Inside this issue:

The Search for Mutagens from Cooked Foods
Food Mutagens

Potent mutagens are found in cooked foods, such as fried beef. This report, the first of two installments, describes our efforts to identify and measure the abundance of more than a dozen potential cancer-causing byproducts in a variety of foods characteristic of the Western diet.
Welcome to Science and Technology Review

WELCOME to the first issue of Science and Technology Review (S&TR), Lawrence Livermore National Laboratory’s monthly magazine. Part of our responsibility as a national laboratory is to inform the country about our work and accomplishments as we address science and technology problems of national importance. As many of our readers are aware, we published S&TR’s predecessor Energy and Technology Review (E&TR) for two decades. Over the years, E&TR evolved as we made changes in format, design, and style to enhance our ability to explain the Laboratory’s work to an audience with, frequently, a more than basic scientific background. S&TR is a natural outgrowth of this evolutionary process. Upon the 20th anniversary of E&TR, we thought it fitting to examine the publication once again to see what could be done to make it still more effective. We confirmed that the journal reflected the wide variety of science and technology being explored at the Laboratory. As part of this review, we prepared a mission statement that reflects the essence of the new publication’s goal:

The purpose of S&TR is to communicate, to a broad audience, the Laboratory’s scientific and technological accomplishments, particularly in the core mission areas of the Laboratory. The publication does more than just inform people about these accomplishments: it helps readers understand them and appreciate their value to the individual citizen, the nation, and the world.

We also recognized that, like everyone else these days, our readership is extremely busy and often does not have the time to read lengthy articles. Feature articles describing the Laboratory’s major accomplishments, projects, and programs will continue to be the mainstay of the journal. We will endeavor, however, to always provide these descriptions in language understandable by educated, interested nonexperts. Feature articles will be supplemented with briefs and research highlights describing, in a few paragraphs or pages, progress in previously reported projects, recent achievements in ongoing programs, and the start of exciting new research efforts. Also included will be a list of recent patents awarded and licensing agreements signed, which will illustrate the scale and scope of the Laboratory’s work. In addition, a new commentary column will provide a forum for members of the Laboratory’s top technical management to give their views on important institutional and programmatic issues and on external events affecting the Laboratory’s current status and future direction.

Finally, we changed the name of our publication to Science and Technology Review to better reflect the Laboratory’s mission of applying a broad range of science and technology in the national interest. In addition to the changes in the journal, we have evaluated its availability and concluded that we could significantly improve the dissemination of technical information about Laboratory programs. During the next several months, we will be adding interested readers to the distribution list. We are also taking advantage of modern information technology by making S&TR available electronically on the Internet via the Laboratory’s home page at http://www.llnl.gov or directly at http://www.llnl.gov/energyreview.html. Electronic access will lead to a wider distribution for the journal and to increased technical interactions with researchers elsewhere. As the Laboratory’s electronic library grows, we will include hypertext links to technical references in S&TR articles. In this way, we will be able to tailor our articles to a nonexpert audience while providing those readers who desire more specifics with easy access to the detailed technical information.

To help us assess whether we are reaching the goals set for S&TR, we encourage you to complete and return the survey printed on the inside back cover of the magazine. This survey will be a regular part of the journal and to increased technical interactions with researchers elsewhere. As the Laboratory’s electronic library grows, we will include hypertext links to technical references in S&TR articles. In this way, we will be able to tailor our articles to a nonexpert audience while providing those readers who desire more specifics with easy access to the detailed technical information.

William Bookless, Scientific Editor

Science and Technology Review

Communicating the Worth of Our Work

THIS past year and a half has been one of the most unsettled periods in the history of the Livermore Laboratory. Our laboratory, like all federal laboratories, has felt the winds of change from Washington. Various committees are questioning the way in which the federal government supports scientific research and the appropriateness of certain programs. Indeed, such questioning is inevitable and necessary in light of trillion-dollar budget deficits, continuing economic uncertainties, and widespread concern about health care, social security, crime and violence, education, and other basic survival issues.

At the same time, many of the problems facing the nation and the world today involve science and technology. For example:

- Ensuring national security, not only by maintaining the U.S. nuclear deterrent but also by stemming the proliferation of weapons of mass destruction.
- Understanding, remediating, and preventing damaging effects of human activities on the environment.
- Solving the mystery of the genetic code.
- Developing advanced technologies, processes, and products (particularly those related to energy production, biotechnology, and electronics) that enhance the quality of life while securing the nation’s preeminence in the global marketplace.
- Improving the quality and reducing the cost of health care.

We believe that national laboratories like Livermore are as important now as they have ever been. Our overriding mission of serving the nation through the application of science and technology remains unchanged. However, as the science and technology required to solve important national problems grows more complex, we must make sure that we explain our work—and the value of that work—in ways that are accessible and meaningful to a broad audience. And we must remember that the value of scientific research (particularly publically funded research) lies, to a large extent, in its ability to solve real-world problems.

No longer is sufficient for scientists to communicate only with other scientists through professional journals or at technical conferences. Neither is it sufficient for laboratories to communicate primarily with each other and with their funding agencies. We must also reach the large numbers of interested, educated nonexperts—government representatives and congressional staffers, community leaders, and the general public—all of whom through their taxes contribute to the Laboratory’s funding and therefore have a vested interest in the Laboratory’s work.

Science and Technology Review is one of the principal mechanisms by which we inform and educate a broad readership about our research programs and accomplishments. Much of the Laboratory’s research is at the cutting edge of science and technology, making it particularly challenging to describe state-of-the-art accomplishments and their significance in widely understood terms. Our goal is that the articles presented here represent the full range of projects at Livermore and convey the challenge and excitement of working at the frontiers of science and technology.
Cutting system assists in Russian nuclear cleanup

The Laboratory is continuing its efforts to assist in the dismantlement of nuclear weapons in the former Soviet Union. In early April, Lab engineers packed a 6.4-m (21-ft) van with a portable “water knife” cutting system designed to permit safe access to warheads and other “hot” materials. The system will be used for standby response in the event of accidents involving the transport of nuclear materials.

Although water knives are not new, adapting a “portable” system is a unique application. The pressure generator alone weighs 771 kg (1700 lb) while the control panels needed for remote use weigh 136 kg (300 lb). To help move the pressure generator, the van has been fitted with a winch; many components have been adapted with lifts that allow assembly by just one person.

The van containing the cutting system was sent to a base near Moscow. Another van containing a second cutting system was shipped from Livermore in early June. In addition, a Laboratory team was dispatched to Russia to provide training in the use of the emergency cutting systems. Assembly of the vans and cutters has been the work of the Disablement Technology Group in the Laboratory’s Nonproliferation, Arms Control, and International Security directorate. The work is part of Department of Energy’s efforts in the Safe Secure Dismantlement Program, an international agreement designed to provide assistance and equipment to Russia, Ukraine, Belarus, and Kazakhstan.

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Study suggests aging, genetic damage connection

A study by Lawrence Livermore scientists has found that older people have more DNA damage than younger individuals. DNA is the molecule that carries the body’s genetic code. On March 14, James Tucker, senior biomedical scientist, reported the results of the three-year study at the Environmental Mutagen Society’s annual meeting in St. Louis, Missouri.

