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Cancer : Induction, Genetics, Detection & Treatment

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Table of contents

Introduction	6
Significance of the terms mutation and carrier	6
Tumor Classification	6
Behaviouristic classification	7
Histogenetic classification.....	7
Relationship between cancer incidence and age	8
Cancer Induction	11
Physical Factors.....	11
The chemical induction of cancer	18
Cancer: A Familiar Name, But What Does It Mean?	34
How Does Cancer Arise?	35
The Cell Cycle.....	37
Oncogenes and Cancer	40
How Oncogenes Cause Cancer?	42
Tumor Suppressor Genes and cancer	43
p ⁵³ and DNA damage	44
Under Standing Prognosis And Cancer Statistics	46
Key points	46
Cancer Diagnosis.....	47
Tumor Markers.....	51
Radiation Therapy For Cancer: Questions And Answers	61
What is radiation therapy ?.....	61
When is radiation therapy used?	61
What is the difference between external radiation therapy, internal radiation therapy (brachy-therapy), and systemic radiation therapy? When are these types used?	61
What are the sources of energy for external radiation therapy?.....	62
What are stereotactic radiosurgery and stereotactic radiotherapy?	63
Who plans and delivers the radiation treatment to the patient ?	63
What are radiosensitizers and radioprotectors?.....	64
What are radiopharmaceuticals? How are they used?.....	64
Chemotherapy For Cancer.	65
How Chemotherapy Works?	65
Chemotherapy Side Effects	67
Gene Therapy For Cancer: Questions And Answers	69
Key Points	69
What are genes?	69
What is gene therapy?	69
How is gene therapy being studied in the treatment of cancer?.....	69
How are genes transferred into cells so that gene therapy can take place?.....	70
What risks are associated with current gene therapy trials?.....	70
What major problems must scientists overcome before gene therapy becomes a common technique for treating disease?	70
How do gene therapy trials receive approval?	71
List Of Abbreviations.....	72
References	73

Table of figures

Figure 1.1: Age-specific incidence rates for cancers of the skin, colon, and pancreas in males and females.	9
Figure 1.2: Estimated contribution of various radiation sources to the total average effective dose equivalent in the U.S population	12
Figure 1.3: The data points for excess solid cancers in a human population exposed an acute dose of low dose linear energy transfer (LET) radiation are frequently fitted by a linear function of dose.	14
Figure 1.5: Structural formulas of typical carcinogenic polynuclear aromatic hydrocarbons.	25
Figure 2.1: Different types of cancer are derived from different tissues within the body.	34
Figure 2.2: Oncogenes are mutated forms of normal cellular genes involved in growth signaling pathways (proto-oncogenes).	35
Figure 2.3: Tumor suppressor genes are genes often involved in the apoptotic pathway.	36
Figure 2.4: Phases of the cell cycle.	38
Figure 2.5: Different proto-oncogene mutations	42
Figure 3.1: Schematic diagram of radio or enzyme immunoassays several variations of these methods have been used to quantitate makers in body fluids.	53

Table of tables

Table 1.1: Histogenetic Classification of Benign Tumors.....	7
Table 1.2: Histogenetic Classification Of Malignant Tumors.....	8
Table 1.3: Extrinsic Carcinogenic Chemicals Discovered By Their Action In Man.....	19
Table 1.4: Examples Of Naturally Occurring Carcinogens.	20
Table 1.5: Endogenous Chemical Carcinogens.	20
Table 1.6: Classes Of Carcinogenic Chemicals.	24
Table 1.7: some direct acting chemical carcinogens	25
Table 3.1: Classification of Tumor Markers Showing Selected Examples	52

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Introduction

The oldest description of human cancer was found in Egyptian papyri written between 3000-1500 BC it referred to tumors of the breast. The oldest specimen of a human cancer was found in the remains of a female skull dating back to the Bronze Age (1900-1600 BC). The mummified skeletal remains of Peruvian Incas, dating back 2400 years ago, contained lesions suggestive of malignant melanoma .and cancer was found in fossilized bones and manuscripts of ancient Egypt. Cancer is not a disease of our modern industrialized age, as some may have believed at one time.

One of the earliest human cancers found in the remains of mummies was a bone cancer suggestive of Osteosarcoma. Louis Leakey found the oldest possible hominid malignant tumor in 1932 from the remains of either a homo erectus or an Australopithecus. This tumor was suggestive of a Burkitt's lymphoma (although that nomenclature was certainly not in use then). Diseases that we know to be rare cancers today have had a long history. Hippocrates is credited with being the first to recognize the difference between benign and malignant tumors. His writings describe cancers of many body sites. The swollen blood vessels around the malignant tumors so reminded him of crab claws, he called the disease karkinos (the Greek name for crab). In English this term translates to carcinos or carcinoma. Knowledge about cancer genetics is rapidly expanding, with implications for all aspects of cancer management, including prevention, screening, and treatment.

Significance of the terms mutation and carrier

A mutation is a change in the usual deoxy nucleic acid (DNA) sequence of particular gene. Mutations can have harmful, beneficial or neutral effects on health, and may be inherited as autosomal dominant, autosomal recessive, X-linked traits. Mutations that cause serious disability early in life are usually rare in the population, because of their adverse effect on life expectancy and reproduction. However, if the mutation is autosomal recessive, that is if the health effect of the mutation is caused only when two copies of the mutation are inherited, carriers (healthy people carrying one copy of the mutation) may be relatively common. "Common" in this context generally refers to a prevalence of 1% or more. Mutation that cause health effects in middle and old age, including several mutations known to cause a predisposition to cancer may also be relatively common. Many cancer predisposing mutations are autosomal dominant, that is, the cancer susceptibility occurs when only one copy of the mutation is inherited. For autosomal dominant conditions, the term carrier is often used in a different way, to denote people who have inherited the genetic predisposition conferred by the mutation

Tumor classification

Why do we bother to classify tumors? Well, if all tumors behaved in the same way, were equally life threatening and could all be treated by the same cocktail of drugs and/or irradiation, then there would, indeed, be little purpose to the exercise. However, tumors from one tissue behave patently differently from those arising in another. However, the relative 5-year survival rate for thyroid and testicular cancers is in excess of 90%. Such a spectacular difference highlights the need to pin point the origin of the primary tumor, but we must add the caveat that differences are not solely attributable to the relative aggressiveness (local growth and metastatic ability) of the tumor types. Sensitivity to various treatment modalities will vary and more importantly, some deep-seated cancers will have reached an advanced stage of growth when they are detected clinically, whereas superficial cancers, e.g. those of the skin, have a much better chance of being detected during their earliest stages of development. Survival figures for most cancers are greatly affected by the extent of disease at the time of detection, suggesting major improvements in overall cancer survival can be achieved through developing

techniques enabling earlier detection. Identification of common properties between individual tumors is invaluable in being able to predict future development and prognosis-patient survival. Aetiology, behavior, histology and immunophenotype superficially would all seem equally helpful. Aetiological considerations (concerning the causative agent of the neoplasm) how ever are not appropriate means of classification, as identical lesions can be caused by different means. For example, Squamous carcinoma of the skin van arise after exposure to agents as varied as UV light, X-rays, contact with arsenic, contact with certain hydrocarbons and a wide range of other chemicals.

Behaviouristic classification

Broadly speaking the greatest distinction of tumor types is between benign and malignant tumors; this is a fundamental difference in tumor state. Benign tumors are generally slow growing expansive masses, often with a pushing margin, and enclosed within a fibrous capsule. Malignant tumors are usually rapidly growing invading local tissue (infiltrative growth pattern) and spreading to distant sites – metastasizing. truly benign tumors exist (e.g. papillomas-benign surface epithelial tumors), as do incontrovertibly malignant ones (e.g. carcinomas-malignant tumors of epithelial tissues) however there are benign tumors that predispose to malignancy (e.g. adenomas of the large intestine), and there are some in situ carcinomas that progress so slowly that they may never achieve malignancy (e.g. some in situ carcinomas of the uterine cervix); thus a spectrum of types of tumor behavior exists. The ability of a tumor to metastasize from its site of origin (the primary tumor) to form a tumor (secondary tumor) at a distant site is unequivocal evidence of malignancy.

Histogenetic classification

The most useful way of classifying tumors is according to the tissue of origin and cell type involved. A tumor thyroid like vesicles and secreting thyroglobulin presents no problems and can be recognized as derived from thyroid tissue, and can be expected to produce characteristic symptoms and behave in a predictable fashion. Alternatively, some tumor cells grow in such a way that they bear no resemblance to any structure or cell type. Such anaplastic tumors require more detailed investigation to discover their histogenesis. A further problem is that sometimes a tumor resembles tissue which is not normally present at the site of origin. A classical example of this is squamous carcinoma of the lung which probably arises from metaplastic Squamous epithelial in the consucting air ways. The suffix –oma usually indicates a benign tumor, but there are exceptions to this rule. For example myelomas and lymphomas are malignant tumors of plasma cells and lymphoid tissue respectively, while other lesions ending with suffix –oma ,e.g. granulomas, are not tumors at all but are collections of macrophages formed in response to infectious agents examples of benign tumors are given in table 1.1

Table 1.1: Histogenetic Classification of Benign Tumors.

Normal tissue	Benign tumor arising
Glandular epithelium	Adenoma
Surface epithelium	Papilloma
Fibroblasts	Fibroma
Cartilage	Chondroma
Striated muscle	Rhabdomyoma
Smooth muscle	Leiomyoma
Blood vessels	Haemangioma
Fat	Lipoma

The names of malignant tumors, with several notable exceptions, are compiled by the name of the tissue with the suffix of the appropriate malignant tumor, e.g. malignant tumors of the bone

are osteosarcomas and malignant tumors of the colonic epithelium are colonic adenocarcinomas table 1.2

Table 1.2: Histogenetic Classification of Malignant Tumors.

Normal tissue	Malignant tumor arising
Epithelium	Carcinoma
Connective tissue	Sarcoma
Bone marrow	Leukemia
More specifically	
Glandular epithelium	Adenocarcinoma
Squamous epithelium	Squamous carcinoma
Fibroblasts	Fibrosarcoma
Cartilage	Chondrosarcoma
Striated muscle	Rhabdomyosarcoma
Smooth muscle	Leiomyosarcoma
Endothelium	Angiosarcoma
Fat	Liposarcoma
Bone	Osteosarcoma
Liver	Hepatocellular carcinoma
Some malignant tumors with typical names	
Skin- melanocytes	Malignant melanoma
Fibroblast/histiocyte	Malignant fibrous histiocytoma
Myeloid stem cells	Myeloid leukaemia
Plasma cells	Multiple myeloma
Lymphoid tissue	Lymphoma/Hodgkin's disease
Sympathetic neurons (neuroblasts)	Neuroblastoma
Germ cells	Malignant teratoma

Tumors of a mixed cell phenotype have, in the past, been difficult to classify histogenetically. Pleomorphic tumors of salivary glands contain an admixture of ductal epithelial cells and myoepithelial cells, but are now believed to arise from a single cell type, the intercalated duct cell which is probably a multipotential stem cell for the salivary gland, and not from two independent cell types.[1]

Relationship between cancer incidence and age

The incidence of the most types of cancer increases as the forth to sixth power of age figure 1.1. The simplest mathematical analysis of such data suggests that five to seven mutations are necessary for malignant transformation of a normal cell. However, there are difficulties with models requiring many mutations for induction of cancer. To create the necessary age-incidence curves, multihit models assume that all of the cells are present for the life time of the individual, that cells with fewer than the required number of mutations have no growth advantage, that each tissue has a very large number of targets for malignant transformation, and that the presence of a putative environmental carcinogens leads to unusually high mutation frequencies.

Because of the apparent requirement for an un-obtainable frequency of mutation, some investigators have proposed that cancer does not arise by mutation but that other events modify the expression of existing genes. For example, some results indicate that changes in patterns of DNA methylation can turn genes on or off, and that the frequency of these events can be as high as 10^{-3} per cell per generation. One difficulty with such epigenetic models is that the high

frequency of epigenetic changes would make the malignant phenotype highly unstable. reversion of the malignant phenotype appears to be very rare, but there is evidence that some murine teratocarcinoma cells can revert at high frequency to normal functional cells.^[2] in this model, either the mutant genes initiating the tumor are turned off, or the initiating events involve inappropriate gene expression rather than mutation; this abnormal expression could then revert to give normal, tissue-specific expression.

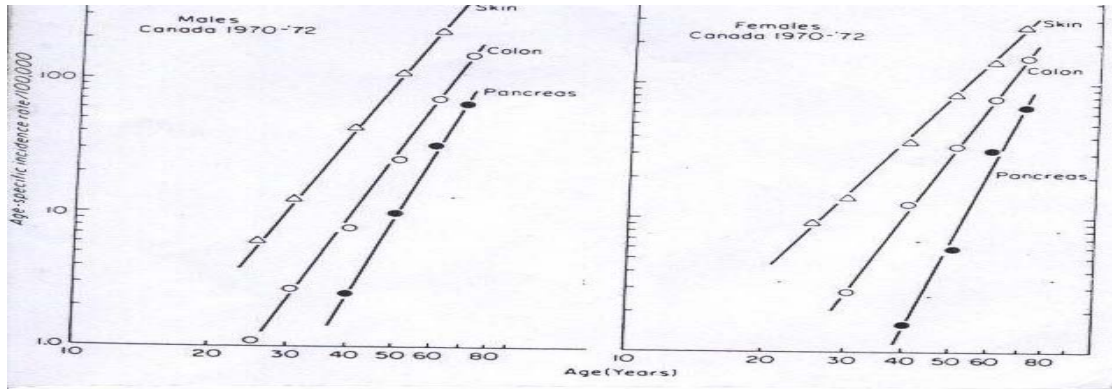


Figure 1.1: Age-specific incidence rates for cancers of the skin, colon, and pancreas in males and females.

cancer induction

Cancer Induction

Physical Factors

This part discusses the induction of cancer by three physical agents: ionizing radiation, ultraviolet radiation, and mineral fibers. This represents a mixture of agents that, on the one hand, occur naturally in the environment, and, on the other hand, are also made or enhanced by humans. Life on earth has always been exposed to ionizing radiations in the form of cosmic rays or radioactivity in the earth. To this must be added artificial radiations from medical radiology and nuclear power, as well as sources enhanced by human activity. This include naturally occurring radon, enhanced because humans live in houses, sealed against the heat or could, in which radon concentrates and decays. Ultraviolet (UV) radiation from sunlight is natural enough, although migration of fair-skinned people to warmer and sunnier climates greatly exaggerates the incidence of UV-induced cancer in humans. Asbestos is also a naturally occurring fiber, but as a carcinogen it too must be classified as enhanced by humans, since only when it is mined and used in commercial products does it pose a hazard. Studies of cancer induction by these physical agents have two distinct aims: to estimate the risk of cancer to the human population following exposure, and to elucidate mechanisms of cancer induction.

Ionizing Radiation

Within 6 years of the discovery of x-rays, a causal relation was suspected between skin cancer and exposure to radiation ^[3]. An association between leukemia and radiation exposure was suspected a few years later ^[4]. There are now many examples of an excess cancer incidence in human populations exposed to radiation. Excellent accounts are available of the experience of the early radiation workers ^[5]. Radiation protection standards in pioneering days were based largely on early effects, such as skin reaction, but the risk of cancer induction is now the dominant factor that determines occupational exposure limits.

Characteristics of ionizing radiation

Radiation is said to be ionizing if it has sufficient energy to eject one or more orbital electrons from an atom or molecule. The important characteristic of ionizing radiation is the local release of large amounts of energy, sufficient to break strong chemical bonds that are biologically important. Ionizing radiations are classified as electromagnetic or particulate. The dose, or quantity, of radiation is expressed in terms of the energy absorbed per unit mass of tissue. The unit is the Gray, defined to be 1j/Kg ^[6]. X-ray gives rise to secondary electrons that are sparsely ionizing; they are said to be low-linear energy transfer (low-LET) radiation. By contrast, α particles, with greater mass and slower velocity, are densely ionizing and are described as high-LET radiation. Exposure to equal absorbed doses of high and low LET radiations does not result in the same biologic effect. The relative biological effectiveness (RBE) of high –LET radiations, such as neutrons and α particles, is greater than that of low-LET radiations, such as X-rays or γ -rays. RBE is the ratio of absorbed dose of a reference radiation, conventionally X-rays, to the absorbed dose of a test radiation to result in the same level of biological effect. This is true for chromosomal aberrations, cell lethality, oncogenic transformation in vitro or cancer induction in vivo. Double-strand breaks may be the most important radiation-induced lesion, and other evidence suggests that the interaction of two double-strand breaks may be responsible for many radiation-induced biologic effects.

Radiation doses to which the human population is exposed

Life on earth has evolved against a background of ionizing radiations arising from cosmic rays and from radioactivity in the earth. In addition, the human population is now exposed to various human-made or human-enhanced sources. The best estimate of the average annual effective dose equivalent from all sources to the US population is 3.6 millisievert ^[7]. The composition of

this dose is illustrated in figure 1.2 only in recent years has the importance of radon as the principle source of radiation to the US general population been recognized. The home is the source of this exposure. Radon is naturally occurring radioactive gas that emanated from the ground. Inside the home, it decays to short-lived radioactive progeny that attach to aerosol particles and are deposited in the tracheobronchial tree. The concern is the risk of lung cancer from the high LET α particles emitted during the decay of the progeny, polonium 218 and polonium 214^[8,9]. The potential exposure to radon varies widely in different parts of the country, depending on the uranium content of rock and soil; an even more uncertain factor is exposure to thoron, another radioactive gas that could contribute to lung dose

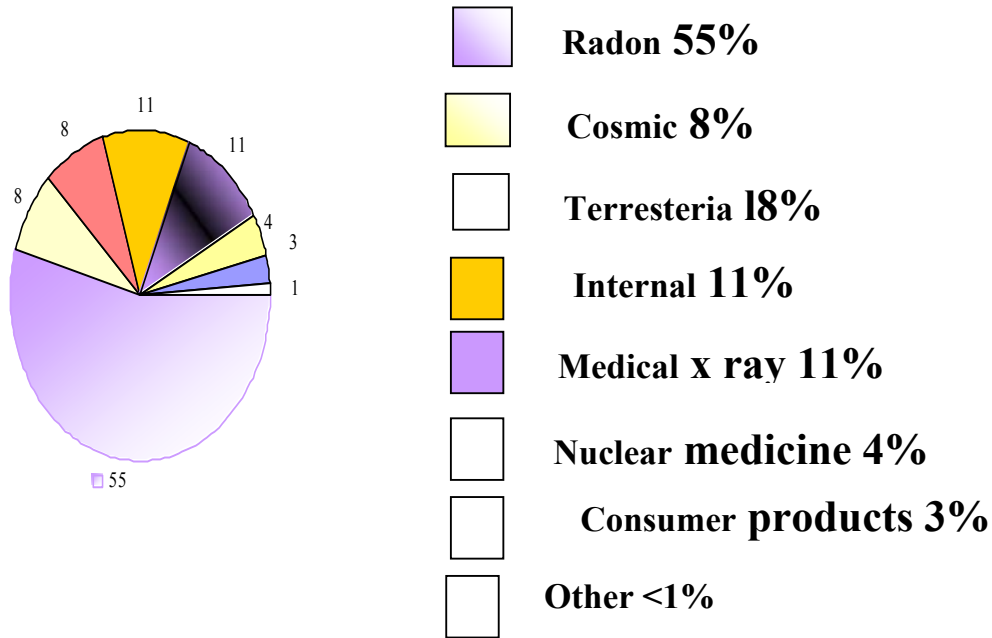


Figure 1.2: Estimated contribution of various radiation sources to the total average effective dose equivalent in the U.S. population

Radiation-induced cancer

There is a wealth of experience concerning cancer and leukemia induced in human populations by radiation, which is briefly summarized here:

1. Skin cancer was common in early X-ray workers, who worked around accelerators before radiation safety standards were introduced.
2. Lung cancer was documented in pitchblender miners and in uranium miners. Breathing radon gas led to the deposit in the lungs of radon progeny, which emit intense α rays^[10].
3. Bone tumors were observed in radium dial painters, who painted luminous dials on clock and watches with paint containing radium, which they ingested by licking the brush into a point^[11].
4. An excess incidence of liver cancer was reported in patients in whom the contrast material thorotrast was used; this contained thorium, which is an α emitter^[12].
5. The survivors of the atomic bomb attacks on Hiroshima and Nagasaki represent the most important single group. Leukemia and a whole spectrum of solid tumors have been observed^[13].

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6. A small excess incidence of leukemia has been observed in patients with ankylosing spondylitis who received radiotherapy for the relief of pain^[14, 15].
 7. Thyroid cancer has been reported in children irradiated for what was perceived to be an enlarged thymus, or epilated with X-rays for the treatment of ring worm of the scalp^[16, 18].
 8. There is an excess incidence of breast cancer in patients receiving radiotherapy for postpartum mastitis^[19], and also in patients with tuberculosis who underwent fluoroscopy many times during the management of artificial pneumo-thorax.

Some of these instances are of little more than historical interest, since the radiation doses involved are not known with any certainty, but others are sufficiently quantitative to allow estimates to be made of the risk of cancer as a function of dose.

Sensitivity of Different tissues

Animal experiments, as well as the historic human experience, shows that exposure of ionizing radiations, in sufficient doses, may result in the induction of cancer. The susceptibility of tissues varies widely, but all appear to be at risk^[20,21]. There is no obvious relation between natural susceptibility and sensitivity to radiation-induced cancer. For example, thyroid cancer has a low natural and high radiation incidence. Breast cancer has a high incidence of both natural and radiation-induced cancer. Colon cancer has a high and low radiation-induced incidence.

Stochastic nature of radiation induced cancer

radiation-induced cancer as well as the genetic effects of radiation are considered to be stochastic late effects a stochastic effect has two characteristics it has no threshold and the severity of the effect is independent of the dose although the probability of its occurring increases with dose the far-reaching implication of this is that any dose however small will carry with it some risk of inducing cancer there are two justifications for this assumption first experimental evidence suggests no threshold although of course it is not practical for those for dose response curves to be taken down to a very low doses second the suggested to mechanisms of cancer include processes such as a point mutation a chromosome deletion that could be the consequence of the passage of a single charge and practical track.

