Recent Progress in the Genetics of Alcoholism

At the time of publication of the *Ninth Special Report to the U.S. Congress on Alcohol and Health* (National Institute on Alcohol Abuse and Alcoholism 1997), twin, family, and adoption studies had very firmly established major roles for both genetics and environment in the etiology of alcoholism in men. Although the earlier studies had failed to detect a genetic component of alcoholism in women, the newest studies at that time were beginning to suggest that alcoholism is as strongly genetically influenced in women as it is in men. Since alcoholism does not follow the simple rules of Mendelian inheritance in multigenerational pedigrees, it was clear that alcoholism is a genetically complex disorder, influenced by multiple genes (their precise number unknown) that interact in an unknown fashion with each other and with similarly unknown environmental factors to produce the disease. It also seemed highly likely that alcoholism is genetically heterogeneous, meaning that individuals in different families develop alcoholism under the influence of different predisposing genes. Some twin studies had also begun to suggest a partially shared genetic influence on both alcohol and tobacco use.

Just two of the genes influencing predisposition to alcoholism were known. A defective allele (variant) of the gene *ALDH2*, common in Asian populations, had long been known to substantially (although not completely) protect carriers from developing alcoholism by making them uncomfortable or ill after drinking alcohol. The *ALDH2* gene encodes aldehyde dehydrogenase, one of the two key liver enzymes involved in the metabolism of alcohol to its final end product, acetate. The illness resulting from the defective allele tended to prevent carriers from drinking enough alcohol to become addicted to it. Newer studies were beginning to suggest that alleles of *ADH2* and *ADH3*, genes encoding two forms of liver alcohol dehydrogenase (the enzyme that carries out the first step in alcohol metabolism in the liver), also protected carriers from developing alcoholism, albeit to a lesser extent than did the defective allele of *ALDH2*. The protective alleles of *ADH2* and *ADH3*, also common in Asian populations, encode forms of alcohol dehydrogenase that metabolize alcohol to acetaldehyde more rapidly than other forms of these enzymes do. This rapid metabolism leads to a greater buildup of this toxic product in the bloodstream after consumption of alcohol, thereby producing feelings of discomfort and illness and tending to discourage carriers of these alleles from consuming large amounts of alcohol.

Finally, there was a large controversy about the role in the etiology of alcoholism of a particular allele of *DRD2*, a gene encoding a particular form of brain receptor for dopamine. Dopamine is a neurotransmitter that plays a central role in brain pathways and that mediates the rewarding properties of alcohol and other drugs of abuse. While a large number of papers had concluded that this allele was associated with alcoholism, an even larger number of papers had reached the contrary conclusion. Questions were raised about the methodological validity of a number of the studies and their corresponding findings, the reasons for the inconsistent findings, and—even when the validity of some of the findings was assumed—their precise biological significance.

Findings from Twin/Family Studies

The classic twin study design compares the resemblances for a trait of interest between monozygotic (MZ, identical) twins and dizygotic (DZ, fraternal) twins, in order to determine the extent of genetic influence, or heritability, of the trait. Heritability can be calculated because MZ twins are genetically identical, whereas DZ twins share only half their genes. The method relies on the "equal-environment assumption," that is, that the similarity between the environments of both individuals in a pair of MZ twins is the same as the similarity between the environments of members of pairs of DZ twins. While earlier

twin studies have been severely criticized for not testing this assumption sufficiently, researchers have taken care more recently to collect data on the twins' environments, thereby allowing correction of results for any deviation from this assumption. While twin studies do not identify specific genes influencing a trait, they do provide important information on the trait's genetic architecture (more general properties of its inheritance pattern, such as whether genes act independently of one another, or in concert, to influence a trait), which aspects of the trait are most heritable, whether the same genes are influencing the trait in both genders, and whether multiple traits share any common genetic influences. When data on twins are augmented by data on their family members, the study is termed a twin/family study and can provide more precise information about whether parents transmit a behavioral trait to their offspring genetically or via some aspect of the familial environment (cultural transmission). When detailed data about the environment are collected, twin and twin/family studies can provide information about how environmental factors interact with genetic predisposition to produce a disease.

