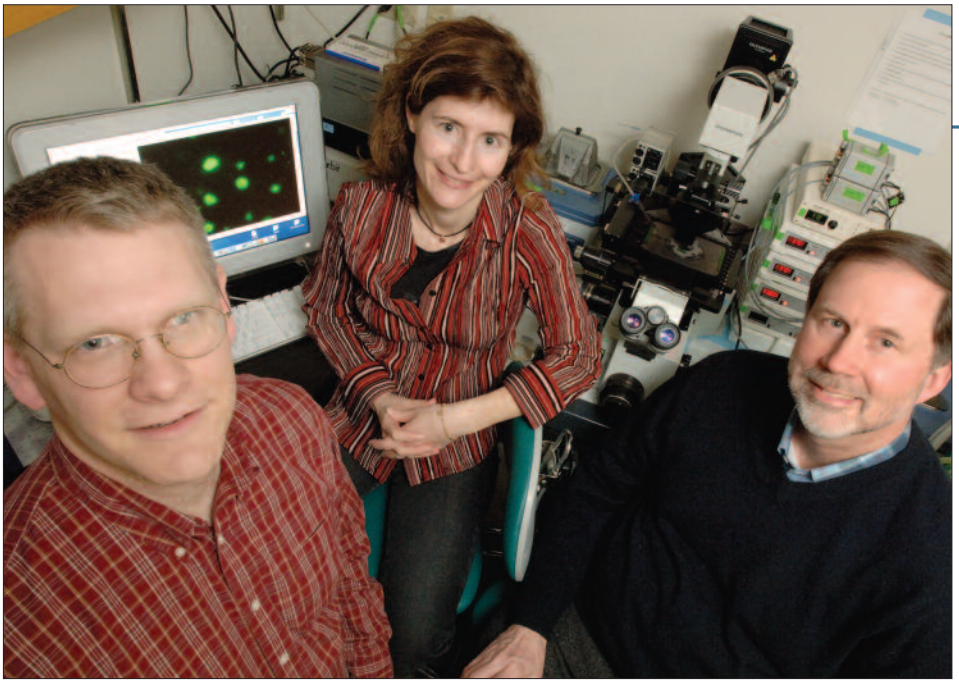

JON GILBERT FOX

Stanton, right—as well as postdocs Anderson, left, and Moreau-Marquis, center—study bacteria in biofilm form.

For a WEB EXTRA with a movie, through a confocal microscope, of *P. aeruginosa* in action, see dartmed.dartmouth.edu/spring08/html/disc_tornadoes_we.php.

Hunting down gray tornadoes in the lungs

They look like tiny gray tornadoes: little *Pseudomonas aeruginosa* bacteria spinning around on their flagella, corkscrewing down, and attaching to the surface of human airway epithelial cells.

"This is very early in the process," explains postdoctoral researcher Sophie Moreau-Marquis, Ph.D., as she watches the drama unfold on her computer monitor. She had filmed the free-swimming planktonic bacteria as they clustered together under the microscope to form biofilms on airway cells from a cystic fibrosis (CF) patient. Bacteria in biofilm communities are far more resistant to antibiotics than are free-swimming bacteria.

Glands: And that's a problem, especially for people with CF, an inherited disease of the mucous and sweat glands that affects the respiratory, digestive, and reproductive systems. *P. aeruginosa* colonize airway cells in the lungs of CF patients. The disease makes their mucus thick and slimy, so it builds up and blocks the airways, making it easy for bacteria to get a foothold and cause infections.

"One of the major problems in cystic fibrosis is that the patients have a persistent infection with *Pseudomonas*," says CF researcher Bruce Stanton, Ph.D. "That bacterial infection is what causes the lung disease. . . . You can give patients antibiotics, [but you] can never get rid of the infection."

So Stanton and other DMS scientists—in the Departments of Physiology and of Microbiology and Immunology—are trying to figure out what triggers the transformation of *P. aeruginosa* from the free-swimming to the biofilm form. Two recent papers highlight their latest work—one on how iron affects the formation of *P. aeruginosa* biofilms, and another on how antibiotic therapy actually facilitates the formation of biofilms.

In order "to outsmart bacteria, you really have to get up early."

