

determine whether any sheep became infected with BSE, but they do suggest that the change in sheep spongiform encephalopathy incidence has been a gradual process, with no large epidemic during the 1980s and 1990s.

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Physiology

Exercise and reduced muscle mass in starlings

Muscles are often viewed as force-producing structures that increase or decrease in size according to their activity^{1,2}. But an increased muscle mass, although desirable for extra power, may also impose unwanted costs. Here we show that flight-muscle mass in starlings induced to perform more take-off flights actually decreases as a result of exercise. Our findings indicate that birds can strategically regulate a lower muscle mass to make themselves lighter and so cut flying costs without compromising their flight performance. This suggests that muscle size may be influenced by factors other than workload.

The muscle use–disuse hypertrophy–atrophy hypothesis is often invoked to explain flight-muscle size in wild birds: for example, enlarged pectoralis muscle (the main force generator for powering flight) is seen as an indicator of increased flight demands^{2,3}. The costs of bearing a larger muscle mass have not been investigated, however. One such cost could be carrying an increased load during flight. We manipulated take-off flight exercise in European starlings, *Sturnus vulgaris*, and measured changes in musculature in response to flight.

Daily flight exercise led to a reduction in

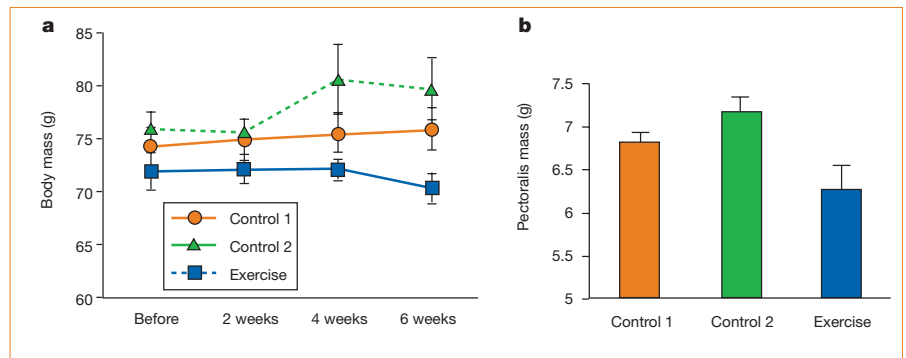


Figure 1 Morphological responses to exercise. **a**, Mean (\pm s.e.) change in body mass during the experiment ($F_{6,45} = 2.35$, $P = 0.047$); and **b**, mean (\pm s.e.) pectoralis muscle mass at the end of the experiment ($F_{2,15} = 3.95$, $P = 0.042$). Eighteen adult starlings were housed under controlled conditions based on an 8-h day and randomly allocated to three groups. ‘Exercise’ group birds were trained to fly between perches at opposite ends of a flight cage (0.6 \times 0.8 \times 2 m) by positive reinforcement with food rewards. Birds had 1-hour exercise trials on most days, with 34 trials over 6 weeks. First control group, control 1: birds were restrained in a small cage and fed *ad libitum* for the 1-h duration of the exercise trials; second control group, control 2: birds were restrained in a small cage and received the same food rewards at the same times as the exercising birds. On four occasions (the day before the experiment and then every two weeks), we recorded take-off force production (by tethered strain gauges), wing area, body mass and fat stores¹¹. After the trials, we measured pectoralis wet mass and fibre length¹²; supracoracoideus wet mass, fibre length and pinnation angle; and gastrocnemius muscle attributes. There was no difference in activity among groups when they were outside their cages. Further details are available from the authors.

body mass (Fig. 1a) and wing loading ($F_{6,45} = 2.28$, $P = 0.052$) compared with controls. Most birds had sparse fat deposits and there was no difference in fat score among treatment groups (Friedman $S_2 = 2.29$, $P = 0.319$). However, birds in the exercise group had reduced pectoralis muscle mass (Fig. 1b; pectoralis fibre length also decreased: sternal, $F_{2,15} = 3.49$, $P = 0.057$; thoracic, $F_{2,15} = 2.68$, $P = 0.101$). There was no effect of exercise on other musculature measured ($F_{2,15} < 1.85$, $P > 0.19$). Although differences in body mass and pectoralis mass appear to exist between control groups at 4 and 6 weeks (Fig. 1a, b), these differences were not significant ($F_{1,11} < 2.17$, $P > 0.171$). The reduction in body mass and pectoralis muscles in these birds did not affect take-off flight performance, which we measured as peak take-off acceleration ($F_{6,45} = 0.42$, $P = 0.859$).

