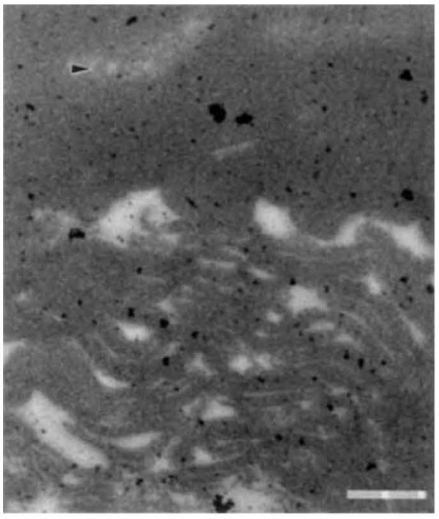

FIG. 2 Transmission electron micrograph showing a cross-section through the exospore of a single member of a dyad. The lightly stained area marked by the arrowhead is the spore surface. The material above this level is the outer region of the adjacent spore of this dyad. The material immediately below the arrowhead is the unlamellated outer exospore. The rest of the micrograph depicts the distinct lamellae of the inner exospore. Scale bar, 0.5 μm.

Exospores constructed of lamellae are nearly universal in modern land plant spores and pollen[2–4]. In most instances, the individual lamellae are highly fused at maturity, in some cases to the point of obscuring their separate status. In most, lamellated layers comprise a minority of the overall exospore thickness, but in hepatics, exospores consist almost exclusively of multiple, clearly separate lamellae. In particular, in the Sphaerocarpales there are several examples of multiple separate lamellae overlain by a more massive layer[5]. No other group of modern plants has this same combination of exospore ultrastructural features.

The lamellate structure of these spores suggests: (1) the early establishment of a developmental pattern common to all subsequent plant groups; and (2) the possibility that these early terrestrial pioneers may have belonged to a group of extant bryophytes.

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1. Gray, J *Phil. Trans. R. Soc.* B**309**, 167–195 (1985).
2. Brown, R.C. & Lemmon, B.E. in *Microspores, Evolution and Ontogeny* (eds Blackmore, S. & Knox, R.B.) 55–94 (Academic, London, 1990).
3. Rowley, J.R. & Southworth, D. *Nature* **213**, 703–704 (1967).
4. Blackmore, S. & Barnes, S.H. in *Microspores, Evolution and Ontogeny* (eds Blacksore, S. & Knox, R.B.) 173–192 (Academic, London, 1990).
5. Steinkamp, M.P. thesis (Univ. California, Santa Cruz, 1973).

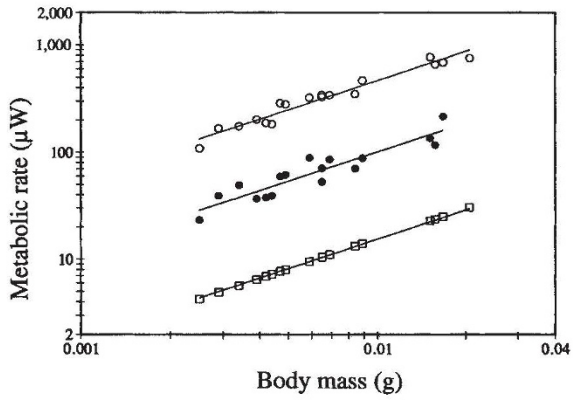
Larger bites of leaf-cutting ants

SIR — The maximum sustainable metabolic rate in non-flying animals ranges from about 8 to 12 times the resting or standard metabolic rate. This "aerobic scope" (maximum sustained metabolic/standard metabolic rate) increases to 20–100 for flying animals. Can scopes of this magnitude be attained without flight? To answer this question we investigated the leaf-cutting ant *Atta sexdens rubropilosa*, which cuts vegetation into convenient, portable fragments that it carries to the nest for processing by a resident fungal garden[1]. By dissection and weighing we ascertained that the mandibular muscles in *Atta* comprise >50% of their head capsule mass, or >25% of total body mass — comparable to the flight motor of flying insects[2].

