ALCOHOL ALERT

National Institute on Alcohol Abuse and Alcoholism No. 61 April 2004

Neuroscience Research and Therapeutic Targets

Alcoholism, like other addictions, is a brain disorder. Research has shown that genes shape how an individual experiences alcohol—how intoxicating, pleasant, or sedating it is—and how susceptible he or she is to developing alcohol use disorders. Research has also shown that chronic heavy drinking causes long-term—and perhaps permanent—changes in the way the brain responds to alcohol. These parallel insights from neuroscience research are paving the way for new medications that will improve alcoholism treatment and relapse prevention.

Addiction science has benefited from rapid progress in cellular and molecular research techniques, from the integration of scientific disciplines in the study of addiction-related behavior, and from the development of more appropriate animal models (1). Research in genetics is paying off in the identification of genes that influence the risk of developing alcoholism (2–7). Many of the genes being identified direct the production of proteins involved in the complex process of signaling between neurons in the brain. For example, genes that encode subunits of receptors for neurotransmitters such as GABA, serotonin, and others have been identified (see below for background on these neurotransmitters). Other genes related to alcoholism risk encode enzymes that metabolize alcohol. Gene discovery offers multiple benefits. Identification of risk-associated genes may provide a means of identifying people at risk. As important, knowing the genes and the proteins they encode is a key to understanding how alcohol interacts with this part of the cell's machinery, how variants of the gene raise or lower risk, and how chronic exposure to alcohol can change gene expression (the translation of genes into proteins) and set the stage for addiction. Some of the genes being identified raise the risk of both alcoholism and so-called comorbid disorders, like depression, that often occur along with alcohol problems. Knowledge of these genes should provide insight into the brain mechanisms that underlie these disorders. Finally, identifying genes provides potential targets for medication development. A recent *Alcohol Alert* on the genetics of alcoholism describes some of the approaches being used to identify genes related to alcoholism risk (8).

This *Alcohol Alert* provides a brief overview of what research is revealing about how alcohol affects the brain and how the resulting changes contribute to alcohol dependence. Also addressed is what research is showing about the effect of stressful life experiences on the brain and how they may contribute to risk of alcohol dependence and relapse to drinking. Beyond understanding how alcohol affects the brain, the hoped-for outcome of this work is the identification of neurologic targets for potential medications. Some of the medications in clinical use or testing that have come out of this work are reviewed below.

Alcohol Alert, *a publication of the National Institute on Alcohol Abuse and Alcoholism, provides timely information on alcohol research and treatment to health professionals and other interested people. This issue is the sixty-first in the series.*

Research in genetics is paying off in the identification of genes that influence the risk of developing alcoholism.

Alcohol Interferes with Brain Cell Communication

Large and often widespread networks of brain cells perform the brain's essential functions: storing information, regulating basic body functions, and directing behavior. The basis of these brain networks is communication from cell to cell by chemical messengers called neurotransmitters. Released into narrow gaps, or synapses, between cells, neurotransmitters cross the synapse and activate proteins called receptors. Receptor activation, in turn, leads to a series of molecular interactions within the receiving cell. Some of the molecular interactions are short-term and remain localized to the area of the cell containing the receptors. Others

U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
Public Health Service • National Institutes of Health

result in lasting changes, at multiple locations throughout the cell, in protein expression, structure, and composition.

Intoxication and other short-term (acute) effects of alcohol are caused largely by temporary, reversible changes in specific receptors and associated molecules. With repeated (chronic) alcohol exposure, long-lasting changes occur in receptors and in the series of chemical interactions they signal. However, neuroscientists have found that receptor changes are only one example of many permanent changes in the brain, collectively referred to as "neuroadaptation," caused by the presence of alcohol. Strong evidence exists that neuroadaptation involves changes at many different levels, from the genetically directed production of critical proteins (9–12) to physical changes in the structure of the cells on both sides of the synapse—that is, both the signaling and the receiving cell.

Unraveling these different aspects of neuroadaptation may be the key to understanding how addiction develops. Recent studies have linked neuroadaptation with tolerance (the need to drink more alcohol to achieve the same level of intoxication) (13,1) and with the symptoms of withdrawal (14). Neuroadaptation also appears to underlie the persistent sense of discomfort, often described as "craving," that can lead to relapse even after long periods of abstinence (14–17).

Unraveling these different aspects of neuroadaptation may be the key to understanding how addiction develops.

Medications from Neuroscience

Based on neuroscience research, scientists are developing medications that potentially could target both the acute responses to alcohol and the neuroadaptations that can accompany chronic drinking. Potential medications may target specific receptor types, the series of chemical reactions set off by receptor activation, or the production of critical protein enzymes involved in these processes within cells. To use these strategies effectively and safely, however, researchers must first understand in detail where and how alcohol exerts its effects.