Tucker’s research team studied chromosomes in blood samples from a group of individuals ranging in age from 20 to 80 as well as in blood taken from the umbilical cords of newborn infants. The scientists found that genetic damage increased with age and shot up dramatically after age 50. “We expected to observe more genetic damage in older people, and this seems to indicate that damage can accumulate through normal living,” said Tucker.

The most common type of damage observed was translocations, where chromosomes break and recombine with other chromosomes. Translocations were found more than 10 times as often in people over the age of 50 than in newborns. Tucker said the results will benefit researchers studying the effects of radiation on people by providing a baseline against which genetic damage in exposed individuals can be measured.

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Radar licensee, AlliedSignal, to develop auto system

Amerigon Inc. of Monrovia, California, has announced an agreement under which AlliedSignal Inc., one of the auto industry’s largest electronic suppliers, will join in the development of an auto radar system based on LLNL technology.

Amerigon Inc. holds the license from the Lab to use the radar advance, called MicroPower Impulse Radar, for automotive safety applications. Amerigon plans possible applications of the radar as a device to signal when vehicles are in a driver’s blind spot, as a backup warning system, as a parking aid, and for triggering side-impact air bags.

The new technology was invented by Livermore engineer Tom McEwan in connection with his work for the Lab’s Nova laser. The radar has been used as a part of a system to measure the balance and power output of the laser’s 10 beams and as the heart of the diagnostics that measure neutrons from the fusion reactions. For $10 to $15 in off-the-shelf components, the MicroPower Impulse Radar can do the same tasks as equipment costing $40,000. Since the new radar technology was announced in March 1994, the Laboratory has received over 2000 calls from businesses and individuals in at least 15 nations.

Contact: Tom McEwan (510) 422-1621 (mcEwan@llnl.gov).

Goal is to spot small, covert nuclear tests

Laboratory researchers are attempting to develop an improved method for identifying covert nuclear tests. Current identification techniques are efficient at identifying detonations of 150 kilotons or greater, the yield limit imposed by the Threshold Test Ban Treaty. They are not able, though, to differentiate effectively between small evasive nuclear tests and other seismic events, such as earthquakes or mining activity. Conceivably, proliferant nations might test with smaller nuclear devices to take advantage of the fact that small explosions are more difficult to identify.

One promising identification method being explored by Lab researchers compares two kinds of seismic waves—called P and S waves—generated by both earthquakes and nuclear explosions and looks for differences in the size of their spectral ratios at many frequencies. A spectral ratio is the ratio of the spectral amplitudes of two signals such as P and S wave.

A weakness of the approach is that it probably would not be able to discriminate between large-scale concentrated chemical explosions and small nuclear detonations, according to Peter Goldstein, the principal investigator working on the detection system for the Lab’s Nonproliferation, Arms Control, and International Security directorate. However, efforts are under way to develop techniques for identifying such explosions.

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Lab and MIT operate tokamak via Internet

Laboratory scientists teamed up with colleagues from the Massachusetts Institute of Technology recently to demonstrate the first transcontinental operation of a fusion experiment via the Internet. Using a Department of Energy subnet of the global Internet known as the Energy Sciences Network, managed from Livermore, MIT scientists successfully operated the Alcator C-Mod reactor in Cambridge over ESnet. On the first full day of remote experiments, 21 of 35 shots were controlled from Livermore.

Although aimed at proving the technical feasibility of running a tokamak remotely, the LLNL–MIT Internet collaboration also allowed scientists to learn more about managing the efforts of research groups working at the same time in different locations on joint equipment.

“This demonstration was the definitive test of controlling a large, complex physics experiment from a remote location,” says physicist Tom Casper, who leads the Laboratory’s effort in the collaboration. “This technology is important not only to fusion, but also to other scientific applications—such as medical systems or environmental work—where real-time access and control are needed from various locations around the country.”

Contact: Tom Casper (510) 422-0978 (casper1@llnl.gov).

Researchers explore MACHOs in dark matter

An international team of scientists reports that Massive Compact Halo Objects, or MACHOs, constitute less dark matter in the Milky Way than was previously thought possible. One conclusion is that some other unknown type of object makes up the dark matter. On the other hand, the findings may mean that the current model for the halo of dark matter in our galaxy is inaccurate. The findings are reported in the April 10 issue of Physical Review Letters.

In their search for MACHOs, the team observed three microlensing events instead of the 15 that were expected if MACHOs made up all of the ubiquitous dark matter. Microlensing is the brightening of a star that indicates the passage of a large object in front of it, thereby magnifying, as if through a lens, the light passing around the object. The September 20, 1993, issue of Nature called this group’s first observation of microlensing, which garnered worldwide attention, the “footprint of dark matter.”

The MACHO collaboration, which consists of 18 researchers from eight institutions, is funded in part by the Laboratory. For the past two years, the collaboration has monitored some 8.6 million stars in the Large Magellanic Cloud with sophisticated camera systems, each of which contains 16 million pictures. The search for dark matter will continue for at least three years at the great Melbourne Telescope at Mount Stromlo Observatory near Canberra, Australia.

Contact: Charles Alcock (510) 423-0666 (alcock@ippg.llnl.gov).

Study of young and old population offers clues to DNA genetic damage

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ACCORDING to a common, rather simplistic notion, we are what we eat. On a far more empirical level, epidemiological studies reveal a connection between diet and adverse health consequences. Many observed differences in cancer rates worldwide, including incidences of colon and breast cancer, are linked to variations in human diets.

Strong evidence suggests that mutations are the initiating events in the cancer process. In other words, the complex sequence of cellular changes ultimately leading to malignant tumors is thought to begin with structural changes—mutations—within the molecular units that make up the genes.

For 17 years, LLNL researchers have been investigating certain biologically active compounds in foods that can trigger tumors in animals, at least after exposure to high concentrations, by producing cellular mutations. At first glance, identifying the mutagens that might put us at risk and understanding how they affect the body appear to be simple matters. In fact, the opposite is true. Consider just a few of the questions that must be addressed to understand the entire picture of diet-induced mutations and possible links to cancer. Exactly what compounds in foods are dangerous, how are the compounds formed during cooking, in what amounts are they present after cooking, and how toxic or cancer-causing are they? What chemical changes take place metabolically at the molecular level after the mutagenic substances are consumed? For example, what role do metabolic enzymes play, how is DNA affected, and how might tumors be triggered in the body’s somatic cells? What chemical, tissue, animal, and human models might be useful to estimate risk to the human population? Are all people affected similarly, or are some resistant to cancer-causing effects? If people vary in cancer incidence, what accounts for the differences in susceptibility?
environmental sciences, and forensics (Figure 1). Our research requires tools such as accelerator mass spectrometry and nuclear magnetic resonance spectrometry, to name a few. The Laboratory is one of the few places that brings together the broad expertise and state-of-the-art analytic tools required to fully understand each important aspect of the problem of mutagens and carcinogens in the human diet. The way we became involved in this field of research has much to do with our role as a national laboratory with interdisciplinary research programs.

Mutagens are the damaging agents that can structurally change the molecular units that make up the genes (that is, the genetic material, DNA) or the relation with extracts from heated animal muscle and found that the extracts were carcinogenic, but the research went no further.

