Firstly, thank you for all the reviewers for their valuable suggestions and questions. I would like to answer them in order. All the added contexts are highlighted in purple.

Reviewer #1:
Scientific Quality: Grade B (Very good)
Language Quality: Grade B (Minor language polishing)
Conclusion: Minor revision
Specific Comments to Authors: The authors demonstrated about high-throughput screening (HTS) to enable to establish new and fast developing technology for the facilitation of in vitro neurogenesis using stem cells and organoids. It is very important technology to study the regulation of disease-mediated circumstance and the development of new drug. This manuscript is written in a well-organized format and covers everything from the description of the HTS system to the in vitro culture system (organoid) and microfluidic device related to neurogenesis. If some minor issues are resolved, it would be good to be published in WJSC.

1. In chapter of "Current in vitro neurogenesis methods", the authors described stem cell differentiation to generate neuronal cells from embryo to adult. However, Figure 1 does not match the description. From the point of view for organoid production, it would be good choice to focus on the neurogenesis of PSCs and write the text. And, the authors should add references to compose Figure 1 to the figure legend.
A: In the organoid generation period, we added the methods of how cerebral organoids are generated from PSCs in line 236-240; we have also added several references in the figure legends of Figure 1.

2. The authors focused on the neural organoid functioning neurogenesis with 3D culture system, and these contents was displayed in Ref. 77 to 87. Organoids induced to a specific part of the brain have different characteristics (induction method, time, conditions, etc.), however the authors listed only the types of brain organoid in parallel without specific mention.
A: This is a very good suggestion and we have added the specific method of generating brain organoids with different regions in line 246-255. In specific, the induction methods are similar to the way of generating function neurons with small molecules and morphogens, as they are also widely used in producing cerebral organoids. For example, when generating midbrain organoids, Shh and FGF8 are important factors for inducing midbrain identity; and dual-SMAD signaling inhibition is a common way for brain organoid generation with PSCs. Significantly, the concentration gradients of Wnt and Shh signaling (using different small molecules as agonists) play important roles for definition of different brain regions in organoids, which are based on the neural pattern mechanism in vivo during embryonic neurogenesis.

Reviewer #2:
Scientific Quality: Grade B (Very good)
Language Quality: Grade B (Minor language polishing)
Conclusion: Major revision
Specific Comments to Authors: I would like to thank the authors for their well written topic. The

manuscript is written with concentrating on the technical point of view, and in my opinion would benefit more and increase the broad-spectrum of readers if a clinical and medical background and introductory paragraphs before the applications part could be added, as this is a part of integrated medicine. English editing certificate is not present, only the manuscript file is uploaded under the name of the required certificate. English language needs revision for grammatical and syntax corrections.

#Title: the word " Prospection" is not used in the right meaning, could the authors use prospects instead? This applies to the whole document.

A: We have revised the title to "Application and Prospects of High-throughput Screening for In Vitro Neurogenesis".

#Introduction: 1- The authors wrote: "Therefore, categories of testing candidates are also developing from molecular aiming at diverse">> could they elaborate molecular what? did they mean molecular markers?

A: We have revised the word "molecular" to "biochemicals", indicating drugs and other biomolecules which could interact with receptors or enzymes in cells.

2- I recommend adding this reference: Azari, H., & Reynolds, B. A. (2016). In Vitro Models for Neurogenesis. Cold Spring Harbor perspectives in biology, 8(6), a021279. https://doi.org/10.1101/cshperspect.a021279 3- Only one reference is from 2021, out of 211 references this seems a very low number, in addition there was a lot of progress last year too in the publication in this area. examples to last year publications: A. Shin, H., Jeong, S., Lee, J. H., Sun, W., Choi, N., & Cho, I. J. (2021). 3D high-density microelectrode array with optical stimulation and drug delivery for investigating neural circuit dynamics. Nature communications, 12(1), 492. https://doi.org/10.1038/s41467-020-20763-3 B. Lam D, Fischer NO, Enright HA. Probing function in 3D neuronal cultures: A survey of 3D multielectrode array advances. Curr Opin Pharmacol. 2021 Oct;60:255-260. doi: 10.1016/j.coph.2021.08.003. Epub 2021 Sep 1. PMID: 34481335.

