none of the mechanisms discussed above can be ruled out. Clarification of this question will require more high-quality PGE analyses coupled with sulphur data, allowing for rigorous comparison as well as determination of the lateral and vertical extent of the high-Pd mantle. □

1. Jagoutz, E. et al. Proc. lunar planet. Sci. Conf. **X,** 2031–2050 (1979).
2. Mitchell, R. H. & Keays, R. R. Geochim. cosmochim. Acta **45,** 2425–2445 (1981).
3. Morgan, J. W., Wandless, G. A., Petrie, R. K. & Irving, A. J. Tectonophysics **75,** 47–67 (1981).
4. Kimura, K., Lewis, R. S. & Anders, E. Geochim. cosmochim. Acta **38,** 683–701 (1974).
5. Morgan, J. W. J. geophys. Rev. **91,** 12375–12387 (1986).
6. Wasson, J. T. & Kallemeyn, G. W. Phil. Trans. R. Soc. Lond. A **325,** 533–544 (1988).
7. Hamlyn, P. H., Keays, R. R., Cameron, W. E., Crawford, A. J. & Waldron, H. M. Geochim. cosmochim. Acta **49,** 1797–1811 (1985).
8. Barnes, S. J., Naldrett, A. J. & Gorton, M. P. Chem. Geol. **53,** 303–323 (1985).
9. Garuti, G., Gorgoni, C. & Sighinolfi, G. P. Earth. planet. Sci. Lett. **70,** 69–87 (1984).
10. Page, N. J. & Talkington, R. W. Can. Mineral. **22,** 137–149 (1984).
11. Govindaraju, M. Geostandard Newsletter (spec. iss.) **VIII,** July (1984).
12. Spettel, B., Palme, H., Ionov, D. A. & Kogarko, L. N. Lunar planet. Sci. **XXII,** 1301–1302 (1991).
13. Greenough, J. D. & Fryer, B. J. Proc. ODP Sci. Res. **115,** 71–84 (1990).
14. Dowling, S. E. & Hill, R. E. T. Aust. J. Earth Sci. **39,** 349–363 (1992).
15. Brügmann, G. et al. Geochim. cosmochim. Acta **57,** 2001–2018 (1993).
16. Barnes, S. J. & Picard, C. P. Geochim. cosmochim. Acta **57,** 79–87 (1993).
17. Fabriès, J., Lorand, J. P., Bodinier, J. L. & Dupuy, C. J. Petrology (spec. lherzolite iss.) 55–76 (1991).
18. Downes, H., Bodinier, J. L., Thirlwall, M. F., Lorand, J. P. & Fabriès, J. J. Petrology (spec. lherzolite iss.) 97–115 (1991).
19. Bodinier, J. L., Dupuy, C. & Dostal, J. Geochim. cosmochim. Acta **52,** 2893–2907 (1988).
20. Reisberg, L. & Lorand, J. P. Nature **376,** 159–162 (1995).
21. Lorand, J. P., Bodinier, J. L., Dupuy, C. & Dostal, J. Geochim. cosmochim. Acta **53,** 3085–3090 (1989).
22. Barnes, S. J. & Francis, D. Econ. Geol. **90,** 135–154 (1995).
23. Hall, G. E. M. & Pelchat, J. C. Chem. Geol. **115,** 61–72 (1994).
24. Keays, R. R. Lithos **34,** 1–19 (1995).
25. McDonough, W. F. Lunar planet Sci. **XXVI,** 927–928 (1995).
26. Hart, S. & Zindler, A. Chem. Geol. **57,** 247–267 (1986).
27. Lorand, J. P., Keays, R. R. & Bodinier, J. L. J. Petrology **34,** 1111–1140 (1993).
28. Borisov, A., Palme, H. & Spettel, B. Geochim. cosmochim. Acta **58** 705–716 (1994).
29. Borisov, A. & Palme, H. Geochim. cosmochim. Acta **59,** 481–487 (1995).
30. Morgan, J. W., Walker, R. J. & Horan, M. F. Paper presented at Workshop on the Formation of the Earth's Core Mainz, 18–20 April (1994).
31. Hertogen, J., Janssens, M. J., Takahashi, H. & Anders, E. Geochim. cosmochim. Acta **47,** 2241–2255 (1983).
32. Morgan, J. W., Janssens, M. J., Takahashi, H., Hertogen, J. & Anders, E. J. Geochim. cosmochim. Acta **49,** 247–259 (1985).
33. Shirey, S. B. Mineralog. Mag. **58A,** 833–844 (1994).
34. Lorand, J. P. Earth planet. Sci. Lett. **93,** 50–64 (1989).
35. Anders, E. & Grevesse, N. Geochim. cosmochim. Acta **49,** 197–214 (1989).

