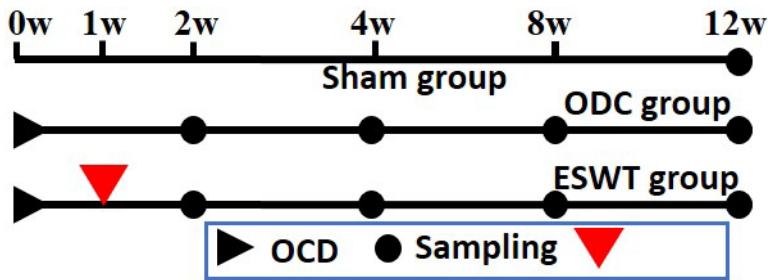


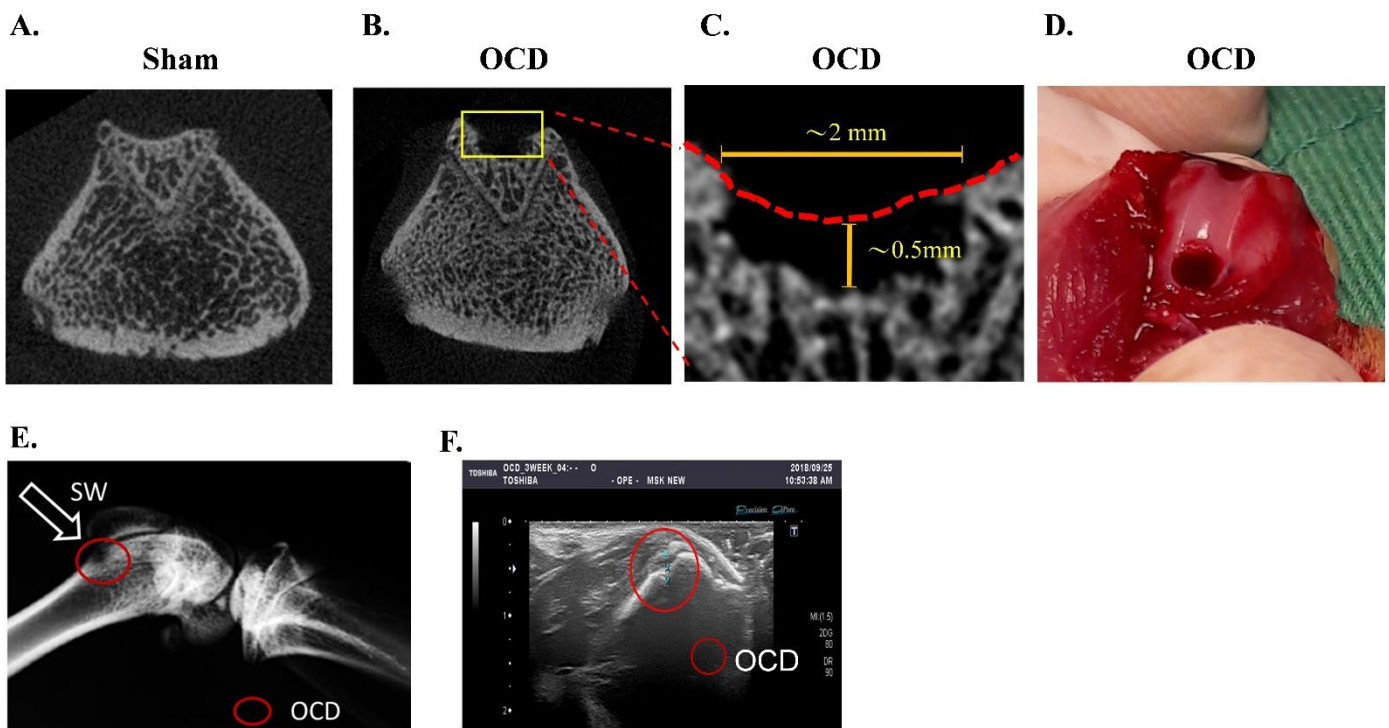
# Bone & Joint Research

## Supplementary Material

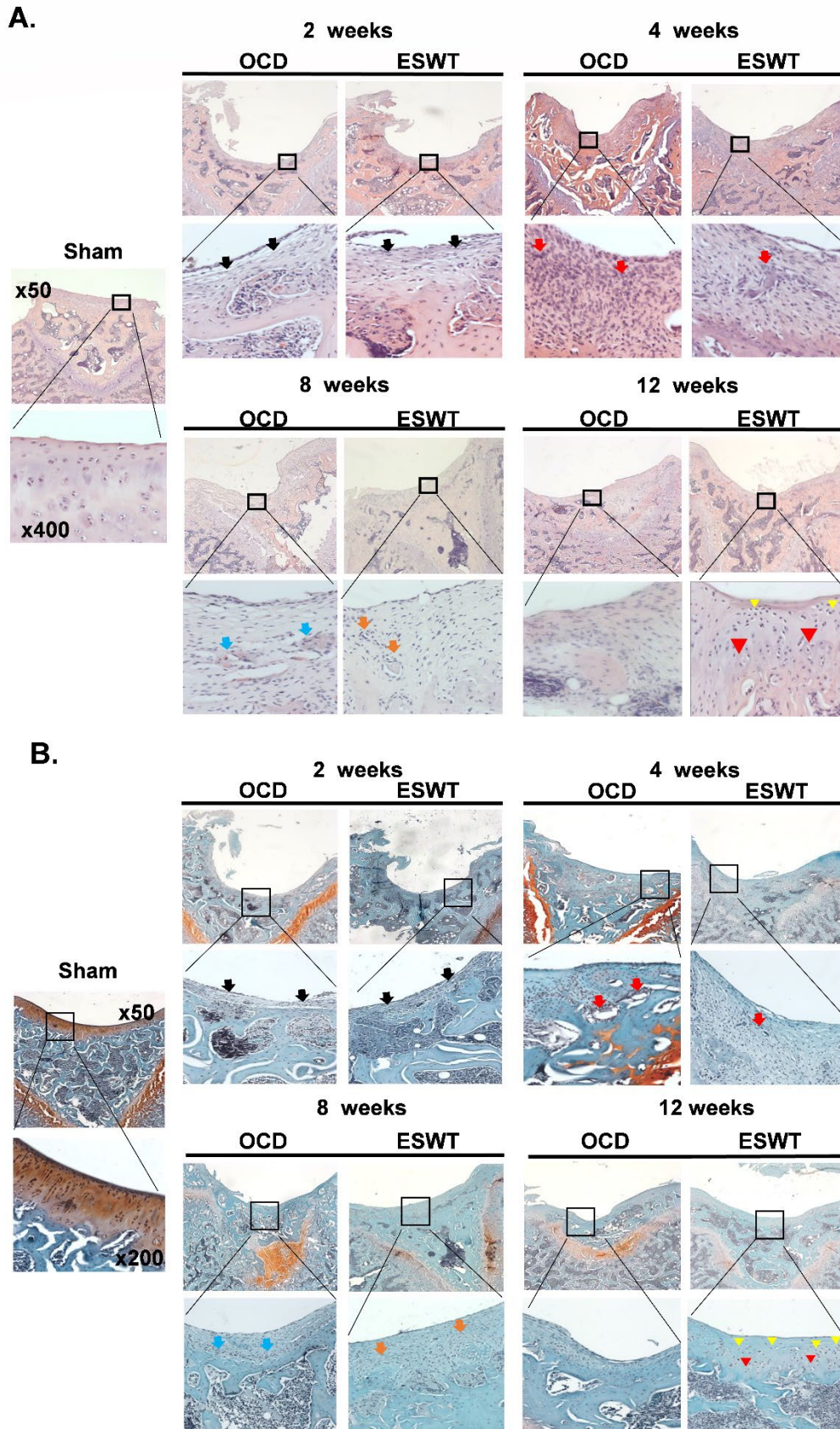
10.1302/2046-3758.138.BJR-2023-0264.R2



**Fig. a.** The flowchart of the experimental design and process. ESWT, extracorporeal shockwave therapy; OCD, osteochondral defect.



**Fig. b.** The osteochondral defect (OCD) rat model. The micro-CT images of A) Sham and B) artificial OCD lesion was taken through the subchondral bone plate and C) subarticular spongiosa in the epiphyseal compartment of the top femoral trochlear groove with 2 mm width and 0.5 mm depth lesion as compared with the Sham group. D) The OCD lesion is in the femoral trochlear groove of the left rat knee. E) A radiograph and F) ultrasound image displaying the OCD lesion and the location of application of focused shockwave (arrow).



