



FIGURE S5. Inhibitory effects of FGF12 on radiation-induced apoptosis in IEC6 cells. The apoptosis of IEC6 cells was assessed by microscopic examination of nuclear morphology using Hoechst 33258, as described previously (18). IEC6 cells were plated in a 3.5-cm dish at a density of 3×10^4 cells/well in 2.0 ml of complete medium 16 h before FGF administration. Then the cells were irradiated with X-rays 24 h before fixation with 1% glutaraldehyde and the condensed chromatin in the nuclei was visualized by staining with 20 $\mu\text{g}/\text{ml}$ of Hoechst 33258. The percentage of apoptotic cells was determined from the examination of 2,000 cells in 10 fields using a fluorescence microscope. Values represent the mean \pm SD. $**P < 0.01$; $***P < 0.001$. Similar findings were observed in at least two independent experiments. **A**, IEC6 cells were treated with 10 ng/ml FGF12B and irradiated with 10 or 20 Gy. FGF12B-treatment significantly reduced apoptosis compared with non-untreated controls at 24 h after irradiation at 10 and 20 Gy. **B**, IEC6 cells were cultured with complete medium containing 1, 10, or 100 ng/ml of FGF12B with or without 5 $\mu\text{g}/\text{ml}$ of heparin 24 h before irradiation at 20 Gy. Each value is shown as the inhibition rate compared with the apoptosis of each control. Heparin significantly decreased the inhibition rate of apoptosis when FGF12B was added to the culture at 10 and 100 ng/ml.