Latent period

there is always a Latent interval between irradiation and the appearance of an Induced malignancy leukaemia has the shortest Latent period excess cases appeared in the survivors of Hiroshima and Nagasaki by 5 to 7 years post irradiation. By contrast, solid tumors have a longer Latency and continue to appear in excess incidence for more than 40 years. Some of the latent interval may be due to the time required for The tumor to reach a sufficient size to be detectable on the other hand then processes of initiation promotion and progression also take time Ideas concerning the latent period have changed in recent years largely as a consequence of the continuing study of the Japanese survivors latency is no longer considered as a fixed time interval but rather solid cancers induced by radiation tend to appear at the age at which the naturally occurring cancer of the same type are seen.^[13] The latent period therefore is longer when exposure is early in life this suggests that age dependent host of factors are important and that the expression of cells initiated in the young are suppressed until the necessary host milieu is present

Dose- response relations

Dose response curves for the incidence of cancer as a function of dose are extremely complex. Current thinking is summarized in a greatly simplified form in figure 1.3 for low LET radiations, excess incidence is proportional to dose at low doses; as the dose increases, the

curve is concave upwards. At some point the curve bends over as cell killing eliminates over the further initiation of more cells to a transformed state. A curve of this general form has been observed for many malignancies in experimental animals notably and leukaemia in Mice and for oncogenic transformation in vitro ^[22,26]. For low -LET radiations at low dose rate, the dose-response curve remains linear over a wider range of doses and appears to be a continuation of the low -dose region of the acute Dose- response curve This pattern of response, observed experimentally, is predicted by models of carcinogenesis that involve interaction of sessions and that allow for repair For high-LET radiations, dose response relationships tend to steep and a linear junction of dose.

As a consequence RBE values are highly, particularly at low doses. in contrast to the situation for low-LET a radiations, the biologic effect of a given dose of high LRT radiation is not reduced by protraction of the does, either continuous low dose-rate, or by fractionation in the case of oncogenic transformation in vitro. Induction of mammary tumors in mice and even the induction of lung tumors in humans by radon inhalation, low dose rates appear to be more effective than high dose-rates biophysical models have been involved to account for this so called inverse dose rate effect.

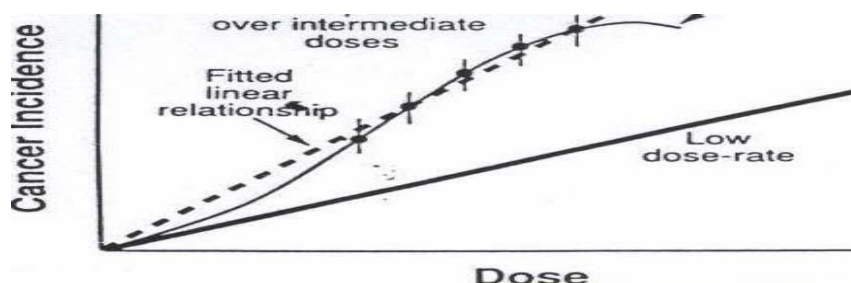


Figure 1.3: the data points for excess solid cancers in a human population exposed an acute dose of low dose linear energy transfer (LET) radiation are frequently fitted by a linear function of dose

Genetic factors

There is no clear proof of the influence of genetic factors on radiation- induced cancer, but some evidences is suggestive. The fact that in mice in there are strain differences in radiation- induced (as well as natural) incidences of cancer attests to the importance of inherited factors, although susceptibility may be related to host factors that influence expression rather than initiation. The cells of individuals with several genetic diseases have been shown to be sensitive in vitro to killing by ionizing radiation ^[27,29] or to the production of chromatic aberrations. Perhaps more to the point is that an elevated risk is associated with some Genetic conditions ^[30], notably ataxia-telangectasia^[31], but an increased susceptibility to radiation- induced cancer has not been shown. Second cancers in patients treated for cancer in childhood have a distinct pattern that reflects Genetic susceptibility, but a gain it is not clear whether this susceptibility extends to radiation-induced cancer.

Age and gender

Although the national incidence of cancer generally increases with age, the risk of radiation-induced cancer is often greater when exposure occurs at younger ages. This is true of cancers of the breast, lung, stomach ^[35], thyroid and connective tissue. The recent reanalysis of the atomic bomb survivors shows that the change of susceptibility with age is particularly dramatic for breast cancer in women. Survivors who were less than 10 years old at the time of exposure are most susceptible with risk decreasing steadily thereafter, to become very small by late middle age. In the case of leukaemias in humans, the age dependency of susceptibility is more complex .the risk of leukaemia was greater in 50 years-old spondylitic patients treated with x-

rays than in patients less than 25 years old. In the recent reevaluation of the atomic bomb survivors by the BEIR V committee, a greater sensitivity was noted for radiation induced leukaemia before than after the age 20 years. The overall risk of radiation-induced cancer is estimated to be slightly higher in women than that in men by about 10%.

The difference can be largely accounted for by gender-specific tumors, principally those in the breast, which appears to be a susceptible Tissue. Thyroid cancer after external radiation may also occur with higher frequency in women than in Men. On the other hand, Male atomic bomb survivors appear to have been at greater risk than female survivors for leukaemia and cancers of the respiratory system.

Risk of Carcinogenesis following radiotherapy

The risk of second malignancies following the radiation therapy is a subject not without controversy. Some investigators report that dose between 40 and 60 Gy. To limited areas do not significantly increase the incidence of second cancers. In contrast, others report excess carcinogenesis when substantial doses are given to healthy organs. Now the combination of chemotherapy and radiation therapy is so commonly used. It is increasingly difficult to dissect out the risk of radiation alone.

Mechanisms of radiation carcinogenesis

Human cancers frequently result from either the activation of a dominant acting oncogene or the deletion of a suppressor gene. These Ideas are particularly attractive because they provide a paradigm for explaining, common pathways for carcinogenesis by quite different agents. Oncogenes are known to be activated by a point mutation, a chromosomal translocation or by gene amplification.^[33, 36] The remove of a suppressor gene is likely to involve a small or large deletion. Radiation is known to be highly effective at producing deletions and chromosome translocations, and rather less efficient at point mutations. These mechanisms are attractive possibilities for radiation-induced cancer, but they have not so far been demonstrated in any specific human malignancy induced by radiation. Activated k-ras and N-ras were reported in a proportion of murine thymic lymphoma induced by X-rays neutrons but are probably not the causative step.

Ultraviolet radiation carcinogenesis

Skin cancers are the most common type of human cancers, basal cell carcinomas occur four times more frequently than squamous cell carcinoma in men, and six times more frequently in women. Both types of cancer occur more frequently in Men than in women and about 70 times more frequently in whites than in blacks.^[37] The cancers usually occur on areas exposed to sunlight, and at higher rates in southern latitudes of the United States. The incidence, particularly of basal cell carcinoma, is increasing but fortunately the cure rate exceeds 95% Solar UV radiation is a potent environmental DNA-damage agent and a known inducer of skin cancer. There is overwhelming evidence that chronic repeated exposure to solar UV is the primary cause of basal cell skin cancers^[38].

The incidence of various types of skin cancer is increasing in epidemic proportions. Because little has evidence suggests that UV radiation is directly involved in melanoma carcinogenesis by causing neoplastic transformation of melanocytes, it has been postulated that the effects of UV radiation on the immune system may play a role in the pathogenesis of this type of skin cancer.^[39-41] The marked increase in the incidence of melanoma in developed nations is a cause of considerable concern because of the high mortality rate associated with this form of cancer. An important question is whether the incidence of melanoma increases with a depletion of the ozone layer. The answer to this question depends on the validity of the causal relationship between melanoma and UVR, which is controversial and in doubt.

Variation in susceptibility to ultraviolet radiation-induced skin cancer

Skin cancer incidence rates vary markedly among different populations. Skin cancer is by far the most common form of cancer among the white populations, but is infrequent among darkly pigmented ethnic groups because of the protection from UVB afforded by melanin. The range of variation is about 50 fold. Those of Celtic ancestry have a particularly high susceptibility to skin cancer. For example, it was found in Philadelphia that those of Irish origin had about five times the incidence of skin cancer as Italians. In Hawaii, it was found that whites had more than 40 times the skin cancer incidence of Asians, While in New Mexico and other parts of the United States Anglos had a skin cancer incidence about six times that of Hispanics. In the case of malignant melanoma, genetic susceptibility and exposure to solar UV radiation are also thought to be the two most important risk factors .people with lesser amounts of skin pigment and those living at Latitudes with increased of UV exposure are at particular risk.

Ozone depletion and skin cancer

The ozone layer in the stratosphere acts as a highly effective absorbing layer that prevents the most biologically effective wavelengths of UVR, especially UVB (280 To 320) from reaching the earth and exposing the human population. Any increase in the fluence of UVB resulting from a depletion of the ozone layer might be expected to increase probability of skin cancer, especially basal cell and squamous cell carcinoma. In the early 1970s Concern about ozone depletion arose for the first time, especially in regard to skin cancer and damage to the eye. Early on it was feared that supersonic air planes would inject nitrogen oxides into the stratosphere resulting in ozone depletion. more recently, chlorofluorocarbons are suspected as a cause of ozone depletion in the stratosphere . There is some evidence of a global reduction in ozone, which causes concern that an increase in the skin cancer may result. Although depletion of the ozone layer leads to an increase in UVB, It has little effect on UVA. Using these new data for the complete UV spectrum and dose-response curves for carcinogenesis in mice, it is estimated that a1% decrease in ozone yields a 1.56% increase in carcinogenic UV, Which in turn may lead to a 2.7% Increase in nonmalanoma skin cancer.

Photo-carcinogenesis appears to involve at least three separate activities of UVR:

- *transformation of normal cells into neoplastic cells
- *the production of antigenic changes in the skin
- *the induction of suppressor T lymphocytes directed against these antigens

Asbestos

Classification of fibers

Asbestos is a broad commercial term for a group of naturally occurring hydrated mineral silicates that crystallize in a fibrous habit. The family of asbestos Minerals can be subclassified into serpentine, Meaning that the fibers are curly and pliable, and amphibole, which are needlelike fibers. Chrysotile, which accounts for 90% of the world's production of asbestos, is the most common fibrous serpentine. crocidolite and amosite are the most common amphibole fibers.

Historic perspective

The widespread use of asbestos attempts to control it, are relatively recent, but the mineral itself has been with us a long time .the Romans wove asbestos into tablecloths that could be tossed into the fire for cleaning.

At the turn of the century, Asbestos was being used on an industrial scale as a fire retardant and thermal insulator, and it was soon shown to cause asbestosis. Its association with lung cancer and mesothelioma in asbestos miners was demonstrated in the 1950s, And in textile weavers,

pipe insulators, and shipyard workers in the 1960s. ^[42-45] The danger of developing asbestos related diseases and fees to extend beyond that of a symbol of oppression and Hauser says it has been documented in family members of asbestos workers and in India he was leaving in the main road of industrial sources of asbestos, by the 1970s and 1980s, the government to action through the whole mission and safety and health administration and the environmental protection agency. Then you regulations will not a problem: Industry bore the cause and the where regarded as human and probably overdue.

Health risk of asbestos

Epidemiologic investigations of the association of mesothelioma with asbestos exposure indicate that mesothelioma is much more likely to result from exposure to amphibole than chrysotile fibers. However, an increase the incidence of lung cancer has been demonstrated from exposure to all types of assistance, an incidence that is synergistically into used by cigarette smoking. ^[46]

Among exposed workers, Lung cancer is the malignant disease most often associated with exposure to asbestos, Followed by Mesothelioma. There is also a modest increase in gastrointestinal cancers and some suggestion of an increase in other cancers such as those of the kidney, pancreas, esophagus, and colon. Mesothelioma is pathignomonic of fiber carcinogenesis, particularly asbestos. These tumors arise from the methelial surfaces of pleural and peritoneum and have a Pleomorphic histologic appearance. The tumors spread over pleural and peritoneal surfaces, do not invade the underlying tissues deeply, but do metastasize. Mesothelioma is difficult to treat and the prognosis, In general, poor. The risks posed by environmental exposure to mineral fibers are thought to be mainly mesothelioma. It appears to have affected significant numbers of people who may have had only low-level exposures to asbestos. Asbestosis, On the other hand, requires intensive exposures which are rare these days even in industry. Lung cancer is by far the most common cancer among asbestos workers, and is still a concern despite modern conditions, although most workers also smoke. Most experts believe, however, that for the general public, Smokers and nonsmokers alike, Lung cancer induction as a result of fiber exposure is less important than mesothelioma.

The electric and magnetic fields

Electric and magnetic fields are topics of much public interest but for which adequate scientific evidence to make judgments is the simply not available. Two quite different situations are of concern. The first is low-frequency electric and magnetic fields around electric power transmission lines. In practice, it is the magnetic component of the field that can penetrate the body and is likely to be of importance. The second is high static magnetic fields. Connected with magnetic resonance imaging (MRI) Systems, both clinical and experimental.

Extra low frequency electric and magnetic fields

Electric and magnetic fields produced , for example , by electric power transmission lines have recently been added to the list all the environmental agents that some believe pose a threat to public health by causing or promoting cancer . Conventional wisdom holds that the fields associated with power systems couldn't be a threat to human health since:

1. The physical interactions of such fees with the body are too weak to have a significant effect on biology at the level of they cell, since they are small compared with thermal noise.
2. Unlike X-rays, power frequency fields do not break chemical, bonds and are therefore unlikely to cause DNA breaks.
3. unlike microwaves, power frequency fields cannot cause significant tissue heating.

The question of an association between cancer and low-frequency of electromagnetic fields arose first because of epidemiologic studies on childhood cancer in Denver. This study reported a positive association between childhood cancer and homes they classified as near high-current configuration distribution lines, which are likely to produce stronger than average magnetic fields. Studies since then have yielded mixed results, two of them found no association^[47,48]. Two others reported positive results including that of savitz, which is the latest and by far the most thorough and complete study. Three studies were designed to examine the association between adult cancer and the residential exposure to electromagnetic fields, but there are so many confounding problems that they do not provide enough evidence to draw any meaningful conclusions. Occupational exposure is another matter.

There is an indication that occupational exposure is associated with an enhanced leukaemia risk. This doesn't of course necessarily imply a causative link, since industrial jobs often involve exposure to other hazardous agents particularly chemical against solvents

Positive correlations between cancer and extra low-frequency electric and magnetic fields (ELF-MF) based on epidemiologic data would be much more credible if there were parallel Laboratory data showing similar effects. Experiments to induce tumors directly in experimental animals or morphologic transformation with cells in vitro by exposure to ELF-MF Have all proved to be negative. However, other biologic endpoints have been reported to be affected by magnetic fields. But no reported response has been replicated widely in different in laboratories. Several studies indicate that ELF-MF can modify cell cycle kinetics.

Furthermore, since changes in cellular membranes caused by ELF-MF have been detected, they may also be of importance in the relation to cancer.^[49] Several studies also indicate that exposure to MF Can modify the gene expression patterns in cells.

In general, Transcription can be increased in salivary glands of the dipteran sciaracrophila After exposure to ELF-MF, And alterations in protein synthesis patterns are also observed. For example, there is some overlap between the proteins synthesized in a response to the heat shock and ELF-MF Treatment, but only the former causes a significant decrease in total cellular protein synthesis. In addition, transcription of β -actin, histone H2B, and myc RNA Are also altered in human HL60 cells after MF Treatment furthermore, peak increases in transcription appeared to occur in a narrow window of exposure parameters.^[50] By making and analyzing c-myc/CAT gene fusions, Lin and colleagues identified a 904 base-pair DNA region upstream of the C-myc gene that responds to ELF-MF Exposure.

The chemical induction of cancer

The molecular structure of chemical carcinogens

Occupational carcinogens:

The majority of chemicals known to be carcinogenic in man have been discovered because of attention was drawn to them. or to related to chemicals by investigations into occupational cancer Table 1.3. The recognition of a particular cancer hazard has tended to follow a predictable pattern. First, astute clinical observation has drawn attention to an excessive number of cases of a particular form of cancer in men working in a given employment. For example, pott (1775) noticed an abnormal number of cases of cancer of scrotal skin among chimney sweeps; Rehn (1895) reported an unusual clustering of cases of bladder cancer in a factory making the dyestuff fuchsin. Finally it has usually proved possible to demonstrate the carcinogenicity for laboratory animals of some contaminants of the industrial environment, and

then, by the use of further tests, to identify the actual chemicals responsible. Occupational skin cancer led to the discovery of carcinogenicity of certain polycyclic hydrocarbons, bladder cancer to that of a number of aromatic amines, some forms of lung and (In the case of nickel) nasal cancer to the knowledge that certain leukaemias to the carcinogenicity of radiochemicals.

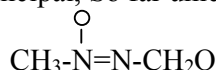
Naturally occurring carcinogens:

Naturally occurring carcinogens are now attracting more attention table 1.4. The fortuitous discovery of metabolites of the mould *asparagillus flavus*, Known as a flatoxins, as a result of the poisoning of some 100,000 Turkey bolts, led to the demonstration of the considerable hepatotoxicity and later hepatocarcinogenicity of a aflatoxin B1 And GII.



It was thought that because mould contamination was more likely in hot moist countries than in temperate climates, aflatoxin or other mould metabolites might be responsible for their relatively high incidence of human liver cancer in some parts of the tropics. Attempts have been made to substantiate this view by comparing the concentration of aflatoxins in foodstuffs purchased by people in areas with high and low incidences of liver cancer ^[51]. Although the results support the hypothesis that aflatoxin may be implicated, the small differences in incidence of liver cancer between the areas under study.

A lake of knowledge of the effects of different ways of preparing food for consumption, and the possible presence of other carcinogenic mould metabolites still leaves the significance of the association in doubt. Other natural products which may induce cancer and which have been found as contaminants in human foodstuffs include cycasin the glucoside of methylazoxymethanol III and a toxic principal, So far unidentified ^[52].



The glucoside of methylazoxymethanol III

Table 1.3: Extrinsic Carcinogenic Chemicals Discovered By Their Action In Man.

Tissue of election of human cancer or related condition	People affected	Class of carcinogen	example
skin	Chimney sweeps Pitch and tar workers Mule-spinners	hydrocarbons	3:4 benzopyrene
Bladder	Chemical workers Rubber workers Dyestuffs workers	Aromatic amines	2- naphthylamine
lung	Metal refiners Nickel processors	metals	Chromium nickel
liver	Chemical workers Liver cirrhosis	Dialkylnitrosamines	
Bone and leukaemia	Dial painters Radiochemical workers	radiochemicals	radium

Table 1.4: Examples of Naturally Occurring Carcinogens.

Source	species	carcinogen
Mould	Aspergillus flavus Aspergillus versicolor	Aflatoxin sterigmatocystin
Plant	Pteris aquilana Senscio, crotolaria heliotropium, etc Cycas circinalis	Compound C ₇ H ₈ O ₄ Pyrrolizidium Alkaloids ,e.g.retrorsine cycasin

The study of such naturally Occurring carcinogens may be of utmost importance in determining the causation of human and animal cancers hitherto regarded as spontaneous in origin.

Endogenous carcinogens

Endogenous carcinogens (i.e. carcinogens produced within the body) have attracted attention as possible causes of naturally occurring tumors (Table 1.5.) physiological levels of circulating hormones are normally maintained within the body by homeostatic regulatory mechanisms, Most of which involve the pituitary gland and other endocrine tissues. Distribution of homeostasis may lead to excessive stimulation of certain endocrine or endocrine-responsive tissues and ultimately, if the stimulus is sufficient and prolonged to cancer. Break down of homeostasis has been brought about by the administration of excessive amounts of oestrogenic substances or by surgical or chemical procedures which lead one or more endocrine tissues to over secrete certain hormones ^[53]. Cholesterol and certain metabolites of tryptophan ^[54] have been put forward as possible endogenous carcinogens.

The induction of these tumors was dependent on the vehicle used: oily solutions were effective whereas aqueous solutions were not. Bischoff (1963) suggested that the carcinogenic effect might be due to oxidation products present in the cholesterol, or that the tumours that rose in a response to a thin film of cholesterol being deposited in the tissues. Thin films or sponges of sufficient area made from a wide variety of materials of different chemical compositions, are able to induce sarcomas in the subcutaneous tissues of rats and mice. The status of tryptophan metabolites as endogenous carcinogens is more difficult to evaluate. Price (1966) Found that all the aromatic amines normally present in human urine were metabolites of the essential amino acid, tryptophan. Patients with bladder cancer, not exposed to occupational bladder carcinogens excreted more of these substances in their urine than subjects without bladder cancer; the difference was especially marked after a loading dose of L-tryptophan.

The metabolites were shown to possess carcinogenic activity by the bladder implantation technique ^[55] but not by more conventional methods. Bladder implantation is not, however, a satisfactory method of carcinogenic testing. More recent investigations of patients with bladder cancer failed to demonstrate as high a proportion of individuals with abnormal tryptophan metabolism as in former series ^[56].

Table 1.5: Endogenous Chemical Carcinogens.

Type of compound	Tissues affected	species	examples
Hormones	Endocrine (pituitary ,ovary) Endocrine dependent (breast)	Various	Oestrogens
Cholesterol tryptophan metabolites	Subcutaneous tissues Bladder	Mouse ,rat mouse	Cholesterol dihydroxy-anthranilic acid

Metabolism of chemical carcinogens with Emphasis on biochemical activation mechanisms

Detoxification reactions :- Among the classes of chemical carcinogens in responsive hosts, one, known as direct-acting, has the proper activity intrinsic in its chemical structures^[57]. Alkylating agents, Such as nitrogen Mustard, Melphan and busulfan, are used extensively in cancer chemotherapy and belong to this group. Others are strained-ring lactones as β _propiolactone, epoxides as butadiene diepoxide, imines as ethylene- and propyleneimine, esters such as methyl or ethyl methanesulfonate, dimethyl sulfate, acylium compounds like dimethylcarbamoylchloride or reactive bis (chloromethyl) ether^[58].

All of these compounds contain "leaving groups" or form reactive carbonium ions, which can interact directly with suitable substrates relevant to carcinogenesis as will be discussed. They are directly mutagenic without need for biochemical activation in bacterial and mammalian cell systems or cause cell transformation readily. Depending on their structure, these agents do undergo metabolism. These metabolic reactions, however, are usually detoxification reactions; that is, they reduce or indeed most often, eliminate any pharmacology effect. Such reactions convert the active entity into an inactive product, generally to such a form that excretory mechanisms via the renal-urinary or the hepatic-biliary pathway lead to elimination.

