genation up to a distance of <200 μm from the closest capillary (*17*). Because gas exchange in explanted lymph nodes must occur exclusively across the surface capsule, it is likely that a steep gradient in tissue oxygen tension was established, with the lowest concentration being in the deep T cell area. Miller *et al.* used a 95% O_2 gas mixture in their incubation chamber (*3*), whereas Stoll *et al.* used 20% O_2 (*4*). Whether and to what extent differences in methodology between the two studies might account for disparities in T cell behavior remains to be determined.

Although two-photon microscopy is used routinely for intravital imaging of living cells and solid tissues, especially in neurobiology and embryology (*18, 19*), it has found surprisingly little application among immunologists (*3, 20*). This situation is set to change with the new reports because the technology is clearly admirably suited for addressing many important immunological questions. For example, the Bousso *et al.* study on page 1876 of this issue (*5*) opens a new frontier in immunological imaging by using two-photon microscopy to observe how thymocytes run the gauntlet of positive selection in three-dimensional thymic organ cultures. As observed with mature T cells in lymph nodes (*3, 4*), engagement of thymocyte TCRs with self antigen presented (in the context of MHC) by thymic stromal cells enhanced the dynamics and duration of T cell–stromal cell contacts (*5*). It will be important to dissect the signals that govern these striking changes in cellular behavior and to determine how these events shape cellular immune responses both inside and outside the thymus. Undoubtedly, six-dimensional imaging strategies that measure the light intensity, color, and motion in space and time of single immune cells in living tissues (and ideally in live animals) will be a critical tool for probing ever more deeply into the mysteries of immunity.

References

1. E. C. Butcher, L. J. Picker, *Science* **272**, 60 (1996).
2. U. H. von Andrian, C. R. Mackay, *N. Engl. J. Med.* **343**, 1020 (2000).
3. M. J. Miller, S. H. Wei, I. Parker, M. D. Cahalan, *Science* **296**, 1869 (2002); published online 16 May 2002 (10.1126/science.1070051).
4. S. Stoll, J. Delon, T. M. Brotz, R. N. Germain, *Science* **296**, 1873 (2002).
5. P. Bousso, N. R. Bhakta, R. S. Lewis, E. Robey, *Science* **296**, 1876 (2002).
6. C. R. F. Monks *et al.*, *Nature* **395**, 82 (1998).
7. A. Grakoui *et al.*, *Science* **285**, 221 (1999).
8. N. Manjunath *et al.*, *Nature* **377**, 535 (1995).
9. M. Gunzer *et al.*, *Immunity* **13**, 323 (2000).
10. W. Denk *et al.*, *Science* **248**, 73 (1990).
11. H. R. Hendriks *et al.*, *Eur. J. Immunol.* **17**, 1691 (1987).
12. R. E. Mebius *et al.*, *J. Cell Biol.* **115**, 85 (1991).
13. J. V. Stein *et al.*, *J. Exp. Med.* **191**, 61 (2000).
14. J. E. Gretz *et al.*, *J. Exp. Med.* **192**, 1425 (2000).
15. R. T. Palframan *et al.*, *J. Exp. Med.* **194**, 1361 (2001).
16. C. R. Mackay, *Nature Immunol.* **2**, 95 (2001).
17. I. P. Torres Filho *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **91**, 2081 (1994).
18. F. Helmchen *et al.*, *Neuron* **31**, 903 (2001).
19. P. T. So *et al.*, *Annu. Rev. Biomed. Eng.* **2**, 399 (2000).
20. C. C. Norbury *et al.*, *Nature Immunol.* **3**, 265 (2002).