Our results contradict the use–hypertrophy hypothesis. The higher nutritional requirements⁴ to fuel exercise cannot explain the changes in pectoralis mass, because daily access to food was not limited. Also, subcutaneous fat stores were not affected and the pectoralis comprises less than 1% fat^{2,5,6}. As sternal pectoralis fibre length decreased by 5% in the exercise group of birds, water loss cannot explain the decrease in muscle size.

Birds in the exercise group performed more flights than controls, so any strategy that reduces energetic flight costs must be adaptive. We propose that birds are regulating a lower body mass level to reduce flight costs, even at the expense of muscle size⁷. Starlings reduce their body mass in response to a reduced wing area⁸. The costs of decreasing pectoralis size are balanced by the benefits of losing 10% of body mass (M).

This idea is also interesting from a scal-

ing point of view: for a given wing area, decreases in pectoral/body mass and wing loading will result in a smaller decrease in force-generating ability (assuming a muscle force α to fibre area $\alpha M^{2/3}$). So a 10% decrease in pectoral/body mass would predict a 4.7% decrease in force generation.

We know that atrophied pectoral muscles retain a high density of mitochondria even though contractile proteins and fat stores are depleted², and that the pectoralis can undergo enzymatic adaptation to increase oxidative metabolism⁵. The smaller pectoralis may therefore be able to maintain its oxidative capacity. The relative abundance of the two fast-twitch muscle fibre types (dependent on either aerobic or anaerobic glycolysis) is unlikely to alter as a result of exercise because the starling pectoralis consists exclusively of fibres of the aerobic type⁹. These observations indicate that pectoralis size cannot simply be linked to functional performance.

The increased flight costs of losing pectoralis mass are offset by the decreased flight costs of losing overall body mass, resulting in a new equilibrium of body condition based on changes in flight demands. Therefore, muscle size is not merely a function of its activity or storage capacity, but also reflects costs of resource allocation within the whole body¹⁰. The size of flight muscle thus represents a dynamic cost–benefit trade-off of a broad range of potential limiting factors.

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Materials

Peeling and sharpening multiwall nanotubes

To realize the full potential of multiwall carbon nanotubes in applications such as biological and scanned probes, it is desirable to develop techniques for controlling their shape and geometry. Here we describe a method by which the outer layers of a multiwall nanotube can be successively removed at the end to produce what is effectively a sharpened structure.

Carbon nanotubes, owing to their unique mechanical and electrical properties, are candidates for a host of applications such as catalysts¹, biological cell electrodes², nanoscale electronic³ and mechanical⁴ systems, and scanned probe microscope and electron field emission tips^{5,6}. Many of these applications would be facilitated by some tailoring of the nanotube. For example, an 'ideal' scanned probe, field emission or biological electrode tip should be long, stiff and tapered for optimal mechanical response and have an electrically conducting tip. In addition, it would be useful to be able selectively to expose nested concentric nanotubes in a nanobearing. Techniques exist for growing nanotubes at preselected sites⁷ and for modifying nanotube ends through chemical etching⁸, but not for finely controlled shaping of the tubes.

We have found a simple and reliable method that allows controlled engineering or shaping of multiwall carbon nanotubes and enables average multiwall nanotubes to be easily converted into tips with ideal geometry for scanned probe, field emission, biological insertion or mechanical nanobearing applications. The shaping process involves the electrically driven vaporization of successive layers (that is, the tube walls) of the multiwall nanotube, with outer layers being removed in turn near the end of the nanotube, leaving the core nanotube walls intact and protruding from the bulk of the nanotube. This peeling and sharpening process can be applied repeatedly to the

same multiwall nanotube until the innermost tube(s) of the smallest diameter protrude, often with a tip having a radius of curvature comparable to that of one single-walled nanotube.