While earlier twin studies have firmly established substantial heritability of alcoholism in men (on the order of 50 percent), they have generally failed to detect heritability in women. This failure may be due, in part, to the lower rate of alcoholism among women than men, thereby necessitating larger sample sizes to achieve statistically significant results. Since the first studies to report a substantial heritability of alcoholism in women (Kendler et al. 1992), others have reported analysis of a sample of volunteer adult Australian twins consisting of 1,328 MZ pairs and 1,357 DZ pairs (distributed among all possible combinations of genders) (Heath et al. 1997). Of these subjects, about 25 percent of the men and about 6 percent of the women met DSM-IIIR criteria for alcohol dependence. (DSM-IIIR refers to the *Diagnostic and Statistical Manual of Mental Disorders, Third Edition, Revised,* a standard classification system for mental disorders [American Psychiatric Association 1987].) Analysis of the concordances for alcoholism among the various classes of twins suggested that about two-thirds of the risk of becoming alcoholic was genetically mediated in both men and women, with the remainder of the risk determined by environmental factors not shared by the two members of a given twin pair. The data provided no evidence for a difference in the degree of heritability in men and women, nor any evidence for genetic factors operating in one gender but not the other. This last conclusion was particularly aided by analyses of data from opposite-sex twin pairs, a type of analysis not previously reported. Using the same subject sample, these researchers have more recently demonstrated that childhood conduct disorder is significantly associated with risk for adult alcohol dependence in both men and women, with genetic factors accounting for most of the association in both genders (Slutske et al. 1998). These findings further emphasize the similarities in factors leading to alcoholism in men and women and suggest either that there are common genetic risk factors for conduct disorder and alcoholism in both genders, or that conduct disorder is itself a genetic risk factor for alcoholism. Since the subject sample for these studies came from the general population and because most of the alcoholics contained therein were relatively mildly affected, it is possible that the conclusions of these studies might not apply to very severely affected alcoholics, such as those identified from treatment centers.

Since individuals who eventually become alcoholic typically begin experimenting with alcohol use during adolescence and then proceed through stages of increasingly heavy use until they become addicted, investigators have long been interested in factors influencing initiation of alcohol use during adolescence. The notion that adolescents learn to use alcohol by modeling the alcohol use of their parents is an old one. Investigators tested this notion in a sample of 1,396 Dutch families, each consisting of a pair of adolescent twins and their parents (Koopmans and Boomsma 1996). The twins' alcohol use resembled that of their parents to some extent. For 17-year-olds, this resemblance could best be explained by genetic similarity of children to their parents, rather than

by children modeling their parents' drinking behavior. For 15- to 16-year-olds, while the resemblance of the children's drinking to that of their parents was explained principally by some aspect of the familial environment, the parents' drinking behavior itself accounted for, at best, only a small part of this resemblance. It appears from this study that children's drinking behavior is influenced primarily by genetic factors and by environmental factors other than their parents' alcohol use. This conclusion is consistent with findings from previous studies demonstrating strong peer influences on adolescent alcohol use.

Many, but not all, alcoholics suffer from medical complications of alcoholism, such as liver cirrhosis, pancreatitis, cardiomyopathy, or psychosis due to brain damage. The inconsistency with which medical complications occur in alcoholism has led to the plausible hypothesis that susceptibility to these complications is influenced by genetic factors independent of those influencing susceptibility to alcoholism itself. Researchers tested this hypothesis using 5,933 male MZ twin pairs and 7,554 male DZ twin pairs from the U.S. World War II Era Veteran Twin Registry (Reed et al. 1996). From this sample, 1,239 subjects had a diagnosis of alcoholism according to ICD-9 criteria (one of the two major classification systems used in *International Classification of Disease, Ninth Revision*, to diagnose mental disorders, including alcoholism [World Health Organization 1977]), 392 subjects had liver cirrhosis, and 242 subjects had alcoholic psychosis. Of the alcoholic subjects, 818 had neither cirrhosis nor psychosis, and 421 had either or both of these complications. From the MZ and DZ concordance rates for the three diseases, the investigators calculated heritabilities of 0.59 for alcoholism (in general agreement with results of other studies), 0.47 for liver cirrhosis, and 0.61 for alcoholic psychosis. For each trait, the remainder of the variance in susceptibility was due to environmental factors not shared by members of a twin pair. Using an analytic method that allowed for simultaneous analysis of all three diseases, the investigators calculated that 85 percent of the overall genetic risk was shared for alcoholism, cirrhosis, and psychosis. The small amount of genetic risk not accounted for by these shared factors was due to separate genetic factors for cirrhosis and psychosis, respectively. Although the role of these disorder-specific genetic factors was small, it was significant; removing these factors from the mathematical model resulted in a significantly worse fit to the data. The conclusion about the largely shared genetic susceptibility to all three diseases differs from that of an earlier analysis of part of these data (Hrubec and Omenn 1981), largely as a result of the more sophisticated analytic methods employed.

