

EXPANSION OF HUMAN MESENCHYMAL STEM CELLS UNDER SERUM-FREE CONDITIONS

D. Sylva¹, J. Rodrigues¹, G. Reilly¹, F. Claeysens²

¹Univ Sheffield, INSIGNEO Inst Silico Med, Sheffield, S Yorkshire, England,

²Univ Sheffield, Kroto Res Inst, Dept Mat Sci & Engr, Sheffield, S Yorkshire, England

Background

- ❖ The lack of uniformity in sera compositions leads to variable and inconsistent *in-vitro* cell behaviour by altering overall cell metabolism.
- ❖ Serum-free media can provide a more consistent performance and avoid masking of biological tests.
- ❖ In this project we aim to test cell expansion with serum and serum-free media to analyse optimal conditions for fast cell expansion

Methods

- ❖ An immortalized human mesenchymal stem cell line (hTERT-MSC Y201) [1] was seeded in a density of 4000 cells/cm².
- ❖ Well-plates were non-coated or coated with different substrates.
- ❖ Media changes varied in different groups
- ❖ Cells were analysed with light microscopy and metabolic resazurin reduction assay on day 1, day 3 and day 7

Table 1- Media types used and composition

Media	Composition
BM3	DMEM (GIBCO) + 10% FBS (GIBCO)
CD1	StemMACS™ MSC Expansion Media Kit XF, human (Miltenyi Biotec), serum-free and xeno-free
HSM	Human Mesenchymal-XF Expansion Medium (Merck), human-serum

Table 2- Different substrates used for coating

Substrates	
NS	No substrate
FS	Fibronectin
GS	Gelatin

Table 3- Different substrates used for coating

Media changes	
N	No media change
P	Partial media change
F	Full media change

Results

- ❖ All media provided high cell metabolism.
- ❖ CD1 had the highest cell metabolism in all media types and showed the best results when coated with fibronectin or gelatin and without any media changes.

