Edexcel GCE

Biology

Advanced

Unit 5: Energy, Exercise and Coordination

June 2010

Scientific Article for use with Question 7

Paper Reference

6BI05/01

Do not return the Insert with the question paper.

Turn over ▶

N37096A

1/1/1/1/1/

N 3 7 0 9 6 A

edexcel

advancing learning, changing lives

Scientific Article for use with Question 7

It's All in the Mind

The link between the brain as a physical organ and what we feel in our conscious mind has long been the subject of research, particularly where we appear to be unable to control aspects of mood or behaviour and where normal life is affected. Stress, pain and depression can be explained in terms of nerve impulses and brain chemistry, and the causes of Parkinson's disease are well understood, but finding reliable ways of correcting problems has proved elusive. Understanding more about how the brain works may well lead to new methods for treating such problems.

Dancing Worms and Deep Depression

In a laboratory in Germany, a tiny worm dances to flashes of light. A flash of yellow and it darts forward. A flash of blue and it jerks back. Yellow, forward, blue, back – right on cue every time.

The worm is not a toy or a robot but a living creature. It has been engineered so that its nerves and muscles can be controlled with light. With each flash of blue its neurons fire electric pulses, causing the muscles they control to clench. A flash of yellow stops the nerves firing, relaxing the worm's muscles and lengthening its body once again.

The worm is in the vanguard of a revolution in brain science – the most spectacular application yet of a technology that allows scientists to turn individual brain cells on and off at will. "It's really changing the whole field of neuroscience," says the worm's developer, neurobiologist Alexander Gottschalk at the University of Frankfurt.

One possibility is that the technology, coupled with a method of getting light into the human skull, could create a Brave New World of neuro-modification in which conditions such as depression or Parkinson's disease are treated, not with sledgehammer drugs or electrodes, but with delicate pinpricks of light. In the long term it is even possible that such treatments could be modified to enhance normal brain function, for example improving memory or alertness.

The technology could also lead to spectacular advances in basic neuroscience, allowing researchers to tease apart the neural circuits that control everything from reflexes to consciousness with unprecedented accuracy. "We'll be able to understand how specific cell types in the brain give rise to fuzzy concepts like hope and motivation," predicts Karl Deisseroth, a psychiatrist at Stanford University in California, who is spearheading some of the work.

These new possibilities materialised when neuroscientists finally cracked a long-standing problem in their field: how to take control of individual neurons.

We've known since the 18th century that zapping neurons with a wire electrode triggers an electrical spike. This approach has provided much of our knowledge of brain function: scientists have inserted electrodes into the brains of flies, mice and monkeys, zapped away, and observed what happened.

Neurologists also use electrodes to treat medical disorders. Depressed patients who don't respond to medication are sometimes given electroconvulsive therapy via scalp electrodes, or vagal nerve stimulation using a device implanted in the neck. Tremor disorders such as Parkinson's disease are sometimes treated with deep brain stimulation using an electrode permanently implanted at a specific spot in the brain. DBS is considered to be a more targeted treatment than standard drug therapy, because it directly affects only a small piece of the brain whereas most drugs act throughout the brain.

But electrodes leave much to be desired, both for basic science and therapy, because even where they only stimulate a few cubic millimetres of brain tissue their targeting is still crude. A cubic millimetre of brain tissue contains tens of thousands of neurons tangled like spaghetti. Some are excitatory– when they fire they cause their neighbours to fire too – while others are inhibitory, quieting their neighbours down. The ultimate aim for many researchers is to target only one of these two types, as precisely as possible.

"If you just jam an electrode into the brain you stimulate all the cell types," says Deisseroth. "You get serious side effects because you get the cell types you want, but also the cell types you don't want."

Electroconvulsive therapy has the worst reputation. It cures some patients, but can erase memories in a few unfortunate souls. The author Ernest Hemingway lost much of his capacity for writing after receiving electroshock therapy for depression in 1961. He committed suicide shortly afterwards.

Vagal nerve stimulation, meanwhile, causes side effects such as pain, hoarseness and rapid breathing. And then there's DBS for Parkinson's disease, in which an electrode implanted in a spot called the subthalamic nucleus (STN) quiets the chaotic activity there. Turning on the electrode can cause strange effects the first time around. It electrifies a few patients' funny bones, causing them to laugh uncontrollably. It plunged one patient into hopelessness, causing her to weep until the electrode was switched off moments later.

