SUPPLEMENTAL TABLES

Table S1. Relative catalase and GPx activity for specific cells and tissues. One unit of catalase activity is defined as the amount of enzyme that decomposes 1 mmol H_2O_2/min. One unit of GPx activity is defined as μmol NADPH oxidized/min. Cited references describe the methods used for determining enzyme activity in each study. To aid comparisons, values are grouped together by study (shading) and tissue/tumor type.

Tissue/Tumor Type	Cell Line Or Tissue	$k_{catalase}$ (U/g protein)	k_{GPx} (U/g protein)	Reference
Glioma	A172	3.7 ± 1.3	5.3 ± 0.6	(105)
Glioma	D54	6.4 ± 0.3	7.6 ± 0.7	(105)
Glioma	U87	5.0 ± 0.4	16.3 ± 1.3	(105)
Glioma	U118	5.8 ± 1.0	8.6 ± 0.7	(105)
Glioma	U251	20.9 ± 1.3	8.9 ± 2.6	(105)
Glioma	U563	17.9 ± 2.2	6.4 ± 0.8	(105)
Glioma	A172R	3.9 ± 0.7	10.0 ± 0.9	(105)
Glioma	RG	16.3 ± 5.1	2.0 ± 0.6	(105)
Glioma	N89-441R	12.3 ± 3.2	6.0 ± 2.5	(105)
Glioma	C6	11.8 ± 0.4	40 ± 4	(60)
Glioma	Human GBM	N.M.	17.71 ± 3.9 (U/g wet tissue)	(92)
Normal Brain		N.M.	45.3 ± 6.9 (U/g wet tissue)	(92)
Hepatoma	Human hepatocellular carcinoma	99 ± 5	N.M.	(11)
Normal Liver		339 ± 12	N.M.	(11)
Hepatoma	HepG2	33 ± 3	N.M.	(23)
Hepatoma	Hepa 1-6	20 ± 1	N.M.	(59)
Normal Liver		99 ± 12	N.M.	(59)
Colon Carcinoma	CT26	35 ± 2	N.M.	(59)
Normal Colon		43 ± 2	N.M.	(59)
Fibroblasts	NIH 3T3	72 ± 5	N.M	(59)

Table S2. Experimental observations for relative HIF1α expression and accumulation over time in chronic conditions of hypoxia (top) and in chronic conditions associated with ischemia or reperfusion (below).

Cell/Tissue type	Time to max. HIF1α (hrs of hypoxia)	O_2	Measured duration of elevated HIF1α	Reference
Cervical cancer, HeLa	4	1%	Over 7 days, loss greatest in first 48 hrs	(35)
Fibrosarcoma, HT1080	4	1%	Over 7 days, loss greatest in first 72 hrs	(35)
Prostate carcinoma, DU145	1	1%	Elevated HIF1α for 4-8 hrs	(52)
Renal cell carcinoma (overexpressing VHL) RCC4-pVHL	4	1%	Over 4 days, loss greatest in first 24 hrs	(35)
Glioblastoma, U87MG	no max detected	0.2%, 2%	1 h to 18 hrs	(97)
Embryonic kidney, HEK293	4	1%	Over 5 days, loss greatest in first 72 hrs	(35)
Embryonic kidney, HEK293	4 to 8	1%	Over 3 days, loss greatest after 8 hrs	
Lung fibroblasts, CCL39	4	1%	Over 7 days, loss greatest in first 48 hrs	(35)
Dermal fibroblasts, NHF	4	1%	Over 7 days, loss greatest in first 72 hrs	(35)
Rat cortical samples	6	10% (live rats)	Over 21 days, 9x-10x elevated HIF1α at 6 hrs, 12 hrs, 1 day, 4 days; 5x HIF1α at 7 and 14 days.	(28)
Mice whole brain, nuclear extracts	3	8% (live mice)	Over 6 hrs	(14)
Cell/Tissue Type	**Time to max. HIF1α (hrs)**	**Condition**	**Measured duration of elevated HIF1α**	**Reference**
Rat cortical samples	1 to 24	N.M.; ischemia, 11-13 min	Over 7 days, highest HIF1α levels between 1 hr and 1 day	(29)
Squamous cell carcinoma, 22B	no max reached	addition of 10 mM pyruvate	Increasing HIF1α levels from 2 to 24 hrs	(63)
Coculture, neurons and astrocytes	0	reoxygenation	HIF1α levels returned to baseline by 6 hrs reoxygenation	(95)
Rat basal ganglia	72	blood injection	4 hrs to 7 days, peak at 3 days	(51)

