expressed differently, and the photosynthetic characteristics of the two progenies changed markedly after carbon-ion irradiation. From mutagenic effects we found that the low dose of carbon ion irradiation caused hormetic effects on the photosynthetic efficiency, thermal dissipation ability and transcriptional regulation of the light harvesting complex II antenna proteins in *Scenedesmus quadricauda*.

4 - 37 DNA Damage in Bone Marrow Mononuclear Cells of Mice Detected by Two Dimensional Comet Assay

Wang Zhuanzi, Liu Fang, Li Wenjian, Wei Wei and Dang Bingrong

Heavy ion irradiation attract a large interest for two applications: radiotherapy and space radiation protection in manned space missions. Exposure to heavy ions radiation results in multiple effects through DNA damage induction. Single-cell gel electrophoresis or comet assay is known for its ability to detect DNA damage at the single cell level and has been used for years to assess DNA damage. It can detect low levels of DNA strand breaks in a short time, just using a few sample cells. DNA double strand breaks (DSBs) are measured at the neutral comet assay condition; under the alkaline comet assay condition both DNA single strand breaks (SSBs) and part DSBs are detected. The two dimensional comet assay is a modification of the two original comet assay, can simultaneously detect DNA SSBs and DSBs in the same human spermatozoa. If the two dimensional comet assay can be adapted to simultaneously assess different DNA break types in other cell types, DNA damage induced by heavy ion radiation can be assessed more accurately. The purpose of this paper is to validate the two-dimensional comet assay as a reliable method to simultaneously detect both DNA single and double strand breaks in the same bone marrow mononuclear cells (BMMNCs).

BMMNCs were incubated with different concentrations of H_2O_2 for 5 min to induce DNA SSBs or incubated with reaction buffer containing DNase I enzyme for different time to induce DNA DSBs, DNase I and H_2O_2 were used successively to induce SSBs and DSBs in the same cells. Then two dimensional comet assay was performed, the images were analyzed with CASPLab 1.2.2 Software.

The electrophoresis results were shown in Fig. 1. The TM and OTM of SSBs and DSBs were analyzed separately.

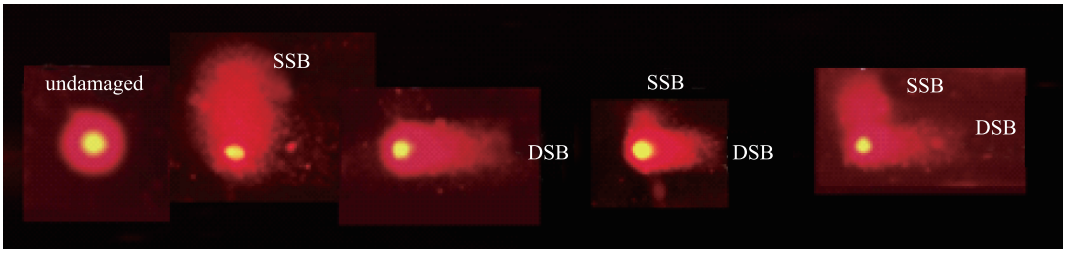

Fig. 1 Two dimensional comet assay detects different comet types.

In the BMMNCs treated with H_2O_2, the yield of SSBs increased significantly with the increase of H_2O_2 concentration, and there was no obvious DSBs (Fig. 2(a)). The results of DNase I treatment were shown in Fig. 2(b), the TM and OTM of DSBs increased significantly with the time of enzyme treatment, while SSBs remained at the control level. In the cells treated with H_2O_2 and DNase I successively, the TM and OTM of X-axis had the same trends as the results of independently DNase I treatment, and the results of Y-axis were similar to the H_2O_2induction, and no significant statistically differences were found (Fig. 2).

This study indicated that two dimensional comet assay is adapted to simultaneously detect BMMNCs SSBs and DSBs. It can be expected to more accurately assess heavy ions radiation induced BMMNCs DNA damage with this technique.

