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Cancer and Radiation Therapy: Present Status

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In a very simple language cancer can be described as a state when the normal growth and differentiation go away resulting into malignant tumours. Cancerous cells continue to divide; live longer than normal cells; appear not to undergo the programmed cell death characteristic of normal differentiation to non-dividing, terminating cells; in addition, they fail to undergo normal differentiated cells. Cancers may be caused by many factors such as chemicals, radiation and hormones. Chemicals are generally associated with skin, lung and bladder tumours, radiation with haematological malignancies, leukaemia, lymphoma as well as thyroid carcinoma, while hormones are associated with cancers in breast and uterus. Growth of malignant cancer depends not just on factors causing cancer but also on body's response to it as well.

It was in the fitness of things that 1989 Nobel Prize in Medicine was awarded to J. Michael Bishop and Harold Varmus of the University of California for their work on oncogene hypothesis in 1976. Their work suggested that normal genes (referred to as proto-oncogenes) controlling growth, development and differentiation somehow become misdirected and mutate into oncogenes in neoplastic cancer cells. While remembering the Nobel Prize winners, let us not forget Alexander Haddow who had indicated as far back as in 1937 that cancer growth arises as a somatic mutation. Oncogenes have been referred to acting like red and green traffic lights regulating cell growth. When they are disrupted, it is like green light getting stuck allowing a tumour to grow unchecked. The discovery has widened our knowledge and insight into the complicated signal systems which govern the normal growth of cells. The importance of Bishop and Varmus' work lies in the fact that we have now a completely different view on how cancer can originate. It is still not very well known how a normal gene gets triggered into becoming an oncogene. Once we know this process, we may also be able to stop this triggering mechanism hopefully some day. We also know by experience that when treating a tumour we often sacrifice and alter many normal cells which cannot be helped. The destruction of the normal cells is the reason for the significant side effects associated with current cancer therapies.

Cancer and Age

Cancer is a disease of old age except some such as leukaemia and bone cancer. Out of 50 million deaths per year in the world, more than 5 million are attributed to cancer. It is estimated that in another 10 to 15 years, cancer deaths in the United States of America alone may account for eight million. The main reasons for this are changes in the health spectrum and domographic structure, changes in life style and changes in environment. More than half the world's population now lives in countries where cancer is among the top ranking

causes of death. In Japan, it is number one and in the United States of America, it is number two. This is because longevity is now highest in Japan. It may also be worthwhile to note the age distribution of population between the age of 10 to 40 years. It is 40% in Africa. 31% in Asia and 12% in Europe and North-America. However, the average age of cancer patients in Europe and North-America is 55.7 years, while in Asia and Africa it is 45.9 and 35.9 years respectively. This must be because the standard of health is poorer in Asia and Africa.

Incidence of Cancer and Deaths

In the United States of America death due to cancer was 107/100,000 population in 1930 and it increased to 180/100,000 in 1980. The increase must also be related to the age since in the fifty years period longevity has also increased. However, when the figure is corrected for longevity factor, even then it comes to 160/100,000 population which also is a significant increase. Unfortunately, a correct estimate of this is not available in India. It is estimated that the incidence of cancer was 80/100,000 in 1947 and has increased to 120/100,000 population at present and is going to increase two to three folds by the year 2000 AD. Longevity has increased in India also, from 37 years in 1947 to 58 years now.

Radiation Therapy

For a long time radiobiologists received very little attention from radiotherapists but the situation changed during the last decade and a half. As a result, the advances in the field of radiation biology are being received with greater interest and understanding by radiotherapy community. This development has brought in dramatic changes and thus the advances have been faster.

Any tumour can be roughly divided to possess three types of cells: (i) necrotic cells which lie in the centre and are far removed from blood capillaries, (ii) hypoxic cells in the middle, and (iii) oxygenated cells which are at the periphery and are nearest to the blood capillaries. It is also very well established that oxygenated cells are more sensitive to radiation than hypoxic cells. Approximately, thirty to fifty per cent of all cancrs need radiation therapy. Single large dose of radiation is harmful, hence fractionated doses are given which is less harmful. Further, in the first dose of radiation the sensitive oxygenated cells are killed when the middle layer hypoxic cells may get converted into oxygenated cells which may be killed by the second dose.

Radioprotectors

It has already been stated in the beginning that the significant side effects associated with cancer therapy occur because of the destruction of normal cells which is difficult to avoid. A great deal of effort has been put in by radiobiologists to search a modifier which would be of help to radiotherapists. It is necessary that such a modifier should either selectively potentiate radiation effects on cancer cells without affecting the normal cells or else they should protect the normal cells without protecting the cancer cells.