Fossil evidence for the origin of the marsupial pattern of tooth replacement

Richard L. Cifelli*, Timothy B. Rowe†, W. Patrick Luckett‡, James Banta*, Reuben Reyes† & Robert I. Howes§

* Oklahoma Museum of Natural History and Department of Zoology, University of Oklahoma, Norman, Oklahoma 73019, USA
† Department of Geological Sciences, University of Texas, Austin, Texas 78712, USA
‡ Department of Anatomy, Medical Sciences Campus, University of Puerto Rico, P.O. Box 365067, San Juan, Puerto Rico 00936, USA
§ Department of Anatomical Sciences, University of Oklahoma, P.O. Box 26901, Oklahoma City, Oklahoma 73190, USA

EXTANT marsupials are distinctive in their pattern of dental development[1], in that only one tooth is replaced postnatally in each jaw. Interpretation of this pattern for marsupials ancestrally is disputed[2-5], partly because ontogenetic data in fossils have been unobtainable. Here we present an ultra-high-resolution X-ray computed tomography (CT) study of the tiny fossil Alphadon, which represents the first evidence of dental development and replacement in a Mesozoic marsupial. In the known pattern of tooth replacement and development, Alphadon is identical to living marsupials, a derived similarity suggesting that this pattern is ancestral to Marsupialia, and that it was established by the Late Cretaceous, at least. This pattern has been correlated with some specialized aspects of marsupial lactation[1,6]. Hence the presence of a marsupial pattern of tooth replacement in Alphadon provides indirect evidence that at least some specialized features of marsupial reproductive processes arose during the Mesozoic.

The adult dentition of primitive living marsupials includes three premolars and four molars, a configuration known to date to the Early Cretaceous[7] and thought to result from a derived developmental pattern within Theria[1]. Of these tooth positions, the last premolar locus (conventionally termed P3) is the only one in the entire dentition at which an erupted deciduous tooth is replaced postnatally by its successor. The homologies of the marsupial dentition are not universally accepted[2-5], and dispute usually

involves ontogenetic evidence. The adult dentition is thought to result from a unique developmental pattern[1], involving succession of vestigial teeth at incisor and canine loci, retardation and non-replacement of deciduous first (dP1) and second (dP2) premolars, normal replacement of a deciduous (dP3) by a successor (P3) tooth at the last premolar locus, and normal differentiation of the molars (M1–4). Thus the anterior two premolars of marsupials appear to be unreplaced deciduous teeth. Until now, little was known of the antiquity of the marsupial tooth-replacement pattern. Single jaws bearing presumed dP3 have been reported from the Late Cretaceous[8] and Paleocene[5,9], but definitive ontogenetic corroboration has been unobtainable.
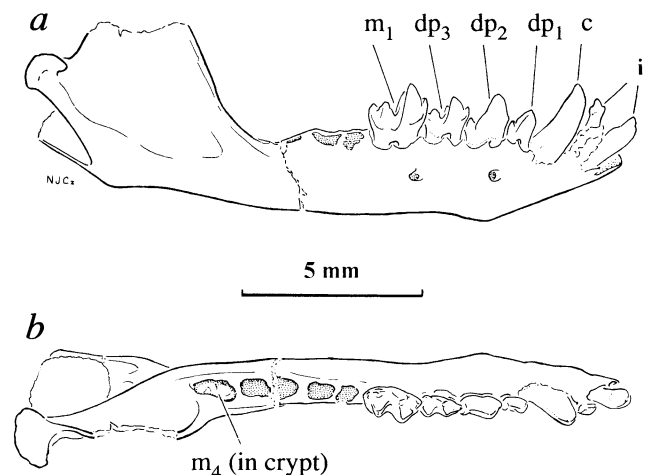
We have extracted ontogenetic data from an exceptionally

