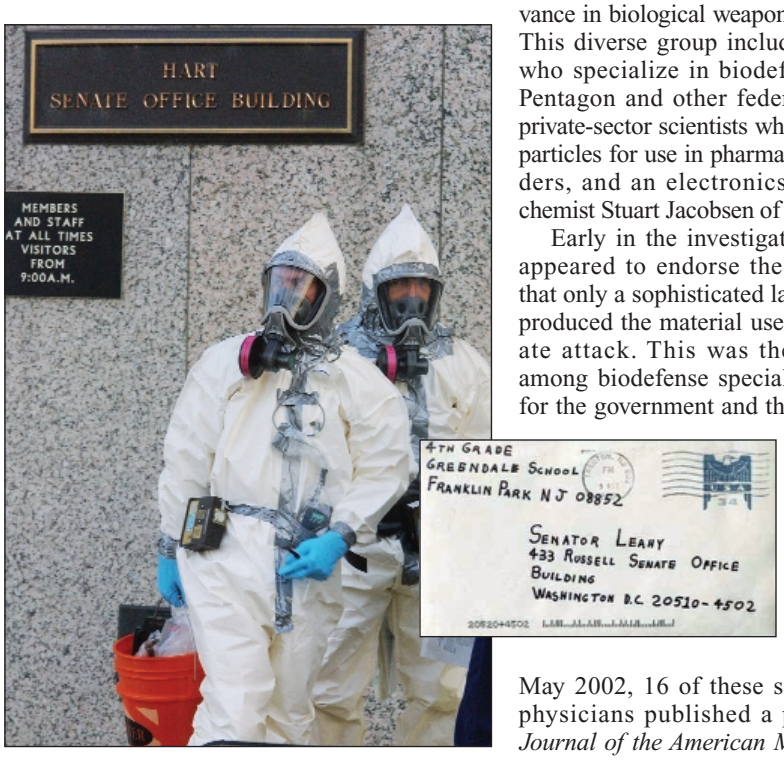
Although the investigation seems focused on the idea that the Senate powder could have been "homemade," some experts say that's improbable

Anthrax Powder: State of the Art?

When the anthrax mailers penned the message, "YOU CAN NOT STOP US. WE HAVE THIS ANTHRAX," the threat included a chilling nuance that remains largely unrecognized. "ARE YOU AFRAID?" asked the attackers. "Yes," should have been the answer, according to some biodefense experts, who think that the anthrax spores mailed to Senators Thomas Daschle (D–SD) and Patrick Leahy (D–VT) in the fall of 2001 represented the state of the art in bioweapons refinement, revealing telltale clues about the source. This view is controversial, however, because others dispute the sophistication of the Senate powder, and a schism now exists among scientists who analyzed it for the FBI.



Rapid release. The powder in letters sent to the U.S. Senate was treated in a sophisticated way to create an aerosol, some researchers say.

One group, comprised mostly of microbiologists and molecular biologists, argues that this material could have been a do-it-yourself job, made by someone knowledgeable but with run-of-the-mill lab equipment on a modest budget. This contingent includes one well-known bioweaponeer, Ken Alibek, who defected from Russia to the United States in 1992.

The other faction thinks that the powder mailed to the Senate (widely reported to be more refined than the one mailed to the TV networks in New York) was a diabolical advance in biological weapons technology. This diverse group includes scientists who specialize in biodefense for the Pentagon and other federal agencies, private-sector scientists who make small particles for use in pharmaceutical powders, and an electronics researcher, chemist Stuart Jacobsen of Texas.

Early in the investigation, the FBI appeared to endorse the latter view: that only a sophisticated lab could have produced the material used in the Senate attack. This was the consensus among biodefense specialists working for the government and the military. In May 2002, 16 of these scientists and physicians published a paper in the *Journal of the American Medical Association*, describing the Senate anthrax powder as "weapons-grade" and exceptional: "high spore concentration, uniform particle size, low electrostatic charge, treated to reduce clumping" (*JAMA*, 1 May 2002, p. 2237). Donald A. Henderson, former assistant secretary for the Office of Public Health Preparedness at the Department of Health and Human Services, expressed an almost grudging respect: "It just didn't have to be that good" to be lethal, he told *Science*.

