SUPPLEMENTAL INFORMATION

Antibody	Concentration and Use	Company
Mouse Anti-Nodal WS65	1:500, WB 1:50, IF	Santa Cruz Biotechnology
Mouse Anti- HIF-1α	1:500, WB 1:50, IF	BD Biosciences
Rabbit Anti-HIF-1α ChIP Grade	25μg for 15 x 10^6 cells, ChIP	Abcam
Rabbit Anti-HIF-2α	1:1000, WB 1: 100, IF 25μg for 15 x 10^6 cells, ChIP	Novus Biosciences
Mouse Anti-DDK 4C5	1:1000, WB	OriGene Technologies
Mouse anti-βActin C4	1:5000, WB	Santa Cruz Biotechnology,
Rabbit Anti-Phospho-Smad2 (Ser465/467)	1:1000, WB	Cell Signaling Technology
Rabbit Anti-SMAD 2/3	1:1000, WB 1:100, IF	Millipore

SI Table 1: Antibodies Utilized for Western Blot (WB), Immunofluorescence (IF) and Chromatin Immunoprecipitation (ChIP) Analyses

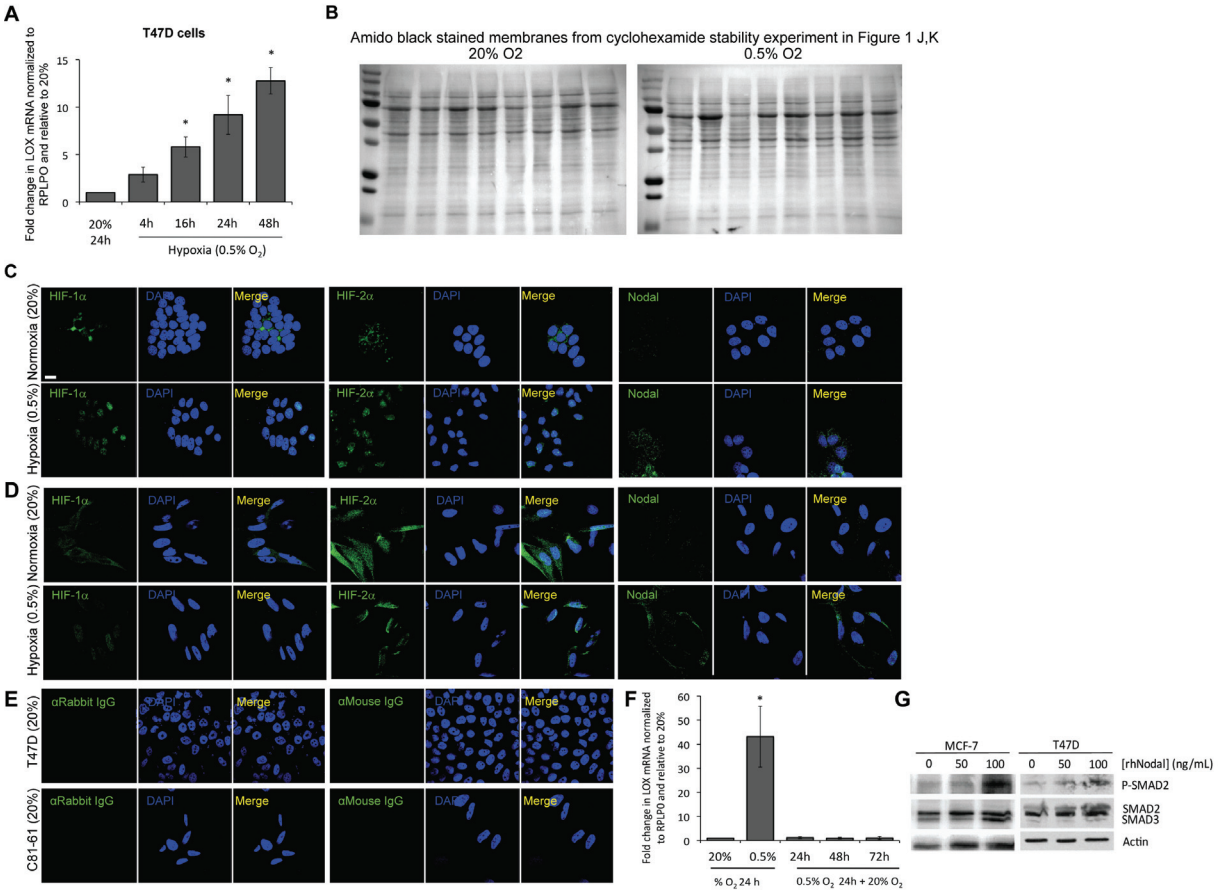
SUPPLEMENTAL FIGURE LEGENDS

Supplementary Figure 1. Validation of experimental treatments and controls pertaining to Figures 1-4. (A) Real-time RT-PCR analysis of *Lox* mRNA in T47D cells exposed to 0.5% O_2 for 0-48 hours. Results validate that LOX is up-regulated over time in response to hypoxia in T47D cells. Gene expression was normalized to RPLPO. Data is presented as mean ± Standard Deviation (SD) relative to 20% O_2 control. Asterisks (*) indicate a significant difference in gene expression compared to 20% O_2 (n=4, $P < 0.05$). (B) Representative amido black stained membranes from cyclohexamide stability assays depicted in Figure 1 to demonstrate equal protein loading. (C) Immunofluorescence of endogenous HIF-1α, HIF-2α or Nodal under normoxic (20% O_2) or hypoxic (0.5% O_2) conditions in T47D cells. (D) Immunofluorescence of endogenous HIF-1α, HIF-2α or Nodal under normoxic or hypoxic conditions in C81-61 cells. (E) IgG isotype controls in T47D and C81-61 cells corresponding to antibodies used for immunofluorescence. (F) Real-time RT-PCR analysis of *Lox* mRNA in T47D cells cultured in 20% O_2 (24 hours), 