This Week in the Journal

JUNE 30, 2005

ORIGINAL ARTICLE

Adefovir Dipivoxil in HBeAg-Negative Chronic Hepatitis B

In the second phase of a randomized, placebo-controlled trial of adefovir dipivoxil for the treatment of hepatitis B e antigen (HBeAg)–negative chronic hepatitis B, patients who had been treated with adefovir during the initial 48 weeks of the trial were randomly assigned to be switched to placebo or to continue to receive adefovir. Patients who were switched to placebo lost the benefits that had been gained during the initial 48 weeks of treatment, and patients randomly assigned to continue adefovir therapy maintained a response. Resistance mutations developed in 6 percent of the patients treated with adefovir dipivoxil for 144 weeks.

SEE P. 2673; EDITORIAL, P. 2743; CME, P. 2766

ORIGINAL ARTICLE

Peginterferon Alfa-2a in HBeAg-Positive Chronic Hepatitis B

After 48 weeks of treatment and 24 weeks of follow-up, patients treated with peginterferon either alone or in combination with lamivudine were more likely to have HBeAg seroconversion than patients treated with lamivudine alone (32 percent and 27 percent vs. 19 percent) and more likely to have HBV DNA levels below 100,000 copies per milliliter (32 percent and 34 percent vs. 22 percent). A 48-week course of peginterferon alfa-2a is more effective than 48 weeks of lamivudine for HBeAg-positive chronic hepatitis B.

SEE P. 2682; EDITORIAL, P. 2743

ORIGINAL ARTICLE

Capecitabine as Adjuvant Treatment for Stage III Colon Cancer

The standard combination of intravenous fluorouracil plus leucovorin for adjuvant treatment of colon cancer was compared with the oral fluoropyrimidine capecitabine in almost 2000 patients with resected colon cancer. With disease-free survival as the primary end point, capecitabine was at least as effective as fluorouracil plus leucovorin. The oral drug had fewer side effects than the intravenous combination.

The finding that a single oral drug is at least as effective as intravenous chemotherapy promises, if confirmed, to be an important step forward in the management of resected colon cancer.

SEE P. 2696; EDITORIAL, P. 2746

ORIGINAL ARTICLE

Daclizumab to Prevent Rejection after Heart Transplantation

Daclizumab is a humanized monoclonal antibody directed against the interleukin-2 receptor and thereby inhibits T-cell proliferation. In this clinical trial, daclizumab reduced the risk of cellular rejection in heart-transplant recipients when it was added to a regimen of cyclosporine, mycophenolate mofetil, and corticosteroids. However, when daclizumab was given concurrently with cytolytic therapy, there was a worrisome increase in the rate of death from infection.

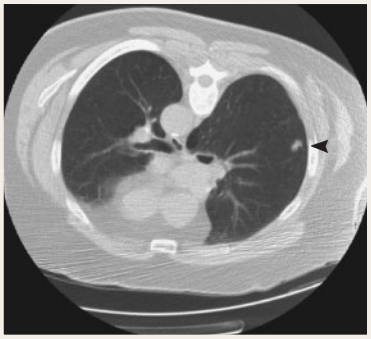
SEE P. 2705; EDITORIAL, P. 2749

CLINICAL PRACTICE

Lung Cancer Screening

A 60-year-old woman who quit smoking 20 years earlier comes for a routine visit. She previously smoked one pack of cigarettes a day for 10 years. Her medical history is otherwise unremarkable. Her husband smoked one pack of cigarettes per day for at least 30 years but stopped smoking a decade ago. She asks whether she and her husband should undergo computed tomographic scanning to screen for lung cancer. What do you advise?

SEE P. 2714; CME, P. 2765



DRUG THERAPY

γ-Hydroxybutyric Acid

The short-chain fatty acid γ-hydroxybutyric acid (GHB), which is synthesized as an analogue of γ-aminobutyric acid (GABA) that would cross the blood–brain barrier, has found limited clinical use as an anesthetic agent and as treatment for narcolepsy and alcoholism. However, during the past decade, GHB has emerged as a major recreational drug in the United States. This review article discusses the mechanisms of action and presents an approach to the treatment of overdose, abuse, and addiction.

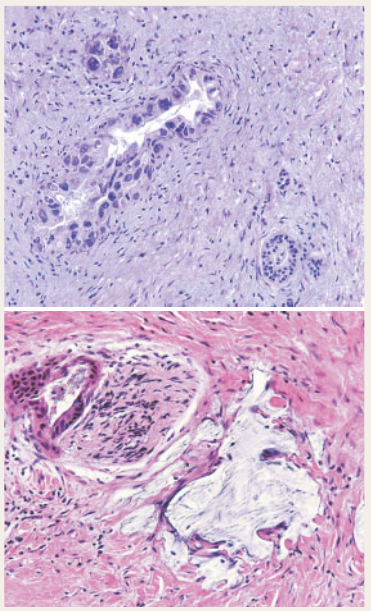
SEE P. 2721; PERSPECTIVE, P. 2671; CME, P. 2767

CASE RECORDS OF THE MASSACHUSETTS GENERAL HOSPITAL

A Man with Locally Advanced Pancreatic Cancer

A 58-year-old man was being evaluated for an abdominal aortic aneurysm when a pancreatic mass was incidentally found; in retrospect, he had experienced vague abdominal discomfort and weight loss for several months. Further evaluation disclosed a pancreatic adenocarcinoma that encased the superior mesenteric and portal veins. Treatment options are discussed.

SEE P. 2734



Genetic Justice

Mark A. Rothstein, J.D.

An interview with Professor Rothstein can be heard at www.nejm.org.

On December 21, 2004, Brandon Moon was released from prison in El Paso, Texas, after having served 16 years of a 75-year sentence for three counts of aggravated sexual assault. Moon, who was 43 years of age at the time of his release, had been convicted in 1988 on the testimony of the three victims, who had had only a fleeting or partial view of their assailant. In 2004, after undergoing DNA testing, Moon was excluded as the contributor of the DNA collected after all three rapes. As a result, Moon became the 154th person in the United States to be exonerated on the basis of DNA evidence that came to light after the person was convicted for a crime.[1]

The Innocence Project, founded in 1992 by Barry Scheck and Peter Neufeld at the Benjamin N. Cardozo School of Law in New York, pioneered the use of forensic DNA testing to provide scientific evidence of guilt or innocence after conviction. As described by Gill in another Perspective article in this issue of the *Journal* (pages 2669–2671), the technology has improved in the past decade, but the basic purpose of forensic DNA testing has not changed. The success of the Innocence Project led to the establishment of similar projects throughout the country, staffed by lawyers and law students working pro bono. Other persons and similar organizations not affiliated with the Innocence Project also use DNA evidence to free wrongfully convicted prisoners.

Cases such as Brandon Moon's elicit mixed emotions. One cannot help feeling sadness at the miscarriage of justice that resulted in the incarceration of an innocent man for 16 years while the perpetrator escaped justice. Furthermore, the passage of time makes the discovery and conviction of the perpetrator unlikely, given the cold trail of evidence and the statute of limitations. Nevertheless, one feels relief that at least some degree of justice has finally been achieved, admiration for the volunteer and public-interest lawyers who worked long hours — in some instances years — to free a wrongfully convicted client, and appreciation of the forensic DNA technology, which can provide compelling new evidence that may cast doubt on past convictions.

The phenomenon of postconviction exonerations based on DNA evidence must be put into perspective. At the end of 2003, more than 2 million people were incarcerated in the United States, and of those convicted, those exonerated during the past decade make up a tiny percentage of the total inmate population. Nonetheless, the exonerations raise three fundamental questions about the U.S. criminal justice system.

First, do these cases of wrongful conviction represent the tip of the iceberg, indicating the existence of deeper structural problems in the criminal justice system? As a study by Gross et al. revealed,[2] most of the exonerations that have occurred since 1989 involved faulty eyewitness testimony, as in the case of Brandon Moon, coerced or false confessions, or perjurious testimony by prison inmates. Other cases involved poor crime-scene processing or poor evaluation of evidence by forensic laboratory workers, ineffective defense counsel, or even police and prosecutorial misconduct. Studies of errors in these cases often indicate the presence of systemic problems in forensics, law enforcement, or the criminal courts.[3] For example, officials in several states ordered the review of hundreds of convictions after they determined that state and local crime laboratories might have made numerous errors in handling and testing evidence or, worse, might have deliberately falsified the results or interpretations of forensic testing.

Second, aside from those already exonerated, how many other innocent people have been convict-

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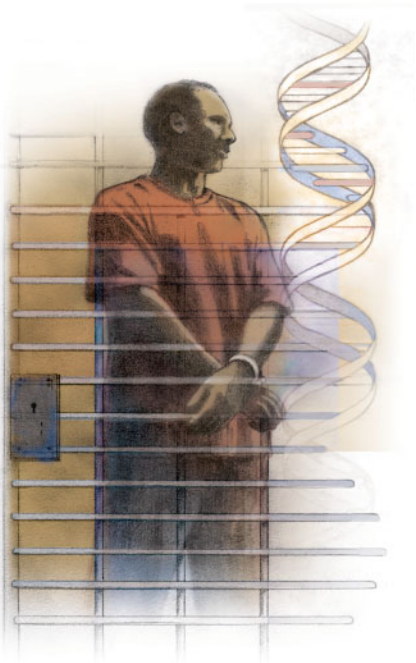
ed of serious crimes and have served, or are now serving, long prison sentences or are facing execution in one of the 38 states that permit capital punishment? Although there may not always be crime-scene evidence available for DNA testing, new federal legislation should make DNA testing more widely available to those who have been convicted. On October 30, 2004, President George W. Bush signed the Justice for All Act of 2004, which grants any inmate convicted of a federal crime the right to petition a federal court for DNA testing to support a claim of innocence. The law provides funding to the states to preserve evidence and to make DNA testing available to those who have been convicted by the state as well. It also increases the financial compensation of wrongfully convicted federal prisoners. Under Texas law, Brandon Moon is eligible for up to $25,000 for each year he was wrongfully imprisoned. Many states, however, have no provision for compensation.



Third, what is the proper role of DNA evidence in criminal investigations? DNA has the power not only to exculpate but also to inculpate. Forensic DNA analysis was first used in the United Kingdom in 1985 to solve two related sexual homicides. The use of such evidence soon became internationally accepted as a method for linking suspects with crime-scene evidence. In 1990, the Federal Bureau of Investigation established a series of federal–state forensic DNA data banks, called the Combined DNA Index System, or CODIS. In every state, certain convicted felons are required to submit a sample of DNA to be typed and the profile entered into a computerized data bank. Law-enforcement officials can then compare the DNA profiles obtained from evidence from a crime scene with all the DNA profiles in the local, state, or national system.

The power of DNA analysis in the investigation of crimes has led many law-enforcement officials to use DNA profiling in increasingly aggressive and controversial ways. One example is the DNA "dragnet," which typically involves a request by police that all men in an area where a sex crime has been committed "voluntarily" submit a sample of their DNA. Although such dragnets have had only limited success in helping to solve crimes, they have caused concern about the infringement of civil liberties. Critics argue that it is unfair to regard as suspects all men who decline to submit a sample and that the DNA profiles of those who volunteer to submit a sample should not be entered into the state DNA data bank.

Another controversial practice is the expansion of the scope of the DNA data banks. Many states have extended the requirement of submission of DNA by convicted felons to submission of samples by convicted misdemeanants, four states authorize the collection of samples from persons when they are arrested, and some forensic experts have proposed the collection of DNA from everyone in the country. These practices and proposals raise the question of whether the benefits to law enforcement of expanding the use of forensic DNA testing are worth the cost in civil liberties.[4] Meanwhile, the exonerations continue to occur at a disquieting pace.

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2. Gross SR, Jacoby K, Matheson DJ, Montgomery N, Patil S. Exonerations in the United States, 1989 through 2003. (Accessed June 9, 2005, at http://www.law.umich.edu/NewsAndInfo/exonerations-in-US.pdf.)
3. Bieber FR, Lazer D. Lessons learned from a miscarriage of justice. Boston Globe. April 12, 2003.
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DNA as Evidence — The Technology of Identification

Peter Gill, Ph.D.

In 1983, in a village near Leicester, England, a local girl named Lynda Mann was found raped and murdered. Three years later, a second girl, Dawn Ashworth, was found dead under similar circumstances. The similarities between the two cases led the police to believe that the same person had committed both crimes. After extensive inquiries, an arrest was made. The suspect confessed to the murder of Lynda Mann but denied having killed Dawn Ashworth. Convinced that they had the right man, the police approached Sir Alec Jeffreys, a professor of genetics at the University of Leicester, with a request to conduct tests using a new method that he called "DNA fingerprinting," which had not yet been used in a real case.

The results were surprising: the suspect was exonerated, and the DNA profiles in the two murder cases were the same, indicating that a single, unknown person had committed both crimes. This finding led to the screening of all 5000 men in the area, using both conventional blood-group methods and DNA testing. The screening failed to identify a suspect — because, as it turned out, the perpetrator, Colin Pitchfork, had paid a colleague to give a DNA sample in his place. When the colleague was overheard bragging to a friend about the incident, Pitchfork was quickly apprehended, analysis of a DNA sample confirmed his guilt in both murders, and he was duly convicted in 1988.[1]

Thus, the first criminal case in which DNA was used provided a vivid demonstration of the method's potential — not only for convicting the guilty but also for exonerating the innocent. It also demonstrated for the first time that a DNA fingerprint could be used to find a perpetrator from within a population.

In 1985, a year after the development of DNA fingerprinting, the polymerase chain reaction (PCR) was discovered.[2] The discovery would revolutionize the field of molecular biology, though the method would not come into routine use in forensic cases until the early 1990s, since new platforms and biochemical tools were needed in order to take full advantage of the potential of PCR. In particular, new automation technology was key, and the advent of the automated fluorescent DNA sequencer in the early 1990s was a major step forward. More generally, forensic DNA analysis has benefited substantially from the Human Genome Project, for the genome could be sequenced only with automated equipment that permitted high-throughput processing. Because forensic science could use the same equipment and biochemical tools that gene sequencing used, new methods were rapidly developed in the early 1990s that would have been considered impossible just a few years earlier.

Perhaps the best example of this adjunct benefit of genomics was the development of national DNA databases. Since its inception in 1995, the National DNA Database for England and Wales has expanded to include more than 2.75 million reference DNA profiles, against which all specimens obtained from the scene of a crime ("crime stains") are routinely compared.[3] The likelihood that a match will be found is approximately 30 percent. Many other countries have since followed suit, and the benefits of such databases are considerable, since persons who commit serious crimes such as murder usually have a previous criminal record. The United Kingdom's policy permits the collection of DNA profiles from all convicted criminals, as well as from anyone suspected of committing a crime that could lead to a prison sentence — and the law allows authorities to retain the DNA profile even if the suspect is found innocent. Consequently, persons who later commit more crimes can be identified and apprehended quickly.

DNA-profiling technology takes several forms. Short tandem repeats (STRs) are universally used as the workhorses of national DNA databases. The length of these stretches of DNA varies from person to person, making them useful as genetic identifiers similar to the bar codes that identify items for sale (see diagram). In the United States, 13 STRs are amplified together in a multiplex PCR. When a complete DNA profile has been obtained, the probability of a match with a randomly chosen person is less than 1 in 1 trillion. Profiling efforts in European countries apply the same principles but generally

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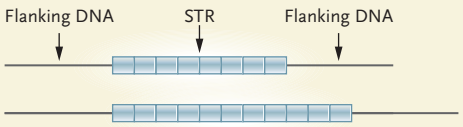

Schematic Diagram of a Short-Tandem-Repeat Marker.
Interspersed throughout the human genome are stretches of DNA comprising short blocks of repeated sequence known as short tandem repeats (STRs). Typically, "forensic" STRs include four bases, arranged in a specific order. Each STR site that is used for forensic analysis has an internationally recognized identifying name. Here, the STR locus is called HUMTH01, and each block consists of the DNA bases thymine, cytosine, adenine, and guanine. In this example, the person carries 8 repeats on one chromosome and 11 repeats on the other (hence, HUMTH01 8.11). The number of repeats carried by each allele is determined by amplifying the DNA with the polymerase chain reaction and then measuring the size of the amplification products. A total of 3 to 14 repeats may be observed at this particular site. Ten separate STR sites, each denoted by two digits, are used in analyses performed in the United Kingdom. Consequently, any person may be defined by a code comprising 20 digits, which can easily be stored in a searchable computer database. The rarity of each genetic combination is estimated on the basis of population surveys and is used to calculate the strength of the evidence, estimated as the likelihood that a random (unrelated) person has the same DNA profile as a suspect.

use fewer STRs; for example, in the United Kingdom, 10 STRs are used, resulting in a match probability of less than 1 in 1 billion.

With the use of conventional STR-based methods, it is possible to analyze small samples that consist of approximately 60 cells; sensitivity can be improved simply by increasing the number of PCR cycles. Theoretically, just a single cell is required to obtain a result, although more are usually analyzed; analysis of such small samples is known as low-copy-number DNA profiling.[4] Caution is required when using this technique, however, because when sensitivity is increased, the potential for analyzing contaminant DNA from a person unrelated to the crime (especially, with contemporary DNA, from a crime-scene investigator or a scientist working on the case) increases as well. Moreover, DNA profiling does not tell us when the sample was deposited, so considerable attention must be paid to the use of ultraclean laboratories and handling with DNA-free materials at all stages of the analysis. The use of low-copy-number DNA profiling has expanded the range of evidentiary samples that can be analyzed, permitting the recovery of a DNA profile even from the skin cells found in a fingerprint.

Low-copy-number analysis does not work in every instance, however. Certain types of evidence, such as hair shafts, have little or no nuclear DNA, and STR analysis often fails with highly degraded materials such as bone.

Fortunately, mitochondrial DNA (mtDNA) may also be used. This type of DNA is found in the cytoplasm of the cell, where there are high copy numbers. However, mtDNA is inherited in the vast majority of cases from the mother, is much smaller than nuclear DNA, and lacks the discriminatory power of STRs; moreover, data derived from mtDNA are incompatible with those in national DNA databases. Hence, mtDNA offers both advantages and disadvantages to the investigator. The main advantage is that it can give results when all other techniques fail — which is why mtDNA analysis is the preferred method for ancient samples. Because of its nearly exclusive inheritance through the female line, mtDNA can help to solve historical mysteries revolving around maternal lineage. Perhaps the best example is the 1994 analysis of remains thought to be those of the Russian royal family, the Romanovs, which revealed mtDNA that matched samples taken from living descendants of the maternal line, such as Prince Philip of Britain.

The male counterpart of mtDNA is the Y chromosome. Unlike mtDNA, it resides in the nucleus and does not have a high cellular copy number. Both Y-specific STRs and single-nucleotide polymorphisms (SNPs) — single-base variants that occur at specific positions in the genome — can be used to characterize the Y-chromosome haplotype. Sometimes in specimens combining material from persons of both sexes, the male component can be completely masked by the female DNA, rendering identification impossible. Under these circumstances, analysis of the Y chromosome is warranted. However, the discriminating power of such analysis is low (as in the case of mtDNA), and the results are incompatible with STR databases. But Y-chromosome DNA is useful in the investigation of historical mysteries involving paternal lineages. The use of mtDNA and Y-chromosome DNA can yield population-specific genetic signatures that offer investigative leads in unsolved cases in which there are no firm suspects. The Y chromosome might also be used as a marker of family surname; people with unusual surnames often have a common genealogy.

The World Trade Center disaster has illustrated the importance of having the entire gamut of techniques available in order to maximize the chances of identifying victims: STRs, mtDNA, and SNPs were all used. More than 19,000 samples of human remains were recovered from more than 2700 victims, and many of these samples were highly degraded, having been subjected to considerable pressure and fire.[5] To date, more than 1500 victims have been positively identified — an undertaking that would have been impossible without DNA technology. DNA profiling is also playing a major role in the identification of victims of the recent Asian tsunami.

The efforts to identify bodies after these disasters underscore the importance of having national DNA databases that can accommodate new types of data, without which there is a danger of becoming locked into old technology. The best way to facilitate change (given that the overriding concern is the maintenance of compatibility) is an important and pressing issue.

Although it is difficult to make predictions, I believe that the use of STRs will probably remain the system of choice for the foreseeable future, because of its advantages over the use of SNPs, including a relative ease of interpretation. SNPs may find a special niche in the analysis of highly degraded material and in the context of massive disasters. Undoubtedly, the usefulness of both methods will benefit from new biochemical tools and new platforms such as DNA microarrays. Automation, miniaturization, and expert systems will all have critical roles in advancing forensic analysis in the coming years, just as they did during the sequencing of the human genome.

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2. Saiki RK, Scharf S, Faloona F, et al. Enzymatic amplification of beta-globin genomic sequences and restriction site analysis for diagnosis of sickle cell anemia. Science 1985;230:1350-4.
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4. Gill P, Whitaker J, Flaxman C, Brown N, Buckleton J. An investigation of the rigor of interpretation rules for STRs derived from less than 100 pg of DNA. Forensic Sci Int 2000;112:17-40.
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Close Calls with Club Drugs

Cynthia G. McGinn, M.D.

Related article, page 2721

Around midnight one night in 1999, I was radioed by the EMTs: they were bringing in two "drunk" college students. This was hardly an uncommon occurrence. Many a college student has been mortified to wake up in the emergency room looking into the eyes of a college administrator who knew that a stressful semester or a campus party could spell danger. Widespread alcohol abuse on college campuses and its sometimes fatal consequences have long raised alarm, and the signs and symptoms of cocaine or marijuana abuse are commonly known.

But there was something different about these students. Apparently healthy college men, they had been drinking at a party when the girlfriend of one noticed that they had disappeared. Worried, she went looking for them. She found them unconscious in the basement of the building, and she couldn't wake them. She might have left them there to sleep it off, but she called for help instead.

The EMTs who responded thought that the students were simply drunk, but the students' level of consciousness decreased alarmingly en route to the hospital. By the time they arrived, both young men were deeply comatose — unresponsive to all stimuli, including pain. As the EMTs were filling me in, I noticed that one of the patients had stopped breathing. Within a few minutes, both of them had to be intubated.

Had these men been left in the basement, they would have been brain-dead within about six minutes after they had stopped breathing, and their hearts would have stopped shortly thereafter. As it was, both required mechanical ventilation for the rest of the night.

What had these students taken? They didn't smell of alcohol. There were no track marks on their arms. There was no evidence of trauma or other disease.

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In fact, what was so alarming was how healthy they looked — they had simply stopped breathing.

None of their friends in the waiting room could tell me what drugs were involved, although there were murmurings of "G," "liquid E," and "vitamins" — street names for γ-hydroxybutyric acid (GHB), 3,4-methylenedioxymethamphetamine (MDMA, or ecstasy), and methamphetamine. Blood and urine drug screens were negative except for alcohol, but the level of ethanol was not high enough to cause respiratory arrest. These patients were not alcoholics, not IV drug abusers. They were student athletes nearing graduation from college with the whole world ahead of them.

As an emergency physician, I had watched the evolution — and treated the clinical consequences — of the abuse of legal and illegal drugs from the hallucinogens of the 1960s to the stimulants and opiates of the 1970s and the cocaine of the 1980s. Now I was encountering the difficulties of diagnosing abuse of the so-called club drugs — which, unfortunately, many people still erroneously believe to be harmless.

The most commonly used club drugs are GHB, MDMA, flunitrazepam, and ketamine. Instructions for making them with the use of chemicals found in household products, over-the-counter medications, and prescription drugs can be found on Internet sites, rendering them accessible and inexpensive. They are also potentially deadly.

In this case, I believe that the culprit was GHB (discussed by Snead and Gibson in this issue of the *Journal*, pages 2721–2732). Clear, odorless, tasteless, and thought to be an aphrodisiac and an amnesic, GHB unfortunately makes a perfect date-rape drug. But the euphoria that begins 20 to 30 minutes after ingestion can quickly evolve into more toxic effects, including dizziness, nausea and vomiting, myoclonic jerks, confusion, agitation, hallucination, and seizure. In combination with alcohol, GHB can cause decreased respiratory drive, coma, and death. There is currently no metabolite that can be measured on routine toxicology screens, so although some laboratories can now test for GHB, the results are not available on an emergency basis.

I don't know whether these students are aware of how close they came to dying. Because they arrived at the hospital when they did, they were able to walk out the following day, remembering little or nothing of the entire event. Given their friends' comments, their profound respiratory depression, the duration of the effects, and the subsequent amnesia, my guess is that they combined GHB with alcohol. But the amnesia made it impossible to get an accurate history.

Clearly, alcohol, tobacco, and marijuana are not the only temptations out there for our children. Club drugs can be taken in pill or liquid form — no snorting, smoking, or needles required. Given the deceptively innocuous qualities of these drugs, it is important to raise awareness of their potentially devastating effects.

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Long-Term Therapy with Adefovir Dipivoxil for HBeAg-Negative Chronic Hepatitis B

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ABSTRACT

BACKGROUND

Treatment with adefovir dipivoxil for 48 weeks resulted in histologic, virologic, and biochemical improvement in patients with hepatitis B e antigen (HBeAg)–negative chronic hepatitis B. We evaluated the effect of continued therapy as compared with cessation of therapy.

METHODS

One hundred eighty-five HBeAg-negative patients with chronic hepatitis B were assigned to receive 10 mg of adefovir dipivoxil or placebo once daily for 48 weeks (ratio, 2:1). After week 48, patients receiving adefovir dipivoxil were again randomly assigned either to receive an additional 48 weeks of the drug or to switch to placebo. Patients originally assigned to placebo were switched to adefovir dipivoxil. Patients treated with adefovir dipivoxil during weeks 49 through 96 were subsequently offered continued therapy. The primary end points were changes in hepatitis B virus (HBV) DNA and alanine aminotransferase levels.

RESULTS

Treatment with adefovir dipivoxil resulted in a median decrease in serum HBV DNA of 3.47 log copies per milliliter (on a base-10 scale) at 96 weeks and 3.63 log copies per milliliter at 144 weeks. HBV DNA levels were less than 1000 copies per milliliter in 71 percent of patients at week 96 and 79 percent at week 144. In the majority of patients who were switched from adefovir dipivoxil to placebo, the benefit of treatment was lost (median change in HBV DNA levels from baseline, –1.09 log copies per milliliter; only 8 percent of patients had levels below 1000 copies per milliliter at week 96). Side effects during weeks 49 through 144 were similar to those during the initial 48 weeks. Resistance mutations rtN236T and rtA181V were identified in 5.9 percent of patients after 144 weeks.

CONCLUSIONS

In patients with HBeAg-negative chronic hepatitis B, the benefits achieved from 48 weeks of adefovir dipivoxil were lost when treatment was discontinued. In patients treated for 144 weeks, benefits were maintained, with infrequent emergence of viral resistance.

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*Other members of the Adefovir Dipivoxil 438 Study Group are listed in the Appendix.

N Engl J Med 2005;352:2673-81.

An estimated 400 million people worldwide are chronically infected with hepatitis B virus (HBV). One million die each year from complications of infection, including cirrhosis, hepatocellular carcinoma, or both.[1] Hepatitis B e antigen (HBeAg)–negative chronic hepatitis B represents a late phase in the course of HBV infection.[2] Mutations in the precore promoter regions, core promoter regions, or both, which prevent the formation of HBeAg, are selected during or after HBeAg loss and seroconversion to antibody to HBeAg (anti-HBe). HBeAg-negative chronic hepatitis B infection is characterized by intermittent periods of exacerbation and quiescence. It frequently follows an aggressive disease course, with low rates of spontaneous recovery.[2-4] Epidemiologic data suggest that the median prevalence of HBeAg-negative chronic hepatitis B varies considerably, ranging from 14 percent in the United States and Northern Europe to more than 33 percent in the Mediterranean area, with an increasing prevalence worldwide.[3]

Current therapeutic options include treatment with interferon alfa, lamivudine, and adefovir dipivoxil. The goal of treatment is HBV DNA suppression, normalization of alanine aminotransferase levels, and reduction in liver necroinflammation. Longer-term objectives include the prevention of cirrhosis, end-stage liver disease, hepatocellular carcinoma, or all of these. It is unknown whether treatment can be stopped or whether long-term therapy is needed.[5]

A one-year regimen of lamivudine has been shown to achieve virologic and biochemical responses.[6-8] However, continued therapy results in resistance in approximately 20 percent of patients per year in most studies.[9] Interferon alfa and pegylated interferon have also shown efficacy; however, the durability of the response after the cessation of treatment is uncertain.[8,10-12]

In an earlier 48-week, placebo-controlled phase of this study, adefovir dipivoxil, as compared with placebo, resulted in significant histologic improvement (in 64 percent of patients vs. 33 percent, respectively), biochemical improvement (normalization of alanine aminotransferase levels, 72 percent vs. 29 percent), and virologic improvement (median reduction in HBV DNA, 3.91 log copies per milliliter [on a base-10 scale] vs. 1.35 log copies per milliliter); no resistance developed in patients treated with adefovir dipivoxil.[13,14] Here, we report the outcomes associated with stopping or continuing treatment with adefovir dipivoxil during a second 48-week randomized, controlled period; we also provide long-term data on treatment with this agent over 144 weeks.

METHODS

Between January 10 and June 7, 2000, 185 patients were enrolled and 184 treated in this international, multicenter, prospective, double-blind, placebo-controlled trial. Patients were randomly assigned to receive 10 mg of adefovir dipivoxil or placebo orally once daily for 48 weeks in a ratio of 2:1. After week 48, 123 patients who had been assigned to adefovir dipivoxil were randomly assigned either to continue with adefovir dipivoxil (the continued-adefovir group; 80 patients) or to switch to placebo (the adefovir–placebo group; 40 patients) for an additional 48 weeks. Three patients did not receive treatment during the second 48 weeks. Of the 61 patients who initially received placebo, 60 received adefovir dipivoxil in this second 48-week period (the placebo–adefovir group). Patients who received adefovir dipivoxil in the second 48 weeks were eligible to continue treatment until week 240.

Liver biopsies were required within six months or immediately before treatment and at week 48. A liver biopsy was optional at weeks 96 and 144. Serum HBV DNA and alanine aminotransferase were measured and blood chemistry assessments were conducted every 4 weeks until week 96 and then every 12 weeks.

Clinical data were monitored and entered into a database by Quintiles, a contract research organization. A central reference laboratory (Covance Laboratories) assessed all laboratory data. Gilead Sciences held all data and conducted the statistical analyses. The academic investigators had full access to the data. Each author contributed to the study design, the interpretation of the results, and the drafts and revisions of the manuscript; all authors had input into and approved the final manuscript. Drs. Hadziyannis, Tassopoulos, Heathcote, Chang, Kitis, Rizzetto, Marcellin, Lim, and Goodman vouch for the veracity and completeness of the data and the data analysis. The study was conducted in compliance with the 1975 Declaration of Helsinki and approved by local regulatory bodies. All patients provided written informed consent.

PATIENTS

Full inclusion criteria have been described previously.[13] Key criteria included HBeAg-negative, anti-HBeAg–positive status and the presence of compensated liver disease, detectable hepatitis B surface antigen (HBsAg) for at least six months, a serum HBV DNA level of at least 100,000 copies per milliliter on polymerase chain reaction (as measured with the Roche Amplicor Monitor; lower limit of detection, 1000 copies per milliliter, previously 400 copies per milliliter[13]), and a serum alanine aminotransferase level between 1.5 and 15 times the upper limit of normal.

ASSESSMENT OF EFFICACY

Primary efficacy end points were the changes from baseline in serum HBV DNA and alanine aminotransferase levels at week 96. Other efficacy end points included the percentage of patients in whom HBV DNA fell below the limit of detection of the assay, the percentage in whom alanine aminotransferase levels returned to normal, and the percentage in whom there was HBsAg seroconversion (i.e., loss of HBsAg and gain of antibody to HBsAg). End points were evaluated at weeks 96 and 144. In a subgroup of patients, histologic features of liver specimens were evaluated by an independent histopathologist (with the use of both Knodell and Ishak scoring systems, which evaluate necroinflammation and liver fibrosis on scales of 0 to 22 [Knodell] and 0 to 24 [Ishak], with higher scores indicating greater severity) who was blinded to patients' treatment assignments and the date on which the biopsy specimens were obtained.[15] Ranked assessment of inflammation and fibrosis was also performed, with severity delineated as improved, no change, or worse as compared with the baseline scores.

ASSESSMENT OF SAFETY

Safety was assessed with the use of laboratory tests and by the reported occurrence of adverse events every 4 weeks for the first 96 weeks and then every 12 weeks. All patients who received at least one dose of adefovir dipivoxil were included in the safety analysis.

RESISTANCE SURVEILLANCE

Genotypic analyses of HBV DNA polymerase mutations were performed on serum samples from patients with HBV DNA levels of more than 1000 copies per milliliter in the 123, 134, and 70 patients who received adefovir dipivoxil through weeks 48, 96, and 144, respectively. The HBV reverse transcriptase (rt) domain (amino acids rt1 to rt344) was sequenced. Sequences at baseline and after baseline were aligned with the use of the MegAlign program (DNAStar).

STATISTICAL ANALYSIS

Statistical analyses included all patients who received at least one dose of the study drug in the second 48 weeks. All HBV DNA values less than the lower limit of detection (1000 copies per milliliter) were assigned a value of 999 copies per milliliter. All tests for significance and resulting P values were two-sided, with a level of significance of 0.05.

RESULTS

CHARACTERISTICS OF THE PATIENTS

A total of 180 patients were randomly assigned to receive treatment in the second 48 weeks of the study. Of these patients, 79 continued to receive adefovir dipivoxil, 40 initially assigned to adefovir dipivoxil received placebo, and 60 were switched from placebo to adefovir dipivoxil. One patient who had been randomly assigned to the adefovir dipivoxil group withdrew from the study before taking medication in the second 48 weeks. At week 96, 125 patients continued to receive adefovir dipivoxil — 70 in the continued-adefovir group and 55 in the placebo–adefovir group. Data are reported up to week 144 for patients who received adefovir dipivoxil from baseline. Baseline demographic characteristics and those related to hepatitis B infection were not statistically different among the three groups (Table 1).

VIROLOGIC RESPONSE

At week 96, serum HBV DNA levels had decreased by a median of 3.47 log copies per milliliter in the continued-adefovir group, as compared with 1.09 log copies per milliliter in the adefovir–placebo group ($P<0.001$) (Table 2). Undetectable levels of HBV DNA were reported in 71 percent of patients in the continued-adefovir group, as compared with 76 percent and 8 percent, respectively, in the placebo–adefovir and adefovir–placebo groups. There was a rapid reduction in serum HBV DNA levels in patients in the continued-adefovir group, with persistent reductions up to week 96. In contrast, the adefovir–placebo group had a rebound in serum HBV DNA levels, with a return to baseline levels within four weeks of the discontinuation of adefovir dipivoxil in the majority of patients (Fig. 1).

Table 1. Baseline Characteristics of the Patients.

Characteristic	Continued-Adefovir Group (N=79)	Adefovir–Placebo Group (N=40)	Placebo–Adefovir Group (N=60)
Age — yr			
Mean ±SD	46±10	46±9.9	46±10.2
Median	47	47	46
Range	26–65	18–64	22–65
Male sex — no. (%)	65 (82)	33 (82)	50 (83)
Race or ethnic background — no. (%)*			
White	55 (70)	26 (65)	39 (65)
Black	4 (5)	1 (2)	1 (2)
Asian	20 (25)	13 (32)	20 (33)
Weight — kg			
Mean ±SD	75±12.2	77±10.0	74±15.2
Median	75	76	74
Range	50–111	60–105	46–135
HBV DNA level — log copies/ml†			
Mean ±SD	6.87±0.86	7.03±0.78	6.91±0.94
Median	7.07	7.16	7.05
Range	3.67–8.42	5.28–8.77	4.42–8.45
Alanine aminotransferase level — IU/liter			
Mean ±SD	140±120.7	152±140.4	149±196.8
Median	98	86	99
Range	24–742	45–657	29–1459
≤ULN — no. (%)‡	7 (9)	0	2 (3)
>ULN — no. (%)	72 (91)	40 (100)	58 (97)
Positive for HBsAg — no. (%)	79 (100)	40 (100)	60 (100)
Prior medications for HBV — no. (%)§			
Interferon alfa	30 (38)	18 (45)	27 (45)
Lamivudine¶	7 (9)	3 (8)	4 (7)

* Race was self-assigned.
† Values were log-transformed with the use of a base-10 scale.
‡ ULN denotes upper limit of the normal range; for men, the level was 43 IU per liter, and for women 34 IU per liter.
§ Some patients had received more than one type of medication.
¶ Lamivudine had been used less than 12 weeks previously in these patients.

In the patients who continued adefovir dipivoxil to week 144, HBV DNA levels remained suppressed at week 144 (median reduction in HBV DNA from baseline, 3.63 log copies per milliliter). In 79 percent of these patients, serum HBV DNA levels were less than 1000 copies per milliliter at week 144.

SEROLOGIC RESPONSE

HBsAg seroconversion (i.e., the loss of HBsAg and gain of anti-HBs) occurred in two patients, one in the continued-adefovir group at week 72 and one in the placebo–adefovir group at week 68 (approximately 20 weeks after the start of adefovir dipivoxil).

BIOCHEMICAL RESPONSE

Median reductions in serum alanine aminotransferase levels at week 96 were 59 IU per liter in the continued-adefovir group, as compared with 29.5 IU per liter in the adefovir–placebo group (P=0.01), and 79.5 IU per liter in the placebo–adefovir group (Table 2). At week 96, a return to normal levels of alanine aminotransferase (upper limit of normal, 37 IU per liter for women and 43 IU per liter for men) was achieved in 73 percent of patients in the continued-adefovir group, 80 percent in the placebo–adefovir group, and 32 percent in the adefovir–placebo group. Patients in the continued-adefovir group had sustained suppression of alanine aminotransferase throughout the study. In contrast, alanine aminotransferase levels returned to pretreatment values or higher in the majority of patients in the adefovir–placebo group within eight weeks of stopping therapy. In 32.5 percent of patients, alanine aminotransferase levels rose sharply — to more than 10 times the upper limit of normal — before returning to baseline levels. None of these elevations were associated with clinical hepatic decompensation. In the patients who continued to receive adefovir dipivoxil to week 144, alanine aminotransferase levels remained suppressed, with normalization in 69 percent of patients.

HISTOLOGIC RESPONSE

A subgroup of 47 patients underwent liver biopsy at week 96. Baseline demographic and disease characteristics of these patients were similar to those of patients in the overall study population. Patients in the continued-adefovir group had a mean reduction of 4.7 points from baseline in the overall Knodell score at week 96 (Table 3) (a mean reduction of 4.4 points at week 48). Among patients in the placebo–adefovir group, there was a mean increase of 0.9 points from baseline at week 48, with a subsequent reduction after the crossover to adefovir dipivoxil of 2.4 points from baseline at week 96, a reversal of the increase observed at week 48. In the adefovir–placebo group, there was a loss of improvement at week 48, with a median reduction of 1 point from baseline at week 96.

Table 2. Virologic and Biochemical Responses at Weeks 96 and 144.*

Response	Continued-Adefovir Group		Adefovir–Placebo Group	Placebo–Adefovir Group
	Week 96 (N=79)	Week 144 (N=70)	Week 96 (N=40)	Week 96 (N=60)
Virologic				
No. of patients assessed	70	67	38	49
Change in serum HBV DNA level — log copies/ml				
Mean ±SD	−3.35±1.18	−3.42±1.27	−1.34±1.24	−3.71±1.05
Median	−3.47	−3.63	−1.09	−3.85
Interquartile range	−4.20 to −2.59	−4.23 to −3.11	−2.19 to −0.40	−4.31 to −3.18
Range	−5.42 to −0.27	−5.42 to −1.18	−4.16 to 0.87	−5.35 to 0.44
P value†	—	NA	<0.001	0.12
Serum HBV DNA level <1000 log copies/ml — no./total no. (%)	50/70 (71)	53/67 (79)	3/38 (8)	37/49 (76)
P value‡	—	NA	<0.001	0.68
Biochemical				
No. of patients assessed	71	67	38	50
Change in serum (alanine aminotransferase) level — IU/liter				
Mean ±SD	−98±118.4	−97±120.13	−63±131.0	−130±213.2
Median	−59	−54	−29.5	−79.5
Interquartile range	−115 to −27	−121 to −28	−68 to 18	−134 to −46
Range	−717 to 51	−707 to 56	−548 to 93	−1429 to 5
P value†	—	NA	0.01	0.21
Normalization of alanine aminotransferase level — no./total no. (%)§	47/64 (73)	43/62 (69)	12/38 (32)	40/50 (80)
P value‡	—	NA	<0.001	0.51

* Negative values indicate a decrease, and positive values an increase. NA denotes not applicable.
† P values were calculated with the use of the Wilcoxon rank-sum test for the comparison between continued treatment with adefovir dipivoxil and the crossover from adefovir dipivoxil to placebo and for the comparison between continued treatment with adefovir dipivoxil and the crossover from placebo to adefovir dipivoxil at week 96. All P values are two-sided, with a level of 0.05 indicating statistical significance; there were no adjustments for multiple comparisons.
‡ Fisher's exact test was used for the comparison between continued treatment with adefovir dipivoxil and the crossover from adefovir dipivoxil to placebo and for the comparison between continued treatment with adefovir dipivoxil and the crossover from placebo to adefovir dipivoxil at week 96.
§ Patients with baseline alanine aminotransferase levels that exceeded the upper limit of the normal range were included in the analysis.