Why is so much of the genetic part of the risk for cirrhosis and alcoholic psychosis due to factors influencing the risk for alcoholism itself? Overall genetic risk for cirrhosis or psychosis refers to those genetic factors influencing the transformation of a normal (nonalcoholic) person into an alcoholic with cirrhosis or psychosis, respectively. The physiologic pathways leading to these medical complications pass obligatorily through alcohol addiction itself, presumably because such addiction is a precondition for the sustained high levels of consumption necessary to bring about the medical complications. There are multiple pathways leading to alcoholism, each with multiple steps. It seems likely that human populations contain a large amount of variation in the genes influencing many of these steps, leading collectively to a large genetic risk. If the physiologic pathways leading to cirrhosis and psychosis, *given that an individual is already consuming large amounts of alcohol*, are relatively simple with relatively few steps, then there will be relatively few opportunities for genetic variation to influence those steps. Alternatively, regardless of the number of steps in this part of the pathway, human populations might contain relatively little variation in the genes influencing these steps. Either of these situations would result in cirrhosis-specific and psychosis-specific genetic factors accounting for only a small part of the overall genetic risk for these complications.

While many observers have noted that alcoholics smoke very heavily, the reasons for this dual substance use have been poorly understood. Recent twin studies are shedding considerable light on the reasons for this phenomenon. In

one such study, researchers analyzed tobacco and alcohol use versus nonuse in a sample of 2,612 adolescent and young adult Dutch twin pairs (Koopmans et al. 1997). At all ages tested (12 through 25 years), regular alcohol use was highly correlated with regular tobacco use. For 12- to 16-year-olds, shared environmental factors were the principal influence on both alcohol and tobacco use. The same environmental factors (peer pressure very likely prominent among them) influenced both smoking and drinking. For 17- to 25-year-old men, both alcohol and tobacco use were highly genetically determined, with shared environmental influences playing a significant but lesser role. For 17- to 25-year-old women, alcohol use was highly genetically determined, and tobacco use was influenced by both genetic and shared environmental factors. The same genetic factors influenced both alcohol and tobacco use, both in young adult men and women. These findings suggest that while initial exposure to alcohol and nicotine is environmentally influenced, persistence in using these substances is under strong shared genetic influence.

Other investigators analyzed alcohol and tobacco use in 173 adult male MZ twin pairs and 183 adult male DZ twin pairs from the U.S. World War II Era Veteran Twin Registry (Swan et al. 1996). In this sample, both alcohol and tobacco consumption were approximately equally influenced by genetic and shared environmental factors. Correlations between alcohol and tobacco use were largely explained by a common genetic factor influencing use of both substances. There were also specific genetic factors influencing the use of either substance individually. Environmental factors influencing alcohol use were apparently different from those influencing tobacco use. These observations applied to the average range of alcohol and tobacco use, not heavy use. The same investigators analyzed heavy alcohol use (more than 67 drinks per month) and heavy tobacco use (more than 30 cigarettes per day) in a sample of 749 MZ twin pairs and 1,267 DZ twin pairs from the same registry (Swan et al. 1997). The threshold for heavy substance use was set at the top 20 percent of the range of quantity consumed, excluding nonusers from the distribution. This study demonstrated a common genetic influence on both heavy alcohol use and heavy tobacco use as well as genetic influences specific to the heavy use of each substance individually.