FIG. 1 Alphadon sp. (OMNH 27380), left mandible (reversed to correspond with CT images in Figs 2 and 4)) from the Upper Cretaceous part of the North Horn Formation, Utah[19]. Alphadon is a relatively long-lived, generalized taxon[20]; its contained species are distinguished primarily on the basis of upper molar features[2,8], so species referral of the specimen is uncertain. The specimen includes I_{2-3}?, the canine (c), dP_{1-2} (conventionally termed P_{1-2}), a molariform dP_3, M_1, alveoli for M_{2-3}, and a partly exposed crypt for M_4; the presence of dP_3 and the pre-eruptive stage of development of the last molar indicate that it is from a juvenile individual. a, Labial, and b, occlusal views.

complete mandible of the Cretaceous marsupial *Alphadon* (Fig. 1), using an ultra-high-resolution X-ray CT scanner (Fig. 2). Comparison was made to a dated developmental series of the small didelphid *Monodelphis domestica*, based on sectioned osteological specimens (Fig. 3), histological sections, and cleared and stained material, and to the small dasyurid *Sminthopsis virginiae*, based on histological sections (Fig. 4).

In dental development, *Alphadon* corresponds closely to living marsupials, which enables estimation of its developmental stage and indicates a tooth-replacement pattern identical in all observable details to the relatively stereotyped, distinctive pattern that characterizes Recent marsupials. The most significant developmental data are the degree of eruption and root formation of the canine, the pre-eruptive stage of the replacement P_3, and the degree of development and rotation of the last molar, all of which correspond to the dentition of *Monodelphis domestica* at 60–70 days of age. The developing P_3 of *Alphadon* lies lingual and deep to the erupted dP_3, as is the common pattern among mammals. Living marsupials are somewhat variable in the exact timing and spatial relationships of the developing P_3; in *Alphadon*, as in *M. domestica* and *S. virginiae*, it lies beneath the anterior root of dP_3, which undergoes some early resorption. The developing P_3 of *Alphadon* is higher in the jaw than in 60–70-day-old *M. domestica*, in this respect more closely resembling the 81-day *S. virginiae* (Fig. 4). In *S. virginiae*, P_3 begins erupting between 102–124 days, after M_4 has completely erupted (W.P.L. and P. Woolley, unpublished data); a similar pattern of late P_3 eruption also characterizes *Monodelphis* and most extant didelphid genera. A greater degree of calcification for M_4 than for P_3 in the juvenile *Alphadon* dentary is consistent with a comparable pattern of late eruption in the Cretaceous taxon. It is unlikely that tooth succession occurred at the anterior two premolar loci of *Alphadon* because there is no evidence of developing teeth in these positions, and the implanted teeth are completely erupted, with fully formed roots.

Many features commonly used to diagnose Marsupialia as monophyletic are either plesiomorphic, controversial or not observable in fossils[10]. The presence of a marsupial pattern of premolar replacement in *Alphadon* suggests that this character complex is diagnostic of the last common ancestor of *Alphadon* and living marsupials. Recent phylogenetic analyses differ on whether *Alphadon* is a basal marsupial or a proximate sister-taxon of marsupials within a more inclusive taxon, Metatheria (see refs 4, 11, 12). As yet there is no evidence, direct or indirect, bearing on the pattern of tooth replacement in deltatheroidans[13] or other putative fossil metatherians which have a postcanine tooth formula similar to that of marsupials (contrary to ref. 13). Despite this comparatively minor phylogenetic equivocation, it seems clear that the pattern of premolar replacement so distinctive of marsupials among living mammals arose during the Mesozoic.