Activation reactions :- not only do chemical carcinogens undergo a variety of structural-dependent detoxification reactions, which facilitate their excretion, but many, called procarcinogens, also are subject to biochemical activation reactions. Often they are toxic and carcinogenic by virtue of such reactions. The relative effectiveness of an agent, under a given conditions, hinges on the efficiency of the activation reactions over the competing detoxification reactions. Genotypic and phenotypic influences such as species, strain, sex and the action of other endogenous or exogenous agents all ultimately express their major effective in the ratio of the activation over detoxification metabolites, although there are also secondary forces, to be discussed, as regards the availability of cellular and molecular receptors factors. Therefore, an understanding of the biochemical activation processes much of which was secured in the last 20 years is crucial to the eventual comprehension of the entire carcinogenic process^[59]. In almost all instances the known activation reactions are mediated by a membrane attached complex enzyme system in the endoplasmic reticulum of cells.

The enzyme systems most often studied are located in the liver a key organ for such reactions, but these systems occur in different amounts in virtually all tissues. Because the early discoveries with these enzymes systems were made in connection with the examination of the metabolic fate of exogenous chemicals and drugs, They were thought of as being related uniquely to the metabolic handling of exogenous agents in the environment. Conny, however, drew attention to the fact that steroid hormones are also metabolized by the same enzymes and that developmentally these enzyme systems are concerned with the metabolism of such endogenous substances. Active work in the entire field of drug metabolism has yielded important insights that could be applied to the specific cases of chemical carcinogens.

On the other hand, few drugs have undergone as comprehensive and detailed examination of metabolism as have carcinogens, so that substantial general advances in this area have resulted their from. Also, the study of chemical carcinogens has given the first indication that such metabolic reaction leads to compounds of higher toxicity and carcinogenicity a sort of lethal synthesis. Contemporary concepts visualize not a single enzyme system, but a family of closely related enzymes located on the endoplasmic reticulum in the cell. During fractionation by ultracentrifugation of the cell components, the enzymes are found in the Microsome fraction. There is evidence for several distinct systems, as shown by differences in UV spectra and

substrate specificity of induced compared to basal enzymes. Also, the enzyme system has been solubilized and fractionated^[60].

A reconstituted was fully competent and active. Studies of overall metabolic fate of drugs and carcinogens, as well as, in vitro tests utilizing Microsome fractions of cells, have yielded valuable information on the properties of these systems. These mixed function oxidases include a hemoprotein component, cytochrome-p450 coupled to cytochrome reductase, itself a flavoprotein figure 1.4 cofactor requirements include NADPH, magnesium ions, and, because the reactions are generally oxidative, oxygen. Studies with isotopic oxygen -18 in the metabolites come from atmospheric oxygen and not from oxygen in water.

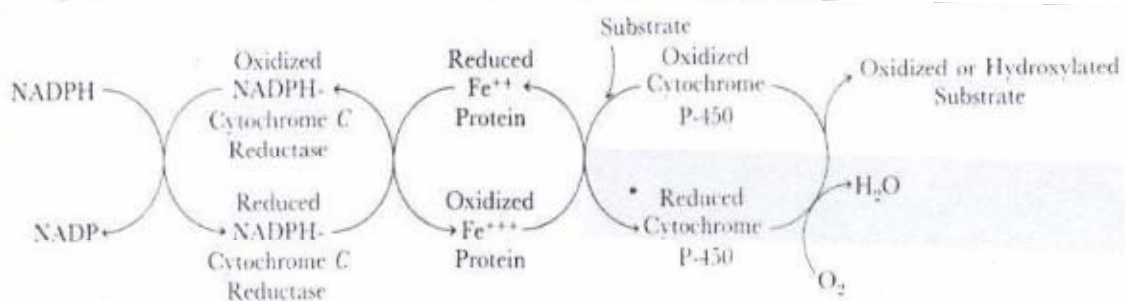


Figure 1.4: the cytochrome p-450 enzyme system, a family of enzymes, present in many tissues.

A special kind of reductive activation reaction, the reduction of certain carcinogenic nitro compounds, particularly of nitroaryl derivatives to the active hydroxylamino compounds, is mediated in part by an enzyme on the endoplasmic reticulum, and also by a soluble enzyme system. Not to be neglected in the overall in vivo metabolic fate of drugs and carcinogens is the contribution made by enzymes in the microbiologic flora of the gut. In the main, these enzymes are hydrolytic or reductive and are important in enterohepatic cycle. Detail investigation of the then curious inhibition of liver tumor induction in rats by 4-dimethylaminoazobenzene, When the poly nuclear aromatic hydrocarbon carcinogen, 3-methylcholanthrene, was fed at the same time, Resulted in the development of an entirely new field of pharmacology, with broad application to medicine.

It was, indeed, discovered by Conney and the Miller that the underlying mechanism rested on the induction, by the hydrocarbon, of an enzyme system that reduced the azo bond in the carcinogenic Azo dye yielding noncarcinogenic inactive products, and thus accounting for the inhibition. Further exploration of this important discovery led to the now well-known concepts of enzyme a regulation by chemicals and drugs. Problems of Addiction and tolerance are now in part explicable by increases in enzyme systems that lead to detoxified or inactive materials, and thus higher dosages of drugs are required to achieve a given pharmacologic effect .the reverse, namely increased toxicity , or synergism in between drugs , also can be traced to such factors . In this case, decreased levels of detoxifying enzymes, or alternately increased levels of enzyme leading to activated intermediates, augment the activity of the agent^[61].

These problems are of particular importance in relation to synergism between carcinogens of different types were considerably reduced dosages of individual agents can combine to yield tumors out of proportion to the quantities of each agent . Moreover, dependent upon the direction in which carcinogenic agents affect the overall activation and detoxification enzymes, Sizeable augmentation or decrease in carcinogenic effect may eventuate. It is mainly because of the actual or probable presence of such interactions that contemporary concepts, embodied in legislation in the United States of safety of food additives and, in part, of drugs have arisen. As discussed elsewhere, Individual carcinogens under specified conditions do exhibit classic dose-

response curves as is true for other drugs or toxicants. The induced tumor yield is proportional to dose and the latent period inversely proportional.

However a no effect dose level in such a test series may be modified to a cancer-inducing dose by addition of an otherwise innocuous material if it affects the enzyme system concerned with activation or deactivation of the carcinogen. Nonetheless, there is experimental and human evidence that low dose levels of certain carcinogens have no effect, Even though they are highly active at higher doses. Yet, Modern analytical technology can the readily detect and quantify such low doses, as for example a few parts per billion of dimethylnitrosamine in some foods. Another aspect of the biochemical activation of carcinogens is that in most instances only a minute Proportion of the dose metabolized to the active agent.

Most of a given material Administered is converted to a number of inactive detoxification products. This point needs to be kept in mind when the fate of environmental chemicals and drugs is being examined. Usually, metabolites produced in largest amount are not carcinogenic. For this reason, the older semi-quantitative methods identified the major metabolites, but often failed to detect the critical carcinogenic metabolite. Assessing mutagenicity of metabolite is a great contemporary aid in discriminating between the important "activation pathway" metabolites and detoxified products. The fact that only a minor fraction of a dose is converted to the active carcinogenic metabolite also bears on the important question of no effect levels^[62].

It may take millions of active molecules to reach the key target, Perhaps as millions of sperms are required to effect fertilization. With carcinogens, the initial reaction may even be partially reversed through repair. Furthermore, Not all cells, at diverse as stages of the mitotic cycle, For example, Are equally sensitive. Even though "carcinogen" Is delivered pharmacology to large numbers of cells, only one cell or a few cells are transformed and become neoplastic.

Structural classes of carcinogenic chemicals

On the bases of conceptual mode of action and pragmatic factors, Cancer producing agents can be divided into eight classes (Table 1.6.)

Direct acting chemicals

A direct acting carcinogenic chemical doesn't require the participation of the host organism to produce an active metabolite. The chemical itself is the ultimate carcinogenic entity. There are relative few such compounds. Because as they are generally highly reactive, the readily undergo detoxification or inactivation. In fact, in some cases their reactivity is so great that their effect can be demonstrated only in vitro. Insertion in an animal system leads to decomposition prior to reaching the sensitive target cells, Hence the inactivity. On the other hand, if such a compound has a sufficient half-life in vivo to reach target Organs, It may be a powerful carcinogen in a broad spectrum of species without regard for other environmental conditions.

Most direct-acting carcinogens are products of industry, as chemical intermediates or developed as drugs for specific purposes. Modern chemical industry depends on reactive substances, by definition. It is important, therefore that such chemicals be tested for carcinogenic potential, so that medical directors and others in industry can take appropriate steps to safeguard their staffs and others who may potentially be exposed. That the risk is real in handling direct-acting agents, mostly alkylating chemicals, is demonstrated by occupational cancers that have been seen where adequate precautions were not take, either because management was unaware of the Hazard or because the staff was not sufficiently briefed. Examples of compounds of this type are listed to in (table 1.7.)

Alkylating agents that are effective in the treatment of some types of neoplastic diseases are often quite reactive and carcinogenic in experimental systems.^[63] Recently, Substantial animal experimentation has redirected attention to this area, Confirming Fully the prediction that such compounds might be carcinogenic. This is consistent with clinical observations. There is a significant increase in secondary neoplasia in patients that had been previously treated with cancer chemotherapeutic drugs. With more information on relative carcinogenic hazards of such drug in experimental systems, the clinician maybe in a position to select the most efficient chemotherapeutic agent with the least carcinogenic risk.

Table 1.6: Classes of Carcinogenic Chemicals.

Type	Mode of action	Example
-Direct-acting ultimate carcinogen	Electrophile, organic compound, Genotoxic,interacts with DNA	Ethylene imine, bis(chloromethyl) ether
-procarcinogens	Requires conversion through metabolic activation by host or in vitro to type 1	Vinyl chloride,benzo (a)pyrene,2-naphthylamine, dimethylnitrosamine
-solid state carcinogen	Exact mechanism unknown; usually affects only mesenchymal cells and tissues;physical form vital	Polymer or metal foils; asbestos
-inorganic carcinogen	Not directly genotoxic, leads to changes in DNA by selective alteration in fidelity of DNA replication	Nickel, chromium
-hormone	Usually not genotoxic; mainly alters endocrine system balance and differentiation; often acts as promoter	Estradiol, diethylstilbestrol
-immunosuppressor	Usually not genotoxic; mainly stimulates "virally induced," transplanted, or metastatic neoplasms	Azathioprine, antilymphocytic serum
-co carcinogen	Not genotoxic or carcinogenic, but enhances effect of type 1 or type 2 agent when given At the same time. May modify conversion of type 2 to type 1	Phorbol esters, catechol
-promoter	Not genotoxic or carcinogenic, but enhances effect of type 1 or type 2 agent when given subsequently	Phorbol esters, phenol, bile acids, tryptophan metabolites

Table 1.7: some direct acting chemical

1,4-Butanediol dimethanesulfonate (Myleran) Dimethyl sulfate	$\text{CH}_3\text{SO}_2\text{O}(\text{CH}_2)_4\text{OSO}_2\text{CH}_3$ $\text{CH}_3\text{OSO}_2\text{OCH}_3$
Bis(2-chloroethyl)sulfide (mustard gas or yperite)	ClCH_2CH_2 S ClCH_2CH_2
Nitrogen mustard (HN_2)	ClCH_2CH_2 NCH ₃ ClCH_2CH_2
Melphalan (sarcolysin)	ClCH_2CH_2 N—C ₆ H ₄ CH ₂ CHCOOH NH ₂
Chlorambucil	ClCH_2CH_2 N—C ₆ H ₄ CH ₂ CH ₂ CH ₂ COOH
Cyclophosphamide† (endoxan; cytoxan)	ClCH_2CH_2 N—P(=O)(N ₂ C ₂ H ₄ O) ClCH_2CH_2
2-Naphthylamine mustard† (chlornaphazine)	ClCH_2CH_2 N—C ₁₀ H ₇
Bis(chloromethyl)ether	$\text{ClCH}_2\text{OCH}_2\text{Cl}$
Benzyl chloride	$\text{C}_6\text{H}_5\text{CH}_2\text{Cl}$
Methyl iodide	CH_3I
Diazomethane	CH_2N_2

Chemicals active after metabolic activation: procarcinogens

Most of the carcinogenic chemicals fall into this class. Metabolic activation is only one of several competing reactions and depends on endogenous and exogenous factors, including species, sex, diet and the effect of other agents. Hence, such compounds are carcinogenic under more selective conditions than are the direct-acting carcinogens. Even so, some are highly active in minute amounts.

Whether a given chemical is oncogenic under a specified laboratory or environmental situation depends strongly on the relative ratio of metabolic activation versus detoxification pathways.^[64] Endogenous or genetic as well as exogenous or environmental conditions that favor activation reactions increase carcinogenicity. In contrast such conditions that increasing detoxification lower carcinogenicity (figure 1.5.)

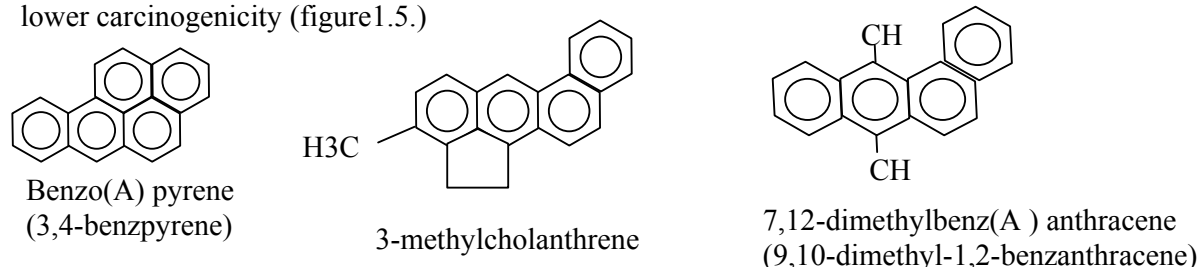


Figure 1.5: structural formulas of typical carcinogenic polynuclear aromatic hydrocarbons.

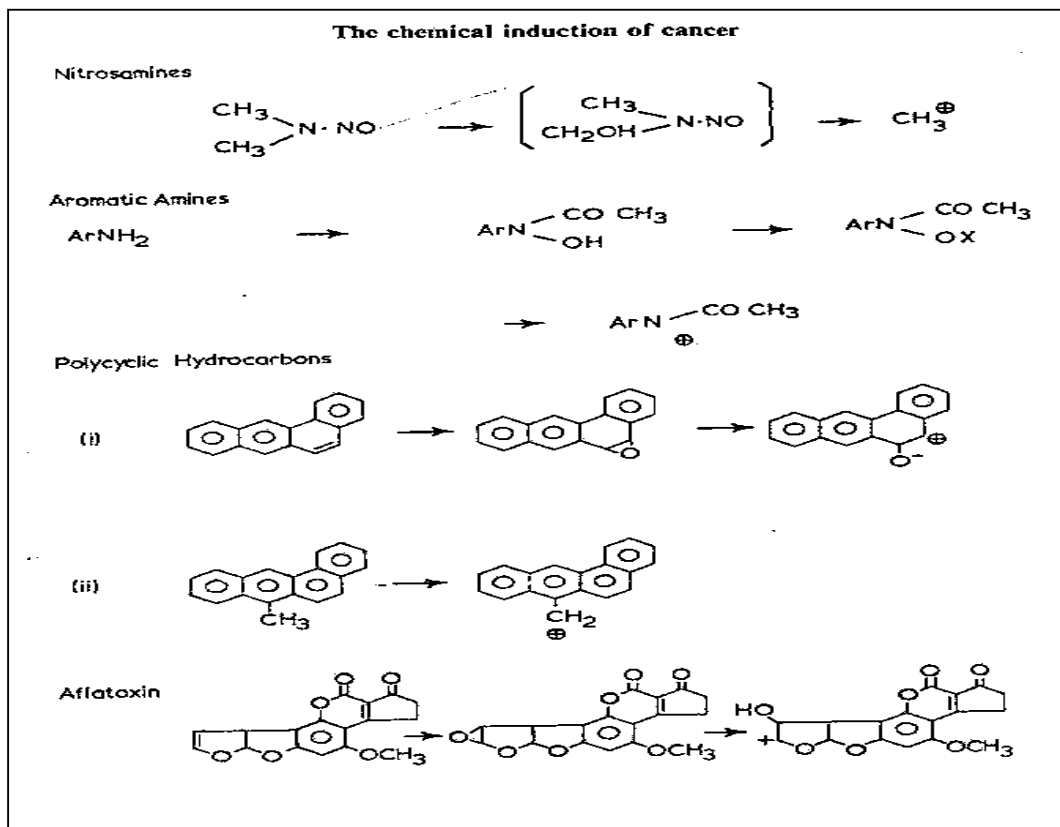


Figure 1.6: The chemical induction of cancer

It was found that many of the tumors induced by aromatic amines occurred at sites of metabolism or excretion of the test compound- kidney, bladder, intestine or liver rather than at sites such as the mouth or stomach, where orally administered to carcinogens would be expected to reach the highest concentrations. This led to the suggestion that the aromatic amines to be metabolize before they become active carcinogens. The first substantial information concerning the nature of metabolic activation followed from the demonstration of a novel metabolic pathway in which is the aromatic amine derivative, 2-acetylaminofluorene (AAF-IV), was converted to N-hydroxy-2-acetylaminofluorene (N-hydroxy-AAF-V) ^[65]

Interaction of carcinogens with tissue components

The presence of covalently bound carcinogen residues on many cellular macromolecules is the consequence of the interaction of the metabolically activated carcinogens with nucleophilic groups in the tissues. Since the earliest demonstration of the carcinogen-protein binding there have been many attempts to link such binding with the ultimate appearance of tumors. Speculation is at present centered around three types of interaction - (a) Carcinogens to DNA; (b) Carcinogens to protein, And (c) Carcinogens to transfer RNA: All three of them are discussed in more details.

Binding to DNA

The idea that cancer might be the result of one or more interactions in the genome of somatic cells has had many adherents, since Boveri (1929) Reported that the chromatin of cancer cells stained irregularly compared to that of normal cells. The somatic mutation hypothesis is particular apposite for speculation because, assuming that mutation occurs in genes with specified actions and that a suitable selection process operates, It is possible to account for the majority of biological phenomena. Substantial evidence is much more difficult to obtain. Earlier objections to the somatic mutation hypothesis were based on the observations that several

important classes of carcinogen, including the polycyclic hydrocarbons and aromatic amines, were inefficient at inducing mutations in the monocellular organisms used in the classical work on chemical mutagenesis.

It was realized that chemical carcinogens require metabolic activation and that the monocellular organisms lack the necessary metabolizing enzymes has been followed by experiments which show that the active metabolites are potently mutagenic and can for example mutate isolated phage^[66]. A mutational hypothesis for the induction of cancer by chemicals as well as by radiation is therefore a real possibility. It shouldn't be thought, however, that every molecule of carcinogen which reacts with DNA necessarily induces a mutation; damage to DNA in most mammalian systems can be repaired by enzymes present in the cells affected. Furthermore, particularly with carcinogens of low molecular weight (for example, the methylating agents such as dimethylnitrosamine), it has been shown that deposition in the DNA molecule at which binding occurs is vitally important in the induction of mutations.

The extent of binding to DNA *in vivo* varies according to the agent, the dose and its susceptibility to metabolic activation. Brookes (1966) in a review of the literature, reported that six moles of sulfur mustard bound to 10.5 nucleotide pairs in DNA in mouse ascites cells whereas only 0.75 moles of benzo (a) pyrene bound to the same number of nucleotide pairs in mouse skin. If this binding is random and leads to simple point mutations, the chance of obtaining the series of specific mutations postulated to be necessary for the induction of cancer would appear to be remote. Yet results obtained in *in vitro* studies suggest that malignant as well as morphological transformation is not uncommon. For example, Chen and Heidelberger (1969) induced many morphological transformations by adding 3-methylcholanthrene (1 microg/ml) to adult C3H mouse ventral prostate cells in culture. Implantation of only 100 of these transformed cells into each of three isologous hosts led to tumors in two mice.

This suggests that a mechanism other than specific somatic mutation may operate in cancer induction. These observations, however, may be artefactual in view of the finding that the addition of a chemical carcinogen, diethylnitrosamine, to rat embryo cells infected with Rauscher virus led to morphological transformation, whereas neither virus nor chemical alone had this effect^[67]. Before accepting that certain results contra-indicate a somatic mutation mechanism for cancer induction, it is necessary to establish that the cells used in transformation experiments with chemicals do not contain incomplete cancer in using viruses.

Binding to Protein

The demonstration that in certain systems carcinogen to protein binding could be correlated with the degree of activity of the carcinogen led to attempts to explain the mechanism of cancer induction. The enzyme deletion hypothesis represented the first of such attempts. It was suggested that binding to cell protein led to the deletion of enzymes controlling growth and thus to cancer. The mechanism by which interaction with a protein could lead to its elimination was not at that time explained. Further work showed that certain proteins had a greater affinity for activated carcinogens than others. Sorof et al. (1960), for example, showed that a group of proteins characterized by their electrophoretic mobility. The H2 proteins had the highest affinity for 4-methylaminoazobenzene metabolites of all proteins in the soluble fraction of rat liver; and a similar group of polycyclic hydrocarbon-binding proteins was demonstrated in mouse skin^[68].

Binding to transfer RNA

Transfer RNA, as its name implies, is responsible for aligning the correct amino acid with messenger RNA in the ribosome during the protein synthesis. Aberrations in transfer RNA could lead to the production of abnormal proteins and thus interfere with the internal economy of the cell. The significance of transfer-RNA in carcinogenesis has recently been discussed^[69]. The

degree of methylation of tRNA is higher in foetal and in cancer tissues than in normal adults and it was believed that one action of methylating carcinogens such as dimethylnitrosamine, might be to increase the level of methylation of tRNA. The methylating carcinogens act on different bases in transfer-RNA than the methyl transferases naturally present in the tissue.

The practical implications of metabolic activation

It is current to practice to test many compounds intended for use as food additives, growth promoters for livestock, Pesticides, or pharmaceutical agents for possible carcinogenicity in animals before they are permitted to be used in the human environment. Difference in capacity for metabolic activation is a major reason why species differ in their response to carcinogens as metabolically activated carcinogens are usually highly reactive, they may be detected and quantified by the use of trapping agents. For example, the use of methionine or guanosine to trap the intermediate produced by the sulphation of N-hydroxyAAF has already been discussed. More recently the use of bacteria in combination with microsomal and activating system has been advocated^[70]. The end point may be the toxicity of the activated carcinogen to the bacteria, Measured by the reduction in the number of viable colonies found on sub- culture. Alternatively, The Induction of mutations by the activated carcinogen may provide a more appropriate parameter.