In the ranked assessment of inflammatory activity, the comparison of scores at baseline and week 96 in the continued-adefovir group showed improvement in 17 of 19 patients (89 percent) and no change in 2 of 19 patients (11 percent); in no patients did inflammation worsen. In the placebo–adefovir group, 14 of 20 patients (70 percent) had improvement, 2 of 20 (10 percent) had no change, and 4 of 20 (20 percent) had a worsening. In the adefovir–placebo group, four of eight patients (50 percent) had improvement, two of eight (25 percent) had no change, and two of eight (25 percent) had a worsening. Improvements were also seen in fibrosis, with patients in the continued-adefovir group having significant reductions from baseline in the Ishak fibrosis score at week 96 (mean [±SD] reduction, 0.63±1.07; median reduction, 1; P=0.031, as compared with the adefovir–placebo group). The improvements in fibrosis at weeks 48 and 96 were extended in patients who underwent a biopsy at week 144.

RESISTANCE PROFILE

A conserved site mutation (rtN236T) was identified in three patients in the continued-adefovir group,

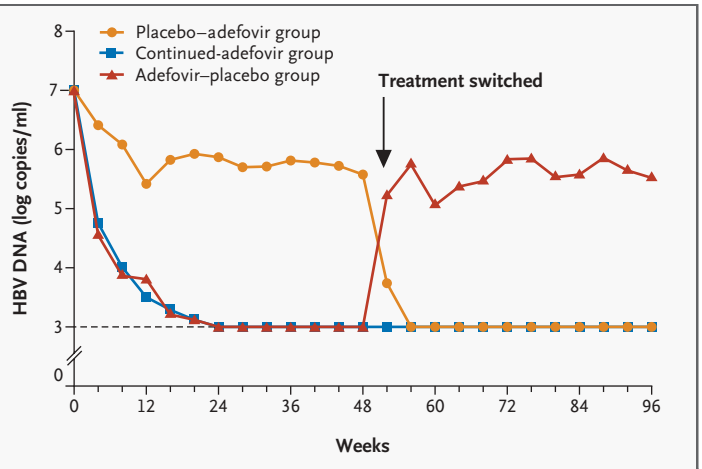

Figure 1. Median Serum HBV DNA Levels through Week 96.
The dashed line indicates 3 log copies per milliliter, which was the lower limit of detection for the assay.

two at week 96 and one at week 144. The emergence of rtN236T was associated with a rebound in serum HBV DNA and alanine aminotransferase levels. In vitro susceptibility testing demonstrated a reduction in susceptibility to adefovir that was 3.9 to 13.8 times that of wild-type virus. One patient was switched to lamivudine at week 104; HBV DNA levels, as evaluated by the Digene assay (lower limit of detection, 150,000 copies per milliliter), became undetectable, and serum alanine aminotransferase levels were normal after six months.[16] Subsequently, resistance to lamivudine developed in this patient; adefovir dipivoxil was restarted, and serum HBV DNA levels again became undetectable.

Another conserved site substitution mutation (rtA181V) in the B domain of HBV polymerase was seen in three additional patients in the continued-adefovir group, two at week 96 and one at week 144. A rebound in HBV DNA levels occurred in two of the three patients. In vitro susceptibility testing demonstrated a reduction in susceptibility that was 2.5 to 3 times that of wild-type virus. For one patient with rtA181V, lamivudine was added to ongoing adefovir therapy; serum HBV DNA levels subsequently were reduced by more than 2 log copies per milliliter.

Of the six patients in whom resistance developed, four had a reduced response to adefovir dipivoxil (serum HBV DNA reduction from baseline, <2.5 log copies per milliliter). The disease characteristics of these patients at baseline were similar to those of the overall patient population. The overall cumulative rate of resistance to adefovir dipivoxil among all patients at 48, 96, and 144 weeks was 0 percent, 3 percent, and 5.9 percent, respectively.

SAFETY

Adverse events during weeks 49 to 96 were similar in severity, nature, and frequency to those during the initial 48-week treatment period. At least one adverse event was reported in 58 of 79 patients (73 percent) in the continued-adefovir group, 41 of 60 patients (68 percent) in the placebo–adefovir group, and 32 of 40 (80 percent) in the adefovir–placebo group. The most common adverse events reported in the continued-adefovir group were headache, abdominal pain, and pharyngitis (Table 4).

The study drug was discontinued because of adverse events in two patients in the continued-adefovir group (a protocol-defined increase in serum creatinine levels of ≥0.5 mg per deciliter [44.2 μmol per liter] and hepatocellular carcinoma) and in three patients in the adefovir–placebo group (jaundice, elevated alanine aminotransferase levels, and a skin disorder).

No notable differences were seen in laboratory values from week 48, with the exception of increases in alanine aminotransferase levels associated with the withdrawal of adefovir dipivoxil therapy. In the adefovir–placebo group, 13 patients (32.5 percent) had alanine aminotransferase levels that were 10 times the upper limit of normal or higher. Elevations of alanine aminotransferase levels were observed in 6 percent of patients who continued to receive adefovir dipivoxil over 96 weeks. No patients had clinical signs of decompensation or required the intervention of an investigator. Of the 13 patients with elevations of alanine aminotransferase levels, 10 had an increase within 12 weeks after the cessation of adefovir dipivoxil therapy.

There were no overall changes in serum creatinine and phosphorus levels. Two patients in the continued-adefovir group had a confirmed increase in serum creatinine levels of 0.5 mg per deciliter or more from baseline. In one case, the highest value remained within the normal range and resolved with continued treatment. In the other case, the highest value was 2.3 mg per deciliter (203.3 μmol per liter), which returned to normal after discontinuation of the study drug. One additional patient in year 3 had a confirmed serum creatinine increase that returned to baseline within eight weeks after the cessation of adefovir dipivoxil. The safety pro-

Table 3. Changes from Baseline in Knodell Scores at Weeks 48 and 96.*

Knodell Score	Continued-Adefovir Group (N=19)		Adefovir–Placebo Group (N=8)		Placebo–Adefovir Group (N=20)	
	Week 48	Week 96	Week 48	Week 96	Week 48	Week 96
Overall						
Baseline	10.02±2.07		12.3±2.25		8.3±3.31	
Change	–4.4±2.39	–4.7±2.7	–4.3±1.49	–1.4±1.92	0.9±4.56	–2.4±4.79
Inflammation						
Baseline	8.37±1.50		10.0±1.31		6.40±2.76	
Change	–4.2±2.32	–4.3±2.71	–3.8±1.83	–0.9±1.96	0.6±3.78	–2.3±3.93
Fibrosis						
Baseline	1.84±1.17		2.3±1.39		1.9±1.17	
Change	–0.2±0.63	–0.4±1.12	–0.5±0.93	–0.5±0.93	0.3±1.17	–0.15±1.27

* Plus–minus values are means ±SD. Negative values indicate a decrease, and positive values an increase. Patients included those for whom biopsy specimens could be assessed at baseline, week 48, and week 96. Baseline values were measured before the first 48 weeks of treatment. Knodell scores (ranging from 0 to 22) evaluate necroinflammation and liver fibrosis. A lowering of scores from the initial biopsy indicates histologic improvement, and an increase indicates histologic worsening.

file over 144 weeks remained consistent with that seen earlier in the study.

DISCUSSION

As shown in other studies, treatment of HBeAg-negative chronic hepatitis B with lamivudine effectively suppresses HBV replication and results in biochemical remission and histologic improvement in more than two thirds of patients.[7,8,13] However, relapse has occurred in the majority of HBeAg-negative patients after the cessation of therapy.[8,17] Similarly, in this study, when treatment with adefovir dipivoxil was discontinued, the virologic, biochemical, and histologic benefits that had been gained in the first 48 weeks were lost. This finding suggests that because HBsAg seroconversion is rare,[2,4,11] long-term therapy will be needed in the majority of patients. Post-treatment flares in serum alanine aminotransferase levels were seen after therapy was stopped. Although these events were self-limiting in this study, it is important to monitor patients carefully after discontinuation of treatment with adefovir dipivoxil.[8,18]

To ensure a favorable risk–benefit profile, any treatment regimen must provide durable efficacy and limited toxicity, with minimal or no emergence of viral resistance. The development of viral resistance over time with the use of lamivudine, which is associated with a loss of clinical response, is common and may become serious in patients with advanced disease.[18] In another study, peginterferon therapy produced a sustained response in terms of normalization of alanine aminotransferase levels for up to 24 weeks after treatment was stopped, and 19 percent of patients had undetectable HBV DNA levels at week 24 of follow-up. However, further follow-up is required to see if this response will be sustained.[19]

Our study demonstrated that with prolonged therapy, adefovir dipivoxil brought about increasing and persistent virologic, biochemical, and histologic responses, with delayed and infrequent development of resistance. Among patients who began adefovir dipivoxil in the second 48 weeks, undetectable HBV DNA levels and normalization of alanine aminotransferase levels were achieved in a significant proportion of patients. However, comparisons of this subgroup of patients with those treated for 96 weeks should be made cautiously, since differences existed in baseline characteristics at the initiation of treatment with adefovir dipivoxil. Our results also suggest that an additional histologic benefit may occur with extended treatment, whereas cessation of treatment results in a reversal of improvement.

The adverse events associated with extended treatment with adefovir dipivoxil were similar in nature, severity, and frequency to those observed over the previous 48 weeks. Although increases in serum creatinine levels have previously been seen with

Table 4. Proportion of Patients with the Most Common Adverse Events and Renal Events.*

Event	Week 49 to Week 96			Continued-Adefovir Group	
	Continued-Adefovir Group (N=79)	Adefovir–Placebo Group (N=40)	Placebo–Adefovir Group (N=60)	Baseline to Week 96 (N=79)	Baseline to Week 144 (N=70)
	number of patients (percent)				
Any event	58 (73)	32 (80)	41 (68)	67 (85)	60 (86)
General					
Headache	12 (15)	4 (10)	5 (8)	23 (29)	19 (27)
Abdominal pain	16 (20)	7 (18)	5 (8)	22 (28)	20 (29)
Asthenia	8 (10)	6 (15)	3 (5)	15 (19)	15 (21)
Flu-like syndrome	6 (8)	4 (10)	5 (8)	14 (18)	14 (20)
Back pain	4 (5)	5 (12)	3 (5)	9 (11)	9 (13)
Pain	4 (5)	2 (5)	4 (7)	11 (14)	12 (17)
Accidental injury	4 (5)	2 (5)	2 (3)	6 (8)	8 (11)
Digestive					
Diarrhea	6 (8)	4 (10)	1 (2)	8 (10)	6 (9)
Dyspepsia	4 (5)	5 (12)†	1 (2)	7 (9)	7 (10)
Respiratory					
Pharyngitis	14 (18)	8 (20)	8 (13)	23 (29)	25 (36)
Increased cough	3 (4)	4 (10)	2 (3)	6 (8)	7 (10)
Bronchitis	2 (3)	1 (2)	1 (2)	6 (8)	9 (13)
Metabolic and nutritional					
Increased alanine aminotransferase levels	2 (3)‡	6 (15)†	1 (2)	3 (4)	3 (4)
Musculoskeletal					
Arthralgia	6 (8)	5 (13)†	1 (2)	7 (9)	6 (9)
Urogenital					
Increased creatinine levels	2 (3)	0	0	3 (4)	3 (4)
Hematuria	1 (1)	0	1 (2)	2 (3)	2 (3)
Kidney calculus	0	0	1 (2)	0	1 (1)
Kidney pain	0	0	1 (2)	2 (3)	4 (6)

* The most common adverse events are those that occurred in 10 percent or more of the patients in any treatment group.
† Fisher's exact test was used for the comparison with the placebo–adefovir group ($P<0.05$).
‡ Fisher's exact test was used for the comparison with the adefovir–placebo group ($P<0.05$).

higher daily doses (>30 mg), the risk is low with a daily dose of 10 mg.

The findings of this study raise two important questions: When should treatment be initiated, and when is it safe to stop? In view of the progressive course of HBeAg-negative chronic hepatitis B[1,3] and the progression of liver damage in patients who received placebo for 48 weeks in this study, it is reasonable to suggest that treatment should not be delayed. However, long-term therapy will be needed for the majority of patients. Therefore, there are several important factors to be weighed before treatment is begun: the patient's age, the severity of liver disease, the risk of disease progression, the risk of resistance, the likelihood of compliance, and the costs associated with long-term therapy.

Treatment with adefovir dipivoxil for 144 weeks resulted in continuing benefits in terms of viral suppression, normalization of biochemical measures, and histologic improvement. These benefits were associated with a delayed and infrequent emergence of resistance, making adefovir dipivoxil an excellent

candidate for the long-term management of HBeAg-negative chronic hepatitis B.

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APPENDIX

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ORIGINAL ARTICLE

Peginterferon Alfa-2a, Lamivudine, and the Combination for HBeAg-Positive Chronic Hepatitis B

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ABSTRACT

BACKGROUND

Current treatments for chronic hepatitis B are suboptimal. In the search for improved therapies, we compared the efficacy and safety of pegylated interferon alfa plus lamivudine, pegylated interferon alfa without lamivudine, and lamivudine alone for the treatment of hepatitis B e antigen (HBeAg)–positive chronic hepatitis B.

METHODS

A total of 814 patients with HBeAg-positive chronic hepatitis B received either peginterferon alfa-2a (180 μg once weekly) plus oral placebo, peginterferon alfa-2a plus lamivudine (100 mg daily), or lamivudine alone. The majority of patients in the study were Asian (87 percent). Most patients were infected with hepatitis B virus (HBV) genotype B or C. Patients were treated for 48 weeks and followed for an additional 24 weeks.

RESULTS

After 24 weeks of follow-up, significantly more patients who received peginterferon alfa-2a monotherapy or peginterferon alfa-2a plus lamivudine than those who received lamivudine monotherapy had HBeAg seroconversion (32 percent vs. 19 percent [P<0.001] and 27 percent vs. 19 percent [P=0.02], respectively) or HBV DNA levels below 100,000 copies per milliliter (32 percent vs. 22 percent [P=0.01] and 34 percent vs. 22 percent [P=0.003], respectively). Sixteen patients receiving peginterferon alfa-2a (alone or in combination) had hepatitis B surface antigen (HBsAg) seroconversion, as compared with 0 in the group receiving lamivudine alone (P=0.001). The most common adverse events were those known to occur with therapies based on interferon alfa. Serious adverse events occurred in 4 percent, 6 percent, and 2 percent of patients receiving peginterferon alfa-2a monotherapy, combination therapy, and lamivudine monotherapy, respectively. Two patients receiving lamivudine monotherapy had irreversible liver failure after the cessation of treatment — one underwent liver transplantation, and the other died.

CONCLUSIONS

In patients with HBeAg-positive chronic hepatitis B, peginterferon alfa-2a offers superior efficacy over lamivudine, on the basis of HBeAg seroconversion, HBV DNA suppression, and HBsAg seroconversion.

More than 400 million people worldwide are chronically infected with hepatitis B virus (HBV).[1] Effective therapy is necessary to prevent the progression of chronic hepatitis B to cirrhosis, hepatocellular carcinoma, and death. Current consensus guidelines from Asia, Europe, and the United States recommend lamivudine, adefovir, or conventional interferon alfa for the treatment of chronic hepatitis B.[2-5] Lamivudine and adefovir suppress HBV replication and result in an improvement in liver architecture on microscopical evaluation during therapy. However, rates of hepatitis B e antigen (HBeAg) seroconversion, an end point that has been associated with improved long-term clinical outcomes,[6,7] are generally low with these agents.[8-10] Lamivudine and to a lesser extent adefovir are also associated with drug resistance,[8,9,11,12] which increases with prolonged use.[12,13] Although there have been very few direct comparisons, rates of HBeAg loss and seroconversion with conventional interferon alfa seem to be slightly higher than the rates with lamivudine or adefovir.[5] Conflicting data on the benefits of combining interferon-based therapies and lamivudine[11,14,15] indicate that the role of combination therapy in the treatment of chronic hepatitis B requires further clarification.

Conventional interferon alfa has suboptimal pharmacokinetics, resulting in an inconvenient dosing schedule and fluctuating drug exposure. Peginterferon alfa-2a, created by attaching a large, branched, 40-kD polyethylene glycol molecule to interferon alfa-2a,[16] has better pharmacokinetics than conventional interferon alfa. This allows for once-weekly dosing, with effective serum concentrations maintained throughout the dosing interval.[17] Peginterferon alfa-2a, like conventional interferon alfa, has a dual immunomodulatory and antiviral mode of action. In a phase 2, proof-of-concept study, peginterferon alfa-2a had better clinical outcomes than did conventional interferon alfa in patients with HBeAg-positive chronic hepatitis B.[18]

The current study was designed to assess the efficacy and safety of three regimens in patients with HBeAg-positive chronic hepatitis B: peginterferon alfa-2a monotherapy, peginterferon alfa-2a plus lamivudine, and lamivudine monotherapy.

METHODS

STUDY DESIGN

This multicenter, randomized, partially double-blind study was conducted at 67 sites in 16 countries in Asia, Australasia, Europe, and North and South America. The study was conducted in compliance with the Declaration of Helsinki and with the principles of Good Clinical Practice. All patients gave written informed consent.

Patients were randomly assigned in a 1:1:1 ratio to receive 180 μg of peginterferon alfa-2a (Pegasys, Roche) once weekly plus oral placebo once daily, 180 μg of peginterferon alfa-2a once weekly plus 100 mg of lamivudine (Epivir-HBV or Zeffix, GlaxoSmithKline) once daily, or 100 mg of lamivudine once daily. Randomization was centralized and stratified according to geographic region and alanine aminotransferase levels. The study comprised 48 weeks of treatment and 24 weeks of treatment-free follow-up.

The study was designed by the sponsor (Roche) in collaboration with expert hepatologists. Clinical data were collected by the Peginterferon Alfa-2a HBeAg-Positive Chronic Hepatitis B Study Group. The sponsor held the data and conducted the statistical analyses. The principal authors had full access to the data and vouch for the veracity and completeness of the data and data analysis. All authors made substantial contributions to the analysis and interpretation of the data and the drafting or revising of the manuscript. All authors approved the final manuscript.

PATIENTS

Adults were eligible if they had been positive for hepatitis B surface antigen (HBsAg) for at least 6 months, were negative for antibodies to HBsAg (anti-HBs antibodies) and positive for HBeAg, had an HBV DNA level of more than 500,000 copies per milliliter, had a serum alanine aminotransferase level that was greater than 1 but less than or equal to 10 times the upper limit of the normal range, and had had findings on a liver biopsy within the previous 12 months that were consistent with the presence of chronic hepatitis B. Exclusion criteria included decompensated liver disease, a coexisting serious medical or psychiatric illness, a neutrophil count of less than 1500 per cubic millimeter, a plate-

let count of less than 90,000 per cubic millimeter, a serum creatinine level that was more than 1.5 times the upper limit of the normal range, a history of alcohol or drug abuse within one year before entry, and coinfection with hepatitis C or D virus or human immunodeficiency virus. Previous treatment for chronic hepatitis B was permitted, but not within the six months before the study.

EFFICACY MEASURES

Efficacy analyses included all randomized patients who received at least one dose of study medication. The study had two predetermined primary measures of efficacy assessed after 24 weeks of treatment-free follow-up: HBeAg seroconversion (defined by the loss of HBeAg and the presence of anti-HBe antibody) and suppression of HBV DNA to levels below 100,000 copies per milliliter. HBeAg and serum HBV DNA were measured at a central laboratory with the use of the AxSYM test (Abbott) and the Cobas Amplicor HBV Monitor Test (Roche Diagnostics), respectively.

Secondary efficacy measures assessed after 24 weeks of treatment-free follow-up included the combined response (HBeAg seroconversion, the normalization of alanine aminotransferase levels, and the suppression of HBV DNA levels to below 100,000 copies per milliliter), HBsAg seroconversion (defined by the loss of HBsAg and the presence of anti-HBs antibody), and the histologic response. A histologic response was defined as a reduction of at least two points in the modified Histologic Activity Index score[19] as compared with the pretreatment score. Scores for this index can range from 0 to 24, with inflammation graded from 0 (none) to 18 (severe) and fibrosis graded from 0 (none) to 6 (cirrhosis). Biopsy samples were scored by an independent histopathologist who was unaware of the timing of the biopsy or the patient's treatment assignment.

SAFETY ANALYSIS

Measures of safety included adverse events, hematologic measurements, clinical chemical measurements, and vital signs. The severity of adverse events was graded on a three-point scale (mild, moderate, and severe), and causality was determined by the investigator. Safety was assessed at baseline; at weeks 1, 2, 4, 6, 8, and 12 and every six weeks thereafter throughout treatment; and as appropriate during follow-up. Safety analyses included all patients who underwent randomization and received at least one dose of study medication and who underwent at least one safety assessment after the baseline assessment.

RESISTANCE AND GENOTYPIC ANALYSES

HBV DNA was extracted from all available serum samples from patients in the two lamivudine groups at the end of treatment (week 48). Mutations in the tyrosine, methionine, aspartate, and aspartate (YMDD) motif of the HBV polymerase gene were identified by means of the INNO-LiPA HBV DR assay (Innogenetics).[20] Genotyping of HBV DNA was performed at baseline on serum samples from all patients by means of the INNO-LiPA HBV Genotyping assay (Innogenetics).

STATISTICAL ANALYSIS

A sample size of 231 patients per treatment group provided the study with a statistical power of at least 80 percent at the 0.0125 level of significance, with a two-sided test, to detect a difference in HBeAg seroconversion rates of 20 percent versus 34 percent or HBV DNA response rates (suppression below 100,000 copies per milliliter) of 30 percent versus 45 percent. The sample size was increased to 250 patients to allow for withdrawals. An overall significance level of 0.025 was chosen because of the two predetermined primary end points. This more stringent overall significance level was adopted for regulatory reasons. For secondary efficacy measures, the level of significance was set at 0.05.

The Cochran–Mantel–Haenszel test, stratified according to geographic region and pretreatment alanine aminotransferase level, was used to compare differences in response rates between the treatment groups. Only if the overall test of the treatment effect was significant were pairwise comparisons performed. Fisher's exact test was used when appropriate. For each treatment group, response rates were computed with corresponding 95 percent confidence intervals. No interim analyses were performed.

Response rates were calculated for all patients who received at least one dose of study drug, according to the intention-to-treat principle. Patients with missing values at week 72 were classified as having no response.

RESULTS

CHARACTERISTICS OF THE PATIENTS

Of the 814 patients included in the analyses, 28 of the 271 patients randomly assigned to receive

peginterferon alfa-2a monotherapy, 25 of the 271 assigned to peginterferon alfa-2a plus lamivudine, and 42 of the 272 assigned to lamivudine monotherapy either did not complete treatment or did not enter or complete the follow-up phase. Baseline demographic and other characteristics were similar among the three treatment groups (Table 1).

Table 1. Baseline Characteristics of the Patients.*

Characteristic	Peginterferon Alfa-2a plus Placebo (N=271)	Peginterferon Alfa-2a plus Lamivudine (N=271)	Lamivudine (N=272)
Male sex — no. (%)	214 (79)	208 (77)	215 (79)
Race or ethnic group — no. (%)†			
White	24 (9)	23 (8)	32 (12)
Asian	237 (87)	236 (87)	232 (85)
Black	4 (1)	4 (1)	3 (1)
Other	6 (2)	8 (3)	5 (2)
Age — yr			
Mean ±SD	32.5±9.6	31.7±10.3	31.6±9.7
Median	31	29	30
Range	18–77	18–66	17–65
Weight — kg			
Mean ±SD	66±13.0	66±14.8	67±14.4
Median	65	64	65
Range	35–128	41–135	40–160
Alanine aminotransferase — IU/liter‡			
Mean ±SD	114.6±114.3	114.9±94.1	102.3±78.4
Median	84.0	81.8	82.1
Range	11.4–1266.0	13.2–642.0	5.9–462.1
HBV DNA — log copies/ml¶			
Mean ±SD	9.9±2.1	10.1±1.9	10.1±2.0
Median	9.8	9.9	9.8
Range	4.4–16.1	3.1–17.9	3.0–16.0
Bridging fibrosis or cirrhosis — no. (%)§	49 (18)	40 (15)	47 (17)
Previous use of conventional interferon alfa — no. (%)	30 (11)	32 (12)	32 (12)
Previous use of lamivudine — no. (%)	31 (11)	24 (9)	42 (15)
Genotype distribution — no. (%)			
A	23 (8)	18 (7)	15 (6)
B	76 (28)	82 (30)	73 (27)
C	162 (60)	156 (58)	162 (60)
D	9 (3)	11 (4)	17 (6)
E, F, or H	0	3 (1)	4 (1)
Mixed	1 (<1)	1 (<1)	1 (<1)

* Percentages may not sum to 100 because of rounding.
† Race or ethnic group was generally assigned by the investigator, but in rare instances was clarified with the patient.
‡ The upper limit of the normal range is 30 IU per liter.
§ The presence or absence of bridging fibrosis and cirrhosis was assessed by local pathologists.
¶ Log to the base 10 was used.

Table 2. Rates of HBeAg, Virologic, Biochemical, Combined, and Histologic Responses.*

Response	End of Treatment (Week 48)			End of Follow-up (Week 72)		
	Peginterferon Alfa-2a plus Placebo (N=271)	Peginterferon Alfa-2a plus Lamivudine (N=271)	Lamivudine (N=272)	Peginterferon Alfa-2a plus Placebo (N=271)	Peginterferon Alfa-2a plus Lamivudine (N=271)	Lamivudine (N=272)
HBeAg response						
HBeAg seroconversion†						
Patients — no. (%)	72 (27)	64 (24)	55 (20)	87 (32)	74 (27)	52 (19)
95% CI — %	21.4 to 32.2	18.7 to 29.1	15.6 to 25.5	26.6 to 38.0	22.1 to 33.0	14.6 to 24.3
P value				<0.001	0.02	
Odds ratio (95% CI)‡				2.0 (1.3 to 3.0)	1.6 (1.1 to 2.4)	
HBeAg loss						
Patients — no. (%)	81 (30)	73 (27)	59 (22)	91 (34)	77 (28)	57 (21)
95% CI — %	24.5 to 35.7	21.7 to 32.6	16.9 to 27.1	28.0 to 39.5	23.1 to 34.2	16.3 to 26.3
P value				<0.001	0.04	
Virologic response						
HBV DNA <100,000 copies/ml§						
Patients — no. (%)	142 (52)	233 (86)	169 (62)	86 (32)	91 (34)	60 (22)
95% CI — %	46.3 to 58.5	81.3 to 89.9	56.1 to 67.9	26.2 to 37.6	28.0 to 39.5	17.3 to 27.5
P value				0.01	0.003	
Odds ratio (95% CI)‡				1.6 (1.1 to 2.4)	1.8 (1.2 to 2.6)	
HBV DNA <400 copies/ml						
Patients — no. (%)	68 (25)	186 (69)	108 (40)	39 (14)	37 (14)	14 (5)
95% CI — %	20.0 to 30.7	62.7 to 74.1	33.8 to 45.8	10.4 to 19.1	9.8 to 18.3	2.8 to 8.5
P value				<0.001	<0.001	
Change in HBV DNA						
Total no. of patients	248	249	249	248	254	241
Mean log copies/ml	−4.5	−7.2	−5.8	−2.4	−2.7	−1.9
95% CI — log copies/ml	−4.1 to −4.9	−6.9 to −7.5	−5.4 to −6.1	−2.0 to −2.8	−2.2 to −3.1	−1.5 to −2.3
Biochemical response						
Normalization of ALT						
Patients — no. (%)	105 (39)	126 (46)	168 (62)	111 (41)	106 (39)	76 (28)
95% CI — %	32.9 to 44.8	40.4 to 52.6	55.7 to 67.6	35.0 to 47.1	33.3 to 45.2	22.7 to 33.7
P value				0.002	0.006	

HBeAg RESPONSE

At the end of treatment (week 48), the percentage of patients with HBeAg seroconversion was highest with peginterferon alfa-2a monotherapy (Table 2 and Fig. 1A). The overall HBeAg seroconversion rates continued to rise during the entire study period in the two peginterferon alfa-2a groups but not in the lamivudine monotherapy group; seroreversion (loss of anti-HBe antibody and re-expression of HBeAg) was substantially less frequent with peginterferon alfa-2a monotherapy (occurring in 13 of 72 patients, or 18 percent) and with combination

Table 2. (Continued.)

Response	End of Treatment (Week 48)			End of Follow-up (Week 72)		
	Peginterferon Alfa-2a plus Placebo (N=271)	Peginterferon Alfa-2a plus Lamivudine (N=271)	Lamivudine (N=272)	Peginterferon Alfa-2a plus Placebo (N=271)	Peginterferon Alfa-2a plus Lamivudine (N=271)	Lamivudine (N=272)
Combined response						
HBeAg seroconversion, normalization of ALT, and HBV DNA <100,000 copies/ml						
Patients — no. (%)	27 (10)	42 (15)	50 (18)	62 (23)	56 (21)	28 (10)
95% CI — %	6.7 to 14.2	11.4 to 20.4	14.0 to 23.5	18.0 to 28.3	16.0 to 26.0	7.0 to 14.5
P value				<0.001	<0.001	
Histologic response¶						
All patients — no.‖				271	271	272
Improved — no. of patients (%)				102 (38)	112 (41)	93 (34)
95% CI — %				31.8 to 43.7	35.4 to 47.4	28.6 to 40.2
Patients with paired biopsy samples — no.**				207	215	184
Improved — no. of patients (%)				102 (49)	112 (52)	93 (51)
95% CI — %				42.3 to 56.3	45.2 to 58.9	43.1 to 58.0

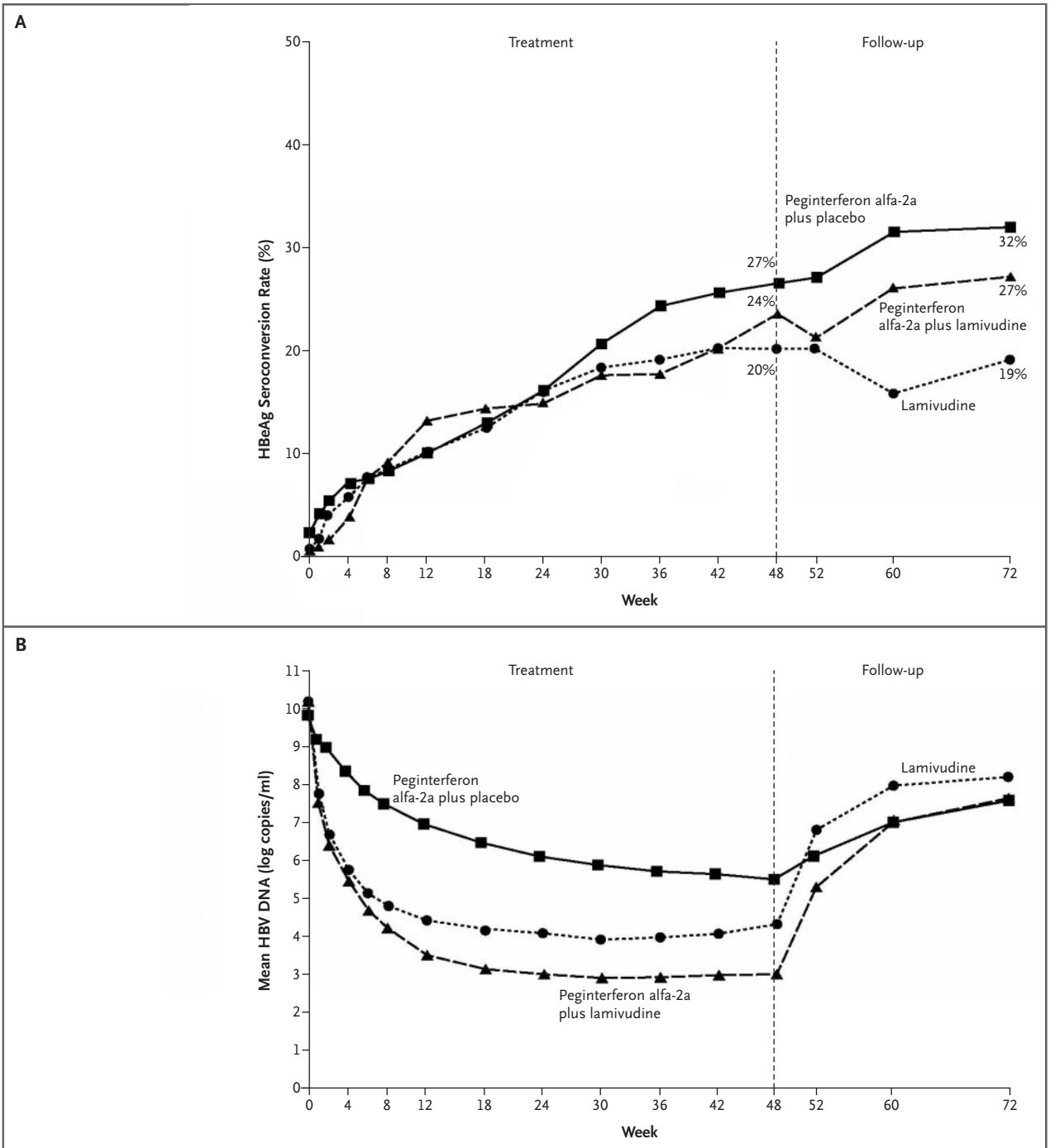
* All P values are from the Cochran–Mantel–Haenszel test for pairwise comparison of each peginterferon alfa-2a group with the lamivudine monotherapy group at week 72. CI denotes confidence interval, and ALT alanine aminotransferase.
† P=0.003 for the overall test of treatment effect, and P=0.23 for the comparison between peginterferon alfa-2a plus placebo and peginterferon alfa-2a plus lamivudine.
‡ Odds ratios are given with 95 percent confidence intervals only for the two primary efficacy outcomes.
§ P=0.007 for the overall test of treatment effect, and P=0.65 for the comparison between peginterferon alfa-2a plus placebo and peginterferon alfa-2a plus lamivudine.
¶ Histologic response was defined as a reduction of at least two points in the modified Histology Activity Index score as compared with the pretreatment score. Scores for this index can range from 0 to 24, with inflammation graded from 0 (none) to 18 (severe) and fibrosis graded from 0 (none) to 6 (cirrhosis).[19]
‖ Patients without paired biopsy samples were classified as having no response. P=0.23 for the overall test of treatment effect.
** Patients without paired biopsy samples were excluded. P=0.79 for the overall test of treatment effect.

therapy (14 of 64 patients, or 22 percent) than with lamivudine monotherapy (23 of 55 patients, or 42 percent; P=0.005 and P=0.03, respectively, by Fisher's exact test). After 24 weeks of follow-up (week 72), the percentage of patients with HBeAg seroconversion was significantly higher with peginterferon alfa-2a monotherapy (32 percent) and combination therapy (27 percent) than with lamivudine monotherapy (19 percent; P<0.001 and P=0.02, respectively) (Table 2 and Fig. 2). At weeks 48 and 72, rates of HBeAg loss closely reflected rates of HBeAg seroconversion (Table 2).

HBeAg seroconversion rates in patients with and without previous exposure to lamivudine or conventional interferon were similar to rates in the overall study population (Table 3). Additional stratified analyses are detailed in Table 3.

VIROLOGIC RESPONSE

At week 48, the percentage of patients with suppression of HBV DNA was highest with combination therapy (Table 2). This changed during follow-up such that at week 72, suppression of HBV DNA levels to less than 100,000 copies per milliliter occurred in a significantly higher percentage of patients receiving peginterferon alfa-2a monotherapy (32 percent) or peginterferon alfa-2a plus lamivudine (34 percent) than in those receiving lamivudine monotherapy (22 percent; P=0.01 and P=0.003, respectively) (Table 2). Rates of suppression of HBV



No. of Patients													
Peginterferon alfa-2a plus placebo	271	269	265	265	262	263	259	258	255	248	254	241	248
Peginterferon alfa-2a plus lamivudine	271	268	268	267	268	263	259	254	255	249	254	244	254
Lamivudine	272	267	267	267	263	263	259	260	244	249	248	228	241

Figure 1. Rates of HBeAg Seroconversion (Panel A) and HBV DNA Levels (Panel B), from Baseline to Week 72.

HBeAg seroconversion was defined by the loss of HBeAg and the presence of anti-HBe antibody. Log to the base 10 was used. The information about the number of patients refers only to Panel B.

DNA levels to less than 400 copies per milliliter at week 72 were 14 percent with both peginterferon alfa-2a monotherapy and peginterferon alfa-2a plus lamivudine, and 5 percent with lamivudine alone (P<0.001 for both comparisons with lamivudine monotherapy). The patterns of HBV DNA levels throughout the study are shown in Figure 1B. Rates of normalization of alanine aminotransferase levels and combined response at week 72 closely reflected the virologic response rates (Table 2).

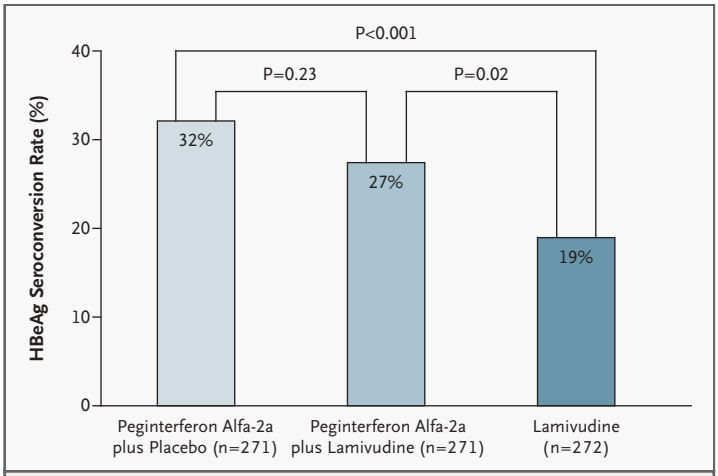


Figure 2. Rates of HBeAg Seroconversion after 24 Weeks of Follow-up.
The percentage of patients with seroconversion in each group is shown at the top of the corresponding bar.

HBsAg RESPONSE

At week 72, HBsAg seroconversion was identified in eight patients receiving peginterferon alfa-2a monotherapy (three Asian and five white patients, five with HBV genotype A and three with genotype C) and in eight receiving peginterferon alfa-2a plus lamivudine (five Asian and three white patients; two with genotype A, one with genotype B, four with genotype C, and one with genotype H). HBsAg seroconversion was not identified in any patients receiving lamivudine monotherapy. The differences in HBsAg seroconversion between peginterferon alfa-2a monotherapy and lamivudine monotherapy, and between peginterferon alfa-2a plus lamivudine and lamivudine monotherapy, were significant (P=0.004 for both comparisons with lamivudine monotherapy, by Fisher's exact test).

HISTOLOGIC RESPONSE

The rate of histologic response was similar among the three treatment groups (Table 2). There was a significant association between improved histologic activity and either HBeAg seroconversion, a virologic response, or a biochemical response at week 72, regardless of the treatment group (P<0.001). Among patients with paired biopsy samples, a histologic response occurred in 133 of 179 patients (74 percent) who had HBeAg seroconversion as compared with 174 of 427 patients (41 percent) who did not have HBeAg seroconversion (P<0.001 by the log-likelihood ratio test).

ALANINE AMINOTRANSFERASE LEVELS

Alanine aminotransferase elevations, defined as a peak value at least five times as great as the baseline value, occurred in 14 patients receiving peginterferon alfa-2a monotherapy (5 percent), 16 receiving combination therapy (6 percent), and 12 receiving lamivudine monotherapy (4 percent). Rates of HBeAg seroconversion in these patients at week 72 were 43 percent, 38 percent, and 25 percent, respectively (Table 3).

