The fate and role of bone graft-derived cells after autologous tendon and bone transplantation in the bone tunnel

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Introduction: Successful anterior cruciate ligament (ACL) reconstruction with autologous tendon graft requires the solid integration of graft at the bone-tendon interface. It has been reported that ACL reconstruction with autologous tendon and bone transplantation in the bone tunnel provided an increase of stiffness of grafted tendon just after the transplantation and promoting integration of grafted tendon at the bone-tendon interface. However, the behavior and biological role of the bone graft-derived cells in the process of remodelling of grafted tendon in the bone tunnel has not been elucidated. We hypothesized that understanding the fate of bone graft-derived cells should be the pilot data to engineer the remodelling of the bone tunnel for regenerating the normal ACL properties.

We newly developed a transplantation model with green fluorescent protein (GFP) transgenic rats for following the bone graft-derived cells. GFP has no immunological rejection in vivo and thus this transplantation model theoretically simulates autologous transplantation. The grafted tendon cells are alive initially after the autologous tendon transplantation, but gradually become extinct and are replaced by host cells. The purpose of this study was to follow the bone graft-derived cells in the bone tunnel after autologous tendon and bone transplantation and to elucidate the fate and role of bone graft-derived cells during the remodelling process of grafted tendon.

Materials and Methods: Twelve-week-old genetically identical female green fluorescent protein (GFP) transgenic rats (n=30) and wild-type rats (n=30) were used. A 3 mm in diameter bone tunnel was drilled from the intercondylar notch to the lateral epicondyle of the right femur of wild-type rats. Achilles tendons were harvested from wild-type rats and bone grafts were harvested from lateral condyle of GFP rats. GFP positive bone grafts were transplanted into the bone tunnel of wild-type rats (A). At 1, 2, and 4 weeks after transplantation, distal femoral epiphyses were harvested and cut in to 14 um serial coronal frozen sections. The sections were then stained with hematoxylin and eosin (HE) and examined with a confocal laser scanning microscopy (LSM 510, Zeiss) to quantify GFP positive bone graft-derived surviving cells.

Results: At 1 week, a large number of GFP positive bone graft-derived cells were found in the interface between the host bone and the grafted tendon and no GFP positive bone graft-derived cells were found in the grafted tendon (B). At 2 weeks, GFP positive bone graft-derived cells were decreased in the interface between the host bone and the grafted tendon but GFP positive bone graft-derived cells were found in the grafted tendon (C). At 4 weeks, no GFP positive bone graft-derived cells were found in the interface between the host bone and the grafted tendon but a few fibroblast-like GFP positive bone graft-derived cells were found in the grafted tendon (D).

Discussion: In this study, a newly developed transplantation model with GFP transgenic rats was useful to follow the bone graft-derived cells after autologous tendon and bone transplantation in the bone tunnel.

At 1 week after the transplantation, a large number of GFP positive bone graft-derived cells were found in the interface between the host bone and the grafted tendon. This means that the bone graft-derived cells are alive until 1 week after the transplantation. At 2 weeks, GFP positive bone graft-derived cells migrated into the grafted tendon. At 4 weeks, GFP positive bone graft-derived fibroblast-like cells were found in the grafted tendon. Therefore, it was conceivable that bone graft-derived cells participated in remodeling of the grafted tendon.

In conclusion, bone graft-derived cells initially survive in the bone-tendon interface and in the proper of grafted tendon. The further examination of the role or character of the bone graft-derived cells is needed for developing the novel strategy to promote the remodelling of the grafted tendon in the bone tunnel.