Supplementary Figure Legends

Supplementary Figure S1. PKR does not affect doxorubicin-induced cell cycle arrest. PKR $^{+/+}$ and PKR $^{-/-}$ MEFs were left untreated (con) or treated with 1 μM doxorubicin for the indicated time periods and subjected to FACS analysis after propidium iodide staining. The percentage of cells in various phases of the cell cycle is indicated. Data are representative of five independent experiments (n=5).

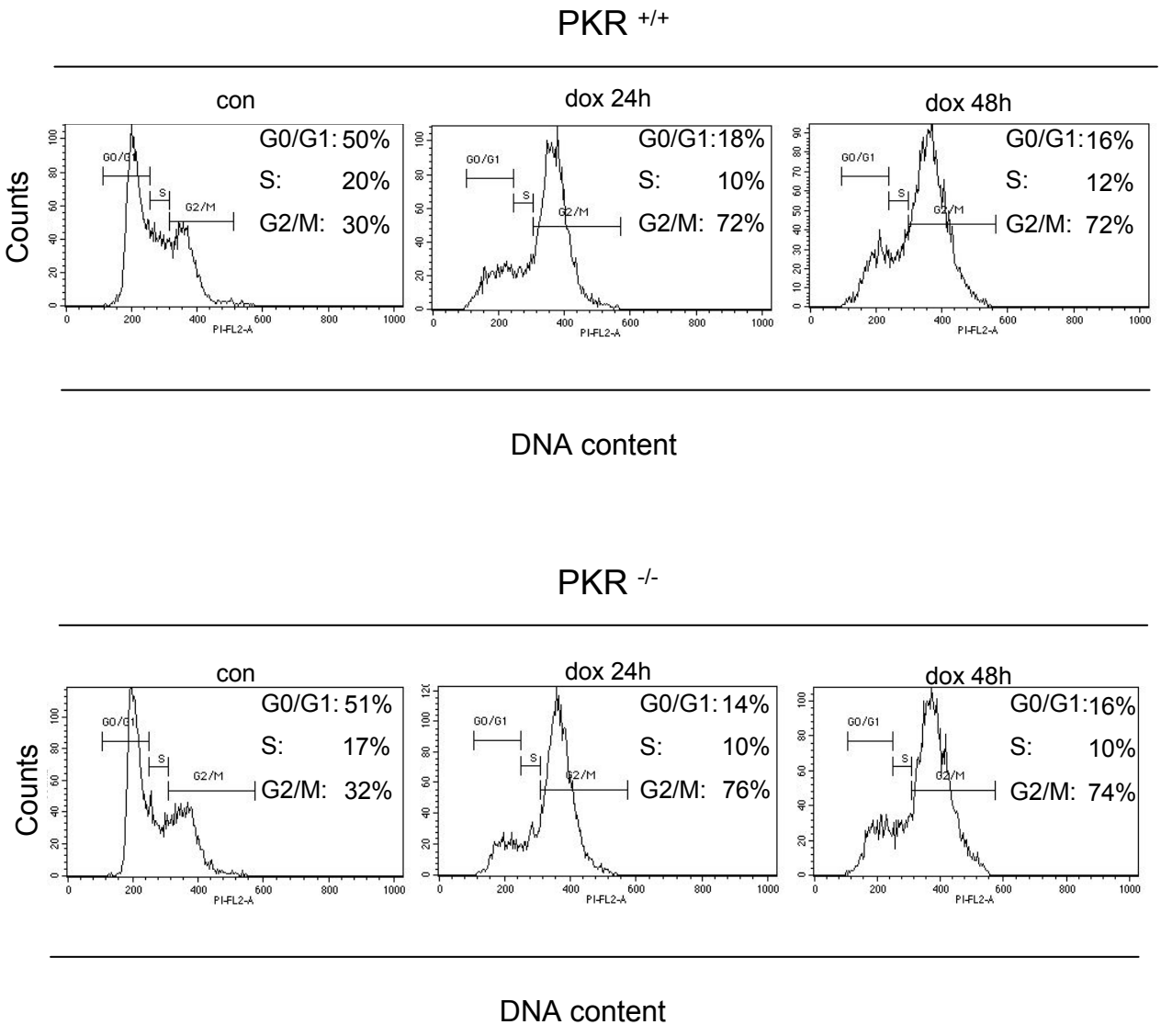
Supplementary Figure S2. PKR induces the expression of eIF2α downstream genes and induces eIF2α phosphorylation in human cancer cell lines upon doxorubicin treatment. **(A)** PKR $^{+/+}$ and PKR $^{-/-}$ MEFs were left untreated (con) or treated with 1 μM doxorubicin (dox) for the indicated time periods. Protein extracts (50 μg) were subjected to immunoblot analysis for phosphorylated eIF2α (panel a), ATF4 (panel b) and actin (panel c). Data are representative of two independent experiments, (n=2). **(B)** Human fibrosarcoma HT1080 cells (panels a-d), human alveolar lung carcinoma A549 cells (panels e,f) or human non-small cell lung carcinoma H1299 cells (panels g,h) were left untreated (lanes 1, 3) or treated with 1 μM doxorubicin (dox) for the indicated time periods (lanes 2, 4). Protein extracts (50 μg) were subjected to immunoblot analysis for phosphorylated eIF2α (panels a,e,g), total eIF2α (panels b,f,h), phosphorylated PKR at T446 (panel c) and total PKR (panel d). Data are representative of two independent experiments, (n=2). The ratios of the levels of the phosphorylated proteins to the total levels normalized to their respective controls are indicated.

