Keeping track of the literature isn't easy, so Outside JEB is a monthly feature that reports the most exciting developments in experimental biology. Short articles that have been selected and written by a team of active research scientists highlight the papers that JEB readers can't afford to miss.

Outside JEB

THE REAL TASTE OF VICTORY

The 'taste of victory' is a phrase many associate with trained athletes preparing for intense competition. Indeed, Ed Chambers and colleagues at the University of Birmingham and Manchester Metropolitan University have started unravelling the physiology behind that phrase. When a cyclist swigs a sports drink prior to a race, we relate the energy boost to the sugary calories ingested. However, there may be more to this effect than the pure metabolic benefits of ingesting carbohydrates, as the benefits are lost if the carbohydrates are infused intravenously. Chambers' team hypothesized that because some taste receptors are connected to the brain region responsible for pleasure and reward, this boost phenomenon may start in the athlete's mouth. They wondered whether carbohydrates stimulate the brain so that it motivates the body to physiologically prepare to perform.

Chambers' team tested this idea with trained cyclists exercising for 1 h. To determine whether sweetness or perceived calories influenced exercise, athletes rinsed their mouths prior to exercise with a 'drink' containing artificial sweetener alone, artificial sweetener and sweet carbohydrate, or artificial sweetener and non-sweet carbohydrate. All three solutions tasted the same so that the athletes were unable to identify whether their mouth-rinse had included calories, which may have influenced their performance.

The results revealed that the athletes completed a set amount of work 2% faster having tasted the sweetened and unsweetened carbohydrate solutions compared with athletes that had tasted the carbohydrate-free, artificially sweetened solution. Athletes also exercised with 3% more power after tasting the sweet carbohydrate solution. This suggested that performance enhancement may be independent of 'sweetness' and more closely related to a drink's perceived caloric value.

Next, Chambers and colleagues decided to use functional magnetic resonance imaging (fMRI), which detects brain activity *via* changes in blood flow, to determine how the athletes' brains responded to the different drinks. The sweet carbohydrate solution activated regions of the brain associated with emotions and eating behaviours, which are suppressed when a person is not hungry. This is a typical response to food, and not thought uniquely related to exercise. Non-sweet carbohydrates activated the same region of the brain but additionally activated regions thought to perceive energy and those associated with motivation and reward.

Chambers' team believes that carbohydrates stimulate non-sweet receptors that activate the brain to enhance performance in similar ways that other stimulants, like caffeine, do. The response tempers messages coming from the body that accompany intense exercise like muscle fatigue, joint pain and increased temperature that could inhibit performance. When stimulated, these new taste receptors tell the brain, 'you can do it!' Chambers' research, therefore, suggests that the brain could be dictating performance before the muscles, heart or lungs. So, if this 'brain-before-body' phenomenon is influencing human performance, imagine what we could be missing when studying other high performance animals! Do other animals even need this motivational input like we do? If so, Chambers' idea could really change the way we assess exercise activities like migration, predator evasion and prey capture in other organisms; the potential for pre-performance brain input could be huge! Meanwhile humans, if you're getting ready for that big race, just remember to treat your tongues first.

10.1242/jeb.023796

Chambers, E. S., Bridge, M. W. and Jones, D. A. (2009). Carbohydrate sensing in the human mouth: effects on exercise performance and brain activity. *J. Physiol* **587**, 1779-1794.

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THERMAL REGULATION

HIGH METABOLIC RATE FLIES LIKE IT COLD

Temperature is a key environmental variable shaping organisms' biology. What enables or constrains species to survive at different temperatures is still not fully understood. Biologists tackle this issue from various points of view. Some investigate why animals prefer and inhabit environments with specific temperatures, while others advance our knowledge of how temperature affects intricate cellular processes. Bridging the gap between cellular level and whole animal behaviour is one of the current challenges in biology.

One approach used recently by a group of Japanese researchers at Kyoto University was to screen for mutant flies that have unusual temperature preferences. The larvae of one mutant, which they called the *atsugari* (*atu*) mutant, preferred and selected a temperature of 18°C when exposed to a temperature gradient, while unmutated larvae preferred 22°C. The team went on to identify how the mutant larvae differed from the unmutated insects and how the changes affected the insects' physiology.

