Investigating potential differences in mesenchymal stromal cells in medial and lateral condyles in patients with knee osteoarthritis

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Background: Osteoarthritis (OA) is as a degenerative disease of the whole joint, with characteristic abnormalities to subchondral bone. We have previously demonstrated higher numbers of multipotential stromal cells (MSCs) in more damaged subchondral areas of OA femoral heads[1] however these MSCs were clearly unable to restore damage to cartilage and instead appeared to be involved in the formation of new bone[2].

Aim: To investigate whether similar MSC behaviour can be observed in more damaged[3] medial femoral condyle compared to the less damaged lateral femoral condyle in OA knees.

Methodology: Femoral condyles were obtained from patients that undergoing total knee replacement (n=16). For the assessment of tissue damage, decalcified samples were evaluated for subchondral bone area, cartilage damage score, osteoclast (TRAP staining) and CD271+[1] MSC distribution. MSCs were extracted from subchondral bone after collagenase digestion and expanded, as previously described [1]. MSC numbers, population doubling (PD) times, and *in vitro* motility were compared between medial and lateral condyles.

Results: As expected, medial condyles presented a tendency of higher degree of damage in the cartilage compared to lateral condyles (OOCHAS score[4] of 15-20 compared to 6-10, respectively, n= 3 pairs). Medial condyles showed a 2.40 fold increase in the bone area than lateral condyles (p-value<0.0001), CD271+ MSCs were found mainly around blood vessels and lining the bone. TRAP-positive cells were found in both condyles, particularly in areas of calcified cartilage and often in the vicinity of CD271+ MSCs (Figure 1, A).

No differences in MSC number measured by colony forming assay in lateral 1.39% and medial condyles 1.50% of total extracted cells, or by flow cytometry (lateral 2.84% and medial 2.04% MSC of total extracted cells) . MSC population doubling (PD) rates were also similar between medial (1.61 days per PD) and lateral (1.64 days per PD) condyles. Following culture, a higher motility capacity (p-value=0.0042) was found in lateral condyle MSCs (95.68% of scratch area covered) compared to medial condyle MSCs (79.98% of scratch area covered) after 24 h (Figure 1, B).

Conclusion: Medial condyles displayed consistently high degree of bone sclerosis, while lateral condyles were less sclerotic and had more variable degree of cartilage damage between different

individuals. Although no gross differences were found in MSC numbers, MSC tissue distribution and proliferation abilities, lateral condyle MSCs had higher motility *in vitro* than medial condyle MSCs. Also, preliminary data indicated gene expression differences between these MSCs. MSC co-localisation with TRAP-positive chondroclasts pointed out potential MSC involvement in early stages of tissue remodelling. Understanding MSC behaviour and their abnormal function in OA can potentially facilitate the development of novel therapies for biological joint restoration.

References

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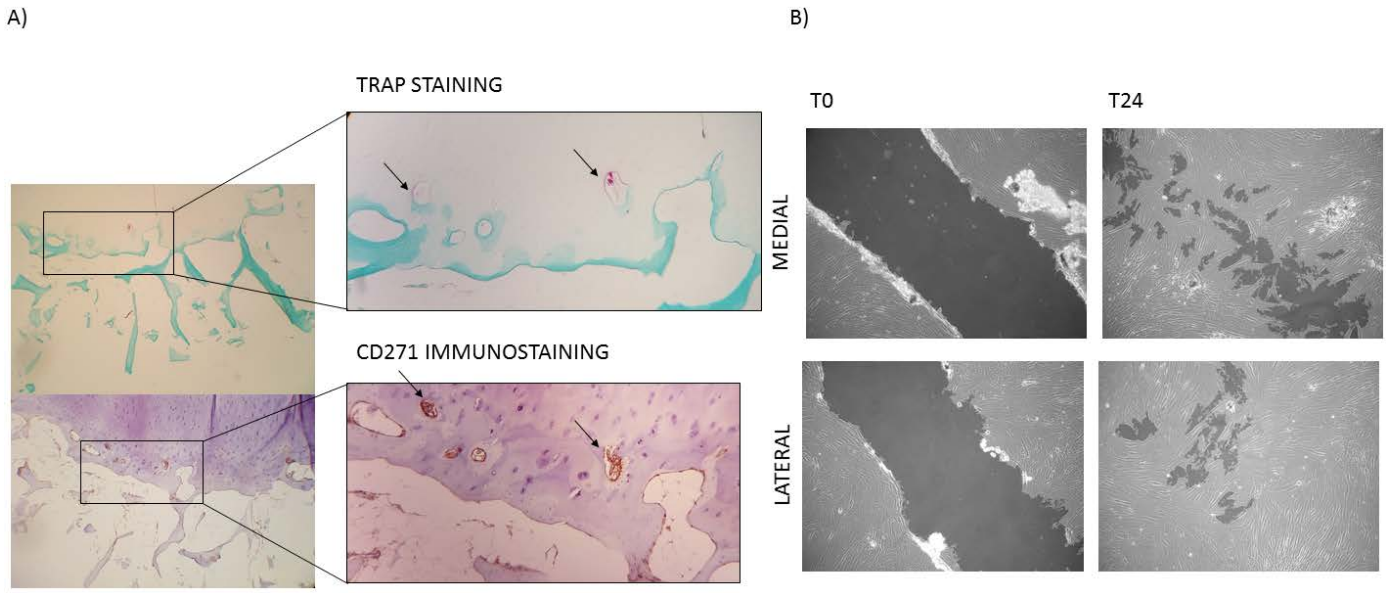

Figure 1. A) Histological images of co-localization of TRAP-positive chondroclasts and CD271+ MSC on the calcified cartilage of condyle. B) Images of scratch performed in the MSC monolayer (t0) and the remained area uncovered by MSCs after 24h.