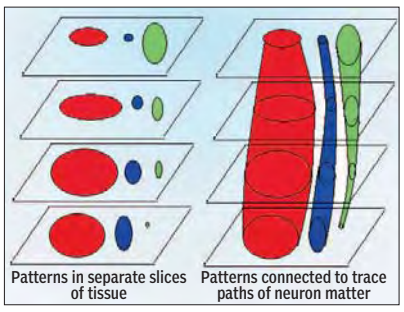
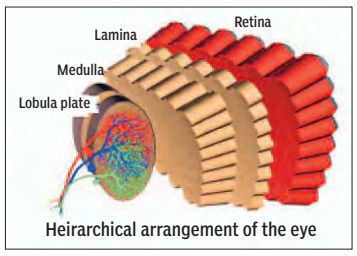
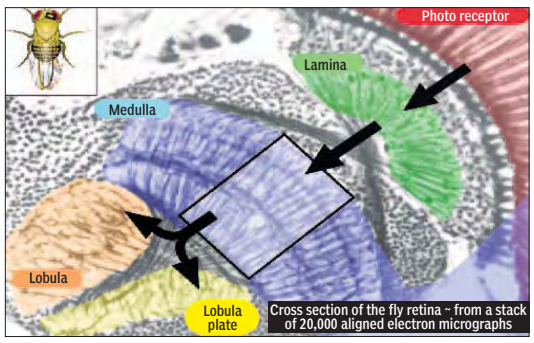
Charting the neural labyrinth

THE FIRST STEPS HAVE BEEN TAKEN TO MAP BRAIN PATHWAYS, SAYS **S ANANTHANARAYAN**

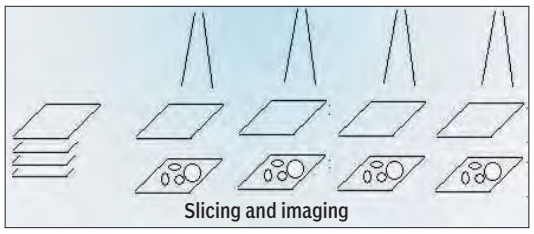




Heirarchical arrangement of the eye



Cross section of the fly retina ~ from a stack of 20,000 aligned electron micrographs


Slicing and imaging

The human brain consists of some 80 billion nerve cells and it works thanks to connections between cell and cell and stronger paths of some connections and weaker paths of others. Mapping such a mass of connections is still quite unimaginable. In fact, given the dimensions and complexity, even the connections of neurons within a small patch of brain matter has not been possible or attempted.

But scientists in Virginia, Halifax, Germany, Austria, Massachusetts, Maryland and London, working in three groups, report in three papers in the journal *Nature* their pathbreaking work in modelling the connections between nerve cells in a small patch of mouse retina, which has revealed the working of different types of cells, closely matching the current understanding which was based on molecular tracing and microscopy of how these cells function.

Two of the groups have built on the work of the first group, to identify the mechanism of the eye in detecting movement. There have been models of motion detection but the steps of neural events, which bring about detection of motion, have not been understood so far. The retina, or the light-sensitive material in the eye, is in fact a part of the brain itself, collecting information of light entering the eye and communicating with other parts of the brain via the optic nerve.

Essentially, the retina is the layer of rods and cones, which are the cells that react when light falls on them, and then the layer of ganglion cells, which transmit visual data from groups of rods and cones to the brain. The eye could thus be considered to be a part processor of the visual data, before the data is interpreted as shapes or patterns by the brain.

Nerve cells communicate by sending an electric signal through a path known as the axon, to the receptors, known as the dendrites, of other cells. The task at hand is essentially to trace the path — axon to dendrite to axon to dendrite — between different kinds of cells involved in the working of the retina, as a sample of brain tissue. But the task is complex, as there are 60 kinds of neurons, closely packed, and 20 kinds of ganglion cells. At the nanometer-scale of the cell-to-cell connections, these paths are not visible in ordinary microscopy. But in ultra thin slices, the structures of different cell material can be readily detected using electron beam microscopy.

Helmstaedter and colleagues studied the path of nerve connection in a patch of 950 nerve cells in a 114 x 80 micrometer area (about a tenth of a millimetre across) of mouse retina using a manual and elaborate but very fine-grained form of layer-by-layer scanning, or tomography. The patch of retina was first sliced into very fine layers. Each slice was then scanned by an electron beam microscope. Once the pattern of cell material in the different layers was identified, the layer images were put together and the original material reconstituted, using computers. The computers first assigned a different colour to each structure in the images of each slice.

They then connected the successive slices to identify the progress of the structures through the depth of the sample, and the computers could identify the connections between cells, the s*ynapses*, along the scan. But to recreate the long branches of the cells, in three dimensions, the computers did not have the required *pattern recognition* capacity.

