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Supplemental Information

The aryl hydrocarbon receptor promotes differentiation during mouse preimplantational embryo development

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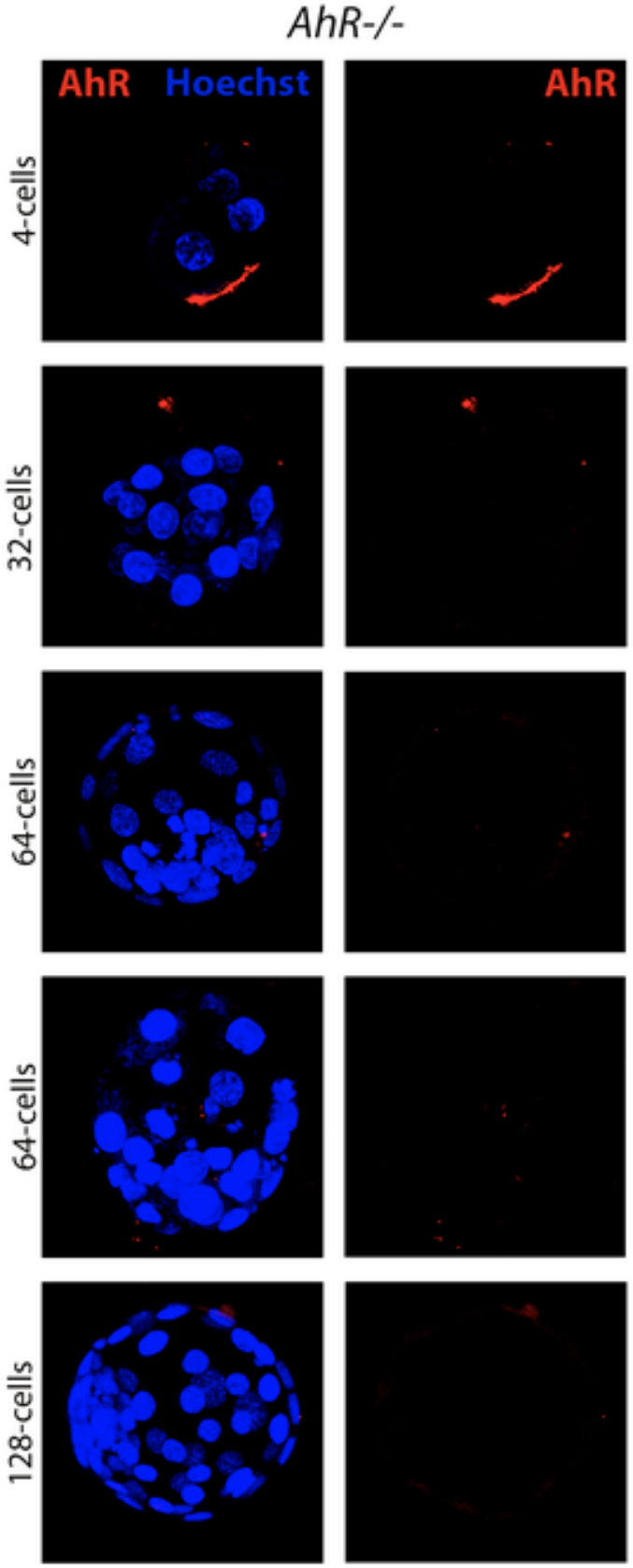
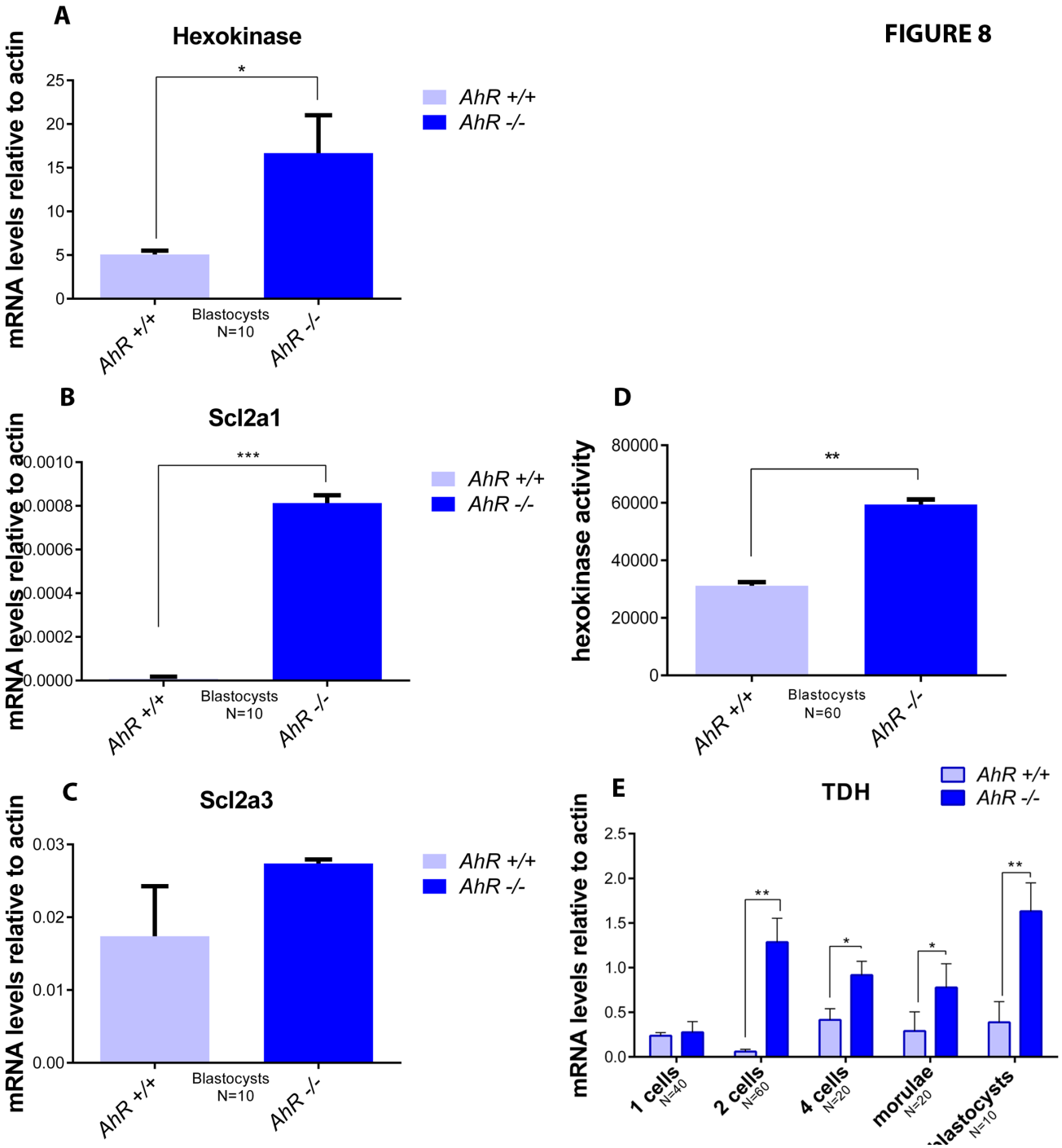


