Guo-Qiang Bi's work shows us that forgetting helps us remember.

## TIMING IS EVERYTHING WHEN IT COMES TO LEARNING AND MEMORY

BY JOE MIKSCH

# FORGET IT?

You catch a whiff of something sweet. You taste bitter coffee. Your eyes take in a towering oak. Your ears hear the teacher tell you “A squared plus B squared equals C squared.” You feel the softness of a velvet Elvis.

Moments later, the input is gone. Yet after the stimulation stops you can conjure up the scent of honeysuckle, the bitter Folgers, the lush tree, the Pythagorean theorem, and the smooth visage of Elvis. You can do this seconds later, hours later, years later. You’ve learned and remembered. So, how the heck did that happen? Well, clearly, your senses gathered information, nerves pumped that stuff into your brain, and it hung around up there in your noodle, available for recall. Simple. Next question.

What? You want to know how that happens? You want to understand how you learned that cloying scent, inhaled for a period of time, was honeysuckle in the first place, and you’d like to know how that odor went from being a bunch of molecules in your snoot to some kind of neuro-something or other rattling around inside your head—what we call a memory? Boy, you’re demanding, but here goes.

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In the mid-1920s, aspiring novelist Donald Olding Hebb was swayed by practical concerns to assume another vocation. A native of Nova Scotia, Hebb graduated from college and became a school principal in Quebec. Seemingly settled in for a nice, comfortable life, Hebb kept his mind sharp by taking graduate classes in psychology at McGill University in Montreal. By 1936, Hebb had not only quit the life of a secondary school administrator, he had earned his PhD in psychology from Harvard University, having taken a particular interest in how brain injury affected intelligence and behavior.

Then came the book. In 1949, Hebb published *The Organization of Behavior: A Neuropsychological Theory*. He intended to posit a "general theory of behavior that attempts to bridge the gap between neuropsychology and behavior." In doing so, the psychologist paved a path that neurologists still tread today, a path that leads from chemical and electrical interactions between neurons to learning and memory.

Hebb proposed that an external stimulus causes a signal to be fired across a synapse, and that when this action is repeated, the synapse is strengthened, making it more likely to stimulate the next neuron to fire. Neurons firing in a series—"cell assemblies," Hebb called them—form a circuit. The brain has then established a pathway in response to a unique stimulus. This is learning. And it is in such circuits where extremely short-term—"working" or immediate and instantly accessible—memory exists, in reverberating cell assemblies, before being transferred to long-term memory.

Hebb laid down the theory, and investigators today are still laboring to reveal the underlying mechanisms that guide what Hebb intuited, notably the University of Pittsburgh's Guo-Qiang Bi.

Bi has figured out quite a bit about how we're able to remember or forget.

He is an assistant professor of neurobiology and member of the Center for the Neural Basis of Cognition. Other scientists describe Bi as self-effacing, humble, and quiet. A visit to his office reveals these adjectives to be apt. It also suggests Bi may be the owner of the most jumbled dry-erase board—red slashes here, blue words there, black scribbles on top of it all—in the land.

As he eagerly explains his work, Bi pops up from his chair, erases a portion of the board with his balled-up hand, draws a pair of neurons, sits down again, jumps up and, poking at the board with a marker, draws neurotransmitters crossing the synapse pointillist-style. Then he sits down again. Sort of a neurology aerobics routine.

From his seat (for a few moments at least), Bi explains that scientists have been exploring Hebb's postulate since it was first published. It is generally accepted, Bi says, that learning and memory take place on the synaptic level.

Increases in neurotransmitters and receptors spurred by neural activity make synapses stronger and the neurons on either side more likely to fire. It is this process that allows information to be stored. But from Hebb until the late 1990s there was a dearth of quantitative information regarding how exactly synapses strengthen—how things happen, in particular, to cause this physical change.

When we learn, Bi says, information doesn't typically come at us in static form. Information flows in time. Consider music: Information is encoded in the sequence of the notes, written down on paper. When a musician plays notes, that information, the tune, comes to us throughout the duration of the song, a function of time.

"For that information to be remembered in the brain, there has to be a conversion from time to space because storage is spatial in the synaptic connections," he says.

"The question is, how can the neuron detect the time and convert that into storage?"