Many chemicals such as epinephrine, histamine and serotonin are known to act as radioprotectors but they are not true radioprotectors as such since they basically cause vasoconstriction and thus reduce basal metabolic rate. True radioprotectors are sulphydryl compounds such as cysteine, cysteamine, cystamine, WR-2721, which basically act as free radical scavengers. The difficulty with these drugs has been that to bring abut a Dose Reduction Factor (DRF) of 2.0 to 3.0, one has to use 50 to 75 per cent of the toxic dose (Sugahara & Srivastava 1976). Nonetheless, the drugs have been useful. American astronauts perhaps carry cysteamine to be taken in case of emergency.

2-Mercaptopropionylglycine (MPG)

Our group has done considerable work on MPG since it is non-toxic and gives a DRF of 1.4 at 1% of the toxic dose. This drug was used in Japan for a long time as a detoxicating agent under the trade name Thiola. Sulphydryl compounds have been classified into three classes according to their toxicity and radioprotective action. Most effective protection was obtained by cysteamine and cysteine followed by AERT and MPG-amide in

that order. Toxicity of these compounds was generally observed in the range of 0.1-2.0 mM (Hikita et al./1975). On the other hand, MPG was found to be non-toxic at 0.02mM and 15mM when it gave significant protection. Extensive work on radioprotection by MPG against gamma radiation-induced damage to liver (Saini et al./1977), bone marrow lymphocytes (Saini et al./1978), testis (Uma Devi & Saharan/1978, Saharan & Uma Devi 1977), small intestine and jejunum (Uma Devi et al. 1978, Uma Devi 1977, Saharan et al./1978), thymus (Saini & Uma Devi 1979a, 1979b, Uma Devi & Saini 1977), and growth-inhibiting effects in utero irradiation in mice (Dev et al. 1982) has been done by our associates. In all these cases an enhanced recovery and accelerated restoration of normal cellular strucures and cell counts were observed in case of MPG-pretreated animals as compared to controls.

In spite of the fact, that sulphydryl compounds are very good radioprotectors, their application in radiotherapy is very much limited because of non-availability of such compounds with twin properties of low toxicity and high DRF. However, two compounds MPG and WR-2721, have been put to clinical tests. While MPG has already undergone clinical trials in Japan (Sugahara & Srivastava/1976), the clinical trials of WR-2721 are currently in progress (Symposium on Perspective in Rdioprotection, March 13-14, 1987, National Bureau of Standards, Bethesda, Maryland, USA). Ayene and Srivastava (1985) have demonstrated radio-sensitization by MPG/Fe complex in microsomes. Enhancement of lipid peroxidation was observed at 0.1mg/ml of MPG instead of protection. However, MPG in the presence of EDTA gave significntly more protection at all doses of radiation. The complex formation was further confirmed by the exogenous supply of ferrous sulphate during irradiation of microsomes. The addition of Fe ions enhanced the formation of lipid peroxides. A further increase in lipid peroxides was observed in the presence of MPG with an abrupt increase to a dose of 133.2 Gy and a slight increase at higher doses of radiation. Similarly greater protective effect of MPG was observed in erythrocytes in absence of MPG/Fe complex formation (Ayene, et al. 1988). Protection to erythrocytes at high concentration was also rendered by MPG at both concentrations used. It also exhibited a similar effect in erythrocytes of low concentration but with a variation in the degree of protection. Further, the data on the effect of $FeSO_4$ provided a clear picture about the role of Fe^{2+} in the enhancement of lipid peroxidation in presence and absence of MPG. It was also demonstrated that both the spontaneous and radiation-induced lipid peroxidation of erythrocytes was increased by $FeSO_4$. The results also showed the enhancement of radiation-induced lipid peroxidation by MPG in the presence of $FeSO_4$. Such complex formation may be responsible for the lesser protective effect of MPG (DRF = 1.4) in mice. Further work needs to be done in *in vivo* systems.