RESISTANCE

At week 48, YMDD mutations were detected in 69 of 254 patients receiving lamivudine monotherapy (27 percent) and 9 of 256 patients receiving peginterferon alfa-2a plus lamivudine (4 percent, P<0.001).

SAFETY

The rate of withdrawal from therapy was low in all three groups (Table 4). The rates of adverse events were similar in the peginterferon alfa-2a and combination-therapy groups but were significantly less frequent in the lamivudine-only group (P<0.001 for the overall comparison). Among the three groups, the incidence of adverse events was similar between Asian and non-Asian patients (79 percent and 82 percent, respectively). The most common adverse events were those known to occur with interferon alfa therapy, including pyrexia, fatigue, headache, and myalgia (Table 4).

Depression, which is a potential concern with interferon-based therapy, was infrequent during the study and was reported by 13 patients (5 percent) receiving peginterferon alfa-2a monotherapy, 16 patients (6 percent) receiving peginterferon alfa-2a plus lamivudine, and 4 patients (1 percent) receiving lamivudine monotherapy.

Table 3. Effect of Baseline Factors and Alanine Aminotransferase Levels during Treatment on HBeAg Seroconversion Rates at Week 72.

Variable	Peginterferon Alfa-2a plus Placebo	Peginterferon Alfa-2a plus Lamivudine	Lamivudine
	no. of patients achieving HBeAg seroconversion/total no. of patients (%)		
Overall study population	87/271 (32)	74/271 (27)	52/272 (19)
Patients with no previous anti-HBV therapy*	66/214 (31)	59/221 (27)	42/208 (20)
Patients with previous exposure to lamivudine			
Yes	10/31 (32)	6/24 (25)	7/42 (17)
No	77/240 (32)	68/247 (28)	45/230 (20)
Patients with previous exposure to conventional interferon			
Yes	13/30 (43)	11/32 (34)	4/32 (12)
No	74/241 (31)	63/239 (26)	48/240 (20)
HBV genotype†			
A	12/23 (52)	4/18 (22)	3/15 (20)
B	23/76 (30)	24/82 (29)	17/73 (23)
C	50/162 (31)	43/156 (28)	29/162 (18)
D	2/9 (22)	2/11 (18)	3/17 (18)
Baseline HBV DNA levels (log copies/ml)			
≤9.07	37/70 (53)	20/56 (36)	24/78 (31)
>9.07–10.26	39/138 (28)	40/147 (27)	21/123 (17)
>10.26	11/63 (17)	14/68 (21)	7/71 (10)
Baseline alanine aminotransferase level (×ULN)‡			
≤2	27/92 (29)	19/93 (20)	19/96 (20)
>2 to 5	36/121 (30)	30/111 (27)	20/129 (16)
>5	24/58 (41)	25/67 (37)	13/47 (28)
Maximum alanine aminotransferase level during treatment (×ULN)‡			
≤5	39/149 (26)	35/150 (23)	33/177 (19)
>5 to 10	28/74 (38)	27/86 (31)	16/64 (25)
>10	20/48 (42)	12/35 (34)	3/31 (10)
Maximum alanine aminotransferase level during treatment (×baseline value)			
≤5	81/257 (32)	68/255 (27)	49/260 (19)
>5	6/14 (43)	6/16 (38)	3/12 (25)

* This group includes patients who had previously been treated with lamivudine, conventional interferon, and peginterferon only.
† This group includes only patients infected with HBV genotype A, B, C, or D.
‡ ULN denotes the upper limit of the normal range, which is 30 IU per liter.

Thirty-three patients had serious adverse events during treatment and up to eight weeks after therapy: 12 patients (4 percent) receiving peginterferon alfa-2a monotherapy, 16 patients (6 percent) receiving peginterferon alfa-2a plus lamivudine, and 5 patients (2 percent) receiving lamivudine monotherapy (Table 4). However, two patients receiving lamivudine monotherapy, neither of whom had cirrhosis or bridging fibrosis at baseline, had hepatic decompensation after the cessation of treatment. One patient required liver transplantation and made a full recovery, and one patient died.

Mean neutrophil and platelet counts were reduced during treatment with peginterferon alfa-2a monotherapy and peginterferon alfa-2a plus lamivudine, yet returned to baseline levels shortly after treatment was stopped. Laboratory abnormalities (alanine aminotransferase elevation, neutropenia, and thrombocytopenia) were the most common reason for dose modification (Table 4).

DISCUSSION

We found that peginterferon alfa-2a alone or in combination with lamivudine resulted in higher rates of sustained HBeAg, HBsAg, virologic, and biochemical response among patients with HBeAg-positive chronic hepatitis B than did lamivudine alone. HBeAg seroconversion is a key objective of therapy for HBeAg-positive chronic hepatitis B, since it is associated with improved long-term clinical outcomes, such as histologic improvement and increased complication-free and overall survival.[6,7]

In this study of patients, predominantly of Asian origin, who had previously been considered to have difficult-to-treat chronic hepatitis B,[21] HBeAg seroconversion rates were significantly higher after 24 weeks of treatment-free follow-up in patients receiving peginterferon alfa-2a alone or in combination with lamivudine than in those receiving lamivudine alone. Previous exposure to lamivudine did not affect the overall rates of HBeAg seroconversion. In accordance with previous findings with interferon alfa therapy,[22] marked elevations in alanine aminotransferase levels were more frequently associated with HBeAg response in patients receiving peginterferon alfa-2a alone or in combination with lamivudine than in those receiving lamivudine alone.

At present, it is not clear whether viral genotype is a predictor of treatment response in chronic hepatitis B, as it is in chronic hepatitis C. Responses to nucleoside or nucleotide analogues are generally consistent among all genotypes,[23,24] whereas higher responses to interferon alfa have been reported for HBV genotype A than for genotype D and for genotype B than for genotype C.[25] The results of our study indicate that HBeAg seroconversion was generally consistent across all genotypes. However, a recent study of peginterferon alfa-2b[26] reported a higher HBeAg seroconversion rate for genotype A. This trend was also observed in our study in the patients receiving peginterferon alfa-2a monotherapy. However, in our study, the number of patients infected with genotype A was very low.

Previous studies have shown that HBV DNA suppression is associated with HBeAg seroconversion.[8,10,27] At week 48 of our study, viral suppression was higher in patients receiving lamivudine monotherapy than in those receiving peginterferon alfa-2a monotherapy. However, despite this more potent suppression of HBV DNA with lamivudine, rates of HBeAg seroconversion at the end of treatment and after follow-up were highest with peginterferon alfa-2a monotherapy. These data indicate that a separate and probably immunomodulatory component influences HBeAg seroconversion with peginterferon alfa-2a. Similarly, among patients receiving peginterferon alfa-2a monotherapy or peginterferon alfa-2a plus lamivudine, who presumably had equivalent immunomodulation related to peginterferon alfa-2a, the increased antiviral activity in the group receiving combination therapy did not improve HBeAg seroconversion rates. Significantly fewer patients receiving combination therapy had YMDD mutants at the end of treatment than did patients receiving lamivudine alone. This suggests that more profound HBV DNA suppression, such as that seen during treatment with peginterferon alfa-2a plus lamivudine, leads to a lower incidence of lamivudine resistance, a finding that concurs with previous studies of HBV.[28,29]

HBsAg loss or seroconversion after therapy is considered the ultimate therapeutic goal of anti-HBV therapy, since it is associated with positive long-term clinical outcomes.[2,4,5,30] In this study, HBsAg seroconversion was identified in 8 of 473 Asian patients (2 percent) and 8 of 47 white patients (17 percent) receiving peginterferon alfa-2a alone or in combination with lamivudine, as compared with none receiving lamivudine alone. These HBsAg seroconversion rates with peginterferon

Table 4. Incidence of Discontinuation of Treatment, Dose Modification, and Adverse Events.*

Variable	Peginterferon Alfa-2a plus Placebo (N=271)	Peginterferon Alfa-2a plus Lamivudine (N=271)	Lamivudine (N=272)
	number of patients (percent)		
Discontinuation			
For safety reasons†	8 (3)	12 (4)	2 (1)
For other reasons‡	9 (3)	6 (2)	12 (4)
Dose modification§			
Total	124 (46)	127 (47)	—
Adverse event	20 (7)	23 (8)	—
Laboratory abnormality	99 (37)	102 (38)	—
Dose missed or dosage error	25 (9)	20 (7)	—
Other	2 (1)	2 (1)	—
Adverse events			
≥1 Reported serious adverse event (weeks 0 to 56)¶	12 (4)	16 (6)	5 (2)
Deaths			
Weeks 0 to 56	0	3 (1)‖	0
Weeks 57 to 72	0	0	1 (<1)**
≥1 Reported adverse event (weeks 0 to 56)††	240 (89)	240 (89)	152 (56)

alfa-2a compare favorably with rates of HBsAg response within 12 months of the cessation of treatment that were shown in studies of conventional interferon in Asian[31-33] and white[7,30] patients.

No statistically significant differences in efficacy were observed between the groups receiving peginterferon alfa-2a monotherapy and peginterferon alfa-2a plus lamivudine after 24 weeks of follow-up, a finding that concurs with a recent study of patients with HBeAg-negative chronic hepatitis B.[29] However, these results do not categorically rule out the possibility that combination therapy, including sequential therapy, may provide clinically relevant benefits.

The tolerability and safety profiles of peginterferon alfa-2a monotherapy and peginterferon alfa-2a plus lamivudine were similar to those reported in patients with HBeAg-negative chronic hepatitis B, and there were no unexpected adverse effects.[29] The safety profile of peginterferon alfa-2a in this study also compares favorably with the profiles described in previous studies of conventional interferon alfa in HBeAg-positive chronic hepatitis B.[11,18] As anticipated, peginterferon alfa-2a alone or in combination with lamivudine was not tolerated as well as lamivudine monotherapy. However, the rate of withdrawal from peginterferon alfa-2a therapy was less than 5 percent.

Depression was reported in 5 percent of patients receiving peginterferon alfa-2a in this study. This incidence is substantially lower than that observed among patients with chronic hepatitis C (16 to 20 percent).[34,35] This finding concurs with data from a recent study of peginterferon alfa-2a in HBeAg-negative chronic hepatitis B.[29]

In conclusion, the results of this large, multinational study show that peginterferon alfa-2a provides significantly improved efficacy over lamivudine in the treatment of HBeAg-positive chronic hepatitis B. Improvement in sustained HBeAg and HBsAg seroconversion rates, as well as sustained virologic and biochemical response rates, indicate that peginterferon alfa-2a offers a therapeutic advantage over available treatments for chronic hepatitis B. The ability to achieve HBeAg and HBsAg seroconversion after a defined period of peginter-

Table 4. (Continued.)

Variable	Peginterferon Alfa-2a plus Placebo (N=271)	Peginterferon Alfa-2a plus Lamivudine (N=271)	Lamivudine (N=272)
	number of patients (percent)		
Adverse events (continued)			
Most common adverse events (weeks 0 to 56)‡‡			
Pyrexia	133 (49)	148 (55)	12 (4)
Fatigue	108 (40)	101 (37)	37 (14)
Headache	76 (28)	81 (30)	27 (10)
Myalgia	70 (26)	77 (28)	8 (3)
Alopecia	55 (20)	78 (29)	6 (2)
Decreased appetite	40 (15)	34 (13)	5 (2)
Rash	27 (10)	22 (8)	10 (4)
Pruritus	26 (10)	26 (10)	5 (2)
Dizziness	25 (9)	32 (12)	11 (4)
Diarrhea	25 (9)	26 (10)	9 (3)
Nausea	24 (9)	27 (10)	6 (2)
Injection-site reaction	24 (9)	15 (6)	0
Arthralgia	24 (9)	24 (9)	7 (3)
Upper respiratory tract infection	21 (8)	15 (6)	29 (11)
Insomnia	20 (7)	23 (8)	10 (4)
Rigors	19 (7)	27 (10)	0
Upper abdominal pain	19 (7)	14 (5)	20 (7)
Sore throat	15 (6)	21 (8)	19 (7)
Gingival bleeding	15 (6)	15 (6)	1 (<1)
Cough	14 (5)	19 (7)	10 (4)
Dyspepsia	14 (5)	6 (2)	9 (3)
Depression	13 (5)	16 (6)	4 (1)

* Values are based on all randomized patients who received at least one dose of study medication and had at least one safety assessment after baseline. Dashes indicate no dose modifications in the group receiving lamivudine monotherapy.
† P=0.03 for the overall test of treatment effect. P=0.06 for the comparison between peginterferon alfa-2a plus placebo and lamivudine alone, and P=0.01 for the comparison between peginterferon alfa-2a plus lamivudine and lamivudine alone.
‡ P=0.36 for the overall test of treatment effect.
§ Some patients who required a dose modification had both an adverse event and a laboratory abnormality. Laboratory abnormalities include alanine aminotransferase elevation, neutropenia, and thrombocytopenia. Other includes circumstances related to patient compliance.
¶ A serious adverse event was one that presented a clinically significant hazard or resulted in a contraindication, side effect, or precaution. P=0.05 for the overall test of treatment effect, P=0.09 for the comparison between peginterferon alfa-2a plus placebo and lamivudine alone, and P=0.01 for the comparison between peginterferon alfa-2a plus lamivudine and lamivudine alone.
‖ All three deaths were accidental and were considered by the investigators to be unrelated to the study medication.
** Life-threatening hepatic encephalopathy developed in this patient, which was considered by the investigator to be related to discontinuation of lamivudine treatment.
†† P<0.001 for the overall test of treatment effect, P<0.001 for the comparison between peginterferon alfa-2a plus placebo and lamivudine alone, and P<0.001 for the comparison between peginterferon alfa-2a plus lamivudine and lamivudine alone.
‡‡ Patients may have had more than one adverse event. The adverse events listed are those reported by at least 5 percent of patients in any treatment group.

feron alfa-2a therapy supports the use of peginterferon alfa-2a as a first-line therapy for patients with HBeAg-positive chronic hepatitis B.

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APPENDIX

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ORIGINAL ARTICLE

Capecitabine as Adjuvant Treatment for Stage III Colon Cancer

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ABSTRACT

BACKGROUND

Intravenous bolus fluorouracil plus leucovorin is the standard adjuvant treatment for colon cancer. The oral fluoropyrimidine capecitabine is an established alternative to bolus fluorouracil plus leucovorin as first-line treatment for metastatic colorectal cancer. We evaluated capecitabine in the adjuvant setting.

METHODS

We randomly assigned a total of 1987 patients with resected stage III colon cancer to receive either oral capecitabine (1004 patients) or bolus fluorouracil plus leucovorin (Mayo Clinic regimen; 983 patients) over a period of 24 weeks. The primary efficacy end point was at least equivalence in disease-free survival; the primary safety end point was the incidence of grade 3 or 4 toxic effects due to fluoropyrimidines.

RESULTS

Disease-free survival in the capecitabine group was at least equivalent to that in the fluorouracil-plus-leucovorin group (in the intention-to-treat analysis, $P<0.001$ for the comparison of the upper limit of the hazard ratio with the noninferiority margin of 1.20). Capecitabine improved relapse-free survival (hazard ratio, 0.86; 95 percent confidence interval, 0.74 to 0.99; $P=0.04$) and was associated with significantly fewer adverse events than fluorouracil plus leucovorin ($P<0.001$).

CONCLUSIONS

Oral capecitabine is an effective alternative to intravenous fluorouracil plus leucovorin in the adjuvant treatment of colon cancer.

Almost 1 million patients receive a diagnosis of colorectal cancer yearly, and half a million deaths from this neoplasm occur annually worldwide.[1] Each year, approximately 230,000 patients with colon cancer are eligible for adjuvant chemotherapy.[1-3] The benefits of fluorouracil-based adjuvant chemotherapy in reducing the risk of relapse and prolonging survival in patients with resected colon cancer are well established, particularly in stage III disease.[4-6] Survival advantages were demonstrated with bolus intravenous fluorouracil plus leucovorin administered according to the Mayo Clinic regimen (five days, monthly, for six months) or the Roswell Park regimen (weekly bolus, six of every eight weeks, for eight months).[5,7-10] The choice between these regimens involves a decision about toxic effects: stomatitis and neutropenia with the Mayo Clinic regimen or diarrhea with the Roswell Park regimen. Infused fluorouracil regimens have also been evaluated, but they have no greater efficacy than bolus fluorouracil plus leucovorin in the adjuvant setting.[11-13] For this reason, six to eight months of treatment with bolus fluorouracil plus leucovorin has been the standard of care worldwide as adjuvant treatment of colon cancer since the 1990s.[14]

There are, however, discrepancies between consensus recommendations and adjuvant treatment in the community.[10,15-17] More effective, better tolerated, and more convenient chemotherapy is required, especially for patients older than 65 years,[18] who are less likely to receive rigorous chemotherapy.[16,17] Moreover, most patients (84 to 89 percent) with cancer would prefer oral chemotherapy, provided efficacy is not compromised.[19,20]

The oral fluoropyrimidine capecitabine (Xeloda, Hoffmann–La Roche) generates fluorouracil preferentially in tumor tissue, by way of a three-step enzymatic cascade.[21] The final stage of conversion to fluorouracil is catalyzed by thymidine phosphorylase, which is appreciably more active in tumor than in healthy tissue.[21,22] As first-line treatment for metastatic colorectal cancer, capecitabine is an established alternative to the combination of fluorouracil and leucovorin. It achieved response rates superior to those achieved with the Mayo Clinic regimen (26 percent vs. 17 percent), with equivalent progression-free survival and overall survival.[23] Capecitabine was also associated with fewer adverse effects than the Mayo Clinic regimen[24] and reduced the use of medical resources.[25]

These data provided the rationale for a phase 3 trial (Xeloda in Adjuvant Colon Cancer Therapy [X-ACT]) to compare capecitabine and the Mayo Clinic fluorouracil-plus-leucovorin regimen as adjuvant treatment in resected stage III colon cancer. The primary objective was to demonstrate that capecitabine was at least equivalent to fluorouracil plus leucovorin in terms of disease-free survival. We report here planned analyses of the primary efficacy end point and the safety end point of the trial.

METHODS

The study was conducted in accordance with the Declaration of Helsinki and its amendments or with the laws and regulations of the country in which the research was conducted, whichever afforded greater protection. Patients gave written informed consent for participation in the trial.

ELIGIBILITY CRITERIA

Patients 18 to 75 years of age were required to be fully recovered after surgery for histologically confirmed stage III colon carcinoma. Surgery had to have been performed within eight weeks before randomization. An Eastern Cooperative Oncology Group performance score of 0 or 1 (with a score of 0 indicating normal activity and 1 indicating the presence of symptoms but nearly full ambulatory capacity) and at least five years' life expectancy were required.

Patients with evidence of metastatic disease, including tumor cells in ascites or microscopic evidence of residual disease, were ineligible for participation. Macroscopic disease was ruled out with the use of abdominal pelvic computed tomography (CT) or magnetic resonance imaging (MRI) and chest radiography. Patients were also excluded on the basis of prior cytotoxic chemotherapy or organ allograft, clinically significant cardiac disease, severe renal impairment, central nervous system disorders, or pregnancy or lactation. Sexually active premenopausal women unwilling to practice contraception were ineligible.

STUDY DESIGN AND TREATMENT

The primary aim of the study was to show at least equivalence in disease-free survival between capecitabine and bolus fluorouracil plus leucovorin. Secondary end points included relapse-free survival, overall survival, and safety. Assessment of the rate of disease-free survival at three years was a prespecified secondary end point. Results with regard to the

quality of life are summarized here. The Quality of Life Questionnaire (QLQ-C30, version 2.0) of the European Organization for the Research and Treatment of Cancer was administered at baseline and before the start of the treatment cycles in weeks 7, 16, and 25 in the capecitabine group and weeks 9, 17, and 25 in the fluorouracil-plus-leucovorin group.

Patients were assigned to 24 weeks of treatment with either eight cycles of oral capecitabine, at a dose of 1250 mg per square meter of body-surface area, twice daily on days 1 through 14 every 21 days, or six cycles of rapid intravenous infusion of leucovorin, at a dose of 20 mg per square meter, followed immediately by an intravenous bolus of fluorouracil, at a dose of 425 mg per square meter, on days 1 through 5 every 28 days. Randomization, with the use of treatment allocation codes (scratch-off labels), was stratified by center and performed with a block size of four. The block size was unknown to investigators and monitors.

EVALUATION OF EFFICACY

Patients were assessed every six months for two years after randomization and then yearly. Each assessment was to include abdominal and pelvic CT or MRI and either thoracic radiography or thoracic CT or MRI. Disease-free survival was defined as the time between randomization and the first relapse, a second primary colon cancer, death from any cause when no evidence of relapse was recorded, or the last date at which the patient was known to be free of disease (censoring time). Relapse-free survival was defined as the time between randomization and the first relapse, a second primary colon cancer, death due to treatment-related toxic effects, or colon cancer if relapse had not been reported. Data on patients without documented relapse or with death unrelated to colon cancer or the study treatment were censored as of the last date on which the patient was known to be free of disease. Overall survival was defined as the time from randomization to death or the date at which the patient was last confirmed to be alive (censoring time).

EVALUATION OF SAFETY

The predefined primary end point for safety was at least equivalence as demonstrated through comparison of Kaplan–Meier estimates of the incidence and onset of all predefined severe (grade 3 or 4) toxic effects of the fluoropyrimidine (i.e., diarrhea, stomatitis, nausea, vomiting, hand–foot syndrome, alopecia, and neutropenia) in the two groups. Adverse events were recorded as previously described.[26] The intensity of and adjustments to the dose of the study drug were recorded throughout treatment.

STATISTICAL ANALYSIS

The intention-to-treat population included all patients who underwent randomization. In accordance with the study protocol, the per-protocol population excluded patients receiving less than 12 weeks of treatment or less than 50 percent of the planned dose of the study drug during this initial period as well as those with major violations of inclusion or exclusion criteria. The population included in the safety analysis comprised all patients receiving at least one dose of the study drug who were followed up for safety. Results of the per-protocol analysis supported the same conclusions as the intention-to-treat analyses and are not presented.

The primary efficacy analysis was planned when 632 events for the end point of three-year disease-free survival had occurred in the per-protocol population. The use of a noninferiority margin of 1.25 for the hazard ratio and a type I error of 2.5 percent ensured 80 percent power to show at least equivalence between the two study treatments. Assuming three-year disease-free survival rates of 70 percent, and allowing for approximately 15 percent of patients to be excluded from the per-protocol population, an enrollment of 1956 patients was planned. A second hierarchical test evaluated equivalence in disease-free survival with an upper limit of the hazard ratio of 1.20. If these analyses proved to be positive, tests for superiority were planned. Analyses for at least equivalence were performed in the per-protocol and intention-to-treat populations; superiority analyses were performed only in the intention-to-treat population, to maintain the most conservative approach. No interim analyses were performed.

Disease-free survival and overall survival were analyzed with the use of proportional-hazards regression and presented as Kaplan–Meier estimates and hazard ratios with 95 percent confidence intervals. Relapse-free survival was analyzed with the use of proportional-hazards regression and presented as a cumulative-incidence plot and hazard ratios with 95 percent confidence intervals. Planned multivariate analyses to evaluate the robustness of the data on disease-free, relapse-free, and overall survival were based on proportional-hazards regression. Subgroup analyses of disease-free survival were also prospectively planned.

The study (trial M66001) was designed and initiated by investigators and employees of the sponsor, Hoffmann–La Roche. The data were collected, managed, and analyzed by the sponsor. The article was prepared by the primary author, with editorial assistance from a medical writer (who was not an employee of the sponsor), on the basis of data and statistical analyses provided by the sponsor. The contents were reviewed and approved by all authors. The decision to publish this report was made by the investigators and the sponsor. The sponsor placed no contractual restrictions on the publication of the data but retained the right to review them before submission of the manuscript for publication. Dr. Twelves vouches for the accuracy and completeness of this report.

RESULTS

PATIENT POPULATION AND FOLLOW-UP

Between November 1998 and November 2001, 1987 patients were enrolled at 164 centers worldwide. Of 1004 patients assigned to capecitabine, 12 percent were excluded from the per-protocol population, and of 983 patients assigned to fluorouracil plus leucovorin, 11 percent were excluded. The reasons for exclusion were balanced between the two groups. In both groups, the median follow-up was 3.8 years, which was the time from randomization to closing of the database for analysis (April 1, 2004). Overall, 33 patients were lost to follow-up (18 in the fluorouracil-plus-leucovorin group and 15 in the capecitabine group).

Baseline characteristics were similar in the two groups (Table 1). There were slightly more patients with carcinoembryonic antigen levels above the upper limit of normal at baseline in the capecitabine group than in the fluorouracil-plus-leucovorin group (8.6 percent vs. 7.0 percent). The proportion of patients with involvement of four or more regional lymph nodes (stage N2 disease), as opposed to involvement of one to three nodes (stage N1 disease), was slightly higher in the capecitabine group than in the fluorouracil-plus-leucovorin group (30.8 percent vs. 29.4 percent).

DISEASE-FREE SURVIVAL

Table 2 summarizes the results for the three major efficacy end points. The primary objective — the determination of whether capecitabine results in disease-free survival at least equivalent to that with fluorouracil plus leucovorin — was met (Fig. 1A). The hazard ratio comparing disease-free survival in the capecitabine group with that in the fluorouracil-plus-leucovorin group was 0.87 (95 percent confidence interval, 0.75 to 1.00). The upper limit of the confidence interval (1.0) was significantly below both predefined margins, 1.25 and 1.20, for at least equivalence ($P<0.001$ for both comparisons), providing confidence that capecitabine is at least as effective as fluorouracil plus leucovorin. The protocol-specified analysis for superiority showed a trend toward superior disease-free survival with capecitabine as compared with fluorouracil plus leucovo-

Table 1. Baseline Characteristics of Patients in the Intention-to-Treat Population.*

Characteristic	Capecitabine (N=1004)	Fluorouracil plus Levcovorin (N=983)
Sex (%)		
Male	54	54
Female	46	46
Age (yr)		
Median	62	63
Range	25–80	22–82
Age group (%)		
<70 yr	81	79
≥70 yr	19	21
ECOG performance score (%)		
0	85	85
1	15	15
Nodal status — (%)		
N1	69	71
N2	31	29
Tumor stage (%)†		
T1 or 2	10	10
T3	76	76
T4	14	14
Carcinoembryonic antigen level (%)		
≤ULN	83	85
>ULN	9	7
Missing data	8	8

* ECOG denotes Eastern Cooperative Oncology Group, and ULN upper limit of normal.
† Higher numbers indicate greater depth of tumor penetration through the bowel wall.

Table 2. Efficacy for the Major End Points over a Median Follow-up Period of 3.8 Years.*

End Point	Total No. of Patients	No. of Patients with Event	Hazard Ratio (95% CI)	P Value for Equivalence	P Value for Superiority
Disease-free survival					
Capecitabine	1004	348	0.87 (0.75–1.00)	<0.001†	0.05
Fluorouracil plus leucovorin	983	380			
Relapse-free survival					
Capecitabine	1004	327	0.86 (0.74–0.99)	—	0.04
Fluorouracil plus leucovorin	983	362			
Overall survival					
Capecitabine	1004	200	0.84 (0.69–1.01)	<0.001‡	0.07
Fluorouracil plus leucovorin	983	227			

* P values for equivalence are one-sided; P values for superiority were calculated with the use of the Wald chi-square test. CI denotes confidence interval.
† The upper limit of the hazard ratio was compared with the noninferiority margin of 1.20, as prespecified in the study protocol.
‡ The upper limit of the hazard ratio was compared with the noninferiority margin of 1.25, as prespecified in the study protocol.

rin (P=0.05). The difference between the three-year rates of disease-free survival (a prespecified end point) in the capecitabine group (64.2 percent) and in the fluorouracil-plus-leucovorin group (60.6 percent) was not significant (P=0.12).

RELAPSE-FREE SURVIVAL

The definition of relapse-free survival was similar to that of disease-free survival, except that patients without a documented relapse and patients who died from a cause unrelated to colon cancer or the study treatment were censored (21 events in the capecitabine group and 18 in the fluorouracil-plus-leucovorin group). Relapse-free survival in the capecitabine group was longer than in the fluorouracil-plus-leucovorin group (P=0.04; hazard ratio, 0.86; 95 percent confidence interval, 0.74 to 0.99). Figure 1B shows the cumulative incidence of relapse (or of death due to the study treatment or colon cancer in patients who had no evidence of relapse). The three-year rates of relapse-free survival (not a prespecified end point) were 65.5 percent in the capecitabine group and 61.9 percent in the fluorouracil-plus-leucovorin group (P=0.12).

OVERALL SURVIVAL

Overall survival in the two groups did not differ significantly (P=0.07) (Fig. 1C). The hazard ratio for death in the capecitabine group as compared with the fluorouracil-plus-leucovorin group was not statistically significant (0.84; 95 percent confidence interval, 0.69 to 1.01). The three-year rates of overall survival (not a prespecified end point) were 81.3 percent and 77.6 percent in the capecitabine group and the fluorouracil-plus-leucovorin group, respectively (P=0.05).

MULTIVARIATE ANALYSES

Included in the prospectively planned multivariate analyses were potentially relevant factors (i.e., treatment, age, sex, nodal status, time from surgery to randomization, elevated carcinoembryonic antigen level) identified from previous trials.[5,6,11] These analyses showed that treatment with capecitabine had a statistically significant effect on disease-free survival (hazard ratio, 0.826; 95 percent confidence interval, 0.709 to 0.962; P=0.01) and overall survival (hazard ratio, 0.788; 95 percent confidence interval, 0.643 to 0.964; P=0.02), as compared with treatment with fluorouracil plus leucovorin. Exploratory analyses suggested that the difference between the results of the multivariate analyses and the unadjusted efficacy analysis was driven by small imbalances in baseline levels of carcinoembryonic antigen and extent of nodal involvement, which favored the fluorouracil-plus-leucovorin group. Other significant variables associated with improved disease-free survival in the multivariate analyses included female sex (hazard ratio, 0.764; 95 percent confidence interval, 0.653 to 0.893), stage N1 disease (hazard ratio, 0.583; 95 percent confidence interval, 0.497 to 0.683), and normal carcinoembryonic antigen levels

(hazard ratio, 0.389; 95 percent confidence interval, 0.312 to 0.485).

SUBGROUP ANALYSES

Subgroup analyses of disease-free survival showed a consistent trend toward benefit from capecitabine over fluorouracil plus leucovorin among the subgroups categorized according to prognostic factors that were used in the multivariate analysis (Fig. 2).

TREATMENT

The median number of chemotherapy cycles received (during the planned 24-week course) was eight in the capecitabine group and six in the fluorouracil-plus-leucovorin group. Eighty-three percent of patients in the capecitabine group and 87 percent in the fluorouracil-plus-leucovorin group completed treatment as planned. The median dose intensity delivered was 93 percent of that planned for capecitabine and 92 percent of that planned for fluorouracil plus leucovorin. In 57 percent of patients receiving capecitabine and 52 percent of those receiving fluorouracil plus leucovorin, the dose of the study drug required modification (for delay, dose reduction, or interruption of treatment). In a similar proportion of patients receiving capecitabine or fluorouracil plus leucovorin, the dose required reduction (42 percent and 44 percent, respectively). More interruptions (15 percent vs. 5 percent) and delays (46 percent vs. 29 percent) were required with capecitabine. Nevertheless, most patients in the capecitabine group completed at least four of the eight chemotherapy cycles without a reduction in the dose of the medication (76 percent vs. 68 percent in the fluorouracil-plus-leucovorin group after three of the six chemotherapy cycles), supporting the use of the standard starting dose in this trial.

PRIMARY SAFETY END POINT

Safety data (excluding the primary safety end point) were reported comprehensively 19 months after the last patient was enrolled.[26] The onset of the predefined key grade 3 or 4 toxic effects was significantly reduced throughout treatment with capecitabine as compared with treatment with fluorouracil plus leucovorin (P<0.001) (Fig. 3). The onset of toxic effects was also delayed with capecitabine as compared with fluorouracil plus leucovorin. Table 3 shows the incidence of the most common treatment-related clinical adverse events and laboratory abnormalities.

A score for global health status on the Quality of Life Questionnaire was prespecified as the primary

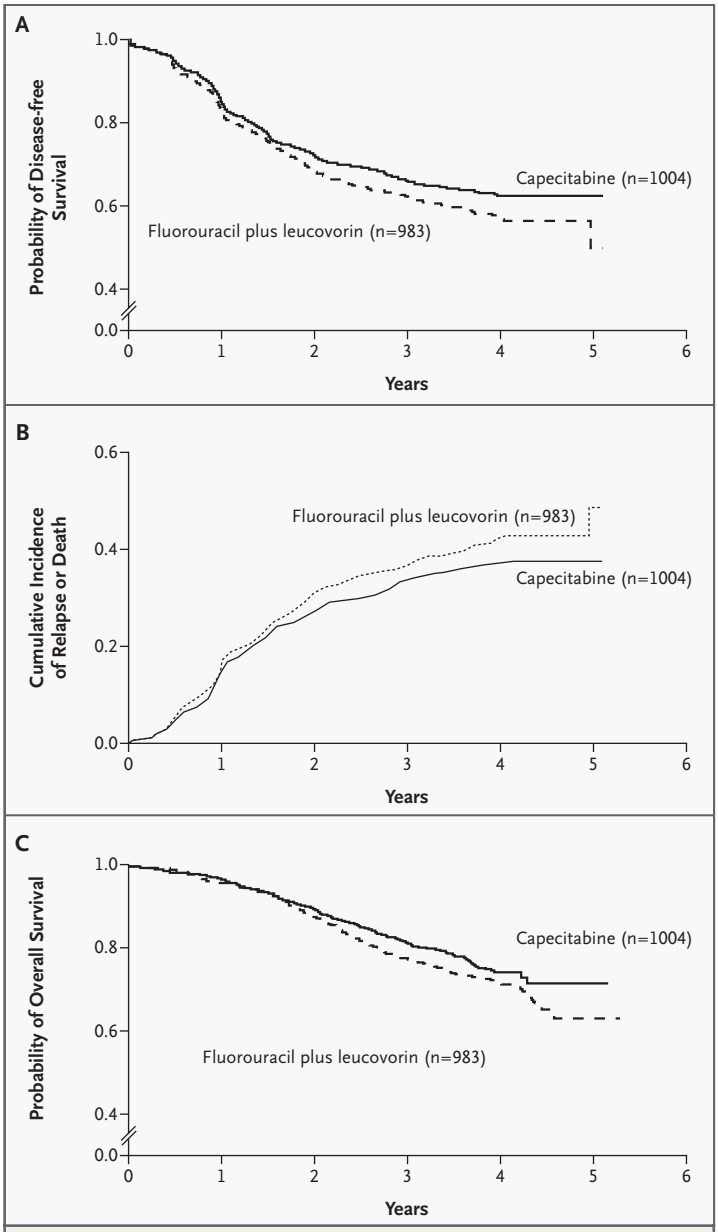

Figure 1. Disease-free Survival, Incidence of Relapse or Death, and Overall Survival among Patients Receiving Fluorouracil plus Leucovorin or Capecitabine (Intention-to-Treat Population).

Panel A shows Kaplan–Meier estimates of disease-free survival. The upper limit of the confidence interval of the hazard ratio was significantly below both the predefined margins, 1.25 and 1.20, for equivalence (P<0.001 in both cases). The analysis for superiority showed a trend favoring capecitabine (hazard ratio, 0.87 [95 percent confidence interval, 0.75 to 1.00]; P=0.05). Panel B shows the cumulative incidence of relapse or death; only deaths related to colon cancer or the study treatment were included. A Cox proportional-hazards model showed that relapse-free survival in the capecitabine group was statistically superior to that in the fluorouracil-plus-leucovorin group (P=0.04; hazard ratio, 0.86; 95 percent confidence interval, 0.74 to 0.99). Panel C shows Kaplan–Meier estimates of overall survival. The analysis for survival showed a trend favoring capecitabine (hazard ratio, 0.84 [95 percent confidence interval, 0.69 to 1.01]; P=0.07).

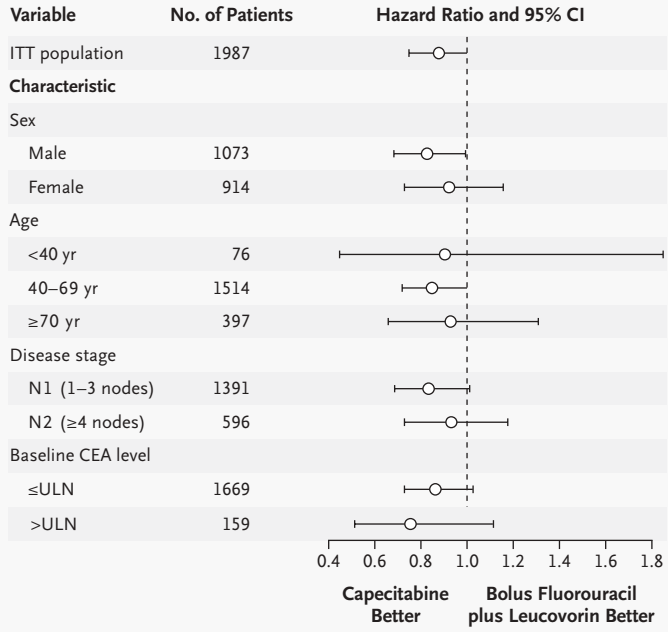

Figure 2. Subgroup Analysis of Disease-free Survival in the Capecitabine Group as Compared with the Fluorouracil-plus-Leucovorin Group (Intention-to-Treat Population).

Data on carcinoembryonic antigen (CEA) levels were missing for 159 patients who were therefore not included in the analysis for this variable. ITT denotes intention to treat, N nodal status, and ULN upper limit of normal.

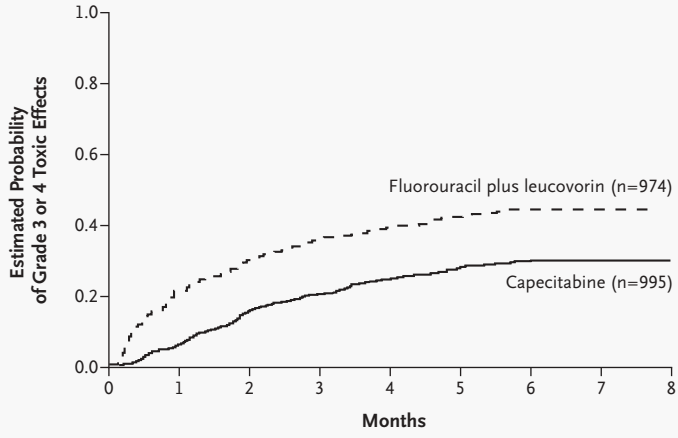

Figure 3. Kaplan–Meier Estimates of the Onset of Predefined Key Grade 3 or 4 Toxic Effects of Fluoropyrimidine.

P<0.001 for the difference between the groups.

measure of the quality of life. In the two groups, the scores remained relatively constant over time. However, at week 25 of treatment, the mean scores for global health status in the two groups showed similar small increases from baseline (<5 percent in raw scores), indicating improvement in the quality of life.

DISCUSSION

This randomized phase 3 trial showed that disease-free survival among patients who received oral capecitabine was at least equivalent to that among those who received fluorouracil plus leucovorin by intravenous bolus as adjuvant treatment for stage III colon cancer. Predefined multivariate analyses reinforced the primary efficacy findings. Although unadjusted analyses of disease-free survival and overall survival showed noninferiority of capecitabine to fluorouracil plus leucovorin, the multivariate analyses suggested that treatment with capecitabine improved the efficacy outcomes. We speculate that small imbalances in the two baseline characteristics with the strongest prognostic influence (i.e., elevated carcinoembryonic antigen levels and nodal status), which favored the fluorouracil-plus-leucovorin group, may have reduced the effect of capecitabine in the unadjusted analyses. Multivariate analyses also confirmed the prognostic significance of female sex, extent of nodal involvement, and elevated baseline carcinoembryonic antigen levels with regard to the three efficacy end points identified in previous trials; as expected, age influenced only overall survival. The only factor identified from previous trials that did not influence outcomes in the current study was time from surgery to randomization. However, because the eligibility criteria in this trial specified an interval of eight weeks or less between surgery and randomization, variability in this measure was limited in ours as compared with earlier trials.

