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Title: Protein kinase CK2 activates Nrf2 via autophagic degradation of Keap1 and activation of AMPK in human cancer cells

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Running Title: Regulation of Nrf2 by CK2

Keywords: CK2, Nrf2, AMPK, Keap1, antioxidants

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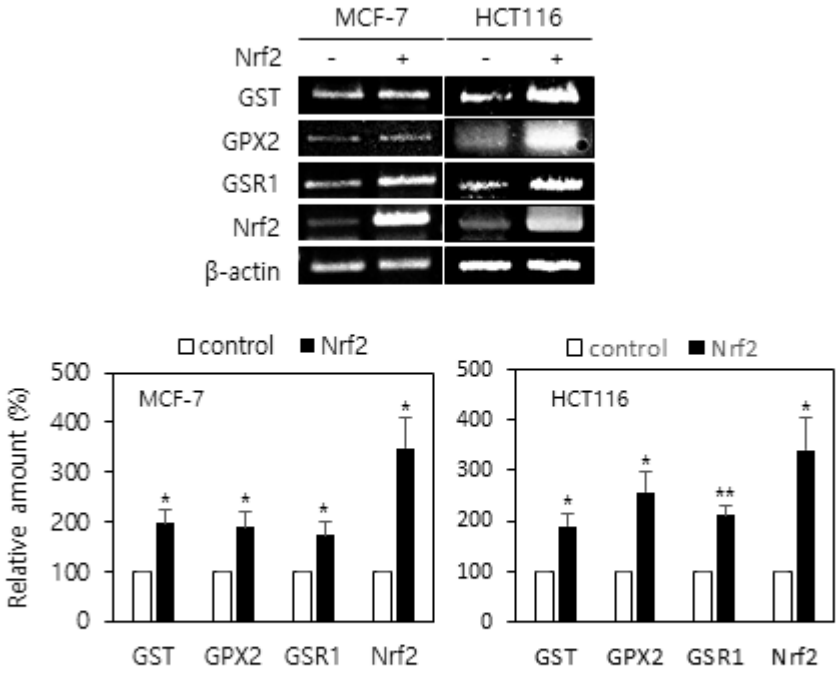
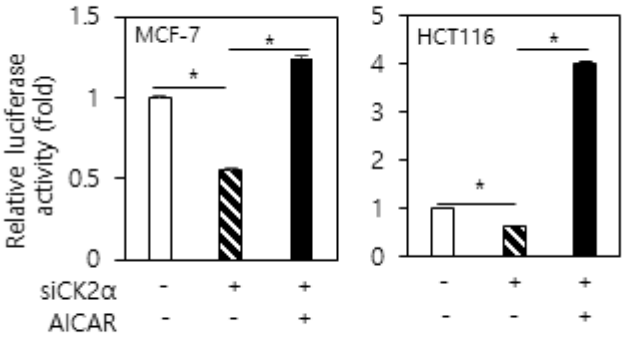
sFig. 1.


MCF-7
HCT116
Nrf2
GST
GPX2
GSR1
Nrf2
β-actin
control
Nrf2
Relative amount (%)
MCF-7
HCT116
GST
GPX2
GSR1
Nrf2

sFig. 2


Relative luciferase activity (fold)
MCF-7
HCT116
siCK2α
AICAR

Supplementary Figure legends

sFig. 1. Nrf2 overexpression upregulates GST, GPX2, and GSR1 in human cancer cells. Cells were transfected with the Nrf2 cDNA for 48 h. Total RNA was extracted from the cells, and RT-PCR was performed using specific primers. PCR products were resolved on a 2% agarose gel (upper panel). Graphs show the quantification of the mRNA level of each gene relative to that of *β-actin* (bottom panel). Data are shown as means ± SEM. **P* <0.05; ***P* <0.01.

sFig. 2. CK2 regulates the Nrf2 activity via AMPK in human cancer cells. MCF-7 and HCT116 cells were co-transfected with the ARE luciferase construct and CK2α siRNA into in the absence or presence of 100 μM AICAR. The firefly luciferase activity was measured 24 h after transfection and normalized to *Renilla* luciferase activity.

Supplementary Table 1. Primers used for RT-PCR analyses

Gene		Primer sequence
CK2α	Forward	5′-AAGACCCTGTGTCACGAACCC-3′
	Reverse	5′-GGCTCCTCCCGAAAGATCATAC-3′
GST	Forward	5’-GACCTCACCCAGGTAATGGA-3’
	Reverse	5’-ATCCGTGCTCCGACAAATAG-3′
GPX2	Forward	5’-ATGGCTTTCATTGCCAAGTC-3’
	Reverse	5’-TTTTTGGACAAGGGTGAAGG-3′
GSR1	Forward	5′-ACTTGCCCATCGACTTTTTG-3′
	Reverse	5’-GTAGGGTGAATGGCGACTGT-3′
NRF2	Forward	5’-CGGTATGCAACAGGACATTG-3′
	Reverse	5’-ACTGGTTGGGGTCTTCTGTG-3′
β-actin	Forward	5′-TCCCTGGAGAAGAGCTACGA-3′
	Reverse	5′-AGCACTGTGTTGGCGTACAG-3′

GST, glutathione S-transferase; GPX2, glutathione peroxidase 2; GSR1, glutathione reductase 1