SUPPLEMENTAL FIGURE LEGENDS

Supplemental Figure 1. HIF1α levels vs. time for cells in the presence or (hypothetical) absence of ROS, where ROS is represented by changes in Fe^{2+} and Asc or 2OG concentrations. ROS may be represented by a decrease (dotted line) or increase (dashed line) in $[Fe^{2+}]_0$ and $[Asc]_0$ or $[2OG]_0$. $[HIF1\alpha]_0$ = 1 μM. (A) HIF1α levels vs. time for cells exposed to ROS, compared to cells without ROS effects. Values are normalized to the maximum $[HIF1\alpha]_{unhydroxylated}$ for each condition. (B) Absolute HIF1α levels vs. time for cells exposed to ROS, where ROS is represented by changes in $[Fe^{2+}]_0$ and $[2OG]_0$. (C) HIF1α levels vs. time in tumors with SDH deficiency, where $[SC]_0$:$[PHD2]_0$ = 500. Values are normalized to the maximum $[HIF1\alpha]_{unhydroxylated}$ for each condition. (D) Absolute HIF1α levels vs. time in tumors with SDH deficiency, where $[SC]_0$:$[PHD2]_0$ = 500. The time step used was 12 seconds; the peaks in $[HIF1\alpha]_{unhydroxylated}$ for the $[SC]_0$:$[PHD2]_0$ = 500:1 in (B), (C) and (D) last for ~1 min. The ratio of $[SC]_0$:$[PHD2]_0$ = 500 was used to represent maximum values found in vitro here, while main Figure 2C showed hypothetical SDH deficiency conditions in vivo, where $[SC]_0$:$[PHD2]_0$ = 5. Further details of this representation can be found in reference (78).

Supplemental Figure 2. (A) Model results show the effects of H_2O_2 on cellular hypoxic response via HIF1α expression from 0 to 144 hours (6 days) in tumor cells exposed to ROS and tumor cells with succinate effects modeled by signaling O_2 depletion. Conditions correspond to those detailed in Table 2. For the succinate O_2-depletion model, $[SC]_0$ = 5.0 μM; k_{SC} = 0.001 min^{-1}. $[HIF1\alpha]_0$ = 1 μM. (B) Cumulative concentration of H_2O_2 produced, transported and metabolized for a tumor cell over 72 hours. Negative values refer to loss of H_2O_2. The

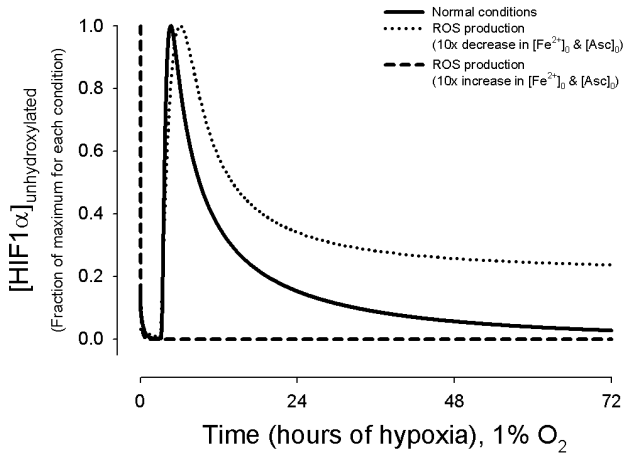
intracellular concentration of H_2O_2 rapidly reached an equilibrium level of 0.25 μM from [H_2O_2] = 0.2 μM (< 1 min).