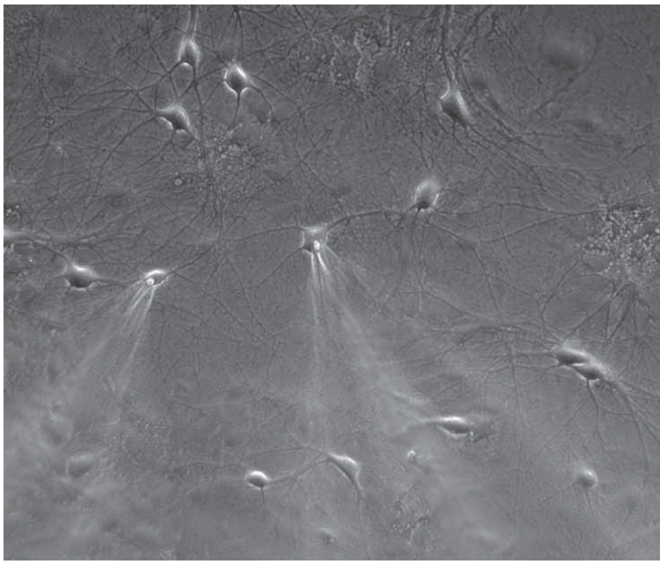
Bi began to answer this question as a postdoc at the University of California, San Diego, in 1998. Seeing how the human brain is home to 20 billion to 50 billion neurons, he thought it was best to start small, so Bi cultured a single pair of neurons from a rat's brain. Bi and his mentor, UCSD's Mu-Ming Poo, building on work published a year earlier by Henry Markram, now of the Brain Mind Institute at the Swiss Federal Institute for Technology in Lausanne, found that for the synaptic connection between the two neurons to be strengthened—for learning to take place—the first neuron must fire within about 10 milliseconds of the second. Otherwise, nothing happens to the synapse. If the second neuron fires before the first, the synapse weakens.

So that's the simplified version of how our brains build connections strong enough for us to remember.

Understanding the process by which synaptic connections weaken is equally valuable, notes Dan Simons, professor of neurobiology at Pitt.

Imagine a scenario in which all the brain's synapses continued to grow stronger. Neural networks would essentially calcify, become so strong and so dedicated to one purpose that learning would cease, Simons says. He adds that Bi's work clarified and expounded upon what others in the field suspected, and Markram began to uncover the year before.

"Guo-Qiang identified some very new and interesting temporal relationships that people hadn't known before," Simons says. "Well, they sort of did, but Guo-Qiang came up with a very careful characterization of these timing relations."

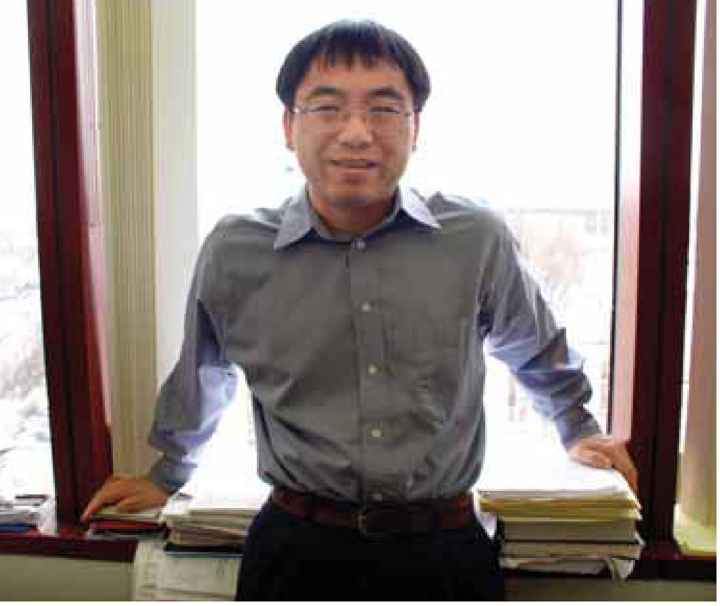


Two glass electrodes (bright spots) record the activity of a network of rat neurons in Bi's lab.

The resulting paper, says one of Bi's former UCSD colleagues, physics professor David Kleinfeld, has become a classic. It led to a more precise understanding of the time factor involved in making memories. The data have been used in more than 300 subsequent papers related to the topic.

"It's very likely that [this] paper will become textbook material," says Kleinfeld.

Two years after that seminal paper was published, Bi found himself recruited to join the faculty at Pitt. He knew he had farther to go with his work and at Pitt he'd have access to what he recognized as one of the largest and most diverse—in terms of disciplines—neurobiology departments in the country. (Since coming to Pitt, Bi has advanced his research by tapping into experts throughout Pitt and at Carnegie Mellon University. "There are quite a lot of resources within walking distance," Bi says.)



Bi says information flows in time.

LEFT: COURTESY G. BI. ABOVE: CAMI MESA.

"The crucial thing with this kind of description is that it's not complete" and is far from representative of how things actually work, Bi says of his 1998 paper.

Simple pairs of neurons and the synapses between them don't account for how we learn and remember. Instead, we use discrete and ever-changing networks of multiple neurons.