The physiologic mechanism of the shared genetic influence on alcohol and tobacco consumption is currently a matter of speculation, as illustrated by the following two hypotheses: Individuals with high reactivity to stress may use both substances for stress relief. Alternatively, use of either substance may induce physiologic tolerance to the other, leading to a need to consume greater amounts of the latter substance in order to experience a subjective effect. Independent twin studies have identified a shared genetic influence between alcoholism and depression (Kendler et al. 1993*a*), as well as between smoking and depression (Kendler et al. 1993*b*). The shared genetic influence on smoking and drinking could thus be related to their respective connections to depression. One study analyzed the relationship between tobacco use and perceived intoxication after consumption of a controlled dose of alcohol in a small sample of Australian twins (194 pairs) (Madden et al. 1997). This exploratory study suggested a complex genetic relationship in women between use of and responses to alcohol and tobacco. There appear to be at least two independent genetic factors involved, one influencing both alcohol and tobacco use, and another influencing smoking and feelings of intoxication after alcohol use but not alcohol use itself. The investigators found that subjects who smoked felt less intoxicated than those who did not. Further laboratory studies are needed to elucidate the biochemical and pharmacologic basis of this finding. The negative genetic correlation between smoking and sensitivity to alcohol, combined with Schuckit's observations that reduced sensitivity to alcohol predicts greater risk of alcoholism (Schuckit 1998), suggests that smoking may increase risk of alcoholism by reducing smokers' sensitivity to alcohol. The results of twin studies on smoking and drinking thus suggest that efforts toward prevention and treatment of alcohol abuse may benefit from inclusion of efforts to abate tobacco use.

Employing a novel conceptualization of "alcoholism treatment seeking" as a trait with both environmental and genetic influences, investigators examined the relationship between alcoholism and propensity to seek treatment for this disorder (True et al. 1996). In a sample of 1,864 MZ pairs and 1,492 DZ pairs of male, primarily Caucasian twins from the U.S. Vietnam Era Twin Registry (composed of pairs of twins who had served in the U.S. armed services between 1965 and 1975), about one-third of the subjects met DSM-IIIR criteria for alcohol dependence. Consistent with other twin studies, genetic influences accounted for 55 percent of the variance in alcoholism risk, with unshared environment accounting for the remainder of the variance. Tendency to seek treatment for alcoholism was highly familial. A mathematical model in which genetic and shared environmental factors each explained almost half the variance in treatment seeking fit the data well. Under this model, the factors influencing treatment seeking were independent of the factors influencing alcoholism itself. While it is not surprising that shared environment influences treatment seeking (through such factors as educational and socioeconomic level), the finding of a genetic influence on the tendency to seek treatment is novel. Perhaps this effect is mediated by known genetic influences on personality.

Findings From Genetic Linkage Studies

Identifying genes influencing predisposition to alcoholism is of critical importance for improving prevention and treatment of alcoholism for two principal reasons. First, it will permit identification of the proteins the genes encode and elucidation of the physiologic pathways in which these proteins function. Every step of every such pathway represents a potential target for prevention or intervention, for example, by design of an appropriately targeted drug. Second, knowledge of the genes influencing predisposition to alcoholism will permit better design of studies to elucidate environmental influences on alcoholism by permitting control of variation at the relevant genes in the subject sample, thereby reducing confusion about whether observed differences between experimental and control subjects are due to environmental or genetic influences.

While twin/family studies (such as those described above) can provide information about the genetic architecture of alcoholism and the relationship between genetic influences on alcoholism and other traits, they do not permit the identification of the specific genes influencing predisposition to alcoholism. Current efforts to identify such genes rely on genetic linkage and association studies. Such studies have received enormous impetus in recent years from the mapping of large numbers of human genetic markers (Broman et al. 1998)—recognizable sites along chromosomal deoxyribonucleic acid (DNA) that act as signposts for researchers—and genes (Schuler et al. 1996) under the Human Genome Project, which is supported by the National Institutes of Health and the U.S. Department of Energy, and from the development of more sophisticated statistical methods for analyzing gene mapping data (Lander and Schork 1994).

