

**LEFTY2 Inhibits Endometrial Receptivity by Downregulating Orai1 Expression
and Store-Operated Ca²⁺ Entry**

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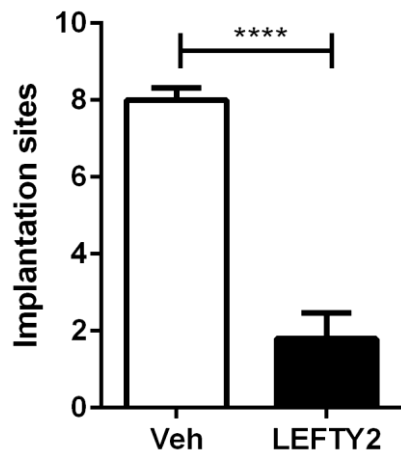
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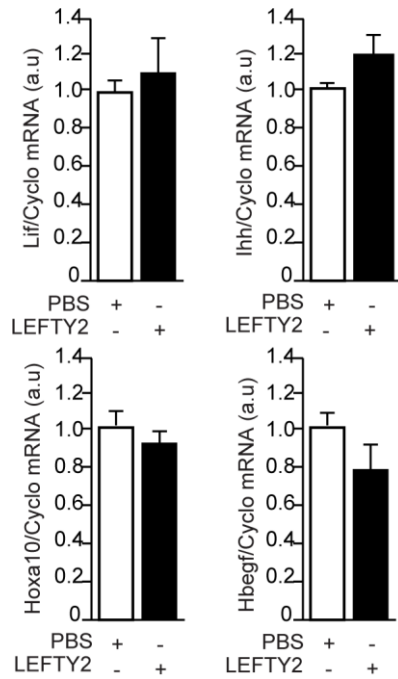
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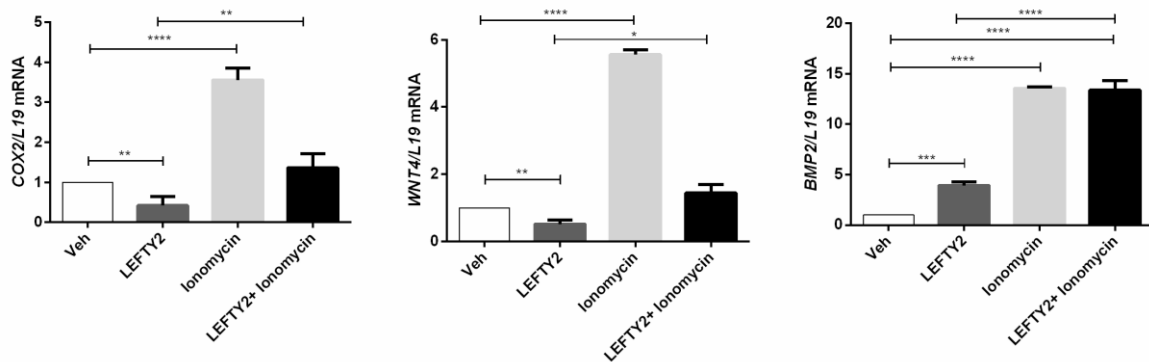
Supplementary Figure 1: LEFTY2 impairs blastocyst implantation in mice.

C57BL/6 female mice mated with fertile males were subjected to laparotomy at 3.0 dpc and both uterine horns gently flushed with either vehicle (PBS) or recombinant LEFTY2 (500 ng/ml). Each treatment group consisted of 5 mice. Bar graph shows arithmetic means \pm SEM ($n = 5$) of the number of implantation sites following uterine flushing with PBS (Veh; White bar) or recombinant LEFTY2 (black bar). ****($P < 0.0001$) indicate significant difference from control mice (Student's t -test).



