

Minipig Derived Mesenchymal Stem Cells from Bone Marrow and Adipose Tissue: Comparison of Adipogenic, Osteogenic and Chondrogenic Differentiation Potential

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INTRODUCTION: For testing the reconstruction of osteochondral defects in vivo by tissue engineering approaches large animal models like pig have to be used to ensure that similar physical forces as in humans act on the constructs. To circumvent the need of immunosuppressive agents for future in vivo studies autologous cells should be used. Hence, bone marrow and adipose tissue from Göttinger minipigs were compared as mesenchymal stem cell sources regarding cell yield, proliferation capacity, surface marker expression and differentiation ability to the adipogenic, osteogenic and chondrogenic lineage.

METHODS: Porcine bone marrow mesenchymal stem cells (poBMSC) obtained from femur condyle were isolated by erythrocyte lysis and subsequent plastic adherence. Porcine adipose derived stem cells (poASC) were isolated from adipose tissue harvested from the inguinal (I) and neck (N) region by collagenase digestion and subsequent plastic adherence. Proliferation capacity was examined by calculating population doubling levels and times. FACS analysis was performed for typical MSC surface markers. Differentiation experiments were done as described for human MSC and lineage conversion was checked by Oil Red O staining (adipogenic), alkaline phosphatase activity and von Kossa staining (osteogenic), Alcian blue staining and GAG quantification (chondrogenic).

RESULTS: PDT of ASC during the first 8 passages was 57 hours whereas BMSC needed 108.4 hours for doubling. This could be decreased to 58 hours by supplementing the culture medium with 1 ng/mL bFGF. Adipogenic and osteogenic differentiation was

observed in 90% of BMSC and ASC experiments performed whereas chondrogenic differentiation was found in all BMSC but only in 1 of 5 ASC approaches.

Table. 1: Differentiation potential of BMSC and ASC derived from minipigs. Adipose tissue was derived from inguinal (I) or neck (N) region.

pig#	Bone marrow			Adipose tissue		
	Adip.	Ost.	Cho.	Adip.	Ost.	Cho.
A	++	-	++	A (I)		
B	+	+	++	B (I)		
C	++	+/-	+	C (I)		
D				D (I)	+	+
E	++	++	+	E (I)	+	+
				E (N)	++	+
F	+	+	+	F (I)	+	+
				F (N)	++	+
	5/5	4/5	5/5		5/5	5/5
					5/5	1/5

DISCUSSION & CONCLUSIONS: Porcine BMSC were found to be superior to ASC regarding their chondrogenic differentiation potential in vitro. If this is due to in vitro conditions or inherent characteristics that can also be observed in vivo has to be elucidated.

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