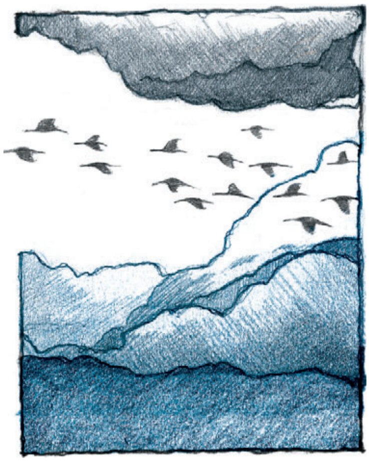
Keeping track of the literature isn't easy, so Outside JEB is a monthly feature that reports the most exciting developments in experimental biology. Short articles that have been selected and written by a team of active research scientists highlight the papers that JEB readers can't afford to miss.

Outside JEB

MANOEUVRABILITY



STABILITY: JUST KEEP ON FLAPPING

If you have ever played with remote controlled helicopters, you will have a grasp of the issues of stability and manoeuvrability in slow flight. Part of the success of the recent, cheap toy helicopters is that they have been made so stable that even a child can fly them. However, this stability can make them somewhat boring to fly – they cannot dart around with great agility. So is there an inevitable conflict between stability and manoeuvrability in slow flight? Ty Hedrick, from the University of North Carolina, teaming up with Bo Cheng and Xinyan Deng, from the University of Delaware, has demonstrated that this may not be the case for flapping animals. Instead, a remarkably simple aerodynamic mechanism accounts for the time taken to recover and regain a steady heading after a perturbation (such as a turn) in slow-flying animals ranging from fruit flies to cockatoos; and changes that would increase this 'stability' would also increase the potential for 'manoeuvrability'.

The mechanism they propose is termed 'flapping counter-torque' (FCT), and is merely what would be expected to happen if the flapping animal just kept on flapping in its normal manner after its perturbation. Consider a turn to the right: as the animal turns to the right, both wings flap forward and back as usual, but because the animal is turning, the left wing moves through the air relatively quickly as it flaps forwards while the right wing moves relatively slowly, and *vice versa* (left wing slow, right wing fast with respect to air) as they flap back again. All else being equal, this results in an aerodynamic 'counter-torque' that acts counter to the direction of turn to stabilise the animal as it continues on its way. Hedrick, Cheng and Deng calculate the way in which this mechanism for recovering from a turn should scale with animal size and wingbeat frequency, and compared this with observations for a range of insects, birds and bats during slow flight involving turns of 60 deg. or greater. As predicted, with similarly shaped animals, the time taken (in terms of number of flaps) to lose half the turning speed is broadly constant, despite vastly different body sizes and wingbeat frequencies. For instance, fruit flies, bluebottles and hummingbirds take around two wingbeats to halve their rate of turning. In addition, this timing changes as predicted with different shapes: hawkmoths', bats' and cockatoos' wings are around double the size of those of the previous group (relative to body weight) and, agreeing with the exceedingly simple FCT model, take less than one wingbeat to lose half the turning rate.

Interestingly, the factors that increase stability due to the passive FCT mechanism, especially higher wingbeat frequency, also increase manoeuvrability. Hummingbirds may provide an example of this: males typically have higher wingbeat frequencies, potentially conferring benefits for both manoeuvrability during display flights and stability when recovering from a perturbation. The cost? Probably energetic: higher flapping frequencies might be expected to require more power, though this has yet to be demonstrated conclusively.

So, Hedrick and colleagues have shown a simple, passive mechanism available to slow, flapping-winged fliers conferring both stability and, potentially, agility. Sadly, this mechanism is unavailable to helicopters; I will have to wait for advances in flapping micro-air vehicles before finding the dream stable-and-yet-also-manoeuvrable toy in my (nephew's) Christmas stocking.

10.1242/jeb.021394

Hedrick, T. L., Cheng, B. and Deng, X. (2009). Wingbeat time and the scaling of passive, rotational damping in flapping flight. *Science* **324**, 252-255.

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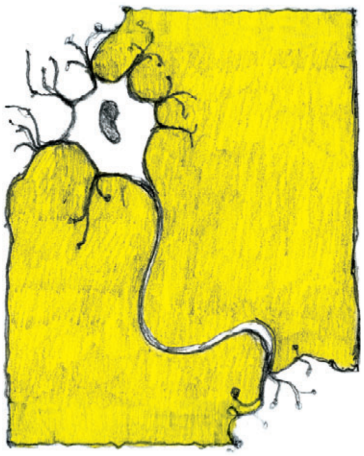
SOCIAL BEHAVIOUR



HOW ANTS SWITCH JOBS

Individuals with well-defined roles can be driven to perform jobs outside their areas of expertise by changes in the environment. We know many animals can make these shifts but, in large part, we know very little about the genetic/molecular basis of environmentally induced switches in behavior. Christophe Lucas and Marla Sokolowski from the University of Toronto recently took on this question by studying shifts in social behavior in the ant, *Pheidole pallidula*. They published their work in a recent edition of the *Proceedings of the National Academy of Sciences*.