The team found that the cold loving mutant larvae had suffered a mutation in the gene encoding the protein dystroglycan (*DmDG*), resulting in a significant reduction in the gene's expression levels to 15% of that of the unmutated flies.

The next step was to find the functional link between the gene mutation and the mutated larvae's low temperature preference behaviour. It turns out that the dystroglycan protein affects cellular membranes and lowering the gene's expression level causes an increase in the cell's calcium permeability. Alterations in the gene's expression caused an increase in the intracellular calcium level in the cold loving mutant larvae. As intracellular calcium levels are an important regulator of cellular energy metabolism through activation of the mitochondrial enzyme pyruvate dehydrogenase (which contributes to ATP production), the team measured the enzyme's activity level. They found that the enzyme had higher activity in the cold loving *atu* mutant larvae, which could explain the higher cellular ATP levels. Also, the metabolic rate of the mutated larvae was almost double that of the wild-type. Drawing these observations together, the team suggest that the mutant larvae's increase in ATP levels and metabolic rates may alter the larvae's thermoregulatory behaviour allowing them to occupy colder locations.

This study shows the close link between energy metabolism and temperature preference in ectothermic animals. The specific gene targeted, *DmDG*, may or may not be a relevant target in nature but the consequence of variation in energetics on the temperature preference behaviour is clearly shown in this study.

10.1242/jeb.021634

Takeuchi, K., Nakano, Y., Kato, U., Kaneda, M., Aizu, M., Awano, W., Yonemura, S., Kiyonaka, S., Mori, Y., Yamamoto, D. and Umeda, M. (2009). Changes in temperature preferences and energy homeostasis in dystroglycan mutants. *Science* **323**, 1740-1743.

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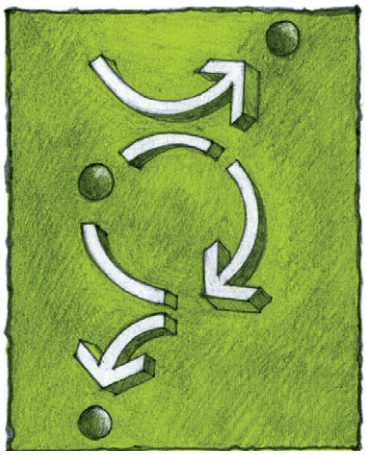
NEUROPROTECTION



HYDROGEN SALINE A REAL GAS

The 2005 announcement in *Science* that the inhalation of hydrogen sulfide gas (H_2S) could induce a reversible hypometabolic state in mice, similar to suspended animation, set off a flurry of speculation and investigation into the mechanisms and potential therapies suggested by the discovery, from the possibility of extended space travel to improving the outcome after a stroke. Breathing the gas decreased body temperature, respiratory and heart rates, and activity levels, all without incurring brain damage. The initial report was followed by the determination that mice could tolerate hours of otherwise lethal hypoxia if exposed to the gas first, suggesting H_2S as a potential therapy for hypoxia-related diseases. Cessation of blood flow (ischemia) and reperfusion, as occurs in stroke or during a heart attack, causes both cell death (apoptosis) and inflammation, and the gas has proved to improve survival by combating both of these pathologies. Hydrogen gas alone is also protective against ischemia–reperfusion injuries, primarily by neutralizing free radicals, but utilizing highly flammable hydrogen gas in a clinical setting poses a safety risk. This led Jianmei Cai and his colleagues at the Medical Universities in Shanghai and Shandong to wonder if injecting saline saturated with hydrogen gas could similarly protect newborn rats against ischemia, as reductions in blood flow or oxygen during birth can lead to significant brain damage.

The investigators first tested three concentrations of hydrogen saline in rat pups subjected to low oxygen (hypoxia) and brain ischemia (reduced blood flow) to determine the best concentration. Hydrogen-saturated saline was injected into the peritoneal cavity and the levels of brain damage determined by staining brain slices to look for dead *vs* live cells; by examining activity levels of an apoptotic marker (caspase-3) that indicates that cells have

died; and by measuring levels of oxidatively damaged lipids, as lipid and protein damage are symptoms of oxidative stress, which continues to damage cells for hours to days after ischemia–reperfusion. The rat pups also underwent behavioral testing to look for overt signs of brain damage.