The results with the Mayo Clinic regimen in our trial were consistent with those in previous studies. If patients with disease stage III are isolated from the more mixed population of the INT-0089 trial,[8] which is the only other study of similar size using this regimen, the three-year disease-free survival rate (63 percent) is similar to that in the group receiving the Mayo Clinic regimen in the current trial.

The significantly lower incidence and delayed

onset of fluoropyrimidine-related grade 3 or 4 toxic effects with capecitabine as compared with fluorouracil plus leucovorin supports the favorable safety data reported with regard to patients with metastatic disease.[26] Overall, there were significantly lower incidences of neutropenia and stomatitis and lower rates of nausea, vomiting, alopecia, and diarrhea in the settings of adjuvant treatment and metastatic disease with capecitabine.[26] The incidence of grade 3 hand–foot syndrome was, however, significantly higher with capecitabine than with fluorouracil plus leucovorin. The higher number of dose delays and interruptions of treatment in the capecitabine group reflected the schedule of twice-daily oral administration. An important element in this approach is educating patients to recognize toxic effects of grade 2 or greater severity and interrupting treatment promptly.

The Multicenter International Study of Oxaliplatin/5-Fluorouracil/Leucovorin in the Adjuvant Treatment of Colon Cancer (better known as MOSAIC) trial showed that adding oxaliplatin to infused fluorouracil plus leucovorin provides a 23 percent reduction in the risk of recurrence, which is a significant and clinically meaningful benefit.[27] Analysis of the survival data is premature, but a meta-analysis of the use of adjuvant fluorouracil has shown that disease-free survival is predictive of overall survival.[28] The safety of infused fluorouracil, leucovorin, and oxaliplatin was acceptable, but peripheral neuropathy, myelosuppression, and gastrointestinal disturbances were significantly more common among those also receiving oxaliplatin than among those receiving infused fluorouracil plus leucovorin alone.

The X-ACT trial shows that capecitabine is at least equivalent to the Mayo Clinic regimen of fluorouracil plus leucovorin in patients younger than 70 years and those 70 years of age or older. The safety advantage of capecitabine over fluorouracil plus leucovorin was also maintained in these subgroups.[29] Our results support capecitabine as an alternative to fluorouracil plus leucovorin in the adjuvant treatment of colon cancer. Capecitabine or oxaliplatin-based therapy should be considered for all patients requiring adjuvant therapy for colon cancer.

Table 3. Most Common Treatment-Related Adverse Events.*

Event	All Grades of Events		Grade 3 or 4 Events (Severe)	
	Capecitabine (N=995)	Fluorouracil plus Leucovorin (N=974)	Capecitabine (N=995)	Fluorouracil plus Leucovorin (N=974)
	percent			
Diarrhea	46†	64	11	13
Nausea or vomiting	36†	51	3	3
Stomatitis	22†	60	2†	14
Hand–foot syndrome	60†	9	17†	<1
Fatigue or asthenia	23	23	1	2
Abdominal pain	10	13	2	1
Alopecia	6†	22	0‡	<1
Lethargy	10	9	<1	<1
Anorexia	9	10	<1	<1
Neutropenia§	32†	63	2†	26
Hyperbilirubinemia§	50†	20	20†	6

* Treatment-related adverse events that occurred in 10 percent or more of patients were included in the safety analysis. The data shown are an update to the data of Scheithauer et al.[26]
† P<0.001.
‡ P=0.02.
§ Diagnosis was based on laboratory values.

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APPENDIX

The following investigators participated in the X-ACT trial. **Argentina** — E. Mickiewicz, G. Pallotta, E. Roca, M.S. Varela, R.C. Wainstein; **Australia** — E. Abdi, A. Barling, S. Begbie, D. Bell, R. Blum, W.I. Burns, P. de Souza, D. Kotasek, J. Levi, K. Pittman, M. Schwarz, C. Underhill, D. Wyld; **Austria** — P. Balcke, M. Baur, D. Geissler, P. Kier, H. Ludwig, K. Mach, D. Öfner, M. Prager, H. Steiner; **Belgium** — J. De Grève, D. Vanstraelen; **Brazil** — L. Camillo-Coura, G. Delgado, S. Lago, A. Malzyner, C. Rotstein; **Canada** — J.P. Ayoub, O. Keller, K. Khoo, R. Rajan, A. Sami, R. Wong; **Croatia** — M. Duvnjak, Z.K. Osijek, R. Ostojic, E. Vrdoljak; **Czech Republic** — I. Bustova, J. Dvorak, J. Fínek, I. Kocakova, M. Kůta, J. Nemec, V. Svoboda, P. Vodvarka; **France** — F.X. Caroli-Bosc, G. Dabouis, J.-Y. Douillard, P. Dufour, E. Gamelin, J.L. Gaudin, M. Giovannini, H. Gouerou, J.E. Kurtz, C. Lombard-Bohas, D. Peré-Vergé, M. Ychou; **Germany** — W. Abenhardt, A. Beham, R. Behrens,

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ORIGINAL ARTICLE

Daclizumab to Prevent Rejection after Cardiac Transplantation

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ABSTRACT

BACKGROUND

Daclizumab, a humanized monoclonal antibody against the interleukin-2 receptor, reduced the risk of rejection without increasing the risk of infection among renal-transplant recipients and, in a single-center trial, among cardiac-transplant recipients. We conducted a multicenter, placebo-controlled, double-blind study to confirm these results in cardiac-transplant patients.

METHODS

We randomly assigned 434 recipients of a first cardiac transplant treated with standard immunosuppression (cyclosporine, mycophenolate mofetil, and corticosteroids) to receive five doses of daclizumab or placebo. The primary end point was a composite of moderate or severe cellular rejection, hemodynamically significant graft dysfunction, a second transplantation, or death or loss to follow-up within six months.

RESULTS

By six months, 104 of 218 patients in the placebo group had reached the primary end point, as compared with 77 of the 216 patients in the daclizumab group (47.7 percent vs. 35.6 percent, P=0.007), a 12.1 percent absolute risk reduction and a 25 percent relative reduction. The rate of rejection was lower in the daclizumab group than in the placebo group (41.3 percent vs. 25.5 percent). Among patients reaching the primary end point, the median time to the end point was almost three times as long in the daclizumab group as in the placebo group during the first 6 months (61 vs. 21 days) and at 1 year (96 vs. 26 days). More patients in the daclizumab group than in the placebo group died of infection (6 vs. 0) when they received concomitant cytolytic therapy.

CONCLUSIONS

Daclizumab was efficacious as prophylaxis against acute cellular rejection after cardiac transplantation. Because of the excess risk of death, concurrent or anticipated use of cytolytic therapy with daclizumab should be avoided.

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Cardiac transplantation has become commonplace in the 22 years since the approval of cyclosporine. Nevertheless, progress in immunosuppression has been slow, in part because the heart is a vital organ and acute allograft rejection can include hemodynamic compromise, irreversible graft injury, and death. Furthermore, the immunosuppressive therapy used to prevent rejection predisposes patients to infection, which continues to be the leading cause of death in the year after cardiac transplantation.[1,2] In a prior report of a single-center evaluation of 55 heart-transplant recipients, there was a striking decrease in the rate of rejection, by a factor of 2.8, among patients receiving daclizumab, a monoclonal antibody against the α subunit of the interleukin-2 receptor, with no increase in the rate of infection.[3]

A common immunosuppression protocol for cardiac transplantation includes cyclosporine, mycophenolate mofetil, and corticosteroids (triple therapy). The results of a multicenter, double-blind, placebo-controlled clinical trial involving 650 cardiac-transplant recipients[4] suggested that treatment with mycophenolate mofetil, a purine analogue, reduced the rate of rejection and improved survival but had a higher incidence of nonfatal, opportunistic infections as compared with azathioprine therapy.

An alternative to standard triple therapy at the time of cardiac transplantation has been the use of augmented immunosuppression, commonly termed "induction immunotherapy," with antilymphocyte antibodies. These cytolytic agents, used in more than 40 percent of initial heart transplantations,[2] include the murine monoclonal antibody muromonab-CD3 and other antithymocyte or antilymphocyte agents (e.g., ATGAM and Thymoglobulin). Retrospective evaluations from a large, multiinstitutional database have suggested that cytolytic therapy reduces the risk of early rejection but increases the risk of infection.[1,5] Despite the widespread use of induction immunotherapy, no randomized, multicenter trial has been conducted in cardiac-transplant recipients.

Daclizumab functions as an immunosuppressant by antagonizing interleukin-2–mediated proliferation of T cells.[6] Daclizumab has been shown to decrease the risk of rejection of renal transplants, with no increased incidence of infection.[7,8] On the basis of a prior report of decreased rejection rates,[3] we conducted a multicenter, double-blind, randomized, placebo-controlled trial designed to test the efficacy of daclizumab induction immunotherapy as prophylaxis against cardiac rejection.

METHODS

PATIENTS

Patients older than 13 years of age receiving their first cardiac allograft were recruited at 31 transplantation centers in the United States, Canada, Germany, and Sweden between August 28, 1999, and April 29, 2001, and after transplantation were randomly assigned in a double-blind manner to receive either daclizumab or placebo in combination with cyclosporine, mycophenolate mofetil, and corticosteroids. Patients known to be in need of cytolytic therapy at the time of transplantation or those requiring ventricular assist devices after surgery were excluded. Each participating institutional review board approved the study protocol, and written informed consent was obtained from all patients.

TREATMENTS

Daclizumab, 1 mg per kilogram of body weight (up to 100 mg), or placebo was administered intravenously within 12 hours after transplantation and on days 8, 22, 36, and 50. Maintenance immunosuppressive therapy included mycophenolate mofetil (1.5 g twice daily) and a microemulsion of cyclosporine (1 to 4 mg per kilogram intravenously or 2 to 6 mg per kilogram orally, initiated within 72 hours after transplantation). The dose of mycophenolate mofetil could be adjusted in the event of adverse effects. The doses of cyclosporine were adjusted to maintain the usual trough levels at each center. Methylprednisolone (500 to 1000 mg) was intravenously administered perioperatively, or oral prednisone or its equivalent was given at a daily dose of 0.5 to 1 mg per kilogram through day 14; the dose was reduced to 0.2 mg per kilogram by day 28 and to 0.10 to 0.15 mg per kilogram by day 90. Corticosteroids could be discontinued electively after day 180. All patients received statins. Antibiotics to prevent *Pneumocystis carinii* pneumonia were administered for one year. Patients at high risk for cytomegalovirus (CMV) disease (CMV-negative recipients whose donors were CMV-positive) received prophylactic ganciclovir (up to 5 to 10 mg per kilogram per day for two weeks, followed by treatment according to the practice of the individual center).

BIOPSY

Endomyocardial biopsies were performed on the days of the second through fifth daclizumab infusions and then every 2 weeks up to 3 months, monthly up to 6 months, and then every 2 months up to 12 months after transplantation. Biopsy specimens were read by the local center pathologist in accordance with the criteria of the International Society for Heart and Lung Transplantation (ISHLT).[9]

END POINTS

The primary efficacy end point was a composite end point at six months consisting of the first of any one of the following: a biopsy showing acute cellular rejection of grade 3A or higher; hemodynamic compromise, treated with inotropic agents and pulsed doses of immunosuppressants, whether or not a biopsy was done and regardless of the grade of the biopsy; death; a second transplantation; or loss to follow-up. Hemodynamic compromise was defined by any or all of the following: an ejection fraction of 30 percent or less or a 20 percent decrease in the ejection fraction from baseline, a fractional shortening of no more than 20 percent or a 25 percent decrease from baseline, or a cardiac index of less than 2.0 liters per minute per square meter or a 25 percent decrease from baseline. Secondary efficacy end points included the composite end point at 12 months, patient and graft survival at 6 and 12 months, and the time to the first occurrence of the composite end point within 6 and 12 months.

STATISTICAL ANALYSIS

The number of patients enrolled in the study was based on a two-sided alpha level of 2.5 percent, an assumed rate of acute rejection of 40 percent in the placebo group, and a statistical power of 80 percent to detect a 40 percent reduction in the rejection rate in the daclizumab group, with an expected dropout rate of 20 percent. The sample size was calculated with the use of the normal approximation of the binomial, with continuity correction.

The Cochran–Mantel–Haenszel general association test, with stratification according to center, was used to test the null hypothesis of no difference between groups. A 95 percent confidence interval was calculated for the difference in the weighted averages (weighted according to center size) of the rejection rates. Logistic regression was used to test for interactions between center and treatment group. The time-to-event data were analyzed with the use of Kaplan–Meier product-limit estimates. Dosing information was summarized at 6 months and 12 months and compared in the two groups by means of the blocked Wilcoxon rank-sum test. Analyses of efficacy were conducted according to the intention to treat unless otherwise noted. Analyses of safety included only patients who had received at least one

Table 1. Baseline Characteristics of the Patients and Donors.*

Characteristic	Placebo Group (N=218)	Daclizumab Group (N=216)	P Value
Patients			
Male sex — no. (%)	177 (81.2)	171 (79.2)	0.60
Race or ethnic group — no. (%)†			0.28
White	193 (88.5)	185 (85.6)	
Black	15 (6.9)	23 (10.6)	
Asian	2 (0.9)	0	
Other	8 (3.7)	8 (3.7)	
Age — yr			
Mean	53.1±11.9	52.4±11.0	0.54
Median	56.0	54.0	
Range	13–74	18–72	
Weight — kg	79.9±15.6	80.0±13.9	0.94
Indication for transplantation — no. (%)			0.88
Coronary artery disease	68 (31.2)	64 (29.6)	
Dilated cardiomyopathy	138 (63.3)	138 (63.9)	
Other	12 (5.5)	14 (6.5)	
UNOS waiting-list status or equivalent — no. (%)‡			0.85
1A	50 (22.9)	46 (21.3)	
1B	83 (38.1)	88 (40.7)	
2	83 (38.1)	82 (38.0)	
Intraaortic balloon pump — no. (%)	8 (3.7)	18 (8.3)	0.04
Left ventricular assist device — no. (%)	16 (7.3)	18 (8.3)	0.70
Most recent PRA <10% — no. (%)	205 (94.0)	194 (89.8)	0.11
CMV status — no. (%)			
Donor and recipient positive	136 (62.4)	136 (63.0)	0.90
Donor positive, recipient negative	50 (22.9)	47 (21.8)	0.77
Donors			
Duration of cold ischemia — min	192.0±67.2	186.0±57.0	0.32
Age — yr			0.08
Mean	29.8±12.5	32.0±13.2	
Median	26.0	28.0	

* Plus–minus values are means ±SD. UNOS denotes United Network for Organ Sharing, PRA panel reactive antibody test, and CMV cytomegalovirus.
† Race or ethnic group was determined by patient report.
‡ Data were unavailable for two patients in the placebo group.

Table 2. Efficacy Results at 6 and 12 Months.*

Result	Placebo Group (N=218)	Daclizumab Group (N=216)	P Value
	no. (%)		
6 Months			
Primary end point	104 (47.7)	77 (35.6)	0.007
First biopsy-proven rejection	90 (41.3)	55 (25.5)	
ISHLT grade 3A	78 (35.8)	50 (23.1)	
ISHLT grade 3B	11 (5.0)	4 (1.9)	
ISHLT grade 4	1 (0.5)	1 (0.5)	
Hemodynamic compromise	3 (1.4)	6 (2.8)	
Death	7 (3.2)	14 (6.5)	
Second transplantation	0	0	
Lost to follow-up	4 (1.8)	2 (0.9)	
12 Months			
Composite end point	116 (53.2)	97 (44.9)	0.06
First biopsy-proven rejection	101 (46.3)	73 (33.8)	
ISHLT grade 3A	89 (40.8)	67 (31.0)	
ISHLT grade 3B	11 (5.0)	5 (2.3)	
ISHLT grade 4	1 (0.5)	1 (0.5)	
Hemodynamic compromise	3 (1.4)	6 (2.8)	
Death	8 (3.7)	16 (7.4)	
Lost to follow-up	4 (1.8)	2 (0.9)	

* The composite clinical end point was the first occurrence of any of the following: cellular rejection, hemodynamically significant graft dysfunction, a second transplantation, death, or loss to follow-up (see the Methods section for additional details). Data on efficacy results at 6 and 12 months were censored at the time of the first event. ISHLT denotes International Society for Heart and Lung Transplantation.

dose of placebo or daclizumab; if a patient received daclizumab any time after randomization, he or she was included in the daclizumab group.

The study was designed by Roche Laboratories, with the assistance of a subgroup of the principal investigators (listed in the Appendix). The data were collected and held by Roche Laboratories. The writing committee had full access to the data and was fully involved in the data analysis, which was performed by the sponsor's statistician. The committee vouches for the veracity and completeness of the reported data.

RESULTS

PATIENT POPULATION

The 434 patients were well matched at baseline (Table 1). There were no significant differences between groups in the percentages of ABO-identical or ABO-compatible allografts and HLA-A, B, or DR antigen mismatches (data not shown).

Adherence to the study protocol was excellent, with 81.8 percent of patients receiving at least four doses of study drug according to the protocol. The concomitant use of maintenance immunosuppression was well balanced: the median daily dose of cyclosporine was 355 mg in the placebo group and 344 mg in the daclizumab group from day 0 to 90 and 313 mg and 300 mg, respectively, from day 91 to 180; the median daily dose of mycophenolate mofetil was 2915 mg and 2918 mg, respectively, from day 0 to 90 and 2716 mg and 3000 mg, respectively, from day 91 to 180; and the median cumulative doses of maintenance corticosteroids were 3401 mg and 3354 mg, respectively, from day 0 to 90 and 900 mg and 855 mg, respectively, from day 91 to 180. By six months, 31 patients in the placebo group (14.2 percent) and 24 patients in the daclizumab group (11.1 percent) were not taking corticosteroids.

EFFICACY

By six months, 104 of the 218 patients in the placebo group had reached the primary end point, as compared with 77 of the 216 patients in the daclizumab group (47.7 percent vs. 35.6 percent), a 12.1 percent absolute risk reduction and a 25 percent relative reduction (P=0.007) (Table 2 and Fig. 1). Corticosteroids were used to treat rejection in 111 placebo recipients, as compared with 78 daclizumab recipients (50.9 percent vs. 36.1 percent). In the group of patients who met the primary end point, the observed difference was heavily influenced by the smaller number of daclizumab-treated patients who had acute rejection of ISHLT grade 3A or higher (55, as compared with 90 in the placebo group; 25.5 percent vs. 41.3 percent) (Table 2). Among the patients who reached the primary end point during the first six months of follow-up, the median time to reach the end point was approximately three times as long among patients in the daclizumab group as among patients in the placebo group (61 vs. 21 days), and this favorable outcome was also observed during the first year of follow-up (median time to reach the secondary end point, 96 vs. 26 days).

OPPORTUNISTIC INFECTIONS AND CANCER

The incidence of any opportunistic infection, including CMV infection and CMV disease, was similar in the two groups (Table 3). Cancer rates were also similar in the two groups (Table 3).

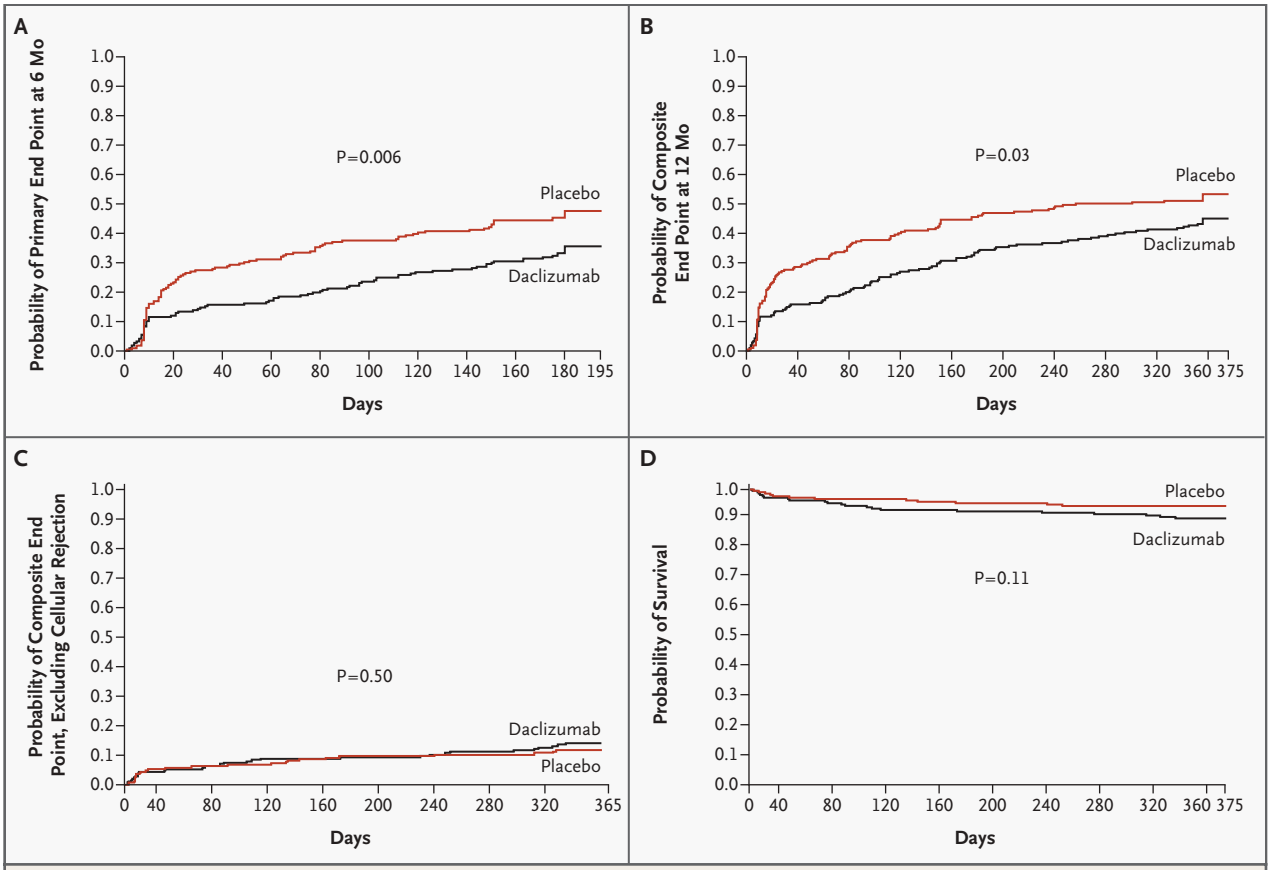

Figure 1. Kaplan–Meier Estimates of the Probability of Reaching the Primary End Point at 6 Months (Panel A), the Probability of Reaching the Composite End Point at 12 Months (Panels B and C), and the Probability of Survival (Panel D).

In Panel A, the primary end point was a composite of cellular rejection, hemodynamically significant graft dysfunction, a second transplantation, death, or loss to follow-up by six months. The log-rank test was used to compare differences between groups. In Panel C, the composite end point excluded cellular rejection. In Panel D, 12 of the 218 patients in the placebo group died, as compared with 21 of the 216 patients in the daclizumab group (5.5 percent vs. 9.7 percent, P=0.11 by the log-rank test). The analyses shown in Panels A, B, and C include all randomized patients, whereas the analysis shown in Panel D includes only patients who received at least one dose of the study drug.

MORTALITY

At one year, Kaplan–Meier estimates of survival according to the intention to treat did not differ significantly between groups (Fig. 1D), even though the majority of patients had a United Network for Organ Sharing (UNOS) waiting-list status of 1A or 1B (Table 1). At six months, 10 patients in the placebo group had died from various causes according to the intention to treat, as compared with 16 in the daclizumab group; at one year, the numbers were 12 and 21, respectively (Table 4). Mortality rates were similar when analyzed according to the actual treatment received (Table 4).

Deaths from infection were more frequent among patients in the daclizumab group who also received cytolytic therapy than among patients in the placebo group who also received cytolytic therapy (6 of 40 [15.0 percent] vs. 0 of 37 [0 percent]) (Table 4). Although cytolytic therapy was prohibited by the study protocol (other than to treat rejection), patients were required to undergo randomization and receive the study drug within 12 hours after their return to the intensive care unit after the heart transplantation.

Predictably, some patients had postoperative changes in their clinical course after randomization, most commonly worsening renal function, for which investigators discontinued cyclosporine and substituted renal-sparing cytolytic therapy (Table 4). Another common indication for cytolytic therapy was suspected acute rejection. A total of 77 patients received cytolytic therapy (37 in the placebo group

Table 3. Rates of Opportunistic Infection or Cancer.*

Variable	Placebo Group (N=207)	Daclizumab Group (N=216)
	no. of patients (%)	
Serious opportunistic infection in 1st 6 mo†	16 (7.7)	15 (6.9)
Bronchopulmonary aspergillosis	1 (0.5)	1 (0.5)
Candida	2 (1.0)	4 (1.9)
CMV	12 (5.8)	11 (5.1)
Herpes zoster	1 (0.5)	1 (0.5)
Any opportunistic infection in 1st 12 mo†	80 (38.6)	71 (32.9)
Bronchopulmonary aspergillosis	2 (1.0)	2 (0.9)
Candida	18 (8.7)	20 (9.3)
CMV	50 (24.2)	43 (19.9)
Herpes simplex	15 (7.2)	12 (5.6)
Herpes zoster	11 (5.3)	9 (4.2)
Cryptococcal meningitis	0	1 (0.5)
Cancer		
By 6 mo	5 (2.4)	7 (3.2)
By 12 mo	11 (5.3)	11 (5.1)
Post-transplantation lymphoproliferative disorder	1 (0.5)	1 (0.5)

* The analysis included all patients who received at least one dose of placebo or daclizumab. CMV denotes cytomegalovirus.
† Some patients had more than one type of opportunistic infection.

and 40 in the daclizumab group), most of them (81.8 percent) within the first 30 days after transplantation. Of the 21 patients in the daclizumab group who died, 8 also received cytolytic therapy within the first 30 days, and 6 of these 8 patients died from infection. In comparison, of the 11 patients in the placebo group who died, 2 received cytolytic therapy within the first 30 days, and neither died from infection.

DISCUSSION

This clinical trial was undertaken to evaluate the efficacy of daclizumab, a humanized monoclonal antibody that binds to the interleukin-2α receptor on activated lymphocytes, to reduce the risk of rejection in patients undergoing cardiac transplantation who were given triple-drug immunosuppressive therapy including cyclosporine, mycophenolate mofetil, and corticosteroids. The primary end point was a composite of any of the following within six months after transplantation: acute cellular rejection of ISHLT grade 3A or higher on endomyocardial biopsy, hemodynamically significant graft dysfunction requiring augmented immunosuppression and inotropes regardless of biopsy results, second transplantation, death, or loss to follow-up. The incidence of the primary end point at six months was 35.6 percent in the daclizumab group and 47.7 percent in the placebo group. This 12.1 percent absolute risk reduction resulted primarily from a reduction in the incidence of acute cellular rejection.

By binding the interleukin-2α receptor on the surface of activated T cells, daclizumab inhibits interleukin-2–mediated proliferation of T cells and thus has an immunosuppressive effect. One dose of daclizumab has an elimination half-life of approximately 20 days.[6] We administered five doses of daclizumab, the first in the immediate post-transplantation period, with four additional doses given through week 7; hence, an immunosuppressive effect was expected to persist for up to 120 days in adults (unpublished data).

Among patients who met the primary end point in the first six months, the median time to reach the end point was three times as long among patients in the daclizumab group as among patients in the placebo group. This prevention of rejection is clinically important, since the reduction in the risk of rejection occurred within the first few weeks after transplantation, when immunosuppression is most intensive and the need to treat acute cellular rejection, usually with high doses of corticosteroids, may increase the incidence of complications, such as impaired wound healing, infection, or glucose intolerance. The decreased overall incidence of rejection in the daclizumab group was attained without an increased incidence of any serious opportunistic infections. This finding has previously been reported in studies of daclizumab after renal transplantation.[7,8]

Survival exceeded 90 percent in both groups at one year and substantially exceeded the UNOS one-year survival rate of 85.3 percent among 4927 first-time cardiac-transplant recipients in the United States during the same period (1999 to 2001).[10] As compared with the UNOS study, we had fewer patients with a UNOS waiting-list status of 1A (22 percent, as compared with 36 percent), more patients with a status of 2 (38 percent, as compared with 27 percent), and similar numbers with a status of 1B (39 percent and 37 percent, respectively), which may have contributed in part to these favorable outcomes. However, in the UNOS study, the one-year survival rates were 81.1 percent among patients with a status of 1A, 87.2 percent among those with

Table 4. Rates of Death and Cytolytic Therapy.*

Variable	Placebo Group	Daclizumab Group	Absolute Difference (95% CI)
Intention-to-treat analysis (randomized population) — no. (%)			
Total no.	218	216	
Deaths			
6 Mo	10 (4.6)	16 (7.4)	2.8 (–2.10 to 7.74)
12 Mo	12 (5.5)	21 (9.7)	4.2 (–1.22 to 9.66)
Analysis according to treatment received (at least 1 dose of placebo or daclizumab) — no. (%)			
Total no.	207	216	
Deaths			
6 mo	10 (4.8)	16 (7.4)	2.6 (–2.45 to 7.60)
12 mo	11 (5.3)	21 (9.7)	4.4 (–1.06 to 9.88)
Causes of death — no.			
0–6 Mo†			
Sepsis	0	6	
Multiorgan failure	3	1	
Anoxic encephalopathy	0	2	
Cardiac arrest	2	0	
Other‡	5	7	
7–12 Mo			
Acute myocardial infarction	1		
CMV infection		1	
Cryptococcal meningitis		1	
Multiorgan failure		1	
Myocardial infarction		1	
Sudden death from cardiac causes		1	
Cytolytic therapy — no. (%)			
0–30 days	31 (15.0)	32 (14.8)	
For renal insufficiency	17 (8.2)	18 (8.3)	
For other indications	14 (6.8)	14 (6.5)	
Cumulative to 6 mo	35 (16.9)	35 (16.2)	
Cumulative to 12 mo	37 (17.9)	40 (18.5)	
Receipt of cytolytic therapy from 0–30 days — no./total no. (%)			
Death within 6 mo	2/31 (6.5)	4/32 (12.5)	
Death within 12 mo	2/31 (6.5)	8/32 (25.0)	
Receipt of cytolytic therapy from 0–12 mo — no./total no. (%)			
Death within 6 mo	2/37 (5.4)	8/40 (20.0)	
Death within 12 mo	2/37 (5.4)	8/40 (20.0)	
Death from infection	0	6/40 (15.0)	

* Cytolytic therapy included muromonab-CD3 and antithymocyte or antilymphocyte agents. CMV denotes cytomegalovirus.
† Causes of death did not differ significantly between groups in intention-to-treat analyses or analyses according to the treatment received.
‡ In the daclizumab group, each of the following caused one death: arteriovenous malformation, heart failure, cerebral hemorrhage, cerebral infarction, mediastinal abscess, pulmonary embolism, and squamous-cell carcinoma. In the placebo group, one death was caused by each of the following: brain herniation, mesenteric occlusion, shock, sudden death from cardiac causes, and thrombotic thrombocytopenic purpura.

a status of 1B, and 87.9 percent among those with a status of 2. Thus, even though our population had fewer patients with a status of 1A and more patients with a status of 2, the overall survival rate still substantially exceeded those in the contemporaneous UNOS study.

More patients in the daclizumab group than in the placebo group died during the first 12 months after transplantation. The annual mortality rate in the placebo group (5.3 percent) was similar to the 6.2 percent observed among 289 patients treated with the same mycophenolate mofetil dosing protocol in an earlier trial.[4] A comprehensive review of the clinical courses of the patients who died revealed that 8 of the 21 patients who died in the daclizumab group (38.1 percent) had also received cytolytic therapy and that 6 of these 8 patients (75.0 percent) had died from infection. Cytolytic therapy has previously been shown to be an independent risk factor for infection after transplantation.[1,5] In contrast, only 2 of the 11 patients who died in the placebo group had also received cytolytic therapy (18.2 percent), and neither death was associated with infection.

The cytolytic therapy given to the daclizumab group that most likely resulted in a very high level of immunosuppression was an unintended result of the double-blind study design. Some investigators, who did not know whether the patient had received daclizumab or placebo, used cytolytic therapy for complications in the early postoperative period, such as serious renal insufficiency necessitating delayed administration of cyclosporine. Therefore, the patients in the daclizumab group who also received cytolytic therapy had a high level of immunosuppression as a result of treatment with up to five drugs (the cytolytic agent, daclizumab, cyclosporine, mycophenolate mofetil, and corticosteroids), which may have increased the risk of serious infection. Furthermore, because of the double-blind study design, investigators continued administering study drug regardless of whether a patient was also receiving cytolytic therapy. This drug combination may have contributed to the increased numbers of patients who died from infection in this trial.

Our findings provide provocative although not definitive evidence of an increased risk of fatal infection when daclizumab is used in conjunction with cytolytic therapy. Such combined ongoing use of daclizumab after the administration of T-cell–depleting antibodies was not intended to be part of the trial design and does not represent the current standard of care. Combined use of daclizumab and cytolytic antibodies should be avoided in routine clinical practice, including in patients undergoing transplantation who are expected to require cytolytic therapy to avoid renal dysfunction from calcineurin inhibitors. Patients who receive daclizumab soon after transplantation and require treatment for suspected or biopsy-proven rejection should preferentially be treated with high-dose corticosteroids rather than cytolytic therapy. If the use of a T-cell–depleting antibody is thought to be indicated, daclizumab therapy should be discontinued.

The clinical relevance of the prespecified primary end point as opposed to other end points, such as long-term allograft survival or freedom from allograft vasculopathy, raises more fundamental questions regarding the prevention of moderate rejection. Evaluations of large registries have shown a relationship between the occurrence of cellular rejection and extended survival.[2] Our 6-month efficacy and 12-month safety design did not permit these issues to be addressed. Future studies of daclizumab and other immunosuppressive approaches in cardiac transplantation will need to account for these important long-term clinical outcomes and whether the use of surrogate markers (e.g., intravascular changes on ultrasonography) will permit truncation of follow-up.

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APPENDIX

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CLINICAL PRACTICE

Lung Cancer Screening

James L. Mulshine, M.D., and Daniel C. Sullivan, M.D.

This Journal *feature begins with a case vignette highlighting a common clinical problem. Evidence supporting various strategies is then presented, followed by a review of formal guidelines, when they exist. The article ends with the authors' clinical recommendations.*

A 60-year-old woman who quit smoking 20 years earlier comes for a routine visit. She previously smoked one pack of cigarettes a day for 10 years. Her medical history is otherwise unremarkable. She feels well and exercises regularly. Her husband smoked one pack of cigarettes per day for at least 30 years but stopped smoking a decade ago. She asks whether she and her husband should undergo computed tomographic (CT) scanning to screen for lung cancer. What do you advise?

THE CLINICAL PROBLEM

With a projection of more than 160,000 deaths from lung cancer this year, the disease now accounts for 30 percent of deaths from cancer in the United States.[1] Since metastatic spread to regional or distant sites is evident in at least three quarters of patients with lung cancer at the time of diagnosis, the five-year survival rate for lung cancer is only about 15 percent. For stage 1 lung cancer, the five-year survival rate exceeds 60 percent. Among women, there has been a 600 percent increase in the incidence of lung cancer during the past 80 years, and rates of death among women with lung cancer in the United States are the highest of any in the world.[2] About 50 percent of adults in the United States have smoked; half of them have quit. After smoking cessation, the risk of coronary artery disease drops promptly, whereas the risk of lung cancer does not.[3] The net result is that lung cancer has recently superseded coronary artery disease as the leading cause of death among current and former smokers.[4-8]

The results of randomized trials that were reported 20 years ago showed no significant reduction in deaths from lung cancer with the use of screening that included a combination of chest radiography and cytologic analysis of sputum. Although methodologic limitations in the studies (such as the frequent use of chest radiography in the control group and low compliance with the prescribed frequency of radiography in the experimental group) may have contributed to the negative results,[9,10] these findings discouraged routine screening for lung cancer with plain films. More recently, the possibility of early detection of lung cancer with high-resolution CT has renewed interest in lung cancer screening (Fig. 1).[11-14] However, as detailed below, decisions regarding the use of this technology must involve a consideration of potential benefits and risks along with the health care costs.[15-17]

STRATEGIES AND EVIDENCE

A number of pilot studies of CT screening have recently been reported. These studies have generally involved groups that are at increased risk for lung cancer, including current and former smokers. Analysis of these results is complicated by a lack of standardized criteria for eligibility and clinical management. Overall, 55 to 85 percent of cancers that are detected in baseline scans and 60 to 100 percent of cancers that are detected in

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annual follow-up scans are stage 1 (Table 1). In contrast, only 16 percent of cancers that are diagnosed in the course of routine clinical care in the United States are stage 1.[1,11,18-21] Since stage 1 lung cancer is the most curable form of this disease, a high frequency of detection of stage 1 tumors is considered a necessary (though not sufficient) indication of a favorable screening outcome.

Although outcome data are lacking in most of these reports, an observational study from Japan reported a reduction in mortality for patients with lung cancers detected by CT screening.[22] Of the lung cancers that were detected by CT scanning of 15,342 subjects, 78 percent were stage 1, with a mean diameter of 1.5 cm, and only 14 percent were either stage 3 or stage 4. The favorability of this staging distribution was greater than that of cancers detected in earlier screening studies with the use of chest radiography. Correspondingly, the overall five-year survival rate improved, from 49 percent for cases detected by chest radiography to 84 percent for those detected by CT.[22] Additional data suggesting a lower rate of death among subjects who underwent CT scanning derive from the International Early Lung Cancer Action Project, a multinational, nonrandomized study that involved an initial screening of more than 26,577 subjects and follow-up screening of 19,555 subjects at more than 30 sites.[23,24] Of 350 lung cancers that were detected, 82 percent were stage 1. With follow-up as long as 100 months (median time, approximately 40 months), the survival rate for subjects with lung cancer was more than 95 percent.

Recently, in a report of a pilot study associated with a large, randomized trial conducted by the National Cancer Institute, the rate of detection of cancer in stage 1 was 40 percent for baseline screening (16 of 40 cancers detected) and only 25 percent for annual follow-up scanning (2 of 8 cancers detected).[25] Factors that potentially contributed to the disparity between these rates and those reported in other series include the small numbers of subjects, the composition of the cohort, and variations among centers in test performance.

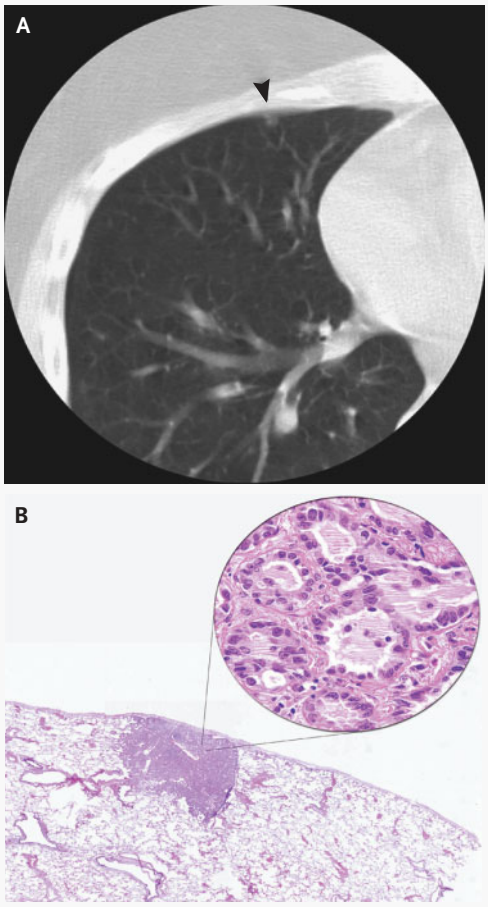

Figure 1. Non–Small-Cell Lung Cancer Detected by Screening.

In Panel A, a CT scan of the chest shows a 5-mm nonsolid nodule abutting the pleura in the right middle lobe (arrowhead). In Panel B, histopathological analysis of tissue obtained from thoracic resection reveals a subpleural adenocarcinoma of a mixed subtype. The tumor nodule shows infiltrative changes along the left border, with the invasive acinar growth pattern of this carcinoma shown on the nodule (inset) (hematoxylin and eosin). Images are courtesy of the Early Lung Cancer Action Project.