These hiccups can be remedied on the spot by adjusting the electrode's position, but others are more problematic. Some studies suggest that a few DBS patients become vulnerable to mood swings and suicide over time. Various explanations have been offered, one of which is that the mood changes are caused by the electrode stimulating not only the right cells but also a few of the wrong ones too.

At least one brain imaging study has shown that electrical stimulation of the STN can accidentally turn on nearby nerve fibres that connect to the limbic system, a network of brain areas that controls mood. DBS can cause the whole limbic system to light up with patterns of brain activity that are eerily similar to those seen in depression.

For years, neuroscientists and neurologists have wanted something better. If they could turn on nerve cells one at a time, leaving everything else alone, they'd be well on their way to targeted therapies, as well as decoding the function of the neural circuits that control complex behaviours. "The goal was to modify a subset of neurons and make them sensitive to light," says Gero Miesenböck, a neurobiologist at Yale University. "By shining light, you can then activate only one type of neuron at a time, while leaving the others alone."

Miesenböck took his first steps towards that goal in 2002 and by 2003 was closing in fast. He inserted a rat gene into fruit flies that caused them to make a membrane protein called P2X2 in certain brain cells. When P2X2 binds to a molecule called ATP, it makes the neuron fire as if zapped by an electrode. Miesenböck made the P2X2 neurons light-sensitive by injecting the flies with a form of ATP that is only activated by exposure to light of a specific wavelength.

He demonstrated the power of his technique using a strain of flies engineered to make P2X2 in just two of the 125,000 nerve cells in their brains – the so-called giant fibre cells, which are known to trigger escape responses. When Miesenböck flashed the correct wavelength of light at the flies, the giant fibre cells fired and, sure enough, the flies jumped and fluttered their wings as though taking flight. Flies with no P2X2 did not respond to the flashes.

When Miesenböck's results appeared in April 2005 they caused an instant sensation. Within an hour of the paper appearing online, Miesenböck's phone rang: it was the US Defense Advanced Research Projects Agency wanting to know if his work had possible military applications (he now works with them). A few days later, Jay Leno spoofed the work on the Tonight Show, pretending to steer a buzzing fly by remote control into the open mouth of President George W. Bush.

For all its power, Miesenböck's approach suffered a killer limitation: the need to inject light-sensitive ATP into the animals' brains. This would complicate efforts to use the technology in humans, but around the time that Miesenböck was making flies jump, another team was already on its way to solving that problem.

Deisseroth, then finishing his medical residency at Stanford, also wanted to find a way to activate neurons selectively. As a practising psychiatrist, he had seen first hand the dark side of electrode therapy. He had administered electroconvulsive therapy over 200 times and had also treated patients for depression using vagal nerve stimulation. On a weekly basis he saw the shortcomings: patients who were still depressed but couldn't have their stimulation level increased for fear of side effects. "It's not very satisfying for someone who wants to engineer precise therapies," says Deisseroth. "So the motivation was enormous."

His opportunity arrived in November 2003 when a team led by Ernst Bamberg at the Max Planck Institute for Biophysics in Frankfurt identified a protein that produces electric pulses in response to blue light. Called channelrhodopsin-2 (ChR2), the protein came from single-celled pond algae. ChR2 allows the algae to detect sunlight, which they need to manufacture sugars. Unlike P2X2, ChR2 is inherently sensitive to light, which meant researchers could probably use it in animals' brains without having to inject them with a light-sensitive compound first.

Deisseroth contacted the German team and asked to collaborate. They agreed and by September 2005 they had managed to express the ChR2 gene in nerve cells in a Petri dish. Flashes of blue light stimulated the neurons to crank out electric spikes as though they had been zapped by an electrode.

The next piece of the puzzle fell into place when the team came across another light-sensitive membrane protein, called halorhodopsin (NpHR), which had been isolated from a desert-dwelling microbe years before. Crucially, NpHR does the exact opposite of ChR2: when exposed to yellow light, nerve cells containing the protein are effectively paralysed and briefly prevented from firing. With both ChR2 and NpHR in their tool kit, neuroscientists could at last turn specific neurons on and off at will. All they had to do was decide which neurons to target, and then genetically engineer the animal so that those cells manufactured the light-sensitive proteins.

