

## **SUPPLEMENTARY TABLES AND FIGURES**

### **Sympathetic $\beta_1$ -Adrenergic Signaling Contributes to Regulation of Human Bone Metabolism**

Supplementary Table 1

Supplementary Table 2

Supplementary Figure Legends

Supplementary Figures 1-5

**Supplementary Table 1.** DXA and central QCT parameters in  $\beta_1$ -selective blocker users versus non-users. Values are age- and sex-adjusted least squares means  $\pm$  SE. P-values are based on linear regression models that include the variables user status, age, and sex.

	<b>Non-users</b>	<b>Users</b>	<b>P-value (adjusted for sex and age)</b>
<b>DXA</b>			
L1-L4, g/cm <sup>2</sup>	1.23 $\pm$ 0.02	1.26 $\pm$ 0.03	0.44
FN, g/cm <sup>2</sup>	0.91 $\pm$ 0.01	0.95 $\pm$ 0.02	0.06
UD Radius, g/cm <sup>2</sup>	0.48 $\pm$ 0.01	0.49 $\pm$ 0.01	0.20
Distal Radius, g/cm <sup>2</sup>	0.69 $\pm$ 0.01	0.71 $\pm$ 0.01	0.23
<b>QCT</b>			
Spine Trab, mg/cm <sup>3</sup>	138 $\pm$ 2.3	145 $\pm$ 4.0	0.13
FN Trab, mg/cm <sup>3</sup>	188 $\pm$ 3.3	201 $\pm$ 5.9	0.06
FN Cortical, mg/cm <sup>3</sup>	537 $\pm$ 5.9	551 $\pm$ 10.6	0.25

Supplementary Table 2. Forward and reverse primer sequences for the genes analyzed in this study.

<b>Gene</b>	<b>Accession #</b>	<b>Forward Primer Sequence (5' to 3')</b>	<b>Reverse Primer Sequence (5' to 3')</b>
<i>RANKL</i>	NM_003701	GGTGGATGGCTCATGGTTAGA	GAGCAAAAGGCTGAGCTTCAA
<i>OPG</i>	NM_002546	TGCTCAGTTTGTGGCGAATAAA	CGTGCATTAGGCCCTTCAAG
<i>ADRB1</i>	NM_000684	TGCTACAACGACCCCAAGTG	AGGTACACGAAGGCCATGATG
<i>ADRB2</i>	NM_000024	TCTTTGAAGGCCTATGGGAATG	TCCACTCTGCTCCCCTGTGT
<i>C-FOS</i>	NM_005252	GAGAATCCGAAGGGAAAGGAAT	TCCGCTTGGAGTGTATCAGTCA
<i>C-MYC</i>	NM_002467	CGTCTCCACACATCAGCACAA	TCTTGGCAGCAGGATAGTCCTT
<i>CRY1</i>	NM_004075	GTGTGGCCAGGTGGAGAAAC	TGCCACCCAAGCTTTTCTTT
<i>CRY2</i>	NM_021117	ATTGAGCTGAATGGGCAGAAG	GGCTGATGATGGCCTGAAAG
<i>PER1</i>	NM_002616	AGTCCGTCTTCTGCCGTATCA	GTTAGGCGGAATGGCTGGTA
<i>PER2</i>	NM_022817	GCCACCCTGAAGAGGAAATG	CCGCTTATCACTGGACCTTAGC
<i>ACTB</i>	NM_001101	CCCAGCCATGTACGTTGCTAT	TCACCGGAGTCCATCACGAT
<i>TUBA1A</i>	NM_006009	GAGTGCATCTCCATCCACGTT	TAGAGCTCCCAGCAGGCATT

## Supplementary Figures

Supplementary Figure 1. Relative mRNA levels of (A) *C-FOS*, (B) *C-MYC*, and (C-F) circadian clock genes in hFOB cells treated for 2 hours with vehicle versus dobutamine ( $\beta_1$ -agonist) or salmeterol ( $\beta_2$ -agonist) (see Methods for experimental details).  $n = 6$  per treatment, with comparisons between vehicle and treatment using the two-sample t-test. \* $P < 0.05$ ; \*\* $P < 0.01$ ; \*\*\* $P < 0.001$ .

