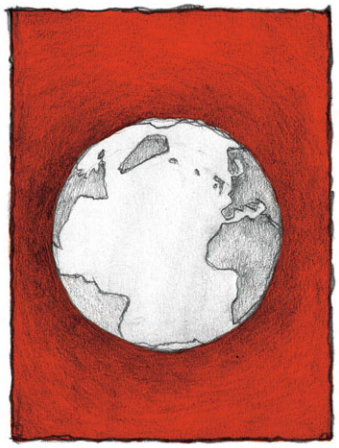
Keeping track of the literature isn't easy, so Outside JEB is a monthly feature that reports the most exciting developments in experimental biology. Short articles that have been selected and written by a team of active research scientists highlight the papers that JEB readers can't afford to miss.

Outside JEB

HEART PERFORMANCE



MATING BOOSTS HEART SIZE

Vertebrates depend on an efficient transport system to supply their bodies with vital nutrients and oxygen. The vertebrate heart is highly versatile and adjusts its performance when a body's demands change in response to factors such as temperature, digestive state, or behavioural responses. In the longer term, other factors influence the heart's workload, such as sexual maturation and reproduction. While cardiac output can be changed short-term by altering heart rate or the amount of blood ejected per heart beat, the only way to change output long-term is to change the heart's size. To investigate what factors influence long-term changes in the heart, Filippo Garofalo and his colleagues from University of Calabria, Italy, investigated the influence of sex and season on heart morphology and performance in the green frog *Rana esculenta*.

Over the course of eight years, the team captured 696 green frogs at different times of year. To see how heart mass differed between the sexes and seasons, they carefully removed the animals' hearts and weighed them. Heart mass increased with body mass in both sexes, however males had bigger hearts for a given body size, even though they are smaller than females. This suggested that males are more active.

Next, to estimate how much blood each heart could pump in a particular time, the team inserted tiny tubes into the veins and arteries of whole hearts and artificially pumped fluid into them. They used pulse pressure – the difference in blood pressure between when the heart is filling with blood and when it is contracting – as a measure of a heart's performance. They found that heavier hearts pumped more fluid and had a higher maximum pulse pressure in both males and females. They also found that female hearts pumped relatively more fluid than male hearts, meaning that they pumped more blood for their size.

The team found out why females' hearts pumped more blood when they investigated which part of the heart was responsible for the increase in overall heart mass. They separated out and weighed each heart's ventricle, which pumps blood to the body, finding that females' ventricles were heavier than males' ventricles, even though they had relatively smaller hearts. The larger ventricles caused the better than expected heart performance in females.

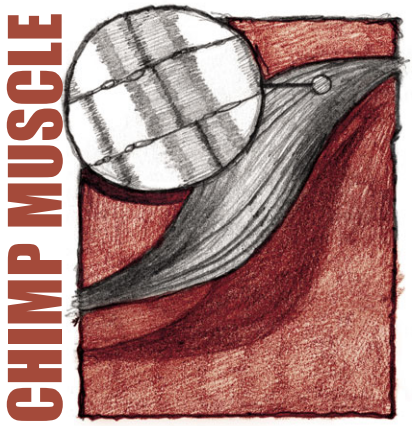
Comparing ventricle size in frogs captured at different times of year, they found that females' ventricles were larger still during the crucial parts of reproductive cycle: spring, when mating occurs; and winter, when animals recover and prepare for the next breeding season. This suggested that there was an extra strain on females at these times of year. Also, they found that as males became sexually mature their ventricles increased in mass, which raises pulse pressure. Wondering what morphological changes occurred in heavier hearts, the team found that larger ventricles had more heart muscle tissue, which would account for their higher pulse pressure.

The authors conclude that when frogs are very active during the breeding season, both sexes tune up their cardiovascular system to cope with the increased metabolic demands: females have to produce a whole clutch of good quality eggs and choose a mate; males use up a lot of energy vocalising to attract females. At these times, females' ventricles are relatively larger, meaning that they can maintain a higher blood flow than the males, provide more oxygen to their tissues, and cope with the enhanced work load caused by their bigger body size and more intense breeding behaviour.

10.1242/jeb.000448

Garofalo, F., Imbrogno, S., Gattuso, A., Spena, A. and Cerra, M. C. (2006). Cardiac morpho-dynamics in *Rana esculenta*: influence of sex and season. *Comp. Biochem. Physiol.* **145A**, 82-89.