By impregnating Parafilm with citrus essences, we created a metabolically inert pseudo leaf that stimulated leaf-cutting behaviour in foraging ants[3], whereupon we immediately placed the pseudo-leaf and attached, cutting worker in an ultra-sensitive flow-through respirometry system with high temporal resolution. Our sample of 18 ants (mean mass 7.89–5.41 mg, range 2.50 – 20.60 mg) cut at a mass-independent rate of 0.103 – 0.074 mm s^{-1}, equivalent to that reported for the cutting of tough or 'dense' leaves (0.1 mm s^{-1}, also mass-independent)[4]. The leaf-cutting metabolic rate was stable and dramatically above both the standard rate and the post-cutting locomotion metabolic rate. We found no evidence for anaerobic metabolism during leaf-cutting; after leaf-cutting, the metabolic rate fell within 30 s to levels characteristic of post-cutting locomotion.

Both leaf-cutting and post-cutting metabolic rates were strong functions of body mass (see figure). The aerobic scope of post-cutting locomotion was 6.7 ± 1.4, equivalent to the figure of 6.8 for unladen locomotion in *A. colombica*[5]. The aerobic scope of leaf-cutting (mean metabolic rate 49.2 ± 6.8 W kg^{-1}) was far larger, at 30.7 ± 3.9. Because of the near-identical mass scaling exponents of leaf-cutting metabolic rate and standard metabolic rates (see figure), this value is mass-independent.

By comparison, even the most prodigious feats of pedestrian locomotion yield aerobic scopes of approximately 12 in vertebrates[6,7] and invertebrates[5,8]. Even flying insects, the most metabolically active animals known, attain aerobic scopes of only 20 –100 (refs 2, 9; M. D. Dickinson and J. R. B. L., manuscript submitted). Leaf-cutting is obviously an energetically extraordinarily intense behaviour and constitutes a new record for metabolic activity not involving flight or flight-motor-associated endothermy. Indeed, when leaf-cutting metabolic rate is expressed specific to mandibular muscle mass it increases from 49.2 to 193.5 ± 33.7 W kg^{-1}, a figure approaching that for insect flight muscle, the most metabolically intense tissue known (300 –3,000 W kg^{-1}).



Leaf-cutting metabolic rate (open circles) and post-cutting, locomotory metabolic rate (closed circles) as functions of body mass in *A. sexdens* at 25 °C. Both regressions shared a mass scaling exponent of 0.895; mass scaling coefficients were 28,741 and 6,180, respectively (overall r^2 = 0.91, P < 0.0001). The relation between body mass and estimated standard metabolic rate[11] is also shown (squares). Leaf-cutting metabolic rate (duration 1-7 min) was elevated 31-fold, and post-cutting rate 7-fold, above the standard rate. Flow-through, CO_2-based respirometry[11] used a Sable Systems TR-3 respirometry system. The respiratory quotient of attine ants during activity is characteristic of lipid catabolism[4], corresponding to 27.6 J per ml CO_2, allowing us to calculate metabolic rate in SI energy units of μW or W kg^{-1}.

Thus, mandibular energetics may play an unexpectedly important role in ant foraging. Specifically, the effects of mandibular energetics on load- size selection and foraging efficiency (at the individual level) and on the occurrence and intensity of recruitment (at the colony level) have probably affected the evolu-

1. Hölldobler, B. & Wilson, E.O. *The Ants* Harvard Univ. Press, Cambridge, Massachusetts, 1990).
2. Casey, T.M. & Ellington, C.P. In *Energy Transformations in Cells and Organisms*, (eds Wieser, W. & Gnaiger, E.) 200–210 (Thieme, G. Stuttgart, 1989).
3. Roces, F. *Anim. Behav.* **40**, 1181–1183 (1990).
4. Roces, F. & Hölldobler, B. *Oecologia* **97**, 1–8 (1994).
5. Lighton, J.R.B., Bartholomew, G.A. & Feener, D.H. *Physiol. Zool.* **60**, 524–537 (1987).
6. Pasquis, P.A., Lacaisse, A. & Dejours, P. *Respir. Physiol.* **9**, 298–309 (1970).
7. Taylor, C.R. *et al. Respir. Physiol.* **44**, 25–37 (1981).
8. Full, R. J. in *Energy Transformations in Cells and Organisms* (eds Wieser, W. & Gnaiger, E.) 175–182 (Thieme, Stuttgart, 1989).
9. Ellington, C.P. *J. exp. Biol.* **160**, 71–91 (1991).
10. Wilson, E.O. *Behav. Ecol. Sociobiol.* **7**, 157–165 (1980).
11. Lighton, J.R.B. & Wehner, R. *J. comp. Physiol.* **163**, 11–17 (1993).