A: We have added those references listed above and also added other references from 2021 and 2022, especially in the "Applications of induced neuronal cells" section.

#Figures: Figures are well-drawn and self explanatory. However, they sometimes lack explanation of the abbreviations. Also, why did the authors chose certain and specific markers in figure 1 while the comment on the figure is relatively a general comment?, please modify, if you want to use a general model do not specify the genetic markers or add the work (for example).

A: We have added the abbreviations in figure legends and changed the figure 1 to the general model in the revised version of Figure 1.

The subtitle " Prospection Developing organoids/spheroids-based HTS system: " . I found the authors using a medical term that is known mainly in psychology, could they redefine the term in light of neurogenesis research or use another term?

A: I am sorry but we did not understand or find out the medical term which is known mainly in psychology, could you please point out that word and we could explain or modify it?

Thank you.

The authors did not explain the concept of "the conversion efficacy" in the text, only in the supplementary table, and this is an important outcome of the research topic in question, and should be explained clearly in the text, along with the research limitations resulting from it, and how to overcome those limitations.

A: We have added the "Limitations and Prospects" section and explained the conversion efficiency and listed some methods to overcome the limitations, including relacing transcription factors to small molecules, activating endogenous loci, and ablating non-neuronal cells selectively and controllably for higher conversion efficiency, in line 835-859.

Overall: I think the authors should explain that 3D brain structures act as "Microphysiological systems (MPS)" to recapitulate the brain physiology, and discuss more the clinical impact of this approach in the text, along with the possible understanding of the pathophysiology of some neurological effects of drugs or diseases. The clinical background is minimally explained in the text, with concentrating on the technical parts of the topic. I think clinicians could benefit more of this review if its impact and benefits on clinical research were clearly delineated. Even in the part " Applications of HTS on neurogenesis" the authors preferred to explain the technical difficulties that could be encountered rather than explaining the clinical impact on the medical research field or why applications is needed in the first place instead of the real world cases studies. Could the authors kindly modify their text? I suggest that after each application the authors add a subtitle "limitations" and explain the limitations in this area instead of in the application part for the presentation to be more clear.

A: Thank you for this suggestion and we have added the "Applications of induced neuronal cells" section (line707-833) and talked about various about the application of the generated induced neuronal cells. Firstly, they can be used for regenerative medicine, especially stem cell therapy to treat NCS diseases and injuries through the transplantation of neural stem cells. We also discussed the clinical trials of neural stem cell therapy for treating neurodegenerative diseases and injuries. Then, the induced neural stem/progenitor cells can be applied to neural tissue engineering, which is also becoming a promising way to treat CNS diseases, especially spinal cord injury. Now the combination of neural stem/progenitor cells and 3D scaffolds, especially collagenI, are gradually appearing in clinical studies. Other applications such as 3D modeling of neurodegenerative diseases and neural development, are also common as the induced functional neurons can mimic the biological characteristics in vivo and allowed for investigation of the mechanisms of diseases and developmental process.

We also added the "Limitations and Prospects" part and discussed the limitations of in vitro neurogenesis and HTS platforms, including the development of organoid-based HTS platform and the possibility of developing microfluidic-based HTS platform, in line 835-935.

Reviewer #3:
Scientific Quality: Grade B (Very good)
Language Quality: Grade B (Minor language polishing)

Conclusion: Accept (General priority)

Specific Comments to Authors: This is an in-depth review of the current state and prospects of HTS in in-vitro neurogenesis. It will be certainly used as a reference to those that want to engage the field. I have corrected some minor wording and language issues as suggested in the file.

A: Thank you for revising and we have also edited the language through the help of American Journal Experts (AJE).