A mutation in the gene responsible for CF "causes the CF airway cells to release more iron," which bacteria use to develop increased resistance to antibiotics, says Stanton. He's the principal investigator of that study, which was published in the *American Journal of Physiology: Lung Cellular and Molecular Physiology* and highlighted in its Editorial Focus section. "The bacteria try to trick the airway cells into allowing them to grow and to be maintained. And the airway cells secrete substances [including iron] that facilitate the development and the growth of the bacteria, as well as antibiotic resistance." In fact, bacteria are 50-fold more resistant to antibiotics in the lungs of CF patients versus in a petri dish.

"This is obviously not good news for CF patients, because it shows that current therapies are unable to eradicate biofilms in *Pseudomonas* infection," says Moreau-Marquis, a member of Stanton's lab. "And now we're starting to understand why." Next, they plan to explore whether iron chelators used in conjunction with antibiotics have a synergistic effect in killing the bacteria.

Tool: Moreau-Marquis and another postdoc—Gregory Anderson, Ph.D., who works in the lab of microbiologist George O'Toole, Ph.D.—helped develop a new tool to study the interaction between *P. aeruginosa* and human airway cells. Previously, biofilms were observed either indirectly—in sputum coughed up by CF patients—or on lab-grown airway cells that had already died. But the postdocs created a flow chamber apparatus, so a constant flow of nutrients feed cells as bacteria nestle in contact-lens-size "pools" on glass slides. The researchers now can observe, under a microscope, live interactions between airway cells and bacteria.

The second study, published in *Infection and Immunity* and highlighted in the journal's Spotlight section, showed that the antibiotic tobramycin downregulates bacterial virulence but promotes biofilm formation. "There's a growing idea that *Pseudomonas* has two different lifestyles," says Anderson. The free-swimming form causes acute pneumonia, and the biofilm form causes chronic infection. Tobramycin may just shift the balance between the two forms. "So we're decreasing the virulence," he adds, but "enhancing the persistence of the bacteria in the lung."

Scary: Making such discoveries "is very exciting as a scientist, but also scary," says Moreau-Marquis. "If you want to outsmart bacteria, you really have to get up early. They are so resistant and so able to bypass everything we know. . . . There's a lot of work to do, that's for sure."

LAURA STEPHENSON CARTER

For a WEB EXTRA about the drug facts box, including a video Q&A and sample facts boxes, see dartmed.dartmouth.edu/spring08/html/disc_drugs_we.php.

Patients deserve data about drugs

"Peaceful, restful sleep" promises an ad for the sleep drug Lunesta. Yet nowhere in the fine print of the ad (or the drug's package insert) is there any data about how many people who take the drug are helped by it or how much sleep they get when taking it. Such "benefits data" is not currently required by the Food and Drug Administration (FDA).

The ad, says DMS's Steven Woloshin, M.D., suggests that popping a Lunesta leads to eight hours in dreamland. "But if you actually look at the data, and this is real data, you fall asleep 15 minutes faster and sleep 37 minutes longer than [with a] placebo."

Drug ads have "huge laundry lists of side effects" but little data.

Common: He and other DMS researchers are also concerned that the information on side effects in drug ads can be confusing. Ads have "huge laundry lists of side effects, and oftentimes they're not highlighting the ones that are more common than placebos' [side effects]," notes DMS's Lisa Schwartz, M.D. Ads also don't make it clear which side effects may be life threatening and which are merely bothersome.

Ads like this give too little data, say DMS experts.

So Woloshin, Schwartz, and H. Gilbert Welch, M.D., created a "drug facts box," to help patients (and physicians) better understand drugs' capabilities. The top part of the box describes what the drug is designed to do, who should consider taking it, and what monitoring is recommended. In the middle is a table with study data showing the drug's benefits and side effects compared to a placebo. At the bottom is its FDA approval date.

Facts: The DMS team then recruited 274 volunteers and presented them with a drug facts box about tamoxifen (a breast cancer preventative) and a survey to test their comprehension of the data.

"People did quite well," says Woloshin. "That was very gratifying." The vast majority, 89%, correctly determined what percentage of women given tamoxifen got a blood clot in their legs or lungs; 71% calculated the absolute difference in the proportion of women who got breast cancer in the tamoxifen group versus the placebo group. Over 66% chose the better drug in two scenarios that called for comparing percentages. Half of those with only a high-school degree and two thirds of those with some college correctly answered at least four out of five questions based on the study data table. The results were published in *Medical Decision Making*.

