Mechanisms of Carcinogenesis and Individual Susceptibility to Cancer

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The population can be divided into four groups, or "oncodemes," depending on the relative contributions of environment and genetics to their risk of cancer. These oncodemes are: 1) background (random mutations in normal people); 2) environmental (environmental carcinogens acting on normal people); 3) environmental/genetic (environmental carcinogens acting on genetic susceptibility); and 4) genetic, with genetic susceptibility being more important than environmental exposure. Most cancer probably occurs in oncodemes 2 and 3. However, the contribution that genetic susceptibility to cancer makes to the total cancer burden is unknown. Genetic susceptibility may be important in occupational settings, where exposure to carcinogens is presumed to be restricted to concentrations believed to confer "acceptable" risks. This approach takes no account of individual susceptibility. The range over which metabolic polymorphisms exert their effects suggests that differences in pharmacogenetics between individuals may be important in occupational carcinogenesis. If this range were extrapolated to risk of human disease, at a given exposure one person might be 10–200 times more sensitive than another. Several metabolic polymorphisms have been linked to susceptibility to cancer. For some of these polymorphisms, genes have been cloned and genotypic tests based on use of the polymerase chain reaction have been developed. Limited data suggest that the effects of these polymorphisms on human cancer are complex. The genotypic testing of individuals occupationally exposed to carcinogens may provide firmer information on the relative contributions of nature and nurture on chemically induced cancer. The use to which such information is put demands wide debate.

Indexing Terms: genetic oncodemes/metabolic polymorphisms

With rare exceptions, the development of cancer in an individual depends on a complex interplay between nature and nurture (environment). It is therefore impossible, at present, to determine the precise contribution that individual susceptibility makes to risk of cancer in a given population. The problem has been addressed by Knudson (1). He suggested that the population can be divided into four groups (genetic oncodemes), depending on the relative contributions of environment and genetics to their risk of cancer. These oncodemes are as follows: 1) background—otherwise normal people who develop cancer as a result of random mutations; 2) environmental—otherwise normal people who develop cancer after exposure to environmental carcinogens (chemicals, radiation or viruses, or combinations of these); 3) environmental/genetic—people with genetic susceptibility to environmental carcinogens; and 4) genetic—people in whom genetic susceptibility to cancer is more important than spontaneous or environmentally induced events.

Most human cancer probably occurs in oncodemes 2 and 3, because exposure to environmental carcinogens is impossible to avoid and because cancers that are entirely due to an inherited predisposition are known to be rare (2–4). That mutation plays a central role in the development of cancer is now widely accepted and is supported by the following evidence (5). Cancer is a genetic disorder of somatic cells—malignant tumors contain cells that carry mutations in or deletions of critical cancer genes. Germline mutations (i.e., mutations that occur in gametes and that are passed from one generation to the next) in such genes result in heritable predispositions to cancer. Most carcinogens bind covalently to DNA and are mutagenic. Many are converted to their carcinogenic forms by cellular metabolism, which is under genetic control. Polymorphisms of genes for xenobiotic metabolism may confer susceptibility or resistance to chemically induced cancers. Germ-line mutations in DNA-processing and DNA-repair genes may result in heritable predispositions to cancer.

Multistep Carcinogenesis and Cancer Genes

It is generally accepted that the malignant phenotype develops as a result of an accumulation of mutations in genes that play a critical role in the control of cell division, growth, and differentiation. Epigenetic phenomena, such as hypomethylation of DNA, also participate in the process (4–6). This stepwise accretion of mutations is accompanied by a series of histopathological changes in which normal epithelium progresses to benign neoplasia (which in some people is reversible); the neoplasia in turn becomes dysplastic, invasive, and ultimately metastatic. The most well-documented example of this process is colorectal cancer (7) (Fig. 1). The development of colorectal cancer is accompanied by mutagenic events in two major classes of genes—protooncogenes and tumor-suppressor genes (6)—that are intimately concerned with control of cell-to-cell signaling, cell division, and programmed cell death. Recently, a third class of genes that predisposes to cancer has been postulated on the basis of the discovery of a gene on chromosome 2 associated with an increased risk of hereditary nonpolyposis colorectal cancer (HNPCC) (8, 9). It is proposed that genes of this
Implicit in the multistep model of carcinogenesis is the requirement for expansion of mutant clones, allowing overgrowth of genetically altered cells that possess a growth advantage over normal cells. Thus, carcinogenesis is a process that requires mutagenesis and mitogenesis (cell division) (5, 6).

