## Effect of Current Osteochondral Allograft Harvesting Techniques on Regional Chondrocyte Viability

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Introduction: Chondrocyte viability directly correlates with the clinical success of osteochondral allograft (OCA) transplantation. It is not known how OCA plug harvest with a powered reamer affects the chondrocyte viability in a human model, or how to best mitigate potential thermal damage. The purpose of this study was to investigate the effect of distal femoral OCA plug harvest and recipient site preparation on regional cell viability when using handheld saline irrigation versus saline submersion.

**Methods**: Thirteen clinical-grade femoral hemicondyles (JRF Ortho) were used in this study. For each condyle, four samples were harvested: (1) 5 mm control piece of cartilage, (2) 15 mm OCA donor plug harvested with a powered coring reamer and concurrent handheld saline irrigation ("Traditional"), (3) 15 mm OCA donor plug harvested while submerged under normal saline ("Submerged"), and (4) 5 mm cartilage from the peripheral rim of a recipient socket created with a 15 mm cannulated counter bore reamer to a total depth of 7 mm ("Recipient").

Samples were stained using Calcein and Ethidium. Live-dead images were acquired using a confocal microscope (**Figure 1**). ImageJ was used to calculate live cell percentages for the whole plugs. Additionally, the 15 mm diameter plugs were divided into the central 5 mm and the peripheral 5 mm for comparison of regional cell viability. The live/dead cell percentages were compared between all groups using Friedman tests. Cell viability was determined to be significantly different if P < 0.05.

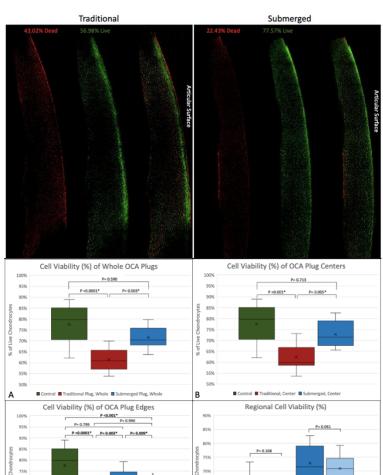
**Results**: Compared to the Submerged group, Traditional plugs had significantly lower percentages of live cells across the whole plug ( $61.42\% \pm 4.98\%$  vs.  $71.54\% \pm 4.82\%$ ; P=0.003), in the plug center ( $62.38\% \pm 6.11\%$  vs.  $72.76\% \pm 5.86\%$ ; P=0.005), and the plug edges ( $60.91\% \pm 4.75\%$  vs.  $70.93\% \pm 4.51\%$ ; P=0.003)

The Traditional plugs had significantly less live cells in all regions compared to Control (77.51%  $\pm$  9.23%; P<0.0001 for all regions). There were no significant differences in cell viability between Control and Submerged plugs (whole: P=0.590; center: P=0.713; edge: P=0.799) (**Figure 2A-C**). There were no intra-group regional differences between the central and peripheral 5 mm plug regions for Traditional (P=0.108) or Submerged (P=0.061) (**Figure 2D**).

Recipient ( $61.10\% \pm 5.02\%$ ) had significantly lower cell viability compared to Control (P<0.001) and Submerged (P=0.009), but was equivalent to traditional (P=0.990) (**Figure 2C**).

**Discussion**: There is a significant amount of chondrocyte death induced by donor OCA plug harvesting with traditional handheld irrigation that is mitigated by harvesting while submerged under saline. Similarly, there is a significant amount of cell death experienced at the periphery of the recipient OCA socket. Mitigating this damage may help improve edge integration of OCA transplants.

Clinical Relevance: Submerged irrigation during OCA graft harvest can increase chondrocyte viability during cartilage restoration procedures.



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**Figure 1.** Representative confocal images of Traditional and Submerged OCA plugs. Each sample is imaged with different lenses to capture only dead cells (Red), only live cells (Green), and all cells along the entire 15 mm diameter of the harvested plug.

**Figure 2**. Box and whisker plots demonstrating the differences in cell viability between the control group and (A) whole traditional and whole submerged plugs, (B) central 5mm of the traditional and submerged plugs, (C) peripheral 5mm of the traditional and submerged plugs and recipient rim, and (D) comparing the central versus peripheral regions \* represents a significant difference at P <0.05.