Some discrepancies on WR-2721 also indicate that caution has to be adopted in the understanding of conditions in which thiol compounds might protect or fail to protect tumours (Lunec et al. 1981). Reports on failure of WR12721 to exhibit differential radioprotection of normal and cancerous tissues have also appeared (Rojas & Stewart 1980, Rojas et al. 1982, Cement & Johnson 1982, Molas et al. 1982). Phillips et al. (1973) have also failed to substantiate the very low protective effect on tumours by WR-2721 reported by Yuhas (1972, 1973) and had shown a wider variation in protection afforded to normal tissues than was previously reported. The importance of dephosphorylation of WR-2721 in the protective efficiency of the drug has been demonstrated by us in erythrocytes (Ayene & Srivastava 1989, Srivastava 1990). Protective and non-protective effect of WR-2721 in microsomes and erythrocytes have been indicated. The effect in erythrocytes was suggested because of the inadequate amount of dephsphorylating enzymes. This was confirmed by the addition of microsomes to erythrocytes that reduced the radiation damage of the erythrocytes. Such dephosphorylation has also been demonstrated in the presence of dephosphorylating enzyme, alkaline phosphatase (Misra & Srivastava/1981,

Collobro Jones et al. 1985, Misra et al. 1990a, 1990b). Research has also been carried out to study and regulate the metabolism, toxicity and radioprotection of WR-2721 by an inhibitor of alkaline phosphatase, levamisole (Brown et al. 1986). Research in the direction of inhibiting the alkaline phosphatase activity in tumour cells may further enhance the chances of using WR-2721 in the differential protection of normal against the cancer cells.

Radiosensitizers

Just like radioprotectors, sensitizers too are of two types, apparent and true. Radiosensitizers are drugs which have the capacity to increase the lethal effects of radiation. If they are non-toxic, then they will be ideal since they could kill the cancerous cells at lower doses of radiation without too much harmful effects on normal cells.

There are many apparent sensitizers such as Atinomycin D which depresses DNA dependent RNA synthesis; Puromycin which selectively depresses protein synthesis; Methotrexate which interferes with the synthesis of DNA by preventing the formation of coenzyme necesary for *in vivo* synthesis of thymidine; or 5-Fluorouracil which kills cells during the DNA synthetic or S phase of their cell cycle. They are not true sensitizers since they are compounds which act only additively with radiation.

True radiosensitizers are drugs such as 5-Chlorodeoxyuridine, 5-Bromodeoxyuridine and 5-Iododeoxyuridine. The basis of their action depends on the fact that the combining size (van der Waals radius) of the atom of chlorine, bromine or iodine is very similar to the methyl group, CH_3. The halogenated pyrimidines as they are called therefore replace the normal DNA precursor thymidine and hence weaken the backbone which breaks during division. Their use, however, limited because of many limitations and also because they get metabolized very soon and hence become ineffective.

Sensitizers of Hypoxic Cells

Many compounds have been developed such as Metronidazole, Misonidazole, Nitroimidazole, etc., which are effective against specific hypoxic cells of tumours since they metabolise slowly and reach the hypoxic cells easily. These drugs increase the oxygen enhancement ratio of the cells upto 3.0. These compounds are being used clinically and have been successful to some extent.

Hyperthermia

The use of hyperthermia alone or in combination with radiation had been suggested over the years but it has found a permanent place in the management of cancer only for about 10 years. In India, unfortunately, it has still to find a place. Miller et al. (1977) and Dewey et al. (1977) have reviewed the early clinical use of hyperthermia. More recently, Arcangeli et al. (1988) have published a review based on the works of seventeen groups of scientists and clinicians. They have described the effect of adjuvant hyperthermia on the radiation response. It has been shown that the frequency of response varies and the tumour regression can be enhanced from 25% to 65% depending upon the tumour and its site. The thermal enhancement ratio of radiation plus hyperthermia and radiation alone comes to 1.4-2.0. The degree of sensitization by hyperthermia and radiation as compared to radiation alone varies from 1.5 to 4.3 depending upon the temperature used from 41 to 43°C.

A few months back (about February-March 1990) Dr Kenneth Alonso and his group of the University of Atlanta Medical Hospital have treated AIDS patients and the blood of patients has been shown to be HIV negative for more than four months. The blood of the patient was passed through a tube the temperature of which was raised to about 43°C. Further, terminal patients are being treated by this method. The doctors do not claim to have checked AIDS but are investigating the reasons for the blood remaining HIV negative for such a long time (Personal communication).

Immunotherapy

The growth of malignant tumour does not depend on cancer itself but it also depends on the body's response to it. If somehow or the other the body's

immune response could be bolstered, then he or she may be able to fight off cancer better. Unfortunately, it has been easier said than done. The situation, hopefully, seems to be changing. In early seventies it was shown that BCG vaccine injections could induce guinea pigs' tumour to regress. Overzealous reports hailed BCG as a "Cancer Cure", which it was not. But the event did give fruitful leads.

