Does ultraviolet B can induce programmed cell death in plant cells?
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Increasing of encompassing level of ultraviolet B (UVB), resulting from lessening of stratospheric ozone layer, has sufficient damaging effect on higher plants. Solar UV wavelengths can reduce plant genome stability, growth and productivity. It was established that short wave ultraviolet spectrum (UVC) can induce processes of programmed cell death in plant cells, but does UVB can stimulate apoptosis in plant remains an open issue to date. We examined apoptotic reaction on UVB impact on plant cells using Nicotiana tabacum cell line BY-2. Cells were exposed with high doses of UVB irradiation, and detection of apoptotic features using most adequate methods was performed. The influence of different UVB doses on viability of BY-2 cells treatment after several irradiation time intervals was studied. UV treatment was performed with covering filter to eliminate UVC irradiation and attenuate irradiation of short wave UVB spectrum (25, 59, 99 and 208 kJ at 305 nm). Development of programmed cell death was estimated with identification of apoptotic morphology using fluorescent microscopy (fluorescein diacetate/propidium iodide/DAPI staining and acridine orange/DAPI staining), nucleosomal DNA fragmentation and TUNEL assay. It was found that UVB effects have dose-dependent character. Cells with high level of vacuolization and shrinkage of cytoplasm were occurred in considerable amount in all irradiated samples. Compared to control, UVB irradiated cells characterized by presence of condensed chromatin at the nuclear periphery (pre-apoptotic nuclei), forming of micronuclei (apoptotic-like nuclei), stretched and lace nuclei. Specific nucleosomal DNA fragmentation after the UVB irradiation was detected also using agarose gel electrophoresis. Additionally, UVB induced apoptosis was verified using TUNEL assay. Positive staining was detected in all irradiated samples compared to non-treated cells. All used assays indicated an increase of apoptotic cells amount in UVB-treated samples that correlates with elevation of irradiation dose. From data obtained we can suppose that UVB overexposure induces programmed cell death in plant cells in dose dependent manner.