By the early 1970s, Bruce Ames at the University of California, Berkeley, had developed a biological test to measure the mutagenic potency (mutagenicity) of substances.

1*In the late 1970s, T. Sugimura, who directed research at the National Cancer Center in Tokyo, applied the Ames method and published the fact that smoke condensate from cooking and the charred surface of broiled fish and beef were mutagenic.

2One year later, Barry Commoner, working at Washington University, St. Louis, used the Ames method to show that cooking temperature and time affect the formation of mutagens in food.

3The news that cooking amino acids (the building blocks of proteins) and muscle-containing foods could be dangerous triggered considerable scientific interest around the world. In 1978, biomedical researchers at LLNL were working on the problem of mutagenic chemicals produced by oil shale retorting and coal gasification.

Because of our combined expertise in chemical analysis (including different types of chromatography and spectrometry), biological analysis (including the Ames mutation assay), and our emerging program in genetics and toxicology, we received a multiyear contract from the National Institute of Environmental Health Science (NIHES) to take a detailed look into the problem of food mutagens. As it turns out, what happens when oil shale and coal are heated is not so different from some of the research issues addressed and analytic methods used in this field of investigation. This series of articles focuses on the first five questions under “Issues” listed in Table 1. A second installment in Science and Technology Review will address the remaining issues.

A simple analogy can help put a key feature of our work into perspective. The compounds we have been investigating for nearly two decades—the aromatic heterocyclic amines—are present in cooked foods at very low levels, in the range of about 0.1 to 50 parts per billion. Isolating material at the part-per-billion level is equivalent to pouring a jigger of Scotch into the hold of a full supertanker and then trying to retrieve it again. Although the compounds we study are present in very small amounts, they are also the most mutagenic compounds ever found, and they produce tumors in mice, rats, and monkeys. Such knowledge, combined with the fact that these compounds are present in many foods characteristic of the Western diet and that certain diets are known to influence tumors at several body sites, gives our research an extra sense of urgency.

LLNL’s Approach

The single aspect that best characterizes our research on food mutagens and carcinogens—and sets our work apart from almost all other efforts around the world—is its multidisciplinary nature. Biomedical scientists at LLNL routinely collaborate with investigators working in analytical chemistry, synthetic chemistry, quantum chemistry, physics, and radiology. Clearly then, isolating, identifying, and assessing the biological activity of mutagenic compounds in food is a difficult problem requiring extensive effort. Table 1 is an overview of some of the research issues addressed and analytic methods used in this field of investigation. This series of articles focuses on the first five questions under “Issues” listed in Table 1. A second installment in Science and Technology Review will address the remaining issues.

Table 1. Some of the required interdisciplinary research, analytic methods, and tools needed to understand the possible connection of mutagens in cooked food to cancer.

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<td>DNA damage and repair</td>
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<td>DNA binding analysis</td>
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<td>DNA adduct analysis</td>
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Figure 1. Cyndy Sailer, one of the researchers in the LLNL food mutagen research group, pours a cooked food sample into an extraction tube to prepare it for subsequent analysis.

*All references are at the conclusion of the third part of this installment on p. 25.
The foods that make up the Western diet, the most common mutagens belong to a class collectively called the amino-imidazoazaarenes (AIAs). Not all the known food mutagens are AIAs, but the commonly found ones are. As shown in Figure 2, AIA compounds have one or two aromatic ring structures fused to the imidazole ring. They also have an amino group (NH$_2$) on the number-2 position of the imidazole ring and can have methyl groups (CH$_3$) of varying number and location.

A Problem of Strategy

Strictly speaking, it is inaccurate to say that cooked foods contain mutagens. More precisely, certain cooked foods contain pre-mutagenic substances (promutagens) that are metabolized by enzymes naturally present in body tissues, leading to the formation of one or more reactive mutagenic substances. Conventionally, however, “promutagen” and “mutagen” are used synonymously, and we have followed that practice here unless the point being made about the research demands a precise distinction.

At the outset of our research, we were faced with problems of strategy. Studying substances that are present at very low concentrations imposes many research constraints. If we focused on only a few foods, as seemed wise, then our results and their implications for public health might be misinterpreted. Instead, we decided to examine the foods that are the principal sources of cooked protein: meats (any muscle-containing food, including fish, eggs, beans, cheese, and tofu). Whereas we initially focused on meats, especially fried beef, we have now expanded the range of foods to include cooked breads and grain products, heated flour from many different plant sources, and meat substitutes.

Over the years, our research has also evolved from relatively simple concepts and approaches to more sophisticated ones. Initially, we had to identify the mutagenic compounds in heated foods because many were not known (that is, neither synthesized nor analyzed). Thus, we focused our efforts on identifying the chemical composition and structure of mutagens, assessing how different cooking procedures affect the formation of mutagens, and determining the amount (abundance) of the mutagenic products. Even though chemical identification and quantification are still important activities, our work has expanded to include many other aspects of the problem.

For example, we developed techniques to help us learn how mutagens are metabolized in the body. We use animals as models to understand complex metabolic pathways and are developing cell-culture methods that model human metabolic systems. One particularly important issue is how metabolites (the intermediate products formed by enzymes) interact with the genetic material. We need to know exactly what takes place at the molecular level, including covalent binding with and structural changes to specific components of DNA. This work taps the skills and facilities in several related research programs, including the Human Genome and DNA repair projects. (See the April/May 1992 and April 1993 issues of *Energy and Technology Review* for more background on these two programs.)

In assessing the effects of low-level exposure to food mutagens, we make use of Laboratory expertise in accelerator mass spectrometry (AMS). Yet another part of the story is the differences among humans in susceptibility to cancer, which has become our newest effort. In essence, our success in recent years is derived not so much from simply applying standard analytical methods by themselves as from combining biological analysis with state-of-the-art analytical tools available at LLNL to study all aspects of the health risks, ranging from dietary exposure to effects in model systems and humans.

In the foods that make up the Western diet, the most common mutagens belong to a class collectively called the amino-imidazoazaarenes (AIAs). Not all the known food mutagens are AIAs, but the commonly found ones are. As shown in Figure 2, AIA compounds have one or two aromatic ring structures fused to the imidazole ring. They also have an amino group (NH$_2$) on the number-2 position of the imidazole ring and can have methyl groups (CH$_3$) of varying number and location.

Of the list of toxic substances known to be produced during cooking, the most important may well be the AIAs. Also referred to as heterocyclic amines, these compounds are potent mutagens produced at normal cooking temperatures in beef, chicken, pork, and fish when fried, broiled, or grilled over an open flame. The pan residuals that remain after frying also have high mutagenic activity, indicating that meat gravies can be a source of exposure. Our research suggests that smoke from cooking meat is mutagenic as well, but any such air exposure is likely to be far less than that from eating the cooked food. Other foods, such as cheese, tofu, and meats derived from organs other than animal muscle, have very low or undetectable levels of AIA mutagens after they are cooked.