Natural products, food contaminants and additives, and Drugs.

Mold toxins. About 1960 an epidemic destroyed half of the commercial turkey Production in Great Britain. Examination of the possible causes soon focused attention on a dietary factor and incriminated to a peanut meal with an apparent Mold contamination by certain species of *aspergillus flavus*.^[71] Cultures of the mold produced a toxin, Small amounts of which were fatal to ducklings. Several chemically closely related toxins were isolated by a combination of the duckling test and the toxin- associated specific fluorescence.

The complex structure of the active components was resolved rapidly by Buchi and Wogan, a major achievement in natural product chemistry. 4 main aflatoxins were identified B1, B2, G1, and G2, The B Series being so named because of a blue fluorescence, and the G Series, Green (figure 1.6.) There are major differences in toxicity and carcinogenicity, however, Aflatoxin B1 Is the most toxic and carcinogenic in rats and several other species, including monkeys. Affected are mainly the liver and less frequently the kidneys, and colon. In the rats, even one micro gram per kilogram of diet was carcinogenic.

This chemical is one of the most potent carcinogens known. Interestingly, Mice (Except for when exposed as newborns) Do not show liver tumors for reasons that are not yet clear, But some strains are sensitive to lung tumor induction. Both rats and Mice develop sarcomas at the point of subcutaneous injection. Aflatoxin G1 is less carcinogenic, and aflatoxin B2 And g2 are only slightly active. Aflatoxin B2 is converted to aflatoxin B1 by enzymatic dehydrogenation, and it is thought that the activity of the B2 analog is due to a small extent of conversion to the B1 isomer. α and β unsaturated lactone part of the molecule is not essential to the carcinogenic effect.

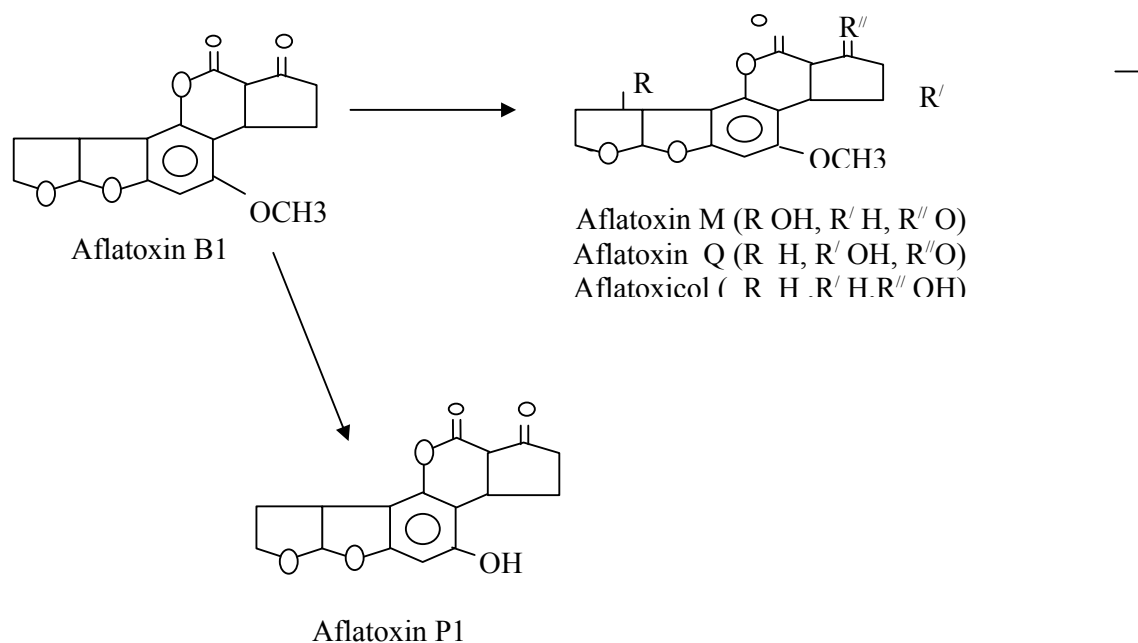


Figure 1.6: the important aflatoxin B1 is metabolized to aflatoxin Pand aflatoxinQ (excrete in urine as conjugate) and aflatoxin M.

A number of other metabolites have been identified. Aflatoxin M1 Was found in the milk of cattle consuming aflatoxin contaminated food. It is carcinogenic. Aflatoxin P1, Stemming from the oxidation of the methoxy group, was thought to be useful in evaluating possible human exposure, but has not been reliably detectable. Other metabolites identified include aflatoxin Q1, Stemming from hydroxylation of the 5-Membered ring system in the coumarin end of the molecule. It would be useful to have available specific sensitive methods to search for aflatoxin metabolites in body fluids, So as to have a measure of aflatoxin intake in studies of populations with the high risk for liver and other types of cancer. Recently, the plasma level of a flatoxicol, formed from aflatoxin B1 by a keto-reductase, was proposed as such a diagnostic indicator of sensitivity.

Drugs and Antibiotics.

Drugs used in cancer chemotherapy can be classified broadly into four types: alkylating Agents, antimetabolites, Antibiotics, and miscellaneous. As may be expected, Alkylating agents are carcinogenic. Their carcinogenic potency varies, and the use of such agents in the clinic is being influenced increasingly by such knowledge. Formerly, it was expected that antimetabolites would not be carcinogenic, since they did not seem to be alkylating agents or metabolized to such. However, several of these agents, including 6-mercaptopurine and azathioprine, among others, are carcinogenic in animals, inducing leukaemias, lymphosarcomas, and reticulum cell sarcomas. This carcinogenicity probably doesn't depend on a genotoxic effect, but rather may be related to immunosuppressive properties of these drugs, facilitating the expression of an oncogenic virus and characteristically yielding the observed tumors.

These animal findings have their Counterpart in man. Treatment of patients with both types of these drugs has resulted in an increased incidence of subsequent neoplasms, especially Leukemias. Among the natural products, Several Antibiotics tested were found to induce tumors. Subcutaneous injection of actinomycin D, or of mitomycin or patulin, Yielded sarcomas at the point of injection, but apparently no distant lesions⁷²¹. On the other hand, griseofulvin,

when fed in the diet at high levels to mice, led to liver tumor induction. It also potentiated the effect of cutaneously applied 3-methylchol-anthrene. One explanation offered for these findings was that griseofulvin might affect immunologic competence. Daunomycin and adriamycin, Used in cancer chemotherapy are effective experimental carcinogens, producing particularly kidney and mammary tumors.

The mode of action of these agents is under investigation. It remains to be determined further whether they are direct-acting or require biochemical activation. According to clinical case reports, the drugs phenylbutazone and diphenylhydantoin have led to leukaemia or lymphoma in man, Perhaps, related to biotransformation to a specific leukemogenic intermediate. Animal experiments with these three compounds have not reliably induced leukaemia or other forms of neoplasia. Thus, studies on the active intermediate by biochemical means cannot be readily devised. It could be, However, that epoxides derived from aromatic ring deserve consideration.

Plant products.

The pyrrolizidine or senecio-alkaloids are constituents of some herbal medicines and teas. These materials have powerful pharmacologic effects. Their salient adverse action is on the liver, where megalocytosis consequent to perturbed cytokinesis occurs, especially when given in low doses chronically. Infrequent administration of large doses followed by lengthy observational Periods results in cancer, mainly in the liver.

More epidemiologic and nutritional studies are needed to establish whether these materials alone or acting together with other harmful components such as the aflatoxins might be related to the presence of liver cancer in man, especially in countries where infants and children are treated with teas and concoctions containing such agents. Several active metabolite intermediates of pyrrolizidine alkaloids have been proposed. Evidence for an oxidative ring rearrangement yielding an active ester intermediate has been presented.

Bracken fern. Bladder cancer occurs sporadically in cattle in some parts of the world.^[73] The Disease was eventually proved to be related to consumption of Bracken fern deliberated exposure of cows reproduced the disease, proving that the plant indeed contains an active carcinogen. Bracken fern led to cancer in the rats, mainly in the small intestine, and especially with Supplementary thiamine, in the bladder. The active component has not yet been identified. In some countries, Cooked or row bracken fern is used for human consumption. More information is required on the relationship of this and other plant materials to human cancer.

Safrole. This component of sassafras oil was formerly added as flavoring agents to root beer and was present in other extracts to which man was exposed. Large doses of safrol induce liver tumors in rats and dihydrosafrol cancer of the oesophagus. On the basis of these findings, Safrole and derivatives are no longer utilized as a food additive. Considering the need to feed high doses for extended periods, an active metabolite is probably produced in only small amounts. This appears to be the more carcinogenic I-oxidation products, which in turn may require esterification, perhaps, by sulfate, as was described for the aromatic amines. Alternatively, the further participation by the allylic Group with epoxidation may also play a role. Allyl alcohol itself is highly hepatotoxic. It is not to known whether it is carcinogenic.

Tannins. The carcinogenicity of complex tannins was discovered inadvertently when tannins-containing salves were applied to burn patients during world war2 in central Europe. These patients develop Liver toxicity. Laboratory investigation showed that subcutaneous injection of such tannins induced Hepatocellular carcinoma in rats, although, oral intake failed to do so. More recently, Tannins have been identified in certain West Indian plants, the extracts of which caused local sarcomas upon subcutaneous injection. Tannins are complex natural products

which not only oxidize easily, but also can act as carriers for contaminants. Thus, their possible carcinogenicity and mode of action require further study. We have found, however, that 2% dietary tannin inhibited colon cancer induction by azoxymethane.

carrageenans. Carrageenans isolated from seaweed are used extensively as food additives.¹⁷⁴ A degraded, low molecular weight carrageenan used mainly in Europe has led to duodenal ulcer formation in animals and man. The natural high molecular weight carrageenan used in the United States has thus far been found safe. However, a recent abstract reports that the administration of 15% carrageenan, a high level in the diet of rats, potentiated colon carcinogenesis.

Tryptophan and derivatives. Suspicion that tryptophan may be involved in cancer production rested on the fact that metabolites like 3-hydroxykynurenine, 3-hydroxy anthranilic acid, and xanthurenic acid led to significant increase in bladder tumors when cholesterol pellets containing them were implanted in mice. Anthranilic acid is not active. Earlier, it was shown that the administration of sizable amounts of tryptophan, together with the carcinogen N-2-fluorenylacetamide, to rats yielded bladder cancer, whereas without tryptophan liver cancer only was seen. Also, hyperplasia was noted in the bladder of dogs fed large amounts of tryptophan for many years. In addition, bladder cancer was seen in a small number of dogs pretreated with limited amounts of 2-naphthylamine or 4-aminobiphenyl insufficient to yield bladder cancer by themselves, when tryptophan was given additionally.

Some tryptophan metabolites are reported as leukemogenic in mice. More research with this amino acid and derivatives to delineate its cancer causing or promoting potential is important, especially with respect to cancer of the bladder a standard national cancer institute (NCI) Bioassay With 5% dietary tryptophan has not shown carcinogenicity in rats or mice. Contemporary views suggest that tryptophan metabolites may not necessarily be carcinogenic, but promote the effect of other agents.

Purines and pyrimidines. Certain synthetic analogs of nucleic acid building blocks, 3-hydroxyxanthine and guanine-3-N-oxide, have carcinogenic potential. Subcutaneous injection in rats yields not only local sarcomas, but also a number of visceral tumors, such as those of the liver. While the evidence is not complete, these derivatives do not seem to arise during normal metabolism of nucleic acids, nor are they known to be present in foods.

Artificial sweeteners

In one experimental series, cyclamate (cyclohexylsulfamate) given to rats in high dosages has led to bladder cancer after an extended latent period. This led to its removal from the market in the United States and several other countries. It was suspected that cyclohexylamine, a metabolite produced by the intestinal flora, might be an active carcinogen. However, extensive tests failed to demonstrate this. The rats utilized in the tests of cyclamate were not free of specific pathogens, and the urinary bladder was infected with the parasite *Trichosomoides crassicaudicum*. The role of this parasite in bladder cancer induction with cyclamate remains to be defined. However, several tests of cyclamate in mice and rats free of specific pathogens have failed to confirm the original study.

With the aromatic amine carcinogen N-2-fluorenylacetamide, the parasite infestation has a synergistic effect in bladder carcinogenesis. Bladder infestation with schistosomes in countries like Egypt is associated with the presence of bladder cancer in man, through as yet unknown mechanisms. cyclamate is not genotoxic. Thus, the bladder cancers in rats, which triggered its ban, may have been due to a mode of action other than direct carcinogenicity. There is no epidemiologic evidence for a correlation between cyclamate intake and bladder cancer in man. Saccharin is another sweetener that has been used for decades. Several retrospective

epidemiologic studies of users such as diabetics have not revealed a cancer risk, though one study has claimed an association between saccharin use and bladder cancer. Most of bioassays of saccharin in which sizable amounts were fed to mice or rats in standard protocols failed to reveal any evidence for carcinogenicity.

However, in three studies saccharin was given to two generations of rats; the second generation of male but not female rats exhibited a low incidence of bladder cancer. Saccharin is not metabolized and exhibits no evidence of genotoxicity, although there are reported genotoxic impurities in trace amounts. Thus, the finding of bladder cancer under specialized conditions may be based on mechanisms other than carcinogenicity, which remain to be investigated.¹⁷⁵¹ Friedell's group has recently observed a promoting effect with saccharin and with tryptophan.

cell cycle and cancer genetics

Cancer: A Familiar Name, But What Does It Mean?

Cancer refers to the hyperproliferation of cells that have lost the ability to be controlled by normal cell signals. Cancer cells have the ability to proliferate independent of their environment and are capable of metastasizing, or colonizing other tissues in the body.

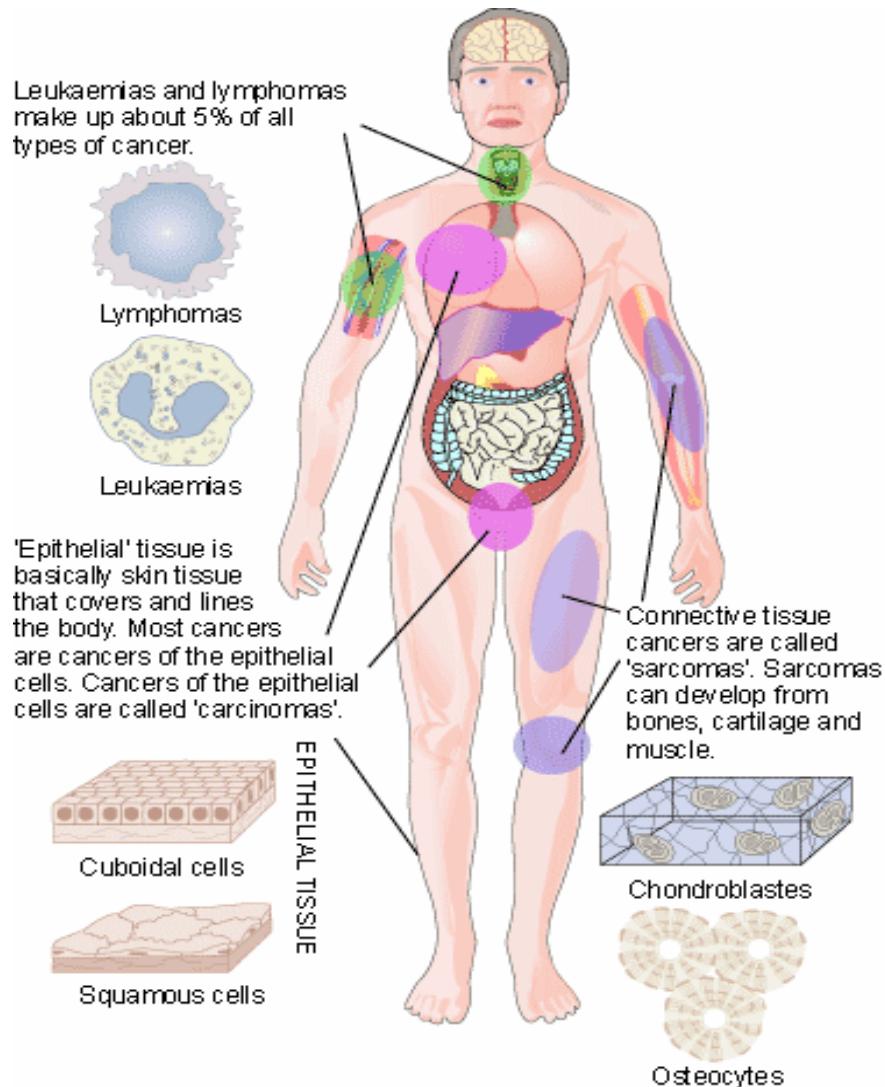


Figure 2.1: Different types of cancer are derived from different tissues within the body.

There are three basic characteristics of early cancer cells. The first is that they have lost the ability to undergo apoptosis, or programmed cell death. Cells that have suffered irreparable DNA damage activate specific proteases and nucleases that destroy the proteins and DNA of the cell, thereby effectively limiting the spread of potentially deleterious mutations. Cancer cells have often obtained mutations in genes involved in regulating this pathway. Secondly, cancer cells have lost the ability to stop dividing. Normal cells require extracellular signals, such as growth factors, in order to activate pro-growth pathways.

This need for extracellular stimulation is one means of regulating cell growth that cancer cells manage to bypass. This means that cancer cells proceed through the cell cycle and continue to divide indefinitely. Thirdly, cancer cells often have abnormal telomerase activity. Telomeres are the specialized regions on the ends of chromosomes that form t-loops - these protect the ends and distinguish double-stranded DNA breaks from normal ends.

Telomerase is an enzyme that acts to lengthen the telomeres on aging chromosomes in certain cell types. A portion of the telomere is lost during each DNA replication. When telomeres are shortened to the point that they cannot form t-loops, cells undergo senescence, where the cell remains in an undividing state, or apoptosis. Cancer cells, which are continually dividing, often express telomerase or have increased telomerase activity in order to circumvent the telomere problem.

These three basic characteristics are the premise for malignancy and enable the accumulation of more genetic and chromosomal abnormalities, which further lead to increased malignant and metastatic phenotypes. There are several different kinds of cancer but they all essentially fall into three basic categories: carcinomas, sarcomas and leukemias/lymphomas (see Figure 2.1). Both carcinomas and sarcomas are solid tumors, or tumors consisting of a dense collection of cancer cells that has its own blood supply. Carcinomas are solid malignant lesions originating in the epithelial tissue. These are often found in glands or the lining of various organs.

Examples of carcinomas would be breast cancer and prostate cancer, and occur most frequently in older patients. Sarcomas are solid tumors of the connective tissue, such as bone and muscle. There is a higher incidence of sarcoma in younger patients than in older ones. Finally, leukemia and lymphoma are malignancies originating in immune or haematopoietic cells and can be found in patients representing a wide age range. These cancers are commonly referred to as disseminated cancers as they do not form masses of cells, but instead are found throughout the vascular system.

How Does Cancer Arise?

Cancer arises most commonly in older patients but can occur at any age. Different cancers are more prevalent in certain age groups and are rarely found in people outside that age range. An example would be prostate cancer, where it is very uncommon for a man under the age of 40 to be diagnosed with the disease. It makes sense that cancer occurs predominantly in the elderly population, as cancer is the result of accumulated genetic mutations. As individuals age they are more likely to have been exposed to chemicals, radiation and other events that cause DNA damage.^[77] As a result, these individuals are more likely to have mutations in genes allowing for cancerous behaviors.

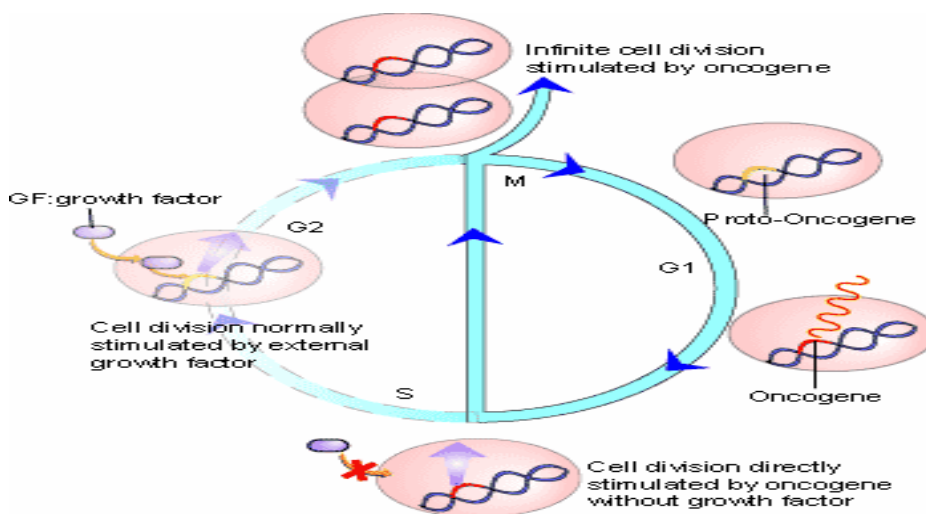


Figure 2.2: Oncogenes are mutated forms of normal cellular genes involved in growth signaling pathways (proto-oncogenes).

When these genes become mutated the cell does not require the presence of pro-growth signals (e.g. growth factors) in order to undergo cell division.

As mentioned above, characteristics like the ability to evade apoptosis; unregulated cell division and increased telomerase activity are all the results of genetic mutations that can lead to cancer. These mutations commonly occur in what are called proto-oncogenes and tumor suppressor genes. Proto-oncogenes are genes coding for proteins involved in cell cycle progression and growth signaling; when mutated these genes are called oncogenes (see Figure 2.2).

Oncogenes are autosomal dominant, meaning that only one mutated allele is necessary for cancerous behavior to develop. Several oncogenes have been identified in many types of cancers, including *myc*; the *bcl* family of genes and *ras* [76]. In contrast, tumor suppressor genes are often involved in regulating apoptosis (see Figure 2.3). Cells that have sustained damage to their DNA apoptosis under the control of tumor suppressors; when both alleles are mutated to be non-functional cells fail to enter the apoptotic pathway. These cells continue to divide and are not subjected to further DNA repair attempts, resulting in increasing genomic instability and further mutations. These mutations often occur in other oncogenes and tumor suppressor genes, conferring more cancerous behaviors. If either of these kinds of mutations exists or occurs in someone then they are at a higher risk of developing cancer.