tion of worker polymorphism and foraging strategies in leaf-cutting ants[1,10], and may be significant in other ant genera.

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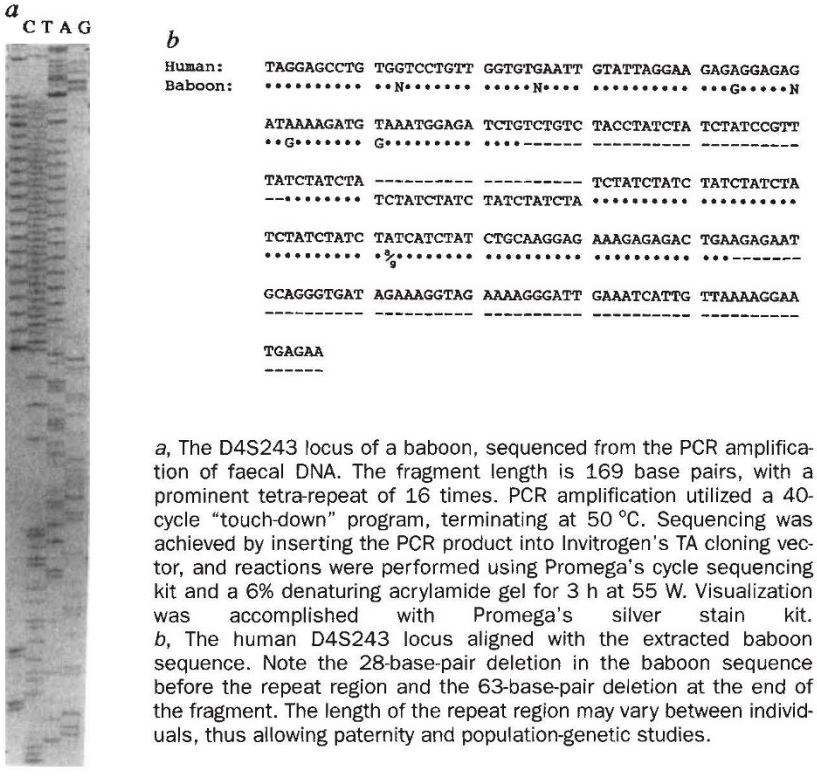
Nuclear DNA from primate dung

SIR — Although hypervariable DNA markers have revolutionized the study of kinship patterns and mating systems in the wild[1,2], these techniques have largely been restricted to species that can be easily captured and handled. Where handling is either unsafe or impracticable, DNA must be obtained non-invasively[3]. Collecting hair or buccal cells can be difficult or impossible in many species, so the ideal system should be based on material that is both plentiful and can be attributed unambiguously to a specific individual. Following the extraction of mitochondrial and chloroplast DNA from bear dung, by Höss *et al.*[4], we investigated whether useful samples of nuclear DNA could be obtained from the faeces of wild primates.

We collected dung from individually recognized olive baboons in the Gombe Stream National Park, Tanzania[5,6], and stored it first in liquid nitrogen, then in a −20 °C freezer. We modified a protocol for extracting mitochondrial DNA from chimpanzee dung[7], combining the techniques of Pääbo[8] and Boom[9] with a phenol/chloroform extraction with hexadecyltrimethylammonium bromide (CTAB) and collagenase. CTAB was added to the overnight incubation in order to break down ingested plant compounds that would inhibit subsequent PCR reactions. Extracted DNA was amplified using human nuclear microsatellite primer D4S243 (ref. 10) and PCR. As a control, we used chelex[3] to extract DNA from hair of several of the same individuals. PCR products were inserted into TA cloning vector (Invitrogen) and transformed into *Escherichia coli*. Colonies were grown overnight, plasmids were isolated and sequenced with the M13 forward primer.