As the investigation dragged on, however, its focus shifted. In a key disclosure, U.S. Attorney General John Ashcroft revealed in August 2002 that Justice Department officials had fixed on one of 30 so-called "persons of interest": Steven J. Hatfill, a doctor and virologist who in 1997 conducted research with the Ebola virus at the U.S. Army Medical Research Institute of Infectious Diseases in Fort Detrick, Maryland. (Hatfill has denied any involvement in the anthrax mailing.) Although the FBI did not spell out its theory, this announcement and leaks to the media from federal investigators indicated that the inquiry had embraced the idea that a lone operator or small group with limited resources could have produced the Senate anthrax powder.

This premise now appears to have run its course. In September 2003, the FBI's Michael Mason admitted that the bureau failed to reverse engineer a world-class anthrax powder like the Senate material and expressed regret that Hatfill had been called a "person of interest." One of the costliest manhunts ever conducted by federal investigators appears to be stymied. The FBI cannot or will not say whether the anthrax powder was foreign or domestic, expensively made or cheaply done, a professional job or the handiwork of an amateur.

But the scientific data amassed so far should provide a wealth of information on the weapon's possible origins, say scientists in the group with expertise in such powders. They argue that the most striking qualities of the Senate powder do not concern the anthrax spores but the way they were processed—specifically, how they were given an electrostatic charge and unusual surface properties.

If the Senate anthrax powder did in fact have these refinements, its manufacture required a unique combination of factors: a strain that originated in the United States, arcane knowledge, and specialized facilities for production and containment. And this raises the discomforting possibility that the powder

October 2001 Letters loaded with anthrax powder reach the Senate.

October 2001 Army lab finds silica in Senate anthrax powder.

April 2002 Media reports "unusual coating" on Senate anthrax spores.

CREDITS: (LEFT TO RIGHT) ALEX WONG/GETTY IMAGES; FBI

was made in America, perhaps with the resources of the U.S. government.

Charged questions

There is no debating that the Senate powder was exceptionally pure and highly concentrated. Nor is there any doubt that it contained the Ames strain, one of the most virulent strains discovered. But what made it truly remarkable, according to biodefense specialists, was its conversion into a cutting-edge aerosol.

That transformation had as much to do with chemistry and physics as with microbiology. Anthrax spores cling to one another if they get too close; sticky chains of proteins and sugar molecules on their surfaces latch onto each other, drawn by van der Waals forces that operate at a distance of a few tens of angstroms. Untreated spores clump into larger particles that are too heavy to stay airborne or reach the narrowest passages in the lung.

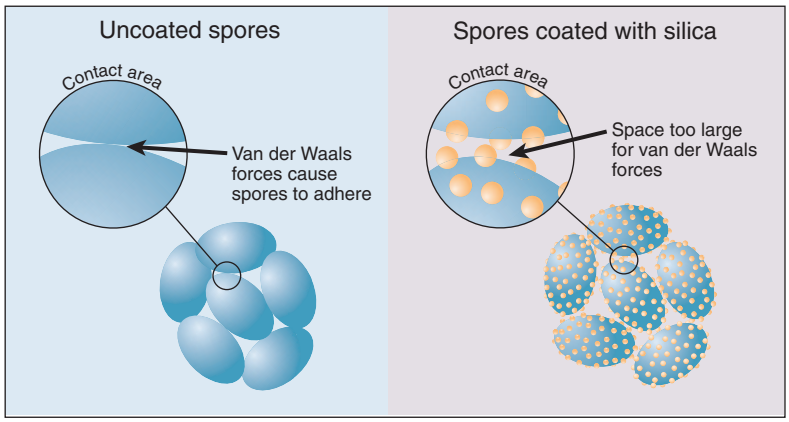
To thwart this clumping, an earlier generation of biological weapons makers—operating out of Fort Detrick when it still made weapons—experimented with ways to prevent the surfaces of germs from getting too close. For example, William C. Patrick III, former chief of Fort Detrick's Product Development Division, pioneered the use of a dusty silica powder with nanometer-sized particles added to nonlethal incapacitating agents such as *Francisella tularensis*, the cause of tularemia (but not *Bacillus anthracis*, the cause of anthrax). "Otherwise," says Patrick, the powder was "very hard to disseminate."

