# **LETTERS**

## Melanin, Nutrition, and the Lion's Mane

IN THEIR REPORT ON THE SEXUALLY SELECTED function of the lion's mane (*Panthera leo*), P. M. West and C. Packer ("Sexual selection, temperature, and the lion's mane," 23 Aug., p. 1339) assert that "[m]ane darkness indicates nutrition and testosterone and influences both female choice and male-male competition" (p. 1339). We contend that there is no evidence in this article or others supporting the idea that the melanin-based color of the lion's mane is dependent on nutritional state.

Image not available for online use.

West and Packer use correlational analyses to link mane color to nutrition. They show that the darkness of the mane is positively correlated with a male's age, natal and breeding site, testosterone titer, belly size, body temperature, and the average ambient temperature. None of these patterns, however, demonstrate that nutrition directly affects coat color. All of the correlations are equally or more consistent with a testosterone- and aggression-dependent control mechanism, as outlined by West and Packer. Testosterone regulates ornamental melanin pigmentation in skin, scales, feathers, hair, and fur (1-4)and mediates aggressive and competitive male behavior (5, 6) in many vertebrates.

In fact, we are not aware of any studies that show direct nutritional control of melaninpigmented fur in mammals. The study cited by West and Packer to suggest an effect of diet on pelage color concerns antelope foraging (7) and does not support this point. Several experimental studies of melanin-based plumage color in birds similarly report no relation between nutrition and melanin pigmentation (8-10).

To conduct the critical tests of the relative effects of hormones, social environment, and diet on pelage color, carefully controlled experiments are needed. Such experiments may not be feasible with large, wild mammals like lions. Field studies of large social animals have provided unique and invaluable insights into how animals interact in natural environments, but they are difficult systems in which to test the environmental controls and signal content of ornamental colors.

GEOFFREY E. HILL<sup>1</sup> AND KEVIN J. McGRAW<sup>2</sup>

<sup>1</sup>Department of Biological Sciences, Auburn
University, 331 Funchess Hall, Auburn, AL
36849–5414, USA. <sup>2</sup>Department of Neurobiology
and Behavior, Cornell University, Seeley G. Mudd
Hall, Room W211, Ithaca, NY 14853, USA.

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#### Response

#### ALTHOUGH WE AGREE THAT TESTOSTERONE IS

likely to be the central factor influencing the color of the lion's mane, we could not rule out the possibility that nutrition also plays a role. Because of space limitations, we were unable to report the results of a survey of captive lions that revealed a positive correlation between mane color and the zinc content in captive diets (Student's T = $2.02, r^2 = 0.083, P = 0.0516, N = 35$  captive males) (1). Dietary zinc actually influences testosterone production (2, 3), so it is possible that the testosterone-aggression model has a nutritional component; body storage of zinc is typically low (4), so lions may need a constant supply in their diet to maintain high levels of testosterone.

Nutritional studies reveal several other links between melanin production and nutrition. Melanin synthesis depends on the oxidation of tyrosine by tyrosinase (5), and diets deficient in tyrosine cause reductions in hair melanin in domestic cats that are corrected by diets high in tyrosine or phenylalanine (6). Copper is critical to the activity of tyrosinase (7, 8), and in captive and wild mammals, a common mechanism of hair depigmentation is copper deficiency either through low copper availability or unbalanced copper/ molybdenum ratios in forage (4, 9). The paper by Tear et al. (10) is one of several (11-13) linking copper deficiencies to decreased hair pigmentation in wild ungulates, and copper deficiencies can cause feather depigmentation in domestic fowl (14). Although copper deficiencies are probably less common in

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carnivores than in herbivores, the fact that copper is generally concentrated in the liver, and the liver is often the first part of a carcass to be consumed (15), suggests that individual carnivores that scavenge rather than hunt their prey (and thus have infrequent access to the liver) could have lower levels of copper in their diets and corresponding decreases in melanin production.

Although the relation between nutrition and melanin production is less clear than that between carotenoids and color patches in some fishes and birds, it would be premature to exclude nutrition as a factor influencing the color of the lion's mane.

#### **PEYTON M. WEST\* AND CRAIG PACKER**

Department of Ecology, Evolution and Behavior, University of Minnesota, 1987 Upper Buford Circle, St. Paul, MN 55108, USA.

\*To whom correspondence should be addressed. E-mail: west0302@umn.edu

References and Notes

- Mane color was assessed according to the protocol described in our Report. Diets were assessed according to advertised nutritional compositions of commercial diets and supplements.
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### Questions of Methodology

#### P. M. WEST AND C. PACKER'S REPORT

"Sexual selection, temperature, and the lion's mane" is a well-written study, but it may be based on some erroneous assumptions.

Why do the authors assume that the lions are attracted to particular stuffed toy "lions"

because of darker mane coloration rather than other variables such as the scent of the synthetic material, human handling, small size, or location? What controls were used?

The use of infrared thermatography to investigate thermoregulation and the effects of mane length is problematic because the temperature of the skin surface is dependent on many factors (e.g., hydration, color of debris, sun exposure, hair replacement cycle, season, subcutaneous circulation, muscle activity, etc.) and may not reflect the true subcutaneous temperature of the animal (1). Were internal temperatures taken (e.g., rectal) to validate the accuracy of the thermography?