Adduct Formation and Mutagenesis

A property common to a wide variety of otherwise disparate carcinogens is the ability to form covalent bonds with DNA to produce chemically stable products known as adducts (12–17). Adducts range in size and complexity from simple alkyl groups (e.g., methyl, ethyl) to bulky multi-ring residues from chemicals such as polycyclic aromatic hydrocarbons, aromatic amines, and aflatoxins. Adducts can link adjacent bases on the same strand (intrastrand cross-links) and can form interstrand cross-links between each strand of the duplex. Adduct formation is characteristic of genotoxic substances, and carcinogens with this property are known as genotoxic carcinogens.

Of chemicals known to be genotoxic, some are intrinsically reactive and can form DNA adducts directly, either with DNA in solution in a test tube or with DNA in a living cell. These directly acting agents include alkylsulfonic esters, epoxides, aromatic N-oxides, aromatic nitro compounds, lactones, alkyl nitrosoureas, and alkyl nitrosamides. They are all electrophilic, i.e., they acquire electrons during chemical reactions. DNA contains many nucleophilic centers—atoms that donate electrons (e.g., N-2, N-7, O-6 atoms of guanine). DNA-adduct formation occurs mainly by the reaction of electrophiles with nucleophilic centers in DNA (17).

Usually a single chemical will give rise to several different DNA adducts. This may be due to the production of several different metabolites from the same chemical, to reaction of a single reactive species with atoms of different nucleophilicities in the DNA molecule, or to a combination of both. Stereochemical and physicochemical constraints also play a part in determining the spectrum of DNA adducts formed by a given compound (13).

The biological consequences of adduct formation depend to a large extent on the nature of the adduct and its precise location in the DNA molecule (17–22). For example, a methyl group at N-7 of guanine is much less mutagenic than the same group at the O-6 position, because the latter participates in hydrogen bond formation during complementary base-pairing while the former does not. However, a bulky adduct such as that formed by aflatoxin B1 at N-7 of guanine is highly mutagenic because it causes gross distortion of the DNA structure. Adduct formation can lead to base substitution, deletion, and addition, and, therefore, to point mutation.

Metabolic Activation of Carcinogens

Many carcinogens only bind to DNA after undergoing metabolism to more reactive species. These are not electrophilic per se, but are converted to electrophiles by
eukaryotic cellular enzyme systems. This process of metabolic activation is a normal biochemical activity of mammals, allowing detoxification and elimination of potentially toxic substances that are generated by normal metabolism, are ingested in the diet, or enter the body in the form of drugs, occupational chemicals, or environmental contaminants. Enzymes with these functions fall into two main classes: phase I and phase II (23, 24).

Most phase I enzymes are members of the cytochrome P450 superfamily of enzymes—"monooxygenases"—that carry out oxidative metabolism by inserting one atom of oxygen into a relatively inert and usually nonpolar substrate. Cytochrome P450s catalyze the biosynthesis and degradation of many normal biochemical substrates, including steroids, fatty acids, prostaglandins, leukotrienes, biogenic amines, and plant metabolites.

Phase II enzymes conjugate reactive intermediates formed by oxidative metabolism with various endogenous molecules such as glucuronides, glutathione, or sulfate to polar, hydrophilic products that are readily excreted from cells and from the body.

The activities of phase I and II enzymes are coordinated in time and space to ensure that potentially reactive and harmful substances are eliminated (23, 24). Sometimes this benign and necessary housekeeping activity is perverted to a more ominous course when a chemically inert and genetically inactive molecule is converted to an electrophilic metabolite capable of reacting with DNA (12, 25). The inactive molecule is the "procarcinogen"; its intermediate metabolites are "proximate carcinogens"; and the electrophilic metabolite that actually reacts with DNA is the "ultimate carcinogen." A variety of different procarcinogens undergo metabolic activation to reactive electrophiles, including polycyclic aromatic hydrocarbons (15), aromatic amines (14), alkyl and aryl nitrosamines (26), and many natural products such as mycotoxins (e.g., aflatoxins) and plant products (e.g., safrole, cypacin) (16).

