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LASER - CELLULAR NUCLEI INTERACTIONS

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Introduction. The effects of an UV laser and of a He-Ne laser radiations on cellular nuclei were analyzed.

Materials and Methods. Suspensions of $5x10^6$ cells/ml or extracted chromatin (the complex of deoxyribonucleic acid -DNA-with proteins from cellular nuclei) of Walker carcinosarcoma, maintained on Wistar rats were utilized. A laser radiation with λ =248 nm from an Iofan 1701 model laser and a He-Ne laser radiation with λ =632.8 nm from a Spectra Physics M 125 laser, in doses of 0.1-1 MJ/m² were used. For cells suspensions as starting material, the determination of chromatin DNA strand breaks was effected by a fluorimetric method, that makes use of a partial treatment with alkali and the percentage of the DNA double strand remained is established. This parameter gives an indication on laser action on chromatin DNA. In the case of extracted chromatin, two parameters were analyzed: the fluorescence energy transfer from dansyl chloride, covalently coupled at chromatin proteins and acridine orange, intercalated between chromatin DNA base pairs and the fluorescence lifetimes of the excited state of the chromatin - ethidium bromide complexes. An Aminco-Bowman spectrofluorimeter and an Edinburg FL 900 CD time resolved fluorimeter were used.

Results. The percentage of remained DNA double strand decrease with the laser dose, more drastically in the UV, than in the He-Ne laser radiation. This fact proves that single and double DNA strand breaks are produced. In the case of double fluorescent labeling of chromatin, the efficiency of the energy transfer decreases with the laser dose, also more evident in the case of UV laser radiation. This behavior denotes a more destabilized tertiary conformation of chromatin, with the destruction of proteins structure, under the influence of laser radiation. Also, depending of the laser dose, the fluorescence decay curves of chromatin-ethidium bromide complexes have different relative contribution for the half-lives for unbound and for intercalated ethidium bromide in chromatin DNA. A stronger increase of the contribution of unbound ligand takes place with the laser dose increase, in the case of UV laser radiation, that in the case of visible laser radiation. This parameter gives a precise ratio of damaged DNA in cellular nuclei.

Conclusions. The used methods of emission spectroscopy are useful for the knowledge of laser action mechanism on cellular nuclei. The modifications in chromatin structure are changes of the total chromatin conformation and the diminution of the double helix DNA proportion. The injuries induced in chromatin are more significant in UV than in visible laser radiation. The results may constitute an indication for more efficient UV laser utilization in certain medical applications.

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MORPHOLOGICAL ANALYSIS OF THE POLARIZED NEAR INFRARED LASER RADIATION EFFECTS ON SKIN WOUND HEALING

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Introduction: The influence of linearly polarized low-intensity laser light irradiation on the healing process of skin wounds has a dependence on the relative orientation between the electric field polarization and the sample preferential direction, as previous studies using a He-Ne laser (632.8 nm) have indicated. Since the GaAlAs laser has been normally used as a low-intensity laser source in the clinical practice, not only as a coadjutant but sometimes as a specific treatment, a study of its polarization orientation on wound healing in rats was performed.

Material and Methods: Three inflammatory lesions were produced on the shaved backs of Lewis male rats, weighing about 300 g, using a cylindrical brass rod cooled to 77K. The used GaAlAs diode-laser (SDL 2382) was operated at the wavelength of 797 nm, and its 99.9% linearly polarized output laser beam was adjusted to an intensity of 3 mW/cm². The first lesion was illuminated with the laser polarization aligned with the rat spinal direction, the second with the perpendicular relative orientation, and the third was not irradiated. Two rats were killed after each irradiation on the 1st, 5th, 8th and 12th day, and the last rats were killed on the 15th day (no more irradiation) to get the tissue sections for morphological study.

Results: No difference was observed between the lesion #1 and lesion #2. Both were recovered by a new epidermis. In both lesions the epithelial layer was ticker when compared with the epithelial layer of the normal skins. The subjacent dermis was formed by a thick layer of a loose connective tissue whose fibroblasts appear to be metabolic actives. In the control lesion the epithelial layer that recovered the scar was very thin compared with the experimental groups. The dermis was formed by a dense connective tissue with few fibroblasts surrounded by a great amount of thick collagen fibers.

Conclusion: Macroscopically, the laser irradiated lesions were healed completely when compared to control lesions. There was no difference between the irradiated lesions with respect to polarization orientation at this wavelength.

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