85159-Answering Reviewers

We are indebted to the reviewers for their insightful comments. Please find below our answers in **Red.**

Reviewer #1:

Scientific Quality: Grade B (Very good)

Language Quality: Grade B (Minor language polishing)

Conclusion: Minor revision

Specific Comments to Authors: The authors have developed an interesting manuscript in the context of the involvement of miRNAs in type 2 diabetes; specifically miR-155. It is a well-developed and coherent letter to the editor. In addition, it contains a significant amount of information contrasted with the appropriate use of bibliographic citations. The work is good, but to improve the quality of the manuscript, I would like to add three suggestions:

1. short title: it contains many abbreviations, perhaps it is not adequate.

Answer: Our title does not contain any abbreviations at all and comprises 17 words as required by the Journal (less than 18 words). We believe the title adequately explains the intention of the minireview.

2. Full name of the miRNA: in the databases it appears as hsa-miR-155-3p or 5p in humans (or mmu in mice). Is there homogeneity in the nomenclature used in the academic articles consulted?

Answer: Indeed, hsa-miR-155 denotes the human miRNA-155 and its two mature forms, miR-155-3p and miR-155-5p, of which miR-155-5p is the most abundant. Most studies have focused on miR-155-5p. In the literature, especially in reviews, hsa-miR-155 is referred to as miR-155. Details can be found in https://www.mirbase.org/cgi-bin/mirna_entry.pl?acc=MI0000681 (where hsa-miR-155 is referred to as "previous ID") and in Elton et al[1] and Desvignes et al[2].

3. What kind of studies are evidenced by the results described in the table? It would be important to clarify whether they are in vitro, in vivo, or cohort studies, and the experimental techniques used.

Answer: References 9, 10, 12, 16, 21-25 are in mouse and human *in-vitro* and *in-vivo* as well as clinical studies in humans. We have added this information to the table in the manuscript.

Reviewer #2:

Scientific Quality: Grade C (Good)

Language Quality: Grade B (Minor language polishing)

Conclusion: Rejection

Specific Comments to Authors: I have carefully studied the manuscript entitled "MiRNA-155 mediates endogenous Angiotensin II Type 1 receptor regulation: implications for innovative Type 2 Diabetes Mellitus management" by Papadopoulos et al. The topic of MicroRNAs in diabetes mellitus is of growing interest and much evidence has been accumulated the last few years. The authors comment on miR-155 effects in type 2 DM, stating that miR-155 is consistently reduced in serum and tissues in T2DM, and propose that strategies to increase an ailing miR-155 production in T2DM might be beneficial. The manuscript, though interesting, exceeds 1,500 words and contains 50 references. Therefore, before considering publication, the authors are kindly proposed to consider resubmitting the manuscript in the form of Minireview.

Answer: We thank the Reviewer for the comments. We leave it to the discretion of Editorial Office of the Journal to decide whether the manuscript will be accepted as a "Letter to the Editor" or as a "Minireview".

Reviewer #3:

Scientific Quality: Grade C (Good)

Language Quality: Grade B (Minor language polishing)

Conclusion: Major revision

Specific Comments to Authors: Thanks for the invitation for reviewing the paper. Major concerns:

1. As we see, author commented miRNA-155 based on Lopez review, however ,the correlation between the two is not very closed. For instance,"MiR-155 is of particular interest in the Lopez review as it is intricately involved in the pathogenesis of DM as well as in the regulation of AT1R and Ang II effects[6, 8-12]."No "MiR-155" was mentioned in Lopez review. This article can be reassembled into article type of review.

Answer: It was precisely the fact that there was no reference to miR-155 in the current review that led us to author a "Letter to the Editor". As miR-155 is pivotally involved in the pathogenesis of both types of Diabetes Mellitus (DM) and the regulation of AT1R and Ang II effects, that mediate DM's detrimental vascular actions, we considered it necessary to comment on it. Whether the current manuscript will be accepted as a "Letter to the Editor" or as a "Minireview", it is left to the discretion of the Editorial Office.