Gottschalk, who collaborated with Deisseroth, was the first to try it. He chose *Caenorhabditis elegans* as his subject – a species of roundworm 1 millimetre long. He engineered the worms to manufacture both ChR2 and NpHR in the muscles along the sides of their bodies and also in the nerves that control those muscles. By flashing a worm with alternating blue and yellow lights, he caused its body to shorten and lengthen in synchrony with the lights – the dancing worm.

"It's a proof of concept," says Gottschalk. "In principle, any behaviour controlled by neurons could be mimicked by turning exactly the right cells on and off using light."

By turning on neurons that sense touch, Gottschalk has also used the technology to make a swimming worm react as if it has bumped into an obstacle, causing it to change direction. And André Fiala at the University of Würzburg in Germany has trained fly larvae to like some odours and dislike others by switching on neurons in the reward centres and aversion centres of their brains.

Nobody is out to create an army of remote-controlled zombie worms, however. The hope is that by learning to induce behaviours and sensations, the research will shed light on the specific neural circuits that control them.

For example, Karel Svoboda at the Howard Hughes Medical Institute in Ashburn, Virginia, is trying to crack the code that brains use to transmit physical sensations. He inserts ChR2 into neurons in a rat's barrel cortex – a brain area that processes electrical signals or "action potentials" received from the base of the animal's whiskers. He then makes the rats turn either right or left by making them "feel" sensations through their whiskers. These feelings are created by pulses of light flashed into the barrel cortex through a small hole in the skull from an LED mounted on the rodent's head.

"We can turn down the light knob and tweak the number of action potentials and really get at the fundamental question of how many action potentials, in what pattern, can be perceived by an animal," says Svoboda.

In another set of experiments, Miesenböck is searching for the neurons that control key behaviours in fruit flies such as mating, grooming and fighting. He is using "off-the-shelf" strains of fly that have been engineered to make it easy to insert a foreign gene into a specific subset of their brain cells – as little as a few dozen of the full complement of 125,000. Miesenböck has tried several fly strains, engineering them to express P2X2 and testing whether flashing them with light leads to noteworthy behaviours. Results so far have been mixed. "Most of the behaviours that you get are a sort of uncoordinated seizure," says Miesenböck, suggesting that too many neurons were reacting to the light to produce a coordinated response.

This illustrates a major obstacle to using light for studying behaviour: the need to improve genetic engineering so that researchers can target exactly the right neurons with their light-sensitive proteins. That goal is being hotly pursued by Deisseroth, Miesenböck and others.

Meeting this challenge will be key to using the technology to treat brain disorders. The most obvious medical goal would be to restore vision in people blinded by diseases such as retinitis pigmentosa, which kills light-detecting cells in the retina. Zhuo-Hua Pan, a visual neuroscientist at Wayne State University in Detroit, Michigan, has spent several years tackling the problem. Working in a strain of blind mice, Pan inserted the gene for ChR2 into surviving cells in the mice's retina in the hope of converting them into new light-detecting cells.

Amazingly, it seems to work. In his early experiments, published in 2006, Pan found that once the gene for ChR2 had been inserted, retinal cells responded to light by emitting electric pulses. Pan monitored the animals' brains and confirmed that the light signal was transmitted through the optic nerve to the visual centres in the brain. "The signal goes to the visual cortex," he says. "The only question is how well the brain integrates this signal." Pan plans to carry out behavioural experiments to determine whether the mice are actually gaining some sort of useful vision.

Deisseroth, meanwhile, hopes to use light to treat Parkinson's disease and depression. His team is preparing to experiment with the ChR2/NpHR combination in rats with a brain disorder analogous to Parkinson's. They are also gearing up to use it in rats that are considered to be a good model for clinical depression owing to their lethargy, poor sleep, reduced eating and a distinct lack of excitement when offered the finer things in a lab rat's life, such as sugar water. The goal of these studies will be to determine not only which brain areas but also which cell types, need to be switched on or off to produce a therapeutic response.

Deisseroth won't say exactly which brain area he is targeting for depression, but one likely suspect is a sliver of the limbic system called area 25. This is overactive in depressed people and can sometimes be dampened down using antidepressants. In a clinical trial in 2005, stimulating area 25 with an electrode quieted its activity and improved mood in four out of six patients. Deisseroth now wants to work out which cells are involved and target them with light.