Supplementary Figure S3. A conditionally inducible form of PKR leads to JNK activation. HT1080 cells expressing a conditionally active form of PKR as a fusion protein with GyrB (Ref. 6) were treated with 100 ng/ml coumermycin for the indicated times. In untreated control cells the equivalent amount of DMSO in which coumermycin was dissolved was added. Protein extracts (50 μg) were subjected to immunoblot analysis for phosphorylated eIF2α (panel a), actin (panel b), phosphorylated JNK (panel c), total JNK1 (panel d) and GyrB.PKR (panel e). Data are representative of two independent experiments, (n=2).

Supplementary Figure S4. PKR induces death independently of ATM and/or DNA-PK in doxorubicin treated cells. **(A)** PKR $^{+/+}$ and PKR $^{-/-}$ MEFs were treated with 30 μM wortmannin (wort; lanes 2, 6), 1 μM doxorubicin (dox; lanes 3, 7) or both drugs (w + d; lanes 4, 8). Protein extracts enriched in histones (50 μg) were subjected to immunoblot analysis for phosphorylated H2AX at S139 (panel a) and total H2AX (panel b). In untreated control cells and doxorubicin treated cells (lane 1, 3, 5, 7) the equivalent amount of DMSO in which wortmannin was dissolved was added. DMSO or wortmannin were added an hour before doxorubicin. Data are representative of two independent experiments, (n=2). **(B)** PKR $^{+/+}$ and PKR $^{-/-}$ MEFs treated with 30 μM wortmannin (wort), 1 μM doxorubicin (dox) or both drugs (wort + dox) for the indicated time periods and subjected to propidium iodide and analysis of death by FACS. Control cells and doxorubicin treated cells received the equivalent amount of DMSO in which wortmannin was dissolved. DMSO or wortmannin were added an hour before doxorubicin. Cell death is represented by the percentage (%) of cells in $SubG_1$. Histograms represent the mean