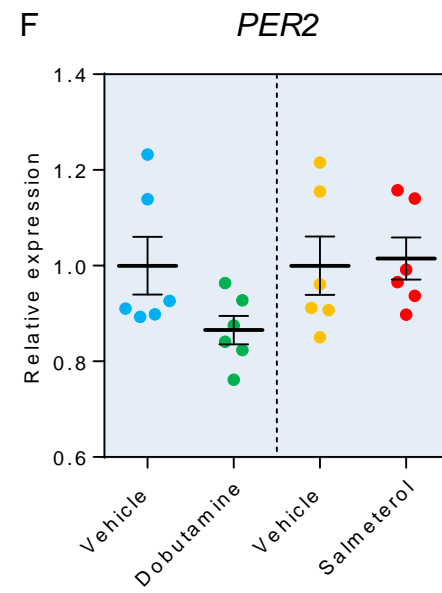
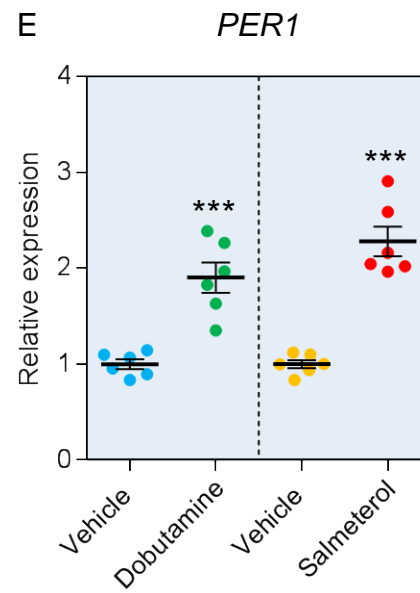
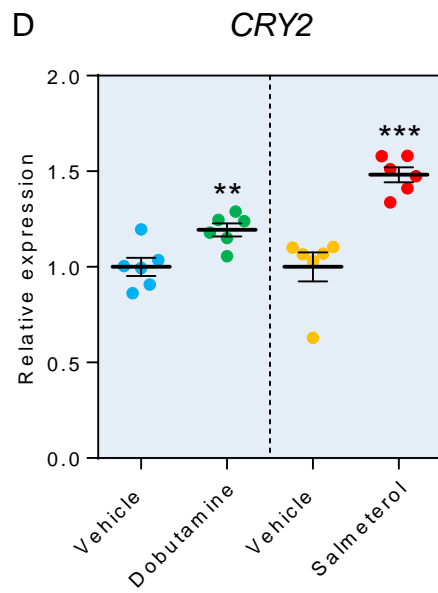
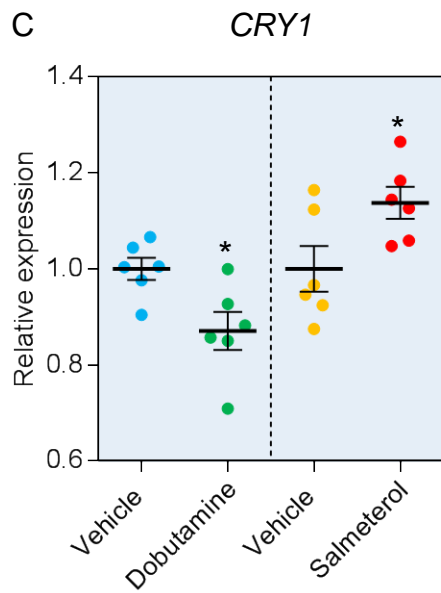
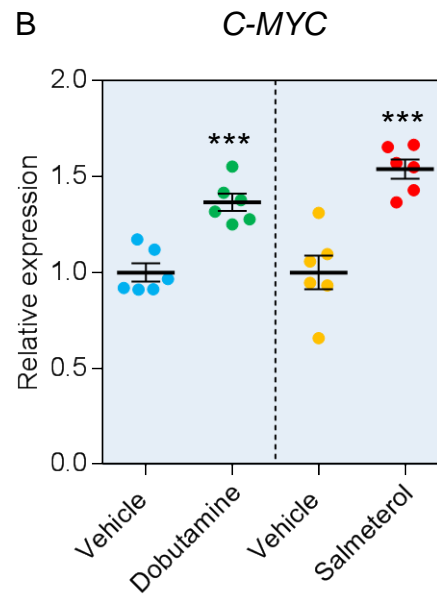
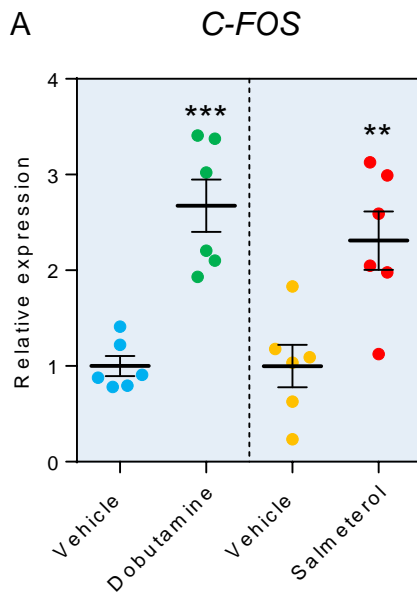
Supplementary Figure 2. Time course of changes in serum CTx following treatment.  $P = 0.009$  for the overall mixed effects model (see Statistical Analyses); \*\* $P < 0.01$  versus the placebo group using Dunnett's two-tailed t-test. Sample sizes over time are as indicated in the CONSORT flow diagram in Figure 2.