Supplementary Figure 1. AhR expression was determined in 4-cells, 32-cells, 64-cells and 128-cells AhR-null mouse embryos by immunofluorescence. Hoechst staining was used to label cell nuclei. Confocal microscopy was used for detection using an Olympus FV1000 confocal microscope.



Supplementary Figure 2. AhR depletion favors a glycolytic metabolism. mRNA was purified from *AhR+/+* and *AhR-/-* blastocysts and the expression of *Hexokinase-HK* **(A)**, Scl2a1 **(B)** and *Scl2a3* **(C)** was quantified by RT-qPCR. Expression levels were normalized by *β-Actin* and represented as $2^{-\Delta\Delta Ct}$. **(D)** Hexokinase activity was measured using pools of 25-30 embryos at the morulae to blastocyst stages by an enzymatic assay. **(E)** Threonine dehydrogenase expression was analyzed at the indicated developmental stages in *AhR+/+* and *AhR-/-* embryos. *p< 0.05; **p< 0.01; ***p< 0.001. Data are shown as mean ± SD. The experiments were performed, at least, 4 times (2 times for D panel) and the number of embryos analyzed is indicated in the x axis.

Supplementary Table S1
Oligonucleotide sequences used for mRNA expression analysis

Gene	Sequence 5´-- 3´
AhR	Fw: ACATCACCTACGCCAGTCGC Rv: TCTATGCCGCTTGGAAGGAT
Nanog	Fw: CAAGGGTCTGCTACTGAGATGCTCTG Rv: TTTTGTTTGGGACTGGTAGAAGAATCAG
Oct4	Fw: AGAGGGAACCTCCTCTGAGC Rv: CCAAGGTGATCCTCTTCTGC
Sox2	Fw: CGTAAGATGGCCCAGGAGAA Rv: GCTTCTCGGTCTCGGACAAA
β-catenin	Fw: GTGCAATTCCTGAGCTGACA Rv: CTTAAAGATGGCCAGCAAGC
Lats1	Fw: GACCCCAGCTAATGGACAAA Rv: GTTTGCAGTCCAGGGACATT
Lats2	Fw: GTGTCCACAAGATGGGCTTT Rv: CTCCATGCTGTCCTGTCTCA
Cdx2	Fw: TCTCCGAGAGGCAGGTTAAA Rv: GCAAGGAGGTCACAGGACTC
Gata3	Fw: CCGAAACCGGAAGATGTC Rv: TCAGCATGTGGGTGGAGT
Mtch1	Fw: CAGAATCCAGGTTCCCAGTT Rv: TCAGGTACTTCCAGCAGTGG
Scl2a1	Fw: CCCCAGAAGGTTATTGAGGA Rv: GGTTCATCATCAGCATGGAG
Scl2a3	Fw: CTTATGGGATTCGCCAAGAT Rv: TACCAGAATCCCAACAACGA
Tdh	Fw: AAGCTTGTCTTGCCCTTGAA Rv: CTTCCCAAATCGTTTCCTCA
Hexokinase	Fw: TGTTCGAGAAGATGGTGAGC Rv: CATACGTGCTGGACCGATAC