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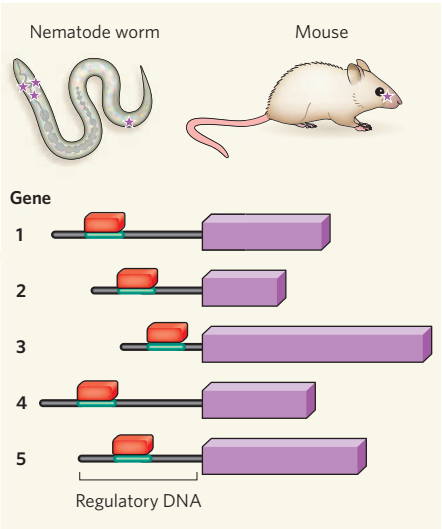

Figure 1 | Orchestrating neurotransmitter expression. Flames and Hobert[1] show that, in neurons of both the nematode worm and the mouse, binding of the same transcription factor (red) to the same short DNA bar-code motif (green) coordinates activation of all five genes (purple) required for expression of the neurotransmitter dopamine. Stars indicate the locations of neurons examined in each species.

down the sequence to a shorter one characteristic of the binding site for the ETS class of transcription factors, and named it the dopamine motif. Of the various ETS proteins they analysed, AST-1 seems to be both necessary and sufficient to activate concerted expression of dopamine-pathway genes by binding to the dopamine motif during development. Moreover, this transcription factor is required for sustained expression of dopamine in mature neurons[1] — an insight that is particularly relevant for a class of neurons the degeneration of which causes Parkinson's disease. So it seems that a simple 'bar code' — in the form of the dopamine motif — specifically and reliably coordinates the expression of genes that drive the differentiation of dopaminergic neurons.

Flames and Hobert[1] extend their findings to mice, thus showing that the regulatory mechanism they describe is evolutionarily conserved. They find that the ETS transcription factor Etv1 is present in dopaminergic neurons in the mouse olfactory bulb. Blocking the expression of Etv1 reduced the number of these neurons, and enhancing its expression increased the number, through the same conserved dopamine motif found in all five mouse dopamine-pathway genes (Fig. 1). What's more, mouse Etv1 could overcome the effects of *ast-1* mutations in the worm, further supporting the conserved function of ETS transcription factors.

Is the mechanism that specifies differentiation of dopaminergic neurons unique? Previous work on similar problems in *C. elegans* set the stage for Flames and Hobert's study. The specification of a group of six mechanosensitive neurons is, for example, governed by the action of two transcription factors that bind to a DNA motif necessary for the development of touch sensitivity[2]. And the identities of the ASE class of sensory neurons and AIY class of interneurons in *C. elegans* are specified by comparable motifs[3,4]. In addition, this general strategy for regulation of gene expression is not restricted to *C. elegans*: in the vertebrate retina, similar regulatory organization determines the fate of photoreceptor neurons through a handful of DNA motifs spread among numerous genes expressed in these cells[5]. What is unique about Flames and Hobert's observations[1], however, is that the mechanism they describe crosses neuronal lineages and spans species, providing cogent evidence for its generality.

It is well established that gene activation can require the binding of combinations of transcription factors to DNA[6]. Does such combinatorial coding mediate differentiation of dopaminergic neurons? Flames and Hobert find that expression of AST-1 in neurons that do not normally express it drives activation of dopamine-pathway genes in some but not all neurons, and only at particular times during development. This result indicates that AST-1 action depends on specific molecular and temporal contexts — a feature reminiscent of the context-dependent expression of 'master genes' such as the *myoD* gene, which can drive muscle development[7], and the *eyeless* gene, which can drive expression of ectopic eyes[8]. Context dependence generally indicates combinatorial coding, and so coding partners for AST-1 are likely to be identified.