LIMITATIONS OF THE AVAILABLE STUDIES

Although these findings appear promising, definitive evidence that CT screening reduces mortality associated with lung cancer is lacking. Because available data are from observational studies, possible biases must be considered that might explain or contribute to the apparent improvement in survival among patients who underwent CT screening, as compared with historically poor survival among patients in whom lung cancer was diagnosed on the basis of symptoms.[10,26] Lead-time bias refers to the apparent increase in survival attributable to the longer interval after a diagnosis made on the basis of a screening test as compared with one made after the onset of symptoms. Length bias refers to the possibility that relatively slow-growing cancers that are

Table 1. A Comparison of Lung Cancers Detected during Screening at Baseline and at Annual Follow-up.*

Variable	Baseline Screening (N=13,122)	Annual Follow-up (N=10,245)
Total tumors detected (no.)	112	55
Stage 1 (%)	55–85	60–100
Stage 3–4 (%)	3–36	0–36
Rate of cancer (%)	0.4–2.7	0.07–1.1
Mean diameter of nodule (mm)	14–21	10–16
Noncalcified nodules (%)	12–46	2.7–14
Patients recalled for workup (%)†	3–12	2.5–11.5
Cancer confirmed by thoracotomy (%)	67–100	66–100

* Data are from Henschke et al.,[11,12] Sobue et al.,[18] Nawa et al.,[19] Swensen et al.,[20] and Pastorino et al.[21]
† Comparable data regarding the number of patients who were recalled for workup after screening were not available from Swensen et al.,[20] so information from the other five studies was used.

less likely to cause symptoms may be preferentially detected by screening. In the latter case, the apparently longer survival with screening may represent the indolent nature of the tumors that were detected rather than a benefit of screening itself. These biases may be particularly problematic when consideration is given to the end points of early screening, such as five-year rates of survival.[27]

A related concern is the possibility of overdiagnosis.[16] Overdiagnosis is said to occur if small tumors preferentially detected by screening would otherwise remain clinically covert until death from other causes. In such cases, screening results that are apparently favorable with regard to stage distribution or five-year rates of survival may not translate into significant reductions in mortality from lung cancer. Although overdiagnosis is possible, available data suggest it is unlikely.[10] Recent reports indicate that small lung cancers that are detected by screening express molecular and biochemical profiles that are indistinguishable from those of lung cancers that are detected after symptoms develop.[28,29]

Generally, as with breast cancer screening,[30] randomized trials have been considered the gold standard for demonstrating that a screening test reduces mortality. However, the rapid advances in CT imaging have greatly complicated rigorous assessment of CT screening, since several generations of important technical improvements are occurring within the time frame required for conducting a single, large, randomized trial.

RISKS OF SCREENING

Possible benefits of screening must be weighed against potential harm. Lung cancers appear on spiral CT scans as noncalcified nodules, but only a small fraction of noncalcified nodules that are detected by screening are lung cancers. Screening may subject persons who do not have lung cancer to risks that include anxiety associated with abnormal findings, procedural complications, and substantial costs. Diagnostic procedures that are indicated by CT results may entail invasive surgery,[16] and resultant morbidity and mortality might undermine potential benefits associated with diagnosing cancers at an earlier stage.

The fraction of detected noncalcified nodules that prompt an invasive diagnostic workup ranges from 3 to 12 percent (Table 1). In one series, 20 percent of the nodule specimens obtained by thoracotomy turned out to be benign.[20,31] Although the surgical management of cases on the basis of positive findings on screening has not resulted in major complications according to reports from several experienced centers, mortality associated with anatomic lobectomy and mediastinal staging may be as high as 3 percent.[32] Since patients with abnormal screening results are likely to have smoked, the potential surgical risk is a critical consideration in counseling.[21,22,33,34]

Features of nodules may help to discriminate between benign and malignant processes, but suggested criteria such as a spiculated distribution of calcium are not sufficiently informative.[23] A strategy that was proposed by the Early Lung Cancer Action Project was to determine the rate of growth of nodules.[24] For noncalcified nodules that were less than 1 cm in diameter, high-resolution CT scanning was repeated at three months and evaluated visually by the radiologist. In this analysis, the radiologist was looking for either a change in volume (of the entire nodule or of the solid component of a partially solid nodule) or the development of a solid component in a previously nonsolid nodule. With the use of these criteria, the recall rate was 12 percent after screening at baseline [24] and fell to 6 percent at follow-up scanning after one year. Among persons who were referred for needle biopsy on the basis of nodule growth, the rate of diagnosis of cancer was 90 percent.

Another proposed strategy is to use positron-

emission tomography (PET) for further evaluation of suspicious nodules. In a recent screening study involving 1035 high-risk persons (i.e., those with a history of more than 20 pack-years of smoking),[21] PET imaging was performed in 42 persons with suspicious nodules. Of the 20 PET scans that were classified as positive, 18 were confirmed to be diagnostic of lung cancer.[21] An alternative diagnostic approach is the use of transthoracic needle biopsy in all patients with positive results on CT screening, with only cases of documented lung cancer referred for surgery.[35] Further research is needed to compare the relative benefits and risks of transthoracic needle biopsy and PET scanning; costs and availability may be limiting factors.

RANDOMIZED CLINICAL TRIALS

The National Cancer Institute initiated the National Lung Cancer Screening Trial in 2002 to evaluate whether CT screening leads to a significant improvement in mortality associated with lung cancer; full accrual was completed in February 2004. The trial includes 50,000 subjects who were randomly assigned to undergo CT screening with multidetector-row scanners (mostly four rows) or chest radiography annually for three years, with planned follow-up for mortality through 2009. A European randomized trial comparing CT screening (with the use of 16-row scanners and computer-assisted detection tools) with standard care among almost 20,000 persons with a history of heavy smoking should be completed around 2010. Elements of data acquisition in these trials are being standardized to facilitate pooled analysis of the final results.[36]

AREAS OF UNCERTAINTY

BENEFITS OF SCREENING

In the absence of data from definitive trials, it is uncertain whether CT screening reduces mortality from lung cancer. If screening proves to be effective overall, more data will be needed to identify the patients who are most likely to benefit from it.[37] The optimal approach to the management of nodules that are detected by screening remains unclear; various strategies are being evaluated in parallel with screening trials.[38,39]

Rates of smoking cessation may be higher after CT screening for lung cancer than they are with counseling alone. In the year after CT screening, 14 percent of the smokers in the Mayo Clinic cohort had quit smoking, as compared with an expected rate of 5 to 7 percent in the general population.[40] In another cohort, a rate of smoking cessation of 23 percent was reported six months after CT screening.[41] These success rates rival those reported after pharmacologic interventions. However, they require confirmation in further studies.

EFFECT OF ADVANCING TECHNOLOGY

The rapid technological advances in scanning methods raise questions about how best to evaluate the value of these tools.[42,43] Ten years ago, it took a typical single-detector CT scanner several minutes to provide views (slices) that were 10 mm thick

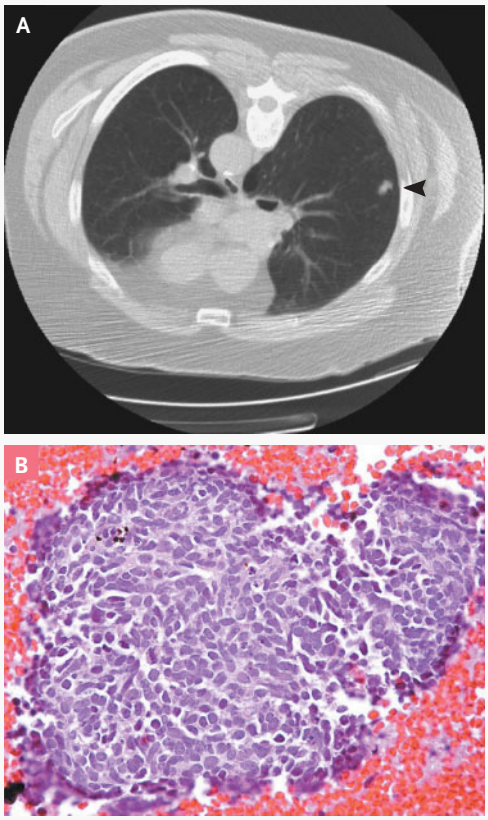

Figure 2. Small-Cell Lung Cancer Detected by Screening.
In Panel A, a prone position CT scan of the chest shows a peripheral 1.1-cm solid nodule in the posterior segment of the right upper lobe (arrowhead). In Panel B, cytopathological features of cells obtained by fine-needle aspiration biopsy are consistent with small-cell carcinoma (hematoxylin and eosin). Images are courtesy of the Early Lung Cancer Action Project.

along the entire length of the thorax, and consequently, the respiratory motion of chest structures seriously compromised the image resolution. After several generations of refinement of multidetector CT scanners, 64-row scanners can create an image of the entire thorax with a slice thickness of 0.625 mm in less than three seconds. These thinner slices may allow for a better characterization of nodules, as well as increase the ability to detect cancers arising in the central airways (such as the small-cell lung cancer shown in Fig. 2).[24,44] Even higher-resolution imaging platforms, now being used in preclinical drug development,[45] are expected to be applied to screening (see Fig. 1 of the Supplementary Appendix, available with the full text of this article at www.nejm.org). Since interpretation of the large quantity of images that are generated requires new processing software, the National Cancer Institute has developed the Lung Imaging Database Consortium to accelerate the development of such software.[46]

MANAGEMENT OF SMALL CANCERS DETECTED BY SCREENING

In the Milan cohort,[21] all patients with lung nodules that were less than 5 mm in diameter underwent annual repeated CT. Only the nodules that were clearly growing, as shown by annual scanning, were diagnostically evaluated. With the use of this conservative approach, all detected cancers were still found to be stage 1, and there were no cancers detected on the basis of symptoms in the interval between the annual screenings.[21] Data are needed to guide the optimal management of small lung cancers that are detected by CT, since the management of these tumors will probably differ from that of larger cancers detected by radiography.[23,47-50]

COSTS AND COST-EFFECTIVENESS

An important concern about widespread CT screening relates to cost. For current smokers, one study projected that the cost of additional health care associated with lung cancer would be $116,300 per quality-adjusted life-year gained; this study used a computer-simulated modeling analysis that was based on assumptions derived from early reports of screening.[15] A recently published analysis used actual cost data from a screening study that relied heavily on noninvasive techniques (such as the evaluation of nodule growth by repeated CT scanning at three months) to guide the workup[51]; this approach was associated with a cost of only $2,500 per person-year of life saved. The great disparity between these estimates underscores the need for further research in this area.

Table 2. Points about Screening for Lung Cancer to Share with Patients.

No data are available from randomized trials, which are ongoing; results are expected in four to five years.
Results from observational studies of CT screening among high-risk patients (i.e., those with a history of heavy smoking) indicate a high rate of diagnosis of lung cancer in stage 1 (a relatively curable stage).
CT screening reveals many noncalcified nodules, only a fraction of which will be found to be lung cancer.
Costly invasive procedures that are associated with serious risks may be required to evaluate some nodules.
A diagnostic workup should be done by physicians experienced in such evaluation.
The selection of a facility with physicians who are experienced and credentialed in multidisciplinary fields (including thoracic surgery, pathology, and pulmonology) is critical to an optimal outcome.
The most effective way for smokers to improve their health is to stop smoking.
There is an increased risk of subsequent lung cancers after curative resection of lung cancer, so ongoing surveillance is essential.
Screening-management trials are available for the evaluation of CT screening.

GUIDELINES FROM PROFESSIONAL SOCIETIES

In a recently updated statement, the American Cancer Society continues to recommend that CT screening not be performed in asymptomatic at-risk persons.[52] Recognizing that many persons with a history of heavy smoking are choosing on their own to be screened (despite the fact that such screening is not routinely covered by insurance), the American Cancer Society recommends that such persons first discuss screening with their physicians and that such testing be done only in experienced centers that are linked to multidisciplinary specialty groups for diagnosis and follow-up.[52]

The U.S. Preventive Services Task Force also has updated its recommendations on the basis of a review of the published literature as of January 2003 (www.ahrq.gov/clinic/uspstf/uspslung.htm). In contrast to its earlier recommendation against screening, the task force now makes no recommendation (either for or against) the use of CT in persons who have no symptoms of lung cancer. If screening is being considered, physicians are advised to discuss with the patient the pros and cons, with an em-

phasis on the lack of studies showing that screening helps people live longer and on reports that false positive test results are common and can lead to unnecessary worry, testing, and surgery.[10]

SUMMARY AND RECOMMENDATIONS

Although CT screening for the detection of lung cancer has appeared to increase markedly the percentage of cases of lung cancer that are diagnosed in stage 1 among persons with a history of heavy smoking, the results of randomized trials are not yet available to assess whether such screening reduces mortality. Furthermore, the promising results reported from experienced centers may not be readily achieved in other settings. Consequently, many persons who seek screening for lung cancer — such as the woman described in the vignette, who does not have the equivalent of a 20-pack-year history of smoking and is therefore at low risk for lung cancer — may have a greater chance of iatrogenic harm than benefit from such screening. At this point, we would not recommend CT screening to such a patient. However, the husband described in the vignette has a smoking history that would be associated with a higher risk. In response to the patient's question, it would be reasonable to discuss the possibility of detection of lung cancer in an early, more curable stage but also the risks, including the possibility of unnecessary invasive procedures. Table 2 summarizes issues to consider in such a discussion. Any patient who is a current smoker should be advised to stop smoking; the evidence in support of this recommendation is far greater than the evidence for CT screening. Patients who are interested in CT screening should be encouraged to participate in screening-management trials designed to help define the best practice.

The views expressed in this article do not necessarily represent the views of the National Cancer Institute, the National Institutes of Health, or the Department of Health and Human Services.

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REVIEW ARTICLE

DRUG THERAPY

γ-Hydroxybutyric Acid

O. Carter Snead III, M.D., and K. Michael Gibson, Ph.D.

THE SHORT-CHAIN FATTY ACID γ-HYDROXYBUTYRIC ACID (GHB) WAS SYNthesized in 1960 in an attempt to create an analogue of the ubiquitous inhibitory brain neurotransmitter γ-aminobutyric acid (GABA) that would cross the blood–brain barrier.[1] GHB turned out to have sedative properties similar to those that had been reported for γ-butyrolactone 13 years earlier.[2] In fact, γ-butyrolactone has since been shown to be biologically inactive,[3,4] since all its biologic and behavioral effects are due to its rapid conversion to GHB by an active lactonase.[5] Although GHB has found limited clinical use as an anesthetic agent[6-8] and in the treatment of narcolepsy[9] and alcoholism,[10] widespread interest has developed during the past 5 to 10 years because GHB has emerged as a major recreational drug and public health problem in the United States. GHB has diverse neuropharmacologic and neurobiologic properties and appears to have dual neuronal mechanisms of action that include activation of both the γ-aminobutyric acid type B ($GABA_B$) receptor and a separate, GHB-specific receptor (Table 1). This complex interaction between GHB and the GHB and $GABA_B$ receptors within mesocorticolimbic dopamine pathways is probably responsible for the addictive nature of GHB and for symptoms of withdrawal from it.

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NEUROPHARMACOLOGIC FEATURES

METABOLISM AND NEUROMODULATORY PROPERTIES

GHB occurs naturally in mammalian brain tissue,[31] where it is derived from the conversion of its parent neurotransmitter, GABA,[32] to succinic semialdehyde through mitochondrial GABA transaminase (Fig. 1). Succinic semialdehyde is then reduced to GHB by cytosolic succinic semialdehyde reductase.[14] GHB may be metabolized through the action of GHB dehydrogenase to succinic semialdehyde, which may be further metabolized either to GABA by GABA transaminase or to succinate through the action of mitochondrial succinic semialdehyde dehydrogenase.[33]

GHB exerts ubiquitous pharmacologic and physiological effects when it is administered systemically to animals (Table 1).[34] However, GHB also has many of the requisite properties of a neurotransmitter or neuromodulator,[35] including a discrete, subcellular anatomical distribution in neuronal presynaptic terminals, along with its synthesizing enzyme. GHB is released by neuronal depolarization in a calcium-dependent fashion.[36] A sodium-dependent GHB-uptake system in the brain has also been described,[37] and an active vesicular uptake system that is most likely driven by a vesicular inhibitory amino acid transporter has been reported.[38]

GHB RECEPTORS

The existence of a specific GHB receptor is suggested by specific, high-affinity GHB-binding sites that are observed in the brains of rats and humans. The kinetics of the GHB receptor are related to the 1-to-4-μm concentration of GHB that is typically found in mammalian brain tissue.[14,31] Although there are contradictory data,[39] evidence suggests that the GHB receptor is presynaptic and G-protein–coupled[15] and that it may

Table 1. Molecular Mechanisms and Physiological Consequences of Ingestion of GHB.*

Variable	References
Molecular mechanisms	
Altered dopamine release (mediated by $GABA_B$ receptors)	Howard and Banerjee[11]
Increased serotonin turnover	Gobaille et al.[12]
Increased level of acetylcholine	Sethy et al.[13]
Increased level of dynorphin A	Maitre[14]
Increased level of 3'-5' cyclic guanosine monophosphate in brain	Maitre[14]
Altered activity of adenyl cyclase (mediated by GHB receptors)	Snead[15]
G-protein activation (mediated by GHB receptors)	Snead[15]
Decreased glucose use in brain	Kuschinsky et al.[16]
Reduced mitogen-activated phosphorylation of protein kinase in brain (mediated by $GABA_B$ receptors)	Ren and Mody[17]
Altered presynaptic release of GABA and glutamate (mediated by GHB receptors and $GABA_B$ receptors)	Hu et al.,[18] Ferraro et al.[19]
Decreased binding to NMDA receptors	Sircar and Basak[20]
Increased plasma concentration of neurosteroids (mediated by $GABA_B$ receptors)	Barbaccia et al.[21]
Physiological consequences	
Hypothermia (mediated by $GABA_B$ receptors)	Quéva et al.[22]
Hypertension (mediated by $GABA_B$ receptors)	Hicks et al.[23]
Tachycardia (mediated by GHB receptors and $GABA_B$ receptors)	Hicks et al.[23]
Increased activity of renal sympathetic nerves (mediated by $GABA_B$ receptors)	Hicks et al.[23]
Decreased minute ventilation	Hedner et al.[24]
Decreased intestinal motility (mediated by $GABA_B$ receptors)	Carai et al.[25]
EEG and behavioral changes, including absence-like seizures and slow-wave sleep, depending on the dose (mediated by GHB receptors and $GABA_B$ receptors)	Snead,[26] Van Cauter et al.[27]
Impaired spatial learning	Sircar and Basak[20]
Increased protection against neurotoxicity	Ottani et al.,[28] Yosunkaya et al.,[29] Guney et al.[30]

* Parenthetical data regarding mediation indicate whether the effects cited are due to an effect of GHB on GHB receptors or $GABA_B$ receptors. When no mechanism is indicated, there are no data regarding mediation. GHB denotes γ-hydroxybutyric acid, $GABA_B$ receptor, γ-aminobutyric acid type B receptor, GABA γ-aminobutyric acid, NMDA *N*-methyl-D-aspartate, and EEG electroencephalography.

function to inhibit the release of GABA.[18] Despite data showing that GHB may be biologically active in its own right, compelling evidence suggests that most of the physiologic and pharmacologic effects of systemically administered GHB are mediated by the $GABA_B$ receptor (Table 1).

GABA RECEPTORS

GABA is ubiquitous in the brain and can activate ligand-gated ion channels — GABA type A ($GABA_A$) and GABA type C ($GABA_C$) receptors — as well as $GABA_B$ receptors. Activation of the $GABA_A$ receptor results in the influx of chloride ions and the generation of a fast inhibitory postsynaptic potential (Fig. 2).[41] There is little evidence to support the hypothesis that GHB interacts with the ionotropic $GABA_A$ receptor.[42]

The $GABA_B$ receptor mediates a slow inhibitory postsynaptic potential. Effector mechanisms associated with the $GABA_B$ receptor include signaling through the action of the adenylate cyclase system and activation of calcium channels and G-protein–coupled, inwardly rectifying potassium channels. The $GABA_B$ receptor is a heterodimer composed of receptor 1 and receptor 2 subunits. The $GABA_B$ receptor is transported from the interior of the cell to the cell surface by the receptor 2 subunit. Postsynaptic $GABA_B$ receptors are coupled to G-protein–coupled, inwardly rectifying potassium channels. Presynaptic $GABA_B$ receptors are subdivided into those that control the release of GABA (autoreceptors) and those that inhibit the release of all other neurotransmitters (heteroreceptors). $GABA_B$ receptors mediate their presynaptic effects through voltage-dependent inhibition of high-voltage–activated calcium channels (Fig. 2).[43]

GHB AND $GABA_B$ RECEPTORS

Because of the structural similarity of GHB to GABA and the pharmacologic $GABA_B$-like effects of GHB, the question of whether the GHB receptor and the $GABA_B$ receptor are the same has been raised, and

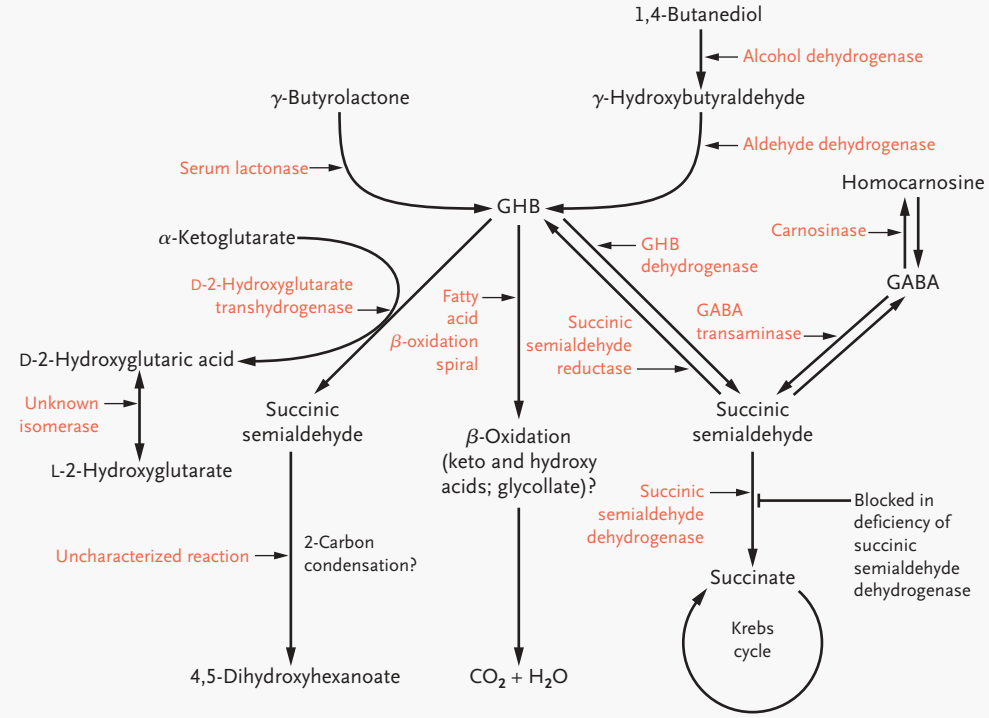

Figure 1. Putative Metabolic Interrelationship of GHB with γ-Butyrolactone and 1,4-Butanediol.

The most important synthetic pathway for γ-hydroxybutyric acid (GHB) entails conversion of γ-aminobutyric acid (GABA) to succinic semialdehyde by mitochondrial GABA transaminase, followed by reduction of succinic semialdehyde to GHB by cytosolic succinic semialdehyde reductase. Mitochondrial succinic semialdehyde dehydrogenase, converting succinic semialdehyde to succinate, couples neurotransmitter metabolism to mitochondrial energy production. This is the enzyme missing in clinical and experimental deficiency of succinic semialdehyde dehydrogenase. A minor pathway for GHB production involves partial oxidation of 1,4-butanediol. Systemically administered γ-butyrolactone is converted by a circulating lactonase to GHB. This lactonase is not present in brain tissue. The most significant catabolic pathway for GHB degradation is the oxidation of GHB to succinic semialdehyde by $NADP^+$-linked succinic semialdehyde reductase. The resultant succinic semialdehyde undergoes further metabolism to either GABA or succinate. A mitochondrial $NADP^+$-independent transhydrogenase is capable of metabolizing GHB to succinic semialdehyde with the production of D-2-hydroxyglutaric acid from L-2-hydroxyglutarate and an end product of 4,5-dihydroxyhexanoate. There is disagreement as to whether there is significant metabolism of GHB through a β-oxidation scheme.

data have been conflicting on this point. However, the results of recent studies, taken in conjunction with older data, suggest that the GHB receptor and the $GABA_B$ receptor are separate and distinct.[22,44,45]

GHB binds to the GHB receptor and the $GABA_B$ receptor with high affinity and low affinity, respectively. Available biochemical data[14,15] suggest that the intrinsic neurobiologic activity of GHB is mediated through the GHB receptor. However, many of the pharmacologic, clinical, behavioral, and toxicologic effects of exogenously administered GHB (Table 1) appear to be mediated through the $GABA_B$ receptor, where GHB may act both directly, as a partial $GABA_B$ receptor agonist,[45,46] and indirectly, on the $GABA_B$ receptor, through GHB-derived GABA.[47] The micromolar concentrations of GHB that are normally present in mammalian brain tissue[31] can activate GHB receptors but are insufficient to activate $GABA_B$ receptors, for which GHB has a weak affinity. However, the supraphysiologic (i.e., millimolar) concentrations of GHB that result from systemic administration[4] of this compound have been shown to compete for binding sites at the $GABA_B$ receptor,[35,48] activate recombinant $GABA_B$ receptor heterodimers,[45,49] and have an electrophysiological effect that is blocked by a specific $GABA_B$ receptor antagonist but not by a GHB antagonist.[46,50]

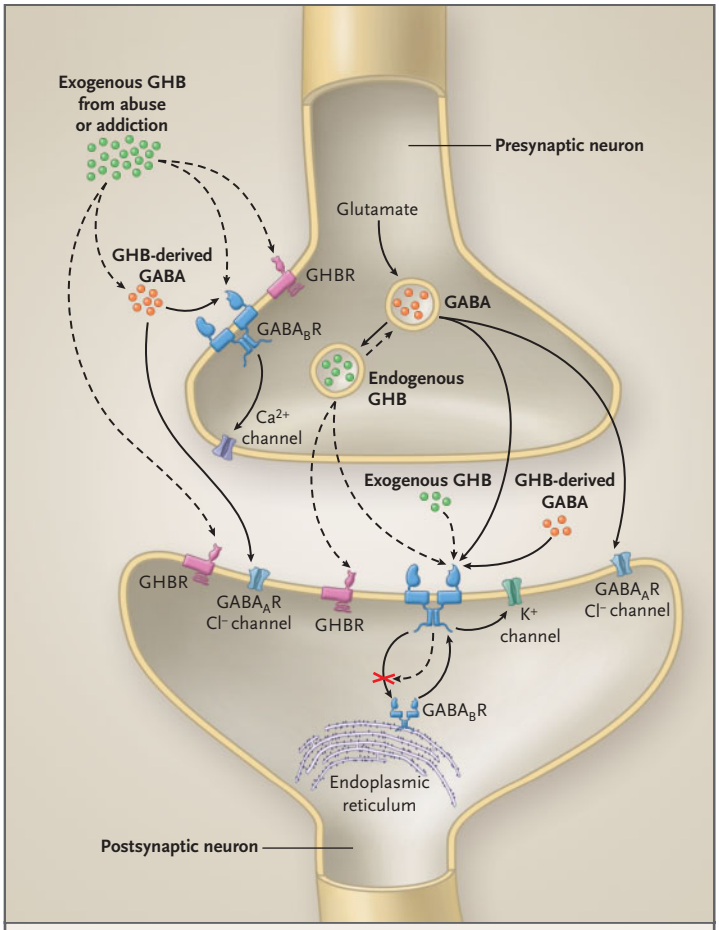

Figure 2. Synthesis and Release of GHB and GABA at Synapses.

The diagram shows the presynaptic and postsynaptic effects of endogenously released γ-hydroxybutyric acid (GHB) (as indicated by dashed arrows) and γ-aminobutyric acid (GABA) (as indicated by solid arrows) and the effects of the exogenously administered GHB, as in abuse and addiction. GABA is synthesized from glutamate in inhibitory neurons and in turn gives rise to GHB. Both GHB and GABA are released on depolarization of the GABA-releasing (GABAergic) neuron. GABA, in forms that are either endogenous or derived from exogenously administered GHB, acts on $GABA_A$ and $GABA_B$ receptors ($GABA_AR$ and $GABA_BR$, respectively). $GABA_A$ receptors are ionotropic and, when activated by GABA, cause fast postsynaptic inhibition by the efflux of chloride ions (Cl^-). $GABA_B$ receptors are metabotropic and, when activated by either GABA or high concentrations of GHB, induce slow postsynaptic inhibition by activating potassium (K^+) currents. Presynaptic $GABA_B$ autoreceptors — when activated by GHB, GABA, or both — reduce the release of GABA by suppressing the influx of calcium (Ca^{2+}). Both endogenous and exogenous forms of GHB have a dual action on the GHB receptor (GHBR) and the $GABA_B$ receptor. GHB that binds with high affinity to the presynaptic GHB receptor decreases the release of GABA; GHB that binds to a low-affinity site on the $GABA_B$ receptor increases activation of cell-surface receptors by inhibiting constitutive and agonist-induced endocytosis. The result is enhancement of GHB function mediated by $GABA_B$ receptors, with a greater effect on presynaptic inhibition than on postsynaptic inhibition. Adapted from Owens and Kreigstein.[40]

In addition to being a weak partial agonist of the $GABA_B$ receptor, GHB may also activate the $GABA_B$ receptor indirectly, through its conversion to GABA (Fig. 2). This hypothesis could explain the inordinately high concentration of GHB required to produce $GABA_B$-receptor–mediated effects, since high micromolar to low millimolar concentrations of GHB are required to produce enough GHB-derived GABA to activate $GABA_B$ receptors.[47] Furthermore, recent data suggest that GHB-derived GABA activates the $GABA_B$ receptor and induces endocytosis of the $GABA_B$ receptor, whereas GHB itself opposes this process and, acting at the $GABA_B$ receptor, causes the $GABA_B$ receptor to be retained on the cell surface, thus prolonging the functionality of the receptor.[51]

Thus, experimental evidence to date suggests that the high concentrations of GHB in brain tissue that would be predicted to accrue from exogenous administration of this compound[4] — as occurs in the clinical scenarios of GHB intoxication, addiction, and abuse — may exert their protean pharmacologic, toxicologic, and behavioral effects primarily through mechanisms mediated by the $GABA_B$ receptor (Fig. 2).

TOXICITY, ABUSE, ADDICTION, AND WITHDRAWAL

GHB TOXICITY

GHB has a half-life of 20 to 30 minutes, plasma levels peak about 40 minutes after oral ingestion, and the compound can be detected in urine for up to 12 hours.[52] GHB has a narrow margin of safety. Doses of 20 to 30 mg per kilogram of body weight lead to euphoria and memory loss, as well as to drowsiness and sleep. However, coma may result when twice this dose (or more) is administered.[53] In some series, GHB was the second most common drug detected in the serum of young people presenting with drug-induced coma, just behind cocaine.[54]

The clinical hallmark of GHB overdose is rapid onset of profound coma, myoclonus, respiratory depression, hypoventilation, and bradycardia. These signs persist for an unusually short time, given the depth of the coma.[53] The usual rapid and uneventful recovery from GHB intoxication can create a false sense of security in the GHB user.[55] The level of consciousness in patients with GHB-induced coma does not correlate with the serum level of GHB.[56] GHB intoxication should be considered in any pa-

tient, particularly any young man, who presents with rapid onset of coma of unknown cause when head trauma, metabolic disorders, central nervous system infection, and increased intracranial pressure have been ruled out.

Death from an overdose of GHB may occur as a result of respiratory compromise, aspiration, positional asphyxia, or pulmonary edema,[53,57,58] as well as traumatic injury or accident, possibly due to the abrupt loss of consciousness induced by GHB.[53,59] Well over half of all patients who present with GHB intoxication have abused other drugs as well.[60,61] Chief among these drugs is ethanol, which is synergistic with GHB in the induction of respiratory depression and hypotension[62] and thus increases the risk of an adverse outcome with an overdose of GHB.

The management of GHB intoxication in a patient who is spontaneously breathing is primarily supportive and includes stabilization of the airway, establishment of intravenous access, oxygen supplementation, and administration of atropine for persistent bradycardia.[53,63,64] Intubation is rarely indicated but should be performed in the presence of marked hypoventilation, hypoxemia, or mucosal ulcerations or in the absence of the gag response.[53] Mucosal ulcerations are of concern because illicit forms of GHB are often made from γ-butyrolactone and sodium hydroxide, an extremely basic mixture that causes mucosal burns. Aspiration of this mixture into the lungs can lead to serious pulmonary complications.[57]

There are no specific antidotes to GHB, nor is there a role for naloxone or flumazenil in the reversal of GHB-induced coma.[65] Activated charcoal is not indicated because of the very short half-life of GHB and the risk of aspiration.[53] Although physostigmine has been used to reverse the clinical signs of GHB intoxication, there is insufficient evidence to recommend its use in the treatment of GHB toxicity.[66] A patient who has recovered within six hours after the onset of symptoms can be discharged, because GHB has a relatively short half-life, and patients usually have a rapid and uneventful recovery from an overdose of GHB. Before discharge, the cause of the GHB toxicity should be determined — in other words, did the overdose occur accidentally during a one-time recreational use, or did it occur in the context of repeated GHB abuse? Discharge plans should be made accordingly, to provide the patient with assistance in dealing with the issues that led to the GHB overdose. This strategy is particularly important in the avoidance of GHB withdrawal if chronic GHB abuse led to the overdose. Any patient with a recovery time that is longer than six hours should be admitted to the hospital.

GHB ABUSE

Since the early 1990s, GHB and its prodrugs, γ-butyrolactone and 1,4-butanediol, have been used and abused by bodybuilders[67] because these compounds were reported to stimulate the production of growth hormone (Table 2).[27] Like γ-butyrolactone, 1,4-butanediol has behavioral and toxic effects caused primarily by its metabolism to GHB by an alcohol dehydrogenase.[72,73] However, the diol itself carries inherent toxicity and is particularly dangerous when used in conjunction with ethanol, which enhances its toxicity, probably because of competition of the two compounds for alcohol dehydrogenase.[74]

By the late 1990s, GHB had become a popular club drug and had gained substantial notoriety both as a major recreational drug of abuse[55,62,68] and as a "date rape" drug.[75] Subsequently, data on the addictive properties of these compounds began to emerge.[59] In 1990, the Food and Drug Administration had banned the sale of nonprescription GHB; in 2000, the agency classified it as a Schedule I substance.[76] However, illicit forms of GHB remain available under a number of names, such as G, liquid ecstasy, grievous bodily harm, Georgia home boy, liquid X, soap, easy lay, salty water, scoop, cherry meth, and nitro.[53,69] In addition, γ-butyrolactone and 1,4-butanediol are still available for purchase on the Internet, where they are advertised for mood enhancement, sleep induction, and bodybuilding.[77]

The abuse of GHB and its congeners, γ-butyrolactone and 1,4-butanediol, probably stems from the euphoria, disinhibition, and heightened sexual awareness said to be experienced after administration of the drug.[69] The psychic effects of GHB have been likened to those of ethanol in combination with reduced anxiety, feelings of euphoria, enhanced sensuality, and emotional warmth.[53] The resultant dreamy, altered sensorium accompanying the use of GHB has made it popular among attendees of so-called circuit parties[77] or "raves".[60] Raves, all-night dance parties attended by large numbers of young people, are characterized by clandestine venues, hypnotic electronic music, and the liberal use of drugs, among them GHB.[78] Circuit parties differ from raves in that they are usually attended by men who are either bisexual or homosexual.[77] When

Table 2. Clinical Aspects of GHB Overdose, Abuse, Addiction, and Withdrawal.*

Feature	Comments	References
Overdose		Couper et al.,[56] Chin et al.,[63] Mokhlesi et al.[65]
Clinical characteristics	Men with history of drug overdose, substance abuse, or psychiatric illness	
	Profound coma of rapid onset associated with myoclonus, hypoventilation, bradycardia, and miosis	
	Clinical symptoms indistinguishable from overdose of benzodiazepine or ethanol	
	Respiratory depression worse when ingested with ethanol	
	Usually rapid and uneventful recovery	
	Difficult to diagnose because of nonspecific nature of symptoms, rapid disappearance of GHB from urine and blood, and failure of routine screens to detect GHB	
Treatment	Supportive treatment with stabilization of the airway, intravenous access, oxygen supplementation, and atropine for persistent bradycardia	
	No specific antidote	
	No indication for activated charcoal, naloxone, or flumazenil	
	Little evidence for efficacy of physostigmine	
	Intubate for serious hypoventilation, absence of the gag reflex, hypoxemia, or presence of mucosal ulcerations	
Abuse		Van Sassenbroeck et al.,[60] Kam and Yoong,[68] McDonough et al.[69]
Clinical characteristics	Increased prevalence in young men	
	Club drug used at raves and circuit parties	
	Abused in conjunction with ethanol, cocaine, and "ecstasy"	
	Taken for euphoria, disinhibition, and heightened sexual awareness	
Treatment	None	
Addiction		Teter and Guthrie,[55] Freese et al.[59]
Clinical characteristics	Rarely occurs in occasional users	
	Occurs in bodybuilders and those using GHB for anxiety and insomnia	
	Frequent and increased dosing prompted by rebound insomnia	
	Use of drug every 2 to 4 hr around the clock	
Treatment	None	
Withdrawal		McDonough et al.,[69] Tarabar and Nelson,[70] Anderson and Dyer[71]
Clinical characteristics	Increased prevalence in men	
	History of bodybuilding, anxiety, or insomnia	
	Use of drug every 2 to 4 hr around the clock	
	Onset of symptoms 1 to 6 hr after last dose	
	Tremor, autonomic dysfunction, anxiety, delirium	
	Symptoms lasting up to 2 wk, possibly recurring in episodic fashion	
Treatment	Supportive care; correction of imbalance in fluids, glucose level, and electrolytes	
	Physical restraint possibly required	
	Sedation with high-dose benzodiazepines	
	No indication for antipsychotic or anticonvulsant drugs	

* GHB denotes γ-hydroxybutyric acid.

used at raves and circuit parties, GHB frequently is ingested along with other illicit drugs, most commonly ethanol, methylenedioxymethamphetamine (MDMA, or "ecstasy"), or cocaine.[60] The abuse of GHB at raves and other party settings appears to be far more prevalent among men than among women.[61,69,79]

GHB poses a serious risk for persons who are infected with the human immunodeficiency virus who are taking protease inhibitors, since these compounds alter the metabolism of GHB through their interaction with the cytochrome P-450 system. The result is that even small doses of GHB in the presence of these compounds may lead to the classic signs of GHB overdose (i.e., coma and respiratory compromise).[80,81]

GHB ADDICTION

GHB is highly addictive.[76] Occasional users of the drug may be at risk for rape, overdose, or death, given the settings in which occasional use occurs, but occasional users are unlikely to become addicted. Frequent users who take GHB as an antidepressant or for sleep, weight loss, or the enhancement of bodybuilding are far more likely to become addicted.[59] Some GHB users describe rebound insomnia or alertness occurring after two or three hours of sleep, an effect that often leads them to take additional doses to return to sleep. Thus, such users may ultimately escalate the dosage to one dose every two to four hours, around the clock.[82] GHB users typically do not see GHB as a drug because of assurances they find in publications and on the Internet that it is a "safe" and "natural" product.[83] Therefore, the GHB user may ignore warnings from friends and family who may comment about increasingly bizarre behavior; users also generally fail to recognize their incipient addiction until withdrawal ensues.[84]

Protocols for the treatment of GHB addiction and systematic detoxification have not been published, to our knowledge. However, it would make sense to consider the use of baclofen, a $GABA_B$ receptor agonist, for such therapy, since this compound appears to be effective in reducing the need for addictive drugs in animal models of GHB addiction as well as cocaine, heroin, and ethanol addiction.[85,86]

GHB WITHDRAWAL

Frequent ingestion of GHB can be associated with severe, potentially life-threatening withdrawal symptoms, necessitating vigorous clinical management, preferably in an inpatient setting.[62,69,82] Although occasional users of GHB may have a mild withdrawal syndrome when the drug is discontinued, those who have been taking GHB every one to three hours can have severe symptoms similar to those of withdrawal from ethanol or benzodiazepine.[70] In dependent persons, withdrawal symptoms may start within one to six hours after cessation of the drug.[69] Although most withdrawal symptoms occur in those who have taken the drug every one to three hours, such symptoms have also been noted in persons who have used the drug every eight hours. In contrast, cessation of GHB prescribed in the context of once-daily dosing for narcolepsy usually does not lead to withdrawal symptoms.[70]

The minimum daily dose of GHB that is associated with withdrawal is reported to be 18 g; for γ-butyrolactone, it is 10 g.[54] However, the major caveat concerning these data is the lack of quality and quantity controls with respect to the ingestion of GHB. Since most patients who present with GHB toxicity or withdrawal have purchased the drug illegally, the purity and size of the described "capful" or "teaspoon" doses are quite variable, ranging from 500 mg to 5 g per dose.[59] As in other forms of addiction and abuse, most patients who present in GHB withdrawal are male.[69] Most of them have been taking GHB for less than two years, and about 75 percent have been using GHB, rather than precursors such as γ-butyrolactone.[54]

GHB withdrawal symptoms may be mild on presentation, but they may increase in intensity and severity over hours or days and culminate in delirium or frank psychosis. The most common features of withdrawal are tremor, tachycardia, restlessness, insomnia, anxiety, nausea, and vomiting. Delirium, often with diaphoresis and hypertension, occurs in people with severe dependence.[59,69] Death from GHB withdrawal caused by pulmonary edema has been reported.[69]

Symptoms of withdrawal from GHB may last up to two weeks. In addition to the acute GHB withdrawal syndrome, a prolonged withdrawal state lasting from three to six months and characterized by dysphoria, anxiety, memory problems, and insomnia has been reported.[87] A person with protracted or untreated symptoms of GHB withdrawal may abuse either alcohol or benzodiazepines in an attempt to relieve anxiety and insomnia.