Inherited Susceptibility to Cancer

Given that somatic mutation, however provoked, initiates cancer, there will always be a background incidence for any given cancer because, by its nature, spontaneous mutation is impossible to prevent and is indeed the driving force of evolution. However, evidence suggests that the background incidence is a small fraction of the total (27) and that most cancer occurs in people who adopt a lifestyle with a particular carcinogenic risk (e.g., smoking; overnutrition) and (or) have a genetic susceptibility to cancer—oncogenes 2 and 3.

Genetic susceptibility to cancer can take several forms. In the most extreme cases a family will exhibit Mendelian inheritance of a mutant cancer gene that predisposes that person to cancer at one or more sites, the predisposition being carried on a single autosomal dominant gene. FAP is such a trait. An affected individual develops hundreds or thousands of colonic polyps during adolescence. Some of these progress to malignancy, and the patient will inevitably develop colon cancer by the third or fourth decade unless a prophylactic colectomy is performed. Offspring of carriers of the FAP gene (a tumor-suppressor gene known as APC) will be at 50% risk of developing FAP. (For other examples of heritable cancer genes see references 2, 4, 8, 9, 28, and 29.)

An enhanced rate of spontaneous mutation, due to defective DNA repair, inefficient "proofreading" during DNA replication, or chromosomal instability, can also be inherited and can also lead to increased susceptibility to cancer. Such disorders are often transmitted as autosomal recessive traits and include ataxia telangiectasia, Bloom's syndrome, Fanconi's anemia, and xeroderma pigmentosum (XP). XP is of particular interest because it is a rare example where the carcinogen (sunlight), the defect in DNA excision repair, and the site of the cancer (skin) can be directly related to each other (28, 29). The recently discovered HNPCC gene may turn out to be a dominantly inherited defect in proofreading during DNA replication (9).

Inherited constitutional chromosomal anomalies are sometimes accompanied by genetic susceptibility to cancer. For example, individuals with trisomy 21 (Down syndrome) have an 18-fold increased risk of leukemia, and men with Klinefelter's syndrome (an extra X chromosome) are at 20-fold excess risk of breast cancer (28, 29).

Metabolic Polymorphisms and Cancer

The extent to which the inherited dispositions to cancer discussed thus far contribute to the total burden of human cancer is unknown, but it is usually estimated to be between 1% and 5%, although this may be a serious underestimation (2). Accurate estimates of the fraction of human cancer that can be ascribed to genetic susceptibility cannot be made until we know far more about the role of metabolic polymorphisms in determining susceptibility to cancer. A polymorphic gene exists, in a given population, as two or more alleles at the same locus. The gene product of each allele will differ (sometimes by only one amino acid), and the differences are detectable at the phenotypic level by variations, e.g., in electrophoretic mobility or enzyme activity. There are numerous examples of metabolic polymorphisms in the human population, their effects being manifest by large differences in response to the same drug by different individuals (24).

Four such polymorphisms have been linked to susceptibility to cancer (Table 1). Polymorphisms in two cytochrome P450 genes, CYP1A (aryl hydrocarbon hydroxylase) and CYP2D6 (debrisoquine hydroxylation), are examples of phase I, oxidative metabolism. Genes NAT2 (acetyltransfer phenotype) and GST1 (glutathione transferase) exemplify phase II, conjugative metabolism (see above). At the time of writing, the effects of these pharmaco genetic polymorphisms on human cancer are still unresolved, with conflicting results in the literature. For example, several case-control studies based on the use of phenotyping have shown that individuals with slow rates of acetylation appear to be at higher risk of bladder cancer than individuals with normal rates (relative increase in risk of bladder cancer in slow acety-
Table 1. Metabolic polymorphisms and human cancer (adapted from reference 5).

