Organic chemist develops vaccines against *C. difficile* and other disease-causing gut bacteria

STORY BY ANDREW VOWLES | PHOTOS BY DEAN PALMER

THE DOOR'S OPEN TO GUELPH'S SUGAR VACCINE LAB

He never quite leaves the chemistry lab behind. Even as he heads home each day, part of Prof. Mario Monteiro remains in U of G's MacNaughton Building with his studies of carbohydrate-based vaccines. And that part of him is still there the next morning. "I do wake up thinking about the research," he says. "It's not like I close the door."

He's not a workaholic. After hours, you can often find Monteiro rinkside watching son Alex playing hockey or poolside serving as an official at his other son Marco's swim meets. But Monteiro says he feels forever driven to seek out relatively untried remedies for serious and sometimes life-threatening diseases caused by nasty bacteria.

Take *Clostridium difficile*, a gastrointestinal (GI) bug that has garnered plenty of headlines in recent years. In most healthy people, it might cause nothing more vexing than a case of diarrhea. In people with weakened immune systems, it's a bigger problem. It's especially a major and growing threat to patients in hospitals and long-term care facilities. If a patient's normal gut microflora has been wiped out by antibiotics, *C. diff* can overrun the intestinal tract, leading to serious disease and even death.

Short of developing new kinds of antibiotics, how best to tackle this threat? Monteiro thinks the answer lies in vaccines – not just conventional protein-based vaccines but ones that target carbohydrates on the bug's surface. It's not quite using sugar to help the medicine go down but using medicine to zero in on sugars and "take down" disease-causing bacteria.

Working with a team of researchers in two labs, Monteiro is studying sugar-based vaccines for a variety of infectious bugs. Their work with *C. diff* is expected to pay off with one of the biggest licensing deals done by U of G's Catalyst Centre. In preclinical work with scientists at Stellar Biotechnologies Inc. in California, Monteiro showed that conjugate (sugar-protein) vaccines can protect against the organism. This past summer, the company announced that it had acquired a licence from U of G to develop therapies against *C. diff* infection based on the chemist's work.

Under its agreement with Guelph, Stellar paid an upfront licensing fee of more than $300,000 in shares; their value has doubled since the deal went public in late August. No vaccine is ready for public use yet. It takes five to eight years on average for a preliminary vaccine to move through the regulatory approval process, but the company plans to begin human trials this year.

