were identified, and that each of these cosmids detected transcripts of the same size in Northern blot experiments, suggest that only one highly expressed gene (that is, *gli*) was present within the amplification unit.

The assignment of the sequences amplified in D-259 MG cells to chromosome 12 (q13 to q14.3) is provocative. A fragile site of the folic acid type has been described at 12q13 (*17*); such sites have been hypothesized to colocalize with proto-oncogenes and play a role in tumor development in affected individuals (*18*). In addition, translocations in the 12(q13 to q14) region have been described in human myxoid liposarcomas and salivary gland tumors (*19*). There is a precedent for genes that are amplified in some tumors to be translocated in other tumors and thus to show similar increases in expression (*1*). This hypothesis can now be tested in liposarcomas and salivary gland tumors with probes for *gli*.

REFERENCES AND NOTES

1. H. Land, L. F. Parada, R. A. Weinberg, *Science* **222**, 771 (1983); K. Alitalo, *Med. Biol.* **62**, 304 (1984); C. M. Croce, *Cancer Res.* **46**, 6019 (1986).
2. S. H. Bigner, J. Mark, D. D. Bigner, *Cancer Genet. Cytogenet.* **24**, 163 (1987).
3. G. R. Stark and G. M. Wahl, *Annu. Rev. Biochem.* **53**, 447 (1984); R. T. Schimke, *Cell* **37**, 705 (1984); J. L. Hamlin, J. D. Milbrandt, N. H. Heintz, J. L. Azizkhan, *Int. Rev. Cytol.* **90**, 31 (1984).
4. I. B. Roninson, *Nucleic Acids Res.* **11**, 5413 (1983).
5. K. W. Kinzler *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **83**, 1031 (1986).
6. Genomic DNA (200 μg) was cleaved with Hind III, and the fragments were separated by electrophoresis through a 1% agarose gel. While in the gel, the DNA was denatured and renatured as described for the analytical assay for amplified sequences (see Fig. 1). However, instead of treating the gel with S1 nuclease, a region of the gel previously shown to have a large number of amplified restriction fragments (2.6 to 5.3 kb) was excised, and the DNA was electroeluted from the gel [I. B. Roninson, H. T. Abelson, D. E. Housman, N. Howell, A. Varshavsky, *Nature (London)* **309**, 626 (1984)]. The eluted DNA was ligated into the Hind III site of the vector pKB358 [T. M. Roberts, S. L. Swanberg, A Poteete, G. Riedel, K. Backman, *Gene* **12**, 123 (1980)]. Only DNA that was double-stranded after the denaturation-renaturation step could recombine with pKB358 to form a viable plasmid, and the library prepared from this DNA was enriched for amplified sequences.
7. S. J. O'Brien, W. G. Nash, J. L. Goodwin, D. R. Lowy, E. H. Chang, *Nature (London)* **302**, 839 (1983); S. J. O'Brien, T. I. Bonner, M. Cohen, C. O'Connell, W. G. Nash, *ibid.* **303**, 74 (1983).
8. A. S. Lidsky *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **82**, 6221 (1985).
9. L. J. van't Veer, A. G. van Kessel, H. van Heerikhuizen, A. van Ooye, R. Nusse, *Mol. Cell. Biol.* **4**, 2532 (1984).
10. J. W. M. Hoppener *et al.*, *Hum. Genet.* **69**, 157 (1985).
