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25. We thank J. Andrews for excellent technical assistance. Hydroxybenzylpindolol was a gift from D. Hauser, Sandoz Pharmaceuticals. Supported by a grant from the American Heart Association. T.K.H. (NS 05126) and B.B.W. (NS 01989) are postdoctoral fellows of the PHS. P.B.M. is an established investigator of the American Heart Association.

10 May 1976; revised 12 July 1976

Oldest Horse Brains: More Advanced Than Previously Realized

Abstract. *Previous interpretations of early horse brains were based on an incorrectly identified fossil endocast, now believed to be from a condylarth. Newly prepared endocasts of* Hyracotherium, *the oldest horse and one of the earliest perissodactyls, reveal a relatively larger brain, with a more expanded neocortex, than existed in the condylarth ancestors of perissodactyls. Fifty million years ago, horse brains had suprasylvian, ectolateral, and lateral sulci, but the frontal lobe was undeveloped.*

Almost 30 years ago, Edinger (*1*) described the brain of Eohippus (*Hyracotherium*), the oldest horse, as being more primitive than that of any living placental mammal and strikingly similar to that of *Didelphis*, the marsupial opossum. That description was important because *Hyracotherium* was the earliest horse and also one of the earliest members of the order Perissodactyla (horses, rhinos, tapirs, and some extinct families). Therefore, knowledge of its brain morphology has relevance for understanding the factors involved in the origin of the order, and also for interpreting subsequent brain evolution in perissodactyls.

Edinger's interpretation of the brain of *Hyracotherium* was based largely on a natural stone endocast with only ventral braincase bones attached (Yale Peabody Museum 11694) from the early Eocene (about 50 to 55 million years ago) of New Mexico. However, there are no teeth or other diagnostic elements that allow definite identification of YPM 11694 as *Hyracotherium* (or as any other perissodactyl). Further, the optic foramen is smaller and more rostrally located in YPM 11694 than in known *Hyracotherium* specimens (*2*), which indicates that YPM 11694 has been incorrectly identified. To see what the brain of the earliest horses actually looked like, I prepared endocasts from three skulls with dentitions, unequivocally identified as *Hyracotherium* (*3*). These endocasts reveal that the brain was considerably advanced, in terms of neocortical expansion, over the condition described by Edinger, and also advanced compared to brains of the archaic ungulates from which the order Perissodactyla arose.

The new *Hyracotherium* endocasts (Fig. 1, A and B), show that the brain had three neocortical sulci. The longest and most prominent sulcus lay immediately dorsal to the rhinal fissure and extended rostrocaudally for most of the length of the cerebrum. Dorsal to the caudal portion of that sulcus lay two shorter and less marked sulci. Comparisons with later perissodactyl endocasts, which reveal the evolutionary history of neocortical folding in the order (*1, 4*), suggest that the sulci indicated on the *Hyracotherium* endocasts represent, from ventral to dorsal, the suprasylvian, ectolateral, and lateral sulci of modern perissodactyls. A shallow notch in the rhinal fissure, which delimited a rostral one-fifth of the cerebrum, marks the later (future) position of the sylvian sulcus. The caudal portion of the rhinal fissure was oriented dorsocaudally, and was overlain by a vascular sinus.

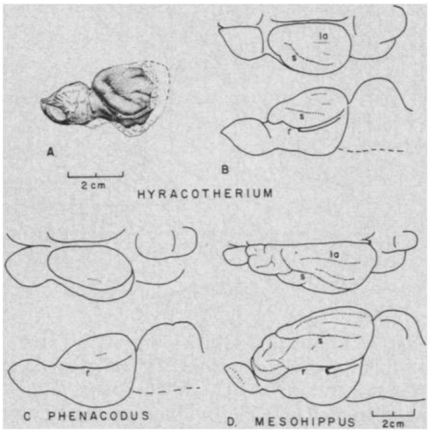

Fig. 1. Fossil endocasts. (A) Oblique view of slightly crushed endocast of forebrain of *Hyracotherium*, a 50-million-year-old horse, American Museum of Natural History (AMNH) No. 55267. (B) Dorsal and lateral views of *Hyracotherium* endocast, composite based on AMNH Nos. 55266, 55267, and 55268. (C) Endocast of *Phenacodus*, a condylarth contemporary of *Hyracotherium*, based on AMNH No. 4369 and Museum of Comparative Zoology No. 4440. (D) Endocast of *Mesohippus*, a 30-million-year-old horse, AMNH No. 9814. Abbreviations: *la*, lateral sulcus; *r*, rhinal fissure; *s*, suprasylvian sulcus; (A, B, and C) slightly more than one-third size; (D) slightly more than one-fourth size. The scales are 2 cm.

A short gap separated the rostral pole of the cerebrum from the olfactory bulbs, which were pedunculate. The olfactory bulbs were large relative to the rest of the brain. The caudal pole of the cerebrum appears to have extended back almost to the cerebellum, and little if any of the midbrain was exposed dorsally. The cast of the cerebellum was not preserved well enough to reveal details beyond gross size and shape.

The *Hyracotherium* endocasts described above are from a species that lived in the latter part of the early Eocene, about 50 million years ago, about 10 million years after the origin of the order. Perissodactyls show little evidence of evolutionary change during the early Eocene, and the oldest known specimens of *Hyracotherium*, which are also the oldest known perissodactyls, differ little in known morphology (cheek teeth) from the later *Hyracotherium* species whose endocasts are here described (*5*). Nevertheless, because of the amount of elapsed time, it is possible that the available *Hyracotherium* endocasts represent a brain morphology somewhat more advanced than that of the earliest perissodactyls. We must await the discovery of additional fossil endocasts to learn whether the brain morphology here described also characterized that of the earliest perissodactyls.