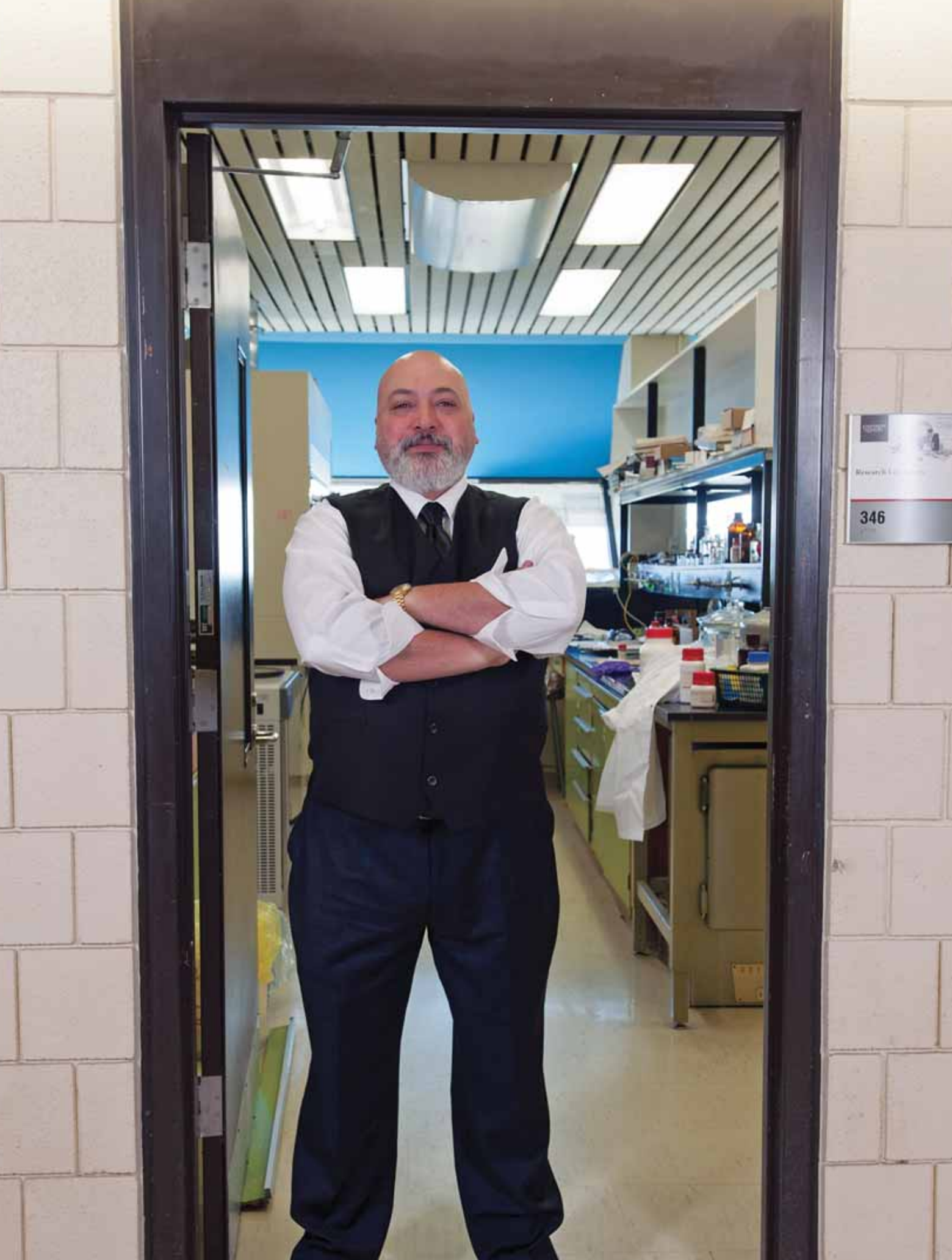
"We expect that this vaccine is going to be a win-win for the University and Stellar," says Dave Hobson, manager of technology transfer at the Catalyst Centre.

C. diff wasn't necessarily Monteiro's main target when he arrived at Guelph in 2004. He had worked for the carbohydrate vaccines division of Wyeth (since absorbed by Pfizer) in the United States before coming here. Earlier, he worked at the National Research Council in Ottawa, where he completed a post-doc with the Canadian Bacterial Diseases Network. He was interested in developing vaccines against a group of bugs that cause a range of GI disorders.

"You sit down and think: How can I use my knowledge?"

Back then, he was looking at *Campylobacter*, another gut microbe that causes food poisoning and neurological problems. This year, the U.S. Naval Office of Research will begin trials with a sugar-based vaccine for *Campylobacter jejuni* developed here in Monteiro's lab.