Unfortunately, unlike worms and fly larvae, humans and rats have opaque heads, so getting light into their brains presents a challenge. Alex Aravanis, a member of Deisseroth's team, has created a device that delivers light to the brain using hair-thin optical fibres. As a proof of concept, he used it to regulate the twitching of a rat's whiskers.

The biggest barrier may not be getting light inside the skull, however, but the need for gene therapy. Whether you're restoring light sensitivity to a damaged retina or silencing cells in area 25, you first have to get those cells to manufacture a light-sensing protein. This means inserting a foreign gene into the brain. Though clinical trials have dabbled in gene therapy for serious inherited conditions like cystic fibrosis, the technique still hasn't received approval for routine use. Inserting genes into the brain, of all organs, would require powerful justification.

Exactly how powerful is currently being explored. In a recent trial, 12 patients underwent gene therapy for Parkinson's disease. The researchers used a modified human cold virus to insert a foreign gene into the neurons of the subthalamic nucleus, aiming to muffle their disordered chitter-chatter. All 12 patients experienced a reduction in their tremors, according to results presented at the October 2006 Society for Neuroscience meeting in Atlanta, Georgia. The work suggests that genes for ChR2 or NpHR could also be delivered deep into the brain, targeted to locations such as the subthalamic nucleus for Parkinson's disease or area 25 for depression.

Neurologist Helen Mayberg at Emory University in Atlanta, Georgia, who conducted the first clinical trial of deep brain stimulation in depressed patients, is optimistic about Deisseroth's work. "We have to keep safety in mind, but this could have absolutely unbelievable implications," she says. "Time will tell."

Deisseroth firmly believes that the technology will lead to better therapies in humans. In the short term, he remains focused on goals that he believes can be reached through animal studies, such as hunting down the neural bases of airy concepts like fear and motivation. "They probably boil down to the activity of a specific kind of cell in a specific part of the brain," he says. Deisseroth hopes that his tiny spotlights of blue and yellow will one day illuminate those elusive neurons and take mind control into a whole new dimension.

Stressed Out

Most people would agree that certain events in their life, such as bereavement, changing jobs, examinations, or even rush-hour travel in big cities, are stressful. We try to avoid stress, but if we cannot, we must try to adapt to it. This adaptation is sometimes referred to as 'toughening up'. Although stress is difficult to define, we know that both avoidance and toughening up are crucial ways of coping with it.

When we cannot cope, stress can lead to irritability and fatigue, and other more serious disorders, such as gastric ulcers, cardiovascular disease, anxiety and depression. Yet not everyone subjected to severe stress suffers from a heart attack or a bout of depression: some individuals are much more vulnerable than others. Many neuroscientists now suspect that the difference in the ability to cope may lie in biochemical changes in the brain involved in the process of adaptation to stress.

Stress causes the adrenal gland, atop the kidney, to release the hormone adrenaline, but many other hormones, including a related one noradrenaline, stream into the blood as well. Adrenaline and noradrenaline act in a similar way, causing physiological changes in the body well described by the everyday language used to illustrate stressful experiences, such as 'hair-raising', 'spine chilling' and 'cold sweat'. These changes probably prepare us to deal with stress by either 'flight or fight', and are remarkably similar to the physiological signs of fear and anxiety. Indeed, drugs that block the actions of adrenaline and noradrenaline can lessen anxiety.

Adrenaline and noradrenaline are also secreted from nerve cells, or neurons, where the compounds act as neurotransmitters, chemical messengers that enable one neuron to communicate with others. Neurons that use noradrenaline as a neurotransmitter feed into nearly all organs in the body, ranging from the iris in the eye, to the gut and the bladder.

The release of such compounds could explain some medical disorders caused by stress. For instance, we know that a high concentration of noradrenaline in the blood, which occurs in people with tumours of the adrenal gland, can fatally damage the muscle of the heart. Stress also increases the levels of this neurotransmitter/hormone in the bloodstream, and could have a similar effect on the heart. The causes of psychiatric disorders, however, are more likely to lie in the brain, so psychopharmacologists have tried to find out how stress alters the way the brain works.

Most studies of the effects of stress on the brain and spinal cord (the central nervous system, or CNS, for short) have looked for changes in the limbic system. This is a collection of regions in the brain involved in the control of emotion and motivation. A network of nerve fibres connects the various parts of the limbic system, linking regions that regulate the secretion of hormones, for instance, to other areas involved in processes such as decision-making and learning.

