EFFECT OF LIPOSOME-ENCAPSULATED RADIOSENSITIZER (AK-2123) ON DEN INDUCED CANCER IN MICE

ABSTRACT

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ABSTRACT

Cancer is one of the major causes of death worldwide. It is caused by the progressive growth of the progeny of a single transformed cell, which is no longer responsive to normal growth controlling mechanisms. Therefore, curing cancer requires special strategy to selectively destroy all the malignant cells without damaging the normal cells. Conventional radiotherapeutic regimen often fails to cure cancer patients because they do not kill all malignant cells. Since, the interaction of radiation with matter is random, the radiation alone will damage both normal as well as transformed cells when the subject is irradiated. The damaged transformed cell may not survive and, thereby, cancer may be eliminated. However, there is a possibility that partially damaged normal cells may undergo transformation in due course of time and reestablish the disease.

The combination of radiomodulatory drugs with radiation has been considered to beneficial for radiotherapy of cancer. It has, indeed, improved the rate of cure of cancer and the outcome of treatment is better than radiation alone. The two different types of radiomodulatory drugs currently used in chemo-radiotherapy are: -

1. Radioprotective drugs that should essentially protect normal cells/tissues from the undesirable damage of radiation, thereby, paving way for application of higher doses of radiation for efficient killing of cancerous cells/tissues.
2. Radiosensitizing drugs that should, in principle, sensitize cancerous cells/ tissues so that they are killed efficiently even by relatively low dose of radiation, thereby, reducing the undesirable damages to normal cells/tissues.

However, the toxicity of drugs to other tissues and various other side effects have limited the application of chemo-radiotherapy in humans. In order to overcome these limitations of chemo-radiotherapy and to improve upon its therapeutic index, concept of drug delivery system has been tried in the last two decades. The concept is based on the fact that certain biologically acceptable carrier may be employed to deliver the drugs to biological target cells/tissues. Of several possible carriers, liposomes has been found to be promising and convenient because it's organized structure could accommodate drugs in the aqueous or lipid phase, depending on their solubility. Liposome is a lipid bilayer that folds back on itself creating a hollow sphere within which, it can enclose or entrap a variety of

substances. Structural versatility and biodegradable nature have made liposomes a more potent biological carrier system in drug delivery and drug targeting.

The work embodied in this thesis is an attempt to evaluate the whole body γ-radiation effect in the presence of AK-2123, a hypoxic cell radiosensitizer on tumor regression in mice. AK-2123 has widely been used in combination with a number of cancer therapies such as thermotherapy, chemo-therapy and radiotherapy.

Mice were chronically exposed to diethylnitrosamine, a potent hepatocarcinogen, for tumor induction. Cancer induction was studied by monitoring the marker enzymes activities, i.e. GGT and AChE, and the rates of nucleic acid synthesis. Histology and electrophoretic studies of surface membrane glycoproteins in liver were also carried out. Regression studies were carried out by administration of AK-2123 either in it's free or liposomal forms, in combination with whole body γ-irradiation 30 min. after the drug administration. The entrapment efficiency of liposomes for AK-2123 was determined using LASER Raman Spectroscopy. For the regression studies the same parameters employed in induction studies were used in addition to some haemopoietic parameters. The liposomal AK-2123 is envisaged to be tested *in vivo* for its radiosensitizing efficacy in normal and transformed system vis-à-vis the free form of AK-2123.

From the experiments conducted it was found that radiation treatment alone resulted in mass destruction of the cells. AK-2123 alone did not exihibit toxic effects on mice whether it was presented in it's free form or encapsulated in liposomes. However, a combination of these three factors i.e. radiation, AK-2123 and liposomes, produced satisfactory results. Liposome encapsulated AK-2123 was found to be more efficient radiomodulator compared to it's free form. The main points emerging from the work embodied in the thesis are: -

- Chronic exposure to NDEA combined with hepatectomy induced cellular transformations in the liver of Swiss albino mice, as substantiated by the pronounced alterations in the activities of the marker enzymes and the synthetic indices of the nucleic acids.

- NDEA treatment resulted in a distinct change in the nature of the hepatocytes such as the variations in the cell shape and size, appearance of more densely stained nuclei and multinucleated cells as elucidated in the histological studies.
- Upon NDEA exposure, the liver glycoproteins exhibited differential expressions. Some proteins were over expressed while others were under expressed as compared to their normal counterparts as revealed by the electrophoretic study.
- Radiation alone inflicted damage to both the transformed and normal cells, as reflected by the marked decline in the activities of the marker enzymes and synthetic indices of the nucleic acids.
- Liposomes obtained by the reverse phase evaporation method were relatively large in size and most of them were unilamellar.
- About 40% entrapment efficiency of AK-2123 into liposomes was achieved as determined by LASER Raman spectroscopy.
- AK-2123 alone, whether present in free or liposome encapsulated forms, had no toxic effect on the liver at a concentration of 200 mg/kg body weight.
- Liposomes retained the properties of the entrapped AK-2123. It's selective migration to the liver ensured that a major proportion of the drug reached the liver intact, as is evident from the levels of the activities of the marker enzymes and nucleic acid synthetic indices, which were in the proximity of the normal levels. This is further supported by the observation that AK-2123 when encapsulated in liposomes showed lesser suppressive effects on the haemopoietic parameters studied, as compared to it's free form.
- AK-2123 in the oxic conditions of irradiation exhibited lesser radiosensitizing effects as compared to the hypoxic mode of irradiation thereby, affording radioprotective effects. It is seen from the fact that none of the transformed groups of animals administered with either free or liposome encapsulated AK-2123 and followed by exposure to radiation in oxygenated conditions, regained their actual normal levels of enzyme activities and nucleic acids' synthetic indices, nor fell below the normal levels.
- Overall, AK-2123 encapsulated in liposomes afforded better radiomodulation than free AK-2123.

Thus, it is seen that treatment of cancer with a combination of radiation, a radiomodifier and a drug delivery system, opens a wide scope for exploitation for the

improvement of existing cancer therapies and the chances of curing and combating the menace of the dreadful cancer.