Cai's group found that 5 ml kg^{-1} of H_2 saline almost completely suppressed the damage that occurred in hypoxic–ischemic rats. The number of live cells in the cortex and hippocampus was significantly higher in hypoxic–ischemic animals that had been treated with H_2 saline than in animals that were hypoxic–ischemic but not treated with H_2 saline, while the volume of dead cells in H_2-treated rats was reduced to non-hypoxic levels. H_2 saline also dramatically decreased levels of oxidatively damaged lipids, apoptotic activity and behavioral deficits. The H_2-treated rats that had been exposed to ischemia also sustained less brain damage than the untreated rats. They were able to escape a water maze in approximately half the time that it took for ischemic rats to escape, had better postural responses, and wandered less in their cages.

The key neuroprotective effect of hydrogen is apparently its ability to neutralize free radicals, and while suspended animation and space travel may yet be a long way off, research such as this offers hope that we can begin to significantly decrease the mortality and morbidity associated with strokes, heart attacks and neonatal brain disorders.

10.1242/jeb.021592

Cai, J., Kang, Z., Liu, K., Liu, W., Li, R., Zhang, J. H., Luo, X. and Sun, X. (2009). Neuroprotective effects of hydrogen saline in neonatal hypoxia–ischemia rat model. *Brain Res.* **1256**, 129-137.

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DO MUSCLE SYNERGIES ACTUALLY WORK?

Animals have a lot of muscles, and many of them do similar things. To an engineer, the multitude of muscles poses a conundrum. When a frog wants to kick its leg back, extending its hip, it could use either the gracilis muscle or the semimembranosus. As they both produce forces that tend to extend the hip, how does the animal decide which muscle to use? And if the animal needs to make a complex movement, how does it manage to sort through the plethora of muscle activation patterns in a reasonable amount of time, when many of the patterns produce roughly the same motions?

Experimental results point to a way out of the conundrum. When researchers measure muscle activation in many muscles, they often see that certain muscles – like the gracilis and semimembranosus – tend to come on together. These patterns, called 'synergies', could mean that the frog doesn't need to make a decision. When it decides to extend its hip, maybe it just activates the 'hip extending synergy' that turns on both the gracilis and semimembranosus.

Synergies could solve both the decision problem – which muscles to turn on – and also the sorting problem – making the decision in a reasonable amount of time – by restricting the number of choices. Rather than choosing an activation level for each muscle individually, the animal would only have to choose between a much smaller number of synergies.

But doesn't the reduction in the number of choices come at a cost? Wouldn't the animal end up turning on some muscles unnecessarily, because they were linked together in a synergy, and therefore end up wasting energy?

According to a recent study in *PNAS* by Matthew Tresch and his colleagues at Northwestern University, the answer is 'no' – provided one chooses the synergies properly. In fact, the group found that only five synergies are sufficient to produce efficient motions in a mathematical simulation of a frog leg.

The researchers developed a computer simulation of the frog hindlimb and 13 muscles. Then they used two different sets of criteria to estimate synergies for the limb. First, they aimed to produce synergies that worked best with the natural biomechanics of the limb, which they called 'natural dynamics synergies'. Second, they calculated synergies that would allow the animal to produce torques in any direction around each joint, called 'joint torque synergies'.

They put their simulated frog leg through a test, asking it to move its leg to six different points. Initially, they calculated the best-case scenario: the most efficient motion, using the smallest muscle activations for each of the 13 muscles individually that would move the leg accurately to each point. Then they compared this best-case pattern to the closest motion produced using the two sets of synergies, rather than activating the muscles individually.

The joint torque synergies ended up wasting a lot of energy, but the natural dynamics synergies did well, producing muscle activations that were about 85% similar to the best-case situation. In support of the idea that synergies could produce the same movements but with fewer choices, they were also able to reduce the simulation to only five natural dynamics synergies – as if there were only five muscles in the limb, not 13 – and still keep more than 80% similarity to the best-case situation.

The benefit of using a few well-chosen synergies was clear, the researchers stated: solving the full, best-case scenario optimization problem took many hours on the computer. But, by using the natural dynamics synergies, they could find a solution in seconds that was nearly as good. Maybe that's how animals do it too.

10.1242/jeb.021550

Berniker, M., Jarc, A., Bizzi, E. and Tresch, M. C. (2009). Simplified and effective motor control based on muscle synergies to exploit musculoskeletal dynamics. *Proc. Natl. Acad. Sci. USA.* **106**, 7601-7606.

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