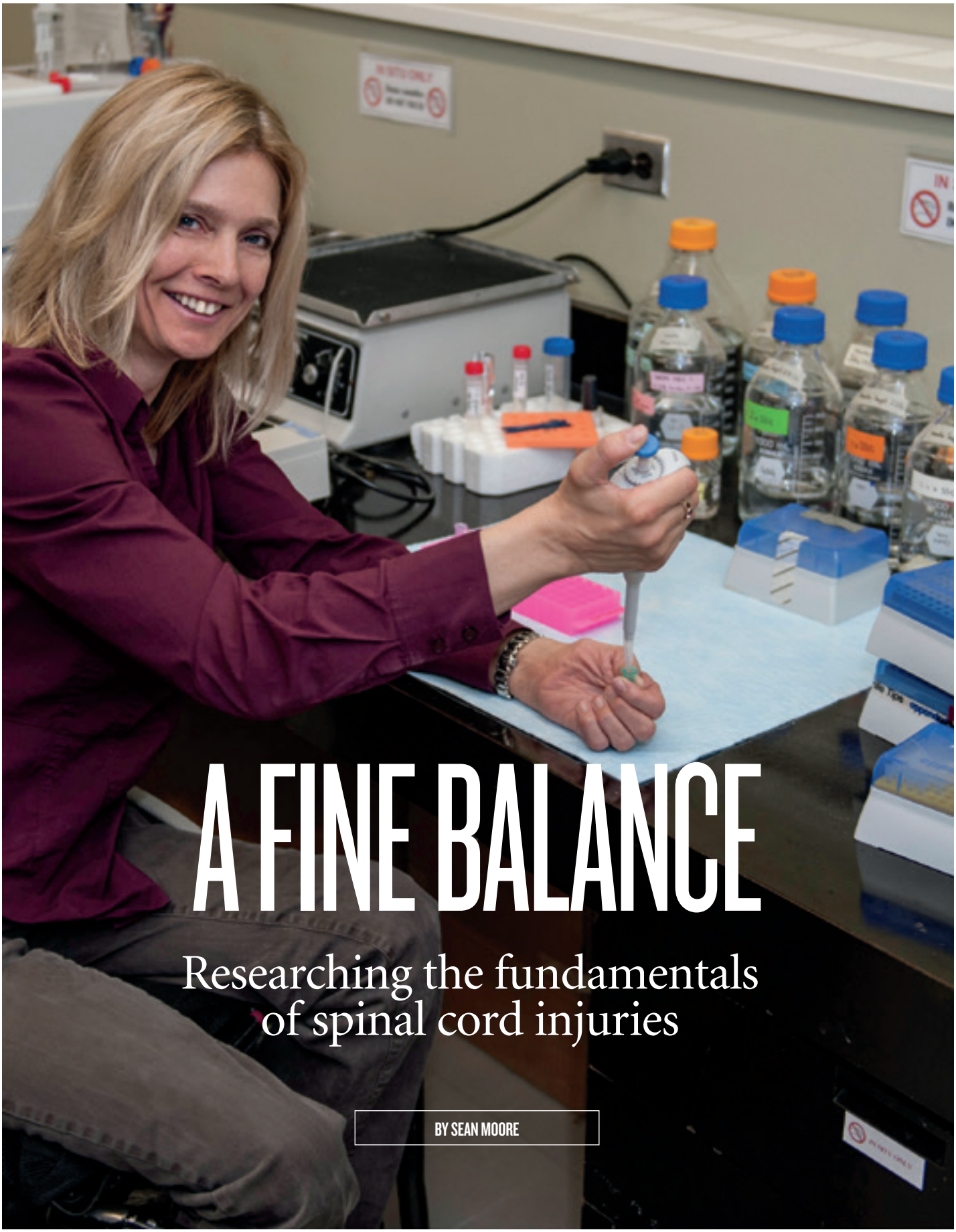