In a separate research arena, pharmaceutical scientists in the 1990s began experimenting with adding electrostatic charges to small particles in medicinal powders designed for inhalation. Adding a like charge of sufficient strength creates an electrostatic field of up to a few centimeters, which makes particles repel one another, creating an "energetic" or self-dispersing powder.

Biodefense scientists say they became aware that such an innovation could be applied to germ-warfare powders with deadly effect, especially deadly because charged particles are more prone to lodge in the lung. Once in the lung, immune cells transport the spores to lymph nodes, where the spores germinate and cause infection. The Senate anthrax spores carried like electrical charges, and some experts believe that they were added deliberately to aid dispersal.

Was it a coincidence that this lethal innovation appeared in the anthrax spores sent to the Senate? Alibek thinks it is possible. The Senate anthrax could have acquired a charge from friction as the envelopes passed through mail-sorting machines. (Alibek also has speculated that the powders mailed to the Senate were more refined than those sent to the New York media and may have come from a different production run.) But his theory raises a question: Why would only the Senate powder acquire a charge from the sorting machines?

Jacobsen, a research chemist who coated sub–5-micrometer particles with silica while working on a program for the Defense Advanced Research Projects Agency (DARPA), is skeptical of this idea. Jacobsen says that friction would add static electricity only to surfaces: "If anything, the sorting machine's pinch rollers and the envelopes should get charged," he says, "not the spores inside."



Spacers. U.S. and Soviet bioweapons specialists discovered that adding silica particles to germ powders made them easier to disperse.

Glassy finish

More revealing than the electrostatic charge, some experts say, was a technique used to anchor silica nanoparticles to the surface of spores. About a year and a half ago, a laboratory analyzing the Senate anthrax spores for the FBI reported the discovery of what appeared to be a chemical additive that improved the bond between the silica and the spores. U.S. intelligence officers informed foreign biodefense officials that this additive was "polymerized glass." The officials who received this briefing—biowarfare specialists who work for the governments of two NATO countries—said they had never heard of polymerized glass before. This was not surprising. "Coupling agents" such as polymerized glass are not part of the usual tool kit of scientists and engineers making powders designed for human inhalation. Also known as "sol gel" or "spin-on-glass," polymerized glass is "a silane or siloxane compound that's been dissolved in an alcohol-based solvent like ethanol," says Jacobsen. It leaves a thin glassy coating that helps bind the silica to particle surfaces.

Silica has been a staple in professionally engineered germ warfare powders for decades. (The Soviet Union added to its powders resin and a silica dust called Aerosil—a formulation requiring high heat to create nanoparticles, says Alibek. U.S. labs have tested an Aerosil variant called Cab-O-

May 2002
JAMA paper says that spores were "weaponized" and "treated."

August 2002
Attorney General John Ashcroft identifies Steven Hatfill as a "person of interest."

November 2002
Capitol Hill aides told that "no silica" was found in Senate powder.

December 2002
FBI and Dugway try to reengineer Senate powder without silica.

September 2003
FBI official says that Dugway reengineering failed; regrets identifying Hatfill as a "person of interest."

Abandoned. Fort Detrick stopped making bioweapons in this defunct (and now demolished) lab more than 3 decades ago.

ILLUSTRATION BY C. CAIN, ADAPTED FROM S. JACOBSEN; (PHOTO) TIMOTHY JACOBSEN/AP

Sil, and declassified U.S. intelligence reports state that Iraq's chemical and biological warfare labs imported tons of both Cab-O-Sil and Aerosil, also known as "solid smoke," in the 1980s).