Naltrexone and acamprosate are two medications that act on receptor systems in the brain on which alcohol is known to have an impact and that have shown some success for treating alcoholism. Naltrexone binds with receptors for endogenous opioids, naturally occurring opiate-like substances that stimulate pleasurable feelings and suppress pain. Animal studies suggest that opiate antagonists like naltrexone block some of alcohol's rewarding effects. Clinical studies have reported that alcohol-dependent patients given naltrexone drink less frequently, and in smaller quantities, than patients given a placebo (18). Naltrexone has been approved by the U.S. Food and Drug Administration (FDA) for alcoholism treatment.

Naltrexone and acamprosate are two medications that act on receptor systems in the brain on which alcohol is known to have an impact and that have shown some success for treating alcoholism.

Acamprosate's precise mechanism of action is not yet known, but it is thought to affect activity of the neurotransmitter glutamate (18,19). In clinical studies in Europe, patients on acamprosate experienced higher abstinence rates, and for those who did relapse, longer periods of abstinence (18). Clinical studies using acamprosate are ongoing in the United States, but it has not yet been FDA approved.

Despite promising results for some patients using naltrexone or acamprosate, not everyone responds. It is likely that different subtypes of alcoholics have different genetically determined traits shaping their response to alcohol and underlying their vulnerability to alcohol problems. For these reasons, the need remains for new medications, with a variety of drugs eventually providing a way to target treatment according to a person's individual biology.

Inhibitory and Excitatory Neurotransmitters: GABA and Glutamate

One of the most powerful actions of alcohol is to reduce the overall level of brain activity by a combination of effects on two key neurotransmitters, GABA (gamma-aminobutyric acid) and glutamate. Alcohol enhances the activity of GABA, the brain's chief inhibitory neurotransmitter. At the same time, alcohol reduces the excitatory effects of glutamate. These actions are the main reason that alcohol is often thought of as a depressant.

Alcohol enhances the activity of GABA, the brain's chief inhibitory neurotransmitter. At the same time, alcohol reduces the excitatory effects of glutamate.

GABA is the neurotransmitter-receptor system that has historically received the most attention in alcohol research, but it remains difficult to exploit therapeutically. Its major receptor type, the GABA-A receptor, is involved in many of alcohol's acute and chronic effects (20–25). Medications that block GABA's ability to bind at the GABA-A receptor also block some of alcohol's effects (1), but because this receptor system plays a role in so many vital brain functions, blocking it has side effects. Current GABA-A–blocking drugs can cause convulsions, a side effect that must be eliminated before this receptor system can be targeted for therapy.

Baclofen, a drug that activates another type of GABA receptor (GABA-B), has recently been shown in a preliminary study to be effective in inducing abstinence from alcohol and reducing alcohol craving and consumption in alcoholics (26). Use of baclofen to treat alcohol-

dependent patients merits further investigation. Treatment for withdrawal commonly involves drugs that act on GABA-A receptors. Investigators are searching for new, safer drugs that increase GABA activity. One such drug—**gabapentin**—is currently being tested (27).

Alcohol reduces the activity of the neurotransmitter glutamate by interacting with NMDA receptors, one of several classes of receptor to which glutamate binds. Preclinical data suggest that reducing NMDA neurotransmission may be effective in treating alcoholism. **Memantine**, a drug that reduces NMDA receptor function, looks promising as an anticraving drug and in treating alcohol dependence. Clinical trials to establish its efficacy are being contemplated (28). More recently, it has been shown that the anticonvulsant drug, **topiramate**, which acts on another class of glutamate receptors (AMPA-kainate receptors), decreases glutamate activity while increasing GABA activity. A recent study reported that alcohol-dependent patients on topiramate had fewer drinking days and had fewer drinks on days they did drink (compared with participants taking a placebo) (29). Topiramate reportedly reduced craving. Additional studies are needed to confirm this work and provide information on how best to use topiramate: on which groups of patients, in what dosage, and with what types of psychosocial therapy.

Receptors Signaling Pleasure: Serotonin and Cannabinoids

Other candidates for drug treatment are aimed at brain receptors thought to be involved with the mood-elevating, rewarding sensations associated with alcohol.