11. N. M. Nau *et al.*, *Nature (London)* **318**, 69 (1985); E. Stavnezer, D. S. Gerhard, R. C. Birmari, I. Balazs, *J. Virol.* **39**, 920 (1981); C. C. Morton *et al.*, *Science* **223**, 173 (1984); C. S. Cooper *et al.*, *Nature (London)* **311**, 29 (1984); Y. Tsujimoto *et al.*, *Science* **224**, 1403 (1984); Y. Tsujimoto, L. R. Finger, J. Yunis, P. C. Nowell, C. M. Croce, *ibid.* **226**, 1097 (1984); C. M. Croce *et al.*, *ibid.* **227**, 1044 (1985); J. Erickson, D. L. Williams, J. Finan, P. C. Nowell, C. M. Croce, *ibid.* **229**, 784 (1985); M. M. Le Beau, C. A. Westbrook, M. O. Diaz, J. D. Rowley, M. Oren, *Nature (London)* **316**, 826 (1985).
12. DNA from D-259 MG was partially cleaved with Mbo I and size-fractionated by centrifugation through a sucrose gradient. Size-fractionated DNA (30 to 45 kb) was cloned in the cosmid vector pTL5 [M. Steinmetz, A. Winoto, K. Minard, L. Hood, *Cell* **28**, 489 (1982)].
13. DMs were isolated from D-259 MG cells by differential centrifugation of metaphase chromosomes as described by C. C. Lin *et al.* [*Chromosoma* **92**, 11 (1985)]. DNA prepared from these DMs contained the equivalent of more than a 1000-fold amplification (that is, 15-fold enrichment by differential centrifugation × 75-fold original amplification). Therefore, an amplified sequence should have had a signal 1000 times as strong as that of any single-copy sequence when labeled DM DNA was used as a hybridization probe. However, hybridization signals from repeated DNA sequences in the DM DNA probe would mask signals from amplified sequences because such repeated sequences occur over 10,000 times in the human genome. This problem with repeated sequences was eliminated by preannealing the hybridization probe to a large excess of human placental DNA as described by P. G. Sealey, P. A. Wittaker, and E. M. Southern [*Nucleic Acids Res.* **13**, 1905 (1985)].
14. A. J. Wong *et al.*, in preparation.
15. No transforming activity was detected in DNA from the D-259 MG cell line in NIH 3T3 transfection experiments performed in collaboration with S. Needleman.
16. DNA was purified from frozen or paraffin-embedded malignant gliomas [as described in A. J. Wong *et al.*, *Science* **233**, 461 (1986)], slot blotted onto nitrocellulose filters, and screened by hybridization with the pKK36P1 probe. Two glioblastoma multiforme tumors (D-259 MG and one other tumor) out of the 45 tested contained more than a tenfold amplification of the sequences detected by pKK36P1. This was confirmed by Southern blotting of Pst I–cleaved DNA, which showed that a 1.55-kb Pst I fragment homologous to pKK36P1 was amplified in both tumors.
17. F. Giraud, S. Ayme, J. F. Mattei, M. G. Mattei, *Hum. Genet.* **34**, 125 (1976); G. R. Sutherland and L. Hinton, *ibid.* **57**, 217 (1981).
18. J. J. Yunis, *Science* **221**, 227 (1983).
19. J. Limon, C. Turc-Carel, P. Dal-Cin, U. Rao, A. A. Sandberg, *Cancer Genet. Cytogenet.* **22**, 93 (1986); M. Mark and R. Dahlenfors, *Anticancer Res.* **6**, 299 (1986).
20. S. E. Goelz, S. R. Hamilton, B. Vogelstein, *Biochem. Biophys. Res. Commun.* **130**, 118 (1985).
21. J. M. Trent, S. Olson, R. M. Lawn, *Proc. Natl. Acad. Sci. U.S.A.* **79**, 7809 (1982).
22. U. Gubler and B. J. Hoffman, *Gene* **25**, 263 (1983).
23. D. W. Cleveland *et al.*, *Cell* **20**, 95 (1980).
24. V. Glisin, R. Crkvenjakov, C. Byus, *Biochemistry* **13**, 2633 (1974).
25. K. H. Cox, P. V. DeLeon, L. M. Angerer, R. C. Angerer, *Dev. Biol.* **101**, 485 (1984); M. E. Harper, L. M. Marselle, R. C. Gallo, F. Wong-Staal, *Proc. Natl. Acad. Sci. U.S.A.* **83**, 772 (1986).
26. Y.-h. Xu *et al.*, *Nature (London)* **309**, 806 (1984).