The most distinctive characteristics of living marsupials are related to their reproductive biology, which is associated with birth of extremely altricial young and subsequent development during a period of extended maternal lactation[14–16]. Particularly characteristic of marsupial lactation is the 'period of fixation' during which young are continuously attached to teats for a prolonged period[17]. The highly modified pattern of replacement in the anterior dentition of marsupials has long been linked to developmental constraints imposed by the 'period of fixation'[1,6]. Neither the replacement pattern nor, by implication, nipple fixation is
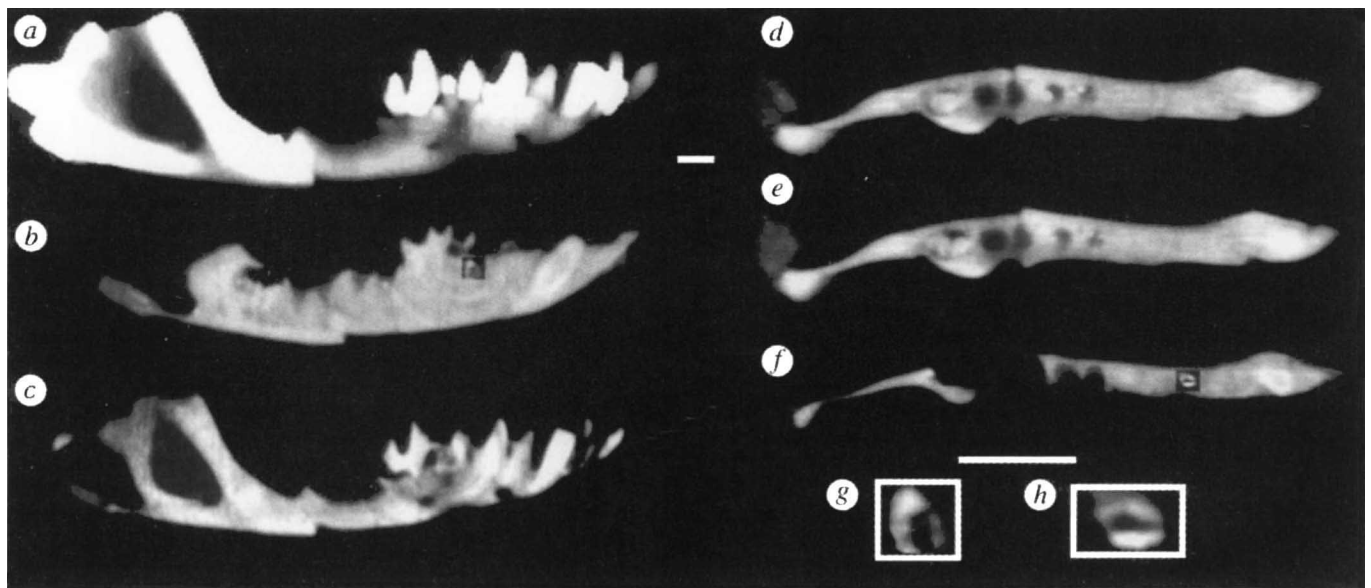

FIG. 2 CT scans of left mandible, *Alphadon* sp. (OMNH 27380). Examination of complete image sequences reveals that the canine has achieved nearly complete eruption and root formation; there is no evidence of unerupted teeth in the incisor/canine region, and we conclude that the erupted, rooted teeth at these positions represent true successors, as they are in extant marsupials[1]. *a–c, g*, Parasagittal sections; *d–f, h*, horizontal (occlusal) sections. *a*, Composite reconstruction of 123 parasagittal sections; note dark area, indicating poor ossification, in the dentary below dP_3. *b*, Section through the lingual part of the jaw, showing canine root, calcified protoconid of P_3 (in box below dP_3; see magnified inset, *g*), and part of the trigonid calcification for M_4. *c*, More labial section, showing space of poor ossification below dP_3 and mandibular foramen. *d*, Occlusal view showing base of M_4 trigonid. Also visible are the alveoli for dP_3, suggesting partial resorption of the roots. *e*, More dorsal section, showing some differentiation of trigonid cusps of M_4 and, possibly, a rudiment of its talonid. *f*, More dorsal section, showing the calcified protoconid (in box; see magnified inset, *h*) of P_3, somewhat lingually placed in the dentary. *g*, Magnified view of box in *b*, showing parasagittal section through calcification of developing P_3. *h*, Magnified view of box in *f*, showing coronal section through calcification of developing P_3. Scale bars, 1 mm.