The neurons of the limbic system rely on many neurotransmitters including adrenaline and noradrenaline. Neurons releasing adrenaline have only recently been discovered and little is known about their function, but we know a bit more about the neurons that release noradrenaline. One popular suggestion is that they serve as an 'alarm system', alerting an individual to a conspicuous threat. In support of this idea, researchers have shown that various forms of stress increase the activity of these neurons. The more active the neurons, the more noradrenaline they release.

When the stress is repeated, as frequently happens in everyday life, further changes take place. Neurons produce more molecules of the enzymes needed to synthesise noradrenaline, for instance, enabling the cells to manufacture and release more of this neurotransmitter. Such changes, which show that neurons are adaptable cells, may underlie the ability to cope with stress.

Recently, researchers have tried to find out how stress alters the way neurons in the limbic system communicate. By and large, neurons are not physically connected to each other, but rely on their neurotransmitter to act as a messenger to transmit a nerve impulse from one cell to another. When active, neurons release molecules of their neurotransmitter, which diffuse across the gap, or synapse, separating one neuron from another. To attract the attention of the next neuron in the chain, neurotransmitters in the synapse have to bind to receptors, proteins that lie in the membranes of nerve cells. A popular analogy is that receptors act as biochemical locks which are opened (or closed) by neurotransmitter 'keys'. Each neurotransmitter has its own specific set of receptors, and each receptor controls a different set of reactions within the target cell. Some receptors, for example, regulate the activity, or firing rate, of target neurons, while others are coupled to enzymes that control the synthesis or release of a neurotransmitter.

There are two main types of receptor for noradrenaline, termed a- and b-adrenoceptors. Noradrenaline activates both types of receptor, but many drugs act on only one receptor type, having no effects on the other. This suggests that there are 'drug keys' that will fit the a, but not the b lock, and vice versa. A radioactive label attached to such a drug enables researchers to count the number of receptors for noradrenaline in a sample of tissue, simply by measuring the level of radioactivity in the sample. This technique, known as radioligand binding, has revealed some interesting links between stress and receptors for noradrenaline.

The first is that stress affects the number of both a- and b-adrenoceptors. A single exposure to stress produces unpredictable changes in adrenoceptors which probably depend on a range of factors, such as the duration and severity of the stress and an animal's prior experience of that particular form of stress. When the stress is repeated, more consistent changes appear. In particular, the number of b-adrenoceptors in the brain falls. This happens in untamed rats even after stress as mild as that of simply handling them for a minute a day.

The most remarkable aspect of this change is that it seems to require several daily sessions. The process of 'taming', judging from outward signs of 'handling stress' such as tremor and attempts to bite any fingers that stray too near to the 'dental defence system', also takes several days of repeated handling. A reduction in the number of receptors for noradrenaline could underlie this process of taming. That is, the changes in receptors might be a component of adaptation to stress.

We now know how this might happen. When noradrenaline binds to the b-adrenoceptors of a neuron, the cell produces more cyclic AMP, an important compound that regulates a wide range of biochemical reactions inside cells, including the synthesis and release of neurotransmitters. After repeated stress, however, noradrenaline is less able to stimulate the cell to produce cyclic AMP. By binding to b-adrenoceptors and modifying the production of cyclic AMP, noradrenaline has a 'hot line' into biochemical processes regulating the function of target neurons. So stress not only increases the release of noradrenaline, but also affects target cells by making their b-adrenoceptors less sensitive to noradrenaline.

The ability of repeated stress to reduce the synthesis of cyclic AMP is especially interesting because studies with radioligand binding show a similar change in the brain cells of animals given anti-depressant drugs for several days. This finding has been an important milestone in research on antidepressant drugs. Eric Stone, working at the State University of New York in the US, has suggested that antidepressant drugs can help to reduce depression precisely because they produce changes in b-adrenoceptors that mimic the adaptation to stress. It follows from this that depression may be caused by a failure of the mechanisms responsible for adaptation to stress. The theory could account for the link between stress and depression, and for the fact that some individuals are particularly vulnerable to stress.

At this point, we must confront another big problem: what does noradrenaline actually do in the brain? Is the increased release of this neurotransmitter responsible for the unpleasant effects of stress, or does it help us to overcome them? The answer to this question is not clear and scientific opinion is deeply divided.