27. We thank C. Jones for providing DNA for hybrid lines F11-1 and F11-13, S. Needleman for performing the NIH/3T3 transformation assay, and P. White and S. Smith for preparing the manuscript. Supported by a National Cancer Institute training grant CA-09243 (K.W.K. and A.J.W.), and NIH grants CA-43722 (S.H.B.), NS-20023 (S.H.B. and D.D.B.), CA-11898 (D.D.B.), CA-29476 (J.M.T.), CA-41183 (J.M.T.), GM-33903 (M.L.L.), and CA-43460 (B.V.).

23 October 1986; accepted 15 January 1987

Superconductivity in Alkaline Earth–Substituted La_2CuO_{4-y}

J. GEORG BEDNORZ, K. ALEX MÜLLER, MASAAKI TAKASHIGE*

La_2CuO_{4-y} ceramics containing a few percent of Ca^{2+}, Sr^{2+}, and Ba^{2+} ions have been prepared. Resistivity and susceptibility measurements exhibit superconductive onsets (as in earlier Ba^{2+}-containing samples). The onset temperature La_2CuO_{4-y} with Sr^{2+} is higher and its superconductivity-induced diamagnetism larger than that found with Ba^{2+} and Ca^{2+}. This is proof that the electronic change resulting from alkaline earth–doping, rather than the size effect, is responsible for superconductivity. The ionic radius of Sr^{2+} is close to that of La^{3+} for which it presumably substitutes.

RECENTLY, BEDNORZ AND MÜLLER (*1*) reported the possible onset of superconductivity in the metallic BaLaCuO system with transition temperatures T_c in the 30 K range. Subsequent dc susceptibility measurements in our laboratory (*2*) supported this, and resistivity and low-field susceptibility data from Tanaka's group (*3*) and Chu's (*4*) group substantiated this finding. The early BaLaCuO samples consisted of up to three phases (*2*); by combining x-ray and diamagnetic susceptibility measurements (*5, 6*), it could be shown that one of them, the La_2CuO_{4-y}:Ba phase, becomes partly superconducting. Identification of flux trapping and the existence of a superconductive glass state in the ceramics was demonstrated by nonergodic magnetic responses (*7*).

Leading to the discovery of superconductivity in the La_2CuO_{4-y} were, first, the known existence of high electron-phonon coupling in oxides such as $Li_xTi_{2-x}O_4$ (*8*) spinel and $BaPb_{1-x}Bi_xO_3$ (*9*) perovskite, and, second, the expected high electron-phonon coupling occurring in polaronic systems as emphasized by Chakraverty (*10*). The existence of Jahn-Teller polarons in a linear chain model was proposed by Höck *et al.* (*11*): If the Jahn-Teller stabilization energy is of the same magnitude as the bandwidth in a metal, the effective mass of the itinerant electrons or holes will become large, that is, a large electron-phonon coupling exists. As a result, such a polaronic system can be favorable for the occurrence of superconductivity. The Cu^{2+} ions in the itinerant compound La_2CuO_{4-y}

IBM Research Division, Zurich Research Laboratory, 8803 Rüschlikon, Switzerland.

*Permanent address: Institute for Solid State Physics, University of Tokyo, Roppongi, Minatoku, Tokyo 106, Japan.

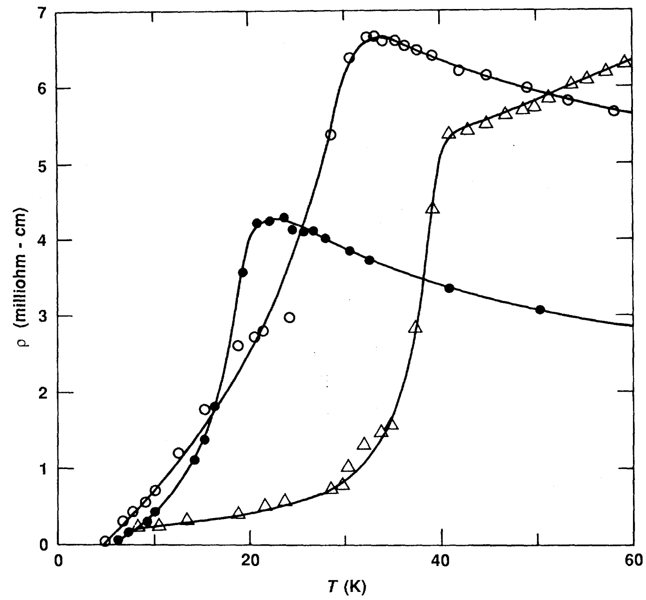

Fig. 1. Resistivity as a function of temperature for calcium (●), strontium (△), and barium (○) substitution with substituent lanthanum ratios of 0.02/1.98, 0.02/1.8, and 0.15/1.85, respectively. The strontium curve has been vertically expanded by a factor of 15.