Without definitive answers to these questions, one may assume that the authors' proposed hypothesis does not reflect the true nature of the physiology of the lion's mane.

#### E. J. NEIBURGER

Tsavo Research Program, 1000 North Avenue, Waukegan, IL 60085, USA.

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#### Response

**NEIBURGER RAISES QUESTIONS CONCERNING** the methodology of our studies of the lion's

mane. Regarding the model experiments, our tests involved three identical models. which were used in rotation so that they were each exposed equally to human handling and to real lions. Each mane was attached to the model with velcro, and the manes were randomly assigned to each model at each test. In areas with heterogeneous vegetation, mane length/color was assigned randomly to the spot with more cover. Our data revealed no significant effect of model or cover. Furthermore, the models were always placed downwind from the experimental subjects, so scent was highly unlikely to play a role in determining the lions' approach.

Neiburger asks whether we validated our infrared measurements with body temperature readings. Other studies have found significant correlations between internal body temperatures and surface temperatures measured with infrared thermography (1), but our field conditions only permitted the remote measurement of surface temperatures. For this reason, all of our arguments referred specifically to surface temperature, and we did not claim that our thermographic data directly demonstrated physiological costs. Instead, we sought (and found) "heat-related" costs

in dark-maned males: a higher percentage of abnormal sperm, as well as lower feeding rates in hot months of the year.

#### **PEYTON M. WEST\* AND CRAIG PACKER**

Department of Ecology, Evolution and Behavior, University of Minnesota, 1987 Upper Buford Circle, St. Paul, MN 55108, USA.

\*To whom correspondence should be addressed. E-mail: west0302@umn.edu

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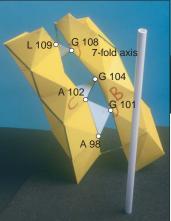
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## Cylindrical Channels from Concave Helices

THE CRYSTAL STRUCTURE OF THE BACTERIAL mechanosensitive channel (MscS) reveals a striking molecular architecture with seven-fold symmetry and a central channel formed from tightly packed  $\alpha$ -helices that partially span the membrane ("Crystal structure of *Escherichia coli* MscS, a voltage-modulated and mechanosensitive channel," R. D. Bass *et al.*, Research Articles, 22 Nov., p. 1582). We were struck by the fact that the  $\alpha$ -helices of the central channel are nearly straight; earlier, we had found  $\alpha$ -helices in an analogous assembly that curved and untwisted in order to lie on

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a cylindrical surface (1). Here, we present a simple model to explain how MscS can form cylindrical channels from straight helices.



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Triangulated representations of the MscS  $\alpha$ -helix. (Left) C $\beta$ /C $\alpha$  atoms. (Right) C $\alpha$  atoms only.

In the wider family of channels related to MscS, there is

absolute conservation of a pattern of glycines (GxxGxxxG). The MscS crystal structure shows that these glycines lie in a group on one side of each helix and form the packing interface with the next helix. The contacting residues of the neighboring

helix are alanines (A), and the molecular symmetry of the channel enforces the sequence pattern AxxGAxGxAxG.

A simplified, triangulated model of the helical interface between neighboring helices can be made by linking points representing the Cβ atoms or, in the case of the glycines, the  $C\alpha$  atoms. This is shown in the left panel of the figure. In the right panel is the triangulated representation using only the Cα positions; this shows that each helix is straight and fairly uniform (in fact, they are untwisted; i.e., they have a helical repeat of roughly 3.5 residues per turn). In contrast,

the  $C\beta/C\alpha$  helix has an apparent concavity on account of the three glycines, and this enables the two straight helices to fit snugly in a right-twisted arrangement.

The same pattern of glycines (GxxGxxxG) is absolutely conserved in the family of bacterial membrane proteins that are involved in proton-dependent transmembrane transport (TolQ, ExbB) and

flagellar rotation (MotA). We suggest that these will probably also form tubular, trans-membrane assemblies with a right-twisted arrangement. However, the residues flanking the conserved glycines are generally  $\beta$ -branched in these proteins and are rarely alanine; hence, the details of packing and stoichiometry are likely to differ from those observed in the MscS architecture.

### C. R. CALLADINE, <sup>1</sup> V. PRATAP, <sup>2</sup> V. CHANDRAN, <sup>2</sup> K. MIZUGUCHI, <sup>2</sup> B. F. LUISI <sup>2</sup>

<sup>1</sup>Department of Engineering, University of Cambridge, Cambridge CB2 1PZ, UK. <sup>2</sup>Department of Biochemistry, University of Cambridge, Cambridge CB2 1GA, UK.

#### Reference

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#### **CORRECTIONS AND CLARIFICATIONS**

**BREVIA:** "Information storage using supramolecular surface patterns" by M. Cavallini *et al.* (24 Jan. 2003, p. 531). In paragraphs four through six, there are several pieces of text that appear in the wrong place. In the last line of the text, the unit should be "10 to 100 Gbit/in²." A corrected PDF is available at www.sciencemag.org/cgi/reprint/299/5606/531.pdf.