Flames and Hobert's data[1] suggest that the conserved dopamine DNA motif and the transcription factor that activates it are key to regulating the expression of this neurotransmitter. More generally, their results support a scheme[9] whereby genes that influence the terminal differentiation of cells — known as terminal selector genes — encode transcription factors that bind to simple, common DNA-selector motifs on cohorts of genes that determine specific neuronal properties. Sherlock Holmes' bemused delight in breaking the Secret Code is surely matched by the satisfaction provided by this compelling demonstration of a shared DNA bar code underlying a crucial aspect of neuronal differentiation. ■

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IMAGING

Nanoscale MRI

P. C. Hammel

Magnetic resonance imaging offers rich three-dimensional pictures, but with limited resolution. Imaging at the nanometre scale has now become possible using highly sensitive force-detection techniques.

Seeing inside a complex object is an invaluable aid to understanding it, so three-dimensional imaging is a pressing objective in areas ranging from molecular and cell biology to investigations of the electronic and structural properties of materials. The challenge is made more difficult by the desire to see the object without altering it in the process. This requires a delicate touch, involving as weak an interaction as possible with the object. But such an approach often conflicts with the need for the high spatial resolution that makes fine detail visible.

Writing in *Proceedings of the National Academy of Sciences*[1], a group led by Dan Rugar reports the success of a delicate yet effective approach to imaging — one that gently excites the nuclear spins in their test objects, particles of tobacco mosaic virus, and records their locations by listening to the spins' oscillating magnetization. An image is derived by recording these signals from a three-dimensional mesh of locations within the object; achieving high resolution requires the mesh to be very fine, so that the volume sampled at each grid location is as small as possible. The authors' sensitive detection of the feeble signals from these elements of nanometre-scale volume, and subsequent reconstruction of the three-dimensional structure of the virus from these signals, marks the arrival of a powerful tool for non-perturbative imaging of a single copy of an object — be it biological, electronic or magnetic — at the nanometre scale.

Rugar and colleagues' approach is inspired by the impressive three-dimensional views of a material provided by magnetic resonance imaging (MRI). Rather than measuring how energetic particles interact with the object

to obtain an image, the technique uses radio waves whose energy is less than a billionth of that of the X-rays used for diffraction studies or the electrons used in an electron microscope. MRI is itself based on nuclear magnetic resonance (NMR), which exploits the intrinsic and plentiful nuclear magnetic spins present in all substances. These nuclear magnets oscillate at a precisely measurable frequency that is determined by fields generated by neighbouring atoms, and by an externally applied field. Hence, these nuclear magnets are embedded, microscopic probes that reveal details of their host's electronic, magnetic and structural properties. Detailed information obtained from NMR has been extensively used for tasks ranging from identifying organic molecules to illuminating subtle features of exotic superconductors.

For imaging, the external field is arranged to vary controllably across the sample, so that the frequency of the nuclear magnetic oscillation will reveal its precise location. This mechanism underlies non-invasive, three-dimensional MRI of regions deep within a sample. Rather than scattering energetic particles, MRI uses low-energy radio waves to excite the nuclear spins so that their oscillation frequency can be measured. A benefit of using magnetic resonance for imaging is that these magnetic resonance signals allow spatially resolved NMR experiments and characterization that enrich the images with detailed microscopic information.

However, the weak interaction that makes MRI so non-invasive is also its Achilles heel: the interaction of the detector with the spin is so small that, in conventional approaches, many spins (10^{12}–10^{18}) are needed to provide a large enough signal to tease out information about the materials. The dimensions of the resolvable volume are limited by the need to detect the weak oscillatory signal of the few spins in the small-volume elements that make up the image. This limits conventional MRI to volumes of several cubic micrometres, and so reduces the usefulness of the technique in solid-state physics, or molecular or cell biology.