Extraction

Analyzing cooked foods for mutagens requires many different methods (Figure 3). The toxic compounds in food must first be...
Step 1. Extract mutagens from cooked food

Step 2. Separate and purify the many different compounds in the complex mixture

Solid-phase extraction

High-performance liquid chromatography (HPLC)

Step 3. Detect Mutagenic Activity

Combine food extract, bacteria, and enzymes

Count revertant colonies (baseline measurement)

Ames/Salmonella Test

Step 4. Subsequent Characterization

Mass spectrometry

Determine molecular weight (MW) and chemical composition

Nuclear magnetic resonance (NMR) spectrometry

Determine definitive structure

Figure 3. Some of the steps required to extract, separate, purify, and confirm the potency and chemical structure of mutagens in cooked food. These steps show a typical sequence of events during research on a given mutagen. However, the sequence shown here can vary depending on whether our objective is to study a known mutagen or to assess the properties of a new candidate. Each of the steps is described in more detail in the text.
needed for the next step—testing for mutagenic potency.

Detection of Mutagenicity

The most widely used detection method for mutagenic potency is the Ames/Salmonella mutation test, which is described in more detail in the box on p. 16. This test for mutagenic activity is exquisitely sensitive and relatively inexpensive. It is also convenient because each analysis requires only 48 hours, and many samples can be analyzed in parallel (Figure 5).

The essential point to remember is that the Ames test (step 3 in Figure 3) gives us a number by which we can express the mutagenic activity of a given compound or food extract. This number by itself for a single mutagen would have little meaning. However, we now have ... other sources, so we can compare the mutagenic activity of many different structural types. When the Ames test is used during initial screening for new mutagens and carcinogens, it serves as a guide to the chemical purification of biologically active molecules. It can also be used to test and compare the potency of newly synthesized chemicals.

Characterization

Once a mutagen has been detected, we can characterize it further through a variety of analytical methods (step 4 in Figure 3). The type and sequence of tests depend on our objective for a given mutagen (Figure 6). For example, we can routinely determine the molecular weight through mass spectrometry and study the detailed chemical composition (the number of hydrogen, carbon, and nitrogen atoms) by high-resolution mass spectrometry (HRMS). In mass spectrometry, complex compounds are broken up into ionized fragments, which are accelerated through a magnetic field until they strike a detector. Because the path of an ionized fragment through the field is determined by its inertia, we can determine the mass of the various ions by their spatial distribution on the detector. Ultraviolet absorbance spectrometry and fluorescence spectrometry are other identification methods that are often combined with chromatography.

Substantially more effort is required if we want to identify a mutagen for the first time. For an unknown compound, we first need information on the atomic composition and the position of atoms in the molecule. This work requires HRMS and nuclear magnetic resonance (NMR) spectra (step 4 in Figure 3).

Chemically extracted before purification. Over the years, we and other researchers have dramatically improved on the original extraction techniques that required various acids or mixed organic solvents in multistep schemes.

We now use solid-phase extraction, which is based on a method first described by G. A. Gross in 1990. After homogenizing cooked food in a blender to obtain a uniform sample, we can extract a sample quickly and efficiently by passing it through a series of small tapered tubes containing chemically activated particles (see step 1 in Figure 3 and Figure 4). The small amounts of organic solvents that are needed during this solid-phase extraction generate a minimum of hazardous waste.

Separation and Purification

We use high-performance liquid chromatography (HPLC) for final separation and purification of the extracted compounds in a food sample (see step 2 in Figure 3). Liquid chromatography is a standard technique in chemistry labs. In HPLC, a liquid mixture is pumped under high pressure through a long, narrow tube filled with fine silica particles. This material differentially retards the passage of different molecular components so that each one exits after a characteristic delay or retention time. Our recent solid-phase extraction method together with HPLC allows excellent quantification from small samples (about a tenth of a hamburger patty, or one bite) and a 1- to 2-day turnaround time for results.

For unknown mutagens, a separation is carried out in several stages. We obtain about 100 fractions at the final stage, where a “fraction” is one portion of the sample material that is captured in a separate vial after exiting the HPLC detector. One fraction at the final stage of separation contains as little as a billionth of the starting material. However, because the extracts from meat and other food products cooked at elevated temperatures are tremendously potent, only a very small sample is needed for the next step—testing for mutagenic potency.

Figure 4. Researcher Cyndy Salmon uses solid-phase extraction to extract a sample by passing it through a series of small cylinders containing small amounts of organic particles.
Our success in detecting and identifying mutagens in cooked foods is made possible by the interplay of different types of chemical analyses, including chromatography and mass spectrometry (Fig. 3), and biological methods. The Ames test is an exquisitely sensitive biological method for measuring the mutagenic potency of chemical substances. The Ames test by itself does not demonstrate cancer risk; however, mutagenic potency in this test does correlate with the carcinogenic potency for many chemicals in rodents.

The test was developed in 1975 by Bruce Ames and his colleagues at The University of California at Berkeley. The Ames method is based on inducing growth in genetically altered strains of the bacterium Salmonella typhimurium. To grow, the special strains need the amino acid histidine. However, when the chemical agent (mutagen) that is being studied is given to bacteria, some of the altered Salmonella undergo mutations. Following a particular type of mutation, the bacteria can grow like the original “wild” (unaltered) strains without histidine. Because the mutant bacteria revert to their original character with regard to the nutrient histidine, they are called “revertants.”

The Ames test yields a number—specifically, the number of growing bacterial colonies—which is a measure of the mutagenic activity (potency) of a treatment chemical. This value is often expressed as the number of revertants per microgram of a pure chemical (mutagen) or per gram of food containing that mutagen. Some pure mutagens result in hundreds of revertants per microgram, but many of the substances we have tested from cooked meats produce hundreds of thousands of revertants per microgram. For example, in one strain of bacterium, the PHp mutagen results in about 2000 revertants per microgram, whereas another cooked food mutagen, IQ, results in 200,000. This illustration at the right shows how a food extract is tested for its mutagenic activity.

In brief, a test begins by placing about 100 million Salmonella bacteria in a petri dish containing a nutrient agar lacking histidine. A few bacteria will spontaneously revert in the absence of mutagens. Counting these revertant colonies gives us a baseline against which to check the validity of our complex laboratory procedures. In a separate but essentially identical histidine-deficient petri dish, another batch of bacteria are given a mutagen plus mammalian enzymes required for metabolism. (Adding such enzymes gives us a more realistic measure of the mutagenicity of a substance for mammals. The enzymes are typically supplied from liver cell extracts of rats given substances to increase levels of metabolizing enzymes.) Revertant bacteria grow into visible colonies. We simply count the colonies (equal to the number of revertants) after a standard time (48 hours) under standard growing conditions (37°C).

Different strains of altered Salmonella bacteria are available for the Ames test. The strains vary in sensitivity to specific mutagens. We used two strains, known as TA98 or TA100, for most of our recent work on fried beef and cooked grains. These strains were generously supplied by Bruce Ames.

Salmonella bacteria are given a mutagen plus mammalian enzymes required for metabolism. (Adding such enzymes gives us a more realistic measure of the mutagenicity of a substance for mammals. The enzymes are typically supplied from liver cell extracts of rats given substances to increase levels of metabolizing enzymes.) Revertant bacteria grow into visible colonies. We simply count the colonies (equal to the number of revertants) after a standard time (48 hours) under standard growing conditions (37°C).