One school of thought has it that the release of noradrenaline in the brain is directly responsible for the harmful emotional effects of stress. This conclusion is based mainly on evidence collected from experiments on animals where researchers electrically stimulated neurons releasing noradrenaline. This procedure causes changes in emotions and behaviour that strongly resemble anxiety and the response to stress. According to this theory, a reduction in the number of b-adrenoceptors after repeated stress helps to counteract the harmful effects of noradrenaline.

If noradrenaline causes the bad effects of stress, we should be able to abolish these effects by preventing the release of noradrenaline. There are a number of ways of doing this, but the simplest is to use a drug such as 6-hydroxydopamine which, when injected into the brain, selectively destroys the neurons that use noradrenaline as a neurotransmitter. When Philippe Soubrie, working at the National Institute of Health and Medical Research (INSERM) in Paris, did this he found that animals were still vulnerable to stress. Other studies have found that injections of noradrenaline into certain regions of the brain could prevent the animals from developing the symptoms of stress. Findings such as these suggest that the release of noradrenaline itself is not responsible for the harmful effects of stress but is a physiological response designed to overcome them. So the release of noradrenaline may be crucial for the process of 'toughening up'.

Even antidepressant drugs do not work in brains bereft of noradrenaline. Researchers have known for some time that removing noradrenaline from the brain prevents antidepressant drugs from reducing the number of b-adrenoceptors. So evidence is growing that noradrenaline helps to reduce the consequence of stress, probably by reducing the number of b-adrenoceptors. Noradrenaline is certainly not the only factor regulating the number of b-adrenoceptors, however. Other hormones are also probably involved in this process. These include the glucocorticoids (also known as stress hormones) which, like adrenaline and noradrenaline, are produced by the adrenal gland.

Studies such as these suggest that all the changes in neurotransmitters and their receptors caused by stress may be interrelated. Thus, drugs acting on one group of neurons may have far-reaching effects in the brain. If this is the case, an imbalance of the function of different neurotransmitters, rather than a disorder of any one in particular, could cause stress-related disorders. Only when we understand all these processes will we be able to explain why some people cannot cope with stress, while others can.

Pain and Gender

Jon Levine was just testing painkillers on people who'd had a wisdom tooth extracted, when he uncovered rather more than he'd bargained for. The women in his study group found that strong painkillers related to morphine, called kappa-opioids, were most effective at numbing pain. But the same drugs didn't work for the men at all. "In fact, the doses used in the clinical trial made pain worse for men," says Levine, a clinical neuroscientist from the University of California in San Francisco.

He was shocked. "The idea that a therapy that had been around for decades could affect women and men in such dramatically different ways was anathema," he says. "It was such an incredible mindset in the field of pain, missing what had clearly gone on in front of their eyes for years."

It's not an effect specific to opioids, either. Another recent study showed that ibuprofen, a widely used anti-inflammatory drug, can be much less effective for women than for men. Researchers at the University of New South Wales found that when they used mild electric shocks to induce pain in healthy young people, only the men got any relief with ibuprofen. It was only a small study, but still worrying, as the drug is often marketed with women in mind.

It's been five years now since Levine first spotted a sex difference, yet we still don't really understand why it exists. And when it comes to testing or prescribing painkillers, or studying pain, nothing much has changed. Remarkably, even many of those involved in pain research are unaware of these findings. "I myself have never been able to get relief from ibuprofen and now I understand why," says Marietta Anthony, a pharmacologist at Georgetown General Clinical Research Center. "This is very dramatic, and has a direct impact for the clinic."

There have always been playful stereotypes of how men and women suffer pain differently. Women are more delicate—but endure childbirth. Men are stoical—until they see a dentist's chair. But these few studies show there's more to the caricatures than meets the eye. Real differences in the underlying biochemistry of male and female pain are revealing themselves. The differences are also starting to suggest some surprising strategies for sex-specific painkillers.

It's perhaps no surprise that the differences have eluded scientists for so long. Pain is multidimensional and highly subjective, and therefore very difficult to study. It varies with the time of day, age, diet, stress, genetic background, location, past and present injuries, and in women, reproductive status and the menstrual cycle.

But not only that. Only 10 years ago, pharmaceutical compounds were tested almost exclusively on men. Women were left out of tests in case their inconveniently fluctuating hormones messed up the analysis. The testers also feared harming a pregnant woman's fetus, while ignoring the obvious safeguard of a pregnancy test and contraceptives.

