Co-Culture of Neonate Mouse Spermatogonial Stem Cells with Sertoli Cells: Inductive Role of Melatonin following Transplantation: Adult Azoospermia Mouse Model

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Abstract : We have recently reported that melatonin as antioxidant enhances the efficacy of colonization of spermatogonial stem cells (SSCs). Melatonin as an antioxidant plays a vital role in the development of SSCs in vitro. This study aimed to investigate evaluation of sertoli cells and melatonin simultaneously on SSC proliferation following transplantation to testis of adult mouse busulfan-treated azoospermia model. SSCs and sertoli cells were isolated from the testes of three to six-day old male mice. To determine the purity, Flow cytometry technique using PLZF antibody were evaluated. Isolated testicular cells were cultured in αMEM medium in the absence (control group) or presence (experimental group) of sertoli cells and melatonin extract for 2 weeks. We then transplanted SSCs by injection into the azoospermia mice model. Higher viability, proliferation, and Id4, Plzf, expression were observed in the presence of simultaneous sertoli cells and melatonin in vitro. Moreover, immunocytochemistry results showed higher Oct4 expression in this group. Eight weeks after transplantation, injected cells were localized at the base of seminiferous tubules in the recipient testes. The number of spermatogonia and the weight of testis were higher in the experimental group relative to control group. The results of our study suggest that this new protocol can increase the transplantation of these cells can be useful in the treatment of male infertility. **Keywords :** colonization, melatonin, spermatogonial stem cell, transplantation

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