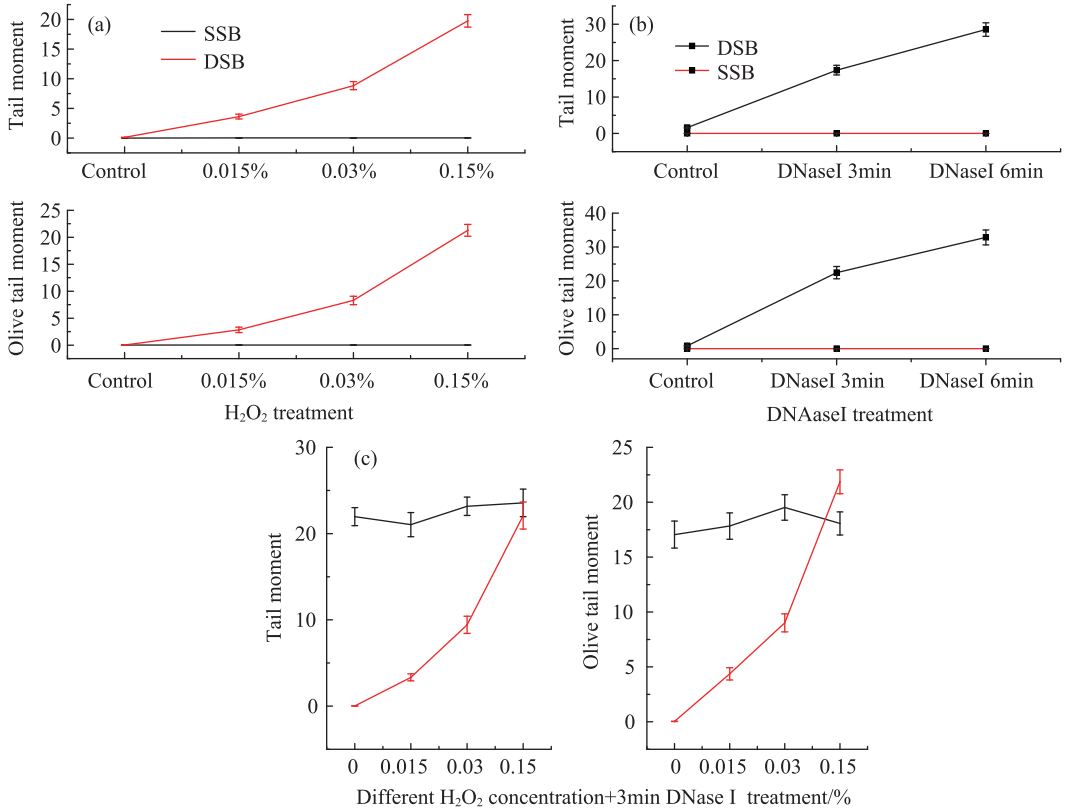

Fig. 2 DNA damage detection with two dimensional comet assay. (a) DNA damage induced by H_2O_2, (b) DNA damage induced by DNase I, (c) DNA damage induced by DNase I and H_2O_2.

4 - 38 DNA Damage in Bone Marrow Mononuclear Cells of Mice Following Total Body Irradiation with Carbon Ions

Wang Zhuanzi, Liu Fang, Li Wenjian, Wei Wei and Dang Bingrong

Among the many special factors in the space environment, space radiation especially high energy heavy ion radiation is widely regarded as one of the crucial factors that hazard the health and safety of astronaut. It is well known that biological effects of radiation are dependent on the linear-energy-transfer (LET) of the emergent beam. Heavy ions have high LET and could induce complex and clustered DNA damage, which is more difficult to be repaired than individual lesions and is even irreparable. The biological effect of heavy ions is higher than conventional radiations such as low-LET γ- and X-rays. So it is important to focus on the radiation biological effects of heavy ions even if their proportion in space radiation is low.

Hematopoietic system is highly sensitive to radiation, and its damage can reduce astronaut's ability to work and radiation tolerance. Bone marrow involved in hematopoiesis primarily. The main purpose of this paper is to evaluate the high-LET irradiation, like carbon ions, caused DNA damage of bone marrow mononuclear cells (BMMNCs) in mice.

120 Female BALB/c mice were randomly divided into 5 groups (24 mice per group) and total body irradiated (TBI) with different doses of carbon ions (LET=31.6 keV/μm) at a dose rate of 1 Gy/min. Sham-irradiation mice were used as controls (0 Gy). On the first, third and eighth day after TBI, BMMNCs were harvested from mice femoral tissues (8 mice/group/time points) and the DNA damage of BMMNCs in mice was evaluated using the two dimensional comet assay.

DNA single strand breaks (SSBs) and double strand breaks (DSBs) results of two dimensional comet assay were analyzed separately and showed in Fig. 1. The results showed that DNA SSBs and DSBs induced by carbon ions irradiation were time and dose-dependent. The TM and OTM of DSBs (Fig. 1(c) and (d)) were much higher than those of SSBs (Fig. 1(a) and (b)). The percentages of DNA damaged cells were also time- and dose-dependent (Fig. 2), and the percentage of DNA DSBs cells at the 1^{st} day after carbon ion exposure (Fig. 2(b)) was much higher than that of SSBs damaged cells (Fig. 2(a)).