With these and other bacteria, he's taking a little-tried approach. He's making vaccines based on carbohydrates or carbohydrate-protein complexes. All vaccines provide disease immunity by priming the body's immune system to "remember" a microorganism during later infection. Most are made from weakened or killed forms of the bacteria, or from toxins or surface protein antigens. Less common are conjugate vaccines that include polysaccharides, or complex chains of sugars on the bug's surface.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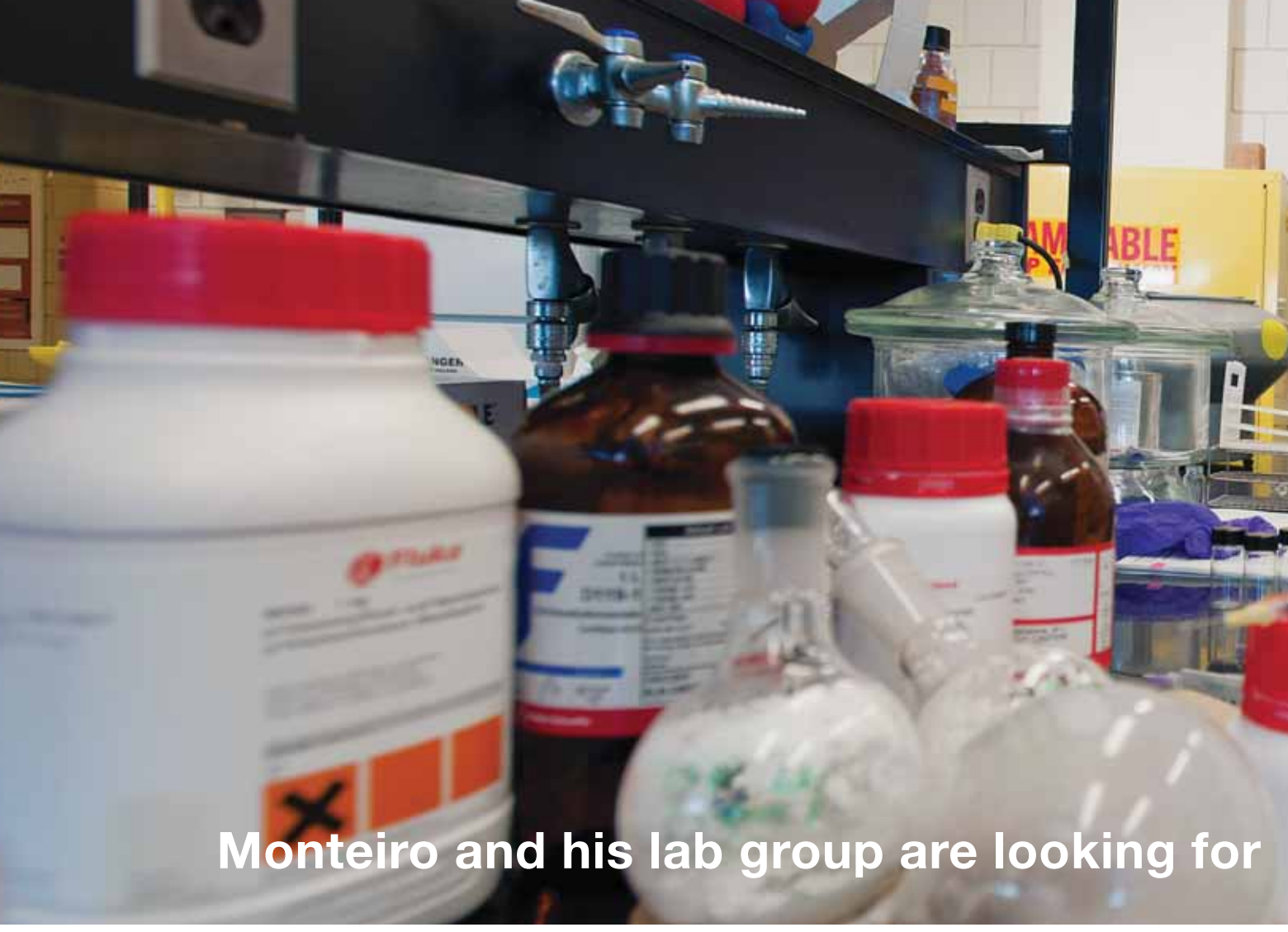

Monteiro and his lab group are looking for

That sugars are immunogenic is not a new idea. In the 1920s, American immunologists Michael Heidelberger and Oswald Avery found that pneumococcal polysaccharides were antigenic. Monteiro figures that potential studies of sugar-based vaccines might have occurred then – but along came penicillin.

Today, a growing threat of antibiotic resistance to current remedies has pushed scientists to look for other drugs, including sugar-based vaccines. Few companies make carbohydrate-based vaccines because of their complex chemistry, but Monteiro says that's changing.

Spurred by success with *Campylobacter* and *C. diff*, he and his lab group are looking to new challenges: "What's beyond the next door?"