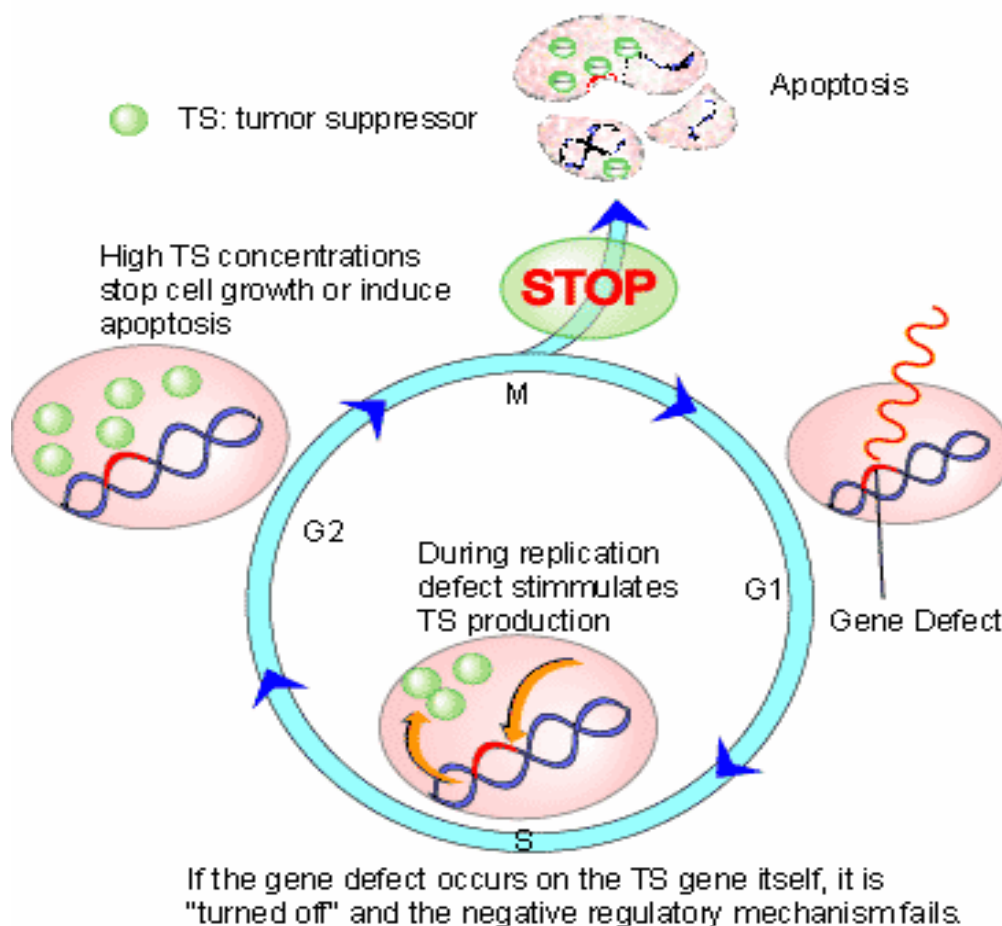


Figure 2.3: Tumor suppressor genes are genes often involved in the apoptotic pathway.

Normally tumor suppressors detect breaks or defects in the DNA - if present in low concentrations these proteins will pause the cell cycle and activate DNA repair mechanisms. If present in high concentrations, tumor suppressors shut down the cell cycle or cause apoptosis. When these genes are mutated to be dysfunctional then the cell does not undergo either of these events.

It is important to note that cancer results from the clonal expansion of one progenitor cell or "mother" cell the one that first obtained a mutation allowing for a "cancerous" phenotype. This means that all cells in a tumor should in fact be genetically identical to the parent cell and exhibit the same mutations, however, because mutations occur at a much higher rate in tumor cells, daughter cells may be more tumorigenic than the parent^[76]. All that is required for cancer to develop is one cell among millions that is capable of developing independent of regulatory signals.

The Cell Cycle

The field of developmental genetics investigates the genetic basis of the changes in form that an organism passes through during its life cycle. One cellular process that is common throughout these changes in form is cell division. The two cell division events that need to be controlled are the entry into the S-phase when DNA is replicated, and the entry into the M-phase when mitosis occurs. In this regard, two timing events need to be monitored by the cell. These are:

1. when to initiate replication (S-phase entry)
2. when to begin chromosomal condensations (M-phase entry)

Related to these events are four factors that appear to control the entry into the M-phase.

- 1) The accumulation of a specific cellular mass is a factor for somatic cells. This is called the mass factor.
- 2) Some cells need to obtain a specific growth rate for mitosis to begin. This is called the growth rate factor.
- 3) The time between successive M-phases appears to be controlled by timer or oscillator genes. This is the time factor and appears to be a factor in embryo cells.
- 4) The entry into the M-phase also requires completion of the S-phase. This insures that daughter cells receive complete DNA complements and is called the completion of chromosomal replication factor.

For the cell to coordinate these different events, it must be able to monitor the cell cycle. An important biological question that needs to be resolved is how the cell knows where it is in the cell cycle. As you would expect, genetics and biochemical characterization have provided an extensive, but incomplete description of the process. Cell cycle research has primarily been performed on mutant strains of the fission yeast (*Schizosaccharomyces pombe*) and the budding yeast (*Saccharomyces cerevisiae*) that have genetic lesions in some phase of the cell cycle.

The cell division cycle (cdc) mutant strains have been quite useful in elucidating important steps. The cell cycle in yeast has two points where it is committed to proceed to the next stage in the cycle. The first point called start occurs near the end of the G₁, and the cell becomes committed to DNA synthesis in the S phase of the cycle. The second commitment point is at the beginning of the M phase when the cell becomes committed to chromosomal condensation and the subsequent mitotic steps.

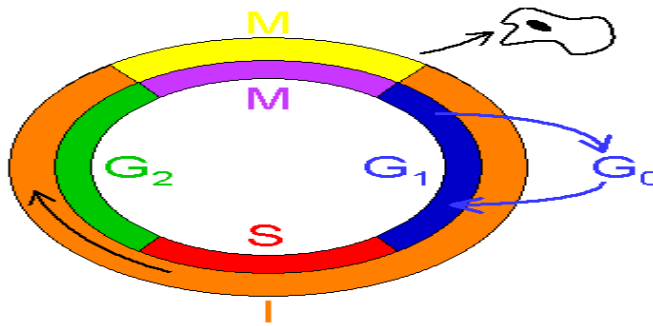


Figure 2.4: phases of the cell cycle

Schematic of the cell cycle. I=Interphase, M=Mitosis. The duration of mitosis in relation to the other phases has been exaggerated in this diagram.(figure 2.4)

The phases of the cell cycle are:

*The G₀ phases is a period in the cell cycle where cells exist in a quiescent state. Interphase- usually 90% of the cycle and can divide into subphases:

*The G₁ phase is the first growth phase when salvage enzymes are synthesized.

*S phase, during which the DNA is replicated, where S stands for the Synthesis of DNA.

*G₂ phase is the second growth phase, also the preparation phase for the cell.

*M phase or mitosis and cytokinesis, the actual division of the cell into two daughter cells.

A surveillance system, so-called "checkpoints", monitor the cell for DNA damage and failure to perform critical processes. Checkpoints can block progression through the phases of the cell cycle if certain conditions are not met. For instance, there is a checkpoint which monitors DNA replication and keeps cells from proceeding to mitosis before DNA replication is completed. Similarly, the spindle checkpoint blocks the transition from metaphase to anaphase within mitosis if not all chromosomes are attached to the mitotic spindle.

If this system senses a problem, a network of signaling molecules instructs the cell to stop dividing. They can let the cell know whether to repair the damage or initiate programmed cell death, a form of which is called apoptosis. Programmed cell death ensures that the damaged cell is not further propagated. For example, a certain protein, called p53, acts to accept signals provoked by DNA damage. It responds by stimulating the production of inhibitory proteins that then halt the DNA replication process. Without proper p53 function, DNA damage can accumulate unchecked. A direct consequence is that the damaged gene progresses into a cancerous state.

Today, defects in p53 are associated with a variety of cancers, including some breast and colon cancers. Some cells, such as neurons, never divide once they become locked in a G₀ phase. However, recent data has shown that neurons undergoing cell death re-enter the cell cycle. addition of cell cycle inhibitors prevent this apoptosis. The following diagrams illustrate the genetic and biochemical information known about the entry into the M-phase of the cell cycle. As you can see from the biochemical diagram a protein complex is formed at the two committal points. Each complex consists of a protein called cyclin, and a protein kinase called p34. When a factor called maturation promoting factor (MPF) was isolated that could initiate mitosis in certain mutant yeast strains whose cell cycle was arrested at this stage. It was the coupling of this type of biochemical research with genetics that defined and elucidated many of the steps in the cycle the

existence of such a complex was described biochemically. To demonstrate the molecular events associated with the cell cycle in more detail, a discussion that links the genetic research with the biochemical research will be presented. In particular, the discussion will concentrate only on the entry into the M-phase. The following table 2.1 represents the mutants, the product of these genes and their role in M-phase entry.

Table 2.1: represents the mutants, the product of these genes and their role in M-phase entry.

Gene	Protein	Function
<i>cdc13</i>	Cyclin	a 45,000-47,000 Dalton protein that complexes with the protein kinase p34 ^{<i>cdc2</i>} to form the MPF; its sequence is 30% conserved over a 200 amino acid stretch in a wide range of species; M-phase entry can be stimulated by adding this protein from clams to frog cells; its degradation appears to be associated with the inactivation of p34 ^{<i>cdc2</i>}
<i>cdc2</i>	p34 ^{<i>cdc2</i>}	a serine-threonine protein kinase of 34,000 Daltons that complexes with cyclin to form the MPF; the inactive form of the protein is phosphorylated at threonine (T) and tyrosine (Y) residues; the phosphorylation appears to be performed by p60 ^{<i>src</i>} in humans; the active form of the protein is dephosphorylated and it functions by phosphorylating a number of proteins; this phosphorylation activity is coupled to the entry into the M-phase; the protein must be associated with a normal cyclin protein for the M-phase to be completed normally; association with deletion mutants of cyclin halts the M-phase before it is completed
<i>cdc25</i>	p80 ^{<i>cdc25</i>}	a protein of 80,000 Daltons that assists with the dephosphorylation of p34 ^{<i>cdc2</i>} by either inhibiting its phosphorylation or promoting its dephosphorylation; its concentration increases as the cell approaches the M-phase suggesting the accumulation of this protein to a specific concentration is required to activate p34 ^{<i>cdc2</i>} ; its increase in concentration appears to be coupled with the completion of the S-phase
<i>suc1</i>	p13 ^{<i>suc1</i>}	a protein of 13,000 Daltons which may be involved in the inactivation of p34 ^{<i>cdc2</i>} late in mitosis by inhibiting its kinase activity or promoting its phosphorylation

It is clear from the genetic and biochemical studies that the appearance of an active MPF occurs at the M-phase committal point. The following cellular events have been associated with the onset of the protein kinase activity of the *cdc2* product

1. chromosomal condensation
2. cytoskeletal reorganization
3. nuclear envelope breakdown
4. cell shape changes

Each of these events is clearly required for cell division to occur. Furthermore, the substrates of the p34^{cdc2} protein kinase are proteins involved in the maintenance of the cell in the G₂-phase. The phosphorylation of these proteins may change their functions and permit the cell to enter the M-phase.

The key substrates of p34^{cdc2} protein kinase are:

1. Histone H1 - the phosphorylation of this protein may be important for chromosomal condensation to occur
2. Centrosomal protein - these proteins are associated with centrioles, the organizing center of the cell for microtubules associated with the cytoskeleton
3. Lamin - this is a protein associated with the nuclear envelope
4. p60^{src} - phosphorylation of the mitotic-specific sites of this protein may influence the cytoskeleton and lead to changes in the cell shape
5. other DNA binding proteins that need to be released for chromosomal condensation to occur
6. The studies on yeast and other organisms have led to the conclusion that a universal control mechanism regulating entry into the M-phase is common to all eukaryotic cells. The key features of the process are as follows the protein kinase activity of p34^{cdc2} is central to the model. This protein is thought to phosphorylate key proteins that lead to the major events in the M-phase. High levels of this protein maintain the cell in the M-phase, and its inactivation is required for exit from the phase.
7. The second key protein is cyclin those complexes with p34^{cdc2} to form the MPF. Cyclin is required for p34^{cdc2} activation. Cyclin degradation is required for the cell to exit the M-phase and probably the inactivation of p34^{cdc2}.
8. The activation of p34^{cdc2} is associated with the dephosphorylation of the phosphorylated tyrosine and threonine residues of the protein. Its kinase activity appears to be associated with the tyrosine residue, so dephosphorylating this site appears essential.
9. Timing of the M-phase entry is associated with two other protein kinases and the accumulation of p80^{cdc25}. This timing event is associated with the dephosphorylation of p34^{cdc2}.
10. p 13^{suc1} interacts with p34^{cdc2} and may be involved in its rephosphorylation at the end of the M-phase.

Oncogenes and cancer

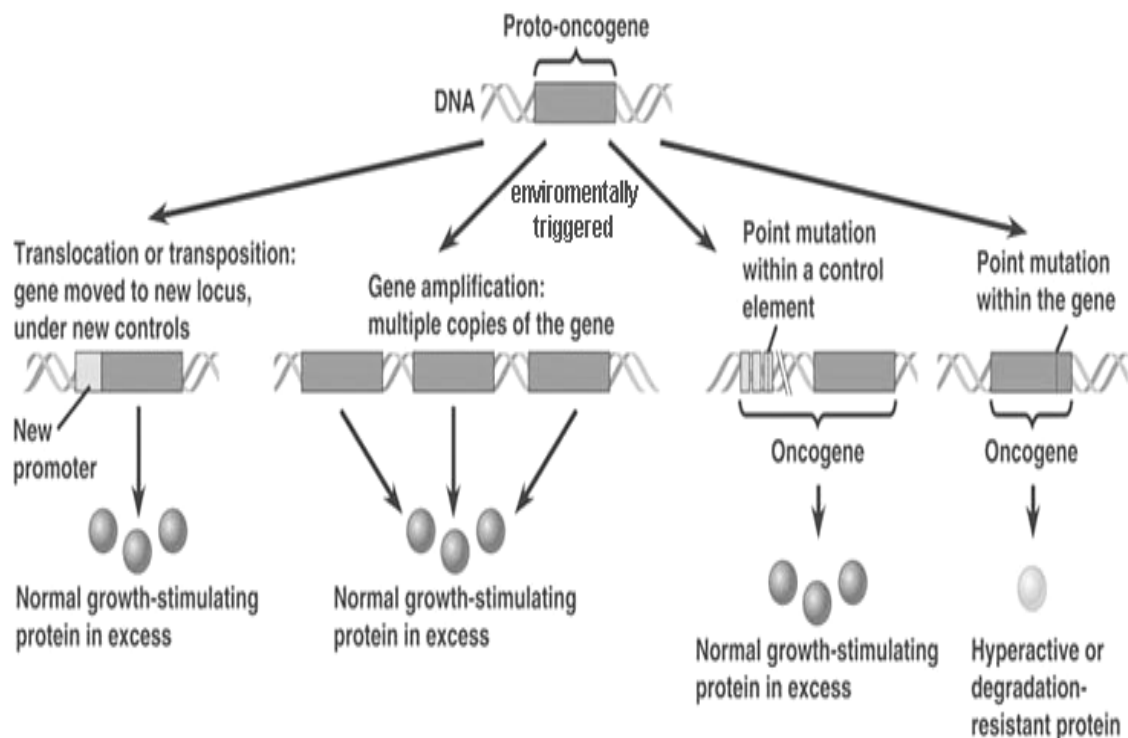
Animals are multi-cellular organisms that require the normal function of all the organs of the body. These organs are developed from different tissues and each of tissues is products of cell division. For the body to function normally, the organs and tissues must communicate to control the development of the cells and tissues. Otherwise, uncontrolled cell growth in one part of the body could infringe on the development of other cells or tissues. Then the normal functions of the individual would be seriously impaired. Research over many years has shown that these control networks have a strong genetic component.

This research has recently benefited from the study of the oncogenes of retroviruses and related protooncogenes that are found in animals. Oncogenes have been shown many times to be associated with cancer and uncontrolled cellular growth. This growth can lead to tumors. Two types of tumors exist. Malignant tumors can induce secondary tumors by the release of cells that can lodge and begin growing in another location of the body. Benign tumors are cells that remain in the initial location. Hybridization of retroviral oncogenes probes to animal DNAs, including humans, demonstrated that copies of many of these genes are present in the genomes of these animals. These are not integrated proviruses, but are complete animal genes which contain their own controlling sequences.

The normal cellular counterparts of these oncogenes are called protooncogenes. In their normal genetic state, protooncogenes are important components of cellular signaling and transcription activation. If a protooncogene undergoes a somatic mutation (figure 2.5.), control of cell growth is lost in the cell in which the mutation occurs and cancer can occur. Those protooncogenes which have been shown to mutate in any individual are called cellular oncogenes and are designated by the prefix "c" (i.e. *c-myc*, *c-abl*) to distinguish them from the viral oncogenes. Those protooncogenes that have not been found to mutate are called normal oncogenes and are designated "n" (i.e. *n-ras*).

Research on retroviral oncogenes was being pursued in parallel with the search by cell biologists for genes which control cell signaling and transcriptional control. At that time, in the early 1980s, it was not widely suspected that these researchers were searching for genes with similar function. The advent of DNA sequencing and the establishment of large DNA sequence databases eventually led to the merger of the two fields. The diagram below shows how the two fields took different routes to realizing that the cellular gene for the epidermal growth factor receptor (EGFR) and the *v-erbB* oncogene encoded the same gene product.

A final point to consider is the evolutionary origin of retroviral oncogenes. Did oncogenic retroviruses exist first and were their oncogenes deposited into the genome of a primitive animal, or did a retrovirus obtain a copy of the gene from a primitive animal. The latter appears to be the case because all the protooncogenes have introns that are not found in the viral oncogenes. The suggested mechanism is that RNA copies of the protooncogenes (lacking the introns) were ligated into a retrovirus genome (also RNA) and the oncogene was then perpetuated upon subsequent replication of the retrovirus. This would stabilize the new gene in the virus.



origins of oncogenes may have been viral elements:

cancer causing viri include:

Epstein-Barr - herpes virus - infectious mononucleosis - Burkett's Lymphoma

Papilloma virus - cancer of the cervix

HTLV-1 - retrovirus - adult leukemia

Figure 2.5: different proto-oncogene mutations

How Oncogenes Cause Cancer?

The change of an oncogene from normal to cancerous function can be caused by a simple point mutation in the sequence of a gene. For example, a change in the ras oncogene, located on human chromosome 11, from guanine to cytosine is frequently associated with bladder cancer. This simple change results in glycine at amino acid #12 being substituted with a valine. This dramatically changes the function of the G-protein encoded by the ras gene. Normally, the protein cycles from an inactive to active state by change the bound guanosine diphosphate (GDP) to guanosine triphosphate (GTP).

The mutation does not allow the release of GTP, and the protein is continuously active. Because the signal delivered by the ras oncoprotein is continuously delivered, the cell continues to grow and divide. This unabated growth leads to the bladder cancer. Deletions of the ligand binding domain of the EGFR oncogene, located on human chromosome 7, results in continuous signal transduction by the epidermal growth factor receptor it encodes. The deletion protein can form a dimer even in the absence of the epidermal growth factor. Dimerization leads to continuous tyrosine kinase activity and uncontrolled activation of the signal transduction pathway associated with this gene.

The following table summarizes the types of molecular changes that can be associated with the activation of an oncogene.

Table 2.2: types of molecular changes that can be associated with the activation of an oncogene.

Molecular Change	Effect On Oncogene
Translocation	Philadelphia chromosome contains a <i>bcr1/abl</i> fusion that activates the <i>abl</i> protein kinase activity; Burkitt lymphoma result from the placement of the <i>c-myc</i> next to an enhancer in B lymphocytes
Point Mutations	See <i>ras</i> discussion above
Deletions	See <i>v-erbB</i> discussion above
Insertional activation	A retrovirus without an oncogene may insert near a protooncogene and activate the protooncogene by increasing its expression 30-100 fold. This activation is not immediate, but can take several months. The LTRs of retroviruses contain powerful promoters and enhancer sequences that presumably are responsible for the increased expression. The insertion can occur on either side of the protooncogene or in its intron. This type of activation has been associated with <i>c-myc</i> and <i>c-myb</i> .
Amplification	Amplification of the protooncogene has also been associated with the onset of cancer. This has been best studied in cell culture

Tumor suppressor genes and cancer

Although the activation of protooncogenes can be accomplished by a number of different molecular mechanisms, it should be pointed out that onset of cancer can be controlled genetically at a higher level. Tumor suppressor genes have been detected in the human genome that prevents the onset of cancer even if one of the above molecular events does occur. These genes have been very difficult to isolate and analyze, but recent evidence suggests several functions.

Retinoblastoma

Is a juvenile eye cancer that is caused by a mutation in the *Rb* gene located on chromosome 13 of humans. This gene suppresses the development of cancer as its dominant phenotype. Therefore both alleles must be mutant for the disease to develop. The *Rb* gene product interacts with a protein called E2F, nuclear transcription factor involved in cellular replication functions during the S phase of the cell cycle. By interacting with E2F it prevents it this function. The *Rb* gene product is only active when it is not phosphorylated by a kinase. It can not interact with E2F when it is phosphorylated. The mutant *Rb* gene product is always phosphorylated and can

not regulate E2F, control of cell division at the S phase does not occur, and normal cells become cancerous.

p53

P53 is a nuclear phospho-protein which, in response to DNA damage, slows progression through the cell cycle and initiates apoptosis if damage is severe.^[78] The tumor suppressor gene TS P53 is altered in the majority of cancers. The p53 protein causes cell-cycle arrest and apoptosis. It acts mainly through p21 to cause cell cycle arrest. It causes apoptosis by inducing the transcription of pro-apoptotic genes such as BAX. Levels of p53 are negatively regulated by MDM2 through a feedback loop. P53 is required for the G1/S check point and is a main component of the G2/M check point^[81].