Supplemental Figure 3. Model results show the effects of H_2O_2 on cellular hypoxic response via HIF1α expression in ischemic cells, ischemic cells exposed to ROS generated by reperfusion in constant hypoxia, and ischemic cells exposed to reperfusion in hypothetical normoxic conditions for 0-12 hrs (A) and 0-72 hrs (B). Reperfusion would likely produce intermediate HIF1α expression levels between those shown for constant normoxia and constant hypoxia. $[HIF1\alpha]_0$ = 1 μM. (C) Long-term expression of HIF1α in ischemic cells, and ischemic cells exposed to ROS generated by reperfusion, where ROS is represented both by H_2O_2 and an increase in Fe^{2+} and Asc. See Figure 6A, for the same conditions, shown from 0-12 hrs. (D) Cumulative change in [H_2O_2] due to production, transport and metabolism for the conditions of ischemia with reperfusion, in hypoxia. ~20 mM [H_2O_2] total was produced (compared to ~36 mM in a tumor cell for 72 hrs), while the graph shows the significance of transport and metabolism. The corresponding H_2O_2 intracellular concentration level reached a value near 2.0 μM, for all times > 30 minutes (see Supplemental Figure 4A). Ischemia and reperfusion conditions correspond to those detailed in Table 2.

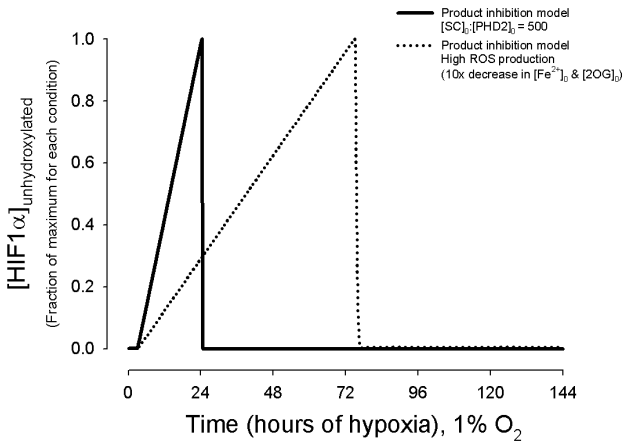
Supplemental Figure 4. (A) Model results show the effect of $[H_2O_2]_{extracellular}$ on HIF1α levels over time in the model's default case representing tumor conditions. (B) Relative expression of HIF1α in the model (diamonds) is compared to experiments measuring relative HIF1 activation through luciferase activity as a function of different H_2O_2 concentrations that were added to the media of cultured HepG2 (human hepatocarcinoma) cells [14] (bars). The experiments

supplemented the media with 2 nM of insulin, expect for the indicated initial value with no added H_2O_2 (- Ins). Results from the experiments and the model were scaled relative to their maximum increase in luciferase activity or HIF1α expression, respectively.

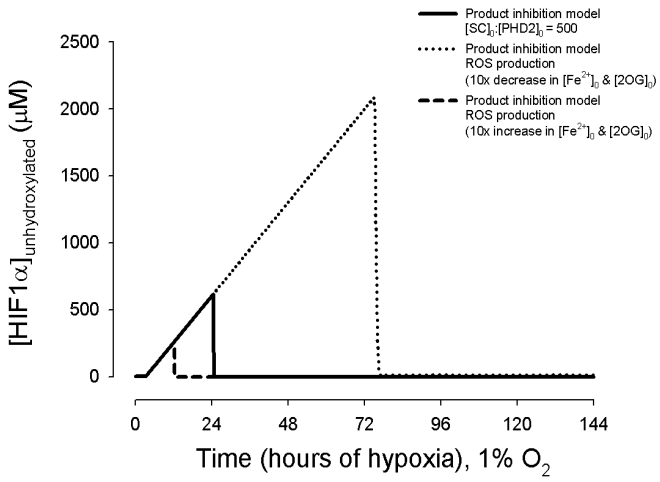
Supplemental Figure 1.