PERSPECTIVES: NEUROSCIENCE

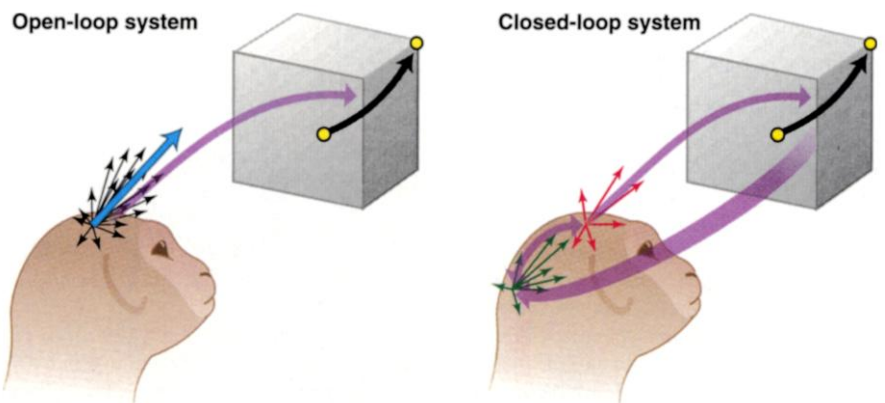
Neurons in Action

Peter König and Paul F. M. J. Verschure

Much of brain research is fueled by the wish to unravel the foundations of thought and action (*1*). Whereas the complex neural processes that underlie consciousness are beginning to be elucidated, the simpler neural circuitry that drives perception and the generation of movement surprisingly remains unclear. For instance, we still do not know whether movement depends on a few localized neuronal circuits or the dynamic state of multiple distributed neuronal systems. Much of the hope surrounding neuroprosthetic devices that translate the activity of neurons in the brain into muscle movements depends on understanding how neurons normally initiate and control skeletal muscles. Toward this goal, work by Taylor *et al.* (*2*) on page 1829 of this issue reveals that the activity of a small group of motor cortex neurons in monkeys can be fine-tuned to carry out complex three-dimensional (3D) movements.

Much of our knowledge about neurons has come from studies of the neuromuscular junction, the point where nerve and muscle meet (*3*, *4*). When parts of the motor system are damaged through injury or disease, repair efforts concentrate on trying to restore the link between neuronal activity and muscle contraction. There have been attempts to achieve this using noninvasive methods, such as electroencephalography (*5*). In this case, recording of the bulk signal of millions of neurons in the motor cortex is analyzed online, resulting in information rates of up to 1 bit/second that can be harnessed to control real-world devices that, for example, dial the phone or switch on the heating.

However, in order to restore speed and fluidity to the motor system, the activity of more neurons needs to be commandeered. The question of exactly how many neurons is crucial for understanding cortical representations of movement in the brain. Experiments using single-neuron electrode recordings in monkeys as they perform a reaching task suggest that movement depends on activation of millions of neurons in a wide area of the motor cortex (*6*). Each neuron in the motor cortex is maximally active for a particular direction of movement, but also fires to a lesser degree in response to movements in other directions. The average of the preferred move-



Think local. Movement of a cursor (yellow sphere) directed by the activity of cortical neurons in the monkey brain. **(Left)** In an open-loop experiment, the preferred direction of movement of a large number of neurons and the weighted average activity of each neuron determines the direction of cursor movement (blue arrow). **(Right)** In a closed-loop experiment (*2*), visual feedback (green arrows) results in adaptation of a small group of motor cortex neurons (red arrows), which are sufficient to accurately control cursor movement. Mapping of the visual cortex onto the motor cortex, and of the motor cortex onto the action performed, is adaptable and can be fine-tuned through learning and visual feedback.

The authors are at the Institute of Neuroinformatics, University/ETH Zürich, Winterthurerstrasse 190, 8057 Zürich, Switzerland. E-mail: peterk@ini.phys.ethz.ch, pfmjv@ini.phys.ethz.ch

ment direction weighted by the activity of neurons then accurately predicts the actual movement performed. This explains many properties of the motor cortex, but also raises several questions. How is the sensory system, with its emphasis on specificity of representation, coupled to the distributed representations of the motor system? How many motor cortex neurons need to be recorded simultaneously in order for real-time movements to be controlled (*7*)?

Taylor *et al.* (*2*) begin to shed light on some of these puzzles. They recorded neuronal activity in the motor cortex of monkeys as the animals made real and virtual arm movements in a computer-generated 3D virtual environment. The monkeys moved a cursor from a central start position to one of eight targets located at the corners of an imaginary cube (see the figure). Their hand movements were represented by two spheres: one being the stationary "target," and the other a mobile "cursor" whose motion was controlled either by the monkey's hand (hand control) or by recorded activity of cortical neurons (brain control). Taylor and colleagues investigated the effects of visual feedback on the monkey's hand movements generated by cortical signals in a closed-loop system. This system differs from the classical open-loop system where animals do not receive visual feedback (see the figure). In the closed-loop system, the monkey's hand movements depended only on the activity of the few recorded neurons, and not on the millions of other active neurons in the motor cortex. Surprisingly, the new study reveals that a few dozen neurons (a tiny fraction of those constituting the motor cortex) are sufficient to generate accurate hand movements.

Intriguingly, the notion of a widely distributed population code is the foundation on which Taylor *et al.* built their study. Yet, their results seem to suggest that effective encoding of motor actions can be accomplished with a very small number of neurons. There are several explanations for this paradox. The first is that learning is crucial. The properties of the recorded cortical neurons became adapted over the course of days, allowing more and more precise control of the cursor. Second, the monkeys relied on visual feedback to fine-tune the properties of the small group of recorded neurons, demonstrating that the visual and motor systems are tightly coupled rather than separate modules. The two new elements in the Taylor *et al.* study—namely, the closed-loop system and visual feedback—are likely to have practical consequences for the design of neuroprosthetic devices. It now seems that only a few recorded neurons, or local groups of neurons, become adapted to actually control movement.