FDA reviewers have expressed interest in the concept and are now working with Schwartz and Woloshin. The researchers' goal is to have the box included in drug package inserts, says Woloshin. They're optimistic about the prospect. The FDA reviewers "really are dedicated public servants," says Schwartz. The researchers are all affiliated with the Center for Medicine, the Media, and the Public at the Dartmouth Institute for Health Policy and Clinical Practice. MATTHEW C. WIENCKE

Depression is prevalent among teens seeking care in the ER, but they're rarely screened for it. A DMS study showed a two-question screen is nearly as sensitive as a 20-question tool.

Identifying *drug*flation

The economy is not the only thing that's inflated. People with inflammatory bowel disease (IBD) overestimate the effectiveness—and underestimate the dangers—of the commonly used drug infliximab (sold as Remicade). So found a team of Dartmouth and Harvard researchers in a survey of 165 patients or parents of patients. When asked about a hypothetical drug that mirrored the benefits and risks of infliximab, a majority of respondents said they would not take the drug. "It is likely that marketing plays some role in both patients' and physicians' beliefs," the authors wrote in the journal *Inflammatory Bowel Diseases*.

An expressive manner

A member of a common molecular family is one of the culprits behind the growth and metastasis of pancreatic cancer, according to a Dartmouth-led study in the *Journal of Clinical Investigation*. GPC1, as it is known, is overexpressed in pancreatic cancer, reported a team led by Murray Korc, M.D., "and attenuation of GPC1 expression dampens [the response to growth factors] and slows pancreatic tumor growth. . . . Taken together, the present findings suggest that targeting GPC1 may ultimately yield novel therapeutic options" for treating pancreatic cancer and its metastases.

For a WEB EXTRA with a video Q&A about the development and function of triterpenoids, see dartmed.dartmouth.edu/spring08/html/disc_trials_we.php.

Chemopreventatives show promise in trials

The discovery of a drug that could arrest cancer—or even prevent it—without nasty side effects would be big news. So findings by Dartmouth pharmacologist Michael Sporn, M.D., and his colleagues will indeed be big news, if they can make the leap from mice to humans.

In more than a dozen papers in the past two years, Sporn and Karen Liby, Ph.D., have demonstrated the potent anticancer effects of a new class of drugs called synthetic triterpenoids. Created 10 years ago by Dartmouth chemists Tadashi Honda, Ph.D., and Gordon Gribble, Ph.D., these compounds have been shown to shrink and prevent cancerous tumors in lab animals, with no toxic side effects. (For more on triterpenoids, see dartmed.dartmouth.edu/winter06/html/compound_interest.php.)

Most drugs target one molecule, or at most a few, but triterpenoids have multiple targets, some of which Sporn and his team are still discovering. This broad range of action helps give the compounds their powerful anti-inflammatory, anti-angiogenic, and cytoprotective properties. But it's also slowed down their development. Big pharmaceutical companies shy away from drugs that "do too much," says Sporn. The broader a drug's range of action, the more difficult—and expensive—it is to prove it's safe, and the more room there is for liability.

Safe: Nevertheless, Sporn was able to get a small, Texas-based company, Reata Pharmaceuticals, interested in testing triterpenoids in humans. Reata recently completed Phase I trials of the triterpenoid RTA 402, a.k.a. CDDO-Me, in patients with late-stage solid tumors and lymphoid malignancies. (Phase I trials include only 20 to 30 patients and look mostly at safety.) RTA 402 was shown to be quite safe. Furthermore, in several patients who had a high tumor burden and whose cancer was progressing, the drug halted disease progression for six months or more. RTA 402 also reduced several immunoregulatory proteins that are markers for a poor prognosis.

"It's important to remember that these patients have advanced cancers and have failed multiple other treatment options," says Melissa Krauth, a vice president at Reata. "Six months without disease progression represents a meaningful benefit. We are also excited to begin testing this drug in patients in an earlier stage of disease, where we would expect to see an even greater effect."

Effect: Reata is now conducting Phase II trials of RTA 402 in patients with pancreatic cancer, one of the deadliest and least-treatable cancers, and metastatic melanoma. "We [think] . . . RTA 402 will produce an even more profound effect when it is used in combination with standard cancer therapies," Krauth adds. "We plan to start testing it in various combinations later this year." The company expects to test RTA 402 in patients with hepatitis, rheumatoid arthritis, multiple sclerosis, and psoriasis, too. Depending on the success of these trials, and FDA timing, the drug could be available by 2010.