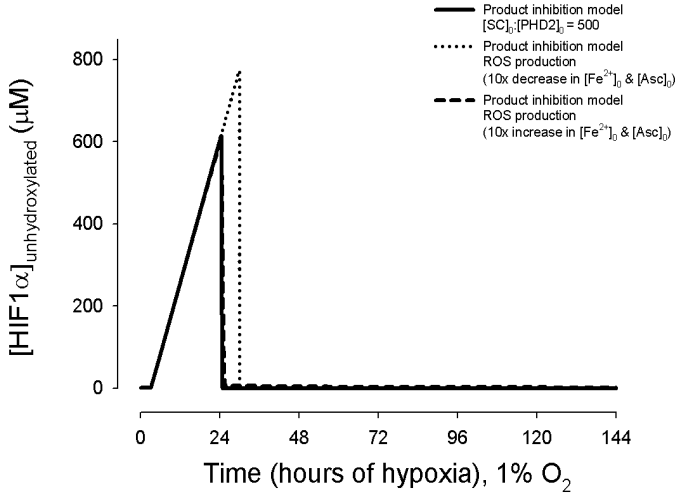
A



B

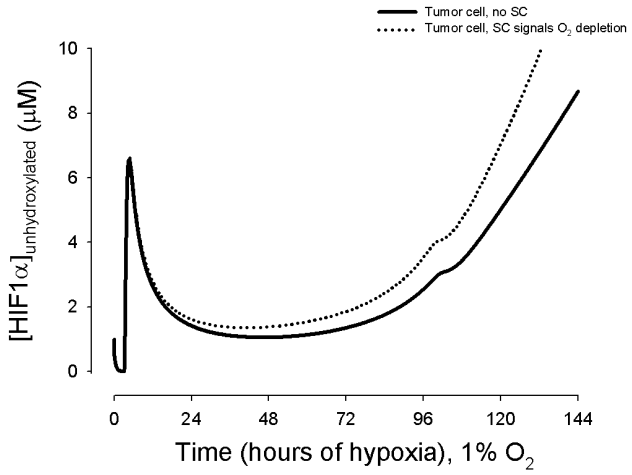


C

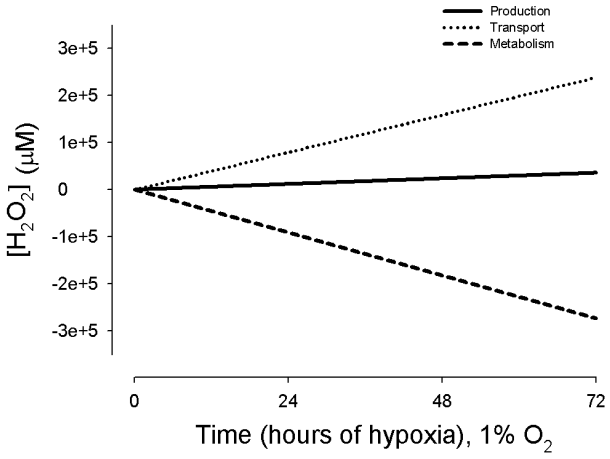


D

Supplemental Figure 2.