"If there's polymerized glass [in the Senate samples], it really narrows the field [of possible suspects]," says Jacobsen, who has been following the anthrax investigations keenly. "Polymerized glasses are exotic materials, and nanotechnology is something you just don't do in your basement."

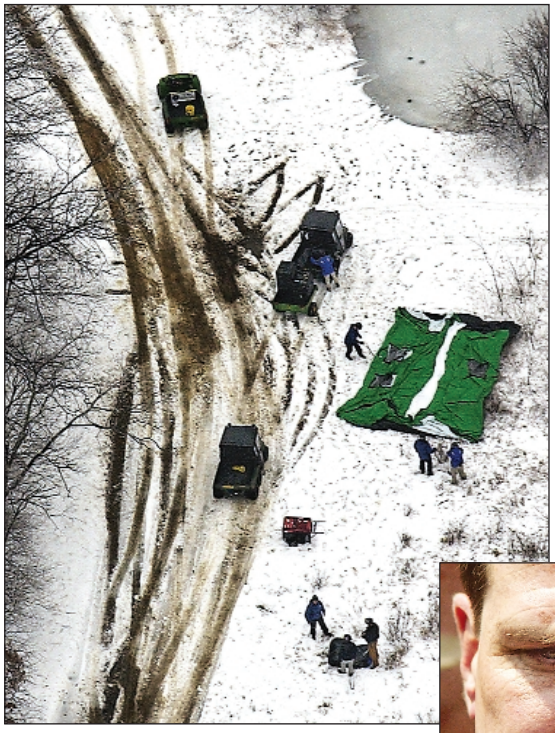
By March 2002, federal investigators had lab results indicating that the Senate anthrax spores were treated with polymerized glass, and stories began to appear in the media. CNN reported an "unusual coating" on the spores, and *Newsweek* referred to a "chemical compound" that was "unknown to experts who have worked in the field for years." When *Science* asked the FBI about the presence of polymerized glass in the Senate powder, an FBI spokesperson said the bureau "could not comment on an ongoing investigation."

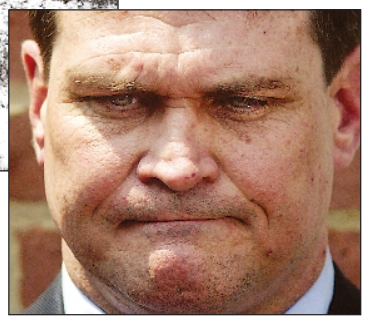
About-face

By the fall of 2002, the awe-inspiring anthrax of the previous spring had morphed into something decidedly less fearsome. According to sources on Capitol Hill, FBI scientists now reported that there was "no additive" in the Senate anthrax at all. Alibek said he examined electron micrographs of the anthrax spores sent to Senator Daschle and saw no silica. "But I couldn't be absolutely sure," Alibek says, "because I only saw three to five of these electron micrographs." Even the astonishingly uniform particle size of 1.5 to 3 micrometers, mentioned in 2001 by Senator Bill Frist (R–TN), now included whopping 100-micrometer agglomerates, according to the new FBI description recounted by Capitol Hill aides. The reversal was so extreme that the former chief biological weapons inspector for the United Nations Special Commission, Richard Spertzel, found it hard to accept. "No silica, big particles, manual milling," he says: "That's what they're saying now, and that radically contradicts everything we were told during the first year of this investigation."

Military scientists did not back off their findings. The August/October 2002 newsletter from the Armed Forces Institute of Pathology (AFIP) reported that a mass spectrometry analysis found silica in the powder sent to Senator Daschle (*The AFIP Letter*, August/October 2002, p. 6). "This was a key component," said the institute's deputy director, Florabel Mullick, in the AFIP newsletter. "Silica prevents the anthrax from aggregating, making it easier to aerosolize," she added. Frank Johnson, chief of AFIP's Chemical Pathology Division, corroborated this in an interview. "There was silica there," said Johnson, "there was no mistaking it." Maj. Gen. John S. Parker, commander of the U.S. Army Medical Research and Materiel Command at the time of the attacks, says he saw AFIP's lab reports. "There was a huge silicon spike" consistent with the presence of silica, he says. "It peaked near the top of the screen."