Meanwhile, Sporn and Liby are studying several even more potent triterpenoids that Honda has synthesized, as well as a class of drugs called rexinoids. Like triterpenoids, rexinoids—not to be confused with their cousins, retinoids, which can have toxic side effects—are also multifunctional. Sporn and researchers at other institutions have shown that rexinoids affect cell growth and differentiation, energy metabolism, and inflammation. In *Clinical Cancer Research*, Liby and Sporn reported that a rexinoid named NRX194204 prevents and treats cancer in mice. The compound reduced total lung tumor volumes by 64% to 81% and caused ER-negative breast tumors—the hardest kind to treat—to either stop growing or regress. Back in 2006, they demonstrated that another rexinoid, LG100268, can prevent ER-negative breast tumors and lung tumors in mice engineered to develop those cancers. However, Ligand Pharmaceuticals, which developed LG100268 for a purpose other than cancer treatment, has abandoned work on it and declined to comment on that decision.

"These drugs are outcasts . . . because . . . they do too much."

"These drugs are outcasts in the pharmaceutical community," says Sporn, "because as the conventional wisdom goes, they do too much." But there is hope that NRX194204 may reach patients. A California-based company, NuRx Pharmaceuticals (formerly Quest International, Inc.), is now conducting Phase I trials with that compound.

Cells: Neither triterpenoids nor rexinoids "fit the paradigm of being monofunctional magic bullets," says Sporn. Instead, they "have multiple actions in several different cell types, particularly in the tumor microenvironment." In other words, they affect not just cancer cells but also surrounding tissues. And this is the key to their success, Sporn points out, because cancer "is the end result of dysfunctional communication between epithelial cells and their microenvironment." As he is fond of saying, there is no such thing as *a* cancer cell.

JENNIFER DURGIN

JON GILBERT FOX

Sporn, rear, is shedding light on chemoprevention.

Another study from SPORT (see below) found that patients who had a diskectomy saw an improvement not only in back pain, but also in leg pain; in fact, their leg pain declined more.

Good SPORT: Study offers more evidence

Hundreds of thousands of people with back pain have surgery every year. Some with a common condition called spinal stenosis have found relief after surgery, but there had been no clear evidence it was better than nonsurgical treatment options. Now, the latest finding from the Spine Patient Outcomes Research Trial (SPORT) shows that people suffering from spinal stenosis, a narrowing of the spinal canal, improve more with surgery than with nonsurgical treatments.

Back: Spinal stenosis is the most frequent reason for low-back surgery in patients over 65. But until SPORT—a seven-year, $21-million, Dartmouth-led study—no one knew for sure if it was the best choice because the options had never been tested in a large randomized trial. The same was true for two other common back conditions: herniated disk with sciatica and a variation of spinal stenosis, where one vertebra has slipped forward over the other, called spondylolisthesis. SPORT showed that surgery helped to varying degrees for those conditions, too (see **dartmed.dartmouth.edu/winter06/html/disc_papers.php** and **dartmed.dartmouth.edu/fall07/html/disc_sport.php** for details).

"For the first time," says James Weinstein, D.O., SPORT's principal investigator, "we have an evidence base on which to advise our patients."

Data: In the stenosis study, 289 patients were randomly assigned to surgery or nonsurgery groups, and 365 could choose to have surgery or not. As the study went on, many randomized patients changed their minds; some assigned to have surgery decided against it, and vice versa. So the paper, published in the February 21 issue of the *New England Journal of Medicine*, contains a dizzying array of numbers.

But the bottom line, says coauthor Tor Tosteson, Sc.D., is in the "as-treated" figures. When the data was analyzed according to what treatment people actually got—surgery or nonsurgical therapy—they found that surgery reduced pain and improved physical function much more. On a 100-point scale, surgery patients' pain improved an average of 28 points (compared with 12 points for nonsurgery patients). And surgery patients' physical function improved an average of 25 points (compared to 10 points for nonsurgery patients).

Surgery reduced pain and improved physical function much more.

Gain: Those treated nonsurgically—with physical therapy or pain medications, for example—improved, too, just not as much as those who had surgery. "Any gain over 10 points on this scale seems to be worth it," says Tosteson. For instance, 10 points can mean being able to lift a bag of groceries or not.

Weinstein and his coinvestigators have now launched a website using data from the stenosis trial to help patients (and doctors) estimate the benefits of surgery versus nonsurgery. There is still a lot of uncertainty, says Tosteson, because people's health histories can vary widely. But for those with spinal stenosis, the decision of whether to go under the knife now has a little less guesswork and a little more evidence behind it.

JENNIFER DURGIN

MARK WASHBURN

Weinstein is the principal investigator for SPORT.