Johnnie B. Andersen
University of Aarhus
johnnie.andersen@ki.au.dk



CHIMP POWER

It was in high school that I first learned of the remarkable genetic similarity between humans and chimpanzees: we share around 98–99% of our DNA with our nearest great-ape relatives. With the recent sequencing of the chimpanzee genome, it is exciting to think that we might soon understand how these relatively small differences between human and chimp DNA translate into rather notable differences in our respective phenotypes. Melanie Scholz of Vrije University and colleagues from Amsterdam and Antwerp recently focussed on one currently underappreciated phenotypic difference between chimpanzees and humans: the incredible strength of chimps.

Anecdotal and scientific evidence indicate that humans have inferior strength to chimps, and most would be hard-pressed to win a rope-tugging contest against a chimp half their size. Since chimps aren't overly endowed with muscle mass, Scholz and her colleagues wondered if there might be something special about the intrinsic properties of chimpanzee muscle that sets them apart from humans. To investigate, they compared squat jumping performance between bonobos (*Pan paniscus*), close relatives of the common chimpanzee, and humans.

During squat jumps, the amount of work generated by the limb muscles closely parallels the potential energy gain of the body. Since a body's potential energy gain is directly related to the height of a jump, estimates of muscle energy output can be made by keeping a close track of a body's vertical movement during a jump, without using any invasive procedures.

To find out if chimps have a superior jumping performance, Scholz and coworkers recorded high-speed videos of squat jumps from three bonobos and four human subjects taking off from a force plate. All three bonobos performed squat jumps higher than 0.7 m. In contrast, the best human subject jumped just over 0.3 m, and the literature reports that top-level athletes jump between 0.4–0.5 m. To estimate the mechanical energy and the power output of the jumps, the team analyzed the movements of the center of mass and the limbs as well as the ground-reaction forces recorded from the force plate. These data were combined with limb anatomical data from previous studies into a mathematical model that determined limb muscle mechanical energy and power output during jumping.

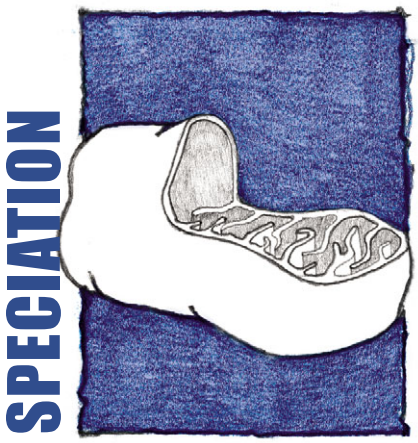
In both humans and bonobos, the mechanical energy and power output required for the best jumps were similar at approximately 450 J and nearly 3000 W, respectively. However, bonobo limb extensor muscle mass is less than half that in a human, suggesting that per gram of muscle, the work and power output of bonobos' muscles are over twice those observed in humans.

The authors suspect that the observed differences in work and power generation could be related to fundamental differences in the ability of a certain mass of muscle to produce force, which could be caused by different forms of the muscle protein myosin. Properties of muscle contraction such as muscle fibre shortening distances or velocities could also be responsible. Regardless, it is possible to make two tentative generalizations. First, some of those small differences in DNA make-up between chimps and humans may well relate to muscle structure and function; and second, assuming chimp muscle properties are widespread among the great apes, King Kong just got a whole lot scarier.

10.1242/jeb.000414

Scholz, M. N., D'Aout, K., Bobbert, M. F. and Aerts, P. (2006). Vertical jumping performance of bonobo (*Pan paniscus*) suggests superior muscle properties. *Proc. Roy. Soc. Lond. B* **273**, 2177-2184.

Gary B. Gillis
Mount Holyoke College
ggillis@mtholyoke.edu



MITOCHONDRIA AND MAKING NEW SPECIES

How do new species arise? Many biologists are intrigued by this question: we are obviously not the same species as chimpanzees, so identifying different species can be simple. However, the process of speciation occurs gradually and is much harder to measure. Scientists want to know what changes occur between reproductive compatability, when two populations fully interbreed; and reproductive isolation, when two populations can't interbreed and become independent species. An early step in this process is called hybrid breakdown, where offspring with parents from different populations have reduced fitness, which gets in the way of successful interbreeding. The causes of hybrid breakdown are poorly understood, prompting Christopher Ellison and Ronald Burton from University of California San Diego to explore how mitochondrial function contributes to this process.