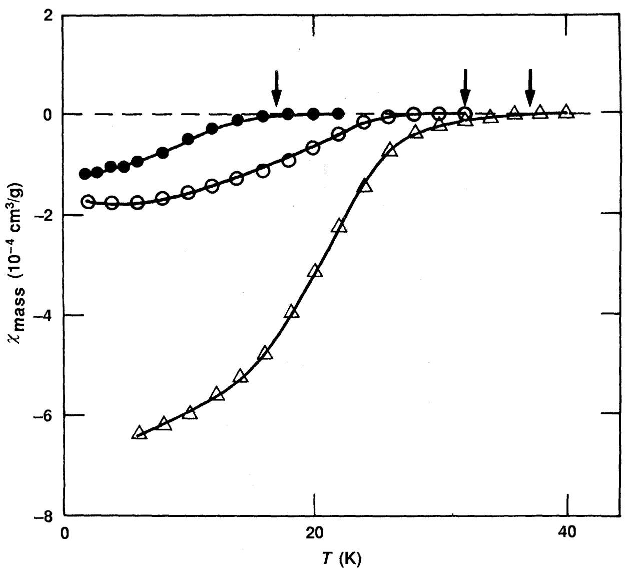

Fig. 2. Magnetic susceptibility of the samples whose resistivities are shown in Fig. 1. The substituents are calcium (●), strontium (△), and barium (○), with total sample masses of 0.14, 0.21, and 0.13 g, respectively. The calcium curve has been vertically expanded by a factor of 10. Arrows indicate onset temperatures.

have this property. However, the experiments showed that Ba^{2+} doping is essential for the occurrence of superconductivity (*1–7*). The ionic radius of Ba^{2+} is 1.34 Å, that of La^{3+} is 1.14 Å, whereas that of Cu^{2+} is 0.72 Å (*12*). By comparing these sizes, one expects that the Ba^{2+} substitutes La^{3+} rather than Cu^{2+}. There are, in principle, two possible mechanisms consistent with these observations which result in the occurrence of superconductivity. (i) The first is an electronic mechanism. The Ba^{2+} substitution causes a mixed-valence state of Cu^{2+} and Cu^{3+}, and this state serves to preserve charge neutrality. It is assumed that the oxygen deficiency *y* is the same in the doped and undoped crystallites, an assumption that needs further investigation. (ii) The second could be a size effect. The larger Ba^{2+} induces an orthorhombic-tetragonal symmetry change as found by room-temperature x-ray diffraction. La_2CuO_{4-y}:Ba samples with nearly tetragonal symmetry exhibit the highest transition temperatures. This symmetry change, however, is also obtained by doping La_2CuO_{4-y} with Sr^{2+} or Ca^{2+} ions, which have radii of 1.12 and 0.99 Å, respectively. We are therefore able to disprove the size-mismatch mechanism (ii) in favor of the electronic, or more accurately, the electron-phonon mechanism (i).

Our oxygen-deficient compounds have been prepared by the coprecipitation process with subsequent low-temperature treatment (*1, 2*) and by high-temperature solid-state reaction from the corresponding oxides and carbonates at 1100°C (*5, 13*). Although in the case of barium substitution, if we start from the ratio (La, Ba):Cu = 2:1, which has been identified as the composition of the superconducting compound (*2*), the low-temperature process yields a two-phase product. A single-phase sample, however, is obtained by proper annealing, whereas in the case of calcium and strontium substitution single-phase samples were prepared without additional treatments (*5*).

As the samples are cooled from room temperature, the resistivity ρ first shows a metal-like decrease. At low temperatures (Fig. 1), a change to an increase occurs in the case of calcium compounds as well as in the barium-substituted samples (*1*). This increase is followed by a resistivity drop, showing the onset of superconductivity at 22 ± 2 K and 33 ± 2 K for the calcium and barium compounds, respectively; ± ranges represent uncertainties due to rounding effects at the onset. In the strontium compound, the resistivity remains metallic down to the resistivity drop at 40 ± 1 K. The presence of localization effects, however, depends strongly on alkaline-earth ion concentration and sample preparation, in particular annealing conditions and the density, which have to be optimized. In strontium-substituted La_2CuO_4 with concentration $x = 0.2$ produced by the high-temperature reaction and subsequent oxygen annealing, Cava *et al.* (*14*) were able to dramatically narrow the transition width to 1.4 K with an onset of 38.5 K. All our samples with low concentrations of calcium, strontium, and barium show a strong tendency toward localization before the resistivity drops occur.