Other candidates for drug treatment are aimed at brain receptors thought to be involved with the mood-elevating, rewarding sensations associated with alcohol. The neurotransmitter serotonin is involved in the regulation of attention, emotion, and motivation. Alcohol alters serotonin neurotransmission (30). In addition, depression is a common co-occurring disorder with alcoholism. One class of drugs used to treat depression—selective serotonin reuptake inhibitors (SSRIs)—increases the availability of serotonin in the synapse. Studies examining whether SSRIs such as fluoxetine (Prozac) and sertraline (Zoloft) might be helpful in treating alcoholism co-occurring with depression have produced mixed results (31–33). What these studies suggest is that individuals vary not only in terms of their risk of alcohol problems, but in how they respond to both pharmacologic and behavioral treatment.

The drug ondansetron reduces the activity of a serotonin receptor (5-HT3) on which alcohol is known to act and has been shown to reduce the desire to drink.

Research on other drugs that affect serotonin neurotransmission also points to variability in individual responses to treatment. For example, the drug **ondansetron** reduces the activity of a serotonin receptor (5-HT3) on which alcohol is known to act and has been shown to reduce the desire to drink in humans. A clinical trial demonstrated that ondansetron was most effective in reducing the frequency and quantity of drinking in early-onset (alcohol-dependent before age 25) vs. late onset (alcohol-dependent after age 25) alcoholics (34). Larger scale studies are needed to confirm these results. Continued research on genetics and neurophysiology should help refine the understanding of what shapes individual responses to drugs and how treatments can be tailored accordingly (35).

Another recent candidate as a target for therapy is a brain receptor (CB1) that responds to endocannabinoids, innate substances that interact with the CB1 receptor in a manner similar to the active ingredients in marijuana. Like serotonin, the endocannabinoid system is involved in the rewarding effects of alcohol (36–40,12,41). A drug that blocks CB1 has been found to reduce alcohol consumption in rats (42–44), suggesting the possibility of using such medications to help people undergoing alcohol detoxification (45).

Cellular Enzymes

An enzyme called protein kinase C (PKC) is the subject of much research because it helps shape the level of sensitivity to alcohol's behavioral effects, at least in part through its interaction with the GABA-A receptor.

Another strategy for developing therapies focuses not on receptors but on enzymes within the cell that are involved in the brain's neuroadaptation to alcohol. Chronic drinking alters the distribution of these enzymes within cells and can change the way brain cells respond to alcohol (46–48,12). An enzyme called protein kinase C (PKC) is the subject of much research because it helps shape the level of sensitivity to alcohol's behavioral effects, at least in part through its interaction with the GABA-A receptor. In a study looking at PKC and alcohol sensitivity, mutant mice that lacked a form of PKC (PKC epsilon) were more sensitive than their littermates to the behavioral effects of alcohol. Without alcohol, the mutant mice did not appear sleepy or sedated, however (46). More recently, the same team reported that the mice lacking PKC epsilon do not work as hard for alcohol (by pressing a lever) as their littermates and do not show the same increases in dopamine that are usually associated with the reinforcing effects of alcohol. Withdrawal-associated seizures are also less severe in these mutated mice (49,50). This suggests that medications developed to inhibit PKC epsilon might reduce alcohol reward as well as help treat seizures, but without the sedating effects of other drugs that act on GABA neurotransmission.

Scanning the Genome

Using gene markers (known variations in genetic material spaced along the DNA chains that make up chromosomes) scientists can scan the entire genome for chromosomal stretches that are associated with alcoholism risk. NIAAA's Collaborative Study on the Genetics of Alcoholism (COGA) has identified stretches on chromosomes 1, 2, 4, 7, 15, and 16 that are associated with alcoholism risk (51–54). Work by other investigators has also found confirming evidence for linkage to chromosome 1 (55), and in American Indians, to chromosomes 1 and 4 (56,57). Recently COGA investigators found evidence that variations in a gene on chromosome 4 encoding the alpha subunit of the GABA-A receptor and, on chromosome 15, in the GABA-A receptor gene *GABRG3* influence alcoholism risk (5,4). These findings bring the work full circle, tying risk associated with a chromosomal location to a specific gene known to be involved in the response to alcohol.

Other emerging methods can be used to assess the activities of thousands of genes at once. This new technology (microarray gene analysis) promises to expand the pool of target molecules for alcoholism researchers and to help them zero in on the brain areas most affected by the disorder. It is possible to compare the patterns of protein production in the brain cells of animals bred to respond differently to alcohol or with different exposure to alcohol; or, of people with and without alcohol addiction (in this case, cells are obtained at autopsy). Comparisons of which genes are active in different brain areas can direct attention to the regions where alcohol exerts its greatest effects and to genes involved in the alcohol response (9,58–61,12,11). A recent paper reported on the identification of a gene that is differentially expressed in brain regions of inbred alcohol-preferring and nonpreferring rats (62). The gene codes for alpha synuclein, a protein that has been shown to be involved in neurotransmission. Another recent study looked specifically at changes in gene expression in mice shortly after a dose of alcohol. Out of about 24,000 genes, the screen identified 61 responding in a significant way to alcohol, including sets of genes that have roles such as protecting cells from injury and glucose metabolism, as well as being involved with behavioral responses to alcohol (63). Further research will clarify the roles of all these genes in the body's response to alcohol.