As the powerhouse of the cell, mitochondria generate energy during respiration in the form of ATP. The final part of respiration involves an electron transport chain that contains five 'complexes', each made up of many enzymes. All of the enzyme complexes, except complex II, contain proteins encoded by both nuclear and mitochondrial DNA, and these proteins must interact properly for mitochondria to work effectively. Offspring can inherit nuclear DNA from either parent, but mitochondrial DNA is inherited only from the mother, and these genomes normally co-evolve to keep proteins in the mitochondria interacting properly. Ellison and Burton suspected that hybrid breakdown is caused when mismatched proteins from genomes that haven't evolved together are combined. This could occur when mitochondrial DNA from a mother in one population is

combined with nuclear DNA from a father in a different population.

To test this idea, they collected marine crustaceans (*Tigriopus californicus*) from several different populations along the western coast of North America. There are strong genetic differences between wild populations, and hybrid breakdown occurs when different populations interbreed and produce offspring. To assess the effects of hybrid breakdown on mitochondrial function, the team interbred individuals from different populations and produced hybrids, then isolated their mitochondria to measure how well they made ATP.

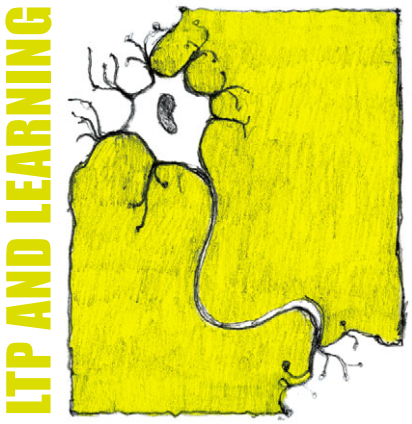
They found that mitochondria from hybrids produced much less ATP than their parents' mitochondria. The authors suspect this reduces hybrid fitness because efficient ATP production is essential for survival; however, they didn't know how ATP production was being reduced. To address this, they measured the activity of enzyme complexes I-IV from the mitochondrial electron transport chain. Complexes I, III and IV had reduced activity in hybrids. The team already knew that these complexes contain proteins encoded by mitochondrial and nuclear DNA, so concluded that incompatibilities between both protein types was causing the mitochondria in hybrids to produce less ATP.

This idea was supported when they found that the activity of enzyme complex II, which is encoded by nuclear DNA only, was the same in hybrids and their parents. This shows that proteins encoded by nuclear DNA are unaffected by interbreeding, while incompatibilities between proteins encoded by nuclear and mitochondrial DNA from different populations might lead to reduced fitness and contribute to hybrid breakdown. By understanding how mitochondria can contribute to this early step in the process of speciation, Ellison and Burton have brought us one step closer to understanding the origin of species!

10.1242/jeb.000430

Ellison, C. K. and Burton, R. S. (2006). Disruption of mitochondrial function in interpopulation hybrids of *Tigriopus californicus*. *Evolution* **60**, 1382-1391.

Graham R. Scott
University of British Columbia
scott@zoology.ubc.ca



UNLOCKING LEARNING

Where are our memories stored? How is information stored in the brain? Philosophers and scientists have been exploring these questions ever since the brain was discovered to control thought and reason. In 1973 Bliss and Lømo described a phenomenon known as long-term potentiation (LTP), where electrical stimulation of neurons during experiments results in an artificial increase, or potentiation, in synaptic strength. Although LTP has been extensively studied as a cellular model of memory formation, until now it has not been directly shown that learning in a living animal actually induces LTP. Jonathon Whitlock and colleagues from The Picower Institute for Learning and Memory at Massachusetts Institute of Technology demonstrate for the first time that a learning task in rats can induce LTP in a brain area called the hippocampus, which plays a role in memory formation.

Given a choice, rats will choose a dark environment over a well-lit one, unless they receive a nasty foot shock each time they enter the dark environment. This type of training is called inhibitory avoidance training and rats learn to avoid the dark environment after a single foot shock, staying in the well-lit one instead. Already knowing that an area of the hippocampus called CA1 is critical for inhibitory avoidance learning, the authors searched this brain area to see if this type of learning forms potentiated synapses, indicating LTP.

To investigate synapse strength, the team implanted several electrodes into the CA1 area of live animals, and stimulated the input neurons to that area. They measured the strength of the synaptic responses both before and after training. The authors found that inhibitory avoidance training caused potentiation at approximately 25% of the recording locations. This was not unexpected, because potentiating every synapse after learning a task is unnecessary to indicate LTP and probably a waste of energy for the network.