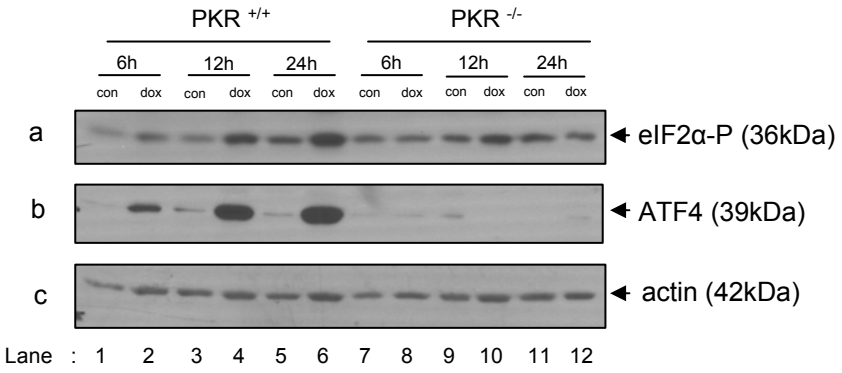
cell death from three independent experiments (n=3). Group statistical significance of the differences as calculated by *ANOVA* is with *P<0.0001 **(C)** PKR $^{+/+}$ and PKR $^{-/-}$ MEFs were treated with 2 μM KU55933, 1 μM doxorubicin (dox) or both drugs (dox + KU55933) for the indicated time periods followed by propidium iodide staining and FACS analysis. Control cells and doxorubicin treated cells received the equivalent amount of DMSO in which KU55933 was dissolved. DMSO or KU55933 were added an hour before doxorubicin. Cell death is represented by the percentage (%) of cells in $SubG_1$. Histograms represent the mean cell death from two independent experiments (n=2). Group statistical significance of the differences as calculated by *ANOVA* is with *P<0.0001.

Supplementary Figure S1.



Supplementary Figure S2.

A



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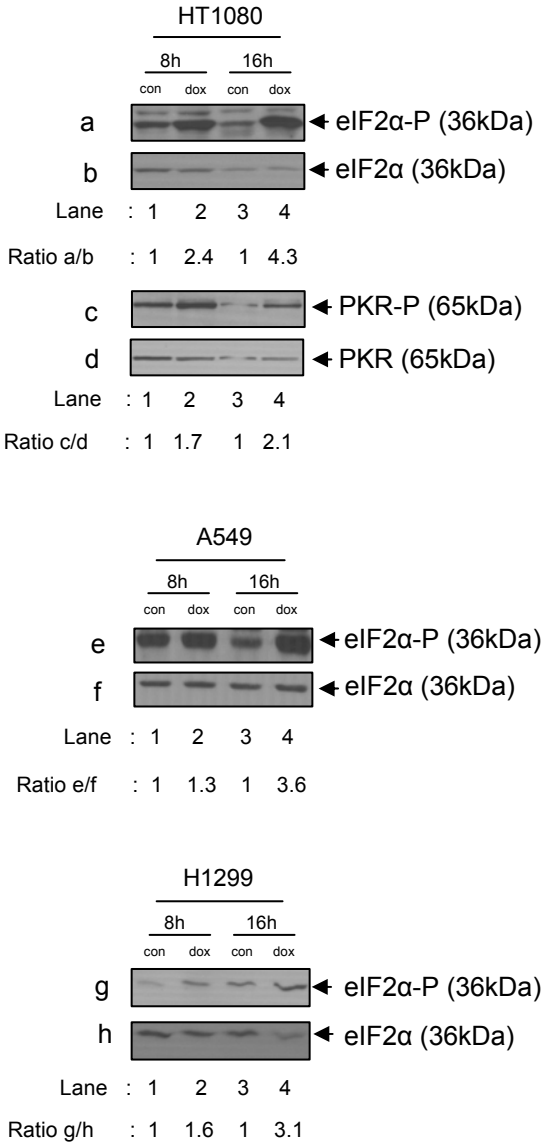
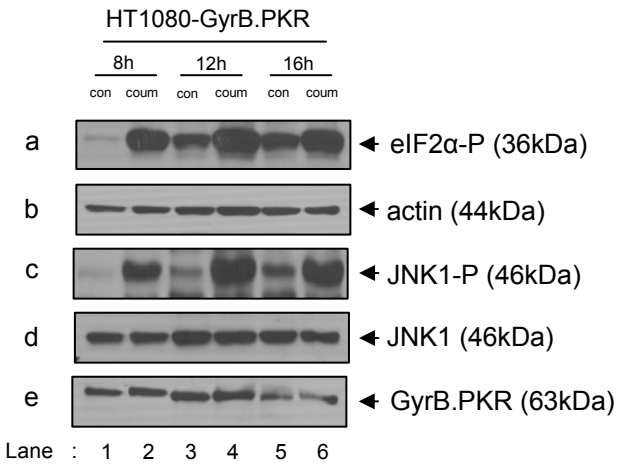


Supplementary Figure S3.



Supplementary Figure S4.