Genetic linkage studies can be designed in either of two principal ways. In the first design, investigators track the inheritance of the disease, along with that of genetic markers spanning the entire genome, through multigenerational families affected by the disease. Various complex statistical analyses permit the determination of which markers are cotransmitted with the disease. In the second design, investigators measure the degree of sharing of different marker alleles by members of pairs of siblings (or other relatives) affected by the disease. On average, simply by chance, siblings are expected to share half of the alleles of most of their genes. However, two siblings affected by the same disease will show more frequent sharing of alleles of markers close to genes affecting predisposition to, or progress of, the disease. Under either design, markers shown to be genetically linked to a disease—that is, inherited with the disease more frequently than would be expected by chance—define a chromosomal region(s) likely to contain a gene(s) influencing the disease. The advantage of this approach to gene discovery is that a sufficiently

comprehensive marker map, such as that now being assembled by the Human Genome Project, permits an unbiased search of the entire genome without requiring any prior physiologic hypothesis about which genes might influence the disease. Linkage studies work best for finding genes when the disease under study is influenced by a relatively small number of genes, each exerting a relatively large effect (Broman et al. 1998; Risch and Merikangas 1996).

The results of the first two systematic searches of the entire human genome (termed "genome scans") for genes influencing predisposition to alcoholism have recently been published. The first study, by the Collaborative Study on Genetics of Alcoholism (COGA), a National Institute on Alcohol Abuse and Alcoholism (NIAAA)-supported consortium of investigators at six centers across the United States, reported results from a primarily Caucasian-American sample of 987 individuals from 105 families (Reich et al. 1998). In order to reduce errors in classification of subjects as alcoholic or non-alcoholic, the study defined alcohol-dependent individuals as those meeting two independent criteria for alcoholism: the DSM-IIIR criteria for alcohol dependence, and the Feighner criteria at the definite level (Feighner et al. 1972). This study found suggestive evidence for genes influencing susceptibility to alcoholism on chromosomes 1 and 7 as well as weaker evidence for a gene on chromosome 2. It also reported modest evidence for a gene reducing the risk for alcoholism on chromosome 4. An independent genome scan, based on 152 subjects from a Southwestern American Indian tribe, has been reported by investigators in NIAAA's own research laboratories (Long et al. 1998). With use of the DSM-IIIR definition of alcohol dependence, this study reported suggestive evidence for a gene influencing susceptibility to alcoholism on chromosome 11 as well as suggestive evidence for a protective gene on chromosome 4 in approximately the same region implicated by the COGA study.

Rather than identifying specific genes, these studies have implicated certain chromosomal regions as containing genes influencing susceptibility to alcoholism. Each implicated region contains hundreds of genes, and determination of precisely which genes located therein influence alcoholism will require higher resolution mapping studies in the future. It is not surprising that the two studies implicated different chromosomal regions because (1) American Indians and Caucasian-Americans (of European descent) have different genetic histories and therefore contain different genetic variation, and (2) the physiologic mechanisms leading to alcoholism in American Indians may be different from those in Caucasians. In view of these differences between the two subject populations, it is of interest that both studies found some evidence for a protective gene in the same region of chromosome 4. Plausible candidates for this gene in this region are *ADH2* and *ADH3*, which encode alcohol dehydrogenases and for which some alleles have been shown to reduce susceptibility to alcoholism in Asian populations (Whitfield 1997), and *GABARB1*, which encodes a subunit of a receptor for the major brain inhibitory neurotransmitter gamma-aminobutyric acid (GABA). The function of this receptor is stimulated by alcohol, possibly accounting for alcohol's sedating and motor-discoordinating effects (Suzdak et al. 1986). Long-term consumption of alcohol alters the brain distribution and function of this receptor, possibly playing a role in the development of alcohol dependence (Mitsuyama et al. 1998). Further studies will be required to determine whether any of these candidates is actually the gene inferred by both linkage studies to be responsible for protection from alcohol dependence.

The investigators responsible for both linkage studies have deliberately described their findings as suggestive rather than definitive. They exercise such caution because the observed sibling allele-sharing patterns (from which they have inferred the locations of the genes) deviate significantly from randomness but not so much so that one can be absolutely certain that the inferred genes are real. Certainty about the locations of the genes will require replication of the results of these studies in independent subject samples.