The mainstay of therapy for GHB withdrawal is supportive care and sedation to prevent injury, hyperthermia, and rhabdomyolysis. Physical restraint may be required in about one third of patients.[69] Benzodiazepines (either lorazepam or diazepam), often in very high doses, are the primary agents used to treat GHB withdrawal[53,69-71,82] because they have a broad therapeutic range, a high threshold for respiratory depression, and are relatively free of cardiovascular complications. Antipsychotic agents are not indicated in the management of GHB withdrawal and have the added disadvantage of lowering the seizure threshold.[70] However, there is no evidence that anticonvulsant drugs are effective in the treatment of GHB withdrawal.[70] In withdrawal that is refractory to benzodiazepines, pentobarbital administered in the intensive care setting is said to be effective.[69-71,88] Multiple relapses after GHB detox-

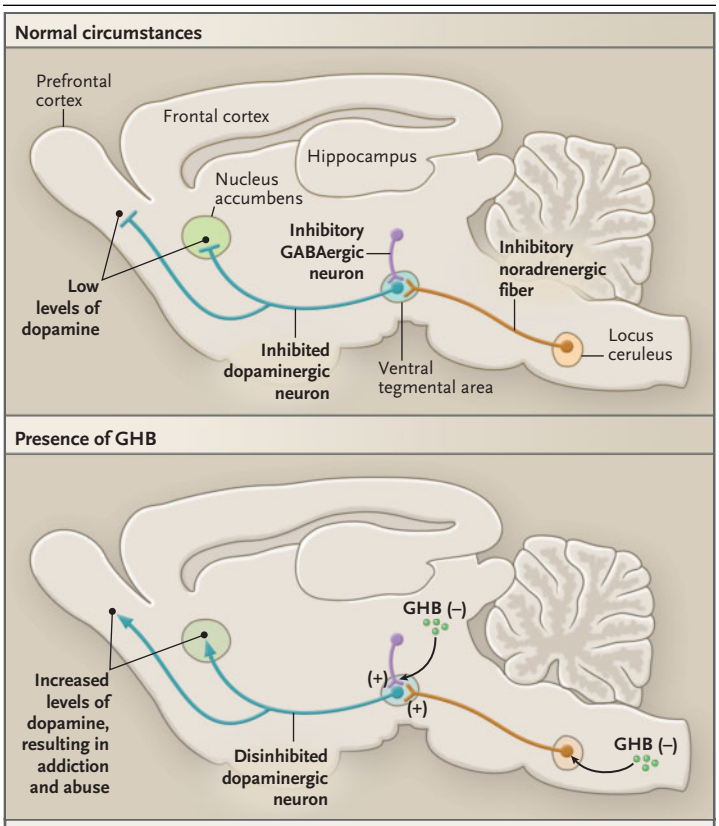

Figure 3. Mesocorticolimbic Dopaminergic Pathways Putatively Involved in the Mechanism of GHB.

These pathways have been defined in the rat brain[96]; a similar map for the human brain does not exist and can only be extrapolated. The dopaminergic neurons in this circuit have their cell bodies in the ventral tegmental area, from which they project to the nucleus accumbens, amygdala (not shown), and prefrontal cortex. Under normal circumstances, the dopaminergic neurons in this circuitry are under inhibitory control by both noradrenergic fibers from the locus ceruleus and GABAergic neurons in the ventral tegmental area. However, in the presence of GHB, which is unable to stimulate postsynaptic $GABA_B$ receptors in the ventral tegmental area,[97] there is a preferential action at $GABA_B$ autoreceptors, with decreased release of GABA and resultant disinhibition of dopaminergic neurons in the circuit. This disinhibition is further accentuated by GHB-mediated inhibition of noradrenergic neurons from the locus ceruleus.[98] The result is increased activity of dopaminergic mesocorticolimbic circuitry in the presence of increased levels of GHB in the brain; addiction then develops. Minus signs denote inhibition, and plus signs disinhibition. Adapted from Gardner and Lowinson.[96]

ification in patients who have gone through addiction and withdrawal are common, as are insomnia, depression, and abuse of other drugs.[84]

GHB-FACILITATED SEXUAL ASSAULT

GHB has received substantial notoriety during the past several years as a date-rape drug — in other words, a compound used to facilitate sexual assault. Low doses of GHB (10 to 20 mg per kilogram) induce short-term antegrade amnesia, increased libido, euphoria, suggestibility, and passivity, all of which contribute to the use of GHB in sexual assaults.[75,89-91] Populations that are at high risk for drug-facilitated sexual assault include single women or men in unfamiliar social settings. The sodium salt of GHB is generally available as a liquid that is colorless, odorless, and water-soluble and tastes slightly salty; this liquid can be easily and surreptitiously added to a drink without detection by the intended victim. Drug administration may occur in a bar or club, when the recipient is inattentive or accepts a drink or an already opened bottle.[92]

Most of the published evidence of GHB in this role is anecdotal; ethanol and benzodiazepines appear to be more commonly used in drug-facilitated assault.[93,94] However, GHB should be considered in cases of sexual assault that occur after drinking and a social encounter, particularly when the patient has a gap in memory. In making a diagnosis of GHB-facilitated sexual assault, it is important to collect samples of blood and urine as soon as possible after the alleged assault and to measure GHB levels with the proper analytic techniques. Since GHB is undetectable by the usual toxicologic screens, laboratory diagnosis of GHB-facilitated assault is challenging. GHB levels may be determined in plasma and urine samples by gas chromatography–mass spectrometry with selected-ion monitoring.[95] Although this analytic technique for the detection of GHB is not readily available, it may be performed by state and national reference laboratories.[56]

PUTATIVE MECHANISMS OF ACTION

The dopamine neurons in the brain are involved in reward-dependent learning; therefore, neurons involved in abuse, addiction, and withdrawal have their cell bodies in the ventral tegmental area and project into the basal forebrain structures, such as the nucleus accumbens, amygdala, and frontal and limbic cortexes (Fig. 3).[96,99,100] Activation of these mesocorticolimbic dopaminergic neurons, with a resultant increase in the output of dopamine in innervated projection structures, has been reported with virtually all major drugs of abuse. Conversely, during abstinence there is a marked decrease in the activity of dopaminergic neurons in the ventral teg-

mental area.[99] Therefore, the mesocorticolimbic circuitry of the brain is a likely target for GHB in abuse, addiction, and withdrawal.

Although a variety of neurotransmitters interact with mesocorticolimbic dopamine pathways,[99] the mechanism of action that would explain the addictive properties of GHB appears to be related to the effects of dopamine mediated by $GABA_B$ receptors[101] in mesocorticolimbic circuitry. However, the reported finding that GHB decreases dopaminergic neuronal activity in the ventral tegmental area and thereby reduces the release of dopamine into the nucleus accumbens[102] poses a conundrum, since drugs of abuse are classically associated with an increase in the neuronal activity of mesocorticolimbic dopamine.

A potential explanation for this paradox may lie in recently published experiments[97] showing that GHB is unable to activate potassium channels mediated by $GABA_B$ receptors in dopamine neurons in the ventral tegmental area. However, GHB was able to activate $GABA_B$ receptor–mediated potassium channels in GABA-releasing (GABAergic) neurons of the ventral tegmental area because of a difference in the expression of potassium-channel subunits between the dopaminergic and GABAergic neurons. GHB also is known to decrease the release of GABA by a presynaptic, GHB-specific action.[15,18] Hence, in GHB abuse and addiction, which are accompanied by an increased concentration of GHB in the brain, GHB may inhibit GABAergic neurons preferentially and decrease the release of GABA through effects mediated by $GABA_B$ receptors and GHB receptors, respectively. The result would be a disinhibition of dopaminergic neurons of the ventral tegmental area with increased dopaminergic activity within that circuitry (Fig. 3), which in turn would lead to the psychic symptoms that accompany GHB abuse and addiction. This hypothesis would also explain the difference between GHB, which is addictive, and the $GABA_B$ receptor agonist baclofen, which is not. In fact, baclofen may be useful in decreasing the reinforcement effects of cocaine, heroin, nicotine, ethanol,[85] and GHB,[86] probably by reducing the release of dopamine in the ventral tegmental area. Finally, GHB has been recently shown to decrease the activity of neurons in the locus ceruleus,[98] providing yet another route by which GHB could disinhibit mesocorticolimbic dopaminergic circuitry (Fig. 3).

In summary, data indicate that the mechanism of GHB abuse, addiction, and withdrawal may be due to inhibition of GABAergic neurons by mechanisms mediated by $GABA_B$ receptors and inhibition of presynaptic GABA release in mesocorticolimbic dopaminergic pathways by a mechanism mediated by GHB receptors, with a resultant disinhibition of dopamine neurons and increase in dopaminergic activity in the mesocorticolimbic circuitry (Fig. 3).

FUTURE DIRECTIONS

The pharmacologic properties of GHB and its $GABA_B$ receptor–mediated effects are well known. However, the neurobiologic function of GHB remains elusive. This function will probably be delineated after the successful molecular cloning of the primary GHB receptor in brain tissue and the subsequent engineering of mice with mutant GHB receptors. These developments will lead to a more precise elucidation of the relationship between the GHB receptor and $GABA_B$ receptor and will facilitate careful investigation of the relative contributions of GHB-induced GABA synthesis, GHB-induced alterations in GABA release, and the signaling pathways involved in GHB-induced alteration of intracellular movement of $GABA_B$ receptors. In a similar fashion, a mutant mouse that is deficient in succinic semialdehyde dehydrogenase[103] may provide insight into the mechanisms of GHB addiction and withdrawal because it has inordinately high levels of GHB and GABA in brain tissue. Given the experimental evidence of the efficacy of the $GABA_B$ receptor baclofen in GHB abuse,[85,86] controlled, prospective clinical trials of this compound in the treatment of GHB addiction and withdrawal and of a $GABA_B$ receptor antagonist[104] in the treatment of GHB overdose will be important.

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CLINICAL TRIAL REGISTRATION

The *Journal* encourages investigators to register their clinical trials in a public trials registry. The members of the International Committee of Medical Journal Editors plan to consider clinical trials for publication only if they have been registered (see N Engl J Med 2004;351:1250-1). The National Library of Medicine's www.clinicaltrials.gov is a free registry, open to all investigators, that meets the committee's requirements.

CORRECTION

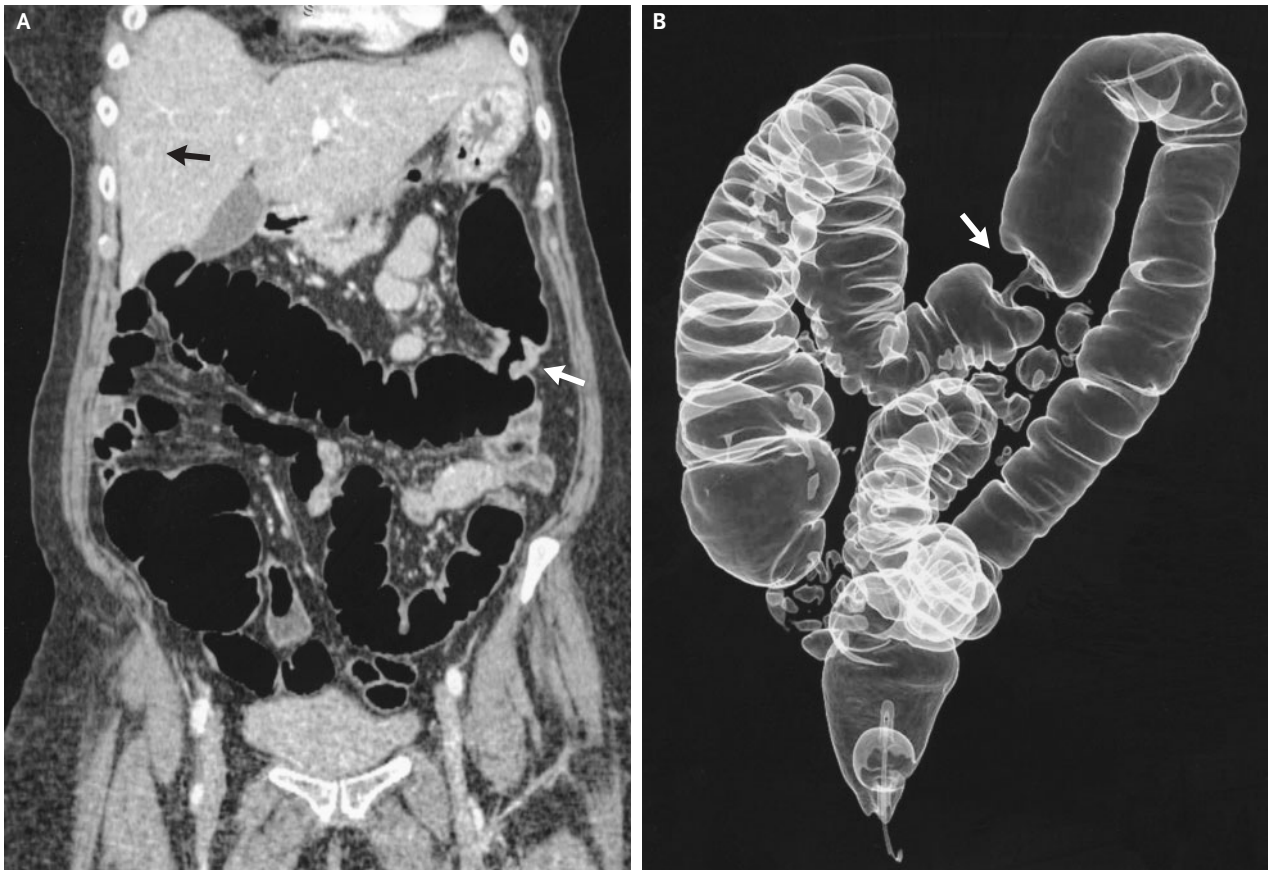
γ-Hydroxybutyric Acid

γ-Hydroxybutyric Acid . On page 2724, lines 8 through 11 under the subhead GHB Toxicity should have read, ``In some series, GHB was the second most common drug detected in the urine of young people presenting with drug-induced coma, just behind cocaine,´´ rather than ``detected in the serum,´´ as printed.

N Engl J Med 2006;354:537-a

IMAGES IN CLINICAL MEDICINE

The Virtual Apple Core of a Colonic Carcinoma



A 67-YEAR-OLD WOMAN PRESENTED WITH A THREE-MONTH HISTORY OF abdominal pain, weight loss, and rectal bleeding. She had never been screened for colon cancer. Laboratory evaluation revealed a hematocrit of 32 percent and normal liver function. The patient was referred for computed tomography (CT) of the abdomen with integrated CT colonography. This technique combines contrast-enhanced CT scanning of the abdomen and pelvis with rectal air insufflation to distend the colon. A coronal multiplanar reformatted image (Panel A) shows a constricting lesion in the distal transverse colon (white arrow), which has overhanging edges that suggest a malignant lesion. A low-attenuation, peripherally enhancing lesion in the liver (black arrow) is consistent with a metastasis. Three-dimensional reconstruction of the colonic air cast — a virtual air-contrast enema (Panel B) — shows an apple-core–like constriction (arrow). Endoluminal three-dimensional CT colonoscopy shows the overhanging distal edge of a lesion compatible with carcinoma of the colon (Panel A of the Supplementary Appendix, available with the full text of this article at www.nejm.org). The appearance of this virtual image is highly similar to the endoluminal photograph (Panel B of the Supplementary Appendix). A biopsy specimen revealed adenocarcinoma. The patient underwent resection of the transverse colon and the single hepatic metastasis. The mesenteric lymph nodes were without tumor. The patient underwent adjuvant chemotherapy and was free of disease at one year.

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CASE RECORDS of the MASSACHUSETTS GENERAL HOSPITAL

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Case 20-2005: A 58-Year-Old Man with Locally Advanced Pancreatic Cancer

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N Engl J Med 2005;352:2734-41.

PRESENTATION OF CASE

A 58-year-old man was seen in the gastrointestinal oncology clinic of the cancer center of this hospital for management of pancreatic cancer.

He had been in his usual state of health until four months earlier, when abdominal computed tomography (CT), performed to evaluate a kidney stone, revealed an incidental finding of a small abdominal aortic aneurysm, for which further evaluation was advised. One month later, abdominal ultrasonography showed an ectatic aorta with a maximal anteroposterior diameter of 2.9 cm; no definite aneurysm was identified. One month later, abdominal and pelvic CT performed with oral and intravenous contrast material disclosed an aneurysm 3.8 cm in diameter, with a mural thrombus. There was a rounded, low-attenuation mass, 1.9 cm by 2.4 cm, in the head and neck of the pancreas, with dilatation of the pancreatic duct and atrophy of the pancreatic tail.

Two weeks later, the patient was evaluated by a gastroenterologist. The symptoms that he described at this evaluation included vague discomfort in the left upper quadrant and left epigastrium, which he said he had had for the preceding year and which usually began one hour after meals and subsided after one to two hours. A trial of histamine H_2–receptor antagonists had not provided relief. The discomfort was associated with a feeling of fullness in his abdomen, which increased with physical exertion but did not awaken him at night. He had lost 2 to 3 kg of weight, which he attributed to a reluctance to eat large meals. The results of a physical examination that included a rectal examination were normal; the stool guaiac test was negative.

Endoscopic retrograde cholangiopancreatography was performed two weeks after the examination by the gastroenterologist; abrupt termination of the main pancreatic duct within the neck of the pancreas was revealed. A cholangiogram showed no abnormalities. Endoscopic ultrasonography revealed an irregular hypoechoic mass in the pancreatic head that was 23 mm in maximal cross-sectional diameter and encased the confluence of the superior mesenteric vein with the portal vein. Fine-needle aspiration of the pancreas was performed. Cytologic examination of a specimen from the neck of the pancreas was positive for adenocarcinoma; specimens from the head and tail of the pancreas did not show malignant cells.

One month later, the patient was evaluated in the gastrointestinal oncology clinic at this hospital. He had borderline hypertension and hypercholesterolemia, coronary artery disease with an inferior-wall myocardial infarction that had occurred nine years earlier, and nephrolithiasis. His medications included a daily regimen of atorvastatin and aspirin and frequent ibuprofen; he had no allergies. He had smoked 20 cigarettes a day for 40 years and had recently decreased his smoking to 5 or 6 cigarettes a day. He consumed one to two cups of coffee and one to two alcoholic beverages a day. He lived with his wife and two children and had a stressful job in sales. His mother had died of a cerebral aneurysm; his father was living, with coronary artery disease.

On physical examination, he appeared well. The weight was 85.9 kg, the height 177.5 cm, the blood pressure 142/78 mm Hg, the pulse 60 beats per minute, and the temperature 35.8°C. The physical examination revealed no abnormalities. The results of urinalysis were normal; the level of CA 19-9 in the blood was 510 U per milliliter; the results of other laboratory tests, including the complete blood count, tests of liver function, and levels of electrolytes, total protein, and albumin, were all within normal ranges.

The day after the patient's evaluation at the gastrointestinal oncology clinic, staging laparoscopy was performed. Biopsy specimens of a peritoneal nodule and a whitish nodule, 2 mm in diameter, in the left lateral segment of the liver were obtained, as were peritoneal washings. Cytologic examination of the peritoneal washings revealed no malignant cells; pathological examination of the liver showed benign hepatic parenchyma; the peritoneal nodule was a benign peritoneal inclusion cyst.

DIFFERENTIAL DIAGNOSIS

Dr. David P. Ryan: May we review the imaging studies?

Dr. Dushyant Sahani: CT scanning of the abdomen, performed after the administration of intravenous and oral contrast material, showed a mass, 1.9 cm by 2.4 cm, in the pancreatic head and neck (Fig. 1A), with dilatation of the pancreatic duct and atrophy of the tail of the pancreas (Fig. 1B). The tumor indented the confluence of the superior mesenteric vein and portal vein and encased more than 50 percent of its circumference (Fig. 1C). No signs of metastatic disease or lymphadenopathy were seen in the abdomen or pelvis.

Dual-phase helical CT, which is the preferred method of evaluating pancreatic carcinomas, is performed after the intravenous administration of a bolus of contrast material and generates images during the phase of arterial perfusion of the pancreas and, after about 20 seconds, during the phase of portal venous enhancement. The rapid acquisition of data with the helical technique also allows for three-dimensional reconstruction. The relationship of the tumor to the surrounding structures (in particular, the vasculature) can be examined in detail, and the presence of liver metastases and enlarged lymph nodes can be evaluated. This method has a very high positive predictive value for unresectability (90 to 100 percent), but it is less accurate for predicting resectability (70 to 80 percent).[1,2] With the introduction of newer multislice CT scanners, the performance of CT has been enhanced still further. A recently published series reported that for detection of vascular invasion, multidetector-row CT yielded a negative predictive value of 100 percent, with no false negative findings, and an accuracy of 99 percent.[3]

In the patient under discussion, this technique was not used, as the pancreatic cancer was unsuspected. Repeated CT scanning was performed later, specifically for staging, but unfortunately, the dedicated protocol was not used, and it was decided not to perform the study a third time. The technique that was used might have underestimated the extent of vascular involvement, but since vascular involvement, including deformation of the vessel by the tumor, was seen even when this less sensitive technique was used, the likelihood that the tumor could have been resected was low.

Dr. William R. Brugge: Endoscopic retrograde cholangiopancreatography was used to examine both the common bile duct and the pancreatic duct. The common bile duct appeared normal. In the pancreatic duct, retrograde injection of contrast material through the papilla revealed narrowing and then complete obstruction in the neck of the pancreas. There was no contrast material in the body or tail of the pancreas. These findings were highly suggestive of a tumor, either primary or secondary, in the neck of the pancreas.

The patient then underwent an endoscopic ultrasonographic examination. The image shows a hypoechoic focus, 2.3 cm in diameter, in the head of the pancreas (Fig. 2A) that is directly adjacent to the confluence of the superior mesenteric vein with the portal vein over an area 1.5 cm long. Even though tumor

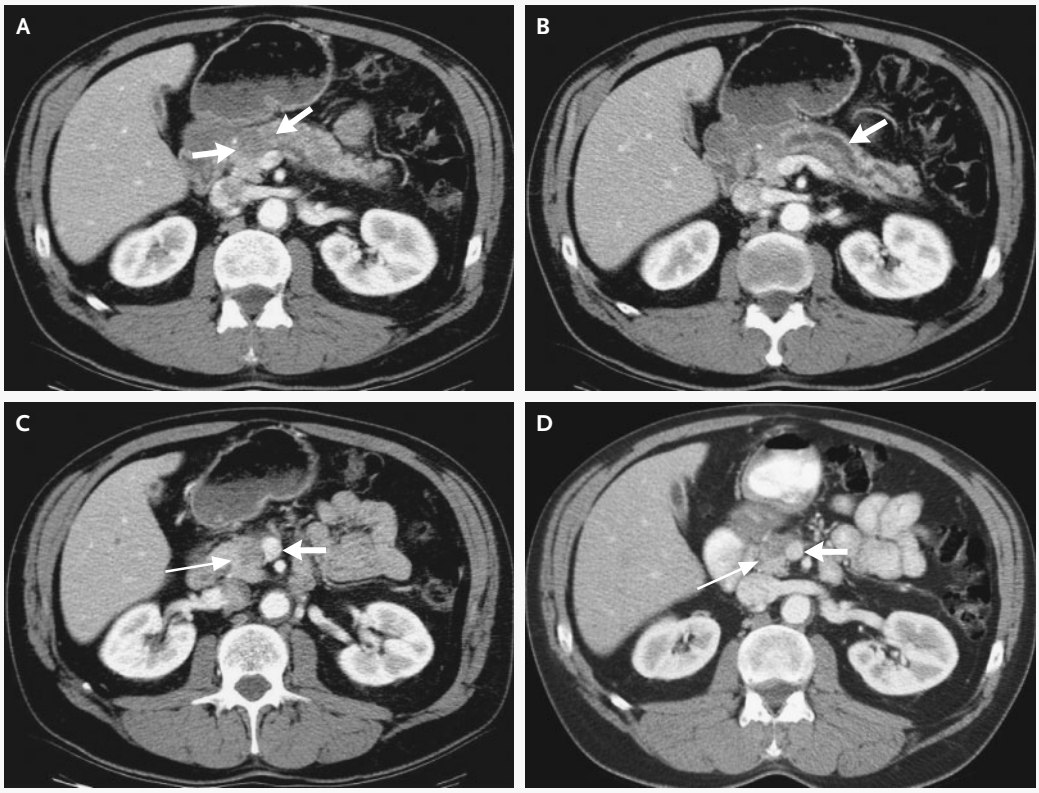


Figure 1. Axial Contrast-Enhanced CT Images through the Pancreas.

A low-density mass is present (arrows, Panel A), which involves the neck and head of the pancreas. Dilatation of the pancreatic duct (arrow) is seen in Panel B, with an abrupt transition in the neck of the pancreas where the mass is located. The mass (thin arrow, Panel C) indents the confluence of the superior mesenteric vein and portal vein (thick arrow) and encases more than 50 percent of its circumference. An axial image obtained after treatment from contrast-enhanced CT (Panel D) shows that the mass no longer indents the vein, and there is less than 50 percent encasement of the circumference of the vein. The thin arrow indicates the mass, and the thick arrow indicates the superior mesenteric vein.

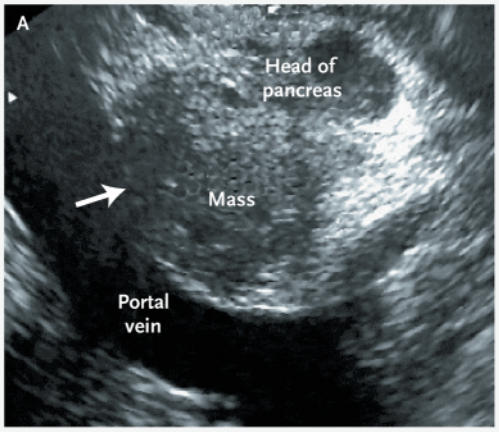
is not seen within the lumen of the vein, the degree of contact is most consistent with local invasion of the vein wall. The anatomical features under discussion are illustrated in a diagram (Fig. 2B).

Fine-needle aspiration guided by endoscopic ultrasonography was performed by placing a 22-gauge needle through the duodenal wall; material was aspirated from the hypoechoic area in the neck of the pancreas.

Dr. Elena F. Brachtel: Cytologic examination of the aspirated material showed cohesive, three-dimensional clusters of neoplastic glandular cells (Fig. 3) with necrosis, mucin, and blood in the background. These cells had irregular, enlarged, and hyperchromatic nuclei, an increased nuclear-to-cytoplasmic ratio, and intracytoplasmic mucin (Fig. 3, inset) — all findings that are characteristic of adenocarcinoma.[4]

ISSUES IN THE MANAGEMENT OF PANCREATIC CANCER

Dr. Ryan: In summary, this 58-year-old man had an incidentally discovered pancreatic adenocarcinoma, 2.3 cm in diameter, with apparent venous invasion at the confluence of the superior mesenteric vein and portal vein. In retrospect, he had had symptoms possibly related to the cancer for about one year. His presentation illustrates a common problem in our multidisciplinary gastrointestinal cancer center: the patient with what is considered a "borderline-resectable" pancreatic cancer. To clarify the multidisciplinary approach in terms of surgical evaluation as well



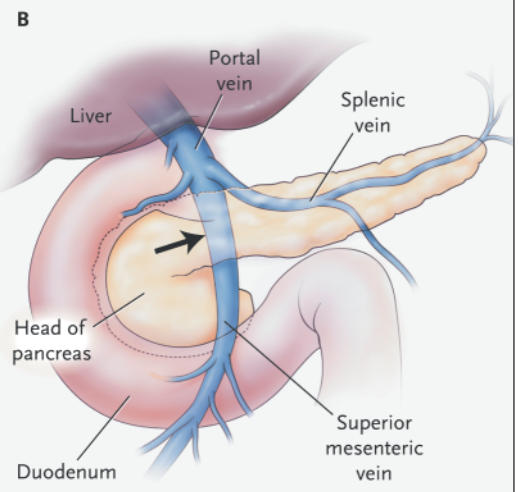


Figure 2. Endoscopic Ultrasonographic Examination.
Panel A shows a hypoechoic mass, 2.3 cm in diameter, in the head of the pancreas, which is in contact with the wall of the superior mesenteric vein for a distance of 1.5 cm. The diagram (Panel B) shows the course of the superior mesenteric and portal veins, which are often invaded by locally advanced cancers of the pancreas. In each panel, the arrow indicates the area in which the tumor in this patient was in contact with the anterior wall of the confluence of the portal and superior mesenteric veins. The patient's anterior aspect is to the right and posterior aspect to the left.

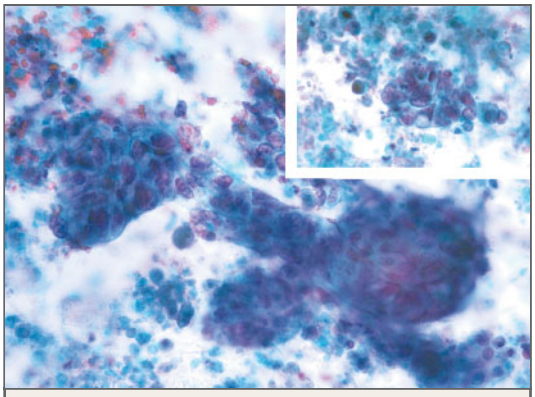


Figure 3. Cytologic Examination of the Specimen Obtained by Fine-Needle Aspiration of the Pancreas (Papanicolaou Stain).
There are large, tightly packed groups of neoplastic glandular cells, with mucin in the background. The groups are three-dimensional, with papillary configurations and nuclear features associated with tumor (crowding of enlarged, irregular, and hyperchromatic nuclei and increased nuclear-to-cytoplasmic ratio). There is abundant single-cell necrosis, also a feature of cancer. The inset image shows tumor cells with intracytoplasmic mucin.

as evaluation for combined therapy, a brief overview of pancreatic cancer is necessary.

Risk factors for pancreatic cancer include increasing age, cigarette smoking, obesity, diabetes mellitus, chronic pancreatitis, and family history; the *BRCA2* gene is emerging as an important locus in familial pancreatic cancer. The patient under discussion was slightly younger than the median age of patients with pancreatic cancer and was a long-time cigarette smoker, but he had no other risk factors. Although 50 percent of patients present with jaundice,[5] his presentation was fairly typical for patients who present without jaundice — namely, nonspecific abdominal symptoms and weight loss.[6]

Most clinicians specializing in the care of patients with pancreatic cancer divide the cases into three categories: resectable, locally advanced, and metastatic. Surgery cures approximately 20 percent of patients who undergo a complete resection, whereas long-term survival for patients with either locally advanced or metastatic pancreatic cancer is rare. Chemoradiation therapy may prolong survival in patients with locally advanced disease, but the median survival is approximately one year. Patients with metastatic disease who are treated with chemotherapy have a median survival of six months. This patient had a tumor that was on the borderline between being resectable and being locally advanced because of the involvement at portal-vein confluence.

The most important decisions regarding the care of patients such as this one with pancreatic cancer are surgical. Can the tumor be resected, by whom, and at which hospital? The surgical mortality rate associated with pancreatic-cancer resection has fall-

en from more than 20 percent in some series in the 1950s and 1960s to less than 3 percent in high-volume single institutions in the 1990s.[7-10] In a recent study using the Medicare database,[11] the mortality rate after pancreatectomy ranged from 14.7 percent with surgeons who performed fewer than two operations per year to 4.6 percent with those who performed more than four per year. Thus, the experience of the surgeon is critical.

SURGICAL MANAGEMENT OF PANCREATIC CANCER

Dr. Carlos Fernandez-del Castillo: At the time of diagnosis, 40 to 45 percent of patients with pancreatic cancer have distant metastases, another 40 to 45 percent have localized tumors that are unresectable (mostly because of vascular invasion), and only 15 to 20 percent have localized and resectable tumors. The treatment and prognosis are very different for these three groups, and therefore, accurate clinical staging is paramount. Currently, the three most frequently used tools for staging are CT, endoscopic ultrasonography, and laparoscopy — all of which were used in the case of this patient.

As Dr. Sahani mentioned, CT scanning has a 20 to 30 percent false negative rate for predicting whether or not the tumor is resectable, because of the presence of small peritoneal and liver implants, and the false negative rate as such constitutes the rationale for the use of laparoscopy in pancreatic cancer. Laparoscopy is particularly useful in patients with tumors of the body and tail of the pancreas, where the frequency of metastatic disease not identified by CT approaches 50 percent (as compared with 10 to 20 percent for tumors located in the head of the pancreas). This diagnostic method is also used in patients with unresectable tumors, since the detection of distant metastases will preclude any survival benefit from radiation therapy. In the patient under discussion, at laparoscopy we found a small peritoneal nodule and a whitish nodule, 2 mm in diameter, in the left lateral segment of the liver, both of which proved to be benign. Peritoneal washings obtained at the time of laparoscopy show malignant cells in a small number of patients (6 percent) without visible metastases and are associated with a poor prognosis; in this patient, they were negative.[12]

In the past several years, endoscopic ultrasonography has emerged as a powerful and versatile imaging method that can identify and guide biopsy of small pancreatic tumors. Although tissue diagnosis is not required by most pancreatic surgeons in order to proceed with resection, it is necessary when neoadjuvant treatment is planned or in cases of unresectable or metastatic tumors. In experienced hands, endoscopic ultrasonography can be as good as CT for staging.[13] Its limitations include the inability to evaluate the liver and the more distal superior mesenteric vein, as well as the fact that its reliability is heavily dependent on the skill of the operator. In this case, endoscopic ultrasonography confirmed venous invasion and provided diagnostic tissue from the tumor.

The contraindications to resection of pancreatic cancer are listed in Table 1. Because some of the elements involved are subjective, it is important that the decision to proceed or not to proceed with resection be made by an experienced pancreatic surgeon. When the portal vein is involved, the proportion of the circumference that is affected is important in determining resectability. If it is less than 50 percent, the tumor can probably be resected with repair of the vein, whereas if it is more than 50 percent, the possibility of resection may be less likely. In the case being discussed, CT and endoscopic ultrasonography suggested involvement of more than 50 percent of the circumference of the vein. Because of this, we thought that the probability that this tumor could be completely resected was low, and we decided together with the medical and radiation oncologist to proceed with neoadjuvant treatment in the hope of reducing the size of the tumor and the degree of vascular involvement, so that resection with clear margins would be possible.

PREOPERATIVE AND POSTOPERATIVE CHEMOTHERAPY AND RADIATION

Dr. Christopher G. Willett: After surgery for resectable pancreatic cancer, local recurrence has been reported in 50 to 85 percent of patients even in subgroups

Table 1. Contraindications to Surgical Resection of Pancreatic Cancer.*

Metastases to the liver, peritoneum, omentum, or any extra-abdominal site
Encasement of celiac axis, hepatic artery, or superior mesenteric artery
Involvement of splenoportal confluence
Involvement of bowel mesentery
Involvement of superior mesenteric vein or portal vein

* The list, top to bottom, represents the range from absolute to relative.

with the most favorable conditions.[14-16] The likely explanation is the presence of residual disease at the retroperitoneal and soft-tissue margins at the time of surgery. Multiple series have now shown that a microscopically positive margin is an important prognostic feature at the time of resection, reducing the median survival to that among patients with locally advanced disease.[17,18] The retroperitoneal margin is the one that is most frequently positive. In this patient, the presence of venous involvement placed him at very high risk for having a positive retroperitoneal margin if resection was the first therapeutic method attempted.

Efforts to improve local control and thus survival include the administration of chemotherapy and radiation therapy, either postoperatively or preoperatively. Of three important randomized trials of chemoradiation in the postoperative setting, one showed a survival benefit with chemoradiation therapy, and the other two did not.[19-21] All the studies have serious limitations, and it is not possible to draw any definitive conclusions because of flaws in the trials. Single-institution trials that have evaluated chemoradiation therapy both for locally advanced tumors and as an adjuvant to resection have shown good local control with satisfactory tolerance when modern irradiation techniques have been used.[15,22]

A more recent approach to improve local control has been the application of preoperative chemoradiation — so-called neoadjuvant therapy — for patients with resectable disease; studies have demonstrated local control in 87 to 100 percent of patients.[23,24] There are two arguments for the use of preoperative chemoradiation therapy. First, it ensures the delivery of chemoradiation therapy to patients undergoing resection, since approximately 20 to 25 percent of patients will not receive postoperative therapy because of a prolonged recovery period after surgery. Second, in approximately 20 to 25 percent of patients, metastatic disease will become apparent while they are receiving preoperative therapy; thus they can avoid an operation that would not be curative. It should be noted that preoperative therapy is not associated with improved median survival rates when compared with historical controls who received postoperative therapy, and only occasionally does downstaging occur.

Dr. Ryan: This patient had a tumor that was on the borderline between being resectable and being considered locally advanced, because of the involvement of about 50 percent of the circumference of

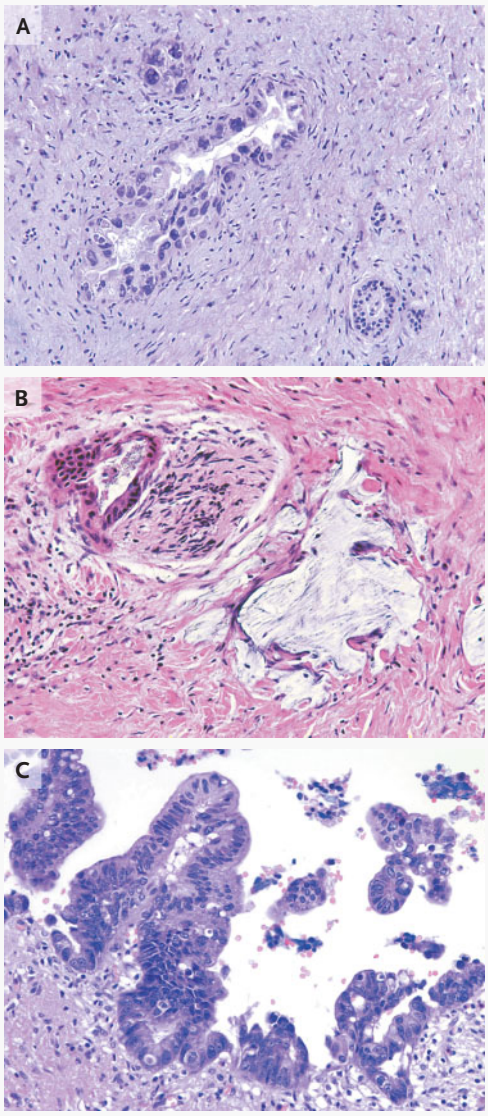


Figure 4. Sections of the Resected Pancreas (Hematoxylin and Eosin).

