**Stem Cell Utilization in Dentistry**

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**Key Words: Stem cell, regenerative medicine, dental pulp stem cells**

**Abstract:**

 **Stem cells research is in its infancy. There are many possible uses yet to be refined and its applications have the potential to be nothing short of extraordinary in the field of medicine. It is the beginning of what is being called ‘regenerative medicine’, which is the use of stem cells to restore tissues in the body. More specifically, dental pulp stem cells have a great capacity for this regenerative medicine in dentistry. They have the potential to restore structural defects, regenerate bone, and heal damaged tissues [1]. However, the lineage of these stem cells and their different subpopulations is relatively unknown. This study seeks to analyze the different subpopulations and their specific regenerative capacities so that dental pulp stem cells might be able to be used in a more effective manner. From this research, it was found that the subpopulation LNGFRLow+THY-1High+ cells had vastly enhanced regenerative capabilities than the other subpopulations of human dental pulp stem cells [2]. Great progress can be made from these findings as it sheds light on how to more effectively take advantage of the regeneration of dental pulp stem cells. It also raises many new questions as to what sets this subpopulation apart and allows it to be so much more effective.**

**Introduction**

Regenerative medicine is quickly reaching new prominence in today’s research, and none more prominent than the use of stem cells. Stem cells are cells capable of becoming many different types of cells, that is, they do not have a specific cell type they are destined to take the form of after development. This flexibility allows them to be utilized in a variety of fashions, and regenerative medicine has arose as a result. Regenerative medicine is the process of replacing or regenerating human cells, tissues, or organs for therapeutic applications [1]. With stem cells being able to become many different types of cells, they can play a critical role in regenerating degraded tissue. Stem cell uses have been especially prevalent in the field of dentistry. Researchers have in fact been able to successfully regenerate tooth structure and dental tissues with dental stem cells in animal studies, but it has yet to be fully developed in humans [1].

There are many applications for dental stem cells, and more specifically, dental pulp stem cells have been found to have vast potential in regenerative medicine (these are often referred to as human dental pulp/progenitor cells (hDPSCs)) [2]. These cells are able to generate both endothelial cells and osteoblasts [2]. That is, they are able to differentiate into dentin, dentin-pulp-like complex (essential parts of tooth structure), and into bone which is why they have potential to be quite useful in the dental field [1]. Not only do hDPSCs have these uses, but they also are relatively easy to surgically extract and can be safely recombined with many materials used to build up tooth structure [1]. Regenerating bone and tooth structure would eventually allow for replacement of filling material as well as the use of crowns. This would allow dental patients to retain material made by their own body and would prevent them from having foreign products permanently inserted into their mouths.

**Recent Progress**

Many of the problems that have arose in attempt to use hDPSCs in the dental field have been in regards to their attachment on plastic materials and tooth structure in the mouth. hDPSCs are able to be used for regenerative therapy because they can expand to become colony-forming unit fibroblasts which are able to adhere to plastic and tooth structure and can generate the large amount of cells needed for proper regeneration and transplantation [2]. Unfortunately, when they attach to these other surfaces, their phenotype often changes in response. This is problematic because the desired tooth structure and tissue will not be properly regenerated. Due to this problem, researchers have worked to isolate purified hDPSCs through identification of selective markers that result from hDPSCs so that specific subpopulations of hDPSCs could be tested to see if any of these subpopulations could be more effective in regenerative therapy in dentistry. The markers chosen were low-affinity nerve growth factor receptor (LNGFR) and THY-1 (Thymocyte differentiation antigen 1) [2]. These markers have been used before to isolate neural stem cells from mammalian fetal peripheral nerves, but this is the first time they were used to see if they could isolate hDPSCs from dental pulp tissue.

 The tissue from which the testing was done came from human donors and consisted of impacted molars. The LNGFR and THY-1 were successfully isolated by using anti-LNGFR and anti-THY-1 antibodies along with flow cytometry [2]. Flow cytometry uses lasers in conjunction with the florescence that the anti-LNGFR and anti-THY-1 antibodies contain. The system detects when the laser makes contact with florescence and uses that as the basis to determine if LNGFR or THY-1 are actually present or not and determine their amounts [2]. From this, five different human dental pulp subpopulations were isolated and obtained, these being LNGFR+THY-1+ cells, LNGFRLow+THY-1High+ cells, LNGFR-THY-1Low+ cells, LNGFR-THY-1- cells, and LNGFR+THY-1- cells [2]. All of these subpopulations of human dental pulp stem cells were tested to see if their clonogenicity was effective. In addition all of these were tested to see if there were any in particular that would be more effective in regenerative therapy than the hDPSCs containing all subpopulations.