Supplementary Figure 3. Time course of changes in serum PINP following treatment.  $P < 0.001$  for the overall mixed effects model (see Statistical Analyses); \* $P < 0.05$  versus the placebo group using Dunnett's two-tailed t-test. Sample sizes over time are as indicated in the CONSORT flow diagram in Figure 2.

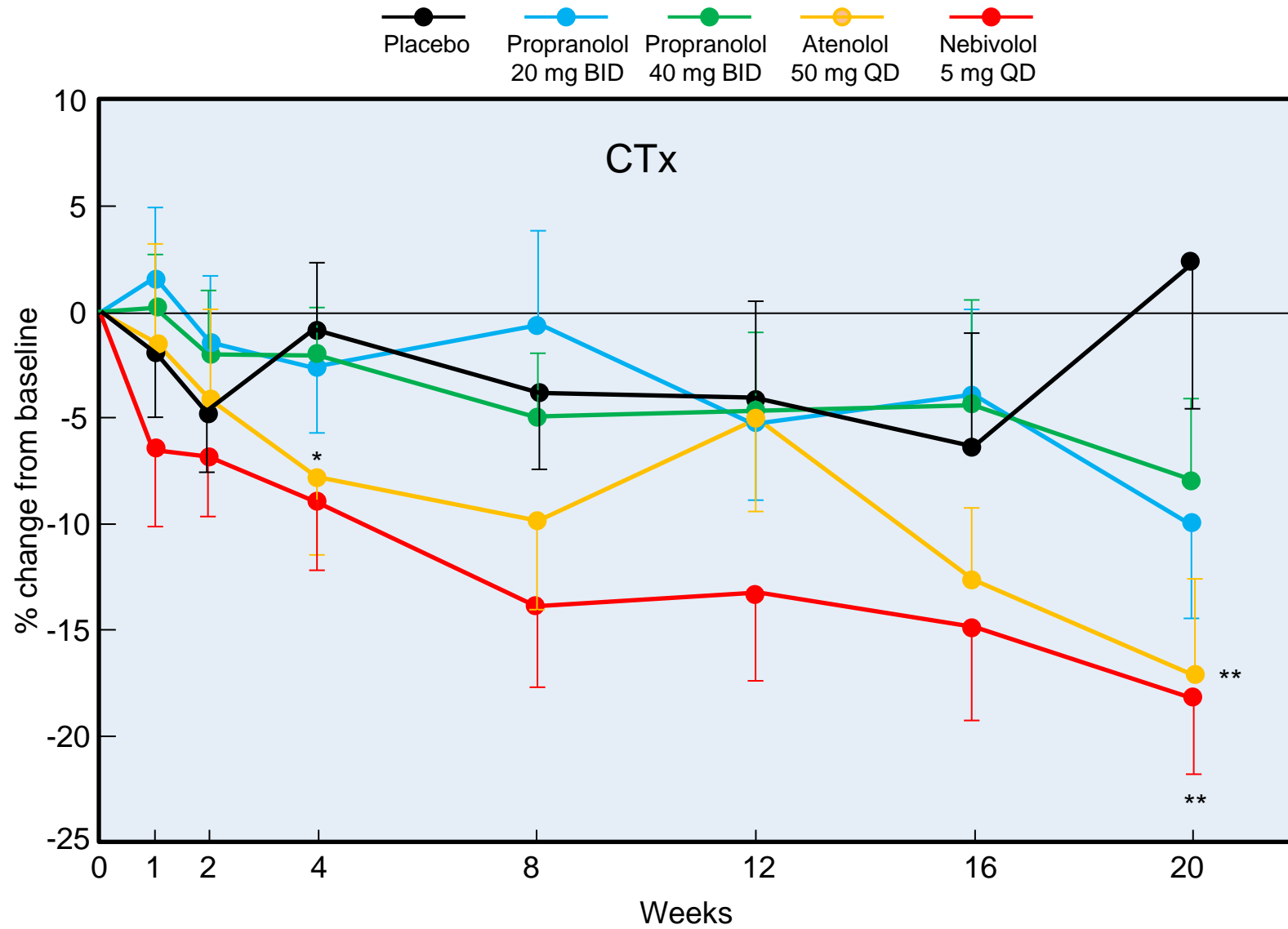
Supplementary Figure 4. Time course of changes in serum TRAP5b following treatment.  $P < 0.001$  for the overall mixed effects model (see Statistical Analyses); \*\* $P < 0.01$  and \*\*\* $P < 0.001$  versus the placebo group using Dunnett's two-tailed t-test. Sample sizes over time are as indicated in the CONSORT flow diagram in Figure 2.