Bi had figured out the precise timing of inputs required to strengthen or weaken the synaptic connection between one pair of neurons. Now it was time to find out how the simple rules he put forth applied to more realistic neuronal networks. He cultured small networks of rat hippocampus neurons in a dish and set about stimulating the network. With adequate stimulation, one cell fired the next and the next and the next. An interesting thing happened when Bi removed the external stimulation. The neurons continued to fire.

There is a reverberation, as Hebb had postulated.

"It won't last forever, but for a few seconds or fractions of seconds, the activity is actually preserved," Bi says. He compares it to your computer's working memory—it's only briefly active.

To translate this residual firing to learning as it is commonly understood, Bi talks about the process involved in remembering a phone number.

We hear the digits once—the input—and a neuron fires. We attempt to cement the number into our memory, so we repeat the number. As we do, the continued input creates a network, a cell assembly, in which this information is stored. The synapses in the network become stronger from the repeated firing, and it is in these synapses where the number—our memory of it at least—dwells. This is long-term memory. Once a strong neural network is established, even a very weak input can trigger the reverberation pattern.

"You remember the phone number, then eventually the person's name will be linked to the phone number. And if you hear the person's name, you can retrieve this phone number by activating this reverberation," Bi says.

After enough repeating, Bi notes, the synaptic network becomes stronger and can be activated.

But quickly after explaining this, Bi throws in a caveat. The above is a mere analogy. To be candid, Bi says, he and others in the field are still, more or less, at the beginning of a long journey toward understanding learning and memory. What transpires in the Petri dish with a limited network of rat neurons may extrapolate to what happens inside our heads. But to really know what's going on in the working human brain, you have to study the working human brain. A tough thing to do.

"What's the mechanism underlying this persistent activity? We don't know," Bi says.

"It's really hard to study this in vivo. There are too many neurons, and we don't know which is activated."

His colleague, Simons, agrees but points out that, well, you've got to start somewhere when it comes to painting a complete picture of learning and memory. And where better than on the most fundamental level?

Bi, Simons says, has started from the bottom and continues to build increasingly complicated physiological systems in the lab. With the basic rules in place, Simons says, Bi's work has great promise.

Here at Pitt, Bi says he's gotten as far as he has thanks to former postdoc Huaixing Wang, graduate students Rick Gerkin and David Nauen, and postdoc Pakming Lau. Next on the Bi lab agenda is the identification and investigation of the chemical processes that mediate what happens in these circuits. Of course, this is no small task either: A legion of molecules is involved in cellular processes. Bi thinks that these chemical interactions are modular, that the perhaps 100 molecules that play a role in detecting and converting neuronal activity into changes in synaptic strength can be grouped together into two or three cohorts. Each cohort, Bi believes, is triggered by a specific type of activity and controls the strengthening and weakening of synapses. In the long term, a precise understanding of how these modules work will help craft what Bi calls "a complete set of rules" regarding the behavior of neural networks.

But what happens when something goes wrong with these networks? Here we can go to cinema for the answer, or at least one plausible possibility.

The 2000 film *Memento* features a character, Leonard Shelby, who seeks vengeance after being assaulted during the murder of his wife. His problem, as the character repeats throughout the movie—"I can't make any new memories." Shelby carries a stack of photos to jog the old ones and tattoos new things he learns onto his skin. The character, Bi says, has a lesion in his hippocampus, a part of the brain rich in neurons, full of connections, and vital for learning and converting short-term memory into long-term memory.

Simons sees potential in Bi's research to assist someone like the fictional Shelby.

"We know that the brain does have capacity for reorganization after brain damage, and we know it has a mechanism to do that," he says. "If we knew the mechanism better—perhaps it relates to something like learning—then maybe we could enhance it either by pharmacological intervention or by physical therapy. This work has very important implications for that."

Likewise epilepsy, in which neurons become too strongly excitatory and fire at the same time. Bi's lab, Simons says, might be able to eventually explain how these abnormal neural patterns end up creating cell assemblies that become highly synchronized and release toxins into the brain.

What else can be gained by establishing the "complete set of rules" that Bi seeks?

Simons sees endless possibilities: tracking how the infant brain is formed by sensory experience, determining why one person perceives a stimulus as threatening and another sees it as benign. All sorts of questions related to the normal development and activity of the brain could be probed with such rules.

Of course, it's not going to be Bi's lab alone that takes neurobiology to this golden moment. If anything is clear after spending time with Bi, it's this: No one, Bi included, is anywhere near unraveling the Gordian knot first presented by Hebb.

But if there ever is a complete set of rules for the operation of learning and memory, neuroscientists will doubtless remember what they learned from Bi. ■