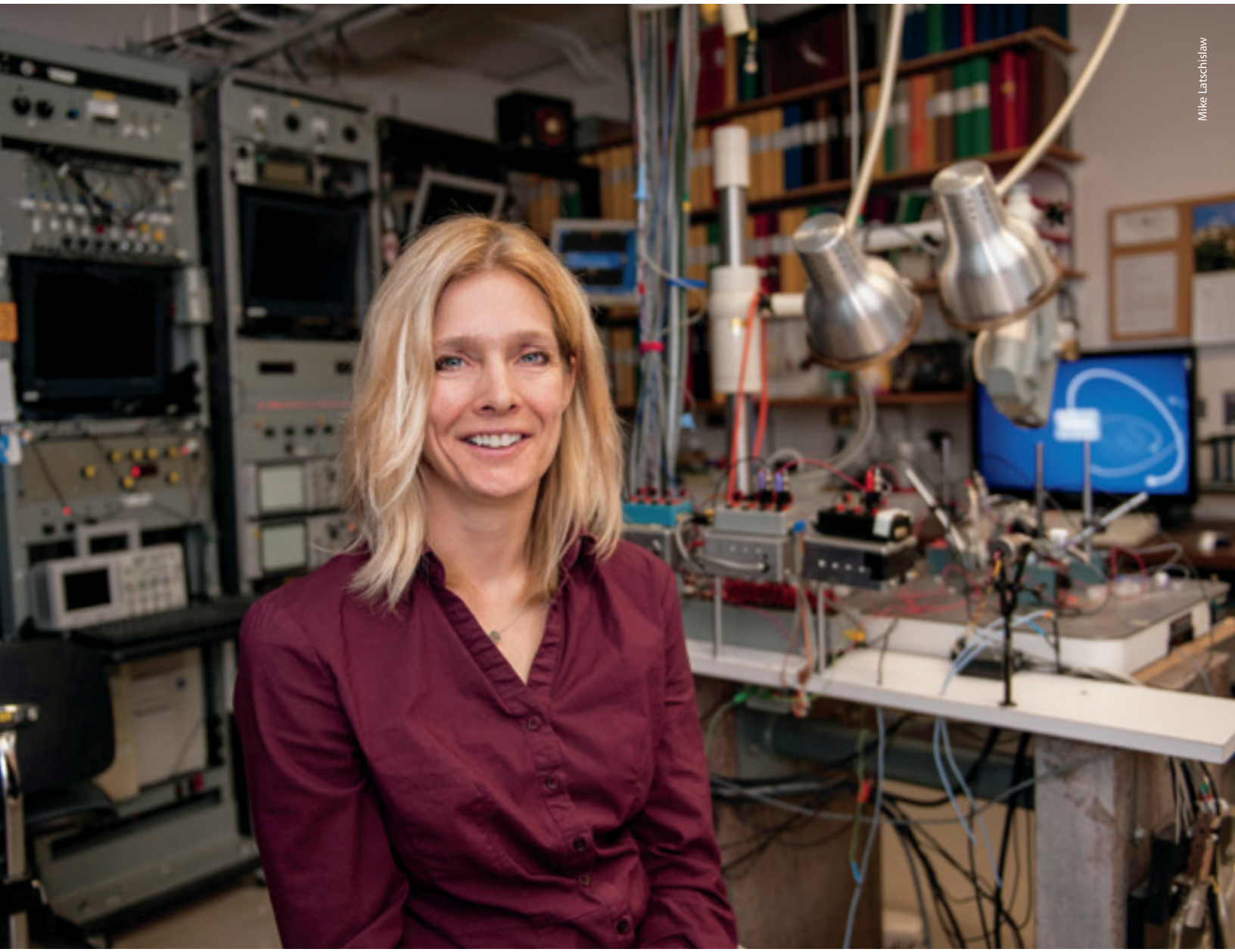