Structure

The core domain structure consists of a beta-sandwich that serves as a scaffold for two large loops and a loop-sheet-helix motif.^[80] The two loops, which are held together in part by a tetrahedrally coordinated zinc atom, and the loop-sheet-helix motif form the DNA binding surface of p53. Residues from the loop-sheet-helix motif interact in the major groove of the DNA, while an arginine from one of the two large loops interacts in the minor groove. The loops and the loop-sheet-helix motif consist of the conserved regions of the core domain and contain the majority of the p53 mutations identified in tumors.

P53 and DNA damage

The tumor-suppressor protein p53 has an important role in determining the fate of cells following exposure to DNA damage and other types of cellular stresses. Following these insults, p53 accumulates in the nucleus of cells, where it can act as a transcription factor. Depending on the cell type and the level of damage induced, p53 can increase the survival of exposed cells by Trans activating genes encoding DNA-repair enzymes and cell-cycle inhibitors, or induce cells to undergo apoptosis. In either case, both mutagenesis and, subsequently, carcinogenesis are suppressed.^[79] Using human cell lines with specific defects in NER, it has been shown that the triggering mechanism for p53 accumulation and apoptosis following UV-light irradiation is dependent on DNA damage. Furthermore, the triggering of p53 and apoptosis is not dependent on DNA-repair-induced DNA strand breaks, but rather on persistent lesions specifically in the transcribed strand of active genes. Studies using human fibroblasts treated with 2-acetylaminofluorene, the polycyclic aromatic hydrocarbon DMBA or cisplatin, which - like UV light - induce bulky DNA lesions, have shown that p53 accumulation and apoptosis are induced at much lower doses in cells that are deficient in the removal of transcription-blocking lesions compared with proficient cells.^[79] The finding that persistent lesions in the transcribed strand of active genes trigger p53 and apoptosis has been confirmed using mouse models with specific genetic defects in GGR or TCR.

cancer detection

Understanding Prognosis and Cancer Statistics

Key points

- Prognosis gives an idea of the likely courses and out come of a disease
- Many factors affect a person's prognosis, including the type, location and the stage of the disease, as well as person's age, general health and response to treatment.
- Survival rates indicate the percentage of people with a certain type and stage of cancer who survive the disease for a specific period of time after their diagnosis, survival rates are based on large group of people.
- Doctors can not absolutely certain about the out come for a particular patient in fact, a person's prognosis may change over time^[82].

People facing cancer are naturally concerned about what the future hold, understanding cancer and what to expect can help patients and their loved ones plan treatment think, about life style changes, and make decisions about their quality of life and finances many people with cancer want to know their prognosis. They may ask their doctor or research for statistics on their own .A prognosis give an idea of the likely course and out come of a disease- that is; the chance that a patient will recover or have a recurrence (return to the cancer) .Many factors affect a person's prognosis. Some of the most important are the type and location of the cancer, the stage of the disease (the extent to which the cancer has metastasized, or spread), or its grade (how abnormal the cancer cells look and how quickly the cancer is likely to grow and spread).

Other factors that , may also affect the prognosis include the person's age , general health , and response to treatment .When doctors discuss a person's prognosis , they carefully consider all factors that could affect that person's disease and treatment , and then try to predict what might happen . The doctors bases the prognosis on information researchers have collected over many years about hundreds or ever thousands people with cancer .When possible the doctor uses statistics based an groups of people whose situations are most similar to that of an individual patient .

The doctor may speak of favorable prognosis if the cancer is likely to respond well to treatment, the prognosis may be unfavorable if the cancer is likely to difficult to control, it is important to keep in mind, however, that a prognosis only a prediction.

The doctor can not be absolutely certain about the out come for a particular patient. Survival rates indicate the percentage of people with a certain type and stage cancer who survive the disease for a specific period of time after their diagnosis .Often, statistics refer to the 5- years survival rates, which means the percentage of people who are alive 5 year after diagnosis, whether they have few or no signs or symptoms of cancer, are free of disease or having treatment. Survival rates are based on large groups of people .they can not be used to predict what will happen to a particular patient. No two patients are exactly alike and treatment and responses to treatment vary greatly.

Cancer patients and their loved ones face many unknowns. Some people find it easier to cope when they know the statistics. Other people find statistical information confusing and frightening, and they think it is too impersonal to be of use to them. The doctor who is most familiar with a patient's situation is in the best position to discuss the prognosis and to explain what the statistics may mean for that person, at the same time it is important to understand that even the doctor can not tell exactly what to expect. In fact a person's prognosis may change if the cancer progresses or if treatment is successful. Seeking information about the prognosis is a

personal decision it is up to each patient to decide how much information he or she wants and how to deal with it.

Cancer Diagnosis

The diagnosis of cancer entails an attempt to accurately identify the anatomical site of origin of the malignancy and the type of cells involved. Cancer can arise in any organ or tissue in the body except fingernails, hair, and teeth. The site refers to the location of the cancer within the body. The body part in which cancer first develops is known as the primary site. A cancer's primary site may determine how the tumor will behave; whether and where it may spread (metastasize) and what symptoms it is most likely to cause. The most common sites in which cancer develops include the skin, lungs, female breasts, and prostate, colon and rectum, and corpus uteri. Secondary site refers to the body part where metastasized cancer cells grow and form secondary tumors. A cancer is always described in terms of the primary site, even if it has spread to another part of the body. For instance, advanced breast cancer that has spread to the lymph nodes under the arm and to the bone and lungs is always considered breast cancer (and the spread to the lymph nodes, bones, and lungs describe the stage of the cancer).

As is the case with other medical conditions, there are many signs and symptoms that may indicate the presence of cancer. These may be observed directly, through imaging technologies, or confirmed by lab tests. However, these signs and symptoms of cancer may resemble those of other conditions. For example, weight loss and abdominal pain can be caused by stomach cancer or an ulcer. Pink or reddish urine can be caused by kidney cancer or a kidney infection. A positive fecal occult blood test can indicate a variety of intestinal problems. A biopsy (removal of tissue for microscopic evaluation) is preferred to establish, or rule out, a diagnosis of cancer. Tissue samples can be easily retrieved from a tumor near the body's surface. If the mass is inaccessible, an imaging exam that enables a tumor to be located precisely and visualized may be ordered before the biopsy is performed.

The histological type is determined by microscopic examination of suspected tissue that has been excised by biopsy or surgical resection. If the histological type is different from what is usually found in the tissue being examined, it can mean the cancer has spread to that area from some primary site. Metastasis can occur by direct extension, via the blood stream or the lymphatic system, or by seeding or implantation of cancer cells. A biopsy, together with advanced imaging technologies, may not only confirm the presence of cancer, but may also pinpoint the primary site and secondary site(s). It is also important to identify the cell type(s). Various histological types have different growth rates and dissimilar prognoses.

More than one histological type of cell may be found in the same site. For example, a tumor whose primary site is skin can be a basal cell carcinoma, a squamous cell carcinoma, or a melanoma. Once cancer has been confirmed, the pathologist tries to determine how closely the cancer cells resemble healthy, mature cells. Such cells are said to be differentiated. Cancer cells that do not look like their healthy counterparts are called undifferentiated, or because they often look like very immature cells primitive. The pathologist assigns a pathological grade to a tumor according to how aggressive the tissue looks under the microscope. Tumor grades can be expressed in words or by a number^[83]. One set of terms consists of well differentiated (grade 1), moderately differentiated (grade 2), poorly differentiated (grade 3), or undifferentiated (grade 4). When tumors are graded by number (1 through 4), a grade-1 tumor has a better natural history than a grade-4 tumor does.

Cancers are further classified according to stage. Staging describes how far a cancer has progressed based on the size of the primary tumor and whether and/or where it has spread. A biopsy is the preferred method to confirm the diagnosis of cancer. Biopsies can provide

information about histological type, classification, grade, potential aggressiveness and other information that may help determine the best treatment.

Common Concerns for Staging

Some common concerns about staging are the following:

The stage of a cancer is sometimes confused with the grade of a tumor by new registrars. Terms such as well differentiated and undifferentiated are tumor grades.

The rules for each staging scheme must be reviewed for each site and histology. The AJCC staging systems and summary staging systems list the sites and histologies to which specific staging schemes apply. The term microinvasion implies invasion through the basement membrane (an anatomic landmark), indicating that the stage is invasive instead of in-situ.

Some cases of cancer are difficult to stage appropriately.

Problem situations include the following:

- 1) Diagnostic tests done on an outpatient basis with results not documented in the hospital health information record
- 2) Tests and biopsies done in a physician's office and sent to freestanding laboratories for assessment

Conflicting information about the exact location, size, and involvement of the tumor

There are many resources available for staging cancers. The registry should have adequate access to appropriate references. Staging manuals for the most commonly used systems (summary staging, AJCC staging, and SEER extent of disease) provide comprehensive guidelines. These should be routinely reviewed at the time of abstracting to verify the staging classification.

The Commission on Cancer of the American College of Surgeons requires that staging be done by the managing physician and recorded in the patient's health information record. This requirement does not negate the need for the registrar to understand staging. Verification is necessary at the registry level to ensure the accuracy and completeness of data. It is imperative that staging be correct, when registry data are reported and analyzed. The patient's treatment is based on the extent of the disease. The prognosis of the disease can be estimated by the stage and other factors such as age, aggressiveness of tumor, and the presence or absence of other medical conditions ^[84]. In certain stages of disease, quality of life issues may influence treatment decisions. The stage of disease is used in research studies and in the analysis of cancers

Physical Examination

For most cancers, the report of the physical examination should include the location of tumor, including site and sub site, direct extension of the tumor to other organs or structures, and palpability and mobility of accessible lymph nodes. The probability of distant site involvement, such as organomegaly, pleural effusion, ascites, or neurological findings should be stated. In a breast cancer case, for example, the physical examination should describe the exact location of the tumor mass, clinical size of the tumor, and the condition of the skin surrounding the tumor, including changes in skin color and texture and attachment or fixation of the mass. The exam should include the entire axial and regional nodal area including the supraclavicular nodes for most cancers, the report of the physical examination should include the location of tumor, including site and sub site, direct extension of the tumor to other organs or structures, and

palpability and mobility of accessible lymph nodes. The probability of distant site involvement, such as organomegaly, pleural effusion, ascites, or neurological findings should be stated. In a breast cancer case, for example, the physical examination should describe the exact location of the tumor mass, clinical size of the tumor, and the condition of the skin surrounding the tumor, including changes in skin color and texture and attachment or fixation of the mass. The exam should include the entire axial and regional nodal area including the supraclavicular nodes. Some organ sites are not easily examined clinically.

A patient suspected of having a gastrointestinal tumor should have external palpation of the liver and abdomen. Females should have both a digital rectal exam and a pelvic exam. Males should have a digital rectal exam. Suspected lung cancer patients should have an assessment of cervical and supraclavicular nodal areas. In all cases, other than lymphomas, nodes must be described by a clinician as "involved" in order to be considered to contain cancer. For example, if it is stated that the nodes are enlarged, they are not considered to contain cancer until there is cytological or pathologic confirmation. If there is matting or fixation, the medical practitioner may state that the nodes are involved with cancer.

Pathologic Examination

The most common diagnosis and accurate methods of diagnosing cancer include microscopic examination of either tissue or cells. Cells examined are usually obtained from fluid around the suspected site of cancer. Tissues examined are usually removed from the primary or metastatic site of a cancer.

There are many kinds of biopsies to remove tissue for a cancer diagnosis. An aspiration biopsy is obtained by using a needle to suction fluid, cells, or tissue into a syringe. A bone marrow biopsy is the removal of bone marrow from one of the body's larger hollow bones such as the femur or pelvic bone. Excisional biopsies attempt to remove the entire tumor. Incisional biopsies remove only a portion of the tissue.

Often, the biopsy specimens are quickly frozen, thinly sliced, and examined to determine the presence or absence of cancer cells (frozen sections). Permanent sections are then made, and the diagnosis from the permanent sections should take precedence over frozen section reports. Surgical resections involve removing more tissue from the body, including margins of normal tissue and/or regional lymph nodes. The pathologist can often determine staging by examining the primary tumor, surrounding tissues, and regional nodes when there is a "total" resection of the tumor. The information from a total resection takes precedence over biopsy reports and operative notes.

Quite often, there are several tissues samples, biopsies, or surgical resections for one cancer. When staging a cancer, it is important to review all pathological reports for the clinical diagnosis, gross description of the specimen and postoperative diagnosis.

The gross description of the specimen should include the total size of the tumor. Both the gross and the microscopic descriptions should state whether the surgical margins are involved by tumor. The pathology report should contain information about the primary site and the spread of the disease in surrounding tissues. It is important to note all areas, organs, or structures involved with tumor.

The pathology report contains the histologic type of cancer and the grade of the tumor (how closely the cancer cells resemble normal tissue). Grade is normally expressed as Grade I through III or as well differentiated, moderately differentiated, and poorly differentiated, respectively. Tumors can also be described as anaplastic or undifferentiated (Grade IV) ^[83].

The final diagnosis of the histological type takes precedence over preliminary reports and frozen sections. The microscopic description takes precedence over the gross description. Occasionally, pathological specimens are sent to other centers for consultation, and the final pathology report may not be signed until all consultations have been returned.

The most important information in a pathology report includes source of the specimen, primary site, tumor size, histologic type of cancer, grade of tumor, and the extent of the disease within the organ of origin and beyond. The type, size, location and number of lymph nodes removed, and number of nodes containing tumor should be noted. This information is often required for accurate staging.

Pieces of chips of tumor should not be added together to determine tumor size. If the patient has received preoperative radiation therapy, the size of the tumor should be recorded as found in radiology reports prior to radiation therapy. Multifocal and multicentric are synonymous terms. The size of the largest of the multifocal tumors should be used for staging.

Autopsy reports are a type of pathology report that contains detailed information about organs and structures of the body. An autopsy is considered to be an ultimate pathology report. In summary, pathology reports, or reports of tissue, contain information about biopsies, frozen sections, tissue aspiration or biopsy of bone marrow, surgical specimens and autopsies.

Cytology reports describe the microscopic examination of cells in body fluids such as sputum, bronchial washings and brushings, pleural fluid, peritoneal fluid, spinal fluid, aspirations from bone marrow, and cervical smears. The Papanicolaou (Pap) smear, used for detection of abnormal cervical cells, is probably the most widely known cytology specimen. Cells can also be obtained by fine-needle aspiration to diagnose cancers of the liver, pancreas, breast, and lung. The most common ways of obtaining cells include brushing the lining of an organ, puncturing the cavity and removing fluid, scraping the lining, or using a swab to obtain secretions.

Thoracentesis is a puncturing of the thoracic, or chest cavity for the removal of fluid. Paracentesis is the puncture of the abdominal cavity for removal of fluid.

There may be multiple cytology reports. It is important to note the source of the specimen, the histologic description, and pertinent findings, along with interpretations

Tumor Markers

Definition of Tumor Marker

- * Substance Produced by a tumor or a host response to a tumor that can differentiate tumor from normal tissue or detect the presence of a tumor with blood testing.
- * Tumor markers are gene Products often Produced in low levels by normal tissues and circulate normally in blood in small quantities.
- * Elevations in blood of tumor markers are usually derived from tissues normally producing that marker or from embryogenically closely related tissues (i.e. CEA from colon or stomach; liver; pancreas).

Ideal Characteristics of Tumor Markers

- 1.Organ specific and Tumor specific.
- 2.Positive only when malignancy is present.
- 3.Positive early in the development of malignancy.
4. Easy to measure in blood.

Clinical Use of Tumor Markers:

Most Tumor Markers are non-specific for a single cancer; they are found with different tumors of the same tissue type (tumor-associated markers).Most tumor markers should not be used as a "cancer screening tool". Most common use: clinical staging, monitoring therapy, detecting recurrence or presence of residual disease. Markers in theory, tumor markers might be useful as an aid tumor diagnosis, and prognosis as method of tumor burden, and as a means for predicting before it can be detected clinically.They might be useful as a guide in the choice and scheduling of treatment ^[85]. However markers are not produced uniquely by tumor their levels, in blood differ quantitatively rather than qualitatively from normal, also the production of markers by likely to be heterogeneous among the cells of the tumor, population. A classification of substances that have been used or proposed as tumor markers shown in Table 3.1Basic properties of the markers and their detection in blood or urine are described.

Properties of tumor markers

▣ Detection of markers

Tumor markers occur in low concentration in plasma and require sensitive techniques for their detection. Radioimmunoassays, immunoradiometric assays or enzyme -linked immunosorbent assays are variations on the same principle and are now the procedures , of choice for the measurement of small quantities of protein in body fluids (fig 3.1). All three procedures depend on the formation of antigen antibody complexes .

A purified preparation of a particular marker used initially to prepare specific antibodies (usually monoclonal antibodies) that recognize the marker. And these antibodies are usually attached to a solid support such as sephadex beads. In radioimmunossay, the quantitation of tumor markers in serum depends on competition for binding to the antibodies between the unknown sample and a known amount of purified and labeled marker. With immunoradiometric of enzyme-linked immunosorbent assays quantitation of an unknown an unknown sample is assayed by it's binding to specific antibodies on the solid support using specific antibodies that form a "sandwich" around the marker antigen (Fig3.1).

The specificity of the foregoing procedures depends on the purity of the markers used to prepare antibodies and the specificity of these antibodies for the particular marker antigen. No substance that is unique to tumors has been isolated to date. It follows, therefore, that anti bodies raised against these markers may cross-react with related molecules. Although highly sophisticated techniques have been developed for the measurement of small quantities of markers caution should be exercised in interpreting the results of these assays.

Table 3.1: Classification of Tumor Markers Showing Selected Examples

Oncofetal proteins	Carcinoembryonic antigen (CEA) Alpha-fetoprotein(AFP)
Hormones	Human chorionic gonadotropin(hcG) Ectopic hormones
Enzymes	Prostatic acid phosphatase Alkaline phosphatase Lactic dehydrogenase Gammaglutamyl transpeptidase(GGT) Neurone-specific enolase(NSE)
Immunoglobulins	
Tumor-associated antigens	CA 125 CA 15.3 Prostate specific antigen(PSA)
Miscellaneous	Polyamines Nucleosides Tissue polypeptide antigen(TPA) Acute-phase proteins

★ Oncofetal Proteins

The oncofetal protein, are normally present during a variable period of embryonic or fetal life do not disappear entirely in the adult and reappear with certain malignancies. They are substances that appear to originate in the tumor itself and enter the circulation as a result of secretion by the tumor or as breakdown products of tumor cells. Classic examples of the oncofetal proteins include carcinoembryonic antigen (CEA) and alpha-fetoprotein (AFP).

(1) Carcinoembryonic antigen

Is a glycoprotein with a molecular weight of about 200,000 Da and is found in the alimentary tract, liver, and pancreas of fetuses between the second and sixth month of intrauterine life. The CEA molecule is heterogeneous; recent data indicate that it is a member of the immunoglobulin supergene family (chapter14, Hammarstrom et al,1989; Barnett et a.,1990; Zimmerman and Thompson,1990). It can be detected in normal adult using sensitive immunohistochemical techniques and is present in small quantities in normal plasma.

Elevated blood levels of CEA are found in several nonmalignant diseases such as cirrhosis and chronic obstructive pulmonary disease, and modest elevations may occur in smokers. Reappearance of the protein or other oncofetal proteins in the presence of a variety of epithelial tumors (e.g., colorectal cancer, breast cancer) has led to the suggestion that "de-repression" may be a characteristic of malignant growth or, alternatively, that stem cell, or other primitive progenitor cells populate the tumor. With the introduction of gene sequencing and of monoclonal antibodies to study the protein, there is evidence to suggest that immunologically

distinct molecular forms exist. Whether they are characteristic of tumors of different organs remains to be determined. The half-life of CEA in plasma appears to be about 6-8 days. This estimate is based on the observation that elevated CEA level return to normal in about two months following successful removal of a CE-producing primary tumor.

(2) Alpha – fetoprotein

is an α -globulin and is a product of the fetal liver, gastrointestinal tract and yolk sac. the protein is normally present in the fetal circulation. there is good evidence to suggest that APF functions as the fetal counterpart of adult albumin because there is considerable homology between the two proteins. Again, the protein gradually disappears from plasma during neonatal life to be replaced by albumin, but never entirely disappears in the adult. Marked increases in plasma AFP concentration are observed in about 80 % of patients with hepatocellular carcinoma and about 60 % of patients with non seminomatous testicular tumors like CEA , AFP levels may be elevated in plasma in the presence of nonmalignant diseases , especially cirrhosis. AFP has a half-life in plasma of about 5.5days.a slower rate of disappearance of the protein, or failure of serum levels to return to normal following surgery, are strongly suggestive of the presence of residual disease (Abelev , 1989)

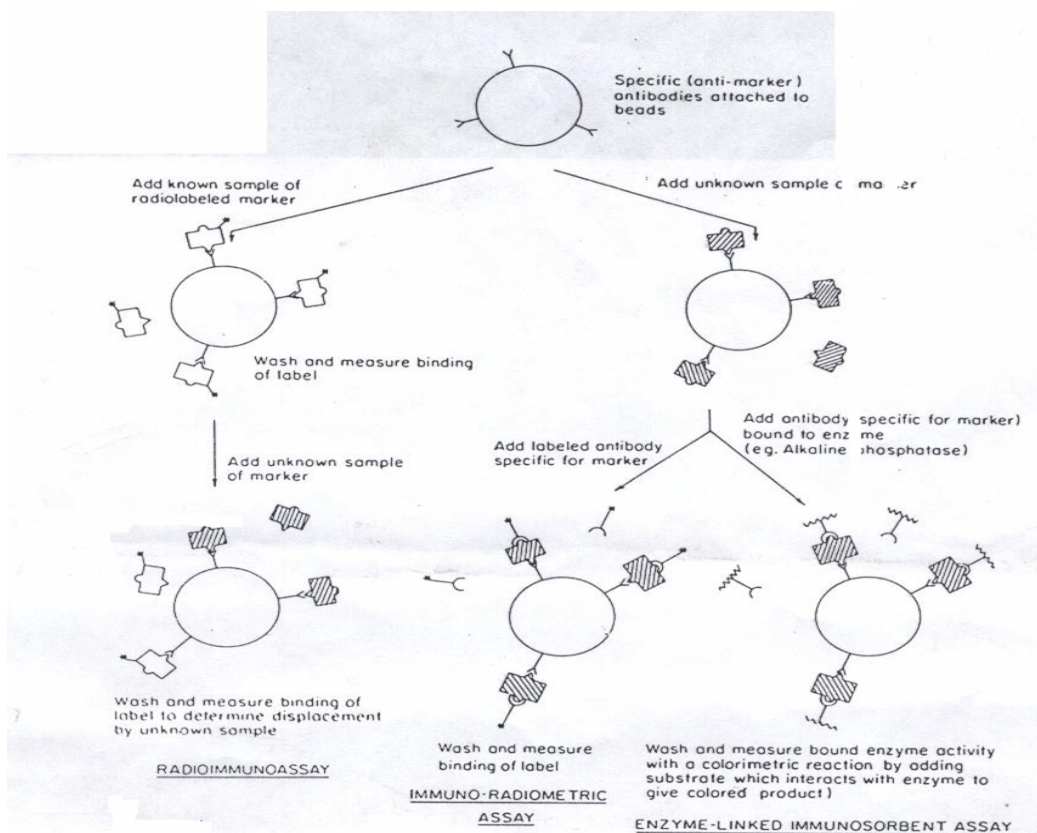


Figure 3.1 schematic diagram of radio or enzyme immunoassays several variations of these methods have been used to quantitate markers in body fluids [85].