Pheidole pallidula colonies consist of morphologically and behaviorally specialized castes: minors ('foragers') and majors ('soldiers'). But individuals can switch jobs at a pinch. If the colony comes under attack, foragers will do some 'soldiering'; likewise, if the colony stumbles on a new food source, soldiers will help out with the harvest.

To get at the molecular underpinnings of this switch, Lucas and Sokolowski first looked for *forager*, a gene involved in controlling how *Drosophila* search for food. The team found a closely related gene in *P. pallidula* (*ppfor*) that codes for the enzyme cGMP-dependent protein kinase (PKG). The enzyme is in the brains of both castes, but is only expressed in a small subset of cells. Intriguingly, when the team assayed PKG activity in brain tissue, they found that soldiers have more PKG activity than foragers. PKG activity is clearly correlated with being a soldier ant.

The team next tested how environmental cues affect PKG activity. First, they gave ants a cue to stimulate foraging (a live mealworm). Afterwards, both soldiers and foragers showed reduced PKG activity when compared with controls (animals who had only encountered a plastic mealworm). In a converse experiment, the team gave ant colonies a cue designed to stimulate defensive behaviors. They placed an 'intruder ant' in an enclosure within the nest. The presence of the intruder caused PKG activity to rise in both castes.

The team's initial experiments provided solid evidence that PKG activity is down-regulated during foraging and up-regulated during colony defense. So is PKG activity alone sufficient to make ants more defensive? To address this, the team spiked colony food with a PKG activator drug, then watched how ants responded to foraging (mealworm) and defensive (alien intruder) stimuli. Indeed, when the insects' PKG levels were elevated, both castes were less interested in foraging, and soldiers spent more time in defensive mode when presented with an intruder.

Lucas and Sokolowski have been able to causally link a single gene product in a small population of brain cells to a complex shift in social behavior. They were able to do this, in large part, because of the unique specializations and behavioral repertoire of ants. As such, this work stands out as an example of how studying social insects can offer unique opportunities to address questions that are often difficult to get traction on in other animals.

This work is also notable because it represents the application of Sokolowski's decades of molecular/genetic expertise in *Drosophila* to a new animal – one with an added dimension of social behavior not seen in fruit flies. Roving out into new territory like this is not an easy thing for a researcher to do, but Lucas and Sokolowski's work shows that the approach can pay off.

10.1242/jeb.021477

Lucas, C. and Sokolowski, M. B. (2009). Molecular basis for changes in behavioral state in ant social behaviors. *PNAS* **106**, 6351-6356.

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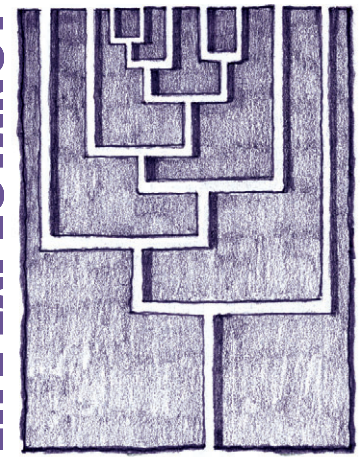
LIFE EXPECTANCY



ELEVATED CO_2 PUTS WORMS ON THE SLOW TRACK TO A LONG LIFE

Gas exchange is a basic requirement for almost all organisms, especially those that rely on aerobic metabolism, because mitochondria consume oxygen and produce carbon dioxide (CO_2) to produce ATP. Because of its critical importance to aerobic metabolism, the biology of oxygen has received a great deal of attention, and subsequently there is a tremendous body of information concerning the ways that cells sense and respond to changes in oxygen concentrations at organismal, organ, tissue and cellular levels. In contrast, far less attention has been paid to the biology of CO_2 in animals. However, CO_2 plays a critical role and is a major contributor to the regulation of pH in the extracellular fluids, and within cells. In fact, in mammalian systems ventilation patterns are largely regulated by levels of CO_2 in the blood and not oxygen. Despite the importance of CO_2 in mammalian physiology, very little work has focused on how cells sense levels of CO_2 and how they respond to elevated CO_2. Kfir Sharabi and colleagues recognized this deficiency in our knowledge and tested the effects of elevated CO_2 on the physiology, development and gene expression of the worm *Caenorhabditis elegans*.

To evaluate the affects of elevated CO_2 on organismal function, the team raised worms in normal air (0.04% CO_2) and air supplemented with 5, 9, 15 and 19% CO_2 at two temperatures, 20 and 25°C. The team then monitored larval development, egg production, egg survival, motility, life span and gene expression patterns using microarrays.