Rare, highly abnormal glands are embedded in dense fibrous stroma, which represent residual carcinoma (Panel A). The nuclei are large, hyperchromatic, and irregular; some of the atypical features may be due to therapy. A preserved islet is present in the lower right corner of the image. There are collections of light blue acellular mucin (Panel B), intimately associated with nerves, indicating that tumor had been present in these areas. The distal resection margin also showed pancreatic intraepithelial neoplasia, grade 3 (Panel C). The duct is lined by papillary excrescences into the lumen; nuclear crowding and atypical features are visible, with no invasive tumor identified.

the confluence of the portal and superior mesenteric veins. After an interdisciplinary consultation, we thought that preoperative therapy would offer improved local control and might increase the likelihood that the tumor in this patient would be resectable. We offered him enrollment in a phase 1 study of concurrent gemcitabine, fluorouracil, and external-beam radiation with a CT-based multifield technique (50.4 Gy in 28 fractions), and he accepted. He had few side effects, his abdominal pain diminished almost to the point of disappearance, and he regained his lost weight.

Dr. Sahani: A post-treatment CT scan shows a mass, 1.7 cm in diameter, in the head of the pancreas, involving slightly less than 50 percent of the circumference of the superior mesenteric vein, without indenting it (Fig. 1D).

Dr. Fernandez-del Castillo: An exploratory laparotomy was performed after the completion of chemoradiation. The tumor was easily dissected from the portal and superior mesenteric veins, and no evidence of metastatic disease was found. A Whipple procedure was performed.

Dr. Brachtel: An ill-defined, firm fibrotic area measuring 2 cm by 2 cm by 1 cm was noted macroscopically in the neck of the pancreas, which was close to the distal resection margin and came to within 1.5 cm of the uncinate and within 1 cm of the retroperitoneal margin. Residual microscopic foci of ductal adenocarcinoma embedded in dense fibrous stroma were present on histologic examination (Fig. 4A). Acellular mucin (Fig. 4B) and pancreatic intraepithelial neoplasia grade 3 were present at the distal resection margin (Fig. 4C), but all resection margins were free of viable tumor, as were four peripancreatic lymph nodes.[25,26]

Dr. Ryan: The final pathological stage of this patient's tumor was T1N0 pancreatic cancer. During his recovery after the operation, the patient had persistent abdominal discomfort, which made the administration of additional chemotherapy impossible. Thus, he did fall into the category of 20 to 25 percent of patients who are not able to receive adjuvant chemotherapy postoperatively. Four years after the Whipple procedure, he remains free of recurrent or metastatic cancer and works full-time.

ANATOMICAL DIAGNOSIS

Ductal adenocarcinoma of the pancreas, with residual microscopic tumor after chemoradiation therapy.

Pancreatic intraepithelial neoplasia, grade 3, present at the distal resection margin. Resection margins and lymph nodes free of invasive carcinoma.

Dr. Ryan reports having received consulting fees and lecture fees from Lilly Oncology.

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EDITORIALS

The Maze of Treatments for Hepatitis B

Anna Suk-Fong Lok, M.D.

Worldwide, there are approximately 350 million carriers of hepatitis B virus (HBV), of whom half a million to 1 million die from liver disease each year. The goal of treatment for chronic hepatitis B is to prevent cirrhosis, hepatic failure, and hepatocellular carcinoma. This goal is best achieved by eradicating HBV before irreversible liver damage occurs. However, the eradication of HBV is impossible to achieve because of the presence of extrahepatic reservoirs of HBV, the integration of HBV DNA into the host genome, and the presence of an intracellular conversion pathway that replenishes the pool of transcriptional templates (covalently closed circular HBV DNA) in the hepatocyte nucleus without the need for reinfection. Thus, withdrawal of treatment is usually accompanied by rapid viral rebound.

Currently, there are five approved therapies for chronic hepatitis B in the United States — interferon alfa-2b, lamivudine, adefovir, entecavir, and pegylated interferon (peginterferon) alfa-2a. Table 1 shows a comparison of the efficacy of these treatments.[1,2] To approve treatments for hepatitis B, the FDA and other regulatory authorities use criteria that are based on responses after one year of treatment. However, very few patients will have a sustained response after one year of treatment. The benefits versus the risks of long-term treatment of chronic hepatitis B have not been properly studied. Given the variable natural course of HBV infection and the high costs of treatments ($5,000 to $15,000 a year, in U.S. dollars), decisions regarding whom to treat and with what and for how long must be carefully weighed.

In their study in this issue of the *Journal*, Lau et al. report that a combination of peginterferon alfa-2a and lamivudine was associated with the greatest degree of virus suppression, followed by lamivudine monotherapy and peginterferon alone.[3] However, the rates of hepatitis B e antigen (HBeAg) seroconversion at the end of 48 weeks of treatment were similar among the three groups.

Unlike lamivudine and other nucleoside or nucleotide analogues, interferon has immune modulatory as well as antiviral effects. Previous studies involving three-to-six-month courses of conventional interferon showed that HBeAg seroconversion frequently occurred a few months after cessation of treatment, presumably because of the lag between immune priming and the decrease in the expression of viral proteins. The study by Lau et al. used a longer duration of peginterferon therapy; nonetheless, a small increment in the rate of HBeAg seroconversion was observed after treatment was stopped, so that at week 72, the two groups that received peginterferon had significantly higher rates of HBeAg seroconversion than the group that received lamivudine monotherapy.

The study reported by Hadziyannis et al. in this issue of the *Journal* showed the results at week 96 and week 144 of a phase 3 clinical trial of adefovir dipivoxil in patients with HBeAg-negative chronic hepatitis B.[6] An earlier report on this trial showed that at week 48, adefovir was associated with significantly higher rates of virologic, biochemical, and histologic responses than was placebo.[4] The study by Hadziyannis et al. showed that these responses were negated in virtually all patients after treatment was stopped at week 48. These disappointing results confirm that current treatments suppress but do not eradicate HBV. They also highlight the inadequacies of the end points of trials used for the approval of treatments for HBV.

Among the patients who continued to receive adefovir from week 49 through week 96 or from week 97 through week 144, the rates of virologic response (defined as a serum HBV DNA level that

Table 1. Comparison of the Percentages of Patients with Responses to Treatments for Hepatitis B.*

Variable	Conventional Interferon Alfa	Untreated Control	Lamivudine	Placebo Control	Adefovir	Placebo Control	Entecavir	Lamivudine Control	Peginterferon†	Lamivudine Control
	12 to 24 wk		52 wk		48 wk		48 wk		48 wk	
Patients with HBeAg-positive chronic hepatitis B										
Loss of serum HBV DNA‡	37	17	44	16	21	0	67	36	25	40
HBeAg seroconversion§	Difference, 18		16–18	4–6	12	6	21	18	27	20
Loss of HBsAg	8	2	<1	0	0	0	NA	NA	3	0
Normalization of alanine aminotransferase§	Difference, 23		41–72	7–24	48	16	68	60	39	62
Histologic improvement¶	NA	NA	49–56	23–25	53	25	72	6	38	34
Durability of response	80–90	NA	50–80	NA	NA	NA	17	NA	82‖	58‖
	6 to 12 mo		52 wk		48 wk		48 wk			
Patients with HBeAg-negative chronic hepatitis B										
Loss of serum HBV DNA‡	60–70	10–20	50–70	NA	51	0	90	72	63	73
Normalization of alanine aminotransferase	60–70	10–20	60–70	NA	72	29	78	71	38	73
Histologic improvement¶	NA	NA	60	NA	64	33	70	61	48	40
Durability of response	20–25	NA	<10	NA	<10	NA	NA	NA	Approximately 30	Approximately 10

* NA denotes not available, and HBsAg hepatitis B surface antigen. The data are from Lok and McMahon,[1] the Entecavir Review Team,[2] Lau et al.,[3] Hadziyannis et al.,[4] and Marcellin et al.[5] All numbers are the percentages of patients in the noted trials.

† Responses to peginterferon monotherapy at week 48 were lower in both patients with HBeAg-positive and HBeAg-negative chronic hepatitis B as compared with responses to a combination of peginterferon and lamivudine, but responses at week 72 (24 weeks after treatment) were similar in the two groups.

‡ The percentages for conventional interferon alfa and lamivudine were determined with the use of hybridization or branched DNA assay, and those for adefovir and entecavir with the use of a polymerase-chain-reaction assay.

§ There were wide variations in response rates among the studies[1,2]; thus, the percentages are based on the results of a meta-analysis.

¶ Follow-up biopsies in the peginterferon trials were performed at week 72 (24 weeks after treatment) and at week 48 or 52 in the lamivudine, adefovir, and entecavir trials.

‖ Additional patients in both groups had HBeAg seroconversion after the cessation of treatment; 32 percent in the peginterferon monotherapy group and 19 percent in the lamivudine group had HBeAg seroconversion at week 72.

was undetectable with the use of polymerase-chain-reaction assay) and biochemical response (normalization of levels of aminotransferases) were slightly higher than the rates after 48 weeks. However, 20 to 30 percent of the patients did not meet these criteria after 144 weeks of continuous therapy.

The main concerns with long-term treatment are side effects, drug resistance, and costs. Adefovir at high doses (≥30 mg per day) has been associated with nephrotoxicity.[4] In the study by Hadziyannis et al., nephrotoxicity was observed in 3 of 70 patients who received adefovir for three years, necessitating discontinuation of the treatment in two patients. Unlike resistance to lamivudine, resistance to adefovir is considered to be uncommon and to emerge later in the course of treatment. In the current analysis, adefovir-resistance mutations were detected in 6 of 70 patients who received adefovir for three years.[6] Thus, although response was maintained in most patients who continued treatment, nephrotoxicity and drug resistance will be of increasing concern with longer durations of treatment.

Ten years ago, conventional interferon was the only approved treatment for chronic hepatitis B. Since then, three orally administered nucleoside or nucleotide analogues and a long-acting (pegylated) interferon have been approved in the United States. Patients with chronic hepatitis B now have more treatment options that have fewer side effects and are more easily administered. This has broadened the indications for treatment for hepatitis B to include patients with decompensated HBV cirrhosis[7]

and patients who require HBV prophylaxis during chemotherapy for cancer.[8] In addition, long-term treatment with lamivudine has been shown to decrease the risk of hepatic failure and hepatocellular carcinoma among patients with cirrhosis and high levels of HBV DNA.[9]

However, these new therapies have brought along new problems. Foremost is drug-resistance mutations.[10] Selection of drug-resistance mutations is accompanied by virologic breakthrough (increased serum HBV DNA levels after initial suppression) and in some patients biochemical breakthrough (increased levels of aminotransferases after initial normalization) and, rarely, hepatic failure and death. In addition, resistance to one antiviral agent may confer resistance to other agents and may limit future treatment options. Another problem is the high rate of relapse when treatment is discontinued. Studies that compared peginterferon and lamivudine all showed a higher rate of virologic relapse when treatment with lamivudine was stopped.[3,5,11] Although adefovir and entecavir have not been directly compared with interferon, existing data suggest that relapse is more common than with interferon.

Given multiple treatment options that are less than ideal, who should be treated, with what, and when can treatment be stopped? The decision to treat or not to treat and the choice of treatment should be made jointly by the physician and the patient and should balance the benefits and the risks (e.g., the likelihood of a sustained response after a defined course of treatment or a maintained response during long-term treatment vs. the risk of progressive liver disease, side effects, drug resistance, and costs). For HBeAg-positive patients, viral suppression without HBeAg clearance is invariably associated with relapse, whereas viral suppression with HBeAg clearance is associated with sustained responses in 50 to 90 percent of patients.[1] For HBeAg-negative patients, relapse is frequent even when the virus has been suppressed to undetectable levels for more than a year.[12] Patients who opt for interferon must be aware of the wide array of potential side effects, whereas those who opt for oral antiviral therapy must be aware of the need for long-term treatment and the risks of drug resistance.

For patients with HBeAg-positive chronic hepatitis B who do not yet have cirrhosis, the goal is to achieve HBeAg seroconversion. Because pretreatment aminotransferase levels are a strong predictor of HBeAg seroconversion (except in the study by Lau et al.),[13] current guidelines do not recommend treatment of patients with normal aminotransferase levels unless liver biopsy shows substantial inflammation or fibrosis.[1,14,15] For patients with HBeAg-negative chronic hepatitis B who do not yet have cirrhosis, a one-year course of treatment is associated with a 15 to 35 percent chance of sustained response after interferon therapy but a less than 10 percent chance after treatment with lamivudine or adefovir.[1,6,10] Given the need for long-term treatment, current guidelines recommend treatment only for patients with elevated aminotransferase levels or histologic evidence of moderate or severe inflammation or advanced fibrosis.[1,14,15] For patients with cirrhosis, the potential gains are higher. Treatment is recommended for patients with high HBV DNA levels, but it is unclear whether patients with low HBV DNA levels will derive the same benefits. It is also unknown whether treatment should be lifelong or whether clinical benefit can be maintained after several years of treatment. Given the propensity for HBV to persist, patients should be closely monitored when treatment is stopped, to avoid fatal flares.

Substantial progress has been made in treatments for hepatitis B in the past decade. However, finding an exit through the maze of new therapies remains a challenge, underscoring the need for careful deliberation before initiating treatment.

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Adjuvant Therapy for Colon Cancer — The Pace Quickens

Carmen Allegra, M.D., and Daniel J. Sargent, Ph.D.

Every year in the United States, approximately 30,000 people receive the diagnosis of lymph-node–positive colon cancer (stage III); worldwide, the number approaches 200,000. The primary therapy for this condition is surgical resection, which cures 50 to 60 percent of patients with average-risk stage III disease.[1,2] During the past 15 years, sequential advances in chemotherapy after surgical resection (adjuvant chemotherapy) have had an irrefutable and substantial benefit, with the 4-year rate of overall survival approaching 80 percent.[3] In this issue of the *Journal*, Twelves and colleagues report on the Xeloda in Adjuvant Colon Cancer Therapy (X-ACT) trial, in which 1987 patients with stage III colon cancer were randomly assigned to adjuvant therapy with either bolus intravenous fluorouracil and leucovorin, given for 5 consecutive days every 28 days, or to oral capecitabine, given twice daily for 14 of every 21 days.[4] Both regimens were given for a total of 24 weeks.

Capecitabine, an oral fluoropyrimidine, was approved in the United States in 2001 for the treatment of metastatic colorectal cancer. The direct comparison of bolus fluorouracil and leucovorin with capecitabine in two large randomized trials involving patients with metastatic colorectal cancer showed that the overall survival was equivalent with the two regimens.[5] The convenience of oral administration, coupled with a favorable profile of toxic effects, formed a compelling basis for testing capecitabine in the adjuvant setting.

Fluorouracil has been the backbone of colorectal-cancer management for almost 50 years. Capecitabine, a prodrug, requires a multistep activation that culminates in its conversion to fluorouracil at the cellular level. Although the plasma half-life of fluorouracil is less than 10 minutes, the half-life of capecitabine is approximately 45 minutes; thus, twice-daily administration of capecitabine results in plasma levels that more closely resemble the levels achieved with protracted fluorouracil infusions than those obtained with daily or weekly bolus administration of fluorouracil. The small benefit of infusion over bolus administration of fluorouracil in advanced disease has not translated into an improved outcome in the adjuvant setting, where these two methods of fluorouracil delivery have similar efficacy.[6,7]

What about the efficacy of capecitabine in stage III colon cancer? On the basis of the similarity in efficacy of infusional and bolus therapy with fluorouracil in the adjuvant setting and the period of time in which the study was conducted, the choice of the control treatment, bolus fluorouracil and leucovorin, in the trial by Twelves and colleagues was appropriate. Despite the lack of central randomization, the demographic characteristics of the participants suggest that the two groups were well balanced. The rate of three-year disease-free survival in the control group (60.6 percent) is consistent with other reports of results with fluorouracil and leucovorin.[3,6] The prospectively defined primary end point was disease-free survival, which is appropriate in the setting of adjuvant therapy for colon cancer; the upper limit of the hazard ratio of 1.20 for noninferiority was also appropriate.[8] On the basis of these considerations, we can confidently conclude that capecitabine is at least equivalent to intravenous fluorouracil and leucovorin, with a P value excluding inferiority of $P<0.001$. Disease-free survival,

relapse-free survival (as distinguished from disease-free survival by the exclusion of deaths from causes other than colon cancer), and overall survival all numerically favored capecitabine, with P values hovering close to 0.05. Although there is a suggestion that capecitabine may be superior to fluorouracil and leucovorin in stage III colon cancer, these data indicate that capecitabine is at least as efficacious as fluorouracil and leucovorin, as the authors have appropriately concluded.

Capecitabine had significantly less overall toxicity than bolus fluorouracil and leucovorin, with most of the difference attributable to a decrease in the incidence of neutropenia. A higher incidence of hyperbilirubinemia of uncertain clinical significance was noted with capecitabine, and as in previous trials in metastatic colorectal cancer, capecitabine was associated with an incidence of 17 percent of severe palmar–plantar erythrodysesthesia (hand–foot syndrome).

The starting dose of capecitabine (2500 mg per square meter of body-surface area given daily in two divided doses) in the study by Twelves et al. required a treatment modification in almost 60 percent of the patients. This dose of capecitabine, specified in the package insert, is generally felt to be poorly tolerated in the U.S. population, a situation that has led many oncologists in the United States to decrease the starting dose for most patients. The reason for the differential tolerance of capecitabine in the United States and other countries is unclear, but it may be related to the abundant supplementation of the U.S. diet with folic acid, a well-known enhancer of the toxicity of the fluoropyrimidines. The efficacy of capecitabine at doses typically given in the United States is critical, particularly in the adjuvant setting, where therapy is given with curative intent. Data in the present report should not be extrapolated to doses not tested, since efficacy at lower starting doses is unknown.

In the treatment of metastatic colorectal cancer, multiagent therapy has become the standard of care in the United States. This standard is based on studies that support the role of fluoropyrimidines and either oxaliplatin or irinotecan, an inhibitor of topoisomerase I, plus the antiangiogenic antibody bevacizumab.[9,10] In the adjuvant setting, the recent Multicenter International Study of Oxaliplatin/5-Fluorouracil/Leucovorin in the Adjuvant Treatment of Colon Cancer (MOSAIC) trial and the National Surgical Adjuvant Breast and Bowel Project (NSABP) C-07 trial have demonstrated that adding oxaliplatin to a biweekly schedule of infused (MOSAIC) or weekly bolus (C-07) fluorouracil and leucovorin significantly prolongs disease-free survival in stage II and III colon cancer, as compared with leucovorin-modulated fluorouracil, with relative risk reductions of 23 percent and 21 percent, respectively.[3,11] Table 1 shows the benefit for three-year disease-free survival of the addition of oxaliplatin to a fluoropyrimidine-based regimen among patients with phase III colon cancer. Randomized investigations comparing bolus or infusion schedules of fluorouracil, leucovorin, and irinotecan in

Table 1. Three-Year Disease-free Survival among Patients with Stage III Colon Cancer Treated with Adjuvant Chemotherapy.*

Trial	Fluorouracil Schedule	Three-Year Disease-free Survival (%)	Hazard Ratio (95% CI)	Absolute Difference in Three-Year Disease-free Survival (%)
X-ACT				
Fluorouracil plus leucovorin	Bolus	60.6		
Capecitabine	Oral	64.2	0.87 (0.75–1.00)	3.6
MOSAIC				
Fluorouracil plus leucovorin	Infusion	65.3		
Fluorouracil, leucovorin, and oxaliplatin	Infusion	72.2	0.76 (0.62–0.92)	6.9
NSABP C-07				
Fluorouracil plus leucovorin	Bolus	65.5		
Fluorouracil, leucovorin, and oxaliplatin	Bolus	72.2	0.77 (0.65–0.92)	6.7

* X-ACT denotes Xeloda in Adjuvant Colon Cancer Therapy, MOSAIC Multicenter International Study of Oxaliplatin/5-Fluorouracil/Leucovorin in the Adjuvant Treatment of Colon Cancer, NSABP National Surgical Adjuvant Breast and Bowel Project, and CI confidence interval.

patients with stage III colon cancer have not demonstrated the superiority over leucovorin-modulated fluorouracil.[12,13]

Given the available data, where does capecitabine fit into the current management of locally advanced colon cancer? On the basis of convenience, efficacy, and favorable rates of toxic effects, capecitabine is an excellent choice when single-agent therapy is desired, provided that the treating physician is comfortable using the starting dose reported in the present investigation. The question of whether capecitabine should replace intravenous fluorouracil in multiagent adjuvant regimens, such as in combination with oxaliplatin, will not be answered until ongoing investigations have been completed. Meanwhile, current evidence favors combinations of oxaliplatin with intravenous fluorouracil and leucovorin as optimal adjuvant therapy.

Although chemotherapy in stage III colon cancer is a well-established standard, much controversy surrounds the use of chemotherapy in stage II disease. Given recent reports of a smaller but consistent therapeutic benefit seen with fluorouracil-based treatment in stage II disease,[2,14] one could consider capecitabine as an option in patients with stage II disease. However, the present trial offers no guidance on this possibility, because it included only patients with stage III disease. A strategy of including patients with stage II or III disease in a clinical trial, as was done in the MOSAIC and NSABP trials, may be preferable in future investigations.

The prospects for a higher rate of cure in patients with locally advanced colon cancer are extremely promising, given the emerging evidence that supports therapeutic targeting of growth factors, growth-factor receptors, and downstream pathways. Both bevacizumab, a vascular endothelial growth factor antibody, and cetuximab, an endothelial growth factor–receptor antibody, have demonstrated benefit in advanced colorectal cancer.[10,15] Their value in less advanced colon cancer is the subject of ongoing randomized investigations worldwide. Clearly, future investigations must identify specific patient populations that do not require postsurgical therapy as well as patients at high risk for relapse who may benefit from additional therapeutic intervention. The present state of generic therapy based on disease stage and histologic site of origin must give way to molecularly guided, personalized therapies, shifting the risk–benefit ratio of chemotherapeutic intervention toward an overwhelming individualized patient benefit — a goal clearly within our collective technological reach.

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Immunosuppression in Cardiac Transplantation

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In this issue of the *Journal,* Hershberger and colleagues present data from a multicenter trial of immunosuppression in cardiac-transplant recipients showing that daclizumab, a monoclonal antibody against the interleukin-2 receptor, reduces the overall incidence of cellular rejection.[1] In discussing these data, I will focus on three areas: the role of and difficulty in carrying out randomized, controlled trials of immunosuppressive regimens in heart-transplant recipients, the end points one is forced to choose in performing such studies, and the clinical relevance of the primary end point of cellular rejection, as well as the possibility of a negative outcome.

Any group that successfully conducts a randomized clinical trial involving heart-transplant recipients should be acknowledged and commended. The first human-to-human cardiac transplantation was performed in 1967. The results of the first randomized, controlled clinical trial of immunosuppression among heart-transplant recipients were published in 1998,[2] more than 30 years later. Since 1998, three other moderate-sized randomized, controlled trials of immunosuppression among heart-transplant recipients, including that by Hershberger et al., have been carried out.[1,3,4]

Why has it been so difficult to conduct what would seem to be logical trials, given the dramatic expansion of our clinical immunosuppression armamentarium? The most obvious reason is the relatively small number of transplantations. In the United States, our average annual heart-transplantation volume is 2500, and this number has not changed appreciably since 1991.[5] Also relevant is the fact that approximately 150 heart-transplantation centers perform this small number of operations. Studies are then focused on the larger cardiac-transplantation centers, all of which are sure that their particular immunosuppression cocktail is clearly the best. Hence, the negotiations among centers regarding which therapies are allowed and which are excluded can be complex. A good example of this is that in all three previously reported trials, approximately 50 percent of the centers used early antibody therapy and 50 percent did not, according to their local protocols. Furthermore, in centers using antibodies, some used monoclonal antibodies, and others polyclonal preparations. Finally, any sign of trouble, given the high stakes involved, resulted in removal of the patient from his or her assigned group, leading to an approximate rate of dropout or crossover of 30 percent in all of these studies. What was left was at best 200 and at worst 100 patients per group who could be evaluated, spread among 30 centers, with half the patients given additional and diverse antibody therapy.

The next important issue in trials assessing immunosuppression after heart transplantation is selecting the most appropriate end point. In 1992, I was involved in the early development of the trial by Kobashigawa et al. comparing mycophenolate mofetil with azathioprine.[2] We decided that biopsy-proven rejection was an "objective" and frequent end point in heart transplantation and should probably be the primary end point of the study. The reasoning behind this decision was the realization that with one-year survival rates at that time approaching 85 percent, the number of patients required to endow a mortality trial with sufficient statistical power to detect a treatment effect would be prohibitive. Conversely, since the incidence of acute rejection on surveillance biopsy was, depending on the center, 50 percent or higher in the first year, the enrollment of small numbers of patients would yield a sufficient number of end points to have an appropriately powered study. The assumption, of course, was that patients in whom rejection occurred might have a poorer overall outcome than those without rejection. Ultimately, in this study, there was a lower rate of death among patients taking mycophenolate mofetil than among those who received azathioprine (6 percent vs. 11 percent), and there was also a lower rate of treated episodes of rejection (66 percent vs. 74 percent), but clearly, many more patients had an episode of rejection than died. In all the subsequent studies, including the study by Hershberger et al., the rate of the primary end point (usually a composite of histologic rejection, hemodynamic compromise, and death) differed significantly between groups, but what truly differed between groups was the incidence of histologic rejection. In the studies by Eisen et al.[3] and Kobashigawa et al.,[4] the mortality rates were similar. In the trial by Hershberger et al., the mortality trends are worrisome.

All of us who regularly treat heart-transplant

recipients know that most patients with histologic rejection have absolutely no change in allograft function, and the histologic findings are usually easily reversed with an augmentation of immunosuppressive therapy. Conversely, many patients with acute hemodynamic compromise do not have the classic findings of acute rejection. Although rejection is a documented cause of death primarily in the first year after transplantation, it has become exceedingly unusual. In a recent multicenter survey of almost 7300 patients, rejection was a cause of death at one year in only 2 percent.[6] Contrast that to the incidence of histologic rejection of 50 to 75 percent in these other studies. Clearly, the results of the multicenter, randomized trials completed to date cannot provide support for a direct relationship between histologic rejection and outcome.

Hershberger and colleagues found that the risk of their primary composite end point of histologic rejection, graft dysfunction, a second transplantation, or death within six months was reduced by treatment with the monoclonal antibody daclizumab. As in prior studies, only 81 percent of the patients received four or five doses of the assigned study drug. The efficacy benefit of the active drug was solely accounted for by the histologic-rejection component of the composite end point. A worrisome finding was that overall mortality was greater in the daclizumab group than in the placebo group (6.5 percent vs. 3.2 percent) at six months, and this trend continued at one year (P=0.11). Also worrisome was the higher incidence of graft dysfunction in the daclizumab group. The authors did an admirable job in trying to sort out reasons for this apparent increase in mortality in the active-treatment group and discovered that most of the deaths were due to infections and occurred in patients who received both daclizumab and a second monoclonal or polyclonal preparation of antilymphocyte antibody. They conclude that although daclizumab reduces the risk of histologic rejection, "concurrent or anticipated use of cytolytic therapy with daclizumab should be avoided."

How does one integrate the results of this and prior heart-transplantation studies into clinical practice? The vast majority of episodes of histologic rejection are treated effectively with a moderate increase in immunosuppressive therapy. Most certainly, the worst part of having to treat rejection is telling the patient about the diagnosis and dealing with the patient's anxiety. I personally do not know how I can "anticipate" a patient's need for antilymphocyte-antibody therapy in order to identify those I can and those I cannot treat with daclizumab, and I would certainly be reticent to limit my options, should the episode of rejection be more serious and require antilymphocyte-antibody therapy.

Finally, what am I willing to trade for this reduction in the risk of histologic rejection? Everolimus therapy reduces the risk of histologic rejection but increases the risk of renal dysfunction,[3] a trade-off that may be acceptable, since the risk of coronary disease after transplantation may be reduced. Tacrolimus reduces the risk of histologic rejection but increases the incidence of diabetes, which may also be considered an acceptable trade-off. In contrast, on the basis of the data from the current trial of daclizumab, I am certain that I would not be willing to trade even a small increase in the risk of death from infection for a reduction in the risk of histologic rejection.

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CORRESPONDENCE

Aspirin in the Prevention of Cardiovascular Disease in Women

TO THE EDITOR: Ridker et al. (March 31 issue)[1] conclude that primary prophylaxis with aspirin to prevent myocardial infarction is ineffective in young, healthy women. However, the majority of patients in this study (84.5 percent) had a 10-year risk of less than 5 percent for an incident myocardial infarction and therefore would not have received aspirin as primary prophylaxis, according to the American Heart Association guidelines. The American Heart Association published recommendations in 2002 stating that aspirin should be used as primary prevention for coronary events in persons with a 10-year risk of an incident myocardial infarction that is greater than 10 percent.[2] Although the results of the study by Ridker et al. are interesting, we need a trial based on practice according to current, established guidelines.

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TO THE EDITOR: The press coverage of the study by Ridker et al. indicated that aspirin does not prevent heart attacks in women. This message is consistent with the abstract's concluding statement, "In this large, primary-prevention trial among women, aspirin lowered the risk of stroke without affecting the risk of myocardial infarction." However, this conclusion is misleading. A more accurate statement would be that very-low-dose aspirin (100 mg every other day) is not effective in preventing myocardial infarction in women.

In the Primary Prevention Project trial,[1] 100 mg of aspirin per day was effective in preventing myocardial infarction in women and in men. However, in the Hypertension Optimal Treatment trial,[2] 75 mg of aspirin per day was effective as prevention in men but ineffective in women. It would not be unreasonable to conclude from these three trials that the minimum dose of aspirin needed for a cardioprotective effect is higher in women than in men and is greater than 75 mg per day. The study by Ridker et al. did not establish that aspirin is ineffective in preventing myocardial infarction in women.

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THIS WEEK'S LETTERS

THE AUTHORS REPLY: Dr. Schwartz correctly notes that the majority of the participants in the Women's Health Study were at low risk for coronary heart disease, as measured by the Framingham risk score. However, 1100 participants did have a risk of coronary heart disease that was 10 percent or greater. Among these high-risk participants, the findings were consistent with the overall findings of the trial, with no significant benefit with respect to the primary end point of major cardiovascular events for women taking aspirin as compared with those taking placebo (61 events in the aspirin group and 60 in the placebo group, P=0.74); there was a benefit for total stroke (17 events vs. 32, respectively; P=0.04) and a trend toward benefit for ischemic stroke (16 vs. 29, P=0.07), and there was no benefit for myocardial infarction (32 vs. 23, P=0.15). Thus, although overall the population had a low risk of cardiovascular events, it is important to note that there was no evidence of a modification of the effect of aspirin according to levels of the Framingham risk score in our study.

Dr. Dalen is correct in pointing out that our trial demonstrated that the specific dose of 100 mg of aspirin every other day was not associated with a reduction in myocardial infarction overall, and he raises the important question of whether this very low dose was inadequate to produce a cardioprotective effect in women. Although we agree that it is certainly possible that the dose was inadequate, there was no direct evidence to support this in the Women's Health Study. We showed that levels of thromboxane and prostacyclin were reduced with 100 mg of aspirin every other day; we observed the expected increased risk of gastrointestinal bleeding, hemorrhagic stroke, nongastrointestinal bleeding, and peptic ulcer, and 100 mg every other day was adequate both to lower the risk of stroke overall and to lower the risk of myocardial infarction as well as stroke in women 65 years of age or older. However, the issue of the lowest effective dose in both women and men requires further research.

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The Ubiquilin 1 Gene and Alzheimer's Disease

TO THE EDITOR: Bertram et al. (March 3 issue)[1] report that in two family-based cohorts, a genetic variant of the UBQ-8i single-nucleotide polymorphism on chromosome 9q22 putatively increased the risk of Alzheimer's disease in an additive disease model. We attempted replication in a similarly ascertained but independent family-based data set based on 288 families in which linkage to microsatellites at 9q22.1 and 9q34.2 was demonstrated in a genome scan.[2] In addition, we analyzed a previously described independent data set based on patients with Alzheimer's disease and 1005 controls.[3]

We found no association between the risk of Alzheimer's disease and UBQ-8i, or any of six additional single-nucleotide polymorphisms within the *UBQLN1* gene, in either of the independent data sets. However, using age at onset as the trait of interest, we found a significant association between the putative UBQ-8i risk allele and an older age at onset in a recessive-disease model only in our case–control data set (Table 1). We found an additional, significant effect related to age at onset only in our family-based data set with a different single-nucleotide polymorphism in *UBQLN1*. Thus, although we found no evidence of risk with any single-nucleotide polymorphism in *UBQLN1*, our results suggest that age at onset may be germane and that additional, detailed examination of *UBQLN1*, including a search for the functional variant (or variants), is warranted.

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1. Bertram L, Hiltunen M, Parkinson BS, et al. Family-based association between Alzheimer's disease and variants in *UBQLN1*. N Engl J Med 2005;352:884-94.
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Table 1. UBQ-8i Allelic Associations.*

Analysis	Family-Based Data Set†		Patient–Control Data Set‡	
	Test§	P Value	Test	P Value
Risk analysis	Family-Based Association Test	0.87	Logistic regression (additive model)	0.23
	Pedigree Disequilibrium Test	0.96		
Analysis of age at onset	Quantitative Transmission Disequilibrium Test	0.21	Linear regression (recessive model)	0.01

* All the patients with Alzheimer's disease met the National Institute of Neurological Disorders and Stroke/Alzheimer's Disease and Related Disorder Association case definition and were evaluated at Duke University Medical Center and Vanderbilt University Medical Center through the collaborative Alzheimer Project.
† In the family-based data set, the mean (±SD) age at onset was 71.1±7.0 years (range, 50 to 59), and 63 percent of the subjects were women.
‡ In the patient–control data set, the mean age at onset was 70.8±6.6 years (range, 51 to 87), and 67 percent of the subjects were women.
§ The Family-Based Association Test is described by Bertram et al.,[1] the Pedigree Disequilibrium Test by Martin et al.,[4] and the Quantitative Transmission Disequilibrium Test by Abecasis et al.[5]

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THE AUTHORS REPLY: We are pleased by Slifer and colleagues' report of a significant genetic association between single-nucleotide polymorphisms in the *UBQLN1* gene and the age at onset of Alzheimer's disease in two separate and independent samples, providing further support for our original finding that *UBQLN1* variants may influence the pathogenesis of Alzheimer's disease.[1] Given the significant age-at-onset effects in their samples, it is puzzling that they did not observe significant effects on the risk of disease, since these two traits are correlated. Possible explanations include allelic heterogeneity, lack of power in the risk analyses, and methodologic differences in sample ascertainment and statistical procedures.

It is also worth noting that the two original linkage signals (according to a binary phenotype definition) reported for the family-based sample analyzed by Slifer et al.[2] were located more than 40 cM away (in either direction) from *UBQLN1* and our linkage peak.[3] Thus, their sample may not be optimal for detecting risk effects of the magnitude described in our initial report.[3] Notwithstanding these differences, the fact that several *UBQLN1* single-nucleotide polymorphisms now show an association with either the age at onset or the risk of Alzheimer's disease in a number of independent samples suggests the possibility of linkage disequilibrium with one or more additional pathogenic variants.

Along these lines, since our original report,[1] we have now found additional evidence of an association between the risk of Alzheimer's disease and the T allele of a *UBQLN1* promoter single-nucleotide polymorphism (rs12345514) in both the National Institutes of Mental Health family sample (P=0.02 by the Family-Based Association Test) and the Consortium on Alzheimer's Genetics family sample (P=0.04). The combined data sets yielded the strongest evidence of an association (P=0.002), which is consistent with our previous findings. Collectively, these data suggest that the pathogenesis of Alzheimer's disease may be influenced by changes not only in the splicing of *UBQLN1*,[1] but also in its expression. Ultimately, meta-analysis of these and additional association studies[4] should provide a more precise measure of the actual contribution of *UBQLN1* variants to Alzheimer's disease.

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Cardiac Revascularization in Specialty and General Hospitals

TO THE EDITOR: We investigated whether the findings of Cram et al. (April 7 issue)[1] regarding severity of illness, mortality, and length of stay among Medicare patients undergoing revascularization in specialty hospitals could be generalized to an all-payer population. We used data for January 2002 through September 2004 from Solucient's all-payer Projected Inpatient Database, which contains information on more than 17 million discharges in the United States annually and has been used for the analysis of patients with cardiac disease.[2] We identified specialty hospitals using the 2003 General Accounting Office definition,[3] modified to include only hospitals with a majority of patients in Major Diagnostic Category 5 (denoting those with diseases of the circulatory system). Nonspecialty hospitals were in the same hospital-referral regions.[4]

We found that patients who underwent percutaneous coronary intervention (PCI) or coronary-artery bypass grafting (CABG) at specialty hospitals were less severely ill than those who underwent one of these procedures at general hospitals, on the basis of the probability of death according to All Patient Refined Diagnosis Related Groups.[5] Although the unadjusted mortality rate for PCI was lower for specialty hospitals, no significant difference remained after adjustment for patient characteristics, admission source, hospital-referral region, payer, and volume of procedures. Unadjusted and adjusted lengths of stay for PCI and unadjusted length of stay for CABG were significantly lower for specialty hospitals. The adjusted length of stay for CABG did not differ significantly between the two types of hospital (Table 1).

Table 1. Risk of Death, Mortality Rate, and Length of Stay in Patients Undergoing PCI and CABG in Specialty Hospitals, as Compared with Nonspecialty Hospitals.*

Variable†	Specialty Hospitals	Nonspecialty Hospitals	Odds Ratio (95% CI)	P Value
PCI				
No. of patients	4123	13,248		
No. of hospitals	8	33		
Probability of death	0.011	0.017		<0.001
Mortality rate (%)				
Unadjusted	0.90	1.83	0.487 (0.340–0.683)	
Adjusted	1.64	1.60	1.031 (0.779–1.353)	
Average length of stay (days)				
Unadjusted	2.43	3.34		<0.001
Adjusted	2.99	3.15		0.64
CABG				
No. of patients	1680	6,155		
No. of hospitals	8	23		
Probability of death	0.032	0.038		0.003
Mortality rate (%)				
Unadjusted	2.62	2.65	0.989 (0.726–1.430)	
Adjusted	2.76	2.61	1.048 (0.746–1.452)	
Average length of stay (days)				
Unadjusted	7.75	8.84		<0.001
Adjusted	8.55	8.62		0.002

* PCI denotes percutaneous coronary intervention, CABG coronary-artery bypass grafting, and CI confidence interval.

† The probability of death is based on All Patient Refined Diagnosis Related Groups (APR-DRG), version 15.0.[5] The adjusted mortality rate and length of stay have been adjusted for the APR-DRG probability of death and length-of-stay weight, respectively, and also for age, sex, payer, admission source, hospital-referral region, volume of PCI or CABG procedures, and year of discharge.

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TO THE EDITOR: In the article by Cram et al., the confidence intervals of the adjusted odds ratios are consistent with a large survival benefit when revascularization is performed in specialty hospitals: a 31 percent reduction in death for PCI and 28 percent for CABG. When adjustment is also made for the hospital's volume of procedures (inappropriately, given its role in the causal pathway related to hospital quality and efficiency), the reduction is 17 percent for PCI and 26 percent for CABG. The analysis

omits longer-term survival and clinically specific process-of-care data in the public domain. In this regard, Baylor Heart and Vascular Hospital has consistently achieved 100 percent performance for these measures while disseminating its best practices to improve care across Baylor hospitals (www.hospitalcompare.hhs.gov/). The low power to detect differences in length of stay is reflected in the confidence intervals of the adjusted odds ratios, which are consistent with a 15 percent shorter length of stay for specialty hospitals. Finally, cost analyses might reveal more efficient resource use within a given length of stay, as has been observed for the use of pharmaceuticals and other supplies in the Baylor cardiovascular specialty hospital.

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TO THE EDITOR: Cram et al. used Medicare Provider Analysis and Review (MedPAR) data to examine relationships between specialty hospitals and general hospitals. Although the study was well conceived, we are concerned about its total reliance on Medicare-specific administrative data. As acknowledged by the authors, claims data may be inadequate to reflect severity of illness meaningfully. This inherent deficiency of administrative data may be understated in the study by Cram et al.

We believe that the conclusions are mitigated by the shortcomings of administrative data. We strongly advocate the use of clinical data in investigations linked to severity of illness. The advantages of clinical data have been clearly illustrated in recent years,[1,2] and significant differences in patient volume and outcome have been demonstrated in comparisons of clinical and administrative data.[2]

We recognize that data from administrative databases provide reasonable information for the assessment of general quality, but the inherent disadvantages of claims data necessarily limit the conclusions. Accordingly, we maintain that data from clinical databases provide the optimal analytic tool for investigations in cardiac surgery.