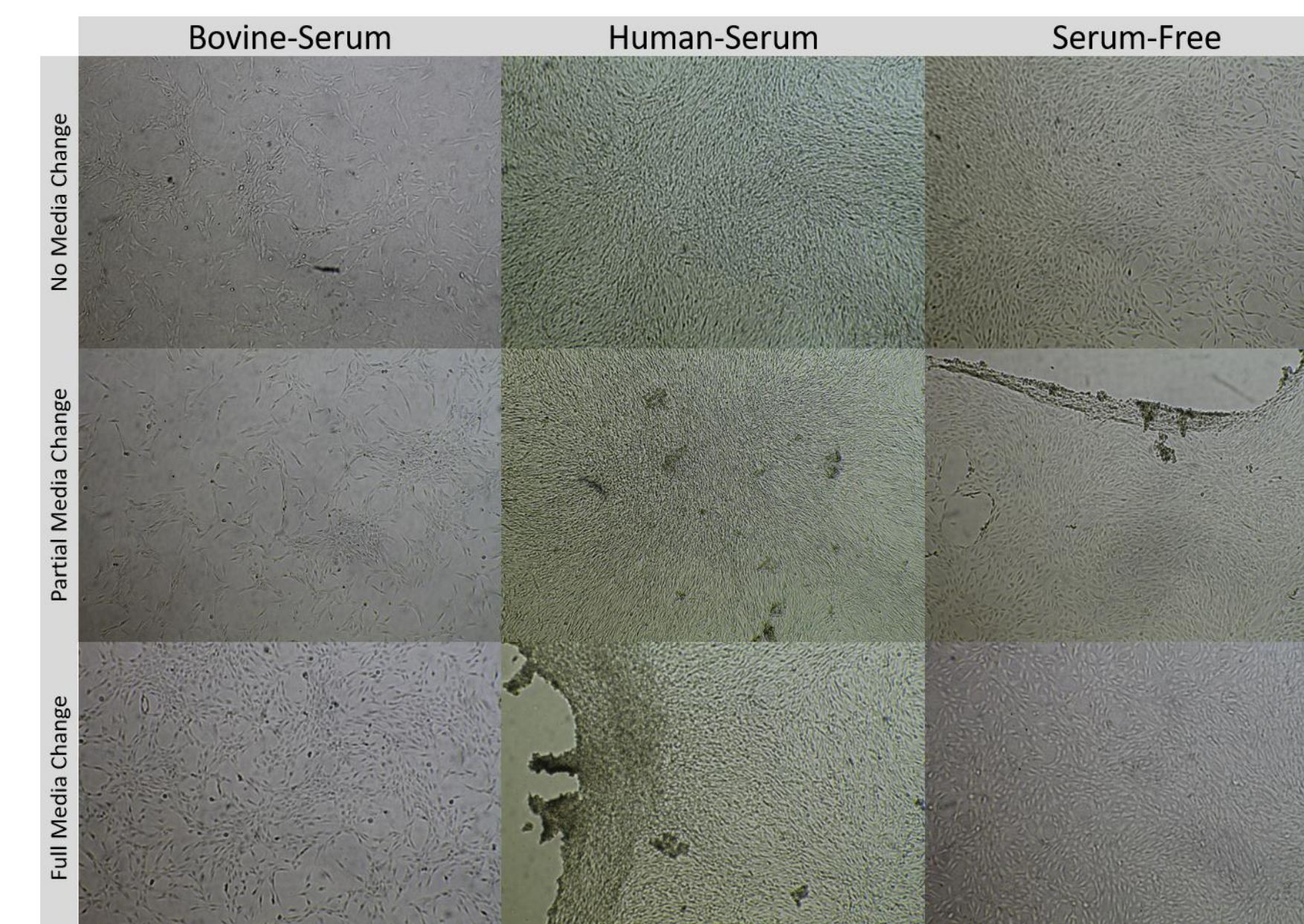


Figure 2- Cells on day 7 with FS coating

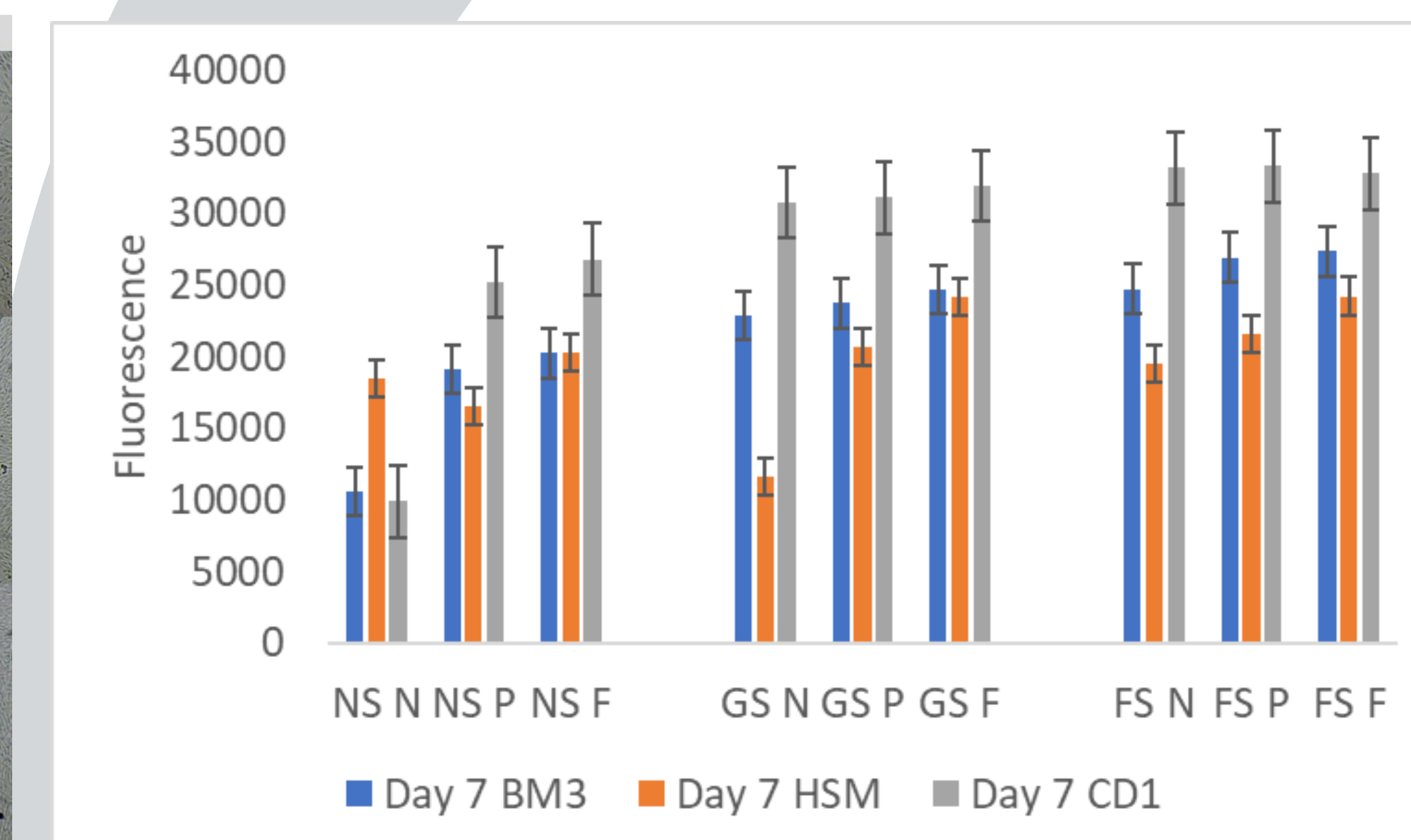


Figure 2- Resazurin metabolic assay on day 7

Discussion and Conclusions

- ❖ Results show that the use of CD1 has the best results when used together with a coating and not treated with any media changes.
- ❖ Serum free media is an overall good candidate for cell expansion

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References

[1] S. James, et al., Stem Cell Reports 4(6) (2015) 1004-1015.