Such studies are now under way. While these first two genome scans have not definitively identified genes influencing predisposition to alcoholism, they nonetheless represent a major step toward that goal. In the intermediate term, further progress will depend on the development of more sophisticated statistical methods that are capable of simultaneously analyzing data not only on alcoholism itself but also on a number of psychological and physiologic traits, such as temperament, sensitivity to alcohol, and various kinds of brain waves that may represent additional dimensions of the disease. These additional dimensions may ultimately constitute essential elements of biologically valid definitions of alcoholism, which will be a natural prerequisite for gene identification. Help in initial localization of disease genes will also come from new statistical methods for analyzing the effects of multiple genes simultaneously rather than one at a time, as most current methods do. Precise gene identification will depend ultimately on the development of a complete gene map of the human genome. It also will depend on use of genetic association studies (see below) that test the association of alcoholism with specific alleles of genes lying within chromosomal regions identified by these and other linkage studies.

Findings From Genetic Association Studies

Genetic association tests measure whether a particular allele of a gene occurs more frequently in individuals affected by a disease than in unaffected individuals. A finding of genetic association can indicate that the gene under study influences the disease. Although such tests require prior knowledge of the gene under study (unlike genetic linkage tests), they are statistically much more powerful than linkage tests for detecting genes exerting only small effects on predisposition to a disease (Broman et al. 1998; Schuler et al. 1996). They are also easier to perform than linkage tests, requiring ascertainment only of disease cases (and sometimes their parents) and controls rather than the entire nuclear families or large, multigenerational families required for linkage studies. However, since an apparent association between an allele and a disease can arise for reasons other than the influence of that allele on the disease, association studies can be highly prone to artifact and need to be carefully designed. Well-designed genetic association studies have the following characteristics (Broman et al. 1998; Schork and Schork 1998; Schuler et al. 1996):

- In case-control studies comparing unrelated affected individuals (cases) with unrelated individuals who are unaffected by the disease (controls), the controls need to be ethnically matched to the cases. Ideally, both cases and controls should come from a population that has arisen from a relatively small number of originating ancestors (Finns, French Canadians, Amish, and so on). This matching is necessary because different populations can have very different allele frequencies for reasons totally unrelated to presence or absence of disease. Comparisons of unmatched samples can thus result in artifactual associations between alleles and disease.

- An alternative approach to avoiding the ethnic matching problem is to use a family-based study design, which compares alleles transmitted to affected offspring by their parents with the parental alleles that were not transmitted, thereby avoiding use of a separate control group. Several analytic methods have been developed for this type of study design, for example, the haplotype relative risk method (Knapp et al. 1993) and the transmission-disequilibrium test (Spielman and Ewens 1996).

- When possible, haplotypes (clusters of genetic polymorphisms, or variations in the DNA sequence, occurring within a small chromosomal region) should be analyzed rather than merely single polymorphisms (Kidd et al. 1996). Such analyses yield a more accurate picture of the relationship between the disease-causing polymorphism and the surrounding chromosomal region.

- The polymorphisms analyzed should cause demonstrated functional alterations in the proteins encoded by the genes under study. In

cases where several genes are known to lie close to the site of the polymorphism being studied, demonstration of functional alteration in the protein product of one of these genes can help decide which of these genes is actually related to the disease.

- Findings should be replicated in independent subject samples.

An excellent example of studies meeting many of the design criteria listed above was a group of seven that focused on the association of the alcohol dehydrogenase genes *ADH2* and *ADH3* with alcoholism. These studies, done in various ethnically matched Asian subject samples, were meta-analyzed in an effort to measure to what extent genetic variation in these genes affects the risk of alcoholism (Reich et al. 1998). Meta-analysis is an approach that involves the use of specialized statistical methods to pool data from several studies. The polymorphisms analyzed have been demonstrated to result in changes in the speed with which the enzymes metabolize alcohol to form acetaldehyde. The results are also consistent across the seven studies included in the meta-analysis. Thus, it can now be regarded as firmly established that alleles encoding faster metabolizing forms of *ADH2* and *ADH3* reduce the risk that carriers of these alleles will develop alcoholism.