Staying abreast of the news

Magnetic resonance imaging (MRI) just isn't worth it as a breast-cancer screening tool for women who have already had a lumpectomy and radiation therapy. That's the conclusion of Dartmouth researchers from the Departments of Surgery and Radiology. After analyzing the records of 471 women who received standard care, they estimated that the total cost of using annual MRIs to detect recurrences would have been more than $7 million. "A total of 2,570 MRIs would have been performed," they wrote in the *Annals of Surgical Oncology*, "but these would have been unlikely to change the therapy or survival of any of our patients."

A big-hearted mouse

When the Grinch's heart grew three sizes in one day, he must have grown a lot of new blood vessels, too—at least according to a recent finding made by the lab of DMS's Michael Simons, M.D. By manipulating a gene in a mouse, the researchers discovered that vessel density controls organ size. "An increase in the size of the vascular bed in the normal heart," wrote Simons and his coauthors in the *Journal of Clinical Investigation*, "leads to increased cardiac mass and . . . increased cardiac performance." In other words, more blood vessels result in a larger—and more powerful—heart.

Weighing what makes a difference

When does a difference make a difference? That's a question that Samuel Finlayson, M.D., and Ian Paquette, M.D., have been pondering, in the context of a recent finding that urban patients tend to be diagnosed with cancer later in the disease's progression than do patients who live in rural areas.

Age: "The difference between the two populations was statistically significant, but the absolute difference was not that great," says Finlayson, an associate professor of surgery. He and Paquette, a resident in general surgery, published their finding in the *Journal of the American College of Surgeons*. They based their study on data collected by the National Cancer Institute about cancers of the colon and lung and controlled for factors such as age, race, gender, marital status, income level, and level of education. Though the difference between urban and rural patients wasn't great, the researchers were interested in the the direction of the difference, for the previous assumption had been that rural patients were diagnosed later.

"What's more notable," Finlayson adds, "is the very high proportion of people presenting with late stages [in] both populations. That's probably the more striking finding—even more striking than the small differences that we saw between the urban and the rural populations."

Colon and lung cancers are "both very common cancers and major health problems in the U.S.," points out Paquette. The two cancers, says Finlayson, "can both present at a very wide range of stages, with very different prognoses."

Colon cancer, when identified early, can be surgically removed; such patients have an average five-year survival rate of greater than 90%. But when it's found at a more advanced stage, colon cancer is virtually incurable. That's why regular screening for colon cancer is highly recommended, especially since the disease's risk factors are not very well understood.

The difference was statistically significant, but "not that great."

For lung cancer, on the other hand, there is no nationally recognized screening recommendation, though an effort to identify one is currently under investigation in a major multicenter trial (DHMC is one of the participants in this trial). Also, there is a clear and undisputed risk factor for lung cancer—smoking—unlike colon cancer, where most cases are sporadic.

"There is certainly a need for a cost-effective way of screening for lung cancer," says Paquette. But, adds Finlayson, "if we were to allocate resources to screen for lung cancer or to prevent smoking, it would probably be more cost-effective to prevent the disease in the first place than to try and find [tumors] after they have evolved."

Inquiring: So a study that was designed to assess whether there's an urban-rural diagnosis differential led to ruminations on the effectiveness, and cost-effectiveness, of screening methodologies. Maybe that's a mark of truly inquiring minds. TINA TING-LAN CHANG

JON GILBERT FOX

Herndon is concerned about a proposal to expand the definition of osteoporosis.

Definition is a bone of contention

Millions more women could be needlessly treated for osteoporosis under new guidelines recommended by the National Osteoporosis Foundation (NOF) and the American College of Obstetricians and Gynecologists (ACOG). A DMS study in the journal *Health Affairs* suggests that the new disease-definition guidelines would come at a net cost of $46 billion.

Osteoporosis makes bones more fragile and likely to break. It affects 44 million Americans, 68% of whom are women. One in two women and one in four men will have an osteoporosis-related fracture during their lifetimes. DMS internist Brooke Herndon, M.D., who led the study, agrees that osteoporosis is a major health concern, especially for postmenopausal women. But she says there's no evidence that treating more women will reduce the number of fractures.

Eligible: Under current guidelines, established by the World Health Organization, 6.4 million women aged 65 years and older and 1.6 million women aged 50 to 64 are eligible for drug therapy. Under the new guidelines, the number of treatment-eligible women would jump to 10.8 million in the 65-plus group and 4.0 million in the younger group. "Expanding disease definitions . . . always means that the number of affected people rises," wrote the DMS authors (who are all members of the VA Outcomes Group in White River Junction, Vt.). "But this group of newly identified patients is at lower risk."