A FINE BALANCE

Researching the fundamentals of spinal cord injuries

BY SEAN MOORE

Mike Latschislaw



Mike Latschislaw

You may be giving the brain more credit than it deserves, at least where walking and running are concerned. When it comes to locomotion, the spinal cord processes information without the brain micromanaging it.

Sometime tomorrow, say when you get up from a chair and walk to get a drink, your brain will send a signal to the spinal cord that says, I'd like to get up and walk over there. Your inner ear—the vestibular system—tells the brain where in three-dimensional space the head is, and the eyes also gather and report data. All of this information descends from the brain and is integrated within the brainstem and ultimately the spinal cord. Simultaneously, muscles are telling cells within the spinal cord how quickly they are expanding and contracting, and the feet and leg joints are reporting on the ground's slope. All this information arrives within the spinal cord, and is integrated within groups of cells called locomotor pattern generators. These cells communicate with each other and then send the appropriate information to motoneurons, to produce the rhythmic motor output we call walking. Whether you are jogging to the bus, prancing through a cocktail lounge, or strolling through a garden, cells within the spinal cord act as a metronome for your steps, telling your muscles the

appropriate pattern of activity needed to do the cha-cha or dash to the bus.

Researchers are only beginning to understand how the spinal cord does this. Even less is known about how cells within the spinal cord function to allow us to balance and remain upright.

How does the spinal cord do all this? That is what researchers at the Spinal Cord Research Centre investigate from many angles. Indeed, this research is something Winnipeg is famous for, but for now only in spinal cord research circles. The centre is in the department of physiology and pathophysiology at the U of M and one of the newest department members is Kristine Cowley, who joined the faculty in 2012 thanks to a Will to Win Classic Professorship. The Will to Win Classic has been supporting spinal cord research for over 30 years and was integral to establishing the Spinal Cord Research Centre where Cowley received her graduate training in neurophysiology. Much of her previous research focus has been on understanding the fundamental workings of the spinal cord on a cellular and systems level, something she says is crucial if we ever want to help people living with spinal cord injury to walk again, or regain their ability to balance and stand.

"We tend to give the brain too much credit in terms of locomotion and balance," Cowley says. "The spinal cord is a centre of integration. And if you want to understand how a rhythmic behavior is produced, you need to start with its simplest modular components and then work outward and upward from there."

To gain insight, we need the approach Cowley is taking —using electrophysiological tools and an *in vitro* spinal cord (a spinal cord removed from its body) to systematically map the pathways involved, and the chemical messengers used, to produce coordinated, balanced stepping.

From an organizational perspective, the spinal cord consists of both ascending and descending pathways, nerves bringing information from the brain to the spinal cord, and from the body and spinal cord to the brain. There are also cells within the spinal cord that integrate information coming in from either the body or the brain; they can influence the activity of motoneurons, which are the neurons that drive muscle activity. Much of Cowley's previous research has been to identify regions of the spinal cord important for generating and coordinating locomotor activity. For example, although the cells that generate and coordinate locomotor-like activity are distributed throughout the entire extent of the spinal cord, including regions related to your legs, trunk and arms, some regions have greater capacity than others.

"Knowing this type of information will be essential if we are to target regeneration or regrowth strategies in the future to recover function after injury," she says.

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In previous research, while working as a research associate in Brian Schmidt's lab at the Spinal Cord Research Centre, Cowley investigated the role of neurons that exist only in the spinal cord, called propriospinal cells. They wanted to know if these cells could relay a locomotor command message from the brain to the neurons activating the legs during stepping. Specifically, if the long descending neural pathways in the spinal cord were severed so there was no continuous neural connection from the brain to the legs, could these propriospinal cells pick up the message at one end, and relay it on to locomotor neurons below the damaged area?

Using an *in vitro* spinal cord in which they could make precise lesions, they cut all direct descending paths on one side of the spinal cord. A bit further down the spinal cord, they cut all the descending paths on the other side of the spinal cord. The only way a message could get from the brain to the leg region would be if the message was relayed through these propriospinal cells. It turned out that after getting rid of all direct descending connections between the brain and spinal cord, and electrically stimulating the brain, locomotor activity could still be produced. So they probed further. Since calcium is needed for synaptic transmission, Cowley removed calcium from the preparation, applied electrical stimulation in the brain stem, and nothing happened. A telling non-event: the propriospinal cells were indeed acting as messengers.

"I'm not someone who says 'wow' in the lab, but this was an interesting finding. We were the first to show this."

Part of her more recent work is seeing what kind of neurotransmitters can be used to activate these cells to increase our ability to get locomotion in both the *in vitro* and the adult *in vivo* research models. Cowley has learned a lot about these cells and how they produce basic rhythm in stepping, but much work remains.

Visit Cowley's office and she can show you a variety of videos showing paralyzed animals and people in harnesses

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or exoskeletons trotting or walking or jerking their legs. News sites report on these findings frequently with headlines like, "Paralyzed rats walk again".

Great. But.

"None of them can balance," Cowley says. "It's a testament to the inherent capacity of the spinal cord to be able to generate steps—but it's a bit of a parlour trick when headlines indicate that voluntary locomotion has been restored in these experimental animals. Even a completely spinalized"—scientifically paralyzed—"animal can produce stepping if you give them training or drugs or electrical stimulation. But none of them will be able to balance and remain upright."

You may come across Internet videos showing people or animals with spinal cord injuries standing upright. But the tools enabling this—epidural stimulators—are crude. It can take up to a year to try to find the correct stimulation to get any response. They can simultaneously activate flexor and extensor muscles, giving the subjects a rigid posture. The researchers don't know what they are activating or where to stimulate to get desired, useful results.