Comparisons of which genes are active in different brain areas can direct attention to the regions where alcohol exerts its greatest effects and to genes involved in the alcohol response.

Neurosystems—The Circuits and Networks of Stress

Neuroscience research is beginning to reveal how different brain regions contribute to the complex process of addiction (64). The brain is subdivided into many specialized regions, which set up connections, or "circuits," with other regions. These circuits, in turn, interact with other circuits to form networks that integrate the functions of the brain.

The pleasurable effects of drugs of addiction, including alcohol, are mediated by the so-called "reward" circuits of the brain (65,16,1). Following short-term exposure to alcohol, these circuits are able to return to their normal level of function. With repeated exposure to alcohol, however, the responsiveness of these circuits changes. For example, studies have demonstrated in rats that the function of neurotransmitters involved in reward is reduced during withdrawal from alcohol, while at the same time, stress-related systems are activated (1). Levels of dopamine, a neurotransmitter associated with reward, are lower in rats following withdrawal than before the rats were made dependent. The rats also react with increased anxiety to stressful situations following withdrawal. Research suggests that the discomfort and distress that result from these persistent changes in brain reward and stress circuits underlie the compelling motivation to drink by alcohol-dependent individuals.

The pleasurable effects of drugs of addiction, including alcohol, are mediated by the so-called "reward" circuits of the brain.

The link between alcohol consumption and stress is complex. Studies suggest that exposure to stress may lead to the initiation and continued use of alcohol (17,66). In fact, many researchers believe that alcohol's stress-relieving effect is what prompts most people to drink (67). While alcohol use may temporarily relieve the symptoms of stress, chronic drinking not only can lead to alcohol-related problems, it may exacerbate the adverse effects of stress, leaving the brain in a state of permanent "physiological stress." This effect may help explain why alcoholics are likely to relapse during stressful life events, even after months or years of abstinence (16,1).

Among the molecules involved in regulating physiological reactions to stress, two in particular have drawn the attention of alcohol researchers. Corticotropin-releasing factor (CRF) is a critical messenger in the stress circuits that work primarily within the brain; neuropeptide Y (NPY) is involved with processes ranging from appetite to memory and stress responses (68–73). In a recent study, both alcohol and CRF increased GABA neurotransmission in mice—except in

While alcohol use may temporarily relieve the symptoms of stress, chronic drinking not only can lead to alcohol-related problems, it may exacerbate the adverse effects of stress, leaving the brain in a state of permanent "physiological stress."

mice missing one of two types of CRF receptors (74). This suggests that the missing receptors (CRF1) are a key link between the GABA-enhancing (and rewarding) effects of alcohol and the neurobiology of stress.

Among NPY's actions, it appears to counteract CRF effects. Research has linked chronic alcohol intake to imbalances in CRF and NPY (68). Both these molecules are logical targets for therapeutic research.

Through basic neuroscience research, scientists are gaining a better understanding of how neuroadaptation sets the stage for alcohol addiction, and how stress can influence both dependence and relapse. Development of effective new medications for alcoholism requires a strategy that takes into account the many different possible interactions of alcohol with the brain, and the genetically determined variability among individuals.

Neuroscience Research and Therapeutic Targets—A Commentary by NIAAA Director Ting-Kai Li, M.D.

Alcohol interacts with the brain in complex ways. Its short-term effects can transiently alter behavior, while long-term exposure to alcohol can result in lasting changes in the brain, or neuroadaptation. Each point in the course of alcohol's interactions with cell surface receptors, intracellular messenger molecules, genes, and brain circuits offers a potential target for pharmacologic intervention. The promise of interdisciplinary research in neuroscience is to link genetic, molecular, cellular, anatomic, and behavioral data and provide both an understanding of how individuals respond to alcohol and a guide for targeting prevention and treatment.