As mentioned in the introduction of this section, many association studies of alcoholism with the dopamine receptor gene *DRD2* have been published (Feighner 1972), with strikingly conflicting results. Many of these studies do not meet the design criteria listed above. However, some do meet these criteria, and results of these best designed studies are consistently negative. Researchers studied the association with alcoholism of an allele of *DRD2* known to produce a receptor defective in intracellular signaling in a Southwestern American Indian tribe, analyzing haplotypes rather than just this single allele (Goldman et al. 1997). Their results—no association of the defective allele with alcoholism—were similar to those of another study analyzing the same allele in Germans (Finckh et al. 1996). A third group of investigators using the transmission-disequilibrium test (one of the analytic methods mentioned above developed for family-based studies) analyzed three different DRD2 polymorphisms in a large U.S. sample (principally Caucasian) from the COGA study and found no association of any of these polymorphisms with alcoholism (defined in any of three different ways) (Edenberg et al. 1998). These negative findings do not mean that the dopamine receptor D2 plays no role in the process of addiction to alcohol. Indeed, there is excellent evidence from pharmacologic studies in both humans and animals that this receptor (as well as the entire dopaminergic pathway) plays a major role in brain reward circuits, the function of which is altered by addiction. Rather, the results mean that genetic variants of this receptor do not explain why some people are more predisposed than others to become alcoholic.

Genes encoding other components of the brain dopaminergic pathway also have been tested for association with alcoholism. While none of the findings has been definitive, two studies have produced suggestive results especially worthy of further pursuit. In one, investigators studied the association with alcoholism of alleles of the tyrosine hydroxylase gene, which encodes an enzyme centrally involved in the synthesis of dopamine, by using cladistic analysis. This analysis makes use of the evolutionary history of the chromosomal region containing the gene under study to afford a more precise and powerful statistical test of association of alleles with the disease (Lobos and Todd 1997). Although the results of this study were ambiguous, the application of cladistic analysis to other association studies deserves further exploration. Other investigators have found an association of a functional variant of the dopamine receptor D4 (a receptor type distinct from D2) with alcoholism in a small sample of severely affected Japanese alcoholics (Muramatsu et al. 1996). Validation of this finding will require replication in a larger subject sample.

As can be seen from the above examples, most genes tested so far for association with alcoholism

have been those that, based on independent physiologic or pharmacologic evidence, have already been suspected to play a role in predisposition to alcoholism. The power of genetic studies to reveal the influence of previously unsuspected genes on predisposition to alcoholism, thereby affording insights into previously unrecognized disease mechanisms, thus remains to be exploited, at least in genetic association studies. The authors of a recent paper have demonstrated that once all of the genes in the human genome have been mapped, and the more common alleles of each of them characterized, it will become possible to conduct statistically powerful tests of all genes in the genome for association with diseases (Schuler et al. 1996). These tests will have greater power than linkage tests to detect genes having small effects on disease predisposition. To hasten the day when such tests become possible, the National Human Genome Research Institute is leading a trans-National Institutes of Health (NIH) initiative to catalogue, as quickly as possible, a large fraction of the common polymorphisms in the human genome (Collins et al. 1997). NIAAA, along with other NIH Institutes, is providing funding for this initiative.

In Closing

The greatest value expected to accrue from genetic studies toward the improvement of treatment and prevention of alcoholism will come from identification of the predisposing genes and the proteins they encode. Since mapping genes that influence genetically complex diseases like alcoholism presents difficult challenges for investigators, progress on such diseases, including alcoholism, has been much slower than progress in gene mapping for single-gene disorders. Despite the difficulty of the problem, NIAAA-supported researchers have taken important steps during the last 3 years by identifying human chromosomal regions possibly containing relevant genes. Twin studies, which explore the relationship between alcoholism and other traits, continue to contribute to the formulation of a more biologically valid definition of the disease and to resolving disease subtypes that may ultimately prove to have differing genetic bases. Achievement of these objectives would greatly expedite the gene search. Further progress toward precise identification of genes influencing predisposition to alcoholism will depend on the development of improved tools for the gene-discovery enterprise. Foremost among these tools will be more sophisticated statistical methods, a complete human gene map, and a catalogue of the major human genetic polymorphisms. Once genes influencing predisposition to alcoholism have been identified, a major new challenge confronting genetic epidemiologists will be to understand how such genes (many of which will have been discovered in families specially selected to be densely affected by alcoholism) interact with environmental factors to influence the development of alcoholism in the general population.

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