**Fig. c.** A) Haematoxylin and eosin (HE) and B) Safranin-O staining of Sham, osteochondral defect (OCD), and extracorporeal shockwave therapy (ESWT) groups are showed at 50x magnification. The boxes indicate image locations at 400x and 200x magnification in HE and Safranin-O staining, respectively.

**Table i.** Mean percentage of positive cells in the articular cartilage of Sham, osteochondral defect, and extracorporeal shockwave groups. Standard deviations are presented in brackets.

Cell type	Group								
		2 weeks		4 weeks		8 weeks		12 weeks	
	Sham	OCD	ESWT	OCD	ESWT	OCD	ESWT	OCD	ESWT
SOX 9	67.48 (9.13)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	24.68 (28.57)*
Aggrecan	26.99 (4.21)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	1.40 (1.44)*
TGF- $\beta$ 1	35.86 (6.74)	46.75 (15.94)	58.44 (21.46)	93.31 (5.58)	92.26 (4.09)	83.24 (11.84)	91.10 (5.68)	58.60 (13.61)	79.15 (11.48)*
BMP-2	19.35 (3.83)	0.68 (1.16)	0.63 (0.71)	0.76 (0.86)	0.77 (1.18)	0.75 (0.81)	0.80 (0.93)	0.37 (0.52)	18.33 (17.06)*
BMP-3	19.80 (5.67)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.13 (0.02)	0.941 (1.10)*
BMP-4	16.48 (3.38)	0.05 (0.14)	0.16 (0.46)	0.13 (0.19)	0.05 (0.15)	0.67 (0.93)	1.41 (1.66)	0.806 (1.14)	14.50 (14.44)*
BMP-5	79.29 (7.98)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.09 (0.276)	0.00 (0.00)	0.44 (0.70)	0.626 (0.743)	40.65 (40.30)*
BMP-7	4.54 (2.09)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.40 (0.53)	0.00 (0.00)	1.41 (1.07)*

\*p < 0.05 as comparing between OCD and ESWT groups.

BMP, bone morphogenetic protein; ESWT, extracorporeal shockwave therapy; OCD, osteochondral defect; SRY-box transcription factor 9; TGF- $\beta$ 1, transforming growth factor  $\beta$ 1.

**Table ii.** Mean relative intensity of cells in bone marrow of Sham, osteochondral defect, and extracorporeal shockwave therapy groups. Standard deviations are presented in brackets.

Cell type	Sham	Group							
		2 weeks		4 weeks		8 weeks		12 weeks	
		OCD	ESWT	OCD	ESWT	OCD	ESWT	OCD	ESWT
TGF- $\beta$ 1	1.00 (0.28)	3.10 (0.66)	4.089 (0.82)	6.42 (1.40)	7.38 (1.50)*	6.837 (1.40)	8.29 (1.00)*	4.47 (0.85)	7.28 (0.97)†
BMP-2	1.00 (0.13)	1.63 (0.32)	2.21 (0.49)	2.51 (0.58)	4.10 (0.55)†	2.07 (0.44)	3.40 (0.70)†	1.45 (0.35)	2.61 (0.42)†
BMP-3	1.00 (0.15)	0.59 (0.15)	0.51 (0.11)	0.35 (0.15)	0.24 (0.09)	0.47 (0.11)	0.50 (0.09)	0.81 (0.11)	0.76 (0.14)
BMP-4	1.00 (0.52)	1.90 (0.63)	4.96 (0.89)†	4.64 (1.16)	10.79 (0.95)†	4.33 (0.93)	7.91 (1.46)†	2.70 (0.56)	6.07 (0.85)†
BMP-5	1.00 (0.11)	0.36 (0.10)	0.76 (0.12)†	0.873 (0.20)	1.51 (0.12)†	1.50 (0.11)	1.88 (0.16)†	1.60 (0.10)	2.09 (0.20)†
BMP-7	1.00 (0.03)	0.02 (0.01)	0.04 (0.02)	0.03 (0.04)	0.06 (0.02)	0.14 (0.04)	0.18 (0.03)	0.08 (0.02)	0.53 (0.16)†

\*p < 0.05 and †p < 0.01 as comparing between OCD and ESWT groups.

BMP, bone morphogenetic protein; ESWT, extracorporeal shockwave therapy; OCD, osteochondral defect; TGF- $\beta$ 1, transforming growth factor- $\beta$ 1.