The Major Food Mutagens

Table 2 is a summary of the 14 major mutagens that have been identified in at least one type of heated food to date. Notice that some of the compounds have the same molecular weights. For example, 4-MeIQx and 8-MeIQx are isomer pairs and so are Trp-P-2 and Me-AoC. The ultraviolet absorbance spectra of two different compounds may be identical when they are isomer pairs and differ only, for example, in the position of a methyl group on one of the rings. The similar properties of isomers make them difficult to separate using chromatography. Likewise, other analytic tools do not always differentiate between isomers. Additional mutagenic isomers have been synthesized for most of the food mutagens in Table 2. The presence of isomers means that we need to apply several different criteria for the identification purposes because no single property, such as an absorbance spectrum, can uniquely identify all of the mutagens. The compounds listed in Table 2 are not the only mutagens or carcinogens in food. Researchers at LLNL and elsewhere have identified other biologically active compounds, including additional aromatic amines, nitrosamines, and hydrazines. However, the heterocyclic amines we have been investigating are among the most abundant and potent substances detected to date. Because of their presence in cooked meats that are common in Western diets and their association with certain types of cancer in laboratory animals, they warrant detailed investigation.

<table>
<thead>
<tr>
<th>Chemical name</th>
<th>Molecular weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-aminopurine</td>
<td>170</td>
</tr>
<tr>
<td>2-amino-1,6-dimethyl-4H-pyrido[3,4-b]indole</td>
<td>176</td>
</tr>
<tr>
<td>2-amino-9H-pyrido[2,3-b]indole</td>
<td>183</td>
</tr>
<tr>
<td>2-amino-1-methyl-5H-pyrido[3,4-b]indole</td>
<td>184</td>
</tr>
<tr>
<td>3-amino-1-methyl-5H-pyrido[3,4-b]indole</td>
<td>197</td>
</tr>
<tr>
<td>3-amino-1-methyl-5H-pyrido[2,3-b]indole</td>
<td>197</td>
</tr>
<tr>
<td>2-amino-1-methyl-6H-pyrido[3,4-b]indole</td>
<td>198</td>
</tr>
<tr>
<td>2-amino-1-methyl-6H-pyrido[2,3-b]indole</td>
<td>199</td>
</tr>
<tr>
<td>3-amino-1,4-dimethyl-5H-pyrid[3,4-b]indole</td>
<td>211</td>
</tr>
<tr>
<td>2-amino-1,4-dimethyl-5H-pyrid[2,3-b]indole</td>
<td>212</td>
</tr>
<tr>
<td>2-amino-3,8-dimethyl-5H-pyrido[2,3-b]indole</td>
<td>213</td>
</tr>
<tr>
<td>2-amino-1-methyl-6-phenylpyridine</td>
<td>224</td>
</tr>
<tr>
<td>2-amino-3,4-dimethyl-4H-pyrido[3,4-b]indole</td>
<td>227</td>
</tr>
</tbody>
</table>
Cooking practices can cause large variations in the total mutagenic activity and in the amount of specific mutagens present in muscle-containing foods. For example, the amount of mutagens in a cooked hamburger from a restaurant varies considerably from one vendor to another and is often several-fold lower than that in a hamburger prepared in our laboratory (and presumably at home). The variation has much to do with the details of food preparation, such as cooking temperature and cooking time. It is becoming increasingly clear that there can be many different routes and rates of formation for the different mutagens we are investigating. Thus, a major concern is to identify the precursors and specific reaction conditions that lead to the formation of mutagens during cooking. With this information, it may be possible to devise strategies to reduce or prevent the formation of mutagens.

**Precursors**

The reactions that produce mutagens in cooked food are not merely the random coalescence of small fragments. We now know that the heterocyclic amines can be formed from single amino acids (the building blocks of proteins) or proteins when these precursors are heated alone. However, the temperatures required to produce mutagens from amino acids or proteins by themselves are higher than those normally used in cooking.

Muscle meats contain creatine and creatinine. At more typical cooking temperatures (greater than 150°C), one or both of these two precursors react with the free amino acids and, in some cases, sugars to form a series of heterocyclic amines more easily.

**Modeling the Formation**

We have modeled the formation of the important mutagen, PhIP (pronounced “fip”), starting with the amino acid phenylalanine mixed with either creatine or creatinine, both of which are found naturally in animal muscle. When phenylalanine and creatine are mixed in the proportion normally found in raw beef and dry heated at 200°C, PhIP is produced in amounts comparable to those found after cooking beef. Figure 7 shows the structures of phenylalanine and creatine and of the PhIP molecule that is produced.

We have modeled the formation of several other food mutagens in additional laboratory experiments. For example, the mutagen IQ can be formed with creatine, creatinine, and any of four different amino acids, again suggesting many different possible routes of formation.

Model reactions can help us identify new mutagens as well. In one case, dry heating three precursors known to be present in meat led us to identify a mutagen with two amino and two methyl groups and a molecular weight of 244. However, the presence of this new mutagen in food has not been verified.

**Variations in Cooking**

During the actual cooking of meat patties, water and precursors move to the hot, drying contact surfaces of the meat where reactions occur. Such migration, with water serving as the transport vehicle, may account for the concentration of precursors near the meat surface, which we have observed in several investigations. However, different cooking practices can lead to very different results. For example, some mutagens are produced at all frying temperatures, whereas others may require higher temperatures. Furthermore, when hamburger patties are grilled at high temperature over an open flame, we can account for less than 30% of the mutagens in the meat. When cooking over an open flame, polycyclic aromatic hydrocarbons (different from AIA food mutagens) arise from fat that drips from the meat—this is an entirely different mechanism than those that produce heterocyclic amines from heated muscle tissue itself. Thus, the formation of mutagens is complex and highly dependent on the details of cooking.

**Preparation Principles**

Given this complicated picture, what statements about food preparation can we make with any certainty? Here is a summary of some of the important things we have learned about the cooking process:

- Food mutagens can be produced both with and without water present.
- Early reports suggested that water is essential to produce food mutagens. In later studies, dry heating actually gives a...
Can the Mutagens in Cooked Beef Be Reduced?

Since mutagens were first observed in cooked meats, researchers in several different laboratories have explored various ways to reduce the amounts produced during food preparation. They have found that mutagenic activity can be lowered by adding antioxidants, soy or cottonseed flour, tryptophan, and various other food additives or sugars either alone or with starch. However, none of these additives is widely used commercially at home.

Consumer acceptance and possible changes in the taste, texture, and nutritional content of the cooked food need to be explored further. Surveys have shown that more than 90% of American homes have a microwave oven. As a practical way to reduce the mutagen and fat content of beef, we studied microwave pretreatment of hamburger for various times before conventional frying either at 200 or 250°C for 6 minutes per side. Our tests used a standard commercial microwave oven set at 80% power for 0 to 3 minutes. The results were dramatic.