References

(1) Koob, G.F. Alcoholism: Allostasis and beyond. *Alcoholism: Clinical and Experimental Research* 27(2):232–243, 2003. **(2) Wernicke, C.**; Samochowiec, J.; Schmidt, L.G.; et al. Polymorphisms in the *N*-methyl-D-aspartate receptor 1 and 2B subunits are associated with alcoholism-related traits. *Biological Psychiatry* 54:922–928, 2003. **(3) Miyatake, R.**; Furukawa, A.; Matsushita, S.; et al. Functional polymorphisms in the $Sigma_1$ receptor gene associated with alcoholism. *Biological Psychiatry* 55:85–90, 2004. **(4) Dick, D.M.**; Edenberg, H.J.; Xuei, X.; et al. Association of *GABRG3* with alcohol dependence. *Alcoholism: Clinical and Experimental Research* 28(1):4–9, 2004. **(5) Edenberg, H.**; Dick, D.M.; Xuei, X.; et al. Variations in *GABRA2*, encoding the alpha2 subunit of the $GABA_A$, are associated with alcohol dependence and with brain oscillations. *American Journal of Human Genetics* 74:705–714, 2004. **(6) Herman, A.I.**; Philbeck, J.W.; Vasilopoulos, N.L.; and Depetrillo, P.B. Serotonin transporter promoter polymorphism and differences in alcohol consumption behaviour in a college student population. *Alcohol and Alcoholism* 38:446–449, 2003. **(7) Ozaki, N.**: Goldman: Kaye, W.H.; et al. Serotonin transporter missense mutation associated with a complex neuropsychiatric phenotype. *Molecular Psychiatry* 8:933–936, 2003. **(8) National Institute on Alcohol Abuse and Alcoholism (NIAAA).** The genetics of alcoholism. *Alcohol Alert* No. 60. Rockville, MD: NIAAA, 2003. **(9) Lewohl, J.M.**; Wang, L.; Miles, M.F.; et al. Gene expression in human alcoholism: Microarray analysis of frontal cortex. *Alcoholism: Clinical and Experimental Research* 24(12):1873–1882, 2000. **(10) Heinz, A.**, and Mann, K. Neurodegeneration and neuroadaptation in alcoholism. In: Agarwal, D.P., and Seitz, H.K., eds. *Alcohol in Health and Disease.* New York: Marcel Dekker, 2001. pp. 225–241. **(11) Crabbe, J.C.** Genetic contributions to addiction. *Annual Review of Psychology* 53:435–462, 2002. **(12) Hoffman, P.L.**; Miles, M.; Edenberg, H.J.; et al. Gene expression in brain: A window on ethanol dependence, neuroadaptation, and preference. *Alcoholism: Clinical and Experimental Research* 27(2):155–168, 2003. **(13) Kiianmaa, K.**; Hyytiä, P.; Samson, H.H.; et al. New neuronal networks involved in ethanol reinforcement. *Alcoholism: Clinical and Experimental Research* 27(2):209–219, 2003. **(14) Hoffman, P.L.**; Morrow, L.; Phillips, T.J.; and Siggins, G.R. Neuroadaptation to ethanol at the molecular and cellular levels. In: Noronha, A.; Eckardt, M.; and Warren, K., eds. *Review of NIAAA's Neuroscience and Behavioral Research Portfolio*. NIAAA Research Monograph No. 34. NIH Pub. No. 00–4579. Bethesda, MD: National Institute on Alcohol Abuse and Alcoholism, 2000. pp. 85–188. **(15) Weiss, F.** Neuroadaptive changes in neurotransmitter systems mediating ethanol-induced behaviors. In: Noronha, A.; Eckardt, M.; and Warren, K., eds. *Review of NIAAA's Neuroscience and Behavioral Research Portfolio.* NIAAA Research Monograph No. 34. NIH Pub. No. 00–4579. Bethesda, MD: National Institute on Alcohol Abuse and Alcoholism, 2000. pp. 261–313. **(16) Koob, G.F.,** and Le Moal, M. Drug addiction, dysregulation of reward, and allostasis. *Neuropsychopharmacology* 24(2):97–129, 2001. **(17) Kreek, M.J.**; LaForge, K.S.; and Butelman, E. Pharmacotherapy of addictions. *Nature Reviews Drug Discovery* 1:710–726, 2002. **(18) National Institute on Alcohol Abuse and Alcoholism (NIAAA).** *Tenth Special Report to the U.S. Congress on Alcohol and Health.* NIH Pub. No. 00–1583. Bethesda, MD: NIAAA, 2000. **(19) Johnson, B.A.**, and Ait-Daoud, N. Medications to treat alcoholism. *Alcohol Research & Health* 23(2):99–106, 1999. **(20) Mehta, A.K.