METHODS. Data were obtained with an industrial scanner capable of two orders of magnitude greater resolution than conventional diagnostic scanners[21]; for this tiny specimen, it was configured with a 120 Kv microfocal X-ray source and a CCD detector that resolved the specimen into 123 parasagittal, 253 horizontal and 468 frontal sections taken at 18-μm (parasagittal, horizontal) and 36-μm (frontal) intervals. Consecutive sections were rendered into digital animations viewable on personal computers, following a protocol described elsewhere[21].

FIG. 3 Partial developmental series of the extant didelphid *Monodelphis domestica*: sectioned osteological specimens of right mandible. Each dated individual is represented by two parasagittal sections: left, region of M_4; right, selected views of the antemolar dentition. The following is a synopsis of relevant features and/or changes in the series (those not included in the figure are enclosed in parentheses). Day 51, canine in early eruption; pulp cavity broadly open with no root formation. (No macroscopic evidence for successor P_3; dP_3 roots complete.) Roots of M_3 short, broadly open to mandibular canal. M_4 calcification limited to protoconid, rotated about 30°. Day 57, canine somewhat more erupted, root beginning to form. Small, dorsally placed crypt for successor P_3 present under anterior part of dP_3, containing P_3 in presumed bell stage; no calcification evident. Roots of M_3 longer, but not extending into mandibular canal. M_4 with all trigonid cusps distinct, although possibly separate in part; trigonid rotated about 45°. Day 65, canine mostly erupted, root nearly complete. Chamber for successor P_3 enlarged, ventrally placed, containing well-developed protoconid cap with early calcification; chamber extends dorsally along lingual margin of anterior root of dP_3. Roots of M_3 extend into mandibular canal. M_4 with small heel; rotation unchanged. Day 70, canine erupted with fully formed but open root. Successor P_3 broader based (some resorption of dP_3 roots apparent). Roots of M_3 more elongate. M_4 talonid complete; rotation of tooth reduced to about 30°. Day 75, canine development appears complete. Protoconid of successor P_3 taller, better calcified. Day 85, both roots of dP_3 show clear resorption; chamber of P_3 more dorsoventrally extended, containing larger, better-calcified P_3. Roots of M_3 extend completely through mandibular canal. M_4 with both roots fully formed; rotation reduced to about 8°.
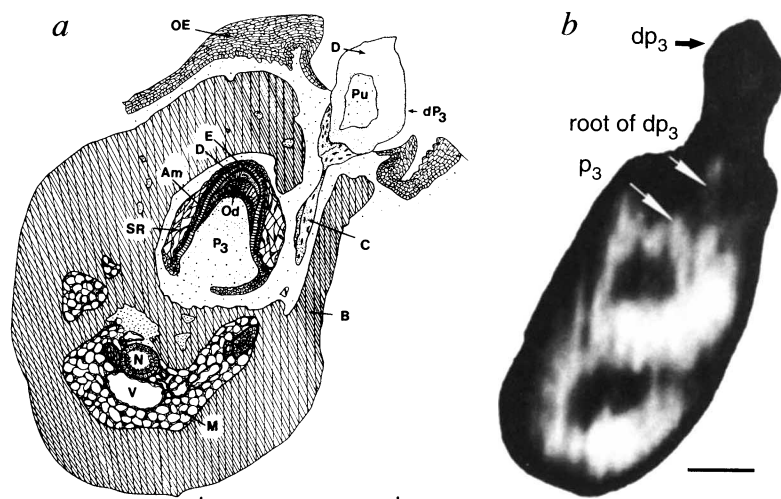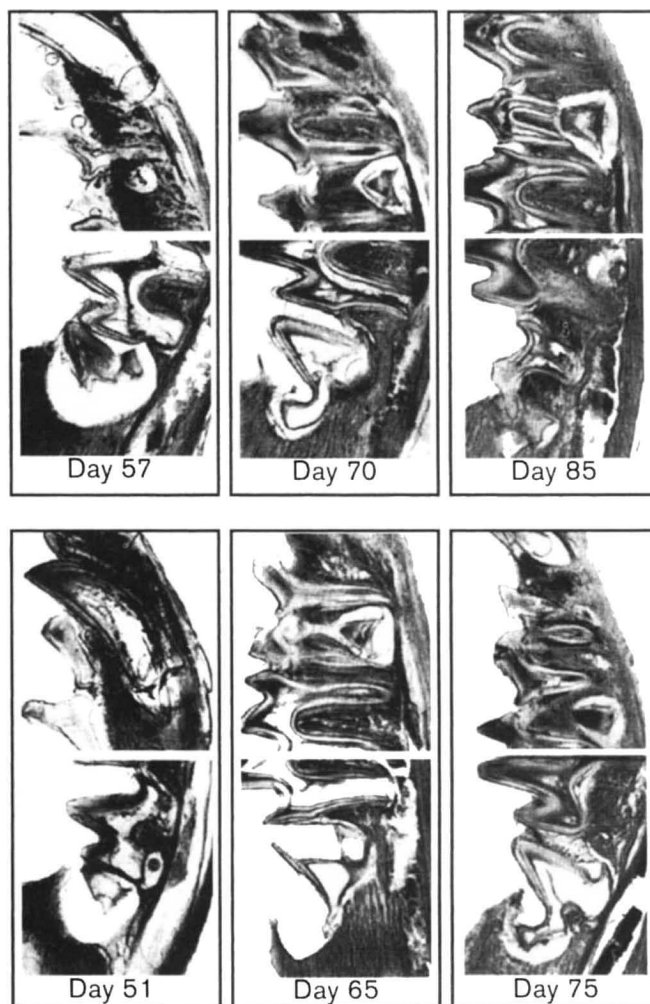