<table>
<thead>
<tr>
<th>Usual name</th>
<th>Gene</th>
<th>Population frequency of polymorphism</th>
<th>Phenotypic features</th>
<th>Human cancer implicated</th>
</tr>
</thead>
<tbody>
<tr>
<td>AH locus polymorphism</td>
<td>AH, single diallelic locus</td>
<td>1 in 10</td>
<td>Induction of high levels of CYP1A1, which oxidatively metabolizes polycyclic aromatic hydrocarbons to reactive species</td>
<td>Lung cancer, especially in smokers</td>
</tr>
<tr>
<td>Debrisoquine hydroxylation</td>
<td>CYP2D6, autosomal recessive</td>
<td>1 in 12</td>
<td>Poor metabolizers have low rate of debrisoquine metabolism owing to low constitutive levels of cytochrome CYP2D6</td>
<td>Cancers of lung, liver, and gastrointestinal tract</td>
</tr>
<tr>
<td>Acetylator phenotype</td>
<td>NAT2 Slow acetylators are homozygous recessive for 1 of 2 “slow” alleles, each having a combination of two different point mutations</td>
<td>1 in 2</td>
<td>Slow rate of acetylation of arylamines and arylhydrazines</td>
<td>Bladder cancer, especially in people occupationally exposed to arylamines; colorectal cancer; lupus erythematosis</td>
</tr>
<tr>
<td>Glutathione-S-transferase</td>
<td>GSTM1 class μ. Null genotype, Autosomal dominant</td>
<td>1 in 2</td>
<td>Individuals with null genotype do not express glutathione-S-transferase μ, which conjugates trans-stilbene oxide with glutathione</td>
<td>Cancers of lung, stomach, colon, and bladder</td>
</tr>
</tbody>
</table>

The molecular epidemiology of polymorphisms that modulate cancer risk in the general population, in which familial clustering of genetic predisposition is rare, may in the long term provide the greatest opportunity to reduce deaths from cancer (3). Knowing the extent to which genetic susceptibility contributes to an individual’s risk of cancer is particularly important in the occupational setting, where, with few exceptions, exposure to noxious chemicals is intended to be restricted to levels believed to confer “acceptable risks.” A debate on who should decide to whom the risk is “acceptable” is beyond the scope of this article. At present, this approach takes no account of individual susceptibility because, as already stated, the contribution of genetic susceptibility to chemically induced cancers is not well established. That differences in pharmacogenetics between individuals may be important in protecting workers from carcinogens is suggested by the range (10- to 200-fold) over which metabolic polymorphisms exert their effects. Nebert (24) suggests that if this range were extrapolated directly to risk of human disease, at any given dose of drug or pollutant one person will be 10–200 times more sensitive to toxicity or cancer than another.

**Ethical Considerations**

Clinicians and genetic counselors are already confronted with problems created by advances in the genetics of cancer (32). For cancer genes giving very high or 100% risks, it might be possible to offer an affected individual prophylaxis or treatment. If genotyping is applied prenatally, abortion might be offered to parents who have conceived an affected fetus. Where prophylaxis or treatment is unavailable and abortion is rejected, some would argue that knowledge is worse than useless and would engender an unacceptable psycholog-
The implications of genotyping populations for particular polymorphisms are just as problematical. It will be difficult to weight the contribution of each polymorphism to overall risk. A metabolic polymorphism may protect against one chemical but increase the cancer risk of another. It may increase cancer risk in one organ but protect another. Polymorphisms of two or more different enzymes in the same person may interact. Suppose that these difficulties can be overcome and it becomes possible to identify genes that confer susceptibility or resistance to cancers induced by exogenous agents (e.g., ionizing radiation, industrial chemicals)—to what uses should such information be put? Will chemical manufacturers demand that all potential employees provide genotypes as part of their job applications? Will it be ethical to put people with “resistant” phenotypes into areas of high exposure, or to deny employment to persons with “high-risk” genetic profiles? Will it be permissible to lower industrial hygiene standards by employing a “resistant” workforce? Will failure to take into account a genetic profile be grounds for remedial action in the courts?

Mapping the human genome is now proceeding apace, and genotyping individuals (prenatally or at term) for susceptibility to disease is now a reality. Discrimination on the grounds of an “undesirable” genetic profile is on the horizon (33). The ethical dilemmas posed by the new genetics are likely to be more difficult to solve than the scientific problems that produce them.

I thank the Cancer Research Campaign and the Medical Research Council for financial support.

References