In each case, at temperatures somewhat lower than the onset of the resistivity drops, the magnetic susceptibility measurements (Fig. 2) show a transition from Pauli paramagnetic behavior to diamagnetism. The changes to diamagnetism occur at 17 ± 1, 32 ± 1, and 37 ± 1 K for calcium, barium, and strontium, respectively. Values of susceptibility χ shown here are equilibrium values obtained by field cooling at 0.5 K/min. Zero-field cooling the samples results in a considerably larger metastable diamagnetism (*2, 7*). For the strontium compound, the value of the volume susceptibility is estimated to be several percent of $-1/4\pi$ at 6 K in a relatively high magnetic field of 0.3 kG, as compared to the Li_xTiO_4 spinel and the oxygen-annealed La_2CuO_4:Sr samples studied by Cava *et al.* (*14*). For lower probing fields, χ is considerably enhanced [see inset of figure 4 in (*2*)]; this is expected theoretically (*15*) in a superconductive glass. Most recently, we proved the existence of such a superconductive glass state in La_2CuO_{4-y}:Ba from measurements of $\chi(T)$ and magnetization $m(T)$ in zero-field and nonzero-field cooled states by the presence of metastability (*7*).

Our resistivity and susceptibility measurements, as a function of temperature, of Sr^{2+}- and Ca^{2+}-doped La_2CuO_{4-y} ceramics (Figs. 1 and 2) show the same general tendency as the Ba^{2+}-doped samples (*1, 2, 7*): that is, a drop in $\rho(T)$, and at a slightly lower temperature a crossover to diamagnetism. The samples containing Sr^{2+} actually have yielded a higher onset temperature than those containing Ba^{2+} and Ca^{2+}. Furthermore, the diamagnetic susceptibility is about three times as large as for the barium samples. As the ionic radius of Sr^{2+} nearly matches the one of La^{3+}, it is clear that the size effect does not cause the occurrence of superconductivity. On the contrary, it is rather adverse, as the data on Ba^{2+} and Ca^{2+} indicate.

The highest T_c for each of the dopant-ions investigated occurs for those concentrations where, at room temperature, the La_2CuO_{4-y} structure is close to the orthorhombic-tetragonal structural phase transition (SPT) (*5*). Thus, this SPT may be related to the substantial electron-phonon interaction enhanced by the substitution. The alkaline earth–substitution of lanthanum is clearly important, and quite likely creates Cu^{3+} ions with two orbitals transforming as orbitals of the cubic e_g group which are half-filled and have a singlet ground state. Therefore, the absence of the third electron in the e_g states, which introduces a Jahn-Teller hole near the Fermi energy, probably plays an important role for the T_c enhancement as investigated theoretically (*16*). Referring to the theory of Höck *et al.* (*11*), one creates Jahn-Teller polaron holes by the doping. However, we cannot yet exclude an enhanced oxygen-vacancy content in the samples on alkaline earth–substitution to preserve charge neutrality. As the concentration of vacancies is finite, the role of pairs of Cu^{2+} and oxygen vacancies in the lattice needs to be investigated (*17*).

REFERENCES AND NOTES

1. J. G. Bednorz and K. A. Müller, *Z. Phys. B* **64**, 189 (1986).
2. J. G. Bednorz, M. Takashige, K. A. Müller, *Europhys. Lett.* 3, 379 (1987).
3. S. Uchida, H. Takagi, K. Kitazawa, S. Tanaka, *Jpn. J. Appl. Phys.*, in press.
4. C. W. Chu *et al.*, *Phys. Rev. Lett.* **58**, 405 (1987); C. W. Chu *et al.*, *Science* **235**, 567 (1987).
5. J. G. Bednorz, M. Takashige, K. A. Müller, *Mater. Res. Bull.*, in press.
6. H. Takagi, S. Uchida, K. Kitazawa, S. Tanaka, *Jpn. J. Appl. Phys.*, in press.
7. K. A. Müller, M. Takashige, J. G. Bednorz, *Phys. Rev. Lett.*, in press.
8. D. C. Johnston *et al.*, *Mater. Res. Bull.* **8**, 777 (1973).
9. A. W. Sleight, J. L. Gillson, F. E. Bierstedt, *Solid State Commun.* 17, 27 (1975).
10. B. K. Chakraverty, *J. Phys. (Paris)* **40**, L99 (1979); *ibid.* **42**, 1351 (1981).
11. K.-H. Höck, H. Nikisch, H. Thomas, *Helv. Phys. Acta* **56**, 237 (1983).
12. H. D. Megaw, *Crystal Structures: A Working Approach* (Saunders, Philadelphia, 1973), p. 26.
13. C. Michel and B. Raveau, *Rev. Chim. Min.* **21**, 407 (1984).
14. R. J. Cava, R. B. van Dover, B. Batlogg, E. A. Rietmann, *Phys. Rev. Lett.* **58**, 408 (1987).
15. C. Ebner and A. Stroud, *Phys. Rev. B* **31**, 165 (1985).
16. D. H. Lee and G. Baskaran, personal communication.
17. See, for example, the study by Y. Takeda *et al.* [*J. Solid State Chem.* **63**, 237 (1986)] on nonstoichiometric $SrFeO_x$ phases.