Only in 1993 did the US make it a legal requirement for women to be included in clinical trials. According to a recent report, on average, 52 per cent of subjects in large-scale trials are women. This looks like progress—but it's not. This figure includes women-only studies such as those investigating hormone therapies or drugs to treat breast cancer. And when testing medications for diseases common to both sexes, women's and men's results are often still lumped together, burying any differences in a statistical quagmire.

In Britain, things are not much better. The Department of Health advises that gender should be taken into account when determining whether a medicinal product is safe and effective. But how strictly this advice is heeded is anyone's guess.

To Marietta Anthony, who was previously acting deputy director of the Office of Women's Health at the FDA, change is imperative. If a drug works differently in men and women, this information should be displayed clearly on the label. "Side effects and efficacy really are different in men and women," says Anthony, "[and] there may be a very fundamental biological reason."

One of the more obvious biological reasons is that men and women tend to suffer from different disorders, mostly the result of a complex bag of hormones, reproductive status and anatomy. So differences in how women and men report feeling pain have often been dismissed as being solely down to the pain's different origins. But origins aside, there's growing evidence that even when the source is the same, the biochemical signals, nerve connections and the way the brain handles pain are all quite different in the two sexes.

Sex hormones are one reason for the differences in pain perception. Women always cry "ouch" first. Whether it's in the clinic or the lab, using the heat of a small laser, the pressure of a tourniquet or electrodes placed on the skin, women are less tolerant of pain. But women's pain sensitivity also yo-yos throughout the menstrual cycle, and just before a period, pain thresholds take a dive. "There is a view that oestrogen is excitatory and could enhance pain transmission in the peripheral nervous system, the spinal cord and in the brain," explains Roger Fillingim from the University of Florida at Gainsville.

Progesterone has quite the opposite effect: it dampens the nervous system's response to any nasty stimulus. And it's most obvious during pregnancy. When progesterone levels rocket in the third trimester, they induce a state of profound analgesia in preparation for labour. Indeed, these hormonal influences are being turned to medical advantage (see "Make your own Valium"). The rest of the time—when not pregnant—women's tolerance generally remains below that of men.

Levine was one of the first to get an inkling of how sex hormones might be setting men's and women's pain thresholds at different levels. His team found that women consistently reported more severe pain than men after removal of a wisdom tooth. Since inflammation is known to underlie most aches and pains, Levine decided to investigate whether inflammatory signals differed between sexes. He gave oestrogen to castrated male rats, and found their pain tolerance plummeted to female levels. And giving testosterone to sterilised females gives them masculine tolerance. In other words, if you switch the sex hormones around, you switch their pain sensitivity around too.

Looking deeper into the biochemical pathways, he has recently found that sex hormones alter the chemical signals involved in inflammation and tissue repair. The female hormone oestrogen quenches the production of bradykinin—a potent inflammatory mediator that protects injured tissues. He believes these differences might account for the different responses to opioids seen in his trial. "As difficult as it is for many of us to acknowledge differences other than in reproductive function, there really are differences between men and women," says Levine.

Another curious difference caused by our distinct physiology is that—especially in women—the visceral organs "talk" to each other, so that pain in one internal organ can be triggered or enhanced by pain in another. Maria Adele Giamberardino at the University of Chieti, Italy, first noticed this effect in women with kidney stones. She has found that when women have painful periods—a condition called dysmenorrhoea—the typical searing back pain from the urinary tract caused by the stones is much more vicious.

Giamberardino's findings ring true to pain specialists. In the clinic, both men and women who suffer from chronic conditions such as irritable bowel syndrome often also experience fibro-myalgia, headaches and chronic pelvic pain. But this coexistence of painful disorders is greater in females than in males. Giamberardino's hypothesis is that the female reproductive organs are highly interconnected with the other organs, and that pain in one organ may trigger painful conditions in others that have linked nerve supplies. The flipside is that these links could become new avenues for treating pain. By tapping into the same communication channels, treating period pains, for example, might help to alleviate other aches.

Our different reproductive organs can also lead to differences in how our diet affects pain ratings, says Beverly Whipple, a neurophysiologist and obstetric nurse from Rutgers University in New Jersey. She noticed that Hispanic women seemed to experience more pain during labour, and at first she attributed this to culture. "I told my students that these women were just more comfortable expressing their pain." Then she became aware of studies in which neonatal rats injected with capsaicin—the chemical that gives chilli its hot bite—did not experience a certain pain-blocking effect that females normally get when pressure is applied to the cervix. Could a diet rich in hot peppers be interfering with the Hispanic women's natural analgesia?

