

## Supplementary Legends

**Supplementary Figure1.** The immunofluorescence figures of nuclear accumulation of NF- $\kappa$ B p65 in A431 and A431<sup>Cx26<sup>-/-</sup></sup> cells 6 h after 5Gy X-ray irradiation.

In the western blot experiment, the expressions of NF- $\kappa$ B p65 were significantly different between A431 cells and A431<sup>Cx26<sup>-/-</sup></sup> cells 6 h after 5Gy X-ray irradiation. Therefore, in the immunofluorescence experiment, we selected the X-ray dose of 5Gy. The time point is still 6 h after irradiation. In the immunofluorescence experiment, we found that the nuclear accumulations of NF- $\kappa$ B p65 showed significantly different between sham and irradiated A431 cells. But the nuclear accumulations of NF- $\kappa$ B p65 were not significantly different between sham and irradiated A431<sup>Cx26<sup>-/-</sup></sup> cells. We think that these results are related to the low expression of Cx26 in A431<sup>Cx26<sup>-/-</sup></sup> cells, which proves that different expression levels of Cx26 have a certain regulatory effect on NF- $\kappa$ B signaling pathway.

**Supplementary Figure2.** The relative optical density analysis results of p-ERK and ERK expression in A431, A431<sup>Vector</sup> and A431<sup>Cx26<sup>-/-</sup></sup> cells after irradiation. (A) The comparison of ERK protein expression levels of A431 cells, A431<sup>vector</sup> cells and A431<sup>Cx26<sup>-/-</sup></sup> cells. (B) The comparison of p-ERK expression levels of A431 cells, A431<sup>vector</sup> cells and A431<sup>Cx26<sup>-/-</sup></sup> cells.

As can be seen from the supplementary Figure 2, after irradiation, the ERK expression

level of A431<sup>Cx26<sup>-/-</sup></sup> cell was generally lower than that of A431 and A431<sup>vector</sup> cell. Moreover, when the irradiation dose was 2Gy and 5Gy, the expression of p-ERK in A431<sup>Cx26<sup>-/-</sup></sup> cell was significantly lower than that in A431 and A431<sup>vector</sup> cell. These results indicate that the knock-out of Cx26 has a certain influence on the expression and phosphorylation of ERK in A431 cells.