Supplementary Figure Legend

Figure S1 Recombinant his-RXRα was unable to interact with GST-hNRF2 mutant fusion protein that lacked residues 209–316 between the Neh5 and Neh6 domains. (A) NRF2 mutant GST fusion proteins were expressed in *Escherichia coli* DH5α cells and purified with glutathione-Sepharose beads The same amounts of GST-hNRF2$^{\Delta DIDLID}$, GST-hNRF2$^{\Delta DIDLID+\Delta 209-316}$ (lane 2), GST-hNRF2$^{17-338}$ (lane 4) fusion protein, GST protein (lane 5), or double amount of GST-hNRF2$^{\Delta DIDLID+\Delta 209-316}$ (lane 3), were incubated with purified his-RXRα. The beads were washed five times before collection by centrifugation. The bead-bound proteins were separated by SDS-PAGE and subjected to immunoblotting using antibodies against either RXRα (upper blot) or GST (lower blot). GST-hNRF2$^{\Delta DIDLID}$ (lane 1) and GST-hNRF2$^{217-338}$ (lane 4) were used as positive controls. GST (lane 5) was used as negative control. The results presented are typical examples from three separate experiments. (B) HEK293T cells were transfected with various pEGFPC1 plasmids encoding GFP-hNRF2$^{17-338}$ (lane 1), GFP-hNRF2$^{339-605}$ (lane 2), and GFP-hNRF2$^{\Delta DIDLID+\Delta 209-316}$ (lane 3). Twenty-four hr post-transfection, the whole cell extracts were incubated with purified His-RXRα. The mixture was subjected to immunoprecipitation using antibody specific to RXRα. The immunoprecipitates were separated by SDS-PAGE and immunoblotted using antibodies against GFP. NRF2 mutant proteins are indicated by * respectively. The molecular mass in kD is indicated. IB, immunoblot.

Figure S2 HEK293 cells were transfected with pEGFP-C1 or pDsRed-C1. Forty-eight hr post transfection, the cells were fixed and examined. Nuclei were stained by DAPI (blue). At least 20 cells were observed per experiment for an equal transfection. Scale bar, 10 μm. Results were confirmed by repeated experiments.

Figure S3 RXRα interacted with Nrf2 *in vivo*. COS7 cells (African Green Monkey SV40-transfected kidney fibroblast) (ATCC, China) were transfected with pcDNA3.1/mNrf2$^{\Delta DIDLID}$-V5. Twenty-four hr later, a fraction of total cell lysate (250 μg protein in 1 ml) was incubated with anti-RXRα antibody 4 hr at 4°C, followed by overnight incubation with protein G-agarose. The IgG was used as negative control. After washing, the immunoprecipitates were analysed by immunoblotting with antibody specific to Nrf2 (upper blot) or RXRα (lower blot). Input, 5% of the cell lysate used for immunoprecipitation; SN, 5% of the supernatant after the immunoprecipitation; Wash, 5% of the supernatant solution after the immunoprecipitates were washed four times; Beads, immunoprecipitates after the washing procedure. IB, immunoblot. The results presented have been replicated over three separate experiments.

Figure S4 tBHQ increased the expression of AKR1C in MCF7 cells. MCF7 cells were exposed to 20 μM tBHQ for 24 hr (lane 2). Cells treated with DMSO (0.1% v/v) were used as negative controls (lane 1). The whole cell lysate was analyzed by Western immunoblotting with antibodies against AKR1C or Actin. Actin was used as loading control. Results from two experiments.

Figure S5 BHA up-regulated ARE-gene battery in mouse small intestine and liver. WT mice were given BHA i.g. daily at a dose of 200 mg/kg for 3 days; and corn oil (vehicle) was used as negative control. Cell extracts from small intestine (A) and liver (B), were analyzed by Western immunoblotting against Nqo1 or Gstm1 (2). Each lane represents a sample from a

single mouse. Data represent immunoprecipitation results from three separate experiments (n = 3).

Figure S6 The interaction of mNrf2 with RXRα in MCF7-RXRα cells. MCF7-RXRα cells were transfected with pcDNA3.1/V5-mNrf2$^{\Delta DIDLID}$ (3). Twenty-four hr later, the cell lysate was immunoprecipitated with antibody specific to V5. After washing, the immunoprecipitates were analysed by Western immunoblotting with antibody specific to RXRα. In parallel, the immunoprecipitation was carried out with IgG as negative control. SN, supernatant; Wash, the supernatant solution after the washes of the beads; Beads, immunoprecipitation after the washing procedure. Results from three separate experiments.

Figure S7 The binding of NRF2 to the ARE sites in MCF7 and MCF7-RXRα cells. Cells were treated with 20 µM tBHQ for 6 hr. The binding of NRF2 to the ARE sites in the promoter of *HO-1* was detected by Chip analysis as described by Materials and Methods. *GAPDH* was used as negative control. PCR reactions were not saturated. Blot represents results from over three independent experiments.