0.5% O_2 (24 hours), or 0.5% O_2 (24 hours) followed by re-oxygenation in 20% O_2 (24-72 hours). Results validate that hypoxia-induced LOX expression does not persist during re-oxygenation. Gene expression was normalized to RPLPO. Data is presented as mean ± Standard Deviation (SD) relative to 20% O_2 control. Asterisks (*) indicate a significant difference in gene expression compared to 20% O_2 controls (n=6, $P < 0.05$). (G) Immunoblot analyses of phosphorylated SMAD-2 and total SMAD-2/3 in MCF-7 and T47D cells cultured in

20% O_2 (24 hours) in the presence or absence of recombinant human Nodal (rhNodal; 50, 100 ng/mL). MCF-7 and T47D cells respond to rhNodal by increasing SMAD-2 phosphorylation. β-Actin is used as a loading control.

Supplementary Figure 2. Sequence validation of DNA fragments isolated via ChIP. (A) Sequence of Nodal NDE PCR product following chromatin immunoprecipitation (ChIP) on T47D cells using HIF-1α antibody. Primer sequences are bolded and CSL binding sites are underlined. (B) Sequence of Nodal 2nd exon PCR product following chromatin immunoprecipitation (ChIP) on T47D cells using HIF-1α antibody. Primer sequences are bolded.

Supplemental Figure 1



Supplemental Figure 2

A

Nodal NDE amplified region (354bp):

TCTCTCTCTCTCAGAGCCTTTCTCCCACCCCCAGGCTCAGAGTTAGGTCATCTCCAACTCAACTTCA
GCACCAGTGACTCCCCGTGGAGGGGAAGTGGGAAGGTGGGAGGCAGAGGCAGCTGGCCTGGGC
TGATGGGAATCTCTGAGAAGCTTCCGGGGTGGGAGGGGAGGGCACCCTCCTGGTGGTCTCTCTGT
TTGTTTCACAAAACCTTCCCATTCCGCGGGGTTCCAAGGCCAGGCTGTGCCTTCACCTCCTGACCC
TGTGGGAAGCGCAAGCCTCGACTTGGCTCCACCCCAAGAGGCTGGGGGGCCATGGGAGCAGGA
GGAGC**TTGAGCTTCAGAGTCAGACAGACC**

B

Nodal 2nd Exon amplified region (202bp):

CTTCTCCTTCCTGAGCCAACAAGAGGATCTGGCATGGGCTGAGCTCCGGCTGCAGCTGTCCAGCC
CTGTGGACCTCCCCACTGAGGGCTCACTTGCCATTGAGATTTTCCACCAGCCAAAGCCCGACACA
GAGCAGGCTTCAGACAGCTGCTTAGAGCGGTTTCAGATGGACCTATT**CACTGTCACTTTGTCCCAG
GTCACC**