Elevated CO_2 slowed the rate of larval development, reduced the total number of embryos produced, and increased the worms' life span at concentrations of 9% or higher. The team also found the largest reduction in the number of embryos produced when the worms were continuously exposed to 19% CO_2 due to a decrease in the number of embryos produced by each individual after reaching adulthood. Exposing embryos to 19% CO_2 for discrete periods of time during development, the team found that the transition from the last larval stage (L4) to the adult stage is the most sensitive period of development in terms of the effects that elevated CO_2 had on each individual's subsequent reproductive output. Worms exposed to high CO_2 during this transition produced only 24% of the number of embryos produced by worms raised in normal air. Focusing on the long-term effects of CO_2 exposure, the team found that exposure to 15 and 19% CO_2 caused abnormal muscle development and reduced motility. Worms raised in 19% CO_2 also suffered a 40% drop in ATP levels compared with worms raised in normal air, indicating a significant physiological effect on metabolism that is presently unexplained.

Using gene expression profiling the team revealed hundreds of genes that change their expression during the first few hours of exposure to 19% CO_2. Elevated CO_2 appears to induce a stress response in *C. elegans*, where a small heat shock protein (hsp 12.3) is upregulated. Importantly, the patterns of gene expression associated with elevated CO_2 were different to those observed in worms exposed to hypoxia.

The results of this study indicate that elevated CO_2 has a profound affect on organismal form and function in this nematode worm. The unique nature of the response of *C. elegans* to elevated CO_2 suggests that in our quest to understand the importance of oxygen delivery, we may have been ignoring the critical other half of the gas exchange equation, the removal of CO_2.

10.1242/jeb.021519

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THERMAL TOLERANCE

THE PROBLEM OF SHIFTING THERMAL TOLERANCES

The interactions of organisms with the range of temperatures in their environments have generated intense interest among researchers for decades, and coupled with the wide acceptance that global climate change is a reality has resulted in a renewed interest in thermal biology. One significant aspect of ectotherm biology concerns the physiological mechanisms that designate the temperature window within which ectotherms (cold blooded animals) can maintain activity. One approach to measure the critical limits of this thermal window involves observing the temperatures where animals lose the ability to remain active by either cooling (CT_{Min}) or heating (CT_{Max}) them. This method is widely used, but recent publications suggested its outcomes are sensitive to cooling or heating rates and the temperatures at which organisms were acclimated, which could potentially complicate thermal studies in unknown ways.

Recently, in *Functional Ecology,* Steven Chown and colleagues reported the first systematic investigation of the effects of variable heating and cooling rates on the means and variances of CT_{Min} and CT_{Max} and the effects of acclimation at different temperatures on a model species, *Drosophila melanogaster*, and a globally important invasive species, *Linepithema humile* (Argentine ants). They acclimated the insects at 15, 20, 25 and 30°C whereafter they measured CT_{Min} and CT_{Max} at heating and cooling rates ranging from 0.05 to 0.5°C min^{-1}.

The results from the flies and ants were generally very different depending on the heating and cooling regimes and the temperature that the insects were acclimated to. The flies had reduced CT_{Min} values at slower cooling rates while acclimation at lower temperatures induced strong downward shifts in CT_{Min} values at all cooling rates. Measurements of critical temperatures in the ants showed that they had reduced CT_{Min} values at fast cooling rates while the acclimation temperature only resulted in shifts in the critical temperatures at faster heating and cooling rates. CT_{Max} means were similar for the two insect species with slower heating rates reducing the insects' measured CT_{Max} values, while the acclimation temperature shifted CT_{Max} values only at slower heating rates. However, the variances of CT_{Max} values increased slightly at faster heating rates while ants' CT_{Max} and CT_{Min} distributions about the mean showed major increases at slower heating and cooling rates.

Explanations for these incongruent results are not readily apparent yet. Nevertheless, the differences in variations about the mean induced by different cooling or heating rates can have profound consequences for estimating the extent to which these thermal tolerances can be genetically inherited by subsequent generations. If slower rates cause increases in variances, while the genetic contributions to thermal tolerance remain constant, one researcher, using slow rates, might conclude that heritability is low, while another, using faster rates, might conclude that heritability is high. This will impact on our interpretations of thermal tolerance evolvability in the light of changing environments. Likewise, the ways that variable effects of acclimation can interact with temperature rates can also lead to contradicting conclusions.

Should researchers therefore do away with these thermal methods? Chown and colleagues says that would likely not resolve the already existing issues. They advise that researchers should, rather, thoroughly document their methods and preferably use standardized rates, $\geq$0.25°C min^{-1}, for comparative studies, and also slower but ecologically relevant rates. And to include both standardized and ecologically relevant rates in datasets for future comparative macro-physiological studies.

10.1242/jeb.021436

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