FIG. 4 Comparison of *a*, *Sminthopsis virginiae*, based on histological section of 81-day juvenile, and *b*, *Alphadon* sp. (OMNH 27380), based on CT section. Both sections are in the frontal plane, taken through the dP_3 and developing P_3. In both, dP_3 and part of its anterior root, labial to the P_3, can be seen; also shown are the mandibular canal, housing a neurovascular bundle while the animal was alive, and the calcification for the developing P_3, near the top of an unossified space within the dentary. In the histological section, most of the anterior root of dP_3 is out of the plane of section; only root cementum (C) is seen. Only the apex of the P_3 protoconid is calcified, and it is capped by a moderately thick layer of dentin (D) and a thin layer of enamel (E). A well-developed neurovascular bundle, composed of nerve fibres (N), venules (V) and small arterioles, is partly enveloped by spongy marrow (M) deep to the developing P_3. Other abbreviations: Am, amelobasts; B, alveolar bone; Od, odontoblasts; OE, oral epithelium; Pu, pulp; SR, stellate reticulum. Scale bar, 1 mm.

likely to have characterized the common therian ancestor of marsupials and eutherians[1,18] because the latter are primitive in replacing most or all of the antemolar dentition. Although characteristics of the reproductive system cannot be observed directly in fossils, the modified developmental pattern in the anterior dentition of marsupials provides indirect evidence of nipple fixation[1]. Thus the developmental pattern seen in *Alphadon* suggests that at least some reproductive specializations of marsupials, including nipple fixation, were probably established during the Mesozoic, earlier than previously suggested[15]. Other aspects of the ancestral therian and metatherian reproductive patterns remain to be elucidated. □

1. Luckett, W. P. in *Mammal Phylogeny* Vol. 1 (eds Szalay, F. S., Novacek, M. J. & McKenna, M. C.) 182–204 (Springer, New York, 1993).
2. Clemens, W. A. in *Mesozoic Mammals* (eds Lillegraven, J. A., Kielan-Jaworowska, Z. & Clemens, W. A.) 192–220 (University of California, Berkeley, 1979).
3. Archer, M. *Mem. Queensland Mus.* **18**, 157–164 (1978).