2. If the article type is letter, the author should focus how could "MiR-155" act as a modulator on ACEI or ARBs? And make the article more shorter. After thorough read, I can only get the correlation with Lopez review. here: "Lopez et al justifiably propose effective interventions through AT1R substrate modulation (ACEi) and/or receptor inhibition (ARBs) to improve glucose homeostasis[6]. Strategies to increase an ailing MIR-155 production in T2DM could prove to be a more appropriate course of action."

Answer: We have rewritten and slightly enlarged the section starting in line 224. We have added an additional reference (ref 51) that further supports the observation that ACEi/ARBs diminish miR-155 and references 52 and 53 regarding ACEi/ARBs' effects on CVD prevention and improvement of glycemic indices in DM and hypertension.

3. 2.The author mentioned miR-155"Minimally detected under normal physiological conditions and mainly expressed in the thymus and spleen " and "In T2DM, miR-

155 levels in plasma, peripheral blood cells, platelets, and urine are significantly and consistently decreased, with surprising congruence between different ethnicities". So how to dectect miR-155?

Answer: Adamkova[3] and Teng[4] state that miR-155 is low in healthy individuals, and its upregulation is generally associated with the activation of an innate immune response. What has been repeatedly described in T2DM is that miR-155 basal secretion and expression levels are consistently diminished in several tissues compared to healthy subjects[5, 6]. Moreover, circulating plasma/serum miR-155 evaluated by quantitative polymerase chain reaction (qPCR) can differentiate between healthy and T2 diabetic subjects[7, 8].

Minor concerns:

1."The syndemic of coronavirus disease 2019 (COVID-19) and T2DM has affirmed the latter's lethal effect". Why COVID-19 with T2DM became a latter's lethal effect. The affects of Renin-Angiotensin System on COVID-19 can be detailed. You can delete this sentence or move this part to discussion on relationship between miR-155 and diabetes.

Answer: The term Syndemics (as introduced by Singer[9] and expanded by the Lancet[10]), is defined as "a conceptual framework for understanding diseases or health conditions that arise in populations and that are exacerbated by the social, economic, environmental, and political milieu in which a population is immersed". Obesity has thus repeatedly been identified as a critical underlying cause of poor outcome in COVID-19, and when combined with other co-morbidities, impacts exponentially on mortality.[11] We feel that this connection represents a global challenge requiring research, public health, nutrition and educational approaches.

Regarding the effects of the RAAS in SARS-CoV-2, we believe this minireview is not the proper forum for its discussion. Reference 13 in the manuscript elaborates in detail the interplay between miR-155 and RAAS.

2."We suggest additional pathways that can modulate AT1R and Ang II effects that are of importance for the pathogenesis of IR, T2DM, and the development of cardiovascular and renal diabetic complications." This sentence can move behind "Since a particular miRNA may target one or many different mRNAs while one mRNA may bind many miRNAs, the host can modulate response feedback, through regulatory gene networks, in a concerted effort to control diverse aspects of cellular processes[7]. "like "additional pathways such as :miRNA can modulate..."

Answer: Amended as suggested.

Final comment This paper reviews "miR-155" from a new perspective that relationship between miR-155 and DM. As a "letter" type ,the content of paper could be more foucused and based on Lopez review. As a "review" type, the conclusion of the paper is not convinced. This paper need major revision.

Answer: we have amended the conclusion of the paper as suggested and we hope that it now adequately concludes our minireview.

References

1 Elton TS, Selemon H, Elton SM, Parinandi NL. Regulation of the MIR155 host gene in physiological and pathological processes. *Gene* 2013; **532**(1): 1-12 [PMID: 23246696 DOI: 10.1016/j.gene.2012.12.009]

2 Desvignes T, Batzel P, Berezikov E, Eilbeck K, Eppig JT, McAndrews MS, Singer A, Postlethwait JH. miRNA Nomenclature: A View Incorporating Genetic Origins, Biosynthetic Pathways, and Sequence Variants. *Trends in Genetics* 2015; **31**(11): 613-626 [DOI: 10.1016/j.tig.2015.09.002]

3 Adamcova M, Kawano I, Simko F. The Impact of microRNAs in Renin-Angiotensin-System-Induced Cardiac Remodelling. *Int J Mol Sci* 2021; **22**(9) [PMID: 33946230 PMCID: PMC8124994 DOI: 10.3390/ijms22094762]