To help out, the experiment used 300 trained students, for 100 hours each, 30,000 hours in all, to trace the paths of the neurons and with this input the computer could join up the coloured patches and create a three-dimensional model of the neuron paths, from rods and cones to ganglion cells, in the tiny bit of mouse retina.

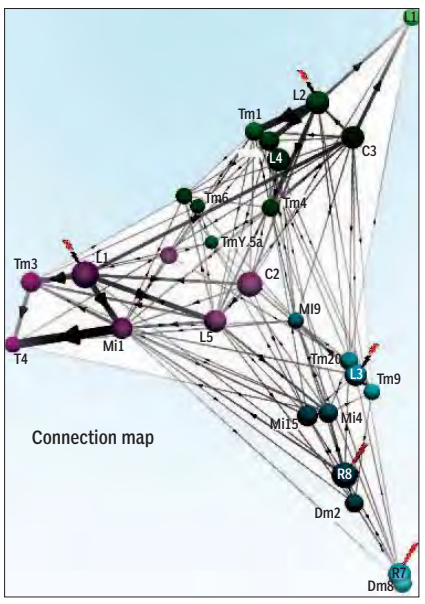
The other groups, of Takemura and colleagues and Maisak and colleagues, studied the detection of motion in the eye of the fruit fly. The fruit fly, which is celebrated for how fast it can detect movement to avoid attack (or being swatted), has the classic *compound eye*, divided into hundreds or even thousands of separate units, called *ommatidia*. The eye is arranged in arrays of six ommatidia, each group of six passing signals, which correspond to one part of the visual field, down to the *lamina*, and a series of nerve bodies, in a *column*.

For detection of motion, what is needed is the change of the image, or signal, coming down in two adjacent columns. Given the complexity of tracing neuronal connections, the study hence looked at a sample of just one column, surrounded by six others. It is known that although photodetector cells by themselves cannot make out motion, somewhere down the line there are cells that clearly indicate motion, up-down or right-left. The directional discrimination is created by neurons called *T4* and *T5*, but they are too small for study of their electrical activity. Maisak and colleagues got around this by genetically coding T4 and T5 cells to change colour when they fired. It was found that the cells formed four groups, which responded to movement up, down, front to back and back to front. And, further, that the cells could discriminate between images with bright edges and those with dark edges, breaking visual data a total of eight ways.

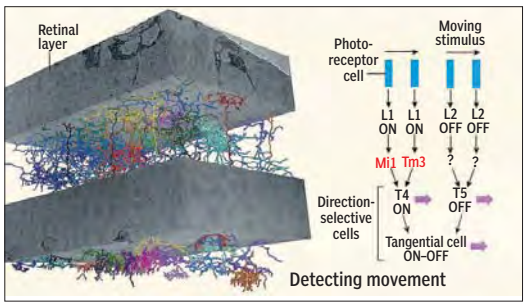
And how do the T4 and T5 cells do all this? Takemura and colleagues found that in the case of T4, just upstream of the cells, there was a pair of neurons, *Mi1* and *Tm3*, which passed down information about points that were narrowly separated in space. Given the nature and timing of the signals, T4 is able to make out the direction of the movement being reported. There is still work to be done, but the Maisak technique of genetically coding Mi1 and Tm3 to respond by glowing in different colours could help discover the last details of how the eye detects direction of movement. But whether the progress made would be able to chart the pathways of the brain itself is an open question. The extent and level of complexity is way above what has been overcome.

Even modest targets would need huge manpower and spending. It is proposed that the work of identifying neural paths through the images of successive slices could be put on the Internet as a game, to make use of public manpower and computing resources. This would create a huge database of basic brain topography and would leave it in the public domain.

THE WRITER CAN BE CONTACTED AT simplescience@gmail.com



Connection map



Detecting movement

PLUS POINTS

800 mph 'trains'

Billionaire inventor and entrepreneur Elon Musk has finally unveiled the "Hyperloop", a proposed public transport system which he claims could take travellers from Los Angeles to San Francisco in just half an hour. The same journey takes one hour by plane and almost six by car. Musk, who published the plans for his ultra-high-speed alternative in a blog post, said it could be delivered to consumers far more cheaply than plane travel, and with the regularity of trains.

The 42-year-old tech mogul inspired Robert Downey Jr's performance as genius industrialist Tony Stark in the movie versions of Marvel Comics' *Iron Man*. He founded Paypal; the high-end electric car-maker Tesla Motors; and SpaceX, a space exploration venture. His stated aim is to establish an 80,000-person colony on Mars.


Elon Musk

Musk first mentioned the Hyperloop more than a year ago, suggesting his invention would be solar-powered, safe and reliable, regardless of weather conditions. He has described the technology involved as a "cross between Concorde, a rail gun and an air hockey table". The concept involves firing frictionless, magnetically levitated pod vehicles through a near-vacuum tunnel, reminiscent of the tube systems traditionally used to transport documents from floor to floor at office buildings and banks.