Other agencies support this view today.





In the cold. The U.S. Justice Department revealed that it was investigating scientist Steven Hatfill (*right*), formerly of Fort Detrick, and searched a nearby pond for clues.

For example, John Cicmanec, a scientist with the U.S. Environmental Protection Agency, says the Department of Homeland Security confirmed to EPA that the perpetrators did, in fact, use silica to weaponize the Senate anthrax spores. According to an abstract that Cicmanec will present at the annual meeting of the Society for Risk Analysis next month, this weaponized form of anthrax is more than 500 times more lethal than untreated spores.

The contradictory military data compelled the FBI to do some explaining. Sources on Capitol Hill say that in an FBI background briefing given in late 2002, Dwight Adams, one of the FBI's top-ranking scientists, suggested that the silica discovered in the Senate anthrax was, in fact, silicon that occurred naturally in the organism's subsurface spore coats. To support his thesis, Adams cited a 1980 paper published by the *Journal of Bacteriology*—a paper that Matthew Meselson, a molecular biologist at Harvard University, says he sent to the FBI. The authors reported that they found silicon, the element, in the spore coats of a bacterium called *B. cereus*, a close cousin of anthrax.

In the 23 years since the *Journal of Bacteriology* published these data, however, no other laboratory has published a report on significant amounts of silicon in the *B. cereus* spore coat, and many bacteriologists familiar with these data consider them an anomaly. Even the authors suggested the finding might have been due to "contamination."

In December 2002, the FBI decided to test whether a high-grade anthrax powder resembling the one mailed to the Senate could be made on a small budget, and without silica. To do this job, the bureau called upon Army scientists at Dugway Proving Ground, a desolate Army test range in southwestern Utah. By February 2003, the scientists at Dugway had finished their work. According to military sources with firsthand knowledge of this effort, the resulting powder "flew like penguins." The experiment had failed. (Penguins can't fly.)

Military sources say that Dugway washed and centrifuged the material four times to create a pure spore preparation, then dried it by solvent extraction and azeotropic distillation—a process developed by the U.S. Chemical Corps at Fort Detrick in the late 1950s. It is not a simple method, but someone familiar with it might be able to jury-rig a lab to get the job done. As recently as 1996, Bill Patrick says he taught scientists at Dugway how to do this.

The FBI-Dugway effort produced a coarse powder. The spores—some dried under an infrared lamp and the others air-dried—stuck together in little cakes, according to military sources, and then were sieved through "a fine steel mesh." The resulting powder was placed into test tubes. When FBI officials arrived at Dugway to examine the results, a Dugway scientist shook one of the tubes. Unlike the electrostatically charged Senate anthrax spores that floated freely, the Dugway spores fell to the bottom of the test tube and stayed there. "That tells you the par-

CREDITS: (TOP TO BOTTOM) SAM YU/*THE FREDERICK NEWS-POST*/AP; RICK BOWMER/AP

ticles were too big," says Spertzel. "It confirms what I've been saying all along: To make a good powder, you need an additive."

Close to home

One doesn't have to look very far to find a powder that more closely resembles the Senate anthrax. The U.S. Army's newest batch of anthrax simulant is a closer match, made with *B. globigii* (BG) spores, which are similar to anthrax but nonlethal. According to military sources, the Danish company Chris-Hansen spray-dried the spores (along with an unidentified "additive") in Valby, a suburb of Copenhagen. Although the spore count varied somewhat from batch to batch, Chris-Hansen says that the average concentration was 500 billion spores per gram, about 100 times more concentrated than the Army's old BG powder. Chris-Hansen shipped the bulk material from Denmark to its New Berlin, Wisconsin, facility in 1996, where, according to Army instructions, it mixed silica into the powder—a product sold commercially under the name Sipernat D 13. Sipernat D 13 is made by Germany's Degussa AG, the same company that makes Aerosil.