In 1991, John Sidles[2] proposed a system for mechanically sensing the weak force that a microscopic ferromagnet exerts on the nuclear magnetic moment in a sample. Tiny forces, he suggested, can be measured by placing the sample under investigation on a compliant cantilever. By observing the slight resulting deflection of the cantilever using, for example, an optical interferometer, extraordinarily small forces can be detected[3]. Force-detected MRI, dubbed magnetic resonance force microscopy (MRFM), has rapidly improved in sensitivity and spatial resolution[4–6]: it has been used to observe a single electron spin[7] and for highly sensitive nuclear-spin detection[8]. MRFM is also a practical materials probe that has been applied to major problems in science[9,10] and technology[11]. Beyond this, it has been shown that techniques used in conventional pulsed NMR are effective for force-detected magnetic resonance[12].

Rugar and colleagues' imaging of individual virus particles[1] is a striking advance in MRFM capability that demanded exceptional detection sensitivity. In particular, the ferromagnetic probe must be brought within tens of nanometres of the cantilever-mounted virus. At these distances, the cantilever experiences many other forces from the nearby surfaces — including, for example, van der Waals forces that are typically thousands to millions of times larger than the nuclear magnetic forces to be measured, and dissipative, electrostatic cantilever-surface forces that produce noise that obscures the signal.

The authors' success is the fruit of a decade of work developing ultrasensitive force-detection techniques. They include excitation techniques[13], which manipulate the spins to produce a distinctive force signal that can be picked out from the background forces, and a nanofabricated antenna[14] that produces a strong radio-frequency magnetic excitation field sufficiently localized that it doesn't disturb the cantilever (the nuclear magnetic forces generate cantilever deflections only at the sub-angstrom level). Finally, the work shows that the noisy signals can be deconvolved into images.

The MRFM procedure will not meet all imaging needs. It is a demanding technique that must be performed in a vacuum and at low temperature. This is a limitation that is shared by electron microscopy of biological specimens, which is nonetheless a highly successful imaging tool. The detection sensitivity of MRFM is improving rapidly, and its history indicates that these capabilities, now at the cutting edge, will soon be routine for MRFM practitioners. But it will be some time before those capabilities can be exploited by the wider microscopy community.

That said, the demonstration[1] of the imaging of viral particles at a resolution down to 4 nanometres heralds the emergence of a new microscope for investigating native biological specimens that will compete with, and complement, electron microscopy and NMR spectroscopy. It uniquely combines non-destructive imaging with the capability of imaging individual copies of specimens such as proteins. The approach is also likely to find wide application beyond biology, in investigations of the chemical and elemental make-up of nanostructures in the physical and materials sciences. ■

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MATERIALS SCIENCE

Nanotubes unzipped

Mauricio Terrones

Nanotubes are single sheets of graphite rolled up into a cylinder. But no one thought that nanotubes could be cut along their axis and flattened out to make such sheets. Until now.

The discovery of buckyballs and carbon nanotubes in the 1980s and early 1990s[1–3] launched the field of carbon nanoscience, and spawned intensive research into the synthesis and applications of these structures. For a long time, it seemed as if the landscape of the carbon nanoworld contained only round objects — spheres and tubes. But in the twenty-first century, flat forms of carbon gained prominence with the discovery of graphene[4] (single layers of graphite) and graphene nanoribbons[5,6]. To realize the practical potential of these newcomers, methods for their mass production are sorely needed. In this issue, two possible solutions are reported — by Kosynkin *et al.*[7] (page 872) and Jiao *et al.*[8] (page 877) — in which nanotubes are 'unzipped' and rolled open to produce nanoribbons.

Graphene is a metal-like conductor, but nanoribbons can generally be either metallic or semiconducting depending on the patterns formed by their edges[5]. Furthermore, nanoribbons less than 10 nanometres wide are expected to be semiconductors, independent of their edge patterns. Narrow nanoribbons are thus excellent candidates for use in electronic devices, such as field-effect transistors, which form the basis of microchips in computers. A thorough exploration of the chemical and mechanical properties of nanoribbons will undoubtedly suggest other applications for these structures, perhaps as sensors, catalysts,