Response to reviewers

Reviewer 1

The author of this manuscript attempted to review the current cellular pathophysiology of neurodegeneration in AD and as potential platforms for drug discovery, and focus on the specific role of mitochondrial dysfunction in AD. However, at present, there are still some concerns need to be carefully addressed before acceptance, the comments are as follow:

Major concerns

- **I think the topic of this manuscript should be mainly focused on iPSC-based mitochondrial dysfunction in AD, while, from this manuscript, there are 6 pages of words describing the mechanism of AD progression, but only less than 5 pages were used to review the iPSC-based research, so, I think the author should make a little bit deduction of words that been used in the initial pages so that give you enough space to describe the development of iPSC-based AD research.**

- We have now changed the title to reflect the emphasis of the review (which is primarily focused on mitochondrial dysfunction in AD and secondarily focused on modelling this in iPSCs), to 'Modelling mitochondrial dysfunction in AD using human iPSCs.'

- **From table 1, the author only summarised the findings that the reference shown, however, the comparison, or advantage and disadvantage of these different references summarize will be benefit for the readers and will give them new directions or avenues for future research. Moreover, I think you can address the different protocols generated neurons for the AD research may have some differences, in other words, human neurons generated from different groups may vary, so, some of the phenotype of the neurons may vary, which may lead to different mechanism investigation have some difference. Please mention them in the table.**

- We have now added advantages and disadvantages columns into **Table 1.** However, we do not see an obvious way to add the different protocols/neuronal phenotypes concisely into this table. Instead we have added a sentence highlighting this important point that different neuronal differentiation protocols may lead to different neuronal phenotypes and therefore different mechanistic findings **(page 11 lines 9-11).**

- **Brain organoid have already been developed by some research groups, and the author have already show some references that organoid had been used for research, I think the authors should separate them as individual part so that you can highlight them the difference between iPSC-derived neurons and brain organoid based mechanism investigation.**

- We have now added a heading entitled, 'AD modelling in 3D culture systems,' which describes various studies that have used iPSC-derived organoids (and other types of neurospheres) to model AD **(page 12 line 18)**.

- **As you may know, the toxicity of tau can be replicated by recombinant pre-formed fibrils, I think you can summarise them with iPSC-derived neurons or brain organoid so that expand the view of current iPSC-based AD research.**

- We have now included some references to work exploring the effects of pre-formed tau fibrils applied to iPSC-derived neurons to model AD **(page 9 lines 6-10)**.

- **The author should provide a clear mechanism of neuron death in AD, although the author have mentioned that there are two mainly pathway, amyloid and mitochondrial cascade, I think the best way is the figure that you can use to clearly show the potential pathway, the upstream and downstream, the mediators, and the potential result of different pathways, and then omit some words for the iPSC-derived neurons development description.**

- **Figure 3** shows a mechanism proposed previously by our group whereby Aβ may cause mitochondrial dysfunction in astrocytes and consequently death in neurons. We have now highlighted the part of this figure that relates to the amyloid hypothesis and added a part to the figure that summarises how the mitochondrial hypothesis fits into this.

Minor concerns

- **From the abstract, I think the author omit some words which belong to the introduction part but not fit for the abstract, the abstract should be briefly summarised that iPSC-based AD mechanism research.**

- We have now rewritten the abstract, ensuring that it covers the key points about iPSC-based AD mechanism research.

- **The author should mention some advantages and disadvantages of iPSC-based research and non iPSC-based AD research, so that give some information to readers to make a decision to choose the ideal model or tools for their research.**

- We have included a section in the review entitled, 'The need for better model systems' that describes the advantages and disadvantages of iPSC model systems compared to other AD model systems. We have now summarised this in an additional table **(Table 2).**

Reviewer 2

This review is excellent, compactly summarising the current understanding of the etiology of AD and the efforts and trials for iPSC-based drug discovery for the treatment of AD. This review will contribute to deepening our understanding of the significance of iPSCs to uncover the pathophysiology of diseases of unknown etiology.