★ HORMONES

1 -Human chorionic gonadotropin (HCG)

Is produced normally by the syncytiotrophoblast cells of the placenta during pregnancy and most pregnancy tests are based on its detection in serum or urine. Elevated levels of HCG are also found in the plasma of almost all women with tumors of placental origin (choriocarcinoma) and in 60 % of men with testicular tumors. The hormone is a glycoprotein related to follicle – stimulating hormone (FSH), luteinizing hormone (LH), and thyroid – stimulating hormone (TSH). It consists of two polypeptide chains, termed α and β . The α chain is homologous with the α chains of the other glycoprotein hormones; the B chains exhibit some differences. Thus use of HCG as a tumor marker depends on immunologic detection of the B chain. Modest elevation of HCG is sometimes observed in patients with tumors of other organs such as breast and large bowel. It is not clear whether this represents true ectopic hormone production, i.e., hormone production by cells that do not produce the substance normally, or rather is due to increased synthesis and secretion of a material produced normally in small quantities by the organ in question i.e., eutopic production

There is great variation in the carbohydrate side chain of HCG in patients with choriocarcinoma and testicular tumors, and molecular forms, or free α or B Chains, can often be identified in the plasma of these patients. The large molecular forms probably represent prohormones that are incompletely processed and that would normally undergo translational modifications prior to secretion. Human chorionic gonadotropin has a half-life in plasma of about 36-48 hours. This short half-life may be useful for early assessment of the efficacy of treatment.

2- Ectopic hormones

Can occasionally be secreted by tumors of nonendocrine organs and give rise to syndromes associated with overproduction of hormones. The fact that the hormones produced are almost invariably polypeptides suggests that the responsible mechanism involves derepression of a single gene. Steroid biosynthesis is much more complex, involving a cascade of enzymes and, unless genomically linked, their activation would entail an unlikely series of events. Small-cell lung cancer is the nonendocrine tumor most commonly associated with ectopic hormone production. Using immunochemical procedures, a great variety of hormones can be detected in the sera of these patients including ACTH, calcitonin, and arginine vasopressin.

In most patients, clinical sequelae are not observed because the synthesis of the hormone is disorganized. Prohormones and fragments of precursors appear in the patient's serum with far greater frequency than the active hormone itself. There is some argument as to whether the secretion of polypeptide hormones in patients with small-cell lung cancer is indeed ectopic in origin. Immunohistochemical techniques have identified cells in the pulmonary bronchioles of the normal lung, which stain for a variety of polypeptide hormones. The cells are often referred to as "pulmonary endocrine" (or APUD = amine precursor uptake and decarboxylation) cells, and it is now believed that small cell lung cancer originates from them, the secretion of ectopic hormones is not unique to tumors since their production also occurs in patients with chronic obstructive pulmonary disease, albeit less frequently and at lower levels.

★ Enzymes

1- prostatic acid phosphatase

Is an enzyme secreted by the normal prostate gland. It can be differentiated from other phosphatases by chemical and immunological means. In patients with prostate cancer, abnormal levels indicate that tumor has extended beyond the prostatic capsule. Synthesis and secretion of prostatic acid

phosphatase is dependent on the action of testosterone and maybe turned off by the administration of estrogens. Unfortunately, the correlation between levels of acid phosphatase and total body burden of prostatic cancer has been rather poor, and consequently values of acid phosphatase are not used to stage the tumor.

2- Alkaline phosphatases

Exist as a number of isoenzymes produced, for example, by liver, bone, or placenta. Elevation of alkaline phosphatase in the plasma of patients with malignancy is usually due to overproduction by either liver or bone and indicates involvement of those organs by metastatic disease. A number of benign disorders are also associated with increased plasma levels of the enzyme. Placental alkaline phosphatase is a normal placental protein that is found occasionally in the plasma of patients with ovarian cancer, testicular seminomas, and other tumors. It appears to be produced by the tumors themselves and may have a role in the diagnosis of mediastinal seminomas when histopathology is unclear as to the nature of the tumor.

3- Lactic dehydrogenase (LDH)

Is a tetramer comprised of combinations of two distinct polypeptide chains designated H for heart and M for muscle. Consequently, there are five possible isoenzymes; the occasional aberrant form has also been described. Elevations of LDH reflect tumor bulk in patients with lymphoma. In addition, enzyme levels, along with levels of β_2 -microglobulin, a lymphocyte membrane peptide and part of the MHC complex, are prognostic of outcome, independent of conventional prognostic criteria in lymphoma. Measurements of total LDH and of ratios of the isoenzymes have been used as indicators of metastases or of tumor burden in other malignancies but have limited value in monitoring the course of disease.

4- Gamma glutamyl transpeptidase (GGT)

Like the hepatic isozyme of alkaline phosphatase, may be elevated in the plasma of patients with hepatic metastases. The bulk of evidence suggests that elevated levels of the enzyme are due to canalicular obstruction, since increases in this enzyme usually parallel those of the hepatic form of alkaline phosphatase. Isozymes of GGT have been described, some of which may be tumor products.

5- Neurone-Specific Enolase (NSE)

Occurs as a dimer whose subunits are designated α and γ . The γ/γ isoform is neurone-specific and is frequently elevated in the plasma of patients with small-cell lung cancer. Its presence provides further evidence for the neuroendocrine origin of this tumor.

★ Immunoglobulins

In multiple myeloma and B-cell lymphomas there is often asynchronous synthesis of the polypeptide chains of immunoglobulins, and an excess of light over heavy chains is frequently formed. Excessive urinary excretion of light chains is almost pathognomonic of these disorders, although incomplete fragments of heavy chains or whole immunoglobulin molecules can be detected in serum or urine. Protein electrophoresis of these fluids will then show a sharp peak, indicating the presence of a monoclonal protein referred to in serum as an M-protein. The use of immunoglobulin markers for diagnosis and for monitoring treatment of lymphoid tumors provides an example of what may be expected of an ideal tumor marker. There is evidence that the level of M-protein reflects the total body burden of disease. While abnormal plasma levels of immunoglobulin can be found in many diseases and monoclonal peaks can occasionally be detected in the elderly population, believed then to represent a "forme fruste" of multiple myeloma, the association of an M-protein and abnormal plasma cells is quite specific for multiple myeloma. In addition, there is a

comparable degree of sensitivity since well over 90 % of patients with this disease will have elevated plasma or urine levels of immunoglobulin or their components at presentation. However, genotypic and phenotypic alterations may sometimes occur in these tumors so that cells may be selected during treatments that have different rates of production of the markers than in the original tumor.

★Tumor-associated Antigens

The widespread availability of techniques for the production of monoclonal antibodies has led to a renewed search for tumor-specific antigens. To do this, tissue cultures of homogeneous tumor cell lines have been established, followed by the generation of monoclonal antibodies to components of the tumor cells. Antigens that are associated with tumors originating from specific organs have been detected, but antigens with absolute specificity have not been isolated for any type of cancer. An example of this approach is the discovery of the

(1) Ovarian antigen CA 125 .

The antigen was isolated and purified from an ovarian cancer cell line. monoclonal antibodies were produced and an immunoradiometric assay developed.^[86] The antigen has proved useful for monitoring treatment of ovarian cancer, but it may also be produced by other tumors, such as those of the lung and large bowel.

(2) CA 15.3

Is one of the new generations of markers detected in an immunometric assay in which the catcher antibody is different from the tracer antibody. [The catcher antibody binds to the solid phase and to the antigen, the tracer binds to the antigen as well and carries the signal-producing mechanism Fig. 3.1]. The catcher antibody reacts with an epitope of a high-molecular weight glycoprotein present in human milk-fat globule membranes. CA 15.3 may be elevated in patients with adenocarcinomas of the breast, ovary, or lung, and it has been used as a guide to treatment of breast cancer. Succession of similar marker has been developed with a least one or the antibodies directed to mucin like cancer-associated antigens.

(3) Prostate-specific antigen (PSA)

Is a 30- to 40-kDa glycoprotein first isolated from human prostate extracts and seminal fluid. As appropriate assays became available for the measurement of PSA in plasma, they were noted to be quite specific for prostatic disease. Because marker levels appear to reflect tumor burden, the assay has assumed a major role in the clinical management of patients with prostatic adenocarcinoma, especially in those who have undergone total prostatectomy. PSA appears to be more sensitive and a better indicator of tumor burden than prostatic acid phosphatase. Unfortunately, the marker cannot be used by itself for diagnosis or screening because modest elevations are observed in patients with benign prostatic hypertrophy.

(4) Carcinoembryonic Antigen (CEA).

-cell surface glycoprotein cell adhesion molecule

-adult normal range: ≤ 3 ng/mL (non smoker) ≤ 5 ng/L (smoker)

*Clinical application:-

- a) Positive in colorectal cancer (approx. 70%), pancreatic carcinoma (approx. 55%), lung carcinoma (approx. 40%), breast carcinoma (approx. 40%), uterine carcinoma (approx.40%).
- b) Not useful for screening for the presence of malignancy: mildly elevated in benign diseases

-
- including smoking (false + ve) and not elevated In some gastrointestinal tumors (false -ve).
- c) Useful adjunct for clinical staging and likelihood of metastases.
 - d) Highly useful in monitoring for recurrent disease following therapy^[84].

(5) Cancer antigen 19.9 (CA 19.9)

1) Glycolipid related to the Le^a blood group antigen. It is denoted as Le^{xa}.

2) Marker for pancreatic carcinoma and colorectal carcinoma.

Elevated CA 19.9 levels have been noted in approximately 80% of patients with pancreatic cancer, 67% of patients with hepatobiliary carcinoma and 50% or less of patients with gastric and hepatocellular carcinoma. Up to 20% of patients with pancreatitis and other benign diseases may have elevations of CA 19.9^[84].

3) CA 19.9 is useful in monitoring recurrence or early relapse.

Unfortunately, detection of early relapse for pancreatic cancer has little clinical significance since there is no known therapy for this disease.

★ Miscellaneous markers

Polyamines, nucleosides, and tissue polypeptide antigen (TPA) are potential markers that reflect cellular proliferation and are increased nonspecifically in patients with cancer. The polyamines spermine, spermidine, and putrescine are products of ornithine decarboxylation and are found in increased concentrations in the urine whenever there is increased cell turnover. The nucleosides dimethyl guanosine and pseudouridine are components of RNA. As with the polyamines, they are released into the circulation in excessive amounts with enhanced cellular proliferation. TPA, which appears to be identical to a cytokeratin, is another nonspecific marker of cell turnover that has been investigated extensively. While these markers of proliferation cannot be used for diagnostic purposes, they be or Sallie use in monitoring the effects of therapy. However, inflammatory or other nonmalignant processes also cause elevation in plasma levels of these substances and may therefore lead to spurious information about the magnitude of therapeutic response.

Acute-phase proteins are under continued investigation as potential markers of malignancy. Most prominent among them are α_1 -acid glycoprotein and C-reactive protein. These proteins are not specific for malignancy but are increased in many inflammatory conditions. If one could rule out accompanying nonmalignant processes, monitoring of disease activity by measuring acute-phase proteins in the plasma might be worthwhile. Increases in acute-phase proteins are usually accompanied by decreases in other proteins in the plasma, notably albumin. They are produced in the liver, and their synthetic process involves diversion of amino acids from the synthesis of other proteins such as albumin. The function of the acute-phase proteins in cancer or other diseases is unknown. This overview of properties of some of the more common tumor markers can be supplemented by referring to a recent and comprehensive review (Sell, 1990)

▣ Sensitivity, specificity and screening

Sensitivity of a marker refers to the proportion of patients with a particular tumor who have elevated plasma levels of the marker. Sensitivity is indicated by the proportion of people who do not have the particular cancer, who have normal plasma levels of the marker. Ideally a marker will have high values of sensitivity and specificity.

A recurrent theme in the sequential evaluation of sensitivity and specificity of a marker substance is the initial finding of high values followed by disappointment as larger studies fail to confirm the results of a pilot investigation. The initial report of an association between CEA and colorectal cancer suggested the discovery of a marker that was extremely sensitive and relatively specific^[87]: 34 of 35 patients with colorectal cancer, studied at a time when tumor tissue was known to be present in the body, had plasma levels of CEA greater than 2.5 mg/ml, and 3 of 32 patients with

cancerous lesions of digestive organs other than large bowel and rectum had elevated CEA levels, while none of 133 controls without colorectal cancer had elevated levels. Subsequent investigators found lower values of about 70% for the sensitivity of elevated levels of CEA in colorectal cancer and variable levels of specificity.

Sensitivity = $\frac{\text{marker elevated}}{(\text{marker elevated} + \text{marker normal})}$

Specificity = $\frac{\text{marker normal}}{(\text{marker normal} + \text{marker elevated})}$

There are several reasons why preliminary results may not be confirmed in larger investigations. Initial studies usually choose patients from a hospital based populations who have advanced bulky tumors. Plasma levels of a marker in these patients are unlikely to reflect those in patients with minimal disease, where marker studies have the highest potential for clinical use. False-positive results in control populations may be rare if these are drawn initially from healthy volunteers, or from patients with benign disease of unrelated organs. Marker elevation is usually more common in patients with other malignancies or in benign disease of the tissue in which the *tumors* under investigation have originated; such patients should be included as controls in larger studies.

Methods for assessing the sensitivity of a marker relate to the detection of early or minimal disease. Preoperative levels of the marker may be correlated with staging at surgery; postoperative elevations of the marker may be correlated with the presence of residual disease at operation, or with the subsequent development of recurrence in patients where there is no apparent residual disease. Attempts to relate plasma levels of markers to clinical data are limited because the clinical assessment of tumor bulk is usually rather imprecise. Examples of the use of the foregoing criteria to measure sensitivity indicate that the CEA test is positive preoperatively in about 25 % of patients with Dukes' stage A (i.e., early) colorectal cancer. CA 125, a marker of ovarian cancer, is positive in about 80% of patients with minimal residual disease (<2 cm) following surgery.

The specificity of a marker may refer to cancer in general or to one type of cancer. If a marker is proposed as specific for a particular type of tumor, the probability of false-positive tests due to tumors of other organs must be established. The frequency of marker elevation must be ascertained in healthy subjects of varying ages in a variety of benign disorders, and particularly in benign diseases originating in the same organ as tumors that cause elevated plasma levels of the marker. If these criteria are applied to the ovarian antigen CA 125, it is found the test is not specific for ovarian cancer since elevation of plasma levels is found in an appreciable number of patients with other tumors such as lung cancer. There is modest elevation in plasma levels of the marker in about 10% of patients with benign gynecologic disease and more marked elevation in about 10% of patients with some other nonmalignant diseases such as cirrhosis of the liver. One can enhance the specificity of a marker by adjusting the threshold for a positive test, but this is accompanied by a fall in sensitivity. At a threshold level for CEA of 7.0 mg/ml, the specificity of marker elevation for colorectal cancer is 95 %. But note that the sensitivity for all colorectal cancers drops from 62 % to 37% and for early disease (Dukes' stages A and B1) from 28 % to 4%.

If several markers indicate the presence of a given type of cancer, use of a combination of markers will increase the sensitivity of tumor detection and if chosen appropriately may lead to only a small decrease in specificity. The ideal choice of markers in combination involves the selection of markers that individually are quite specific and that are complementary. Human chorionic gonadotropin and AFP, as markers of non seminomatous testicular tumors, are ideal in this respect; each marker, by itself, has a specificity of more than 90 % and is positive in about 60% of patients bearing these tumors. There is some overlap, but the markers are complementary to the extent that one or the other is positive in about 95 % of patients. The usefulness of a marker in screening is measured by the predictive value of a positive test, which depends not only on sensitivity and specificity but also on the prevalence of disease. This is by considering the use of a very sensitive marker with 90 %

specificity to screen for a common type of human cancer, with a prevalence of 1 in 1000 people. For each 1000 people that are screened, the marker would be elevated in 100 (since the specificity is 90 %), but only 1 of these will have disease. Such a marker might, however, have some use as an aid to diagnosis if patients are first preselected by the occurrence of symptoms, by performance of clinical and radiological tests, or in populations that are seriously at risk. Human chorionic gonadotropin and AFP in testicular cancer, and immunoglobulin products in myeloma, are useful diagnostic aids when used in this way. However, most epithelial tumor markers are neither sufficiently sensitive nor specific for diagnostic purposes. To establish whether they can be used for monitoring the status of disease, information is required about the relationship between plasma levels of markers and tumor burden.

cancer treatment

Radiation Therapy for Cancer: Questions and Answers

What is radiation therapy?

Radiation therapy (also called radiotherapy, x-ray therapy, or irradiation) is the use of a certain type of energy (called ionizing radiation) to kill cancer cells and shrink tumors. Radiation therapy injures or destroys cells in the area being treated (the “target tissue”) by damaging their genetic material, making it impossible for these cells to continue to grow and divide. Although radiation damages both cancer cells and normal cells, most normal cells can recover from the effects of radiation and function properly.

The goal of radiation therapy is to damage as many cancer cells as possible, while limiting harm to nearby healthy tissue. There are different types of radiation and different ways to deliver the radiation. For example, certain types of radiation can penetrate more deeply into the body than can others. In addition, some types of radiation can be very finely controlled to treat only a small area (an inch of tissue, for example) without damaging nearby tissues and organs. Other types of radiation are better for treating larger areas. In some cases, the goal of radiation treatment is the complete destruction of an entire tumor. In other cases, the aim is to shrink a tumor and relieve symptoms. In either case, doctors plan treatment to spare as much healthy tissue as possible. About half of all cancer patients receive some type of radiation therapy. Radiation therapy may be used alone or in combination with other cancer treatments, such as chemotherapy or surgery. In some cases, a patient may receive more than one type of radiation therapy.

When is radiation therapy used?

Radiation therapy may be used to treat almost every type of solid tumor, including cancers of the brain, breast, cervix, larynx, lung, pancreas, prostate, skin, spine, stomach, uterus, or soft tissue sarcomas. Radiation can also be used to treat leukemia and lymphoma (cancers of the blood-forming cells and lymphatic system, respectively). Radiation dose to each site depends on a number of factors, including the type of cancer and whether there are tissues and organs nearby that may be damaged by radiation.^[88] For some types of cancer, radiation may be given to areas that do not have evidence of cancer. This is done to prevent cancer cells from growing in the area receiving the radiation. This technique is called prophylactic radiation therapy. Radiation therapy also can be given to help reduce symptoms such as pain from cancer that has spread to the bones or other parts of the body. This is called palliative radiation therapy.

What is the difference between external radiation therapy, internal radiation therapy (brachy-therapy), and systemic radiation therapy? When are these types used?

Radiation may come from a machine outside the body (external radiation), may be placed inside the body (internal radiation), or may use unsealed radioactive materials that go throughout the body (systemic radiation therapy). The type of radiation to be given depends on the type of cancer, its location, how far into the body the radiation will need to go, the patient’s general health and medical history, whether the patient will have other types of cancer treatment, and other factors.^[89] Most people who receive radiation therapy for cancer have external radiation. Some patients have both external and internal or systemic radiation therapy, either one after the other or at the same time.

a) External radiation therapy usually is given on an outpatient basis; most patients do not need to stay in the hospital. External radiation therapy is used to treat most types of cancer, including cancer of the bladder, brain, breast, cervix, larynx, lung, prostate, and vagina. In addition, external radiation may be used to relieve pain or ease other problems when cancer spreads to other parts of the body from the primary site.

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- i) Intra-operative radiation therapy (IORT) is a form of external radiation that is given during surgery. IORT is used to treat localized cancers that cannot be completely removed or that have a high risk of recurring (coming back) in nearby tissues. After all or most of the cancer is removed, one large, high-energy dose of radiation is aimed directly at the tumor site during surgery (nearby healthy tissue is protected with special shields). The patient stays in the hospital to recover from the surgery. IORT may be used in the treatment of thyroid and colorectal cancers, gynecological cancers, cancer of the small intestine, and cancer of the pancreas. It is also being studied in clinical trials (research studies) to treat some types of brain tumors and pelvic sarcomas in adults.
 - ii) Prophylactic cranial irradiation (PCI) is external radiation given to the brain when the primary cancer (for example, small cell lung cancer) has a high risk of spreading to the brain. Internal radiation therapy (also called brachy therapy) uses radiation that is placed very close to or inside the tumor. The radiation source is usually sealed in a small holder called an implant. Implants may be in the form of thin wires, plastic tubes called catheters, ribbons, capsules, or seeds. The implant is put directly into the body. Internal radiation therapy may require a hospital stay. Internal radiation is usually delivered in one of two ways, each of which is described below. Both methods use sealed implants. Interstitial radiation therapy is inserted into tissue at or near the tumor site. It is used to treat tumors of the head and neck, breast, and perianal and pelvic regions, prostate, cervix, ovary. Some women treated with external radiation for breast cancer receive a “booster dose” of radiation that may use interstitial radiation or external radiation.
 - iii) Intra-cavitary or intra-luminal radiation therapy is inserted into the body with an applicator. It is commonly used in the treatment of uterine cancer. Researchers are also studying these types of internal radiation therapy for other cancers, including breast, bronchial, cervical, gallbladder, oral, rectal, tracheal, uterine, and vaginal.
- b) Systemic radiation therapy uses radioactive materials such as iodine 131 and strontium 89. The materials may be taken by mouth or injected into the body. Systemic radiation therapy is sometimes used to treat cancer of the thyroid and adult non-Hodgkin’s lymphoma. Researchers are investigating agents to treat other types of cancer.

What are the sources of energy for external radiation therapy?