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THE AUTHORS REPLY: We are pleased that Dr. Young and colleagues found the results of our analyses of data on Medicare patients to be largely generalizable to an all-payer population. Their analyses are timely, since roughly half of revascularization procedures are performed in non-Medicare patients.

We generally agree with the issues raised in Dr. Ballard's letter, but we would note that it is not customary to focus on the lower bounds of a 95 percent confidence interval when interpreting study results. A more balanced interpretation of our results is that the odds of death for patients at specialty hospitals was 11 percent lower for PCI and 16 percent lower for CABG after adjustment for patient characteristics. We agree that it is not unreasonable to argue that, because specialization is closely tied to procedural volume, comparisons of specialty and general hospitals should be based on risk-adjusted (but not volume-adjusted) mortality rates. However, given that some have argued that there is an inherent advantage in specialization above and beyond the "volume effect," we felt it was important to control for volume in our analyses in an attempt to tease apart these differences.

We believe that our analyses indicate that the lower unadjusted mortality at specialty hospitals is largely attributable to the lower average risk of the patients such hospitals treat and to the hospitals' greater procedural volumes. Perhaps most informative were the results of stratified analyses that directly compared lower-volume and higher-volume specialty and general hospitals and did not show significant differences. Finally, we agree with Dr. Ballard that other indicators of quality, such as process-of-care measures, which are difficult to derive from claims data, should ideally be examined.

We also agree with Dr. Edwards and colleagues that analyses based on administrative data are subject to a number of potential methodologic limi-

tations when the quality of care is assessed. However, the lack of registries with detailed clinical information that can be used to track patient outcomes on a national level makes it impossible to conduct comparative analyses of specialty and general hospitals, since the validity of such analyses is predicated on the availability of data from all hospitals in the markets studied. We believe that, in the absence of national clinical registries, use of available administrative databases to provide empirical insight into important questions relevant to policy is preferable to not addressing these questions; however, the limitations of such analyses should be clearly recognized.

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Polycystic Ovary Syndrome

TO THE EDITOR: Ehrmann's review article on the polycystic ovary syndrome (March 24 issue)[1] misses the liver issue. Alanine aminotransferase activity is abnormal in 30 percent of patients with the polycystic ovary syndrome in whom causes other than nonalcoholic fatty liver disease were excluded.[2] The high rates of prevalence of impaired glucose tolerance and type 2 diabetes mellitus among patients with the polycystic ovary syndrome confirm that chronic nonalcoholic liver disease and its consequences are an important issue in these patients.[3] Accordingly, the use of thiazolidinediones to treat patients with the polycystic ovary syndrome requires serious caution. The Food and Drug Administration recommends that liver enzymes be checked at the start of therapy, quarterly during the first year, and periodically thereafter. Moreover, even though the short-term studies presented look promising, caution would seem to be indicated because long-term therapy with thiazolidinediones in obese mice with diabetes results in severe hepatic centrilobular steatosis.[4] Long-term data will be important to obtain from studies of patients with the polycystic ovary syndrome.

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TO THE EDITOR: Autoimmune thyroiditis should be added to the group of disorders associated with the polycystic ovary syndrome. Sex steroid hormones appear to be the most important environmental factors contributing to the development of autoimmune thyroiditis.[1] Furthermore, a recent prospective study demonstrated a high prevalence of autoimmune thyroiditis among women with the polycystic ovary syndrome.[2] Since hypothyroidism has been shown to make the polycystic ovary syndrome worse,[3] physicians should carefully check thyroid function and thyroid-specific autoantibodies in such patients.

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TO THE EDITOR: We were concerned about Dr. Ehrmann's statement that "there is an increased prevalence of endometrial hyperplasia and carcinoma in women with the polycystic ovary syndrome." The common occurrence of risk factors for endo-

Cardiac Revascularization in Specialty and General Hospitals

TO THE EDITOR: We investigated whether the findings of Cram et al. (April 7 issue)[1] regarding severity of illness, mortality, and length of stay among Medicare patients undergoing revascularization in specialty hospitals could be generalized to an all-payer population. We used data for January 2002 through September 2004 from Solucient's all-payer Projected Inpatient Database, which contains information on more than 17 million discharges in the United States annually and has been used for the analysis of patients with cardiac disease.[2] We identified specialty hospitals using the 2003 General Accounting Office definition,[3] modified to include only hospitals with a majority of patients in Major Diagnostic Category 5 (denoting those with diseases of the circulatory system). Nonspecialty hospitals were in the same hospital-referral regions.[4]

We found that patients who underwent percutaneous coronary intervention (PCI) or coronary-artery bypass grafting (CABG) at specialty hospitals were less severely ill than those who underwent one of these procedures at general hospitals, on the basis of the probability of death according to All Patient Refined Diagnosis Related Groups.[5] Although the unadjusted mortality rate for PCI was lower for specialty hospitals, no significant difference remained after adjustment for patient characteristics, admission source, hospital-referral region, payer, and volume of procedures. Unadjusted and adjusted lengths of stay for PCI and unadjusted length of stay for CABG were significantly lower for specialty hospitals. The adjusted length of stay for CABG did not differ significantly between the two types of hospital (Table 1).

Table 1. Risk of Death, Mortality Rate, and Length of Stay in Patients Undergoing PCI and CABG in Specialty Hospitals, as Compared with Nonspecialty Hospitals.*

Variable†	Specialty Hospitals	Nonspecialty Hospitals	Odds Ratio (95% CI)	P Value
PCI				
No. of patients	4123	13,248		
No. of hospitals	8	33		
Probability of death	0.011	0.017		<0.001
Mortality rate (%)				
Unadjusted	0.90	1.83	0.487 (0.340–0.683)	
Adjusted	1.64	1.60	1.031 (0.779–1.353)	
Average length of stay (days)				
Unadjusted	2.43	3.34		<0.001
Adjusted	2.99	3.15		0.64
CABG				
No. of patients	1680	6,155		
No. of hospitals	8	23		
Probability of death	0.032	0.038		0.003
Mortality rate (%)				
Unadjusted	2.62	2.65	0.989 (0.726–1.430)	
Adjusted	2.76	2.61	1.048 (0.746–1.452)	
Average length of stay (days)				
Unadjusted	7.75	8.84		<0.001
Adjusted	8.55	8.62		0.002

* PCI denotes percutaneous coronary intervention, CABG coronary-artery bypass grafting, and CI confidence interval.

† The probability of death is based on All Patient Refined Diagnosis Related Groups (APR-DRG), version 15.0.[5] The adjusted mortality rate and length of stay have been adjusted for the APR-DRG probability of death and length-of-stay weight, respectively, and also for age, sex, payer, admission source, hospital-referral region, volume of PCI or CABG procedures, and year of discharge.

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TO THE EDITOR: In the article by Cram et al., the confidence intervals of the adjusted odds ratios are consistent with a large survival benefit when revascularization is performed in specialty hospitals: a 31 percent reduction in death for PCI and 28 percent for CABG. When adjustment is also made for the hospital's volume of procedures (inappropriately, given its role in the causal pathway related to hospital quality and efficiency), the reduction is 17 percent for PCI and 26 percent for CABG. The analysis

omits longer-term survival and clinically specific process-of-care data in the public domain. In this regard, Baylor Heart and Vascular Hospital has consistently achieved 100 percent performance for these measures while disseminating its best practices to improve care across Baylor hospitals (www.hospitalcompare.hhs.gov/). The low power to detect differences in length of stay is reflected in the confidence intervals of the adjusted odds ratios, which are consistent with a 15 percent shorter length of stay for specialty hospitals. Finally, cost analyses might reveal more efficient resource use within a given length of stay, as has been observed for the use of pharmaceuticals and other supplies in the Baylor cardiovascular specialty hospital.

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TO THE EDITOR: Cram et al. used Medicare Provider Analysis and Review (MedPAR) data to examine relationships between specialty hospitals and general hospitals. Although the study was well conceived, we are concerned about its total reliance on Medicare-specific administrative data. As acknowledged by the authors, claims data may be inadequate to reflect severity of illness meaningfully. This inherent deficiency of administrative data may be understated in the study by Cram et al.

We believe that the conclusions are mitigated by the shortcomings of administrative data. We strongly advocate the use of clinical data in investigations linked to severity of illness. The advantages of clinical data have been clearly illustrated in recent years,[1,2] and significant differences in patient volume and outcome have been demonstrated in comparisons of clinical and administrative data.[2]

We recognize that data from administrative databases provide reasonable information for the assessment of general quality, but the inherent disadvantages of claims data necessarily limit the conclusions. Accordingly, we maintain that data from clinical databases provide the optimal analytic tool for investigations in cardiac surgery.

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THE AUTHORS REPLY: We are pleased that Dr. Young and colleagues found the results of our analyses of data on Medicare patients to be largely generalizable to an all-payer population. Their analyses are timely, since roughly half of revascularization procedures are performed in non-Medicare patients.

We generally agree with the issues raised in Dr. Ballard's letter, but we would note that it is not customary to focus on the lower bounds of a 95 percent confidence interval when interpreting study results. A more balanced interpretation of our results is that the odds of death for patients at specialty hospitals was 11 percent lower for PCI and 16 percent lower for CABG after adjustment for patient characteristics. We agree that it is not unreasonable to argue that, because specialization is closely tied to procedural volume, comparisons of specialty and general hospitals should be based on risk-adjusted (but not volume-adjusted) mortality rates. However, given that some have argued that there is an inherent advantage in specialization above and beyond the "volume effect," we felt it was important to control for volume in our analyses in an attempt to tease apart these differences.

We believe that our analyses indicate that the lower unadjusted mortality at specialty hospitals is largely attributable to the lower average risk of the patients such hospitals treat and to the hospitals' greater procedural volumes. Perhaps most informative were the results of stratified analyses that directly compared lower-volume and higher-volume specialty and general hospitals and did not show significant differences. Finally, we agree with Dr. Ballard that other indicators of quality, such as process-of-care measures, which are difficult to derive from claims data, should ideally be examined.

We also agree with Dr. Edwards and colleagues that analyses based on administrative data are subject to a number of potential methodologic limi-

tations when the quality of care is assessed. However, the lack of registries with detailed clinical information that can be used to track patient outcomes on a national level makes it impossible to conduct comparative analyses of specialty and general hospitals, since the validity of such analyses is predicated on the availability of data from all hospitals in the markets studied. We believe that, in the absence of national clinical registries, use of available administrative databases to provide empirical insight into important questions relevant to policy is preferable to not addressing these questions; however, the limitations of such analyses should be clearly recognized.

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TO THE EDITOR: Autoimmune thyroiditis should be added to the group of disorders associated with the polycystic ovary syndrome. Sex steroid hormones appear to be the most important environmental factors contributing to the development of autoimmune thyroiditis.[1] Furthermore, a recent prospective study demonstrated a high prevalence of autoimmune thyroiditis among women with the polycystic ovary syndrome.[2] Since hypothyroidism has been shown to make the polycystic ovary syndrome worse,[3] physicians should carefully check thyroid function and thyroid-specific autoantibodies in such patients.

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TO THE EDITOR: We were concerned about Dr. Ehrmann's statement that "there is an increased prevalence of endometrial hyperplasia and carcinoma in women with the polycystic ovary syndrome." The common occurrence of risk factors for endo-

metrial carcinoma, obesity, and type 2 diabetes in patients with the polycystic ovary syndrome does not necessarily suggest that the syndrome is associated with an increased prevalence of endometrial carcinoma. In fact, the recent review of the relevant literature by Hardiman et al.,[1] which was mentioned by Dr. Ehrmann to support his statement, did not confirm such an association, and the authors concluded that the evidence of an increased risk of endometrial carcinoma in the polycystic ovary syndrome was incomplete and contradictory.[1]

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1. Hardiman P, Pillay OS, Atiomo W. Polycystic ovary syndrome and endometrial carcinoma. Lancet 2003;361:1810-2. [Erratum, Lancet 2003;362:1082.]

DR. EHRMANN REPLIES: Dr. Braillon rightly calls attention to the increased prevalence of nonalcoholic steatohepatitis among women with the polycystic ovary syndrome[1] and the importance of periodic assessment of hepatic function in women with this syndrome who are taking a thiazolidinedione. However, recent evidence suggests that the thiazolidinediones may attenuate, rather than exacerbate, the biochemical and histologic abnormalities of nonalcoholic steatohepatitis associated with insulin resistance.[2] Thus, although the thiazolidinediones are not formally recommended for the treatment of nonalcoholic steatohepatitis associated with the polycystic ovary syndrome, neither are they necessarily contraindicated when nonalcoholic steatohepatitis is present. Prospective studies are needed to clarify the short-term and long-term effects of the thiazolidinediones when nonalcoholic steatohepatitis is present in women with the polycystic ovary syndrome.

Drs. Mascitelli and Pezzetta's assertion that thyroid dysfunction (i.e., autoimmune thyroid disease) and the polycystic ovary syndrome are causally linked must be viewed with caution. Although sex steroids (androgens, estrogens, and progestins) may modulate the immune response in humans,[3] these effects are complex and often conflicting. In addition, because both autoimmune thyroid disease and the polycystic ovary syndrome occur commonly in women, a statistical association between these conditions may be evident, but this does not imply causality. Although it is reasonable to recommend the assessment of thyroid function in women with the polycystic ovary syndrome, there is little evidence of a pathogenetic link between these two disorders.

Finally, Drs. Mikhail and Cope are correct in remarking that on the basis of the review by Hardiman et al.,[4] data on the risk of endometrial carcinoma among women with the polycystic ovary syndrome are incomplete and contradictory. Yet it must be acknowledged that Hardiman et al. go on to conclude that in the absence of data to confirm the contrary, "there is little choice other than to advise oligomenorrheic women with PCOS [polycystic ovary syndrome] that they may be at increased risk of developing endometrial cancer." I suspect that all would be in agreement with this recommendation.

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Hypothyroidism Due to Ethionamide

TO THE EDITOR: Ethionamide (Trecator-SC, Wyeth-Ayerst) is an antimycobacterial drug used as a second-line agent in the treatment of multidrug-resistant tuberculosis. It is structurally similar to methimazole, has been shown to inhibit thyroid hormone synthesis, and was last reported to cause hypothyroidism in 1984.[1] Before that report, only three other cases of hypothyroidism related to ethionamide were documented.[1] We now report the case of a 57-year-old Indian-American woman with Takayasu's arteritis, coronary artery disease, and multidrug-resistant tuberculosis in whom hypothyroidism developed while she was receiving 250 mg of ethionamide twice daily for three months. She

had previously been euthyroid (thyrotropin level, 2.1 μU per milliliter; normal range, 0.35 to 5.5), but after treatment (Fig. 1) her thyrotropin level increased to 19.4 μU per milliliter.

Ethionamide was discontinued for other reasons, and her serum thyrotropin level decreased to 5.4 μU per milliliter. Ethionamide was restarted six months later because of suspected recurrent multidrug-resistant tuberculosis. Within four months of restarting ethionamide, the patient had symptoms of severe hypothyroidism. An echocardiogram revealed a reduced ejection fraction (45 percent, a decrease from 60 percent six months earlier) and a new 1.5-cm pericardial effusion. Clinical evaluation revealed a small goiter, a thyrotropin level of 136 μU per milliliter, a thyroxine level of 1.6 μg per deciliter (normal range, 4.5 to 10.9), and an elevated thyroglobulin level (232 ng per milliliter; normal range, 4 to 40). A test for thyroid peroxidase antibodies was negative. The patient was treated with 88 μg of levothyroxine daily, which was reduced to 50 μg per day after ethionamide was discontinued once the course of therapy was completed. Four weeks later, the thyrotropin level was 1.2 μU per milliliter. The patient reported feeling well, and a repeated echocardiogram showed a normal ejection fraction and a marked decrease in the size of the pericardial effusion.

Although ethionamide-induced hypothyroidism is noted as a possible adverse effect in the package insert,[2] as well as in summary statements about treatment of multidrug-resistant tuberculosis,[3] it appears that this complication is not widely known among practicing physicians. Ethionamide is not generally included in references that list medications that may cause hypothyroidism.[4] Hypothyroidism due to ethionamide is, at least in part, reversible after withdrawal of the drug. There is emerging evidence from a World Health Organization global project that although tuberculosis strains that are sensitive to standard antimycobacterial drugs remain the most prevalent, there are epidemics and several "hot spots" of multidrug-resistant tuberculosis around the world, for which second-line agents, such as ethionamide, are commonly used.[3,5] Thus, physicians treating patients who have multidrug-resistant tuberculosis should be aware of this potential complication of ethionamide therapy. Thyroid function should be carefully monitored in all patients treated with ethionamide,

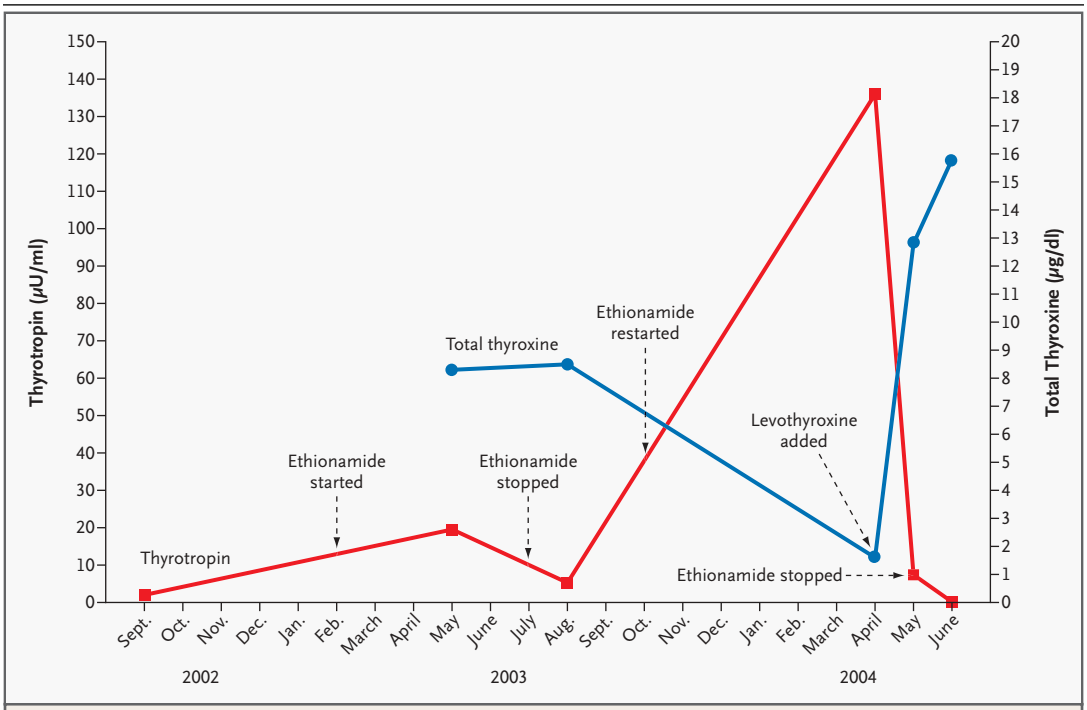

Figure 1. Thyroid Function in the Patient over Time.

and ethionamide should be included in any list of drugs that may cause hypothyroidism.

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Gestational Exposure to Lovastatin Followed by Cardiac Malformation Misclassified as Holoprosencephaly

TO THE EDITOR: In 2004, we reported central nervous system and limb anomalies that followed exposure to statin drugs in the first trimester of pregnancy (April 8, 2004, issue).[1] One case, in which there had been exposure to lovastatin, was described as involving holoprosencephaly on the basis of three separate reports of a "cerebral/brain ventricular septal defect," with accompanying cardiac malformations that had been submitted to the Food and Drug Administration adverse-event database. We recently learned that the manufacturer considered the structural anomalies to be solely cardiac, and therefore we requested source documentation to clarify the conflicting reports. A detailed clinical report that was located among archival documents clearly described an atrial septal defect, a ventricular septal defect, and aortic hypoplasia leading to cardiac failure, with secondary central nervous system dysfunction. It was apparent that a data-extraction error had occurred, incorrectly categorizing the ventricular septal defect as an intracranial anomaly. Correcting this misclassification reduces our reported number of lovastatin-exposed fetuses with midline central nervous system anomalies from three to two. We still believe that the preponderance of the evidence supports the hypothesis that early gestational exposure to statin drugs may be teratogenic and that prospective studies should be initiated.

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BOOK REVIEWS

DNA AND THE CRIMINAL JUSTICE SYSTEM: THE TECHNOLOGY OF JUSTICE

(Basic Bioethics.) Edited by David Lazer. 414 pp. Cambridge, Mass., MIT Press, 2004. $67 (cloth); $27 (paper). ISBN 0-262-12265-0 (cloth); 0-262-62186-X (paper).

Over the past decade, DNA technology has become fixed in the popular consciousness as a reliable tool for the identification of the guilty and the exoneration of the innocent. Arguably, the public's expectations regarding DNA testing are such that a failure to use it in any major criminal prosecution would have to be explained to the jury. This book takes the popular acceptance of forensic DNA identification as fact and focuses on some of the policy issues that have emerged as the technology has come to prominence. The book consists of a collection of essays representing diverse political and philosophical positions on these issues; its objectives are to identify areas of consensus and to clarify points of contention. At this level, the book succeeds; the essays are written by top-flight authorities, and the policy positions are well articulated.

The principal focus of the book is on policies regarding forensic DNA databases. There are a number of important questions here: Who should be included in the databases? What is appropriate use? Who should have access? Some parts of these questions mirror questions raised with regard to medical and genetic databases, but there are also important issues specific to forensic databases — notably, the control of such databases by law enforcement (a suspect governmental entity to some; a protector of rights to others), the constitutional questions involving Fourth Amendment protections and privacy rights, and the concern that database information may be used inappropriately to study the genetics of criminal behavior (whatever that may be) or for other purposes unrelated to forensic identification per se.

If there is fault to be found with the book, it is in the lack of empirical data with regard to what has been learned from the use of forensic databases during the past decade. The effort to balance discussions of individual privacy and the public good would have benefited from the inclusion of hard data about what is actually working and what is not. In fairness to the editor, there exist no studies on this subject, but it would have been helpful to identify the deficiency.

If I have a general quibble about the book, it is that the emphasis on database issues eclipses the discussion of other significant policy concerns. A notable example is the spare attention given to one of the most striking consequences of forensic DNA testing — the exoneration of a large number of wrongfully convicted persons. That these exonerations have brought to light systematic flaws in the way the legal system works is well described, but there is no elaboration of policy options or their implications. This is unfortunate, for restoring trust in the justice of our legal system should be a high priority.

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THE FUTURE OF THE BRAIN: THE PROMISE AND PERILS OF TOMORROW'S NEUROSCIENCE

By Steven Rose. 344 pp., illustrated. New York, Oxford University Press, 2005. $28. ISBN 0-19-515420-7.

In 1990, president George H.W. Bush inaugurated "The Decade of the Brain." In *The Future of the Brain*, Steven Rose suggests that the first 10 years of the new millennium be designated "The Decade of the Mind." This juxtaposition of brain and mind crystallizes the book's message.

Rose is a neuroscientist who studies the molecular mechanisms of memory in animals. He is also a consummate essayist who has written extensively for the general public, conveying complex science in simple terms and at the same time questioning how well or how poorly science fares when assaulting fundamental questions about life. In his book *Lifelines: Life beyond the Gene* (Oxford, England: Oxford University Press, 2003), Rose attacks the radical re-

ductionism of molecular biologists who argue that everything we do is dictated by our genes. He now applies a similar line of reasoning to challenge the neuroscientific reductionists who believe that all feelings and thoughts can be explained by molecular neuroscience. Rose has been aptly dubbed the "conscience" of neuroscience.

What most bothers Rose is encapsulated in the first sentence of the first chapter: "'Better Brains' shouted the front cover of a special edition of *Scientific American* in 2003." Rose is distressed by the tendency of scientists and their apostles in the media to make too many promises to the public. He is particularly vexed by pharmaceutical solutions to society's ills, such as the quest of large pharmaceutical concerns to create a "smart pill," as well as stimulants that many of us feel are overused in the treatment of hyperactive children.

In contrast to many authors who espouse a thesis and then bludgeon the reader with 500 pages on what could have been circumscribed in a single paragraph, Rose develops his thesis gently and elegantly, laying his groundwork in a series of chapters that summarize what we know about the brain. He almost literally begins from the beginning, explicating the genetic code and then reviewing the evolution of the nervous system from that of the most primitive organism to that of the human. He proceeds with a detailed analysis of the development of the brain, showing how ontogeny recapitulates phylogeny. He reviews the ways in which the adult brain regulates perception, conceptualization, and emotions through diverse neurotransmitter systems. He describes what is known about the aging brain. Chapters are devoted to mental illness as well as to the major drugs used to treat real or imagined disabilities.

In contrast to many books that "dumb down" science for the lay public, Rose displays respect for the intelligence of his audience. Although the book is accessible to the educated nonscientist reader, Rose's discussions of science with regard to the brain are sufficiently sophisticated that the professional researcher has much to learn. A unique strength of Rose's presentation is his emphasis on the historical background of present-day knowledge. Of equal importance is his critical analysis of the epistemological aspects of neuroscience. For instance, in the chapter dealing with mental illness, he relates the classic study of David Rosenhan, "On Being Sane in Insane Places" (published in *Science* in 1973), in which Rosenhan and a team of volunteers sought admission to psychiatric hospitals by claiming that they heard voices. Once inside the hospital, they behaved normally and maintained that the voices had ceased. The physicians caring for them regarded their protestations of normality as evidence of abnormality and were reluctant to release them. To add insult to injury, Rosenhan subsequently announced that another group of "pseudo-patients" would be presented to psychiatric hospitals in the vicinity, whereupon there was an epidemic of diagnoses of pseudo-patients — who never existed.

Rose is highly critical of the excesses of psychopharmacology, perhaps too much so. He devotes substantial space to the use of stimulant drugs such as Ritalin (methylphenidate) to treat attention deficit–hyperactivity disorder. He properly questions how one can diagnose as a "disease" a condition characterized by a child's being more active or paying less attention than "the majority" of children in the classroom. He is dubious about the reality of a disorder that is diagnosed in England at 1/10 the rate in the United States. It is easy to ridicule the field of psychiatry when a diagnosis is made on the basis of aberrant behavior, rather than something concrete such as a bacterial infection. It is also true that psychiatric diagnoses are influenced by fashion, especially that of available new medications. Thus, when lithium was introduced for the treatment of mania in the early 1960s, the diagnosis of this condition increased severalfold within two years. Nonetheless, the most critical psychiatric researchers would agree that bipolar disorder is an illness and that there is genuine reality underlying at least a substantial number of cases of attention deficit–hyperactivity disorder.

Rose is at his best when, by integrating a vast body of knowledge, he illuminates the limitations in our use of existing neuroscience to control, predict, and excuse behavior. For instance, he reviews studies showing that abnormal variations of the gene for the enzyme monoamine oxidase, which degrades neurotransmitters such as serotonin and dopamine, predispose persons to violent behavior. Will possession of such a gene be used in court to excuse murder? In discussing the role of determinism as a link between brain and behavior, Rose quotes Francis Crick, who maintains that we are "nothing but a bunch of neurons," whereas Rose would argue that our free will and individual responsibility are independent of the molecular details of genes and neurons.

In summary, Rose has provided a powerful con-

tribution to the literature. His book will be of interest to both lay and professional readers, to scientists, philosophers, and anyone else who is curious about the workings of the brain and the promise and perils that brain research holds for the future.

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TO DO NO HARM: ENSURING PATIENT SAFETY IN HEALTH CARE ORGANIZATIONS

By Julianne M. Morath and Joanne E. Turnbull. 354 pp. San Francisco, Jossey-Bass, 2005. $42. ISBN 0-7879-6770-X.

Manifestos are typically the declarations of political and social revolutions. They are almost unheard of in health care. But in *To Do No Harm*, Julianne Morath and Joanne Turnbull boldly apply that label to the patient-safety movement. The book conveys the passionate desire for change that one would expect from a manifesto. This passion, coupled with the book's comprehensiveness, will lead most readers with any clinical background or interest to find it worth reading.

The book synthesizes existing information and concludes (as other books have) that the culture of health care is broken and must change. The proposed solution is for leaders to educate themselves about safety, adopt new ideas, make changes, and thereby change the culture and improve safety. Thus, persons responsible for improving patient safety in hospitals and other health care settings are the primary audience. These readers will find the book remarkably comprehensive. A unique contribution of the book is that it brings together safety information and practices that have been rapidly adapted from other industries and disciplines since the publication of *To Err Is Human*, by the Institute of Medicine (Washington, D.C.: National Academies Press), in 2000. This is an important service, because many of these ideas come from nonmedical disciplines and may be less accessible to persons within the field of health care. The book is practical and easy to read, and it will motivate leaders to make changes.

The first three chapters serve as a basic introduction to the research, concepts, and terms involved in patient safety. The remaining chapters keep to the main theme of empowering leaders to change the culture. These chapters discuss the importation of skills and knowledge from industries and organizations with high levels of reliability (such as nuclear power, commercial aviation, and naval aircraft carriers); error-reporting systems; accountability (including disclosures to injured patients and the responsibilities of leaders); external influences on safety (e.g., regulators, licensure boards, and the malpractice system); and strategies to accelerate change. The organization of the book along these lines means that many individual topics (such as reporting and the assessment of the safety of the culture) are repeated, but this is useful given the importance of these topics. Each chapter has one or more sections labeled "Concept to Action" that are illustrative case studies of actions taken in the attempt to improve safety. There is an informative glossary, a list of resources, references to additional reading, and appendixes that contain tools, policies, and other information. The book rarely deviates from addressing its primary audience of health care leaders. However, health care providers without leadership roles and even students and other trainees will find it a good introduction to the subject of patient safety.

Although the authors' choice to view patient safety as a "movement" with a "manifesto" adds energy, urgency, and focus to the book, it also results in predictable limitations (and predictable criticisms from a researcher such as myself). For example, the authors state, "Harm-free care is our goal. Our response is a matter of attitude, belief, and will." But we also need research to help us understand the strengths, weaknesses, and cost-effectiveness of the proposed safety interventions. The authors uncritically accept the premise that practices adopted from other industries will work in health care. I suspect that many readers will be taken aback by the confidence with which many new interventions are presented. In addition to "attitude, belief, and will," leaders need data to help guide decisions about which interventions are worthwhile. Should a hospital invest time and effort in near-miss reporting systems, or should it devote that time and effort to ensuring that patients receive therapies proven to save lives, such as aspirin and beta-blockers after myocardial infarction? Of course, this is not purely an either–or situation, as most readers will come to understand. The book does a good job of describing the close relationships that exist among the areas of safety, quality, and risk management. To some degree, the lack of skepticism in this book with regard to safety interventions reflects the young

age of the field. Critical assessments of patient-safety practices are just now beginning to appear.

Another limitation is the focus on the practices of high-reliability organizations. Given the focus on leadership and management, it is surprising that the authors do not rely more heavily on the disciplines of business management, organizational psychology, and sociology.

Nevertheless, *To Do No Harm* can help to equip leaders with a broad array of knowledge and tools with which to improve patient safety. The book admirably synthesizes the current practices and presents them in an engaging and passionate manner. It remains to be seen whether these practices will in fact help us to do no harm.

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NOTICES

Notices submitted for publication should contain a mailing address and telephone number of a contact person or department. We regret that we are unable to publish all notices received. Notices also appear on the Journal*'s Web site (www.nejm.org/meetings). The listings can be viewed in their entirety or searched by location, month, or key word.*

UNIVERSITY OF MICHIGAN MEDICAL SCHOOL

A course entitled "Cancer Update 2005: Highlighting Recent Advances in the Prevention, Diagnosis, and Treatment of Breast Cancer, Melanoma, Sarcoma, and Prostate Cancer" will be offered in Bay Harbor, Mich., July 8–10.

Contact Registrar, Department of Medical Education, University of Michigan Medical School, P.O. Box 1157, Ann Arbor, MI 48106-1157; or call (800) 800-0666 (national) or (734) 763-1400 (Michigan); or fax (734) 936-1641.

MAYO SCHOOL OF CME

The following courses will be offered in Rochester, Minn., unless otherwise indicated: "Internal Medicine Review" (July 6–12); "Shoulder Arthroscopy" (July 9); "Mayo Clinic Psychiatric Genomics: Applications for Clinical Practice" (Aug. 1–5); "Selected Topics in Rheumatology" (Victoria, B.C., Canada, Aug. 11–14); "Mayo Clinic Pediatric Days" (Chicago, Sept. 8 and 9); "Mayo Clinic Nutrition in Health and Disease" (Chicago, Sept. 8 and 9); "Mayo Clinic Gastroenterology and Hepatology Board Review" (Chicago, Sept. 8–11); "Mayo Clinic Update in Hepatology and Liver Transplantation" (Minneapolis, Sept. 16 and 17); "10th Annual Mayo Cardiovascular Review Course: Cardiology Boards and Recertification" (Sept. 24–29); "Subspecialty Board Review: Interventional Cardiology Board Review" (Sept. 30 and Oct. 1); "Subspecialty Board Review: Electrophysiology Boards and Recertification" (Sept. 30–Oct. 2). "Geriatric Update for the Primary Care Provider" (Oct. 6); "Genomics in Clinical Practice" (Oct. 20 and 21); and "Current Concepts in Primary Eye Care" (Nov. 10).

Contact Mayo School of CME, 200 First St. SW, Rochester, MN 55905; or call (800) 323-2688 (national) or (507) 284-2509 (Minnesota); or fax (507) 284-0532; or see http://www.mayo.edu/cme; or e-mail cme@mayo.edu.

THE AMERICAN SOCIETY FOR CELL BIOLOGY

The "45th Annual Meeting" will be held in San Francisco, Dec. 10–14. Deadline for submission of abstracts is July 28.

Contact The American Society for Cell Biology, 8120 Woodmont Ave., Suite 750, Bethesda, MD 20814-2762; or call (301) 347-9300; or fax (301) 347-9310; or e-mail ascbinfo@ascb.org; or see http://www.ascb.org.

SOCIETY OF COMPUTED BODY TOMOGRAPHY AND MAGNETIC RESONANCE

The "Fifteenth Summer Practicum" will be held in Napa, Calif., Aug. 7–11.

Contact SCBT/MR, c/o Matrix Meetings, P.O. Box 1026, Rochester, MN 55903-1026; or call (507) 288-5620.

CALL FOR APPLICATIONS

The Mount Sinai School of Medicine is accepting applications for two integrated fellowships: "Hematology-Oncology/Palliative Medicine" and "Geriatrics/Palliative Medicine," which will provide intensive one-on-one mentoring and rigorous clinical, educational, and research training. Applications are being accepted on an ongoing basis for academic years 2006–2008.

Contact Hertzberg Palliative Care Institute, Brookdale Department of Geriatrics, Mount Sinai School of Medicine, 1 Gustave Levy Pl., Box 1070, New York, NY 10029-6574; or see http://www.mssm.edu/palliative/fellowship; or e-mail palliativecare@mssm.edu; or call (212) 241-1446.

VA—NATIONAL MEDICAL MUSICAL GROUP

The group is recruiting new members for its symphony orchestra and chorus. The MMG holds concerts around the country and overseas, including annual Flag Day/Independence Day and Veterans' Day concerts. Physicians, dentists, nurses, other healthcare personnel, faculty and students, both VA and non-VA, and their families and friends may apply.

Contact VA–National Medical Musical Group, 1700 17th St., NW, Suite 508, Washington, DC 20009; or call (202) 797-0700; or e-mail vanmmg@hotmail.com; or see http://www.medicalmusical.com.

UNIVERSITY OF TORONTO

The following courses will be offered in Toronto: "Urology Update 2005" (Oct. 21 and 22) and "International Congress on Glaucoma Surgery" (May 25–28, 2006).

Contact Continuing Education, Faculty of Medicine, University of Toronto, 500 University Ave., Suite 650, Toronto, ON M5G 1V7, Canada; or call (888) 512-8173 or (416) 978-2719; or fax (416) 971-2200; or see http://www.cme.utoronto.ca; or e-mail ce.med@utoronto.ca.

AMERICAN HEADACHE SOCIETY

The following meeting will be held: "The Scottsdale Headache Symposium" (Scottsdale, Ariz., Nov. 18–20).

Contact American Headache Society, 19 Mantua Rd., Mount Royal, NJ 08061; or call (856) 423-0043; or fax (856) 423-0082; or e-mail ahshq@talley.com; or see http://www.ahsnet.org.

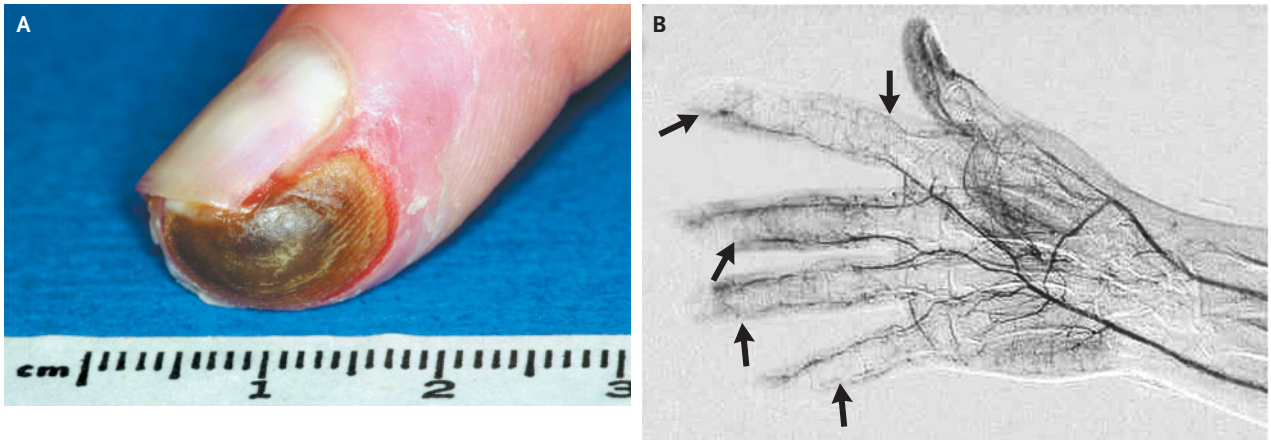
LUCILE PACKARD CHILDREN'S HOSPITAL

The following courses will be offered: "Practical Childhood Obesity Management: A Guide for the Practitioner" (Stanford, Calif., July 7); "13th Annual Pediatric Update" (Stanford, Calif., July 8 and 9); and "Palliative and End of Life Care for the Adult and Child" (Kauai, Hawaii, Nov. 7–9). The courses are jointly sponsored by Lucile Packard Children's Hospital at Stanford and Stanford University School of Medicine.

Contact Lucile Packard Children's Hospital, CME Programs, 725 Welch Rd., MC 5517, Palo Alto, CA 94304; or call (650) 497-8554; or fax (650) 497-8585; or see http://www.cme.lpch.org.

IMAGES IN CLINICAL MEDICINE

Calcific Arteriopathy



France Joyal, M.D.
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A 38-year-old man who had undergone renal and pancreatic transplantation five years earlier was referred because of acral gangrene of both hands. The patient also had type 1 diabetes and was a nonsmoker. Within the previous month, painful paraungual blue spots had developed on three of the patient's fingers, including the fourth digit of his left hand (Panel A); ulcers had then developed on the affected fingers. His renal function had deteriorated, a fact attributed to the effects of infection with polyomavirus type BK. The systemic blood pressure was 130/80 mm Hg, and the segmental arterial pressures in the upper limbs were more than 300 mm Hg, with normal pulse signals. Arteriography revealed diffuse arterial occlusions (Panel B, arrows). The serum calcium level was 2.52 mmol per liter, the phosphorus level was 1.04 mmol per liter, and the creatinine level was 2 mg per deciliter (177 μmol per liter). The patient was treated with calcium antagonists, antiplatelet therapy, and heparin. However, within six months, ischemia had progressed in all digits except for the thumbs, and he underwent partial amputation of the second, third, and fourth fingers of both hands. Microscopical examination of an amputated digit showed ischemic necrosis with inflammatory infiltration, medial calcifications of medium-sized arteries, and intimal hyperplasia with luminal thrombosis. Despite the amputations, the patient continues to have ischemic pain, and only the thumbs have adequate perfusion.