The initial Chris-Hansen production run wasn't exactly what the Army wanted, military sources say, so this batch of anthrax simulant was further enhanced at Dugway Proving Ground. An official at Chris-Hansen, speaking on condition of anonymity, says he doesn't know if the Army added an electrostatic charge or a coupling agent to the powder, and the Army won't discuss it. But unlike the powder that Dugway reverse engineered earlier this year, the most recent batch of simulant—according to military sources—has great "hang time."

A government scientist who had a sample of the Army's anthrax simulant described it for *Science*: When he shook a test tube filled with it, a dense fog of particles swirled to the top in roiling eddies. After 10 minutes, the powder still hadn't settled. This scientist observed two other marked similarities with the Senate material: "There appears to be a lot of static charge," he said. When he suspended the preparation in water, he saw mostly "single spores." When Canadian military scientists used this silica-laced simulant in 2001 to assess the risk from anthrax spores delivered by letter, the aerosol behaved like the one that would later contaminate Senator Daschle's office with real anthrax spores; the weaponized BG particles spread across a 50-cubic-meter room in less than 2 minutes.

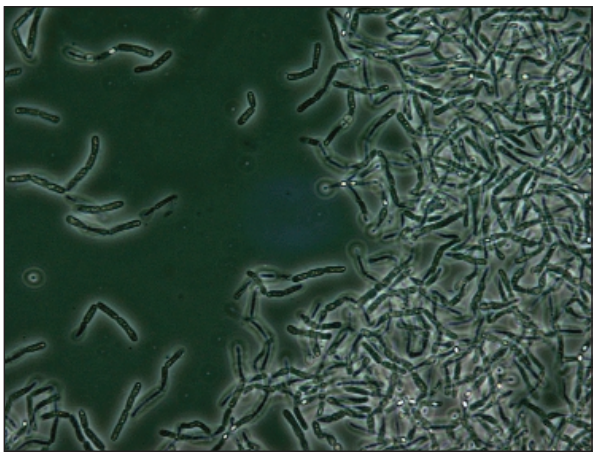
This new batch of "energetic" simulant was light-years beyond the old U.S. weapon in its refinement, experts say. Divulging the specifications of the weapon, the last foreman in charge of drying and milling anthrax spores at Fort Detrick, Donald Schattenberg, told *Science* that the old U.S. anthrax powder contained no additives. "We didn't use silica or bentonite" (a clay that contains a high percentage of fine-particulate silica), says Schattenberg. "We made little freeze-dried pellets of anthrax," he says, "then we ground them down with a high-speed colloid mill." The resulting powder contained growth media residue (called "menstruum") and vegetative cells, making it less concentrated, according to William P. Walter, who says he worked on every batch of anthrax spores ever produced at Fort Detrick. This extraneous material accounted for a significant amount of the powder's volume and mass.

Orley Bourland, who once managed the entire operation, says the old weapon had no electrostatic charge and contained only 20 billion to 30 billion spores per gram. These facts were corroborated by more than half a dozen veterans of the former U.S. weapons program, including Edgar "Bud" Larson, who scoffs at the suggestion that the Senate powder was the product of a secret one-man operation. "I think that's very unlikely," Larson said. "I don't think anyone could make this product covertly."

So far, only Dugway Proving Ground has acknowledged making aerosols with Ames strain spores. According to a memorandum from U.S. Army Test and Evaluation Command dated 19 July 1995, Dugway began experiments with a liquid preparation of the Ames strain starting in February 1994. This was part of what the Army called "bioprofiling": an effort to "establish a 'library' of information," said the memo, to help defend against biological attack. In December 2001, *The Baltimore Sun* broke the story that Dugway had been making dried anthrax using live spores, and *The Washington Post* reported that Dugway used the Ames strain in its anthrax powders. Dugway released a statement acknowledging that its scientists have been doing this work to develop an "effective bioaerosol collection" but insisted that "All anthrax used at Dugway has been accounted for."

The Battelle Memorial Institute, a nonprofit organization based in Columbus, Ohio, is possibly the only corporation in the world known to possess both the Ames strain as well as a "national security division" offering the services of a team of "engineers, chemists, microbiologists, and aerosol scientists supported by state-of-the-art laboratories to conduct research in the fields of bioaerosol science and technology." On its Web site, Battelle calls this research group "one-of-a-kind."