The energy (source of radiation) used in external radiation therapy may come from the following:

- a. X-rays or gamma rays, which are both forms of electromagnetic radiation. Although they are produced in different ways, both use photons (packets of energy). X-rays are created by machines called linear accelerators. Depending on the amount of energy the x-rays have, they can be used to destroy cancer cells on the surface of the body (lower energy) or deeper into tissues and organs (higher energy). Compared with other types of radiation, x-rays can deliver radiation to a relatively large area.
- b. Gamma rays are produced when isotopes of certain elements (such as iridium and cobalt 60) release radiation energy as they break down. Each element breaks down at a specific rate and each gives off a different amount of energy, which affects how deeply it can penetrate into the body. (Gamma rays produced by the breakdown of cobalt 60 are used in the treatment called the “gamma knife.”)
- c. Particle beams use fast-moving subatomic particles instead of photons. This type of radiation may be called particle beam radiation therapy or particulate radiation. Particle beams are created by linear accelerators, synchrotrons, and cyclotrons, which produce and accelerate the particles required for this type of radiation therapy. Particle beam therapy uses electrons, which are produced by an x-ray tube (this may be called electron-beam radiation); neutrons, which are produced by radioactive elements and special equipment; heavy ions (such as protons and helium); and pi-mesons (also called pions), which are small, negatively charged particles produced by an accelerator and a system of magnets. Unlike x-rays and gamma rays, some

particle beams can penetrate only a short distance into tissue. Therefore, they are often used to treat cancers located on the surface of or just below the skin.

d. Proton beam therapy is a type of particle beam radiation therapy. Protons deposit their energy over a very small area, which is called the Bragg peak. The Bragg peak can be used to target high doses of proton beam therapy to a tumor while doing less damage to normal tissues in front of and behind the tumor. Proton beam therapy is available at only a few facilities in the United States. Its use is generally reserved for cancers that are difficult or dangerous to treat with surgery (such as a chondrosarcoma at the base of the skull), or it is combined with other types of radiation. Proton beam therapy is also being used in clinical trials for intraocular melanoma (melanoma that begins in the eye), retinoblastoma (an eye cancer that most often occurs in children under age 5), rhabdomyosarcoma (a tumor of the muscle tissue), some cancers of the head and neck, and cancers of the prostate, brain, and lung.

What are stereotactic radiosurgery and stereotactic radiotherapy?

Stereotactic (or stereotaxic) radiosurgery uses a large dose of radiation to destroy tumor tissue in the brain. The procedure does not involve actual surgery. The patient's head is placed in a special frame, which is attached to the patient's skull. The frame is used to aim high-dose radiation beams directly at the tumor inside the patient's head. The dose and area receiving the radiation are coordinated very precisely. Most nearby tissues are not damaged by this procedure. Stereotactic radiosurgery can be done in one of three ways. The most common technique uses a linear accelerator to administer high-energy photon radiation to the tumor (called "linac-based stereotactic radiosurgery"). The gamma knife, the second most common technique, uses cobalt 60 to deliver radiation. The third technique uses heavy charged particle beams (such as protons and helium ions) to deliver stereotactic radiation to the tumor. Stereotactic radiosurgery is mostly used in the treatment of small benign and malignant brain tumors (including meningiomas, acoustic neuromas, and pituitary cancer).

It can also be used to treat other conditions (for example, Parkinson's disease and epilepsy). In addition, stereotactic radiosurgery can be used to treat metastatic brain tumors (cancer that has spread to the brain from another part of the body) either alone or along with whole-brain radiation therapy. (Whole-brain radiation therapy is a form of external radiation therapy that treats the entire brain with radiation).^[87] Stereotactic radiotherapy uses essentially the same approach as stereotactic radiosurgery to deliver radiation to the target tissue. However, stereotactic radiotherapy uses multiple small fractions of radiation as opposed to one large dose. Giving multiple smaller doses may improve outcomes and minimize side effects. Stereotactic radiotherapy is used to treat tumors in the brain as well as other parts of the body. Clinical trials are under way to study the effectiveness of stereotactic radiosurgery and stereotactic radiotherapy alone and in combination with other types of radiation therapy.

Who plans and delivers the radiation treatment to the patient?

Many health care providers help to plan and deliver radiation treatment to the patient. The radiation therapy team includes the radiation oncologist, a doctor who specializes in using radiation to treat cancer; the dosimetrist, who determines the proper radiation dose; the radiation physicist, who makes sure that the machine delivers the right amount of radiation to the correct site in the body; and the radiation therapist, who gives the radiation treatment. Often, radiation treatment is only one part of the patient's total therapy. Combined modality therapy, the use of radiation with drug therapy, is commonly used. The radiation oncologist also works with the medical or pediatric oncologist, surgeon, radiologist (a doctor who specializes in creating and interpreting pictures of areas inside the body), pathologist (a doctor who identifies diseases by studying cells and tissues under a microscope), and others to plan the patient's total course of therapy.^[88] A close working relationship between the radiation

oncologist, medical or pediatric oncologist, surgeon, radiologist, and pathologist is important in planning the total therapy

What are radiosensitizers and radioprotectors?

Radiosensitizers and radioprotectors are chemicals that modify a cell's response to radiation. Radiosensitizers are drugs that make cancer cells more sensitive to the effects of radiation therapy. Several compounds are under study as radiosensitizers. In addition, some anticancer drugs, such as 5-fluorouracil and cisplatin, make cancer cells more sensitive to radiation therapy.

Radio-protectors (also called radioprotectants) are drugs that protect normal (noncancerous) cells from the damage caused by radiation therapy. These agents promote the repair of normal cells that are exposed to radiation. Amifostine (trade name Ethyol) is the only drug approved by the U.S. Food and Drug Administration (FDA) as a radioprotector. It helps to reduce the dry mouth that can occur if the parotid glands (which help to produce saliva and are located near the ear) receive a large dose of radiation. Additional studies are under way to determine whether amifostine is effective when used with radiation therapy to treat other types of cancer. Other compounds are also under study as radio-protectors.

What are radiopharmaceuticals? How are they used?

Radiopharmaceuticals, also known as radionucleotides, are radioactive drugs used to treat cancer, including thyroid cancer, cancer that recurs in the chest wall, and pain caused by the spread of cancer to the bone (bone metastases). The most commonly used radiopharmaceuticals are samarium 153 (Quadramet®) and strontium 89 (Metastron™). These drugs are approved by the FDA to relieve pain caused by bone metastases. Both agents are given intravenously (by injection into a vein), usually on an outpatient basis; sometimes they are given in addition to external beam radiation. Other types of radiopharmaceuticals, such as phosphorous 32, rhodium 186, and gallium nitrate, are not used as frequently. Still other radiopharmaceuticals are under investigation.

Chemotherapy For Cancer.

Many people fear chemotherapy because they have heard that it can have uncomfortable side effects. But side-effect management has come a long way over the last few decades. Today, many side effects once associated with chemotherapy can be prevented or controlled. With some types of chemotherapy, you may experience only minimal side effects. And chemotherapy may be your best option for a successful outcome. You can help achieve a successful outcome by understanding how side effects can impact your treatment. Learn how best to manage chemotherapy side effects.

Chemotherapy is the general term for any treatment involving the use of chemical agents to stop cancer cells from growing. Chemotherapy can eliminate cancer cells at sites great distances from the original cancer. As a result, chemotherapy is considered a systemic treatment. More than half of all people diagnosed with cancer receive chemotherapy. For millions of people, chemotherapy helps treat their cancer effectively, enabling them to enjoy full, productive lives.

A chemotherapy regimen (a treatment plan and schedule) usually includes drugs to fight cancer plus drugs to help support completion of the cancer treatment at the full dose on schedule. Most doctors agree that staying on your chemotherapy schedule gives you the best opportunity for a successful result^[88]

To get the most from chemotherapy, it's important to stick to a schedule of treatment. Find out more about chemotherapy cycles and schedules.

How Chemotherapy Works?

Chemotherapy is designed to kill cancer cells. Chemotherapy can be administered through a vein, injected into a body cavity, or delivered orally in the form of a pill, depending on which drug is used. Chemotherapy works by destroying cancer cells; unfortunately, it cannot tell the difference between a cancer cell and some healthy cells. So chemotherapy eliminates not only the fast-growing cancer cells but also other fast-growing cells in your body, including hair and blood cells. Some cancer cells grow slowly while others grow rapidly. As a result, different types of chemotherapy drugs target the growth patterns of specific types of cancer cells. Each drug has a different way of working and is effective at a specific time in the life cycle of the cell it targets. Your doctor will determine the chemotherapy drug that is right for you.

Discussing the Effectiveness of Cancer Treatment.

Understand the goals and risks of each treatment option so you can work with your doctor to decide which treatment is best for you. Balance potential benefits against the risks of treatment. Some risks of cancer treatments may include time away from family and friends, uncomfortable side effects, or long-term complications. Cancer treatment may be inconvenient, prolonged, or unavailable close to home. These are important considerations when evaluating treatment options, but they are not typically mentioned in medical journals reporting the results and benefits of new treatments. Once you and your doctor have decided on a treatment plan, talk with your doctor about all you can do to make sure you get the full dose of your cancer treatment on schedule..

Importance of Full Dose on Schedule

Studies show that for certain types of cancer, chemotherapy produces the best long-term results when patients receive the full dose on time, every time.

Your doctor will develop a treatment plan scientifically designed for you, based on your type of cancer, its stage of advancement, and your overall health. It will consist of specific chemotherapy agents, at specific doses and intervals. These are called your *scheduled cycles*. Generally, treatments are given daily, weekly, or monthly. Your doctor will help you determine the most effective treatment schedule for you.

The goal is to make your chemotherapy as effective, timely, and problem-free as possible. But while your chemotherapy treatment works to fight your cancer, it also can cause side effects such as a lowered white blood cell count. A low white blood cell count means your immune system isn't as strong as it could be which can increase your risk of infection. It also can require your doctor to change your dose or schedule of your chemotherapy. A chemotherapy-induced low white blood cell count, caused by healthy cells lost during chemotherapy, is an expected side effect of your treatment. Therefore, you can plan ahead so it is less likely to disrupt your treatment schedule. A low white blood cell count typically occurs after the administration of certain types of chemotherapy and may continue for several days.

To help reduce side effects like low white blood cell count that may interfere with your treatment schedule, learn more about managing chemotherapy side effects. Under certain circumstances, your doctor may decide your body is too weak to receive chemotherapy. A low white blood cell count can temporarily disrupt your cancer treatment or result in having your chemotherapy dose decreased. These changes to your treatment plan could make your cancer treatment less effective than it should be. To get the most from chemotherapy, it's important to stick to a schedule of treatment. Find out about chemotherapy cycles and schedules. Count

- Low white blood cell count
- Low red blood cell count
- Low platelet count
- Nausea
- Vomiting
- Hair loss
- Fatigue

Some side effects may be temporary and uncomfortable. Some can cause dose reductions and treatment delays or even be life-threatening. For example, one of the most serious potential side effects of chemotherapy is a low count of infection-fighting white blood cells—a condition called neutropenia (new-troh-PEE-nee-ah). Neutropenia can interrupt your chemotherapy schedule and put you at risk for infections that may require hospitalization and may even be life-threatening. Fortunately, significant progress has been made in the development of "proactive" therapies that help you manage the side effects of chemotherapy—ideally, before they interrupt your treatment schedule. Take an active role in managing side effects. Learn all you can, use your tools for organizing your cancer information to note any side effects you experience, and be sure to discuss them with your doctor.

Full Dose on Schedule

Certain side effects may prevent doctors from delivering your full dose of chemotherapy on schedule.

This can be a problem, since your best results typically depend on receiving treatment at the full dose and on your scheduled plan.

Impact of Delaying Treatment or Reducing Doses

A 2003 study reported in *The Journal of Clinical Oncology* by Dr. Gary Lyman, an oncologist and Professor at the University of Rochester, New York, found that many breast cancer patients did not receive the full doses of their chemotherapy on schedule

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- Most delays and reductions in the planned doses of chemotherapy were due to concerns about side effects.
 - Delays or reductions in the planned doses of chemotherapy were likely to affect the likelihood of a successful outcome. In many cases, such delays and reductions were preventable.
 - "The odds of curing breast cancer are highest when a patient completes a full course of chemotherapy,

Chemotherapy Side Effects

In the last 20 years, scientists have made a great deal of progress in developing therapies to help prevent and manage the side effects of chemotherapy. Newer supportive care treatments have led to vast improvements in the management of symptoms associated with cancer treatment. Many people don't experience side effects at all, and you are unlikely to experience all the side effects you read about below. All chemotherapy options are designed to treat cancer; unfortunately, they often affect parts of your body not directly affected by the cancer itself. This undesired result is referred to as a complication of treatment, or a side effect.

Side effects may be *acute* (short-term), chronic (long-term), or permanent. Side effects may cause inconvenience, discomfort, and even death. Additionally, certain side effects may prevent doctors from delivering the prescribed dose of chemotherapy at the specific time and schedule of the treatment plan.^[91]

Since the expected outcome from chemotherapy is based on delivering the full chemotherapy dose on schedule, it is important to understand chemotherapy cycles and schedules. Side effects from chemotherapy can include pain, diarrhea, constipation, mouth sores, hair loss, nausea and vomiting, as well as blood-related side effects. In this section, you can learn more about the importance in diagnosing and monitoring blood-related side effects. These may include low white blood cell count (neutropenia), low red blood cell count (anemia), low platelet count (thrombocytopenia), and related fatigue

CBC and Related Side Effects.

The CBC, or complete blood count, helps your doctor look for side effects of chemotherapy, which include changes in the three types of cells in your blood. Because chemotherapy kills fast-growing blood cells as well as cancer cells, side effects involving your blood are an almost-to-be-expected result of chemotherapy. Your first step in managing blood-related side effects is understanding CBC, or your complete blood count. Side effects involving blood include the following

Neutropenia

Neutropenia (new-troh-PEE-nee-ah) is the scientific name for a low infection-fighting white blood cell count. A low white blood cell count may leave your body vulnerable to infection and too weak to receive chemotherapy at the full dose on schedule.^[91] This could lead your doctor to delay your current treatment or reduce your doses until your count reaches sufficient levels. If not properly treated, infection can lead to hospitalization. To help reduce the risk of treatment delays due to blood-related side effects, find out more about the risks associated with low white cell blood count.

Anemia

Anemia (ah-NEE-mee-ah) is the scientific name for a low red blood cell count. A low red blood cell count may cause you to feel fatigued or sluggish because there is not enough oxygen circulating in your body. This condition can be effectively managed with one of several treatments, including blood transfusion if necessary. There are also other ways to manage low red blood cell count.

Thrombocytopenia

Thrombocytopenia (throm-boh-sy-toh-PEE-nee-ah) is the scientific name for a low platelet count. A low platelet count may cause you to experience bruising or excessive bleeding. Learn more about the risks of low platelet count. All of these side effects may be related to your chemotherapy. All are diagnosed through your CBC test. You can manage them to help reduce the possibility that they will compromise your treatment. A journal provides a place for you to keep track of your blood counts throughout your chemotherapy. In addition to blood-related side effects, chemotherapy can result in other side effects that can interfere with treatment if not managed properly.

Gene Therapy For Cancer

Key Points

- Gene therapy is an experimental treatment that involves introducing genetic material into a person's cells to fight disease)^[92]
- Researchers are studying gene therapy for cancer through a number of different approaches
- A gene can be delivered to a cell using a carrier known as a "vector." The most common types of vectors used in gene therapy are viruses^[90]
- The viruses used in gene therapy are altered to make them safe; however, some risks still exist with gene therapy
- A clinical trial using gene therapy must be approved by at least two review boards at the scientists' institution, as well as the U.S. Food and Drug Administration and the National Institutes of Health Recombinant DNA Advisory Committee^[92,91]

The Ethical, Legal, and Social Implications (ELSI) Program was established in 1990 to identify, analyze, and address the implications of human genetics research

What are genes?

Genes are the biological units of heredity. Genes determine obvious traits, such as hair and eye color, as well as more subtle characteristics, such as the ability of the blood to carry oxygen. Complex characteristics, such as physical strength, may be shaped by the interaction of a number of different genes along with environmental influences.

A gene is part of a deoxyribonucleic acid (DNA) molecule. Humans have between 50,000 and 100,000 genes. Genes carry instructions that allow the cells to produce specific proteins such as enzymes. During the creation of proteins, cells use another molecule, ribonucleic acid (RNA), to translate the genetic information stored in DNA. Only certain genes in a cell are active at any given moment. As cells mature, many genes become permanently inactive. The pattern of active and inactive genes in a cell and the resulting protein composition determine what kind of cell it is and what it can and cannot do. Flaws in genes can result in disease.

What is gene therapy?

Advances in understanding and manipulating genes have set the stage for scientists to alter patients' genetic material to fight or prevent disease. Gene therapy is an experimental treatment that involves introducing genetic material (DNA or RNA) into a person's cells to fight disease. Gene therapy is being studied in clinical trials (research studies with humans) for many different types of cancer and for other diseases. It is not currently available outside a clinical trial.

How is gene therapy being studied in the treatment of cancer?

Researchers are studying several ways to treat cancer using gene therapy. Some approaches target healthy cells to enhance their ability to fight cancer. Other approaches target cancer cells, to destroy them or prevent their growth. Some gene therapy techniques under study are described below.

- In one approach, researchers replace missing or altered genes with healthy genes. Because some missing or altered genes (e.g., p53) may lead to cancer, substituting "working" copies of these genes may keep cancer from developing.

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- Researchers are also studying ways to improve a patient's immune response to cancer. In this approach, gene therapy is used to stimulate the body's natural ability to attack cancer cells.
 - In some studies, scientists inject cancer cells with genes that make them more sensitive to chemotherapy, radiation therapy, or other treatments. In other studies, researchers place a gene into healthy blood-forming stem cells to make these cells more resistant to the side effects of high doses of anticancer drugs.
 - In another approach, researchers inject cancer cells with genes that can be used to destroy the cells. In this technique, "suicide genes" are introduced into cancer cells. Later, a pro-drug (an inactive form of a toxic drug) is given to the patient. The pro-drug is activated in cancer cells containing these "suicide genes," which leads to the destruction of those cancer cells.
 - Other research is focused on the use of gene therapy to prevent cancer cells from developing new blood vessels (angiogenesis).

How are genes transferred into cells so that gene therapy can take place?

In general, a gene cannot be directly inserted into a person's cell. It must be delivered to the cell using a carrier, or "vector." The vectors most commonly used in gene therapy are viruses. Viruses have a unique ability to recognize certain cells and insert their DNA into the cells. In some gene therapy clinical trials, cells from the patient's blood or bone marrow are removed and grown in the laboratory. The cells are exposed to the virus that is carrying the desired gene. The virus enters the cells and inserts the desired gene into the cells' DNA. The cells grow in the laboratory and are then returned to the patient by injection into a vein. This type of gene therapy is called *ex vivo* because the cells are grown outside the body. The gene is transferred into the patient's cells while the cells are outside the patient's body. In other studies, vectors (often viruses) or liposomes (fatty particles) are used to deliver the desired gene to cells in the patient's body. This form of gene therapy is called *in vivo*, because the gene is transferred to cells inside the patient's body.

What risks are associated with current gene therapy trials?

Viruses can usually infect more than one type of cell. Thus, when viral vectors are used to carry genes into the body, they might infect healthy cells as well as cancer cells. Another danger is that the new gene might be inserted in the wrong location in the DNA, possibly causing cancer or other harmful mutations to the DNA.

In addition, when viruses or liposomes are used to deliver DNA to cells inside the patient's body, there is a slight chance that this DNA could unintentionally be introduced into the patient's reproductive cells. If this happens, it could produce changes that may be passed on if a patient has children after treatment. Other concerns include the possibility that transferred genes could be "over expressed," producing so much of the missing protein as to be harmful; that the viral vector could cause inflammation or an immune reaction; and that the virus could be transmitted from the patient to other individuals or into the environment. Scientists use animal testing and other precautions to identify and avoid these risks before any clinical trials are conducted in humans.

What major problems must scientists overcome before gene therapy becomes a common technique for treating disease?

Scientists need to identify more efficient ways to deliver genes to the body. To treat cancer and other diseases effectively with gene therapy, researchers must develop vectors that can be injected into the patient and specifically focus on the target cells located throughout the body.

More work is also needed to ensure that the vectors will successfully insert the desired genes into each of these target cells. Researchers also need to be able to deliver genes consistently to a precise location in the patient's DNA, and ensure that transplanted genes are precisely controlled by the body's normal physiologic signals. Although scientists are working hard on these problems, it is impossible to predict when they will have effective solutions.

How do gene therapy trials receive approval?

A proposed gene therapy trial, or protocol, must be approved by at least two review boards at the scientists' institution. Gene therapy protocols must also be approved by the U.S. Food and Drug Administration (FDA), which regulates all gene therapy products. In addition, trials that are funded by the National Institutes of Health (NIH) must be registered with the NIH Recombinant DNA Advisory Committee (RAC). The NIH, which includes more than 20 institutes and offices, is the Federal focal point for biomedical research in the United States.

List Of Abbreviations

DNA : deoxy nucleic acid
AAF : acetylaminofluorene
UV : Ultraviolet
UVR : Ultraviolet radiation
low-LET : low-linear energy transfer
RBE : relative biological effectiveness
GY : Gray
BEIRT committee
MRI : magnetic resonance imaging
ELF-MF : extra low frequency electric and magnetic field
AAF: acetylaminofluorene
RNA: ribonucleic acid
tRNA: transfer ribonucleic acid
NCI : national cancer institute
G1 phase: first gap
G2 phase: second gap
S phase : synthesis of DNA
M phase : mitosis
TS: tumor suppressor
Cdc: cell cycle division
MPF : maturation promoting factor
EGFR : epidermal growth factor receptor
GDP : guanosine diphosphate
GTP : guanosine triphosphate
E2F : transcriptional factor
RB : retinoblastoma
CEA: carcinoembryonic antigen
P ACP : prostatic acid phosphatase
HCG : human chorionic gonadotropin
GGT: gamma glutamyl transpeptidase
NSE : neurone-specific enolase
CA : cancer antigen
PSA : prostatic specific antigen
TPA : tissue poly peptide antigen
IOPT : intraoperative radiation therapy
PCI : prophylactic cranial irradiation
FDA : food and drug administration
CBC : complete blood count
ELSI : ethical , legal, and social implications
NIH : national institute of health
RCA : recombinant DNA advisory committee

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