In an interview with *Bloomberg Businessweek*, Musk said the tubes would be mounted above ground on columns. The pods would move as fast as 800 mph. "The safe distance between the pods would be about five miles," he said, "so you could have about 70 pods between Los Angeles and San Francisco that leave every 30 seconds. It's like getting a ride on Space Mountain at Disneyland." Passengers would experience a G-force no greater than that of a sports car. The ride, he said, would be "supersmooth".

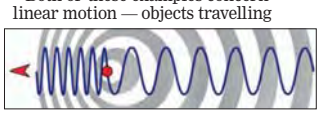
Yet Musk also said last week that the technology remained "extremely speculative", and he was too preoccupied with Tesla and SpaceX to go to work on the Hyperloop himself. Instead, he released the plans so that other firms and interested individuals might develop and improve on them.

TIM WALKER/THE INDEPENDENT

In a spin

Scientists at the Universities of Glasgow and Strathclyde have discovered rotational speed can be determined by measuring the Doppler Shift – the same effect utilised in radar speed guns. The Doppler Shift is a phenomenon everyone is aware of, if perhaps not by name, and is most often experienced by the sound of a siren from a police car or ambulance rising and falling in pitch as it passes by. It is a result of the frequency change due to the position of the observer relative to the source, so each sound wave from an approaching siren is generated a little closer to the observer each time and a little further away as it travels away. The effect is utilised in radar speed guns. When the waves generated by the gun reflect off a car, the change in frequency is detected and speed calculated.

Both of these examples concern linear motion — objects travelling



along a straight path — but scientists at the University of Glasgow have discovered the Doppler Shift can be used to detect rotational motion, too, but only when using "twisted light". Researchers at the School of Physics and Astronomy used a beam of light that had "angular momentum" – or spin – and pointed it at rotating objects. Even though the light hit the object head-on, because the light was twisted like a corkscrew, it was actually hitting the object at a slight angle. The light is scattered and a Doppler Shift is observed. Light without this spin would see no such shift.

Martin Lavery, research assistant who conducted the experiments, said, "Detecting changes in wave frequency cased by the Doppler Effect is a well established method of measuring the speed of an object travelling along a linear path. In our study, we wanted to look at this effect for rotating objects, for example where it could be used to detect speed by looking at the spinning wheels of a car. This research is the first realisation that there is an angular equivalent to the Doppler Effect that allows rotational speed to be determined head-on when using twisted light beams. The research was undertaken by Professor Miles Padgett and Lavery at the University of Glasgow and Professor Stephen Barnett and Fiona Speirits at the University of Strathclyde.

A POTENT INDUCER

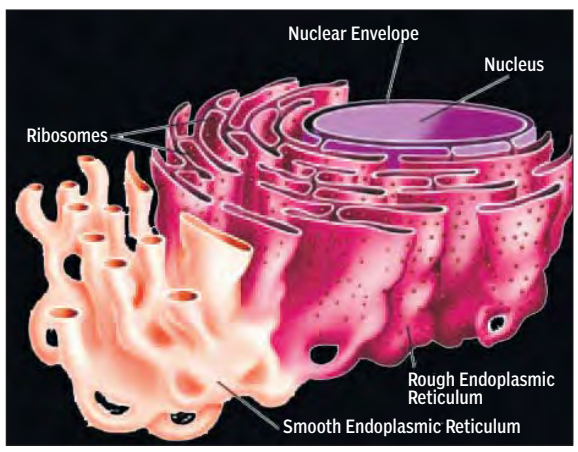
TAPAN KUMAR MAITRA EXPLAINS DRUG DETOXIFICATION

Smooth Endoplasmic Reticulum is involved in several different cellular processes, including drug detoxification. A reaction common to most pathways for drug detoxification and steroid biosynthesis is hydroxylation, the addition of hydroxyl groups to organic acceptor molecules. In each of these cases, hydroxylation depends on a member of the cytochrome *P-450* family of proteins. Members of this family are especially prevalent in the smooth ER of hepatocytes (liver cells) but are also found in lung and intestinal cells. An electron transport system in the smooth ER transfers electrons stepwise from either of the reduced coenzymes *NADPH* or *NADH* to a cytochrome *P-450* protein. The reduced form, *pf P-450*, can then donate an electron to molecular oxygen (O_2), activating the molecule for hydroxylation.