Figure 1A: Visual representation of the cloning capabilities of hDPSC subpopulations LNGFRLow+THY-1High+ and LNGFR+THY-1+

 It was found that the subpopulations LNGFRLow+THY-1High+ and LNGFR+THY-1+ had the greatest cloning abilities of the five subpopulations (figure 1A). Their capabilities

of successful use in dental regeneration were also tested, engrafting them into calvarial defects. LNGFRLow+THY-1High+ was found to have the longest time of cell survival when compared to LNGFR+THY-1+, and additionally it was found to generate new bone in the engrafted site, which was determined by confocal microscopy analysis [2]. The use of LNGFRLow+THY-1High+ was then compared to bone regeneration by the standard hDPSCs. With hDPSCs, there was bone regeneration, but it was sparse, and as expected, full adherence was problematic as well. However, when replaced with LNGFRLow+THY-1High+ cells high levels of wound healing and bone regeneration took place, indicating LNGFRLow+THY-1High+ cells from hDPSCs are more effective than the hDPSCs in adherence to surfaces and proper regeneration [2]. So that being said, this study provided evidence that the problems that have arose from using hDPSCs in regenerative therapy is because they contain five different subpopulations, and only one of which (LNGFRLow+THY-1High+) is fully effective. For example, previous studies done testing regenerative therapy did not have this specific subpopulation of hDPSCs isolated so the regenerative capacity was not in its purist form [2]. It is not known if the other subpopulations inhibit the LNGFRLow+THY-1High+ cells from taking full advantage of their osteogenic capabilities, but from this study, it was found that using LNGFRLow+THY-1High+ cells as opposed to the hDPSCs containing all subpopulations, was much more effective in regeneration.

**Discussion**

 This study has provided further understanding to the composition of hDPSCs, and has provided evidence to how they can be more effectively used in regenerative therapy. The lineage of hDPSCs had not been looked at, and this study sought to accomplish just that. Through the study of its lineage, subpopulations of hDPSCs were able to be identified and further analyzed. From this analysis, cells derived from hDPSCs with better regenerative capacities were found, specifically the subpopulation LNGFRLow+THY-1High+. Results presented from the study were mostly valid, as all subpopulations had the same tests performed on them testing their regenerative abilities. However, only the subpopulations with the greatest regenerative abilities (LNGFRLow+THY-1High+ and LNGFR+THY-1+) were chosen to do further testing on their therapeutic potential. This certainly does not invalidate their results, but their results could be further solidified had analysis besides just cloning abilities been performed on all subpopulations. That being said, their results showed increased validity in that the LNGFRLow+THY-1High+ and LNGFR+THY-1+ subpopulations were not just analyzed for their short term osteogenic capabilities, but also long term, as long term is what would actually be applicable to regenerative medicine. By analyzing over a longer period of time, and revealing that the LNGFRLow+THY-1High+ cells still are able to survive and result in new bone formation for over four weeks, the study is further impactful and may indeed have effects on how regenerative medicine moves forward. However, questions still need to be answered in regards to what the distinguishing factor of LNGFRLow+THY-1High+ cells is which allow them to regenerate bone better than the hDPSCs, and why the other subpopulations are largely unsuccessful.

**References**

[1] Jain, Aditya, and Ramta Bansal. “Current overview on dental stem cells applications in regenerative dentistry.” *Journal of Natural Science, Biology and Medicine*, vol. 6, no. 1, 2015, p. 29., doi:10.4103/0976-9668.149074.

[2] Yasui, T., et al. “Purified Human Dental Pulp Stem Cells Promote Osteogenic Regeneration.” *Journal of Dental Research*, vol. 95, no. 2, 2015, pp. 206–214., doi:10.1177/0022034515610748.