We demonstrate the method in a transmission electron microscope (TEM) configured with a custom-built mechanical/piezo manipulation stage with electrical feed-throughs to the sample. Figure 1 shows high-resolution TEM images of a conventional arc-grown multiwall carbon nanotube at different stages in the peeling and sharpening process. The left end of the nanotube (not seen in the image) is attached to a stationary zero-potential gold electrode. To the right (also not shown) is a larger nanotube that serves as the 'shaping' electrode: it is attached to the manipulator, whose potential can be controlled externally.

Figure 1a shows the nanotube in its pristine, as-grown state. In Fig. 1b, the shaping electrode has been momentarily brought into contact with the nanotube and a carbon onion has been inadvertently transferred from the shaping electrode to the nanotube, but the applied voltage (2.4 V) and current (170 μ A) are below the shaping threshold and no peeling or sharpening has taken place. Figure 1c shows what happens when the shaping electrode is brought into intimate contact with the tip of the nanotube at 2.9 V and 200 μ A: almost immediately, many layers of the nanotube are peeled away near its end and it now has a stepped diameter and is significantly sharpened. The carbon onion has been displaced to a benign position further down the tube. The newly exposed tip of the nanotube appears to be undamaged.

For Fig. 1d, the peeling and sharpening process has been repeated, resulting in a multiwall nanotube with highly desirable characteristics for many practical applications. The dominant protruding segment now consists of a three-walled electrically conducting nanotube with a radius of just 2.5 nm. Although we used an *in situ* TEM configuration to follow the sharpening and peeling process, this is not essential as the process could be performed blind and monitored only from the electrical characteristics of the nanotube. In addition, the 'shaping' electrode can readily be replaced by any conventional conducting substrate.

The physics behind our novel peeling and shaping process is intriguing. It is unlikely that uniform Joule heating of the nanotube would result in the observed behaviour. It is more likely that multiwall nanotubes conduct ballistically⁹, and the energy to break the carbon bonds and remove the nanotube layers originates from highly localized dissipation at defect scattering sites, located primarily at the ends of the tube. The ensuing avalanche of dissipation and bond-breaking would then lead to cata-

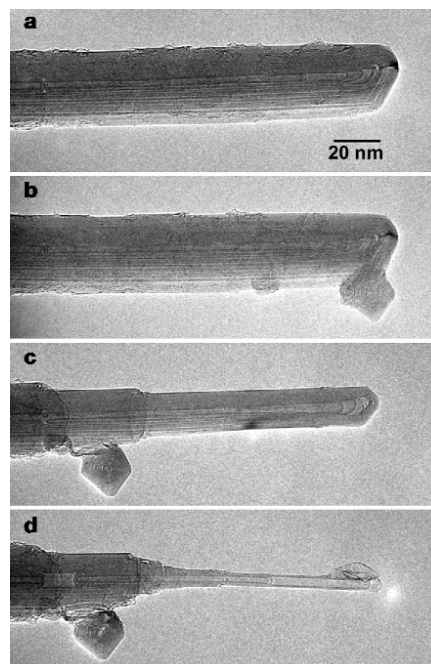


Figure 1 Transmission electron microscope images of a multiwall carbon nanotube being shaped. **a**, Nanotube in its pristine form: it contains approximately 37 walls and has an outer radius of 12.6 nm. **b**, A carbon onion has been inadvertently transferred to the nanotube end from the shaping electrode, but no attempt has been made to shape the nanotube. **c,d**, Results of the subsequent peeling and sharpening processes: the onion has simultaneously been displaced to a benign position down the tube axis. The shaped, or 'engineered', nanotube in **d** is thick and mechanically rigid along most of its length (not seen in the image), but tapers stepwise to a fine sharp tip that is electrically conducting and ideal for scanned probe microscopy or electron field emission applications. The final long nanotube segment contains three walls and has an outer radius of 2.1 nm.

strophic failure over a significant portion of the nanotube shell.

The fact that only the outer layers are affected indicates that electrical current in multiwall nanotubes may flow mainly in the outer carbon layers of the tube, in agreement with conclusions from magnetotransport experiments¹⁰.

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