4. Reig, O. A., Kirsch, J. A. & Marshall, L. G. in *Possums and Opossums: Studies in Evolution* (ed. Archer, M.) 1–89 (Surrey Beatty, Sydney, 1987).
5. Marshall, L. G. & de Muizon, C. *Natn. geogr. Res.* **4**, 23–55 (1988).
6. Wilson, J. T. & Hill, J. P. Q. *Jl microsc. Sci.* **39**, 427–588 (1897).
7. Cifelli, R. L. *Proc. natn. Acad. Sci. U.S.A.* **90**, 9413–9416 (1993).
8. Clemens, W. A. *Univ. Calif. Publ. geol. Sci.* **62**, 1–122 (1966).
9. Muizon, C. de in *Fósiles y Facies de Bolivia* (ed. Suarez Soruco, R.). *Rev. téc. Yac. Petrol. Fisc. Bolivia* **12**, 575–624 (1992).
10. Muizon, C. de *Nature* **370**, 208–211 (1994).
11. Marshall, L. G., Case, J. A. & Woodburne, M. O. in *Current Mammalogy* Vol. 2 (ed. Genoways, H. H.) 433–505 (Plenum, New York, 1990).
12. Szalay, F. S. *Evolutionary History of the Marsupials and an Analysis of Osteological Characters* (Cambridge Univ. Press, New York, 1994).
13. Kielan-Jaworowska, Z. & Nessov, L. A. *Lethaia* **23**, 1–10 (1990).
14. Lillegraven, J. A. *Evolution* **29**, 707–722 (1976).
15. Tyndale-Biscoe, H. & Renfree, M. *Reproductive Physiology of Marsupials* (Cambridge Univ. Press, 1987).
16. Renfree, M. in *Mammal Phylogeny* Vol. 1 (eds Szalay, F. S., Novacek, M. J. & McKenna, M. C.) 4–20 (Springer, New York, 1993).
17. Hill, J. P. & O'Donoghue, C. H. Q. *Jl. microsc. Sci.* **59**, 133–174 (1913).
18. Luckett, W. P. *J. mamm. Evol.* **2**, 255–283 (1994).
19. Speiker, E. M. *Int. geol. Congr.* 21 **5**, 14–24 (1960).
20. Eaton, J. G. *J. vert. Paleont.* **13**, 105–124 (1993).
21. Rowe, T. B., Carlson, W. D. & Bottorff, W. W. *Thrinaxodon Digital Atlas of the Skull* (Univ. Texas Press, Austin, CD-ROM, 623Mb, 1993).

Productivity and sustainability influenced by biodiversity in grassland ecosystems

David Tilman*, David Wedin† & Johannes Knops*

* Department of Ecology, Evolution and Behavior, University of Minnesota, St Paul, Minnesota 55108, USA
† Department of Botany, University of Toronto, Toronto, Ontario M5S 3B2, Canada

THE functioning and sustainability of ecosystems may depend on their biological diversity[1–8]. Elton's[9] hypothesis that more diverse ecosystems are more stable has received much attention[1,3,6,7,10–14], but Darwin's proposal[6,15] that more diverse plant communities are more productive, and the related conjectures[4,5,16,17] that they have lower nutrient losses and more sustainable soils, are less well studied[4–6,8,17,18]. Here we use a well-replicated field experiment, in which species diversity was directly controlled, to show that ecosystem productivity in 147 grassland plots increased significantly with plant biodiversity. Moreover, the main limiting nutrient, soil mineral nitrogen, was utilized more completely when there was a greater diversity of species, leading to lower leaching loss of nitrogen from these ecosystems. Similarly, in nearby native grassland, plant productivity and soil nitrogen utilization increased with increasing plant species richness. This supports the diversity–productivity and diversity–sustainability hypotheses. Our results demonstrate that the loss of species threatens ecosystem functioning and sustainability.

