Peer Review File

ReviewA

Comment 1: As regards the AMI patients, there is no information at all as to how they were treqted, and in the case of the STEMI patients, how soon after the onset of chest pain. This could easily affect the results. There is also no information as to how soon after admission (and initial treatment) the blood samples were taken.

Reply 1: The MPS test the blood samples were taken before coronary angiography, Biochemical parameters of blood samples were collected within 24 hours of hospitalization. The STEMI patients need to emergency PCI, whose chest pain were within 12 hours.

Changes in the text: I will add "To avoid coronary angiography and its associated interventional effects on peripheral blood MPs levels, 2 mL peripheral blood was collected from all patients prior to coronary angiography and MP levels were subsequently detected" and "The STEMI patients need to emergency PCI, whose chest pain were within 12 hours" to the article.

Comment 2: The information about the control group, other than that they had been found to have normal coronary angiograms. Therefore they were not "normal controls" in any respect. We need to know whether they had valvular disease or chest pain: if the latter, they may have had coronary artery spasm, which is associated with endothelial damage. Comparisons between groups may be less than edifying, depending on who were the controls.

Reply 2: The normal controls have chest pain, but the pain is not associated with heart disease, so they had not coronary artery spasm.

Changes in the text: We add "coronary artery spasm" to exclusion criteria.

Comment 3: The manuscript is littered with grammatical errors, which must be corrected. There are also sentences which defy understanding (see line 57, for example). However, there also are factual mistakes. For example, in the Abstract, it is stated (WRONGLY) that EMP and RMP levels were higher in patients with single, than with multi-vessel disease.

Reply 3: I have carefully corrected them.

Changes in the text: The levels of EMPs and RMPs in multi-vessel were higher than in the single-vessel l disease.

Comment 4: It would have helped a great deal to correlate EMPs with peak troponin levels, or, better still, with plasma concentrations of other markers of endothelial

damage, such as Syndecan-1. The use of a series of univariate analyses to understand variability in MP concentrations is suboptimal: I would much prefer addition of a multivariate analysis.

Reply 4: We will take consideration of correlating EMPs with Syndecan-1 and adding multivariate analysis in future.

Changes in the text: No change

Comment 5: The finding that EMP concentrations are higher in patients with multivessel than single-vessel CAD is worthy of attention, I think. This may be evidence supporting multi-vessel inflammatory activation in paitnets with AMI, which provides the theoretical underpinning for PCI of all stenosed vessels during index admission. In this specific case, it might possibly be that signalling from MP to vessel is important

Reply 5: The future research will pay more attention to the EMPs of patients with multivessel artery.

Changes in the text: No change.

ReviewB

Comment 6: In the Materials and Methods section, please clarify in 2.3 first sentence: why is PFP stated here when above the method provided was for isolation of MPs that were then frozen prior to analysis. In the same paragraph, does 'dissolved' mean 'thawed'?

Reply 6: We have added detailed methods, the dissolved means thawed.

Changes in the text: Blood samples were centrifuged at 3 500 g at room temperature for 10 minutes to separate the bleeding cells, and the supernatant was platelet-rich plasma (PRP). PRP centrifuged a large number of platelets after centrifugation at 2 000 g and 10 ° C for 10 min, the supernatant was platelet-poor plasma (PPP), which followed by three centrifugations at 23,000 g at 4°C for 45 min. The supernatant was discarded, and the precipitate was platelet-free (PFP).

Comment 7: Explain the purpose of using the Ca vector A23187 and annexin V. Does this provide an assessment of 'function'? It is possible to assess MPs by the monoclonal antibodies alone without A23187 and annexin V to identify MPs present in the blood circulation of the patients. Please explain and clarify the method used in this study.

Reply 7: Ca vector A23187 promotes the release of MPs in PFP, annexin V adsorbs MPs

Changes in the text: No change

Comment 8: Were beads of standard size used to determine the proper gating for the MP region? Also how do you determine that the "MPs" were not just debris?

Reply 8: MPs are 0.1 to 1um in diameter, which have cell membrane phosphatidylserine and contained information like mRNA, microRNAs (miRNAs), receptor and specific proteins of parent cell

Changes in the text: No change

Comment 9: Give a more descriptive explanation of how the flow cytometric data was expressed. What was the numerator and denominator (10,000 counts, annexin V + ...) of the calculated % reported out? It is important to clearly state how the data was analysed in order for readers to compare to their own experimental systems.

Reply 9: The number of cells in the portal was 10000wh each time, reading at a flow rate of 35μl / min for 30 seconds, counting the number of EMPs and RMPs .The final peripheral endothelial cell- and red blood cell-derived MPs are expressed as percentages.

Changes in the text: The number of cells in the portal was 10000wh each time, reading at a flow rate of 35μl / min for 30 seconds, counting the number of EMPs and RMPs and analyzing the fluorescence percentage of endothelial cells and red blood cells labeled with specific monoclonal antibodies to further characterize EMPs and RMPs. The final peripheral endothelial cell- and red blood cell-derived MPs are expressed as percentages.

Comment 10: In paragraph 2.2 why are only the EMPs listed. Shouldn't this also state EMPs as well?
Reply 10: The same separation method to obtain EMPs and RMPs.
Changes in the text: No change

Comment 11: In the Discussion several statements are made that should be referenced and defined as opinion or as validated mechanisms. See lines 162 (are they really procoagulant?), 167 (why state 'rarely released'?), and 168 (reference the atherosclerosis mechanism).
Reply 11: EMPs have the procoagulant function according to the research(16), the state should be released,
Changes in the text: we delete the "rarely".

Comment12: What is meant by 'distal perfusion", line 170?
Reply 12: Distal perfusion means circulating blood.
Changes in the text: Recent studies have shown that RMPs are released into the circulating blood due to worsening thrombosis conditions and can be detected in patients with STEMI.

Comment 13: Line 182, the MPs could also be an 'innocent bystander', the result of some other more direct acting mechanism.
Reply 13: The results suggest that EMPs and RMPs may be involved in AMI and may be correlated with several coronary artery diseases
Changes in the text: No change.

Comment 14: The significance/relevance of Figure 1 is difficult to understand without any reference point for comparison. It may be better to eliminate this figure unless there

is a particular reason to include it.

Reply 14: we change the title of Figure 1

Changes in the text: TEM show the structure and size of MPs

Comment 15: Define all abbreviations. In particular, what is cTNT line 94 and TC line 130 and Table 1?

Reply 15: we should define all abbreviations.

Changes in the text: we have changed those.

Comment 16: Some minor English grammar corrections are needed in the paper.

Reply 2: we should pay more attention to English grammar

Changes in the text: we have changed the English grammar errors.