How We Fried the Burgers We Studied

We examined the production of PhIP and other mutagens in beef at different cooking temperatures and times. The box at the right gives the details on how we prepare our fried beef. Figure 8 shows the mutagenic activity, as measured by the Ames test, of all the mutagens combined in a gram of beef的重要单源食材的来源。然而，我们发现在不同实验室使用不同的分析方法时，结果通常是一致的，尽管不同的实验室使用不同的分析方法时，结果通常是一致的，尽管不同的实验室使用不同的分析方法时，结果通常是一致的，尽管不同的实验室使用不同的分析方法时，结果通常是一致的。
patty fried at 150°, 190°, and 230°C. We found no detectable heterocyclic amines after frying at 150°C for 2 to 4 minutes. In general, increasing either the temperature or time of cooking (specifically, frying on a solid metal restaurant-type grill) causes a dramatic increase in both the mutagenic activity and the total amount of mutagens produced, especially PhIP and 8-MeIQx. For the most part, as shown in Table 3, the amount of individual mutagens in fried beef increases proportionately with the cooking temperature. A clear exception to this trend is the compound PhIP, which is produced at much greater concentrations at higher temperatures relative to the other mutagens we have studied. When the cooking temperature and time are increased, the PhIP content of fried beef patties increases nearly exponentially.

**Table 3. Content of four different mutagens in fried beef patties (expressed as nanograms of mutagen per gram of beef) cooked at different times and temperatures.**

<table>
<thead>
<tr>
<th>Cooking time per side, min</th>
<th>150°C</th>
<th>190°C</th>
<th>230°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>IQ</td>
<td>none</td>
<td>none</td>
<td>none</td>
</tr>
<tr>
<td>4-MeIQx</td>
<td>none</td>
<td>none</td>
<td>0.15</td>
</tr>
<tr>
<td>8-MeIQx</td>
<td>none</td>
<td>0.25</td>
<td>0.4</td>
</tr>
<tr>
<td>4,8-DiMeIQx</td>
<td>none</td>
<td>none</td>
<td>1.6</td>
</tr>
<tr>
<td>PhIP</td>
<td>none</td>
<td>none</td>
<td>1.3</td>
</tr>
</tbody>
</table>

**Mutagens from Grain?**

We also recently used the Ames test to assess the mutagenic activity in many heated foods derived from grain products. Our studies include cooked breads (white, pumpernickel, crescent rolls, and pizza crust), breadsticks, heated flour from many different grain sources, breakfast cereals, grain crackers, and meat-substitute patties after frying. These foods were either tested as purchased without additional cooking (for example, grain crackers and a grain beverage powder) or were cooked according to package instructions. In some studies, we deliberately overcooked the grain products for twice the cooking time at the specified temperature setting to see if the mutagen content would increase with continued cooking, as it does in muscle meats. Our studies generally demonstrate increased mutagenic activity in grain foods with cooking time, but the exact composition of the food is important. For example, when wheat gluten (the protein in wheat seed) is heated alone at 210°C in a beaker, it shows a potent, time-dependent mutagenic response (Figure 9). Because breadsticks are high in wheat gluten, they also show some activity when heated normally and much higher activity when overcooked. In fact, the mutagenic activity of breadsticks cooked for double the regular heating time is 20% that of a hamburger fried 6 minutes per side at 210°C. In all cases, overcooking grain foods leads to much higher mutagenic activity. Cooked rye flour (containing no gluten), on the other hand, showed no detectable activity, and rice cereal showed very little. Fried tofu (soy bean curd) was not mutagenic, and the measured level of activity in meat-substitute patties (which are made from vegetable proteins) after frying was about 10% or less than that of a beef patty cooked under the same conditions.

Table 4 summarizes the mutagenic activity, as measured by the Ames/Salmonella test, for a variety of cooked-grain food products. The results are expressed as mutagenic activity from the Ames test, so they cannot be directly compared with those in Table 3. (Recall that the numbers in Table 3 represent a different measure, namely the content by weight of individual mutagens.) Because we do not yet know the identity of the mutagens present in cooked grain products, we cannot provide their content by weight. However, to allow for some comparison between cooked grain and meat, we have included the values of mutagenic activity for hamburger cooked for three different times at the end of Table 4.

Overall, the level of mutagenic activity measured in heated nonmeat foods is lower than that in cooked meats. It is important to recognize that the cooked grains we studied lack the creatine and creatinine levels that explain the formation of mutagens in muscle meats during cooking. We are currently investigating the question of why foods high in gluten are quite mutagenic in the absence of creatine and creatinine. We suspect that the amino acid, arginine, can substitute for the creatine and creatinine precursors found in meat, but it may be a less effective substitute.

**Figure 9.** The mutagenic activity of wheat gluten increases dramatically when heated at 210°C for up to 2 hours. This potent response tells us that one or more highly mutagenic chemicals, still unidentified, are formed with continued cooking at high temperature.

### Table 4. Mutagenic activity of nonmeat food products (expressed as the number of revertants [mutants] per gram from the Ames/Salmonella test using the TA98 strain of bacteria). Results for hamburger are given for comparison.

<table>
<thead>
<tr>
<th>Mutagen activity, revertants per gram</th>
</tr>
</thead>
<tbody>
<tr>
<td>Food sample</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Chemical-grade gluten</td>
</tr>
<tr>
<td>Food-grade wheat gluten</td>
</tr>
<tr>
<td>Cornmeal</td>
</tr>
<tr>
<td>Garbanzo flour</td>
</tr>
<tr>
<td>Tofu flour</td>
</tr>
<tr>
<td>Rye flour</td>
</tr>
<tr>
<td>Wheat flour for bread</td>
</tr>
<tr>
<td>Food samples cooked double the time directed</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>White bread</td>
</tr>
<tr>
<td>Pumpernickel</td>
</tr>
<tr>
<td>Breadsticks</td>
</tr>
<tr>
<td>Crescent rolls</td>
</tr>
<tr>
<td>Pizza crust</td>
</tr>
<tr>
<td>Graham crackers</td>
</tr>
<tr>
<td>Grain beverage</td>
</tr>
<tr>
<td>Commercial meat substitutes fried at 210°C for 6 minutes per side</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Gluten-based patties (various samples)</td>
</tr>
<tr>
<td>Tofu</td>
</tr>
<tr>
<td>Falafal</td>
</tr>
<tr>
<td>Tempeh burger</td>
</tr>
<tr>
<td>Tofu burger</td>
</tr>
<tr>
<td>Soy-based patties</td>
</tr>
<tr>
<td>Glutinous wheat, bean-based patties (230°C)</td>
</tr>
<tr>
<td>Hamburger fried at 210°C for 6 minutes per side</td>
</tr>
<tr>
<td>Hamburger fried at 250°C for 6 minutes per side</td>
</tr>
<tr>
<td>Hamburger fried at 250°C for 6 minutes per side</td>
</tr>
</tbody>
</table>

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### Table 3. Content of four different mutagens in fried beef patties (expressed as nanograms of mutagen per gram of beef) cooked at different times and temperatures.

<table>
<thead>
<tr>
<th>Mutagen</th>
<th>Cooking temperature of grill, °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>150°C</td>
<td>190°C</td>
</tr>
<tr>
<td>IQ</td>
<td>none</td>
</tr>
<tr>
<td>8-MeIQx</td>
<td>0.7</td>
</tr>
<tr>
<td>4,8-DiMeIQx</td>
<td>1.6</td>
</tr>
<tr>
<td>PhIP</td>
<td>1.3</td>
</tr>
</tbody>
</table>

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### Table 4. Mutagenic activity of nonmeat food products (expressed as the number of revertants [mutants] per gram from the Ames/Salmonella test using the TA98 strain of bacteria). Results for hamburger are given for comparison.
efficient mutagen precursor in cooked grain products.

