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Induced pluripotent stem cells (iPS cells) generated from somatic cells through reprogramming hold great promises for regenerative medicine. However, how reprogrammed cells survive, behave in vivo, and interact with host cells after transplantation still remains to be addressed. There is a significant need for animal models that allow in vivo tracking of transplanted cells in real time. In this regard, the zebrafish, a tropical freshwater fish, provides significant advantage as it is optically transparent and can be imaged in high resolution using confocal microscopy. The principal goal of this study was to optimize the protocol for successful short-term and immunosuppression-free transplantation of human iPS cell-derived neural progenitor cells into zebrafish and to test their ability to differentiate in this animal model. To address this aim, we isolated human iPS cell-derived neural progenitor cells from human fibroblasts and grafted them into (a) early (blastocyst)-stage wild-type AB zebrafish embryos or (b) 3-day-old Tg(gfap:GFP) zebrafish embryos (intracranial injection). We found that transplanted human neuronal progenitor cells can be effectively grafted and that they differentiate and survive in zebrafish for more than 2 weeks, validating the model as an ideal platform for in vivo screening experiments. We conclude that zebrafish provides an excellent model for studying iPS cell-derived cells in vivo.

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The use of autologous (or syngeneic) cells derived from induced pluripotent stem cells (iPSCs) holds great promise for future clinical use in a wide range of diseases and injuries. It is expected that cell replacement therapies using autologous cells would forego the need for immunosuppression, otherwise required in allogeneic transplantations. However, recent studies have shown the unexpected immune rejection of undifferentiated autologous mouse iPSCs after transplantation. Whether similar immunogenic properties are maintained in iPSC-derived lineage-committed cells (such as neural precursors) is relatively unknown. We demonstrate that syngeneic porcine iPSC-derived neural precursor cell (NPC) transplantation to the spinal cord in the absence of immunosuppression is associated with long-term survival and neuronal and glial differentiation. No tumor formation was noted. Similar cell engraftment and differentiation were shown in spinally injured transiently immunosuppressed swine leukocyte antigen (SLA)-mismatched allogeneic pigs. These data demonstrate that iPSC-NPCs can be grafted into syngeneic recipients in the absence of immunosuppression and that temporary immunosuppression is sufficient to induce long-term immune tolerance after NPC engraftment into spinally injured allogeneic recipients. Collectively, our results show that iPSC-NPCs represent an alternative source of transplantable NPCs for the treatment of a variety of disorders affecting the spinal cord, including trauma, ischemia, or amyotrophic lateral sclerosis.

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