Independently, Brecht and colleagues have shown that injecting a current into a single neuron in the rat cortex elicits a short sequence of action potentials that lead to detectable whisker movements (*8*). Thus, the cortical representations of the motor system may not be that global after all. This is reassuring given the similarities in the anatomical organization of sensory and motor cortical structures. The functional properties of different cortical areas, like their anatomical organization, may turn out to be variations on a common theme.

References

1. A. K. Engel *et al.*, *Conscious Cogn.* **8**, 128 (1999).
2. D. M. Taylor, S. I. Helms Tillery, A. B. Schwartz, *Science* **296**, 1829 (2002).
3. P. Fatt, B. Katz, *J. Physiol.* **115**, 320 (1951).
4. E. D. Adrian, *The Basis of Sensation* (Norton, New York, 1928).
5. J. R. Wolpaw *et al.*, *IEEE Trans. Rehabil. Eng.* **8**, 164 (2000).
6. A. P. Georgopoulos, A. B. Schwartz, R. E. Kettner, *Science* **233**, 1416 (1986).
7. J. Wessberg, *Nature* **408**, 361 (2000).
8. M. Brecht, M. Schneider, B. Sakmann, T. Margrie, *Soc. Neurosci. Abstr.*, in press.

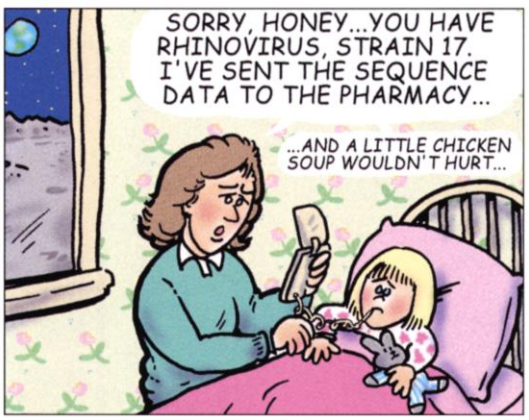
PERSPECTIVES: ANALYTIC CHEMISTRY

Everyone's a (Future) Chemist

Mark A. Burns

Miniaturized chemical analysis systems ("labs-on-chips") (*1*–*4*) have the potential to revolutionize analytical chemistry. A roomful of equipment and several trained technicians may be replaced with a few small, battery-powered devices. These portable systems could make complex chemical analysis available to the untrained, providing individuals with better tools for investigating the world around them (see the figure).

What stands in the way of further advancing this technology? An analogy with the computer industry may help to understand the challenges ahead. The computer industry was born not when the transistor was invented but when versatile and efficient integrating systems on and off the chip were invented (*5*, *6*). For microfabricated integrated chemical systems, one of the biggest challenges has been how to control fluid flow on a micrometer scale. Many individual components now exist, from pipes and fluid channels to the pumps and valves needed for full fluidic control. The difficulty lies in integrating them into the same device with other analysis components.



CREDIT: JOE SUTLIFF

Over the past 15 years, the length scale of fabrication has continued to decrease from the micrometer-scale that is widely used in the production of computer chips to the submicrometer scale (*7*). Recent advances in molecular-level engineering and assembly ("nanofabrication") and soft lithography are expanding the pool of microfabrication tools and materials (*8*, *9*). By combining these and other fabrication or manipulation techniques, simple but powerful components can be constructed.

An example of combining different techniques for component development is given by Terray *et al.* on page 1841 of this issue (*10*). The authors use latex spheres manipulated by optical traps to pump fluids. To fabricate their devices, they use standard channel-etching procedures combined with latex spheres that serve as pump vanes, and optical traps to control the spheres' motion. The beauty of their approach is that they have accounted for future integration into larger, more complex devices: Multiple pumps are controlled on the same device with the aid of a piezoelectric mirror.

A range of other microfabricated pumps, valves, and fluid-handling systems have been reported. Some pumping systems rely on surface tension, exploiting the surface forces that dominate fluid motion at micrometer-length scales (*11*–*13*). For aqueous liquids, electroosmotic pumping provides electronic control of liquid motion. This method has been used exten-

The author is in the Department of Chemical Engineering and Department of Biomedical Engineering, University of Michigan, Ann Arbor, MI 48109–2136, USA. E-mail: maburns@umich.edu