Old therapies focussed on non-specific stimulators of immune system including BCG but more recently several compounds such as lymphokines, cytokines, interferons and inter-leukins have been developed which are anti-viral proteins— glyco-proteins— and are especially useful for cancers of viral origin. The new therapies use tumour cells, often prepared from patient's own concern, to elicit specific immune responss to the particular tumours from which the patients suffer.

Development of several vaccines is in the pipeline. Although vaccines do seem to prevent metastases, enough time has not elapsed since the study began to establish whether it improves 5-year survival rates which are considered to be "gold standards" for successful cancer therapy. Although many are now optimistic about vaccine results, but people are cautious not to claim them aer at this stage. However, people
had been trying to develop cancer immunotherapy for 20-30 years which has only recently started giving encouraging results.

Tumour Angiogenesis Factor

In early seventies, an observation was made that when tumour grows in size, blood capillaries start proliferating, growing nearer and nearer to the tumour. This was a reaction to the growth of tumour since the blood capillaries can supply oxygen and nutrition upto only approximately 150 μm. A pharmaceutical firm donated 23 million US Dollas to the Harvard Medical School to carry on research to find out if any tumour angiogenesis factor was involved in this. After 12-13 years of work, when the scientists were on the verge of giving up the pursuit, they found some clue when they got a compound which they named as Angionin in 1985. They found 1mg of the compound in 2000 litres of fluid in which cancer cells were grown. The molecular weight of angionin is 14,400 and has 123 amino acids and is 35% identical to ribonuclease. Biotechnology can now play a role to develop "anti-angionin factor".

Tumour Necrotic Factor

Lymphocytes and macrophages are known to produce a substance called Cachectin which is a tumour necrotic factor that helps in the process of necrosis of cancerous cells. It is a polypeptide and may be in dimeric, trimeric or pentameric forms. The molecular weight of each sub-unit is 17,000. It was sequenced in 1986 and has now been produced biotechnologically. Numerous clinical tests are in progress.

It had also been observed long time back that a cancer patient reacts to some drug and recovers but later on when a relapse takes place, the same patient does not respond to the same drug to which he/she had responded earlier. Scientists had been busy to find out the cause of such a phenomenon. It has again been shown very recently in 1986 that as a reaction to the drug, the cells produce certain glycoproteins whose molecular weight has been determined to be 170,000. This glycoprotein attaches itself with the membrane of the cells and thus alters the cellular membrane. This changed cellular membrane now acts as a pumping mechanism and the next time the patient is given the same drug, it is pumped out of the cells before it could act on the cells.

Photochemotherapy

Some pharmaceutical firms are now turning to photo-activated chemicals to formulate highly specific weapons against a variety of serious diseases including cancer. The first successes in this area were achieved by the scientists of the American firm Johnson and Johnson, who developed 'Photofrin' a derivative of natural substances called porphyrins which are normally harmless components of haemoglobin in red

blood cells. When exposed to sunlight, porphyrins emit an intense red fluorescence that produces ozone which destroys cells. Since 1985, another American pharmaceutical company, Cyanamid, began clinical testing of the drugn on cancer patients at more than 50 centres.

What makes these drugs so effective are lasers and fibre-optic probes which deliver specific wavelengths of light needed to set off the drugs directly on cancerous tissue. For this, the drug is first injected into the patient where it collects in tumour cells, since they retain it more easily than healthy cells. After two days, the drugs are excreted from the healthy cells and are retained only by the cancerous cells. A beam of low-powered laser light is channelled through a fibre-optic probe which activates the drug and selectively kills the cancerous cells, the normal cells remaining unaffected.

The origin of this development also lies in the past. About 100 years back, a Danish physician, Niels Finsen, used light from an arc lamp to treat a tubercular condition of the skin called lupus vulgaris. This disease was very common in northern latitudes, especially during winter months. Because of Finsen's discovery, lupus vulgaris practically disappeared from Scandinavian countries. Finsen's treatment was so original and successful that he was awarded one of the first Nobel prizes in 1903.

Conclusion

In conclusion this can only be said that the fight for the cure of cancer is still on and will perhaps continue for some time to come. In general, one third of cancers are preventable, one third are curable and the remaining one third at present can only be treated with pain relief measures.

In India, approximately 50% cancers belong to oral cancer, whose detection in early stages is very much easier than many other cancers and hence are totally or nearly curable and cervical cancer which takes five years to develop localised cancer and another ten years to reach the advanced stage. The latter can again be detected early and if proper care is taken it can virtually be cured 100%. Thus, in India, preventive oncology should play a big role.

In the end, I will like to thank the Indian National Science Academy for awarding me this prestigious Aryabhata Memorial Medal.

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