Nuclear DNA extracted from dung and hair reveals that the baboon sequence at the D4S243 locus aligns with the human sequence, exhibiting three base substitutions, four ambiguous codes, and deletions of 28 and 63 base pairs (see figure). Human microsatellite primers successfully amplify baboon DNA in a significant number of cases (J. Rogers, personal communication), and paternity exclusions can usually be achieved by amplifying six polymorphic loci. PCR amplification has been successful in 39 out of 40 faecal samples tested so far. The ability to amplify microsatellite regions from faecally extracted nuclear DNA opens a new realm of possibilities for studying organisms that are difficult to catch or handle[11]. Dung should be considered essential material for paternity and population genetic studies of arboreal or endangered mammals.



a, The D4S243 locus of a baboon, sequenced from the PCR amplification of faecal DNA. The fragment length is 169 base pairs, with a prominent tetra-repeat of 16 times. PCR amplification utilized a 40-cycle "touch-down" program, terminating at 50 °C. Sequencing was achieved by inserting the PCR product into Invitrogen's TA cloning vector, and reactions were performed using Promega's cycle sequencing kit and a 6% denaturing acrylamide gel for 3 h at 55 W. Visualization was accomplished with Promega's silver stain kit. *b*, The human D4S243 locus aligned with the extracted baboon sequence. Note the 28-base-pair deletion in the baboon sequence before the repeat region and the 63-base-pair deletion at the end of the fragment. The length of the repeat region may vary between individuals, thus allowing paternity and population-genetic studies.

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1. Burke, T. *Trends ecol. Evol.* **4**, 139 (1989).
2. Queller, D.C., Strasssman, J.E. & Hughes, C. R. *Trends ecol. Evol.* **8**, 285 (1993).
3. Morin, P.A. *et al. Science* **265**, 1193 (1994).
4. Höss, M. *et al. Nature* **359**, 199 (1992).
5. Goodall, J. *The Chimpanzees of Gombe* (Harvard Univ. Press, Cambridge, 1986).
6. Packer, C. *et al. Nature* **373**, 60–63 (1995).
7. Goldberg, T. *Biotechniques* (in the press).
8. Pääbo, S., Gifford, J.A. & Wilson, A.C. *Nucleic Acids Res.* **16**, 9775–9787 (1988).
9. Boom, R. *et al. J. clin. Microbiol.* **28**, 495–503 (1990).
10. Hudsen *et al. Genomics* **13**, 622 (1992).
11. Kohn, M. *et al. Molec. Ecol.* (in the press).

Comparison of deep ice cores

SIR — Comparison of the GRIP and GISP2 deep ice cores from central Greenland[1,2] has confirmed the occurrence of exceptionally large, rapid changes in many climatic indicators over approximately the past 100,000 years[3,4]. Similar rapid changes occur within deeper ice with a warm isotope signature (identified as the Eemian, Sangamonian, or stage 5e[3,4]), but differences in their patterns between the two cores raise the possibility that at least one record was disturbed by ice-flow processes[1,2]. Disturbances might include boudinage, open folding or similar processes that would distort the time-depth relation but leave layers in stratigraphic order, or overturned folding that would disturb stratigraphic order. The GRIP and GISP2 steering bodies have initiated comparative studies of the two cores, including visible stratigraphy and crystal fabrics in selected sections of both cores, which are reported here. The most important results of these studies include the following.

(1) The small diameters of the cores and the lack of any highly reliable stratigraphic 'up' indicators prevent us from distinguishing overturned from right-side-up layers in structures much larger than the core diameter (13 cm for GISP2 and 10 cm for GRIP before sampling). We thus cannot invalidate or confirm the instability of the Eemian record.

(2) Both cores contain structures above the Eemian (which was identified as 2,790–2,865 m at GRIP[3,4]) that are larger than the core diameter and that could represent inverted strata. A 10–20-cm long region of layers dipping 20 ° relative