One challenge has led the team into looking at gastrointestinal problems associated with a different disorder: autism. More than 80 per cent of children with autism suffer from chronic, severe GI symptoms, and 75 per cent of those suffer from diarrhea; currently, doctors use antibiotics to handle most infections. Scientists have found that a relative of *C. diff* called *Clostridium bolteae* often shows up in higher numbers in the GI tracts of autistic children than in those of healthy kids.

Monteiro has looked at this microbe along with Guelph microbiology professor Emma Allen-Vercoe, who cultures hard-to-grow species of bugs in her lab in the Department of Molecular and Cellular Biology. Allen-Vercoe cautions that no one has yet shown that specific GI bacteria cause mental disorders, although research has shown that gut microbiota can affect mood and behaviour.

Last year, Monteiro and PhD student Brittany Pequegnat announced that they had developed a carbohydrate-based vaccine intended to reduce GI symptoms caused by *C. bolteae*. It's the first-ever vaccine developed for gut bacteria common in autistic children.

"An anti-autism vaccine is actually an anti-diarrheal vaccine," he says. "Eighty per cent of kids with autism have digestive issues." Monteiro says it might take more than a decade for a vaccine to work through preclinical and human trials, and even longer before a drug is ready for market.

Adds Pequegnat: "I would hope that it is a step in the right direction and aids in subsiding some of the symptoms of autism spectrum disorders. It would be nice to give them a little more comfort in their lives."

She's the primary author on the *C. bolteae* paper published last summer with researchers from U of G and Stellar Biotechnologies. Pequegnat studied applied pharmaceutical chemistry here for her undergrad, including completing co-op placements under Monteiro's advisement. She had asked him for a reference letter in applying to another university for her master's. Instead, he persuaded her to study vaccines at Guelph.

new challenges: “What’s beyond the next door.”

“I was always interested in pharmaceutical research,” she says. “My best friend was diagnosed with diabetes in Grade 7. It changed how I thought about life. It helped that I was good at math and science.” She decided she wanted to help people cope with illness and improve their lives.

Pequegnat began her PhD this year. “I love being in the lab. I never felt more in the right place than when I was in the lab.”

She says her supervisor is always supportive. She also likes his work ethic, including his focus on practical results. “It’s not about the recognition. It’s about seeing what you can do to help people.” Good science, she adds, “is not for you. It’s for the end result.”

Monteiro echoes the importance of the work ethic. His main peeve: students who are not really interested in research. “When I give them the opportunity, and I don’t see them trying their best: that gets me down.” Like swimmers or hockey players, he says, “young scientists should be dedicated, hard-working. Hockey players play or practise every day. You don’t score goals or become Gretzky without practising.”

Some of that conviction likely stems from his own student days. Monteiro was 14 when his family arrived in Canada from Portugal in 1981. Only a few years earlier, he had been helping to milk his grandfather’s sheep in central Portugal. Then he was taking English as a second language classes and attending high school in Toronto.

He paid for university by working two jobs: serving tables at weddings and banquets on weekends, and cleaning schools on weeknights. “Monday mornings I couldn’t stand by the bench because my feet were in blisters,” he says. “That’s part of my fibre. Even when I used to clean schools, I thought I was the best cleaner there was. There has to be pride in what you do.”

He completed his PhD at York University with polysaccharide chemist Gerald Aspinall. Monteiro chose chemistry partly because he’d done well with the physical sciences in high school and partly because he felt the life sciences would have called for better English than he had at the time. Besides, organic chemistry gave him the chance to draw molecular structures. “I doodle a lot.”

He had considered studying fine art but changed his mind. Monteiro feels that art and science are parallel worlds, both requiring fine observational skills, an ability he says he honed growing up in the solitude of Portugal’s hills.

“People don’t really spend much time thinking about what’s involved in a success story. The stars have to align. It’s about having a vision.” He says the bugs – and the need for inventive ways to stay ahead of them – will always be with us. “There has to be a global need for the solution you’re trying to conjure up.” ■