2 February 1987; accepted 18 February 1987

Avascular Necrosis: Occurrence in Diving Cretaceous Mosasaurs

BRUCE ROTHSCHILD AND LARRY D. MARTIN

A study of vertebrae of extinct giant marine lizards showed the presence of avascular necrosis in two of the three most common genera of these mosasaurs, *Platecarpus* and *Tylosaurus*. This bone disease was invariably present (involving 5 to 66% of vertebrae) in these genera, but absent in a third genus *Clidastes*. Differential occurrence of avascular necrosis may be related to decompression syndrome, suggesting different habitat and diving habits of the respective genera.

THE BONE PATHOLOGY, AVASCULAR necrosis, was found to be common in the skeletons of extinct giant marine lizards, the mosasaurs. The phenomenon was identified by its pathognomonic radiologic appearance (*1–3*), that of a linear region of radiolucency. This focal avascular necrosis (Fig. 1A) is quite different from the regional vascular changes resulting in tail loss in iguanids and the hoof and distal extremity loss occurring in grazing animals, secondary to ergotism. Whereas bone necrosis may occur in the latter circumstances, it is quite different from the linear necrotic pattern herein described in mosasaur vertebrae, where it is identical to the phenomenon known as avascular necrosis in man. Ischemic necrosis of vertebrae, presenting with an intravertebral radiolucent cleft is rare. All reported human cases that we have identified appear to manifest vertebral collapse, in addition to the intravertebral radiolucent cleft. Resnick considered this radiolucent cleft sign "virtually pathognomonic of bone necrosis" (*1*) as did Maldague (*2*). The vertebrae involved in man typically have included the tenth thoracic through the third lumbar, generally as an isolated phenomenon (limited to one vertebrae). Spinal involvement in avascular necrosis has thus been diagnosed by its radiologic appearance.

During study of a mosasaur vertebra with infectious spondylitis, we selected what appeared to be a normal vertebra for sectioning in order to characterize normal vertebral anatomy, and found a linear region of loss of tissue definition. The similarity of radiographs of this vertebra to those of vertebral bodies with avascular necrosis occasionally observed in man led to characterization of the nature of the bony change and a survey of the frequency of the pathology in caudal vertebrae in the University of Kansas Vertebrate Paleontology mosasaur collection.

Vertebral material in the collection belongs mostly to three genera of mosasaurs, *Platecarpus*, *Tylosaurus*, and *Clidastes*. Avascular necrosis was identifiable in all specimens of *Platecarpus* and *Tylosaurus* in the collection and in none of *Clidastes* (Table 1). The percentage of vertebrae involved in a given individual (Table 1) was greater for *Platecarpus* (mean ± SEM, 25.2 ± 5.5) than for *Tylosaurus* (9.1 ± 1.2) ($t = 2.36$, $P < 0.03$).

The vertebrae had no external evidence of vertebral collapse. Their size was the same as that of unaffected vertebrae. Radiologic examination revealed a radiolucent cleft (Fig. 1A). The cut surface revealed a linear area of loss of bone definition (Fig. 1B), confirmed on scanning electron microscopy. This can be distinguished from vascular channels by absence of sharply defined, radiodense margins. The relative localization of this abnormal zone within a given vertebrae corresponds to localization of vertebral avascular necrosis in man (*4, 5*) which in man represents a watershed region of vascular supply (*6*). Localization of perforating vertebral vessels in *Platecarpus* (Fig. 1C) is consistent with their location in man. Figure 1D shows on transverse section the vascular supply in *Platecarpus*, which is similar to that reported in man (*6*). The fossil occurrences differ from reported cases of avascular necrosis in man only in that vertebral collapse does not occur in the mosasaur specimens (*4*), and osteoporosis, reported in 65% of human cases (*7, 8*), was not identifiable in the mosasaur specimens. The relatively uniform width of the clefts would not be compatible with a diagnosis of metastatic carcinoma. Absence of irregular finger-like radiolucen-

B. Rothschild, Museum of Natural History, Lawrence, KS 66045, and St. Elizabeth Hospital Medical Center and Northeast Ohio Universities College of Medicine, Youngstown, OH 44501.
L. D. Martin, Museum of Natural History and Department of Systematics and Ecology, University of Kansas, Lawrence, KS 66045.