To find out, Whipple set up a study with Mexican women whose consumption of chillies ranged from one or two a week to three a day. "We found that the women who ate a diet high in hot chilli peppers do not get the pain-blocking effect," she says.

The physiological differences don't stop at our reproductive organs and hormones, however. They run all the way to the brain. In a study soon to be published, Anthony Jones, director of the human pain research group at the University of Manchester, has scanned the brains of people experiencing pain from a variety of natural causes. Although many parts of the brain are engaged when a person is in pain, Jones pinpointed one main area of disparity between the sexes. "Women tend to process pain more in one part of the brain concerned with attention and emotion," he says. He suggests that the experience of pain is bound to differ between men and women. "Women tend to process things in a more affective way," he says. For women, pain depends on how much attention they pay to a tender spot. So when it comes to treatment, for women it may be as important to provide them with distractions, coping mechanisms and psychological care as painkilling drugs.

Distractions may work in a different way for men. It seems to be important for men to act tough in public. In experiments performed at the State University of New York, Fredric Levine and Laura Lee De Simone found that men's pain thresholds soared if an attractive female technician was conducting the tests. Women, however, seemed immune to the charms of hunky men. And according to Knox Todd, a specialist in the assessment and treatment of pain at Emory University in Atlanta, Georgia, the differences make their way into the clinic. "What we see in the emergency department is that males make a public display of stoicism, ask for no pain medication, and keep up a good public front." But their stoicism evaporates as soon as men leave the hospital to go home, he says.

But who wins out in the end? Is having a higher pain threshold good or bad? To women, pain is a wake-up call to sort out the problem before it gets too big. Men, who can put up with more, postpone asking for help until it's too late. Women's prompt action could be at least part of the key to their longer life expectancy. In the meantime, a message to dithering males: stop procrastinating, make that dental appointment, and your niggling shoulder pain might get sorted into the bargain. And to overdue pregnant women: ignore the advice that a curry will bring on labour. Chillies are the last thing you need when the contractions kick in.

Sex hormones might complicate our understanding of pain, but one day they might help us beat it, too. Locked inside your brain is the most powerful sedative, anti-anxiety drug and painkiller rolled into one. This magical compound derives from the sex hormone progesterone and, if medicinal chemists get it right, it may soon lead to an analgesic to rival morphine.

Scientists have known since the 1940s that progesterone—the female hormone we usually associate with the Pill and making babies—is also an incredibly potent sedative. Now researchers have found that it is the breakdown products of progesterone that have such a potent anaesthetic and analgesic effect. "During pregnancy, for example, as a woman comes close to term, the levels of these breakdown products of progesterone are extremely high," says Jeremy Lambert, a neuro-pharmacologist at the University of Dundee in Scotland. Only the natural hormone will do—the synthetic compounds used in contraceptive pills do not work in the same way.

Fortunately, this natural analgesia and anxiolysis is not exclusive to women. There are enzymes in the brain and spinal cord of both men and women that produce similar breakdown compounds, known as neurosteroids, from cholesterol or progesterone. In mounting doses, they may act as analgesics, anticonvulsants and even anaesthetics.

Researchers are now intent on harnessing these effects. The trick is to untangle one neurosteroid action from another: to induce pain relief without knocking you unconscious and without affecting fertility. Colin Goodchild, an anaesthesiologist at Monash University in Victoria, Australia, may have already hit on a compound—alphadolone—that can do exactly that. "It can work as a pain-relieving drug without causing sedation," says Goodchild.

Goodchild hopes that alphadolone may eventually replace opioids such as morphine, or at least reduce their usage. Progesterone metabolites might also lead to an "all-natural" sleeping pill and antiepileptic with few, if any, side effects. "I think neurosteroids are going to be the pharmaceuticals of the future," says Goodchild.

Acknowledgments

'Remote control brains: a neuroscience revolution', Douglas Fox, © New Scientist (Issue 2613)
'Why stress gets on your nerves', Clare Stanford, © New Scientist (Issue 1679)
'His pain, her pain', Lisa Melton, © New Scientist (Issue 2326)