**, and Ticku, M.K. An update on $GABA_A$ receptors. *Brain Research Reviews* 29(2–3):196–217, 1999. **(21) Buck, K.J.**, and Finn, D.A. Genetic factors in addiction: QTL mapping and candidate gene studies implicate GABAergic genes in alcohol and barbiturate withdrawal in mice. *Addiction* 96(1):139–149, 2000. **(22) Loh, E.W.**, and Ball, D. Role of the $GABA_A\beta2$, $GABA_A\alpha6$, $GABA_A\alpha1$, and $GABA_A\gamma2$ receptor subunit genes cluster in drug responses and the development of alcohol dependence. *Neurochemistry International* 37(5–6):413–423, 2000. **(23) Cagetti, E.**; Liang, J.; Spigelman, I.; and Olson, R.W. Withdrawal from chronic intermittent ethanol treatment changes subunit composition, reduces synaptic function, and decreases behavioral responses to positive allosteric modulators of $GABA_A$ receptors. *Molecular Pharmacology* 63(1):53–64, 2003. **(24) Blednov, Y.A.**; Jung, S.; Alva, H.; et al. Deletion of the $\alpha1$ or $\beta2$ subunit of $GABA_A$ receptors reduces actions of alcohol and other drugs. *Journal of Pharmacology and Experimental Therapeutics* 304(1):30–36, 2003. **(25) Kralic, J.E.**; Wheeler, M.; Renzi, K.; et al. Deletion of $GABA_A$ receptor $\alpha1$ subunit-containing receptors alters responses to ethanol and other anesthetics. *Journal of Pharmacology and Experimental Therapeutics* 305(2):600–607, 2003. (**26) Addolorato, G.**; Caputo, F.; Capiristo, E.; et al. Baclofen efficacy in reducing alcohol craving and intake: A preliminary double-blind randomized controlled study. *Alcohol and Alcoholism* 37(5):504–508, 2002. **(27) Bonnet, U.**; Banger, M.; Leweke, M.; et al. Treatment of acute alcohol withdrawal with gabapentin: Results from a controlled two-center trial. *Journal of Clinical Psychopharmacology* 23(5):514–519, 2003. **(28) Bisaga, A.**, and Evans, S.M. Acute effects of memantine in combination with alcohol in moderate drinkers. *Psychoparmacology* 172:16–24, 2004. **(29) Johnson, B.A.**; Ait-Daoud, N.; Bowden, C.L.; et al. Oral topiramate for treatment of alcohol dependence: A randomized controlled trial. *Lancet* 361(9370):1677–1685, 2003. **(30) Lovinger, D.M.** Serotonin's role in alcohol's effects on the brain. *Alcohol Health & Research World* 21(2):114–120, 1997. **(31) Cornelius, J.R.**; Salloum, I.M.; Ehler, J.G.; et al. Fluoxetine in depressed alcoholics. A double-blind, placebo-controlled trial. *Archives of General Psychiatry* 54(8):700–705, 1997. **(32) Moak, D.H.**; Anton, R.F.; Latham, P.K.; et al. Sertraline and cognitive behavioral therapy for depressed alcoholics: Results of a placebo-controlled trial. *Journal of Clinical Psychopharmacology* 23(6):553–562, 2003. **(33) Pettinati, H.M.**; Volpicelli, J.R.; Luck, G.; et al. Double-blind clinical trial of sertraline treatment for alcohol dependence. *Journal of Clinical Psychopharmacology* 21(2)143–153, 2001. **(34) Johnson, B.A.**; Roache, J.D.; Javors, M.A.; et al. Ondansetron for reduction of drinking among biologically predisposed alcoholic patients. *Journal of the American Medical Association* 284(8):963–971, 2000. **(35) Kranzler, H.R.** Medications for alcohol dependence. *Journal of the American Medical Association* 284:1016–1017, 2000. **(36) Childers, S.R.**, and Breivogel, C.S. Cannabis and endogenous cannabinoid systems. *Drug and Alcohol Dependence* 51(1–2):173–187, 1998. **(37) Basavarajappa, B.S.**, and Hungund, B.L. Neuromodulatory role of the endocannabinoid signaling system in alcoholism: An overview. *Prostaglandins, Leukotrienes and Essential Fatty Acids* 66(2–3):287–299, 2002. **(38) Hungund, B.L.**; Basavarajappa, B.S.; Vadasz, C.; et al. Ethanol, endocannabinoids, and the cannabinoidergic signaling system. *Alcoholism: Clinical and Experimental Research* 26(4):565–574, 2002. **(39) González, S.**; Cascio, M.G.;