The diversity–productivity hypothesis is based on the assumption that interspecific differences in the use of resources by plants allow more diverse plant communities to utilize more fully limiting resources and thus attain greater productivity[6,8,17,18]. A related hypothesis is that nutrient leaching losses from ecosystems should be a decreasing function of plant diversity because of greater nutrient capture and/or immobilization in more diverse ecosystems[4,5,17]. Taken together, these lead to the diversity–sustainability hypothesis; that the sustainability of soil nutrient cycles and thus of soil fertility depends on biodiversity. But the relationships between biodiversity and ecosystem functioning remain controversial[3,10,13,18–23] because existing field data are from studies that lacked direct experimental control of biodiversity or sufficient replication. Only direct control of diversity allows attribution of responses to diversity rather than to other correlated factors[8,18,20–23]. Moreover, the species composition of plots must be

randomly determined to avoid biases caused by the traits of particular species[20,21].

Here we report results of a field experiment in which the number of plant species was experimentally controlled (Fig. 1 legend). Our 147 plots, located on nitrogen-limited soil[24], were planted with either 1, 2, 4, 6, 8, 12, or 24 species. The species assigned to each plot were chosen by a separate random draw of the appropriate number of species from a pool of 24 North American prairie species. The impacts of diversity on plant productivity, nutrient capture and nutrient leaching were observed during the second year of growth. We also sampled a native grassland to determine the relationships between these variables in an undisturbed mature ecosystem.

Treatments created an experimental biodiversity gradient. Plant species richness, Shannon diversity (H'), and effective species richness ($e^{H'}$; Fig. 1a) were all significantly correlated with the number of species seeded into the plots (Pearson's $r = 0.81$, $r = 0.74$, $r = 0.75$, respectively; $n = 147$, $P < 0.001$ for all). Two measures of peak standing crop (our estimate of plant productivity) were positively correlated with the species-richness treatment (total plant cover: $r = 0.39$, $n = 147$, $P < 0.001$, Fig. 1b; biomass estimated by light penetration: $r = 0.27$, $n = 147$, $P < 0.001$). Both estimates were similarly dependent on observed plot species richness ($r_{cover} = 0.55$, $r_{biomass} = 0.42$, $n = 147$, $P < 0.001$ for both) and effective species richness ($r_{cover} = 0.29$, $r_{biomass} = 0.29$, $n = 147$, $P < 0.001$ for both). Thus, greater plant diversity led to greater productivity during the second year of ecosystem establishment.

Rooting-zone extractable soil NO_3^- was a decreasing function of species richness (Fig. 1c). Extractable NH_4^+ had a similar pattern ($r = -0.18$, $n = 147$, $P = 0.03$). This indicates that more species-rich plots more fully utilized soil mineral nitrogen, the main limiting resource[24]. Below the rooting zone, soil NO_3^- concentrations

TABLE 1 Factors influencing plant total cover (productivity) in the biodiversity experiment

Variable	Parameter	Student's t	P
Intercept	18.9	3.82	0.0002
Species richness	1.31	6.72	0.0001
Rooting zone NO_3^-	−26.6	−2.20	0.03
Rooting zone NH_4^+	7.82	1.65	0.10
Root mass	2.96	5.00	0.0001

Multiple regression of total plant cover (dependent variable) on listed variables, each measured in all 147 plots. Overall $F_{4,142} = 28.5$, $P < 0.0001$, $R^2 = 0.45$.

TABLE 2 Factors influencing soil NO_3^- in the rooting zone in the biodiversity experiment

Variable	Parameter	Student's t	P
Intercept	0.33	12.0	0.0001
Species richness	−0.005	−2.74	0.007
Plant total cover	−0.001	−1.56	0.12
Root mass	0.003	0.14	0.89

Multiple regression of rooting zone soil NO_3^- on listed variables, each measured in all 147 plots. Overall $F_{3,143} = 7.39$, $P < 0.0001$, $R^2 = 0.13$.