When no further LTP can be induced at a synapse, it is said to be saturated. Previous reports have shown that learning can result in saturation at certain synapses, indirectly indicating that learning induced LTP. Having found that the training potentiated about 25% of the recorded synapses, the authors then applied a stimulus to CA1 known to induce LTP across synapses, and measured the response to see if they could induce further LTP. They saw no further potentiation at recording locations already showing LTP from the first experiment, showing that the synapses were saturated and could not increase in strength any further. In contrast, stimulation did cause LTP at locations that did not show LTP before training.

The authors suggest that other researchers could not find LTP specifically induced by learning because synapses potentiated by learning are sparsely and widely distributed, making them difficult to detect in a vast sea of unmodified connections. Their data showing potentiation at only some of the recording locations supports the idea that associative memories are stored in many locations, and also means that these types of memories can be recalled when retrieval cues only partially match the original situation. Sparse distribution could also render the memory more robust in case some synapses fail to respond. If this were true, memory recall for inhibitory avoidance training would still be successful if LTP was disrupted at just a subset of the electrodes showing learning-induced LTP after training.

10.1242/jeb.000398

Whitlock, J. R., Heynen, A. J., Shuler, M. G. and Bear, M. F. (2006). Learning induces long-term potentiation in the hippocampus. *Science* **313**, 1093-1097.

Susan Sangha
Westfaelische Wilhelms Universitaet Muenster
sangha@uni-muenster.de

METABOLISM

BMR UNDER THE SPOTLIGHT

Basal Metabolic Rate (BMR) is the lowest amount of energy used by a resting, fasting individual who is not consuming any energy to warm up or cool down, and is one of the most studied aspects of animal physiology. However, scientists haven't yet answered all the questions associated with BMR, as a recent publication by Greg Russell and Mark Chappell from the University of California, Riverside, tells us. They tested a common assumption that BMR is consistent over an animal's lifetime in the deer mouse *Peromyscus maniculatus*, and found some unexpected results.

For decades, physiologists have extensively measured the BMR of many warm-blooded animals and linked their findings to an animal's fitness. By doing this, scientists are assuming that offspring inherit their BMR from their parents, meaning that BMR should be consistent over an animal's lifetime, but will change between generations in response to selection pressure. A few previous studies have measured consistent BMR over an animal's lifetime, supporting this assumption. However, other studies have shown the opposite: that environmental factors such as altitude and temperature influence BMR. With this in mind, Russell and Chappell wanted to confirm if the assumption that BMR is consistent and responds to selection pressure was accurate in deer mice, and if BMR responds to changes in altitude and temperature.

First, the team investigated whether altitude and temperature influence BMR. They bred two groups of experimental captive mice: one group were born and bred at 340 m, and the other group in the mountains, about 4000 m above sea level. Measuring the animals' BMR, they found no difference between low- and high-altitude mice, suggesting that altitude alone doesn't affect BMR. To find out how temperature and altitude together affect BMR, the team divided both the high-altitude and the low-altitude mice into two groups, keeping one group at a warm 21°C and the other at a chilly 5°C. They found that the combination of high-altitude and 5°C temperatures raised BMR, but the other groups were unaffected. This finding agrees with previous studies which showed that cold temperatures and high-altitude influence BMR during an animal's lifetime.

To find out if BMR remained consistent in the mice, supporting the assumption that it responds to selection pressure, the team measured BMR in the mice again after 1–2 months of adapting to their new temperatures. Expecting to find the same BMR in both measurements, the research duo were astonished to find out that BMR in deer mice was not consistent over time. This was the team's most surprising result and heavily contradicts previous studies that show BMR consistency.

The authors conclude that – at least in deer mice – researchers can't make the assumption that BMR responds to selection pressure because it is not consistent over an animal's lifetime. Instead, the researchers propose that selection could affect BMR indirectly, for example by acting on physiological traits that experience selection, such as maximal aerobic capacity, which in turn influence BMR by increasing or decreasing metabolic demands. Russell and Chappell's findings show that researchers should be cautious when assuming that BMR changes in response to selection pressure in all mammalian species, and will probably start a lively discussion on the evolutionary basis of BMR and the assumptions that researchers make in experiments.

10.1242/jeb.000422

Russell, G. A. and Chappell, M. A. (2007). Is BMR repeatable in deer mice? Organ mass correlates and the effects of cold acclimation and natal altitude. *J. Comp. Physiol. B.* **177**, 75-87.

Teresa Valencak
Veterinary University Vienna
Teresa.Valencak@vu-wien.ac.at