Before we can evaluate the risk associated with cooked grains, we need to determine the mass of mutagens in each food and to identify the specific mutagenic compounds that are present. Except for very low levels of PhIP in wheat gluten (accounting for only 4% of its mutagenic activity), our analysis did not reveal any of the other mutagens found in cooked meat or listed in Table 2. Because the mutagens in cooked grain appear to be as potent as the heterocyclic amines—and such potency is unusual, we suspect that the mutagenic compounds may be new heterocyclic amines similar to those we have found in cooked meats. However, more work needs to be done before we understand the entire picture.

What About Fumes?

Some studies have suggested the possibility of an increased risk of respiratory tract cancer among cooks and bakers. When foods rich in protein are heated, several mutagens are formed. Our research on food mutagens is associated with cooked grains appear to be as potent as the heterocyclic amines—and such potency is unusual, we suspect that the mutagenic compounds may be new heterocyclic amines similar to those we have found in cooked meats. However, more work needs to be done before we understand the entire picture.

Cook to Manage Mutagens

Our research on food mutagens is not specifically designed to generate practical advice for diet- and health-conscious individuals. Many questions remain unanswered in this highly complex field of investigation. Although food mutagens are extremely potent, our preliminary estimates of risk are not alarming primarily because of their low concentrations in food. Nevertheless, the amount of mutagens ingested can be reduced by choice of diet and by modifying cooking practices.

Cooking Tips Summary

• Fried beef has very high mutagenic activity. Its popularity suggests that this food may be the most important source of heterocyclic amines in the typical Western diet.
• Most, but not all, of the mutagenic activity in fried beef can be accounted for by known heterocyclic amines. The single mutagen PhIP accounts for most of the combined mass of mutagens in fried beef cooked well-done.
• The fumes generated during the cooking of beef have about one-third of the mutagenic activity measured in the fried meat itself. Occupational exposure over long periods could pose some risk, but probably much less than that from consuming the meat.
• The fat content and thickness of meat have little effect on mutagen content, whereas the method and extent of cooking have major effects. Frying, broiling, and barbecuing muscle meats produce more heterocyclic amines and mutagenic activity, whereas stewing, steaming, and poaching produce little or no mutagenic activity. Roasting and baking show variable responses.
• Both cooking temperature and time can be manipulated to minimize the formation of mutagens. Increasing the frying temperature of ground beef from 200 to 250°C increases mutagenic activity about six- to sevenfold.

Reducing cooking temperature and time can significantly lower the amounts of mutagens generated and subsequently consumed in the diet.

• Microwave pretreatment of meat, followed by pouring off the clear liquid before further cooking, can dramatically reduce the formation of heterocyclic amine mutagens, even if the meat is cooked well-done.
• Most nonmeat foods, including cooked grain products, contain lower levels of mutagens than cooked meats.
• At least in rodents, we know that food mutagens trigger cancer in several different target tissues, such as the liver, colon, breast, and pancreas. In a follow-up installment in Science and Technology Review, we will address the health risks to humans that may arise from exposure to heterocyclic amines. For this intriguing part of the story, we will show how these highly toxic compounds can react with the most critical molecules of all, DNA.

With a connection established between food mutagens, DNA damage, and the potential for cancer, we will then try to make sense of what all the numbers on mutagenic activity and mutagen content in food mean for the average person.

Key Words: Ames/Salmonella assay, amines, aminoimidazolecarboxamidines (AICAs), carcinogenic, DNA adducts, heterocyclic amines, high-performance liquid chromatography (HPLC), mutagens—airborne, in cooked foods, in fried beef; mutagenicity, 2-amino-1-methyl-6-phenylimidazoles (4,5-bipyridine [PhIP]), 2-amino-1-methylimidazo[4,5-f]quinoline (IQ), 2-amino-3,4-dimethylimidazo[4,5-f]quinoline (MeIQx).

References

About the Scientist

JAMES FELTON joined the Biomedical Sciences Division of Lawrence Livermore National Laboratory as a Senior Biomedical Scientist in 1976. He is currently the Group Leader of the Molecular Toxicology Group at the Laboratory. He received his A.B. in Zoology from the University of California, Berkeley, in 1967 and his Ph.D. in Molecular Biology from the State University of New York at Buffalo in 1973. From 1969 until 1976, he was a Fellow of the National Institute of Health, initially in New York and later in Maryland.

In more than 147 professional publications, James Felton has explored the role of diet in carcinogenesis and mutagenesis. He has been a part of the Laboratory’s research on food mutagens since it began 17 years ago and has led it for the past 8 years.

To view this article with interactive links to these references, visit our Internet homepage at http://www.llnl.gov/tl/str.html. After August 1, click on references in color for immediate access to additional specific information.

For further information contact James S. Felton (510) 422-5606 (felton1@llnl.gov) or Mark G. Knize (510) 422-8260 (knize2@llnl.gov).
The engine does not idle; rather, it shuts down each time the energy-storage device is fully charged. To complete the power train, an electric motor is coupled to the wheels by a feature of regenerative braking. Thus, kinetic energy returns to the storage device when the brakes are applied.

If the engine/generator in a hydrogen-powered vehicle supplies enough power for a fully loaded vehicle to climb hills at cruising speeds, then it performs much like today’s gasoline-powered automobiles. However, if the engine/generator supplies just enough power for average energy consumption, then it can serve as a range extender. The difference in power required for cruising versus hill climbing is about a factor of four. We are designing a fully capable concept car that can cruise and climb hills.

The Design Team’s Challenges

LLNL researchers are working on the technical details of a new hydrogen piston engine with investigators at Los Alamos National Laboratory and Sandia National Laboratories, California. Essentially, LLNL is responsible for the initial system studies, engine design, and combustion kinetics. Los Alamos investigators perform the computational fluid-dynamics modeling (combustion modeling) and integrate this information into our vehicle simulation codes. Researchers at Sandia’s Combustion Research Facility then do the engine-performance and emissions testing.

The need for a highly efficient vehicle and power train is driven by the associated problem of onboard storage of hydrogen fuel. Onboard fuel storage is perhaps the single most difficult task associated with our project. Table 1 shows two options we are considering for fuel storage: a cryogenic tank for liquid hydrogen or a high-pressure tank for hydrogen gas. Without increased efficiency, the onboard fuel tank would need to be about three times the volume listed in Table 1 and three times the times the size shown in the illustration; that is, the tank would become so large as to be impractical. We are applying the hybrid vehicle evaluation code (HVEC) developed at LLNL as a guide to select components that maximize efficiency and thus reduce fuel-tank volume and weight.

HVEC incorporates a wide range of details and complexity. The code calculates power-train dimensions, fuel economy, time to accelerate to 60 mph (96 km/h), hill-climbing performance, and emissions. Our basic premise is that we need to generate electrical energy at efficiencies of about 42%, based on a generator that is 95% efficient and an engine efficiency of about 46%.

