

## PEER-REVIEW REPORT 1

**Name of journal:** Neural Regeneration Research

**Manuscript NO:** NRR-D-18-00596

**Title:** Adult neural stem cell plasticity

**Reviewer's Name:** Kyle D. Fink

**Reviewer's country:** USA

**Date sent for review:** 2018-08-27

**Date reviewed:** 2018-09-10

**Review time:** 14 Days

Manuscript Rating Question(s):	Scale	Rating
The subject addressed in this article is worthy of investigation. (3 as the best score)	[1-3]	2
The information presented was new. (5 as the best score)	[1-5]	4

### COMMENTS TO AUTHORS

The review paper discusses stem cell plasticity, specifically describing how current in vivo and in vitro methods can be utilized to analyze the plasticity of neural stem cells. The chimera assay provides an in vivo method for determining if transplanted cells exhibit plasticity and can therefore contribute to the development of an animal. However, evaluating NSC plasticity with the chimera assay has yielded differing results when tested in mouse, chick, porcine and zebrafish models. In vitro methods, which co-culture adult stem cells with stem cells or factors of a specific tissue can also be used to determine the plasticity of NSCs or NPCs. Recent studies suggest that co-culturing mouse NPCs and skin cells directs an ectodermally-restricted conversion of NPCs to keratin1-positive cells. Method optimization is necessary in order to determine if NSCs have plasticity in vitro and in vivo and if NSC plasticity is relevant for therapeutic applications.

#### Strengths:

- Describe different in vivo and in vitro models for evaluating plasticity.
  - o Include previous literature which evaluated NSC plasticity in different animal models (mouse, chick, porcine and zebrafish) as well as the strengths and weaknesses to using the animal model.
- In the discussion, provide context for why optimizing the in vivo and in vitro plasticity methods is necessary. Also, provide support for why identifying an efficient way to derive, define and culture NSCs before transplantation is important.

#### Weaknesses:

Clarity, missing supporting information or did not completely describe a cited papers results.

The paper would also benefit from NSC plasticity diagrams/graphics.

- Page 1 at line 14 (Problem: clarity)
  - o "With the ability to create induced pluripotent stem cells from somatic cells now available, the properties of multipotent stem cells are being reevaluated." Is this because those iPSCs are then

differentiated into multipotent stem cells, which allows researchers to study the properties of multipotent stem cells?

- Page 1 at line 19 (Problem: clarity)

o "Advancements in biotechnology now allow for better methods to investigate stem cell plasticity, such as the relative influence of external versus intrinsic factors on cell fate, and to therapeutic transplantation of NSCs."

o The end of this sentence doesn't connect well with the beginning, which makes the overall messaging confusing. My edit: "Advancements in biotechnology now allow for better methods to investigate stem cell plasticity, such as the relative influence of external versus intrinsic factors on cell fate, and to determine the therapeutic potential of transplanting NSCs."

- Page 1 at line 31 (Problem: missing supporting information)

o I think what is missing is why NSCs might be a better alternative for creating rare cell types. The authors mention that NSCs would be better than ESCs (I'm assuming they are referring to ethical issues), but do not provide a clear reason why, and also do not mention why it would be better than iPSCs (difficult to create, expensive, time consuming).

- Page 1 at line 55 (Problem: clarity)

o "...the cells in question are transplanted into a developing embryo..." what cells are transplanted? Embryonic stem cells? adult stem cells?

- Page 2 at line 17 (Problem: missing supporting information/did not completely describe a result)

o "An in vitro version of this assay.....or are co-cultured with embryonic stem cells and grown for a limited time." Is the in vitro method more or less effective than the in vivo method for determining plasticity? The authors could include a concluding statement in this paragraph that compares the in vivo and in vitro method.

- Page 2 at line 28 - 38 (Problem: missing supporting information)

o "These methods allow for controlled administration of factors which are known to promote differentiation of a particular fate." I think a specific example would be beneficial and highlight what type(s) of adult stem cells this has been successful with. Since the paper mainly focuses on NSCs, the authors could speak to how NSC potency is affected by co-culture with mature neurons/glia cells....Although moving the sentence at line 28 on Page 4 could also work since it discusses NPC co-culture with mouse skin cells.

- Page 4 at line 14 (Problem: missing supporting information)

o "Similar to the results in mice, findings of plasticity following xenotransplantation into zebrafish are variable." What were the findings in zebrafish and how did they vary?

- Page 4 at line 48 (Problem: did not completely describe a cited paper's results)

o "...transplanted cells retained their neural phenotypes despite their locations outside the central nervous system." What neural phenotypes did the authors look at?

- Page 5 at line 9 (Problem: clarity)

o "They found that adult- and fetal- derived cells recapitulated the hematopoietic system." Not sure if the authors are saying that the NPCs were found in the hematopoietic system and/or performed the function of resident hematopoietic cells.

- Page 5 at line 16 (Problem: missing supporting information & reorganize?)

o "Neural progenitor differentiation into skeletal muscle cells has been demonstrated after transplantation into regenerating muscle of adult mice." Why/how does this occur?" Some support for this statement is on Page 2 in paragraph 3.

- Page 8 at line 6 (Problem: more information)



o "The use of stem cell plasticity for therapeutic application may be considered." Therapeutics for what?

Other comments:

Review paper might benefit from reorganization (one example listed below).

- Describing what the chimera assay is (currently Page 1 at line 54) and then subsequent paragraphs which focus on how adult NSC plasticity was evaluated in the assay (currently Page 3 at line 7).

Overall, the paper is interesting and highlights an important stem cell property. The paper would benefit from editing sentence structure and providing additional information to support the content.