As subcontractors, Battelle scientists have made anthrax powders for use by the Army and U.S. intelligence agencies, but rarely by Fort Detrick, which specializes in vaccine development. Charles Dasey, spokesperson for the parent agency, the U.S. Army Medical Research and Materiel Command, says that as far as he is aware, the only dried anthrax spores made at Fort Detrick since it stopped making weapons were made by Battelle scientists working there for DARPA. This material, made in a biosafety level 3 suite in the Diagnostic Systems Division, contained killed Ames strain at a concentration of 326 million spores per gram—several orders of magnitude less concentrated than the Senate powder and crude by current standards.



They're back. Abandoned as a superpower weapon years ago, anthrax spores have returned as an instrument of terror.

Battelle is capable of more sophisticated work, as it also makes one of the world's most advanced medicinal powders. Battelle's pharmaceutical division, BattellePharma, also in Columbus, is one of the few companies anywhere developing electrostatically charged aerosols for inhalation. BattellePharma's Web site boasts that the company's new "electrohydrodynamic" aerosol "reliably delivers more than 80% of the drug to the lungs in a soft (isokinetic) cloud of uniformly sized particles." Other powders, boasts the Web site, only achieve 20% or less.

None of this argues that Battelle or any of its employees made the Senate anthrax powder. But it is evidence that Battelle was a logical place to start looking for clues. Officials from Battelle and the Army declined to comment on any aspect of anthrax powder manufacture.

The FBI says it has interviewed and polygraphed scientists working at both Dugway and Battelle. No "person of interest" at either facility has been named, and no evidence has been made public indicating either as a point of origin.

CREDIT: CDC, COURTESY OF LARRY STAUFFER/OREGON STATE PUBLIC HEALTH LABORATORY

A dose of reality

Today, there is no firm evidence to link Iraq—or any other government—to the anthrax attacks. But some weapons experts such as Spertzel are still inclined to look for a sponsor with deep pockets, and they say Hussein's regime cannot be ruled out. Spertzel's main point, however, is that only a state-run facility or a corporation has the resources to make an anthrax powder as good as the one mailed to the Senate.

The amateur anthrax scenario appears to have lost some credibility with the failure of the FBI's attempt to reverse engineer a high-quality powder using basic equipment. If the Army couldn't do it in a top-notch laboratory staffed by scientists trained to make anthrax powders, skeptics ask, who could do it in a garage or basement?

The silica dust might still provide a trail to the killers, say chemists who specialize in silica. According to military sources, since the abandonment of the offensive biological warfare program, the U.S. Army has continued to experiment with various brands of silica nanoparticles added to germ-warfare powders produced in small quantities. These include WR-50 and WR-51 (manufactured by Philadelphia Quartz Co.), Cab-O-Sil (Cabot Corp.), and Sipernat D 13 (Degussa AG). Each brand is made differently, so each has a unique chemical signature, says Jonathan L. Bass, a Pennsylvania-based analytical chemist who used to do research with silica at PQ Corp. (formerly Philadelphia Quartz). "It'd be a laborious process, and some of the differences would be hard to detect," says Bass, "but if a known brand of silica was used by the killers, I think I could trace it back to a specific company." A coupling agent should also provide a unique chemical signature that could narrow the field.

Two years on from the attacks, public discussion of the silica additive has all but ceased; the discussion about polymerized glass has yet to occur. Instead, the FBI has devoted much of its effort to the idea that a low-budget amateur operation could have produced a "weaponized" form of anthrax powder without a sophisticated additive.

"ARE YOU AFRAID?" asked our unknown assailants 2 years ago. "Yes," is still the answer, but of whom?

—GARY MATSUMOTO

Gary Matsumoto, an investigative journalist in New York City, is writing a book on biodefense.

High-Energy Physics

To B or Not to B?