Supplementary Figure 5. Time course of changes in serum osteocalcin following treatment.  $P < 0.001$  for the overall mixed effects model (see Statistical Analyses); \* $P < 0.05$ ; \*\* $P < 0.01$ , and \*\*\* $P < 0.001$  versus the placebo using Dunnett's two-tailed t-test. Sample sizes over time are as indicated in the CONSORT flow diagram in Figure 2.

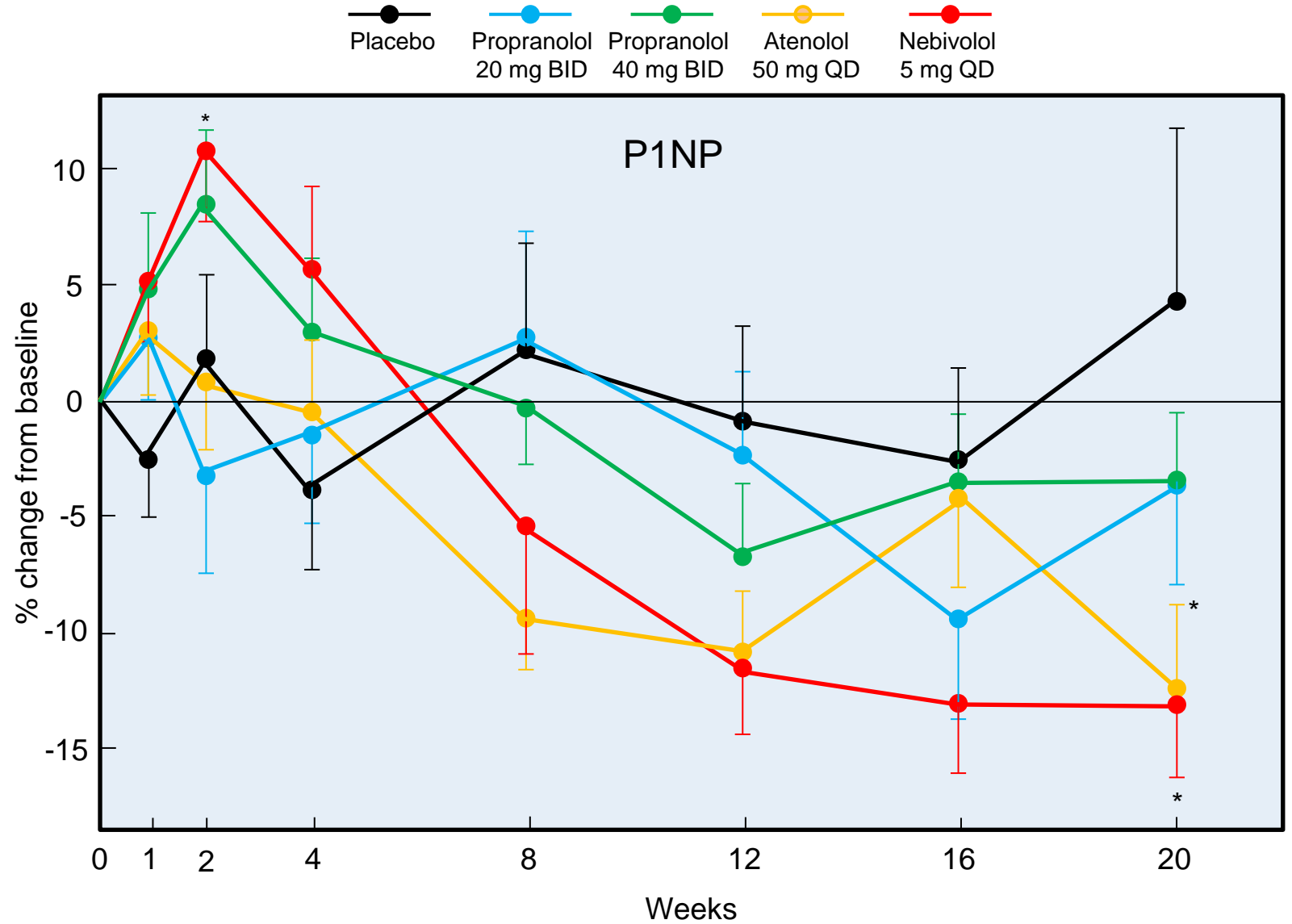
# Supplementary Figure 1



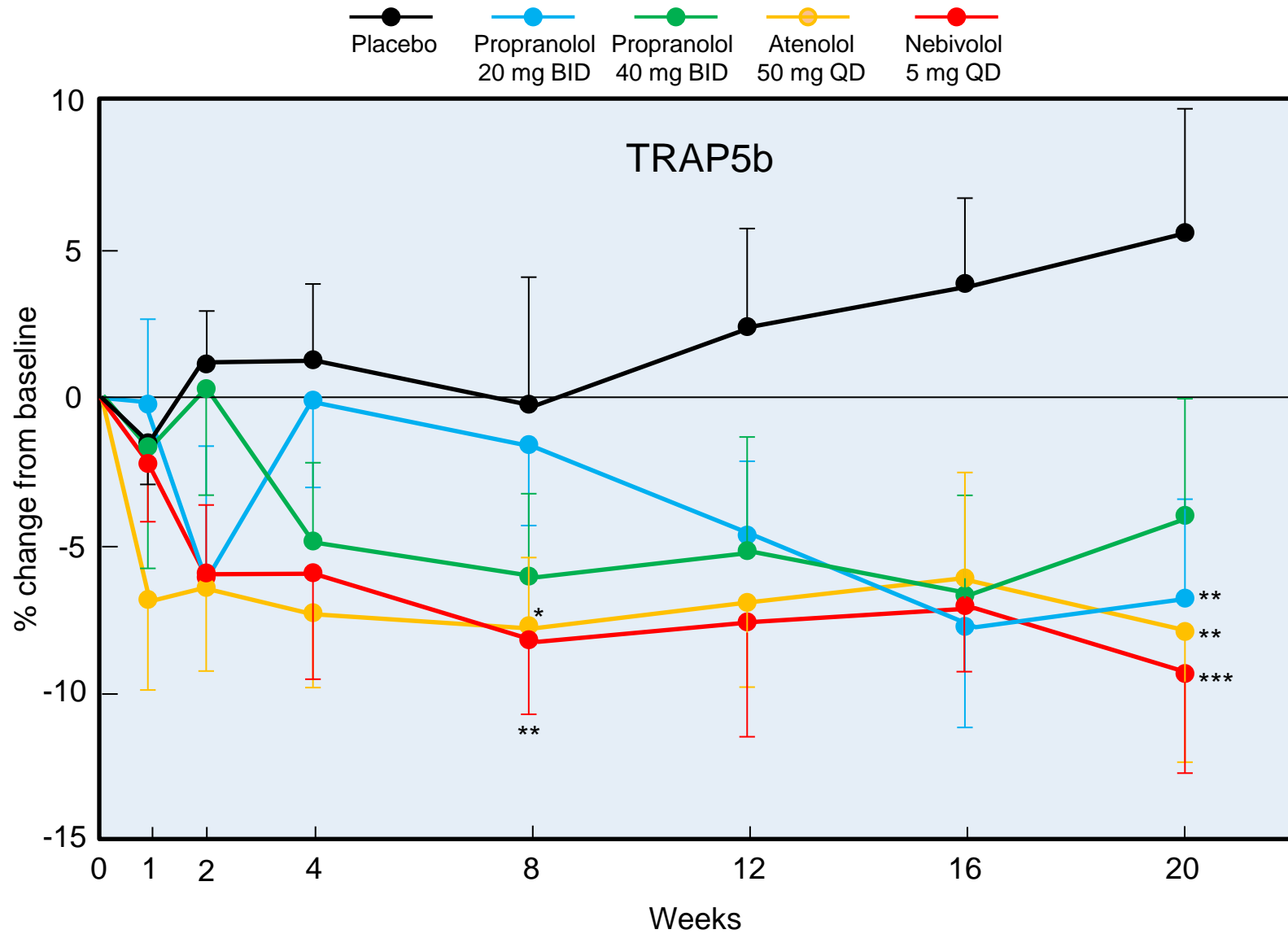
Supplementary Figure 2



Supplementary Figure 3



Supplementary Figure 4





Supplementary Figure 5