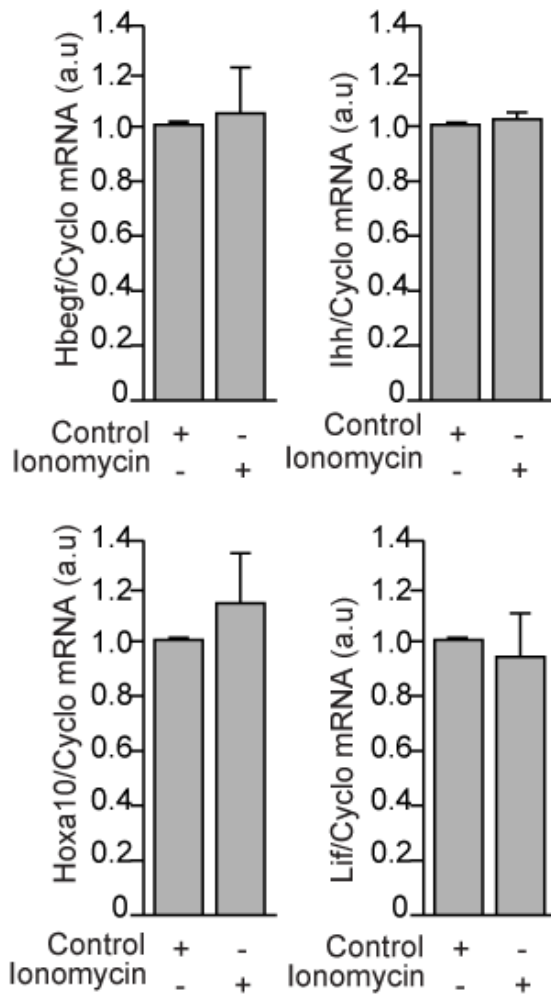
Supplementary Figure 2: Receptivity genes insensitive to LEFTY2.

Arithmetic means \pm SEM (n=6) of transcript levels encoding murine *Lif*, *Hbegf*, *Hoxa10* and *Ihh* levels after flushing with PBS (black bars) and with LEFTY2 (500 ng/ml; grey bars). Transcript levels were determined by qRT-PCR, normalized to the levels of *Cyclo* (housekeeping) mRNA and expressed in arbitrary units (a.u.). No significant difference was seen.



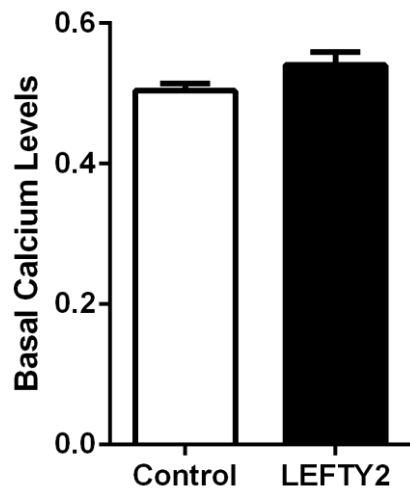
Supplementary Figure 3: Differential regulation of Ca²⁺-dependent receptivity genes in response to LEFTY2 and Ionomycin.

Primary HESC cultures were treated with 8-Br-cAMP and MPA for 6 days with or without LEFTY2 (25 ng/ml) or Ionomycin (1 μ M) or in combination. Transcript levels encoding human *COX2*, *WNT4* and *BMP2* were determined by qRT-PCR and normalized to *L19* (housekeeping) mRNA and expressed in arbitrary units (a.u.). The data shown are arithmetic means \pm SEM (n=6). **($P<0.01$) and ***($P<0.001$) indicates significant difference when using Student's *t*-test.



Supplementary Figure 4: Receptivity genes insensitive to Ionomycin.

Primary HESC cultures were treated with 8-Br-cAMP and MPA for 6 days with or without Ionomycin (1 μ M). Arithmetic means \pm SEM (n=6) of transcript levels encoding human *LIF*, *HBEGF*, *HOXA10*, *IHH* transcript levels determined by qRT-PCR, normalized to the levels of *L19* mRNA and expressed in arbitrary units (a.u.). No significant difference was seen.



Supplementary Figure 5: Basal Calcium Levels

Arithmetic means \pm SEM (n = 6, each experiment 15-30 cells) of basal Ca^{2+} of Control (Veh) and LEFTY2 treated cells. No significant difference was seen.