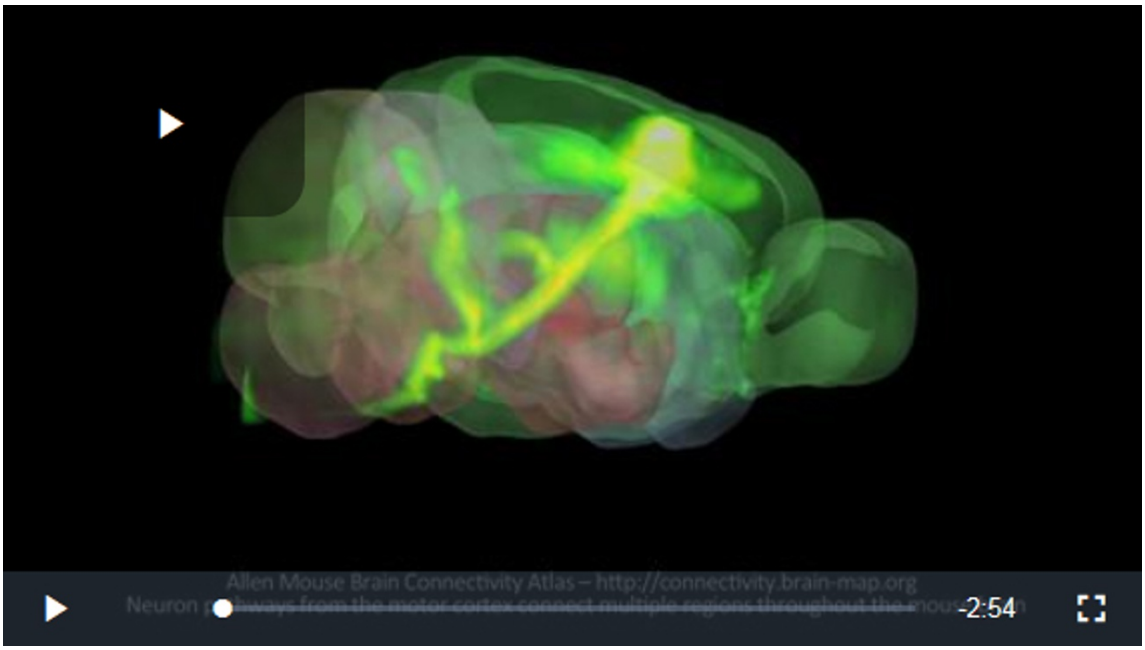
NATURE | NEWS

RNA activity mapped across cells

Technique adds spatial dimension to studies of gene expression.

Brendan Borrell

27 February 2014



Scientists can now take snapshots of where and how thousands of genes are expressed in intact tissue samples, ranging from a slice of a human brain to the embryo of a fly.

The technique, reported today in *Science*[1], can turn a microscope slide into a tool for creating data-rich, three-dimensional maps of how cells interact with one another — a key to understanding the origins of diseases such as cancer. The methodology also has broader applications, enabling researchers to create, for instance, unique molecular 'barcodes' to trace connections between cells in the brain, a stated goal of the US National Institutes of Health's Human Connectome Project.

Previously, molecular biologists had a limited spatial view of gene expression, the process by which a stretch of double-stranded DNA is turned into single-stranded RNAs, which can in turn be translated into protein products. Researchers could either grind up a hunk of tissue and catalogue all the RNAs they found there, or use fluorescent markers to track the expression of up to 30 RNAs inside each cell of a tissue sample. The latest technique maps up to thousands of RNAs.

Mapping the matrix

In a proof-of-principle study, molecular biologist George Church of Harvard Medical School in Boston, Massachusetts, and his colleagues scratched a layer of cultured connective-tissue cells and sequenced the RNA of cells that migrated to the wound during the healing process. Out of 6,880 genes sequenced, the researchers identified 12 that showed changes in gene expression, including eight that were known to be involved in cell migration but had not been studied in wound healing, the researchers say.

"This verifies that the technique could be used to do rapidly what has taken scientists years of looking at gene products one by one," says Robert Singer, a molecular cell biologist at Albert Einstein College of Medicine in New York, who was not involved in the study.

The method hinges on 'fossilizing' the RNA in place in the cell and sequencing it. First, the researchers affix a slice of tissue on a surface and wash away the cellular membranes, keeping the cells' scaffolding, RNA and proteins in place. Next, the researchers add chemicals to 'reverse-transcribe' each short segment of RNA, converting it into circular fragments of single-stranded DNA. Then they

add more chemicals to make hundreds of copies of each DNA circle, which form clusters called nanoballs. These nanoballs are chemically linked together to form a durable, transparent matrix that approximates the original layout of the cell, and then analysed by SOLiD sequencing, a method that uses digital imaging to capture the colours and locations of fluorescent probes as they interrogate the DNA.

The technique has applications beyond understanding gene-expression patterns, says Jay Lee, a medical doctor and biologist in Church's lab. At present, the most advanced technology for labelling and mapping neurons, Brainbow, is limited to 100 simultaneous hues. Lee says that it's now possible to create 1 trillion different molecular barcodes from small strands of RNA. He is also working on techniques to add barcodes to proteins inside the cell.

Lee says that the technique reminds him of a scene from the science-fiction film *The Matrix*, in which the character Neo sees the binary source code underlying his environment. "This sounds a little corny," he concedes, but he adds, "I want biology to be like that."

Nature | doi:10.1038/nature.2014.14787

References

1. Lee, J. H. *et al*. *Science* http://dx.doi.org/10.1126/science.1250212 (2014).