PHOTODYNAMIC EFFECT OF THE HE–NE LASER WITH HpD ON THE ULTRASTRUCTURE OF RHABDOMYOSARCOMA CELL

Chen Ting-I and Guo Zhong-he

Institute of Radiation Medicine, Academy of Military Medical Science, Beijing, China

*The Chinese PLA General Hospital, Beijing, China

INTRODUCTION

Photodynamic effect of Hematoporphyrin Derivative (HpD) on malignant cells has been widely studied; cells which have taken up HpD are killed "in vitro" or "in vivo", when the compound is activated by light. The light excitation of HpD produces singlet oxygen which, by its strong oxidizing power, breaks down the structure and alters the biological function of major biomolecules, including proteins, nucleic acids, cholesterol, fibrinogen, and various kind of enzymes etc. 1,2. As all of these are the essential materials in the structure and function of the cell, photodynamic action of this kind may induce extensive damage or alteration in the ultrastructure of cells. It has been reported that photodynamic action dissolves the cell membrane, oxidizes phospholipid and cholesterol, causes damage of enzymes in membrane transferring system, inhibits Ca++ transportation of Chinese hamster ovarian cells 3, depresses oxidative phosphorylation in mitochondria as well as oxygen consumption 4, breaks down on DNA chain 5, and kills the tumor cells 6 especially those at G1–S stage of the cell cycle 7. The singlet oxygen action causes wide-spread damage of the cell which can be revealed easily by observing the ultrastructure under electron microscopy. Coppola 8 reported that mitocontria may be more sensitive to light and HpD, and therefore become the first organelles to be affected. We have observed the photodynamic action on tumor cells. The results are presented in the following.
Fig. 1. Cells in same field show various degree severity of lesion as shown by the different changes of the liable organelles in different cells. x 4,000.

MATERIALS AND METHODS

The PLA-802# Rhabdomyosarcoma cells of human larynx (Alveolar type) were cultured in RPMI1640 medium for 24 hours. HpD was added to the medium at a concentration of $10^{-5}$ M and kept in the dark for 20 minutes. The cells were irradiated inside the flask by an He-Ne laser with the power density 10 mW/cm$^2$ for 20 minutes.

The cells were harvested from the flask, centrifuged, then fixed, embedded in EPON-812, and prepared for ultrathin section with LKB microtome. After staining with lead and uranium, the preparations were observed under JEM-6C electron microscope*.

RESULTS

Photodynamic action of the He-Ne laser and HpD induced various and extensive changes in ultrastructure of PLA-802 cell. In the electron-microscope observation field, one might disclose cells damaged at various degrees as shown by the structure changes in differ-

---

# Cells are provided by Department of Pathology of The Chinese PLA General Hospital.

* The EM sections were prepared by Xiao Lou, and Zhan Si-min.