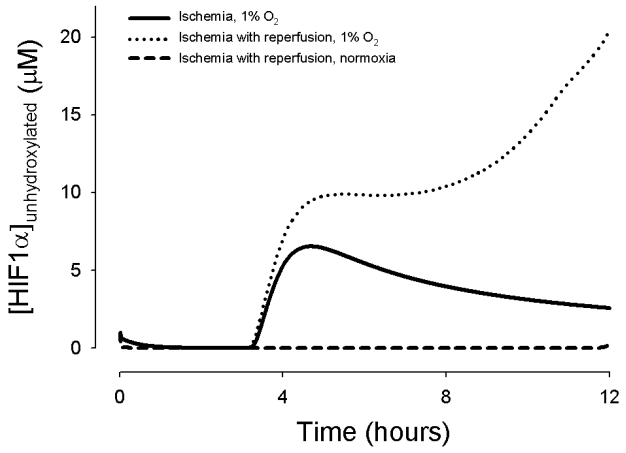


A

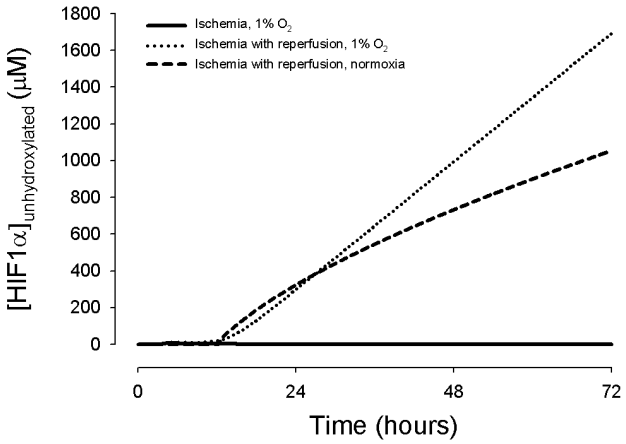


B

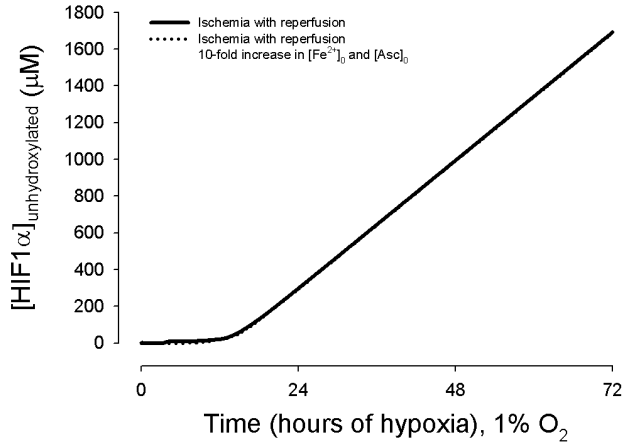
Supplemental Figure 3.



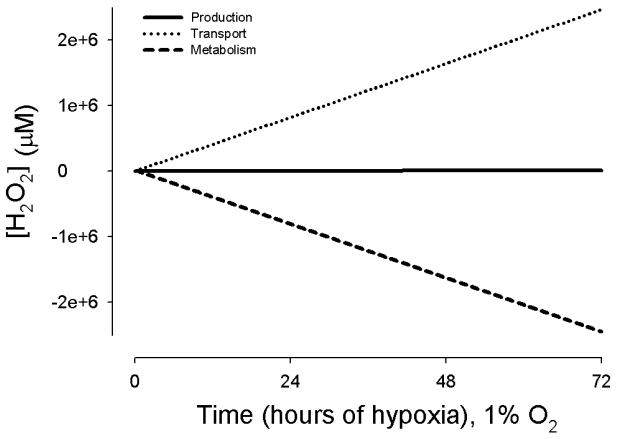
A



B

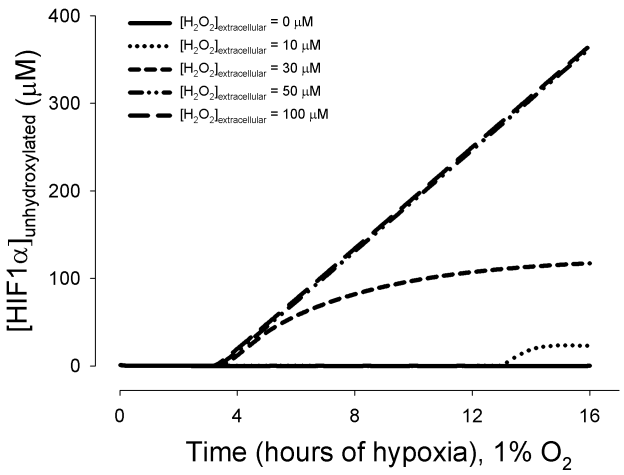


C

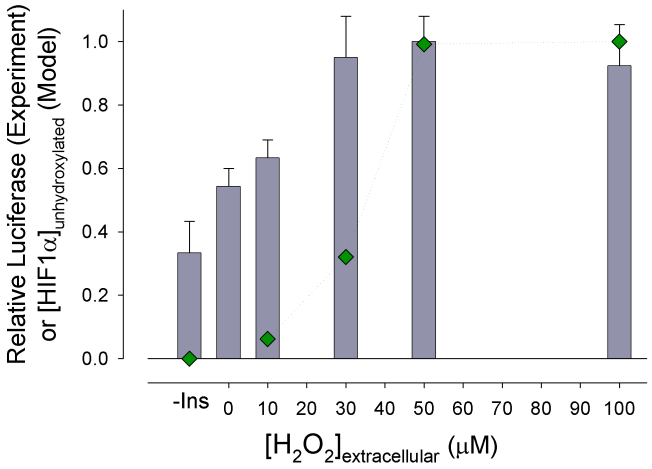


D

Supplemental Figure 4.



A



B