “of”.

**Reply: we have modified our text as advised**

Line 332: please substitute “will need integration into the design of” with “needs to be investigated in”.

**Reply: we have modified our text as advised**

Line 335: please substitute “related” with “relates”.

**Reply: we have modified our text as advised**

Line 338: please substitute “source of synthesis, releasing” with “ source of synthesis and release of”.

**Reply: we have modified our text as advised**

Line 345: please substitute “a risk” with “the risk”.

**Reply: we have modified our text as advised**

Line 355: please delete “associations”.

**Reply: we have modified our text as advised**

Line 374: please substitute “associations” with “an association of”.

**Reply: we have modified our text as advised**

Line 379: please substitute “nurtured” with “led to”.

**Reply: we have modified our text as advised**

Line 382; please delete “plays”.

**Reply: we have modified our text as advised**

Line 391: “left researchers a bit puzzled” is not scientific “ may be “remains poorly understood”

**Reply: we have modified our text as advised**

Line 412: please substitute “has been shown to be expressed higher” with “shows higher expression”.

**Reply: we have modified our text as advised**

Line 413-414: please change to “Both cardiomyocytes and cardiac fibroblasts respond to higher glucose concentrations with osteopontin expression”.

**Reply: we have modified our text as advised**

Line 416: please substitute “ provide this relation” with “show such an association”.

**Reply: we have modified our text as advised**

Line 418: please substitute “evidences” with “has”.

**Reply: we have modified our text as advised**

Line 420-421; please re-structure the sentence; it is not understandable in current form.

**Reply: The sentence was changed: “In the setting of HF, levels of osteopontin increase in concordance with the severity of HF”**

Line 423: “patients” instead of “patient”.

**Reply: we have modified our text as advised**

Line 428-429: please change to “in studies with higher proportions of patients chronic kidney disease or large coronary artery occlusive disease” (if I correctly understood the sentence)?

**Reply: Thank you for your proposal. We have modified our text as advised**

Line 430-431: please change to “ seem to be promising biomarkers, but more clinical trials are required to elucidate their role”.

**Reply: Thank you for your proposal. We have modified our text as advised**

Line 442: please add “of” before “GDF-15”.

**Reply: We have modified our text as advised**

Line 444: please “delete “have”.

**Reply: We have modified our text as advised**

Line 455: should be “A pooled analysis”.

**Reply: We have modified our text as advised**

Line 464: please deleted “reported”. Place an “a” before “systematic”.

**Reply: We have modified our text as advised**

Line 465: please delete “provided”.

**Reply: We have modified our text as advised**

Line 482: please delete “which was provided”.

**Reply: We have modified our text as advised**

Line 495: add “that” before “elevated”.

**Reply: We have modified our text as advised**

Line 499: add “it was” before “no longer”.

**Reply: We have modified our text as advised**

Line 503: please substitute “has yielded” with “showed”.

**Reply: We have modified our text as advised**

Line 510: “play” not “plays”.

**Reply: We have modified our text as advised**

Line 512: “Precursors” are you meaning “precursor (or progenitor) cells” here?

**Reply: We substituted Precursors with Progenitor cells**

Line 514: delete “for”.

**Reply: We have modified our text as advised**

Line 518: please add “a marker to assess” before “multiple”.

**Reply: We have modified our text as advised**



Line 520: “the number” instead of “a number”.

**Reply: We have modified our text as advised**

Line 526: Data is plural (sing Datum), thus it should be “Data confirm”.

**Reply: We have modified our text as advised**

Line 563: please substitute “specially” with “specifically”.

**Reply: We have modified our text as advised**

Line 564: is the word “besides” used with the meaning “except for”?

**Reply: We have modified our text using the word “especially”**

Line 566: what does “while their quality is qualified and high” mean? To what part of the sentence it refers?

**Reply: We changed the sentence as “...their quality is quite high”**

Line 567: you did not use meta-analytic methodology; so there are no results subject to heterogeneity.

**Reply: We deleted this sentence**

Line 567: please substitute “a lot of” with “a large”.

**Reply: The sentence was deleted**

Line 568: please change “do not distinguish HF patients with pre-DM and over DM” to “not distinguishing patients with HF with pre-DM versus those with DM”.

**Reply: The sentence was deleted**

Lines 569-560: Please change to “However, although not desirable, we believe that these limitations do not compromise the accuracy of the reported findings or lead to their misinterpretation”.

**Reply: We have modified our text as advised**

Line 572: please substitute “on a” with “for”.

**Reply: We have modified our text as advised**

Line 573-576: Please change to “Conventional biomarkers, such as NPs, cardiac troponins are found to have an optimal ability to diagnose and predict HFrEF and HFpEF and are recommended by current on HF. However, their utility to stratify individuals at risk and manage patients with HFpEF is limited due to their high variability in in the presence of cardiovascular disease and metabolic comorbidities.”

**Reply: We have modified our text as advised**

Line 579: please substitute “deeply investigated” with “under intense investigation”.

**Reply: We have modified our text as advised**

Line 580: “power” instead of “potency”.

**Reply: We have modified our text as advised**

Lines 581-583: please change to: “Large clinical studies are required to better elucidate whether multiple biomarker approach including both conventional and alternative biomarkers will be clinically useful and cost effective.”

**Reply: We have modified our text as advised**