Our conceptual design of a hydrogen hybrid vehicle features a large fuel tank for pressurized hydrogen. It has a gasoline-equivalent fuel efficiency of 80 mpg and a driving range of 380 mi (608 km).
January 1995 provided an effective test of CARS’s ability to simulate local precipitation and atmospheric variables, which are then averaged over individual watersheds. The Russian River basin was among the hardest-hit areas, with an estimated $800 million in flood-related damage.

Large-scale forecast data (80-km resolution) from the National Weather Service were used as input to the CARS system, and MAS simulations (20-km resolution) were run producing precipitation fields for all of California for this time period. MAS’s ability to calculate rainfall and snowfall separately was essential for predictions of river flow, since snowfall does not immediately affect river flow.

California’s complex terrain can cause considerable differences in the precipitation received by areas only a few miles apart. As a result, accurate estimates of local precipitation are essential for accurate estimates of river flow in mountainous areas. To illustrate this dependence, CARS computations were made for the area-averaged daily rainfall for the entire Russian River basin (approximately 7000 km$^2$) and compared with calculations of the Hopland watershed (a smaller area, about 660 km$^2$) within the Russian River basin, north of the Hopland gauging station. The simulated daily rainfall for the two areas differs by factors of two to three (Figure 2a).

To evaluate CARS’s ability to predict river flow and flooding, simulated precipitation values for the Hopland watershed were compared with the National Weather Service’s California–Nevada River Forecast Center to model river flow). CARS successfully simulated the amounts and timing of rainfall over the Hopland watershed, except on January 10, where the model overestimated rainfall by a factor of two (Figure 2b). Upon further examination, this overestimation was found to have resulted from excessive amounts of water vapor flux in the input data for the CARS runs, clearly demonstrating the dependence of regional predictions on accurate large-scale data.

Figure 2c plots the observed and simulated daily-mean river flow volume of the Russian River at the Hopland gauge station from January 1 through January 12. CARS simulated the river flow rate to within 10% accuracy during the flood stage. The overestimation of modeled river flow for January 11 was due in part to the overpredicted rainfall for January 10, as noted above. For the low flow periods before flooding, simulated river flow exceeded the observed river flow mainly because of uncertainties in the initial water content of the soils, a difficult variable to simulate.

These successful predictions of precipitation and river flow demonstrate the applicability of the CARS system to short-term, local weather forecasting. Such modeling will not replace human weather forecasters; rather, modeling can provide another type of data to assist forecasters. As Janow Kim, one of CARS’s developers, remarks, “The value of front-forecasting is that the forecasters have the experience to interpret data from various sources. Our goal is to create a modeling system that can help improve the accuracy of a forecast and the time span for which it is valid.”

Improving short-term weather forecasts is but one step toward the long-range goal of understanding and predicting global climate change and its regional impacts. Having successfully simulated the Russian River situation, CARS’s developers are moving ahead on several fronts.
CARS’s hydrology simulation model is being extended to include other major river systems in California, specifically the inflow to Lake Shasta, the Feather River, and the American River. This expansion will make it possible to use CARS for simulating local weather and river flows over northern California’s major watersheds.

In collaboration with the National Weather Service, the CARS system is being used for experimental weather prediction for the southwestern United States. Simulations are also being run to test CARS’s ability to assess water resources over seasonal, multiyear, and decadal time scales, to model the effect of such global phenomena as El Niño on regional climate, and to determine the effects of pollutants such as carbon dioxide and aerosols on climate change.

References


To view this article with interactive links to these references, visit our Internet homepage at http://www.llnl.gov/str/str.html. After August 1, click on references in color for immediate access to additional specific information.

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Awards
Dana Isherwood, the laboratory’s legislative analyst, and Dick Post, a laboratory associate in the Energy Directorate, were elected fellows of the American Association for the Advancement of Science (AAAS) in recognition of their scientifically or socially distinguished efforts on behalf of the advancement of science or its applications.

Ralph Jacobs, director of New Technology Initiatives in the Laser Program at LLNL, was elected fellow of the American Physical Society. He was honored for “fundamental and applied contributions to the research and development for a wide variety of gaseous, solid, and liquid laser media.”

Tom McEwan and his “radar on a chip” were honored in April by the Federal Laboratory Consortium for excellence in transferring technology from a laboratory to private business. The Consortium is an association of Department of Energy research facilities that assists the U.S. public and private sector in using technologies developed by federal research laboratories.

Secretary of Energy Hazel O’Leary presented laboratory representatives with the Management and Operation Contractor of the Year Award on March 31 in recognition of its outstanding achievement in providing substantial contracting opportunities for small businesses. She cited our socioeconomic program assisting small, women- and minority-owned business in securing procurement contracts with the Laboratory as the best of its kind in the DOE complex.

The 1994 O. E. Lawrence Award has been awarded to Michael Campbell, head of the Laboratory’s lasers program, and John Linell, scientific director for Inertial Confinement Fusion for distinguished leadership in helping to propel laser-driven inertial confinement fusion to the forefront of physics research. The award was established in 1959 in memory of Ernest O. Lawrence to recognize outstanding contributions in the field of atomic energy. Dr. Campbell was also the winner of the 1995 Edward Teller Medal. This award was established in 1989 to commemorate Teller’s contributions to fusion energy.

The Northern California Section of the American Institute of Chemical Engineers has named a hazardous explosives cleanup process developed at the Laboratory as Project of the Year. The award cited the project’s principal investigators: Ravi Upadhye, Bruce Watkins, Cesar Pruneda, and Bill Brummond. The process uses molten salt to safely dispose of waste explosives and explosive-like materials.
Research on Food Mutagens

Of all the toxic substances produced during cooking, the most important are likely to be the heterocyclic amines. For 17 years, LLNL researchers have been identifying these food mutagens, measuring their abundance in cooked foods typical of the Western diet, working to understand how they can trigger malignant tumors in laboratory animals that have been exposed to high mutagen doses, and estimating the importance of human exposures. Our success is largely a function of the interdisciplinary approach we have taken to quantify food mutagens and to study their biological effects. LLNL investigators were the first to identify five of the most important mutagens in heated food, including PhIP and DiMeIQx. We have shown that fried beef may be the most important single source of heterocyclic amines in the human diet and that PhIP accounts for most of the combined mass of mutagens in fried beef cooked well-done. Most nonmeat foods contain low or undetectable levels of these types of compounds, but some cooked protein-containing foods, such as those high in wheat gluten, have significant levels of unknown aromatic amine mutagens. Cooking time and temperature significantly affect the amounts of mutagens generated. For example, reducing the frying temperature of ground beef from 250 to 200°C lowers the mutagenic activity by six- to sevenfold. Microwave pretreatment of meat and discarding the liquid that is formed also greatly reduces the formation of heterocyclic amines. Our related work on dose and risk assessment will be described in a forthcoming article.

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Abstract

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What Do You Think?

This issue of Science and Technology Review (formerly Energy and Technology Review) represents a modified approach to communicating the work of the Laboratory. The intent of our changes is to make this publication more interesting to a broader audience. Please give us your reactions to our changes by answering the questions in section 1 and 2 below and faxing them back to us at (510) 422-8803. You can also mail your response to the address below. If you photocopy this form first, you won’t have to remove it from the publication.

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