ABSTRACT

Cancer is indeed a dreaded disease. Hardly there is any other malady which would evoke greater fear and anxiety than cancer and its ranks worst killer next only to heart attack.

There are certain fairly reliable method for the alleviation and cure of cancer, particularly, when detected early: viz, radiotherapy, surgery and chemotherapy. Though these modalities have been successful, by and large, all suffer from certain limitation and disadvantages.

Surgery is quite effective when the cancer or tumour lesion is localized and easily accessible. For example, the most convenient approach for breast cancer is surgery. But surgery is hardly advisable if the afflicted region is vital or inaccessible (lung, brain etc.) and also when the patient is too young, debilitated or too old. Over and above the surgery does not ensure 'uprooting' the tumour. Radiotherapy is next successful modality, in that it can "uproot" cancerous growth; but here the most important problem is the radiation attacks tumour tissue as much as normal ones. Further, there is a good chance of radiation itself (gamma ray, X-ray) inducing cancer when spilled over normal tissues. Chemotherapy i.e., use of potent antitumour drugs is the only approach if the tumour is not-localized or
inaccessible. But the side effect of these drugs are indeed too heavy a price to pay. Often, a combination of above three modalities with different proportion leads to better results.

Under these circumstances, scientist started looking for alternative, or at least a complementary modality. When laser was invented in 1960 many started to work on the use of laser for this application and this efforts continues now in full swing.

Laser is a coherent light. Coherence means orderliness in the emission of photons (quantized light particle) in space and time. This property makes the laser beam to have monochromaticity (single wavelength) and directionality (collimation) which in turn makes the lasers to produce a high optical energy density a few thousand times better than any non laser light source. Since the laser beam can be focused to a spot of 1 µm diameter, it can be transported through optical fibers with negligible loss and the optical fiber can be inserted into the body through natural orifice. This mean, we can deliver a high dose of optical energy on the surface or the interior of the body. The high energy density from laser can do localized heating or photochemical reaction. Of these, localized heating is not specific or selective. That is, both the normal and cancerous cells would get killed. But the laser induced
photochemical reaction, with the aid of a sensitizer labeled to the cancerous cell, would generate highly toxic singlet oxygen and photo radicals on the heads of the tumour lesion leading to the specific localized tumour cytotoxicity. This is essence of Photo Dynamic Therapy (PDT).

In the last decade, serious investigation and clinical trials have been carried out in the many developed countries of the world and considerable success has been reported. We started in a humble methodological way to explore the possibility of using lasers for detection and treatment of cancers. The thesis gives details of our goals, approach and results. To the best of our knowledge, ours would be the first systematic investigation in this interdisciplinary, applied area of research in India.

We have used two types of lasers viz, pulsed and continuous wave lasers, with two lasers for each type. They were: ultraviolet, pulsed nitrogen laser gas laser of peak power of 100 kW and pulse duration 10 ns; visible, pulsed dye laser of 10 kW power and duration 5 ns. The CW laser used were: He-Ne laser at 632.8 nm of power 15 mW and Argon ion laser at 488 nm and 514.5 nm each of power about 1 W.

The photosensitizers used were: HPD and DHE of porphyrin family; Eosin-Y, Rose Bengal and Fluorescein of Xanthene family and AlPCS of phthalocyanine family. With
those lasers and with these tumour labeling sensitizers, we have studied the combined effects of laser radiation and sensitizer on a set of cell lines in-vitro and also on induced rat fibrosarcoma and spontaneous human oral carcinoma, in-vivo.

The thesis begins with the introduction of cancer cell biology and gives a brief outline about the causes for cancerous growth and conventional methods of treatment indicating the merits and demerits of each. After having pointed out the possible advantages of laser in this area, a fairly detailed account of laser interaction with tissue is given in Chapter 2.

Chapter 3 deals with the principle and operational characteristics of Photodynamic therapy and thus, medical applications of lasers have been narrowed down to the domain of interest for the present work. These three chapters are introductory and prepare the background to view our results in proper perspective.

Chapter 4 deals with the instrumentation, experimental techniques and methodology of the present work. A brief outline of lasers in general, and the operational characteristics of lasers used for this work also are given. This is followed by the irradiation method and details of cell viability analysis.
Chapter 5 describes results of Photo Dynamic Activity (The combined effect of photon, a sensitizer and oxygen on cell in-vitro is commonly known as Photodynamic Activity (PDA) and the same for clinical trial is called Photo Dynamic Therapy (PDT) on human erythrocytes. This is a semi-model system study, since erythrocytes are static and lack nucleus. From the experience gained by the PDA on human erythrocytes, we went on to study the PDA on fibrosarcoma, a fast growing tissue tumour cell line. These cells were obtained from chemical induction of tumours on albino rats. The in-vitro study was carried out as a function of nature of the laser used, fluence, nature of the sensitizers used and its concentration. This study was done under aerobic and subaerobic condition by bubbling nitrogen gas also. The latter was done to assess the importance of oxygen content in the photochemical reaction environment.

In order to assess the photosensitivity of the tumour cells of the different origin, PDA study similar to that for fibrosarcoma was carried out on two types of cultured epithelial cells, He-La and HEp-2. He-La is a tumour cell associated with human cervical cancer and HEp-2 with human larynx cancer.

These in-vitro studies showed that the cell damage is chiefly due to sensitizer intermediated photochemical
reaction and that Argon ion laser with DHE or EY is the best combination for clinical trial.

Based on this guideline, in vivo study on rats carrying induced fibrosarcoma was done. The results confirmed that in few cases the tumour growth could be suppressed and even normalcy could be achieved if right combination of laser and sensitizer could be used particularly at the early stages of tumour. The control set of animals which had the application of laser alone or sensitizers alone or no treatment withered and died in few days whereas the ones which had PDT lived healthy life for four months.

Then a pilot study on spontaneous cancerous and precancerous lesions (oral carcinoma in situ) on human being was also attempted. Here again, we could bring about the remission in one case and alleviation in few cases of carcinoma. Three cases of leukoplakia were also treated by the technique and full or partial cure could be achieved.

PDT is indeed a complex phenomenon involving many unknown parameters and is in the process of getting understood. Being an interdisciplinary subject and hampered by the lack of adequate analytical instruments we could make only a humble beginning. But, we hope, we have begun well, and in the right direction.