Fernándo-Ruiz, J.; et al. Changes in endocannabinoid contents in the brain of rats chronically exposed to nicotine, ethanol, or cocaine. *Brain Research* 954:73–81, 2002. **(40) Schmidt, L.G.**; Samochowiec, J.; Finckh, U.; et al. Association of a CB1 cannabinoid receptor gene (CNR1) polymorphism with severe alcohol dependence. *Drug and Alcohol Dependence* 65(3):221–224, 2002. **(41) Wang, L.**; Liu, J.; Harvey-White, J.; et al. Endocannabinoid signaling via cannabinoid receptor 1 is involved in ethanol preference and its age-dependent decline in mice. *Proceedings of the National Academy of Sciences* U.S.A. 100(3):1393–1398, 2003. **(42) Hungund, B.L.**, and Basavarajappa, B.S. Role of brain's own marijuana, anandamide and its cannabinoid receptors (CB1) in alcoholism. *Recent Research Developments in Neurochemistry* 3(Part 1):9–26, 2000. **(43) Freedland, C.S.**; Sharpe, A.L.; Samson, H.H.; and Porrino, L.J. Effects of SR141716A on ethanol and sucrose self-administration. *Alcoholism: Clinical and Experimental Research* 25(2):277–282, 2001. **(44) Vacca, G.**; Serra, S.; Brunetti, G.; et al. Boosting effect of morphine on alcohol drinking is suppressed not only by naloxone but also by the cannabinoid receptor antagonist, SR 141716. *European Journal of Pharmacology* 445(1–2):55–59, 2002. **(45) Lallemand, F.**; Soubrié, P.H.; and De Witte, P.H. Effects of CB1 cannabinoid receptor blockade on ethanol preference after chronic ethanol administration. *Alcoholism: Clinical and Experimental Research* 25(9):1317–1323, 2001. **(46) Hodge, C.W.**; Mehmert, K.K.; Kelley, S.P.; et al. Supersensitivity to allosteris $GABA_A$ receptor modulators and alcohol in mice lacking PKCε. *Nature Neuroscience* 2(11):997–1002, 1999. **(47) Nuwayhid, S.J.**, and Werling, L.L. $\sigma_1$receptor agonist-mediated regulation of *N*-methyl-D-aspartate-stimulated [^{3}H]dopamine release is dependent upon protein kinase C. *Journal of Pharmacology and Experimental Therapeutics* 304(1):364–369, 2003. **(48) Proctor, W.R.**; Poelchen, W.; Bowers, B.J.; et al. Ethanol differentially enhances hippocampal $GABA_A$ receptor-mediated responses in protein kinase Cγ (PKCγ) and PKCε null mice. *Journal of Pharmacology and Experimental Therapeutics* 305(1):264–270, 2003. **(49) Olive, M.F.**; Mehmert, K.K.; Messing, R.O.; and Hodge, C.W. Reduced operant ethanol self-administration and *in vivo* mesolimbic dopamine responses to ethanol in PKCε-deficient mice. *European Journal of Neuroscience* 12:4131–4140, 2000. **(50) Olive, M.F.**; Mehmert, K.K.; Nannini, A.; et al. Reduced ethanol withdrawal severity and altered withdrawal-induced c-fos expression in various brain regions of mice lacking protein kinase C-epsilon. *Neuroscience* 103:171–179, 2001. **(51) Foroud, T.**; Bucholz, K.K.; Edenberg, H.J.; et al. Linkage of an alcoholism-related severity phenotype to chromosome 16. *Alcoholism: Clinical and Experimental Research* 22(9):2035–2042, 1998. **(52) Foroud, T.**; Edenberg, H.J.; Goate, A.; et al. Alcoholism susceptibility loci: Confirmation studies in a replicate sample and further mapping. *Alcoholism: Clinical and Experimental Research* 24(7):933–945, 2000. **(53) Reich, T.**; Edenberg, H.J.; Goate, A.; et al. A genome-wide search for genes affecting the risk for alcohol dependence. *American Journal of Medical Genetics (Neuropsychiatric Genetics)* 81:207–215, 1998. **(54) Song, J.**; Koller, D.L.; Foroud, T.; et al. Association of $GABA_A$ receptors and alcohol dependence and the effects of genetic imprinting. *American Journal of Medical Genetics Part B (Neuropsychiatric Genetics)* 117B:39–45, 2003. **(55) Lappalainen, J.**; Kranzler, H.R.; Petrakis, I.; et al. Confirmation and fine mapping of the chromosome 1 alcohol dependence risk locus. *Molecular Psychiatry* 9:312–319, 2004. **(56) Ehlers, C.L.**; Gilder, D.A.; Wall, T.L.; et al. Genomic screen for loci associated with alcohol dependence in Mission Indians. *American Journal of Medical Genetics Part B (Neuropsychiatric Genetics).* Early View, published online (www3.interscience.wiley.com), June 7, 2004. **(57) Long, J.C.