"There's a lot of confusion in clinic, because almost every woman over the age of 50 has low bone density," says Herndon. "What's so low that you should do something about it? What level of 'low' actually matters to the patient?"

The authors pointed out that expanded disease definitions can mean larger markets for pharmaceutical firms and that WHO, NOF, and ACOG all receive funding from the drug industry. They would like to see an "independent organization, such as the Institute of Medicine, review the evidence and develop an unconflicted definition of osteoporosis requiring treatment." LAURA STEPHENSON CARTER

DMS's David Goodman, M.D., found a 200% difference in physician supply across the U.S. For every physician who has moved to a low-supply area, four moved to a high-supply area.

Assessing old and new, with cost in mind

Most women probably don't even know if their mammogram is recorded on film or digitally, says Anna Tosteson, Sc.D. But the transition from film to digital imaging has raised questions—including whether there's a difference in the cost-effectiveness of the two methods.

A professor of medicine at DMS, Tosteson recently looked into that question in concert with the American College of Radiology Imaging Network (ACRIN). In phase one of the organization's Digital Mammography Imaging Screening Trial (DMIST), other ACRIN investigators focused on the clinical effectiveness of the two methods. In DMIST's second phase, Tosteson's team added cost to the analysis, to see if digital mammography's higher cost could be justified by better health outcomes.

"Some consideration ought to be given" to technology's value.

Phase: The first phase showed that in younger women and women with dense breasts, digital mammography resulted in more screen-detected cases of cancer and fewer projected deaths from cancer. But digital was no better than film in women over 50 with non-dense breasts.

Tosteson's multisite team revisited the records of the nearly 50,000 asymptomatic women involved in phase one. They entered findings from the two types of mammography, and the $50 higher Medicare reimbursement for a digital screening, into a University of Wisconsin breast cancer model that simulates women's life history. This yielded a cost per quality-adjusted life-year (QALY) for each method; the numbers were run for the entire sample, as well as for the age and breast-density breakdowns.

FLYING SQUIRREL GRAPHICS

There are big differences in the cost-effectiveness of film and digital mammography, Tosteson found.

Dense: The results, published in the *Annals of Internal Medicine*, were striking. The cost to use digital instead of film mammography for all women is over $300,000 per QALY gained. If digital is used only for younger women and those with dense breasts, where its effectiveness is higher, it falls to between $26,500 and $84,500 per QALY gained.

Asked what inspired the study, Tosteson explains that "the U.S. spends more on health care than any other country . . . and there's a feeling that part of what's driving [that] are technological innovations. So if you have something big like digital mammography coming along, before it's adopted—fully adopted—some consideration ought to be given to the economic value." The team's goal was not to limit access to technology but to assess its "value . . . if you look at longer-term endpoints like life expectancy or quality-adjusted life expectancy."

Method: About 33.5 million mammograms are done in the U.S. each year, and the digital method appears to be gaining ground rapidly. Tosteson says 20% of all mammograms were digital in 2007, up from only 7% in 2005. (At DHMC, all mammograms are done digitally.) Why make the digital–film comparison when film seems headed for obsolescence? The ACRIN paper pointed out that it's important "to gain insight into how various population subgroups are affected by technologic innovation." James DiClerico

Air line guide

An increasingly popular way to deliver oxygen to infants with respiratory problems may be easier but should not replace the standard method, DMS researchers recently reported in *Pediatrics*. The team measured the pressure inside the mouths of 27 infants as they received oxygen through heated, humidified, high-flow nasal cannula therapy. "Only in the smallest infants with the highest flow rates, with the mouth fully closed, can clinically significant but unpredictable levels of continuous positive airway pressure be achieved," wrote Zuzanna Kubicka, M.D., and her coauthors. "Our results and those of others also raise important safety and monitoring issues."

Organ lesson

Despite efforts to ensure equal access to organs, country-dwellers are up to 20% less likely than their urban counterparts to get a heart, liver, or kidney transplant, concluded a study led by DMS surgeon David Axelrod, M.D. Patients in "rural regions and small towns face multiple barriers to health-care access," wrote Axelrod and his coauthors in the *Journal of the American Medical Association*, "including the need to travel long distances, the lack of locally available specialty services, and difficulty in receiving follow-up care." ■