The net reaction is shown in the following equation, where R represents the organic hydroxyl acceptor, and the electron donor is either NADPH or NADH: RH + NAD(P)H + H^+ + O_2 à ROH + NAD(P) $^+$ +H_2O While NADPH is the electron donor for drug detoxification and steroid biosynthesis, NADH serves as an electron donor during the hydroxylation of fatty acids. In addition to NADPH or NADH, the second essential molecule for hydroxylation is O_2. As indicated by reaction, one atom of the oxygen molecule is used to hydroxylate the substrate and the other is reduced to water. Enzymes that catalyse such hydroxylation reactions are called mixed-function oxidases or mono-oxygenases.

During drug detoxification, hydroxylation increases the solubility of hydrophobic drugs in water. This alteration is critical because most hydrophobic compounds are soluble in the lipid layers of membranes and are therefore retained by the body, whereas water-soluble compounds are more easily flushed away by the blood and subsequently excreted from the body.

The elimination of barbiturate drugs, for example, is enhanced by hydroxylation enzymes associated with smooth ER. This can be demonstrated by injecting the sedative *phenobarbital* into a rat. One of the most striking effects is a rapid increase in the level of barbiturate-detoxifying enzymes in the liver, accompanied by a dramatic proliferation of smooth ER. A concomitant effect, however, is that increasingly higher doses of the drug are necessary to achieve the same sedative effect in habitual users of phenobarbital. Further, the mixed-function oxidase induced by phenobarbital is of such broad specificity that it can hydroxylate and therefore solubilise a variety of other drugs, including such useful agents as antibiotics, anti-coagulants and steroids. As a result, the chronic use of barbiturates decreases the effectiveness of many other clinically useful drugs.



Another prominent member of the cytochrome P-450 protein family found in smooth ER and involved in hydroxylation is part of an enzyme complex called *aryl hydrocarbon hydroxylase*. This is involved in metabolising polycyclic hydrocarbons, organic molecules composed of two or more linked benzene rings. While hydroxylation of such molecules can be important for increasing the solubility of toxic substances in water, the oxidised products are often more toxic than the original compounds.

Research has revealed that aryl hydrocarbon hydroxylase can convert potential carcinogens into their chemically active forms. Mice synthesising high levels of this hydroxylase exhibit a higher incidence of spontaneous cancer than normal mice, whereas mice treated with an inhibitor of aryl hydrocarbon hydrolase develop few tumors.

Significantly, cigarette smoke is a potent inducer of aryl hydrocarbon hydroxylase.

THE WRITER IS ASSOCIATE PROFESSOR AND HEAD, †HEAD, DEPARTMENT OF BOTANY, ANANDA MOHAN COLLEGE, KOLKATA

Nightingales prove it

THEIR MATING SONGS IMPROVE WITH AGE, LEAVING YOUNGER RIVALS FAR BEHIND, WRITES **ROGER DOBSON**

Older male nightingales have perfected an art that would be the envy of men having a mid-life crisis: a trick that makes them more attractive to females than their younger male competitors. Their mastery of successful courtship is achieved with a dazzling array of up to 100 trills a second, far more than their younger competitors can manage, and more than any other investigated bird, according to new research.

That ability, backed up by a sophisticated playlist of about 200 songs, means that they are probably seen as better mates by young trill-seeking females. Singing so many trills at peak frequency requires a lot of physical effort and, as a result, it has evolved as a sign on fitness, say the researchers.

"Females could assess the age of the male singer by the trill rate, and mate preferably with older ones," says zoologist Valentin Amrhein, who led the study at the University of Basel, Switzerland. "This makes sense for the females because older males have more experience with defending their territory or with raising young, and therefore have a better reproductive performance."

The research, being published in the *Journal of Avian Biology*, shows that older birds can come up with 100 trills a second, making them the fastest singers. They also performed about 200 different song types, but the researchers think it is the immediate impact of the trills that is attracting the females. It would take more than an hour for the male to go through his whole song list.

"Since the performance of these sounds is very demanding, the rate at which they can be repeated is limited. Trying to sing rapidly increasing sounds in fast repetition is very hard for us humans as well," says Dr Amrhein. "Singing rapid broadband trills comes at a certain price for the male nightingale, so trilling is a good indicator for mate quality."

Nightingale populations have been in sharp decline in recent decades. Between 1995 and 2009, the British nightingale population decreased by 57 per cent, which, experts say, may be because suitable breeding habitat is becoming scarcer.


Valentin Amrhein

Meaning "night songstress", the nightingale earned its name because it is often heard singing at night as well as during the day. Writers throughout history have attached romantic symbolism to this detail. The nightingale is commonly considered something of a muse for poets, who have drawn inspiration from its melodious song.

Coleridge and Wordsworth evoked the nightingale as the voice of nature itself, while *Ode to a Nightingale* by John Keats depicts the bird as an idealised poet who has achieved the kind of artistic success that the narrator longs for. In *Romeo and Juliet*, the bird's song signifies that love transcends death.

THE INDEPENDENT