**; Knowler, W.C.; Hanson, R.L.; et al. Evidence for genetic linkage to alcohol dependence on chromosome 4 and 11 from an autosome-wide scan in an American Indian population. *American Journal of Medical Genetics (Neuropsychiatric Genetics)* 81:216–221, 1998. **(58) Thibault, C.**; Lai, C.; Wilke, N.; et al. Expression profiling of neural cells reveals specific patterns of ethanol-responsive gene expression. *Molecular Pharmacology* 58(6):1593–1600, 2000. **(59) Mayfield, R.D.**; Lewohl, J.M.; Dodd, P.R.; et al. Patterns of gene expression are altered in the frontal and motor cortices of human alcoholics. *Journal of Neurochemistry* 81(4):802–813, 2002. **(60) Rimondini, R.**; Arlinde, C.; Sommer, W.; and Heilig, M. Long-lasting increase in voluntary ethanol consumption and transcriptional regulation in the rat brain after intermittent exposure to alcohol. *FASEB Journal* 16(1):27–35, 2002. **(61) Saito, M.**; Smiley, J.; Toth, R.; and Vadasz, C. Microarray analysis of gene expression in rat hippocampus after chronic ethanol treatment. *Neurochemical Research* 27(10):1221–1229, 2002. **(62) Liang, T.**; Spence, J.; Liu, L.; et al. Alpha-synuclein maps to a quantitative trait locus for alcohol preference and is differentially expressed in alcohol-preferring and nonpreferring rats. *Proceedings of the National Academy of Sciences of the U.S.A.* 100(8):4690–4695, 2003. **(63) Treadwell, J.A.**, and Singh, S.M. Microarray analysis of mouse brain gene expression following acute ethanol treatment. *Neurochemical Research* 29:357–369, 2004. **(64) Roberts, A.J.**, and Koob, G.F. The neurobiology of addiction. *Alcohol Health & Research World* 21(2):101–106, 1997. **(65) Hernandez-Avila, C.A.**; Oncken, C.; Van Kirk, J.; et al. Adrenocorticotropin and cortisol responses to a naloxone challenge and risk of alcoholism. *Biological Psychiatry* 51(8):652–658, 2002. **(66) Kreek, M.J.**, and Koob, G.F. Drug dependence: Stress and dysregulation of brain reward pathways. *Drug and Dependence* 51:23–47, 1998. **(67) Sayette, M.A.** An appraisal-disruption model of alcohol's effects on stress responses in social drinkers. *Psychological Bulletin* 114:459–476, 1993. **(68) Slawecki, C.J.**; Somes, C.; and Ehlers, C.L. Effects of chronic ethanol exposure on neurophysiological responses to corticotropin-releasing factor and neuropeptide Y. Alcohol and Alcoholism 34(3):289–299, 1999. **(69) Thiele, T.E.**; Miura, G.I.; Marsh, D.J.; et al. Neurobiological responses to ethanol in mutant mice lacking neuropeptide Y or the Y5 receptor. *Pharmacology, Biochemistry and Behavior* 67(4):683–691, 2000. **(70) Caberlotto, L.**; Thorsell, A.; Rimondini, R.; et al. Differential expression of NPY and its receptors in alcohol-preferring AA and alcohol-avoiding ANA rats. *Alcoholism: Clinical and Experimental Research* 25(11):1564–1569, 2001. **(71) Lappalainen, J.**; Kranzler, H.R.; Malison, R.; et al. A functional neuropeptide Y Leu7Pro polymorphism associated with alcohol dependence in a large population sample from the United States. *Archives of General Psychiatry* 59(9):825–831, 2002. **(72) Thorsell, A.**; Rimondini, R.; and Heilig, M. Blockade of central neuropeptide Y (NPY) Y2 receptors reduces ethanol self-administration in rats. *Neuroscience Letters* 332(1):1–4, 2002. **(73) Pandey, S.C.**; Carr, L.G.; Heilig, M.; et al. Neuropeptide Y and alcoholism: Genetic, molecular, and pharmacological evidence. *Alcoholism: Clinical and Experimental Research* 27(2):149–154, 2003. **(74) Nie, Z.**; Schweitzer, P.; Roberts, A.J.; et al. Ethanol augments GABAergic transmission in the central amygdale via CRF1 receptors. *Science* 303:1512–1514, 2004.

Full text of this publication is available on NIAAA's World Wide Web site at
http://www.niaaa.nih.gov

U.S. DEPARTMENT OF
HEALTH AND HUMAN SERVICES

NIAAA Publications Distribution Center
Attn.: *Alcohol Alert*
P.O. Box 10686
Rockville, MD 20849–0686

Official Business
Penalty for Private Use $300

PRSRT STD
POSTAGE AND FEES PAID
NIH/NIAAA
PERMIT NO. G-824