Comparative Cellular and Functional Characterization of BMAC and SVF for Cartilage Regeneration

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INTRODUCTION: Orthobiologics prepared at the point of care are increasingly used for musculoskeletal conditions due to their regenerative potential and ease of application. Among these, bone marrow aspirate concentrate (BMAC) and stromal vascular fraction (SVF) are two prominent cell-based therapies. Despite their widespread use, there is a lack of side-by-side studies comparing their cellular composition and functional effects on joint tissues. This study aims to fill that gap by directly characterizing BMAC and SVF and assessing their paracrine effects on chondrocytes.

METHODS: SVF and BMAC samples were collected respectively from 39 (26M, 13F; 56 ± 9 years) and 28 (17M, 11F; 55 ± 8 years) osteoarthritis (OA) patients undergoing intra-articular injection procedures. Cell counts and viability were assessed using automated cell counting. Flow cytometry was performed (SVF = 27; BMAC = 23) using markers to identify mesenchymal stromal cells (MSCs) (adipose-ASCs: $CD45^{-}CD31^{-}CD34^{+}CD90^{+}CD105^{-/L}CD146^{-}$; bone marrow-BMSCs: $CD45^{-}CD271^{+}$). Soluble factor (SVF = 12; BMAC = 8) profiling was performed using multiplex ELISA-based immunoassays to quantify cytokines, chemokines, growth factors and receptors. Functional *in vitro* assays were carried out by treating immortalized human articular chondrocytes inflamed with IL1β with SVF or BMAC added in transwell-systems. Gene expression was measured across seven experiments (10 SVF, 10 BMAC samples) scoring a panel of 84 inflammation-related genes. Parametric or non-parametric tests were applied based on data normality. Significance was set at $p \leq 0.05$. The study was approved by the local Ethics Committee and written informed consent was obtained from all participants.

RESULTS: Donor characteristics were comparable between SVF and BMAC groups (age: 56 ± 9 vs. 55 ± 8 years; BMI: 27 ± 3 vs. 27 ± 4; Kellgren–Lawrence grade: 2.3 ± 0.5 vs. 2.3 ± 0.4; all p > 0.05; Figure 1A). BMAC showed significantly higher WBC, RBC, and platelet counts than BMA (WBC: 6.0-fold, PLT: 4.5-fold, RBC: 1.4-fold increase, p < 0.0001), and than SVF (WBC: 213-fold, RBC: 49-fold, PLT: 25-fold, p < 0.0001) (Figure 1B). MSC quantification by flow cytometry revealed similar percentages in BMA (0.0011% ± 0.0007) and BMAC (0.0013% ± 0.0009), but a 7-fold higher MSC/mL in BMAC (859 ± 649 vs. 126 ± 81). SVF ASCs were 0.67% ± 1.13 with 1093 ± 1335 cells/mL; MSC/mL did not significantly differ between SVF and BMAC (Figure 1C). Proteomic analysis identified 121 soluble factors, with BMAC showing an average 4.8-fold higher concentration. Twelve proteins were ≥10-fold enriched in BMAC (p ≤ 0.05), including KIT and CSF1R (>1000 ng/mL). HGF was more abundant in SVF (BMAC/SVF = 0.1). Functional annotation highlighted growth and immune response clusters, with BMP4/5/7, TGFB1 and FGF4 associated with cartilage development. Gene expression in IL1β-stimulated chondrocytes (N = 8) showed 32 detectable transcripts. PCA showed BMAC and SVF counteracted IL1β-induced inflammation, with partial overlap. BMAC significantly reduced *IL8* and *CCL20*, while both treatments downregulated *IL1β*, *IL6*, *CXCL1*/2, and *CSF2* to different extents (Figure 2). Correlation analysis linked *IL8* downregulation to WBC (r = –0.81), *CCL23* (–0.83), *CCL18* (–0.81), and *PLG* (–0.81), and *IL1β* to *CCL23* (–0.80) and *IL13RA1* (–0.82) (Figure 3), suggesting cellular and molecular components of BMAC and SVF may influence their immunomodulatory effects.

DISCUSSION: Despite a similar MSC count, BMAC was richer in blood-derived elements and growth factors and exerted a more pronounced anti-inflammatory effect. This may stem from the presence of immunomodulatory components such as platelets, leukocytes, and specific cytokines and receptors. The findings highlight the complexity of orthobiologics and the need for further clinical and mechanistic studies to clarify their therapeutic roles in OA.

SIGNIFICANCE: This study provides a direct, side-by-side comparison of BMAC and SVF, two point-of-care orthobiologics increasingly used for osteoarthritis treatment, revealing key compositional and functional differences. These findings offer mechanistic insight into their distinct biological effects and support the rationale for tailored clinical use based on product-specific profiles.

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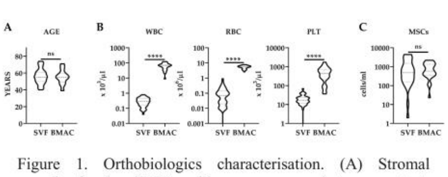

Figure 1. Orthobiologics characterisation. (A) Stromal vascular fraction (SVF) and bone marrow aspirate concentrate (BMAC) donor age, body mass index (BMI) and Kellgren–Lawrence (KL). Violin plots with quartiles and median are shown. $N = 39$ for SVF and 28 for BMAC. ns stands for not significant. (B) Haematological data for SVF and BMAC. Violin plots with quartiles and median are shown. $N = 39$ for SVF and 27 for BMAC. **** for p value < 0.0001. (C) Mesenchymal stromal cells (MSCs) content per millilitre in SVF and BMAC. Violin plots with quartiles and median are shown. $N = 27$ for SVF and 23 for BMAC. ns stands for not significant.

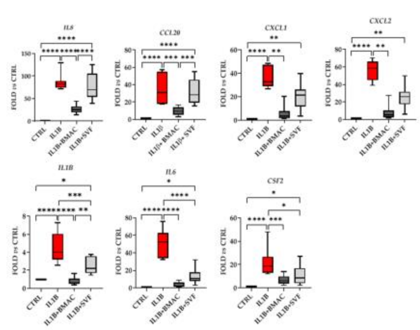

Figure 2. Gene expression modulation in inflamed chondrocytes (IL1B) by bone marrow aspirate concentrate (BMAC) or stromal vascular fraction (SVF) treatment. Untreated (CTRL) cells set as 1. * for p value ≤ 0.05, ** ≤ 0.01, *** ≤ 0.001 and **** ≤ 0.0001. $N = 7$ for CTRL and IL1B, $N = 10$ for IL1B + BMAC and IL1B + SVF.

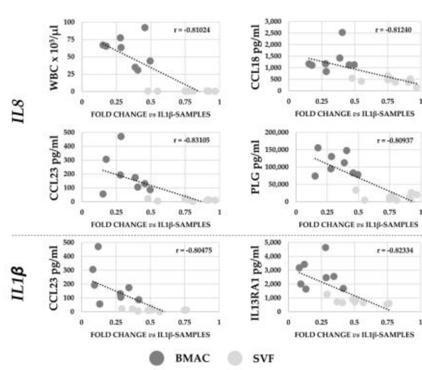

Figure 3. Correlation analyses of BMAC and SVF parameters (WBC or soluble proteins) with modulation in *IL8* and *IL1β* expression in inflamed chondrocytes. $N = 8$ for BMAC and $N = 10$ for SVF.