When your brain sends a command for you to stand up, we don't know what pathways it takes—for example, dorsal or ventral. And once you're standing, scientists don't know which areas remain activated in the spinal cord.

"Is there a spinal stance generator? We don't know. There is evidence from animal literature from 40 or 50 years ago suggesting it may exist. But we can use these *in vitro* preparations to understand the neural circuitry that is involved in maintaining an upright posture and responding to balance perturbations."

Understanding this will take some time. And when we do understand it, will it be enough to help someone like Cowley walk again? Maybe not. But once we know the fundamentals, once we understand balance, we can use this knowledge to design therapeutically useful interventions. These interventions may not completely restore function, but they could be used to slow the musculoskeletal decline that is currently inevitable after spinal cord injury.

Every wheelchair-bound person will develop osteoporosis in their legs at some point, and within five to eight years after injury the inevitable fractures begin; reports suggest 50 per cent or more will suffer at least one low-impact or spontaneous fracture in the decades after injury.

"This has huge social and medical costs," Cowley says.

What is more, if people with spinal cord injuries are to ever walk again, their bones must be up to the task.

If you can get people to stand with appropriate stimulation of the spinal cord, maybe you can turn the stimulator on, stand up, reach a few things, and sit down. Or maybe you can turn it on and stand up long enough to put stress on the bones and delay or prevent the bone density loss that leads to osteoporosis.

After Cowley earned her PhD, she began collaborating with others at the U of M like Brian MacNeil in the School of Rehabilitation Medicine. They worked to develop an adult rat model of spinal cord injury to test whether their findings *in vitro* regarding the neural basis of stepping and stance and balance could be translated to the adult. This also enabled Cowley to begin investigating ways to reduce secondary complications related to spinal cord injury. Cowley developed an adult rat model with which to test activity-based therapies: for their potential to reduce osteoporosis and musculoskeletal decline after spinal cord injury. Cowley will soon begin investigating how spinalized rats respond to standing on their hind legs, while supported, on a vibrating plate. Will this preserve bone density? She will be the first to investigate this—other researchers have examined sheep and the results were remarkably positive, but the animals were not spinalized.

By combining the resources within the Spinal Cord Research Centre and the small animal imaging facility at the U of M, teams like Cowley's can investigate these

potential treatments in a controlled, systematic research model that will take mere weeks in an animal model rather than the years needed to determine if the therapies work in humans with spinal cord injury.

She also collaborates with Dr. Karen Ethans, the medical director of the spinal cord injury programs at the Health Sciences Centre, and Dr. Barbara Shay, head of physical therapy at the U of M. Together they are trying to find exercises that people with spinal cord injuries can do that will effectively lower rates of diabetes, cardiovascular disease and obesity in this population. This type of research has an almost immediate potential for clinical applications for the roughly 80,000 Canadians living with a spinal cord injury.

"In many ways," Cowley says, "Winnipeg is the ideal place for both studying how the spinal cord functions, within the Spinal Cord Research Centre, and for more clinical research, working with people living with spinal cord injury. We have a centralized spinal cord injury rehabilitation facility with long-term follow up for the province, as well as connections to community based organizations like the provincial Canadian Paraplegic Association."

Cowley herself is the former executive director of the Canadian Paraplegic Association, a role that has influenced her research career.

Why this research? The motivations are multiple.

Cowley's motivation in researching the role of the nervous system in stepping and balance comes from her general interest in figuring out how things work. After sustaining a spinal cord injury at C8, just after finishing second year university, she realized there were many physical barriers to completing a medical degree, so she decided to focus on research instead. Since she has always been interested in how things work, physiology made sense.

As executive director of the Canadian Paraplegic Association, she observed the many hundreds of people living out the decades after injury dealing with these multiple and currently untreatable secondary consequences of spinal cord injury. That experience motivated her in her work to reduce muscle deterioration.

Her interest in the effect of exercise on spinal cord injury comes from years competing as a Paralympic wheelchair track athlete, and seeing the benefit of training on those living with spinal cord injury.

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"What's interesting is if you survey people with spinal cord injuries, they don't rank walking as their number one thing. Balance ranks higher than stepping in terms of 'what are the things each person would like to recover.' And if you think about it, it makes sense because balance affects everything we do."

Would she prefer balance to stepping?

"Yes, I'd like to stand up. But that's not motivating this. This is motivated from a number of years of looking at locomotor research and saying, 'Well, what's missing? What do we need to do?'"

She looks at her computer screen again, saying, almost to herself, "We can generate stepping, but how are we going to remain upright?"