

456 Figure 3 Real-time PCR of cartilage expressed genes in WT and *Bgn^{-/-}Fmod^{-/-}*
457 treated without or with BMP-2. A) Type II collagen (*Col2*), B) Type X collagen
458 (*Col10*) C) *Sox9*, D) *ihh*. #*p*<0.01.

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460 Figure 4. Immunohistochemistry (IHC) and real-time PCR of genes expressed in
461 fibrocartilage and bone. A) IHC of type I collagen in WT (left panels) and *bgn^{-/-},*
462 *fmod^{-/-}* deficient condyles (right panels) treated without (top panel) or with (bottom
463 panels) BMP-2. B) Real-time PCR of type I collagen (*Col1*), C) *Runx2* D) osterix
464 (*Osx*) and E) *Wisp1* mRNA in WT and *Bgn^{-/-}Fmod^{-/-}* condyles treated with or
465 without BMP-2. **p*<0.05, #*p*<0.01

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467 Figure 5. Assessment of bone resorption A) Tartrate Resistant Acid Phosphatase
468 (TRAP) staining of osteoclasts in WT (left panels) and *Bgn^{-/-}Fmod^{-/-}* deficient
469 condyles (right panels) treated without (top panel) or with (bottom panels) BMP-2.
470 Real-time PCR to measure levels of B) TRAP (*Acp5*) C) *Rankl* D) and AdamTS4
471 mRNA.

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473 Supplementary:

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475 Table S1. Primer sequences used for Real-Time PCR

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477 S1. Diagram showing an outline of experimental approach used to treat and
478 analyze the TMJ from WT *Bgn^{-/-}Fmod^{-/-}* mice.

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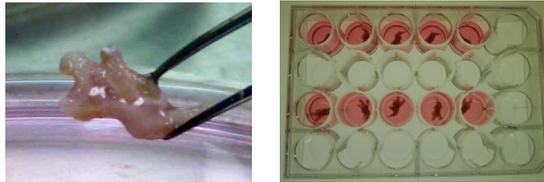
480 S2. Low power view of condyle showing fibrocartilage, cartilage and subchondral
481 bone layers. The Safranin O red stained area was used to quantify the relative
482 area of cartilage present in treated and untreated WT and *Bgn*^{-/-}*Fmod*^{-/-} condyles.

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Table 1 Primer sequences used for real-time PCR

<i>Acan</i>	forward 5' CCCGGTACCCTACAGAGACA3', reverse 5' ACAGTGACCCTGGAAGTTGG3',
<i>Col2A1</i>	forward 5' CGAGGGCAACAGCAGGTTACATAC3' reverse 5' GTCAATAATGGGAAGGCGGGAGGTC3'
<i>Col10A1</i>	forward 5' GGGATGCCGCTTGTCAGTGCTAAC3' reverse 5' TGGGTCGTAATGCTGCTGCCTATTG3'
<i>Sox9</i>	forward 5' TCAGATGCAGTGAGGAGCAC3' reverse 5' CCAGCCACAGCAGTGAGTAA3'
<i>IHH</i>	forward 5' ACGTGCATTGCTCTGTCAAG3' reverse 3' CTCGATGACCTGGAAAGCTC3'
<i>Col1A1</i>	forward 5' CACCCTCAAGAGCCTGAGTC3' reverse 5' GCTTCTTTTCCTTGGGGTTC3'
<i>Runx2</i>	forward 5' GCAGTTCCCAAGCATTTCAT3' reverse 5' CACTCTGGCTTTGGGAAGAG3'
<i>osx (SP7)</i>	forward 5' AAGTGTGTGTGCCGTGGATA3' reverse 5' GTCGAGAGGACTGGGGTACA3'
<i>Wisp1</i>	forward 5' ATCGCCCGAGGTACGCAATAGG3' reverse 5' CAGCCCACCGTGCCATCAATG3'
<i>Acp5 (Trap)</i>	forward 5' CAGCAGCCAAGGAGGACTAC3' reverse 5' ACATAGCCCACACCGTTCTC3'
<i>Rankl</i>	forward 5' ATCGTGGTGTATGTCAGCCA3' reverse 5' GTCATTCTGCACATTGTCCG3'
<i>Adamts4</i>	forward 5' CGCTGACCGCCAATGCCAACTG3' reverse 5' CGCTGACCGCCAATGCCAACTG3'
<i>S29</i>	forward 5' GGAGTCACCCACGGAAGTTCCGG3' reverse 5' GGAAGCACTGGCGGCACATG3'



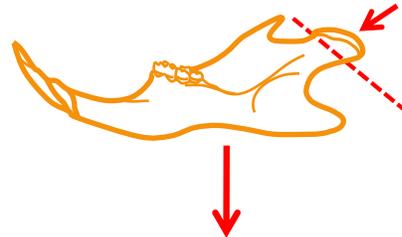
isolate whole mandible condyles from WT or *Bgn*^{-/-}*Fmod*^{-/-} mice



3 (histology) or 5 weeks (mRNA)

culture explants 24 -well plate for 24h

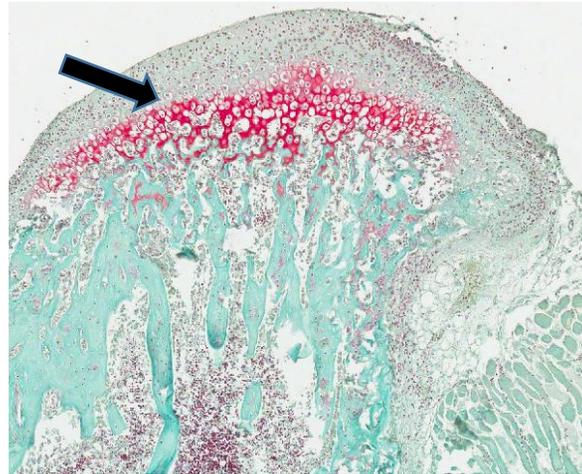
+/- BMP-2 48 hours



collect explants
microCT, histology and mRNA analysis

S1 Experimental Design

Fibrocartilage
Cartilage
Subchondral Bone



**S2. Safranin-O positive region (red stain) of the condyle under low power
Showed the region of the condyle that was measured (arrow).**