hMSC Interaction with Environmental Hyaluronan Influences Chondrogenic Potential
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Introduction
Hyaluronan (HA) is a major component of synovial fluid and cartilage matrix. Mesenchymal stem cells (MSCs) have the capacity to repair damaged tissue and can differentiate down the chondrogenic lineage. Increasing the chondrogenic potential of hMSCs would impact their therapeutic application in cartilage repair. Our objectives were to evaluate the effect of HA exposure on the subsequent chondrogenic potential of hMSCs.

Materials and Methods
hMSCs were seeded onto surfaces coated with either 1mg/ml Supartz or Durolane, or onto plastic with 1mg/ml HA in solution. Chondrogenesis was induced by pellet culture and 10ng/ml TGF-β3. GAG content was assessed by staining with Safranin O, and DMMB assay. Immunohistochemistry: Preformed using DakoCytomation Envision+ System-HRP (DAB) and anti-collagen II antibody (NeoMarkers). Immunofluorescence was preformed with either anti-CD44 (Cell Signalling) or anti-CD168 (Ab Cam) antibodies and a PE labelled secondary antibody.

Results
HA exposure influenced the subsequent chondrogenic potential of hMSCs. Most notably, GAG accumulation was abolished in hMSC pellets expanded in the presence of Durolane in solution on day 14 (Fig. 2A). Similarly, CollIII production was markedly reduced (Fig. 1I). GAG content was also significantly reduced on day 21 (Figure 2B). Correspondingly, expression of the HA receptor CD168 was increased in hMSCs expanded in a Durolane solution (Fig. 3F), while expression of CD44 was unchanged (Fig. 3C).

Discussion and Conclusions
Expansion in the presence of Durolane in solution significantly reduced the chondrogenic potential of hMSCs. These results suggest that interaction of hMSCs with bound Durolane through CD44 did not inhibit chondrogenesis, while interaction with Durolane in solution through CD168 significantly reduced the subsequent chondrogenic potential of hMSCs. These results are important to note in the therapeutic application of Durolane for cartilage repair.

Disclosure
The authors have nothing to disclose.