

Supplementary Materials for

Protein kinase A–dependent phosphorylation stimulates the transcriptional activity of hypoxia-inducible factor 1

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Other Supplementary Material for this manuscript includes the following:

(available at www.sciencesignaling.org/cgi/content/full/9/430/ra56/DC1)

Data file S1 (Microsoft Excel format). Summary of MS/MS data for R1a peptide and protein group identification.

Table S1. List of primer and shRNA nucleotide sequences.

Category	Target	ID	Sequence (5'-3')
Mutagenesis Primers	HIF-1 α (human)	S31A-fwd S31A-rev	ATCTCGGCGAGCTAAAGAATCTGAAGTTTTTATGAG CTGGCTGCATCTCGAGAC
		T63A-fwd T63A-rev	GATGAGGCTTGCCATCAGCTATTTG ACAGAGGCCTTATCAAGATG
		S692A-fwd S692A-rev	TAACGTGTTAGCTGTGCGCTTTG GGGCTTCTTGGATGAGATTTTTTC
		T700A-fwd T700A-rev	GAGTCAAAGAGCTGCAGTTCCTGAGG AAAGCGACAGATAACACG
		S727A-fwd S727A-rev	ACATGATGGTGCACTTTTTCAAG TCCATTTTTTCGCTTTCTCTG
		S760/761-fwd S760/761A-rev	AGGATGCAAAGCTGCTGAACAGAATGGAATGGAG TGGATCCCCGGAATTCC
		S809A-fwd S809A-rev	TATACAAGGCGCCAGAAACCTACTGC GGAGCATTAACCTCACAATC
RT-qPCR Primers	<i>HIF1A / Hif1a</i> (human / rat)	Fwd. Rev.	CCACAGGACAGTACAGGATG TCAAGTCGTGCTGAATAATACC
	<i>CA9</i> (human)	Fwd. Rev.	TCTCGTTTCCAATGCACGTACAGC AGTGACAGCAGCAGTTGCACAGT
	<i>Ca9</i> (rat)	Fwd. Rev.	CCTCTCTGCTGGTGACATCC GTCCCACTTCTGTGCCTGTG
	<i>PDK1 / Pdk1</i> (human / rat)	Fwd. Rev.	ACCAGGACAGCCAATACAAG CCTCGGTCACCTCATCTTCCAC
	<i>CD39 (ENTPD1)</i> (human)	Fwd. Rev.	ACTATCGAGTCCCCAGATAATGC CCTGATCCTTCCCATAGCACAA
	<i>CD39 (Entpd2)</i> (rat)	Fwd. Rev.	TTCTGCTACTTTGCGTCCC CCTGTGTCATTCTCCTTGTC
	<i>CD73 (NT5E)</i> (human)	Fwd. Rev.	CCAGTACCAGGGCACTATCTG TGGCTCGATCAGTCCTTCCA
	<i>CD73 (Nt5e)</i> (rat)	Fwd. Rev.	ACCAACACCTTTTCTACACAG CTCAACCTTCAGATAGCCCAG
18S RNA (human / rat)	Fwd. Rev.	CGGCGACGACCCATTGGAAC GAATCGAACCTGATTCCCCGTC	
shRNA	<i>PRKACA</i> (human)	shCa-1	CCGGCAGCCCACTTGGATCAGTTTGCTCGAGCAAAC TGATCCAAGTGGGCTGTTTTTG
		shCa-5	CCGGTAGATCTACCAAGCGCTTTGCTCGAGCAAAG CGCTTGGTGAGATCTATTTTTG

Table S2. Antibodies used for immunoprecipitation and immunoblot assays.*

<u>Antibody target</u>	<u>Commercial source</u>	<u>Catalog number</u>
CREB (IB)	Novus Biologicals	NBP1-90364
FIH-1 (IB)	Novus Biologicals	NB100-428
GST (IB)	Santa Cruz Biotechnology	sc-459
HIF-1 α (IB)	BD Biosciences	610959
HIF-1 α (IP)	R&D Systems	AF1935
KAT3B/p300 (IP)	Novus Biologicals	NB100-616
Penta-Histidine-HRP (IB)	Qiagen	1014992
Phospho-Ser-133 CREB (IB)	Cell Signaling	9191
PKA-Ca (IB)	Novus Biologicals	NB100-92207
PKA-R1a (IP)	Novus Biologicals	NBP1-47935
Rat and human actin (IB)	Santa Cruz Biotechnology	sc-1616
Rat and human CD39 (IB)	Santa Cruz Biotechnology	sc-33558
Rat and human CD73 (IB)	Novus Biologicals	NBP1-85740
Rat HIF-1 α (IB)	Novus Biologicals	NB100-479
Rat PKA-R1a (IP)	BD Biosciences	610166
Yeast Gal4 DBD (IB)	Santa Cruz Biotechnology	sc-577

* Unless otherwise stated, all antibodies were raised against proteins of human origin.