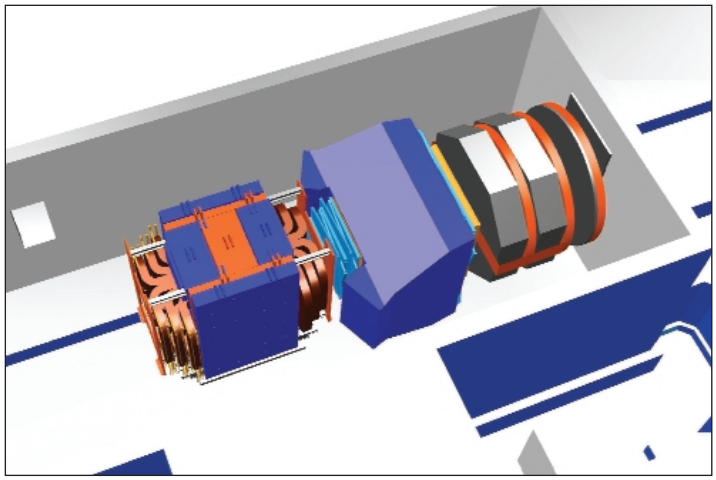
To compare matter and antimatter, physicists hope to use Fermilab's gigantic atom smasher to study particles called B mesons. But can they afford the machine's last hurrah?

Good things may come to those who wait, but don't try telling that to physicist Joel Butler. A researcher at the Fermi National Accelerator Laboratory (Fermilab) in Batavia, Illinois, Butler has been stumping for more than a decade to use the laboratory's enormous Tevatron collider to study the subtle flaw in the mirrorlike symmetry between matter and antimatter, an imbalance without which the universe would remain void. All Butler and colleagues need is a detector specifically designed to snare particles called B mesons—whose behavior may already be hinting at new particles, and which the Tevatron pumps out by the billions each year anyway.

But although two other laboratories have devoted themselves to cranking out B mesons and researchers plan to study them at a gargantuan collider currently under construction in Europe, physicists working on the Fermilab project have found themselves mired in reviews, redesigns, and political wrangling. In 2000, lab management approved plans for the new detector, known as BTeV, which should be able to measure matter-antimatter asymmetries better than the other experiments. But the U.S. Department of Energy (DOE), which funds Fermilab, has yet to ask Congress to fund BTeV's construction. "It's really up to the DOE to give us the green light," Butler says. "In a sense, we've been waiting for three-and-a-half years."

Things are looking up for BTeV, however. In October, DOE's High Energy Physics Advisory Panel (HEPAP) urged the department to fund and accelerate the $140 million project. And on 10 November, DOE listed BTeV among the 28 major facilities it hopes to build within the next 2 decades (*Science*, 14 November, p. 1126). Still, DOE has yet to show researchers the money, and some worry that, with work on the European collider proceeding apace, time is running out. "If we don't get started in 2005 we're out of business, because we've got competition," says Sheldon Stone of Syracuse University in New York.

BTeV researchers hope to start taking data in 2009, 2 years after the Large Hadron Collider (LHC) at the European particle physics laboratory CERN near Geneva, Switzerland, revs up. BTeV would then be the only particle detector running at the Tevatron, which currently supplies particle collisions for two detectors, CDF and D0, that are searching for exotic new particles. But far more is at stake than the fate of the aging collider, says Fermilab director Michael Witherell. "BTeV could be the only experiment approved this decade for a U.S. accelerator," Witherell says. "To not do it would really send a message of backing off high-energy physics."



CREDIT: FERMILAB

Going my way? The 15-meter-long BTeV detector, shown here in an artist's conception, would straddle Fermilab's Tevatron collider (blue line) and would snag B mesons moving to the right.

Angling for antimatter

Life, the universe, and everything owes its existence to the fact that matter and antimatter aren't quite exact opposites. Known as CP violation, that imbalance explains why, in the moments after the big bang, matter and antimatter did not annihilate one another and leave the cosmos empty. Thanks to CP violation, a little matter remained to form nuclei and atoms, stars and galaxies, physicists and DOE officials.

CP violation was discovered in 1964, when researchers ob-