4 Teng G, Papavasiliou FN. Shhh! Silencing by microRNA-155. *Philos Trans R Soc Lond B Biol Sci* 2009; **364**(1517): 631-637 [PMID: 19008191 PMCID: PMC2660923 DOI: 10.1098/rstb.2008.0209]

5 Corral-Fernández NE, Salgado-Bustamante M, Martínez-Leija ME, Cortez-Espinosa N, García-Hernández MH, Reynaga-Hernández E, Quezada-Calvillo R, Portales-Pérez DP. Dysregulated miR-155 expression in peripheral blood mononuclear cells from patients with type 2 diabetes. *Exp Clin Endocrinol Diabetes* 2013; **121**(6): 347-353 [PMID: 23616185 DOI: 10.1055/s-0033-1341516]

6 Khamaneh AM, Alipour MR, Sheikhzadeh Hesari F, Ghadiri Soufi F. A signature of microRNA-155 in the pathogenesis of diabetic complications. *J Physiol Biochem* 2015; **71**(2): 301-309 [PMID: 25929727 DOI: 10.1007/s13105-015-0413-0]

7 Akhbari M, Khalili M, Shahrabi-Farahani M, Biglari A, Bandarian F. Expression Level of Circulating Cell Free miR-155 Gene in Serum of Patients with Diabetic Nephropathy. *Clin Lab* 2019; **65**(8) [PMID: 31414764 DOI: 10.7754/Clin.Lab.2019.190209]

8 Polina ER, Oliveira FM, Sbruzzi RC, Crispim D, Canani LH, Santos KG. Gene polymorphism and plasma levels of miR-155 in diabetic retinopathy. *Endocr Connect* 2019; **8**(12): 1591-1599 [PMID: 31751306 PMCID: PMC6933831 DOI: 10.1530/ec-19-0446]

9 Singer M, Clair S. Syndemics and public health: reconceptualizing disease in bio-social context. *Med Anthropol Q* 2003; **17**(4): 423-441 [PMID: 14716917 DOI: 10.1525/maq.2003.17.4.423]

10 The L. Syndemics: health in context. *Lancet* 2017; **389**(10072): 881 [PMID: 28271823 DOI: 10.1016/s0140-6736(17)30640-2]

11 Hill MA, Sowers JR, Mantzoros CS. Commentary: COVID-19 and obesity pandemics converge into a syndemic requiring urgent and multidisciplinary action. *Metabolism* 2021; **114**: 154408 [PMID: 33080269 PMCID: PMC7831812 DOI: 10.1016/j.metabol.2020.154408]

Editor-in-Chief Review

JOURNAL EDITOR-IN-CHIEF (ASSOCIATE EDITOR) COMMENTS TO AUTHORS

Since this hypothesis is interesting, it should be given a hypothetic illustration to link these pieces of components in order to help the readers to well understand and keep it in mind. Therefore, I would suggest informing the authors for its acceptance with an opportunity to further enhance its potential impact by adding the illustration.

I have replied to the comment but as the system only allows one file upload, I have not been able to upload the manuscript file where the (Figure 1) insertions are seen and highlighted in yellow.

I therefore, attach here the manuscript file with the insertions and the figure 1 and legend file as well.

The insertions are as follows:

Page 5/21, in the paragraph starting "MiR-155 is of particular interest…" inserted (Figure 1) at the end of this sentence.

Page 6/21, in the paragraph stating "In T2DM, miR-155 Levels in plasma,…", inserted (Figure 1) (Table 1) at the very end of this paragraph.

Page 6/21, second paragraph, starting "Aging, obesity, sarcopenia,…" inserted (Figure 1) at the end of this sentence.

Page 7/21, mid-page, sentence starting "From the sum of these actions,…" inserted (Figure 1) (Table 1) at the end of this sentence.

Page 8/21, in the paragraph starting "AT1R substrate modulation (ACEi),…" inserted (Figure 1) (Table 1) at the end of the paragraph's second sentence.

Page 9/21, in the last sentence before CONCLUSION, inserted (Figure 1) at the end of this sentence.

Page 9/12, in CONCLUSION, first sentence, inserted (Figure 1) (Table 1) at the end of this sentence.

All insertions highlighted in yellow.