Table S1: Studies on impacts of hypoxia on the differentiation and activity of PBMC-derived osteoclasts and osteoclast-like giant cells

Study	Author	Type of Study & Sample	Hypoxic Conditions	Methods	Results
1.	Gorrisen *et al.* (2018)	*in vitro;* CD14⁺ monocytes from human peripheral blood mononuclear cells (PBMCs) were cultured with M-CSF and RANKL supplementation.	5% O_2 throughout the experiment (24 h, 1, 2 and 3 weeks)	a. Osteoclasts were identified by TRAP staining. b. Resorption was measured through bisphosphonate fluorescence visualized with confocal microscopy. c. Gene expression of *CA II*, *ITGB3*, *TRAP*, *CatK* and *DCSTAMP* was measured by RT-qPCR.	a. Hypoxia reduced the intensity of TRAP positive osteoclasts.. b. Hypoxia delayed TRAP-positive osteoclast formation. c. Hypoxia decreased mRNA levels of *CA II*, ITGB3, *TRAP*, CatK and *DCSTAMP*. d. Hypoxia increased single and double nucleated cells but not multinucleated. e. Total bone resorption capacity was not affected by hypoxia exposure over the 21 days. f. Hypoxia decreased senescence marker p21and SA-β-gal expression
2.	Murata *et al.* (2017)	*in vitro;* CD14⁺ monocytes from human peripheral blood mononuclear cells (PBMCs) were cultured with M-CSF and RANKL supplementation.	2% O_2 for 3 h before RANKL was added.	a. Osteoclasts were identified by TRAP staining. b. Resorption was measured through the Osteo Assay Surface plate method. c. Gene expression of *ITGB3*, cathepsin K *CatK* and *CTR* was measured by RT-qPCR.	a. Hypoxia increased the number of TRAP-positive osteoclasts. b. Resorption was higher in cultures under hypoxia c. Hypoxia decreased *ITGB3*, *CatK* and *CTR* expression
3.	Hulley *et al.* (2017)	*in vitro;* CD14⁺ monocytes of PBMCs from leukocyte cones (NHS Blood and Transplant) were cultured with M-CSF and RANKL supplementation for 9 days.	2% O_2 for 24 h.	a. Osteoclasts were identified by TRAP staining. b. VNR-positive osteoclast was detected by immunocytochemistry. c. Resorption was measured through toluidine blue staining method.	a. Number of VNR-positive osteoclasts was not different between hypoxia and normoxia on day 2-3, 4-5, 6-7 and 8-9 of differentiation. b. Resorption was higher in cultures under hypoxia on day 4-5, 6-7 and 8-9 of differentiation.
4.	Utting *et*	*in vitro;*	Hypoxia: 0.2-12%	a. Osteoclasts were identified by	a. Hypoxia increased the osteoclast number

	al. (2010)	Mononuclear cells were isolated from healthy male and female human PBMCs aged 20-50. The mononuclear cells obtained were cultured with M-CSF supplementation.	O_2 for 10 days.	TRAP staining.	b. Hypoxia increased the number of nuclei per osteoclast c. Resorption was higher ~~resorption pit formation per osteoclast and total amount of resorption were higher in cells under~~ under hypoxia (2% O_2).
5.	Knowles *et al*. (2009)	*in vitro;* Osteoclast-like giant cells of giant cell tumor of bone (GCTB) were isolated from tumor tissue and used as a model of mature human osteoclast.	Hypoxia: 8, 5, 2 and 0.1% O_2 for 24 h.	a. Resorption area was visualized using toluidine blue staining. b. CTX-1 level in culture media was measured by ELISA. c. TRAP and CatK activities were measured by spectrometry. d. VNR was visualized by immunohistochemistry.	a. Resorption area was higher in cultures under 5 and 2% O_2, while was lower in 0.1% O_2 compared to normoxia.
		in vitro; Mononuclear cells derived from PBMCs were cultured with M-CSF and RANKL supplementation.	Hypoxia: 8, 5, 2 and 0.1% O_2 for 24 or 72 h. a. 24 h 2% O_2 b. 24 h 2% O_2 + 24 h normoxia c. 72 h hx/rx (reoxygenation) : 3X of 1 h normoxia + 23 h 2% O_2 d. 7 days: 2% O_2 on days 14-21 e. 21 days hx/rx: 21X of 1 h normoxia + 23 h 2% O_2		a. Resorption area and TRAP activity were higher in cultures under 5 and 2% O_2 for 24 h. b. The mRNA level of CTX-1 release in media and CatK was higher in 2% O_2 for 24 h. c. Number of VNR-positive cells was higher in cultures under 8% O_2, while was lower in cultures under 2 and 0.1% O_2 for both 24 and 72 h. b. Number of toluidine blue-positive cells was lower in cultures under 0.1% O_2 for 24 h, 2 and 0.1% O_2 for 72 h. a. Number of toluidine blue-positive cells was lower in cultures under 2% O2 on days 14-21, while cell number was not different between other conditions and normoxia.

6.	Muzylak *et al*. (2006)	*in vitro;* PBMCs isolated from healthy feline blood of 2 years old. Cells were cultured with M-CSF and RANKL supplementation.	Hypoxia: 12 or 2% O_2 on days 1-14 or 11-14.	a. Osteoclasts were identified by TRAP staining. b. Resorption was visualized by biotin-conjugated wheat germ agglutination lectin staining after reacted with TRITC-streptavidin.	a. Hypoxia lowered the number of TRAP-positive osteoclasts when exposed to 12 and 2% O_2, b. The size of each osteoclast was bigger in 2% O_2 compared to 20% O_2 exposure on days 1-14. c. The size of TRAP-positive osteoclasts was bigger while their number remained unchanged in cultures exposed to 2% O_2 on 11-14 days. d. Resorption percentage was higher in cultures exposed to 12 and 2% O_2 on days 1-14 and 11-14.

Note: Normoxia or reoxygenation (rx): 20-21% O_2. Abbreviations: Oxygen (O_2), Quantitative reverse transcriptase polymerase chain reaction (RT-qPCR), messenger RNA (mRNA), Tetramethylrhodamine (TRITC), Enzyme linked immunosorbent assay (ELISA), Tartrate-resistant acid phosphatase (TRAP), Carbonic anhydrase II (CA II), Integrin subunit beta-3 (ITGB3), Cathepsin K (CatK) and Dendritic cell-specific transmembrane protein (DCSTAMP), Calcitonin receptor (*CTR*), Vitronectin receptor (VNR) & hypoxia (hx).