The fossil record suggests that perissodactyls arose in the late Paleocene (about 60 million years ago), from a group of archaic ungulates, the phenacodontid condylarths (*6*). The only phenacodontid endocasts known are from *Phenacodus*, a genus that survived through the early Eocene, and coexisted with *Hyracotherium* and other early perissodactyls before becoming extinct. The brain of *Phenacodus* (Fig. 1C) was more primitive than that of *Hyracotherium* in having a less expanded neocortex: note the higher rhinal fissure, fewer neocortical sulci, and less expanded frontal pole. The available evidence of phenacodontid phylogeny suggests that both the earliest perissodactyls and *Phenacodus* evolved from the middle Paleocene phenacodontid genus *Tetraclaenodon* (*6, 7*). If that is correct, the endocast of *Phenacodus* indicates the maximum degree of neocortical expansion one would expect in the phenacodontid condylarth ancestor of perissodactyls. Thus, expansion of the neocortex may reflect one of the adaptations responsible for the emergence and early success of the order Perissodactyla.

The new *Hyracotherium* endocasts provide a baseline against which to compare later horse brain evolution. After *Hyracotherium*, the next good record is from endocasts of *Mesohippus*, a 30-million-year-old ancestor of modern horses (*8*). The brain of *Mesohippus* (Fig. 1D) had a more expanded neocortex than is seen in *Hyracotherium*, with the occipital and frontal lobes in more extensive contact with cerebellum and olfactory bulbs, respectively. The ectolateral and lateral sulci were longer in *Mesohippus*, but the greatest change appears to have been in the frontal lobe, which was considerably larger and more convoluted in *Mesohippus* than in *Hyracotherium*. Thus, in the first 20 to 30 million years of horse brain evolution, there was continued expansion of the neocortex, with the most marked increase in the area of the frontal lobe.

The *Hyracotherium* endocasts suggest a brain volume of about 25 cm^3, and associated skeletal materials suggest a body weight of about 9 kg (*9*). A useful index of relative brain size is the encephalization quotient, or E.Q., proposed by Jerison (*10*), which is the ratio of a given species' brain size compared to the brain size one would expect in an average living mammal of that species' body weight (*11*). The E.Q. of *Hyracotherium* was about 0.47, meaning that it had a brain about half the size one would expect in an average living mammal of the same body weight as *Hyracotherium*. For the condylarth *Phenacodus*, I estimate an E.Q. of 0.22; for *Mesohippus*, an E.Q. of 0.77; and for modern horses and zebras, E.Q.'s of 0.95 to 1.09 (*12*). In other words, the relative brain size of *Hyracotherium* was about twice that of the contemporaneous condylarth *Phenacodus*, but about half that of modern horses; and during the 20 to 30 million years between *Hyracotherium* and *Mesohippus*, relative brain size increased by about 65 percent.

The evolutionary trends toward increased amount of neocortex and increase in relative brain size are probably correlated (*13*), and are seen in many other groups of mammals besides horses (*10, 14*). The functional significance of those trends is unclear, and their elucidation remains one of the important unsolved problems in studies of mammalian brain evolution. Expansion of the frontal lobe, seen in evolution from *Hyracotherium* to *Mesohippus*, is a common trend in ungulate brain evolution (*15*). Extrapolation from cortical maps of living ungulates suggests that increased tactile sensitivity of the lips, reflected in expansion of lip region representation in somatic sensory cortex, was at least in part responsible for frontal lobe expansion in ungulates. Increased lip sensitivity, necessary for manipulation of vegetation prior to ingestion, would be expected in mammals specializing in browsing or grazing.

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11. The relationship of brain weight (E, grams) to body weight (P, grams) in a large sample of living mammals is described by the equation: $E = 0.12 P^{0.67}$ (*10*).
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15 July 1976

3,4,3′,4′-Tetrachloro Azoxybenzene and Azobenzene: Potent Inducers of Aryl Hydrocarbon Hydroxylase

Abstract. *Two unwanted contaminants, 3,4,3′,4′-tetrachloroazoxybenzene (TCAOB) and 3,4,3′,4′-tetrachloroazobenzene (TCAB), formed in the commercial synthesis of 3,4-dichloroaniline or of herbicides made from 3,4-dichloroaniline, were responsible for three outbreaks of acne among chemical workers. TCAOB and TCAB are approximately isosteric to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin and 2,3,7,8-tetrachlorodibenzofuran, two well-known contaminants that cause acne. All four of these agents are potent inducers of hepatic aryl hydrocarbon hydroxylase activity and compete for stereospecific binding sites in the hepatic cytosol, which are thought to be the receptor sites for the induction of this enzyme. Among the chlorinated azoxy and azobenzenes, the potency of a congener to induce aryl hydrocarbon hydroxylase activity correlates with its binding affinity for the hepatic cytosol specific binding sites and its capacity to induce acne; this relation between structure and activity parallels that observed for the chlorinated dibenzo-*p*-dioxins and dibenzofurans.*

3,4-Dichloroaniline is the starting material for the synthesis of a number of commercially important herbicides (acylphenylamides, phenylcarbamates, and phenylureas). During the synthesis of 3,4-dichloroaniline or its further conversion to herbicides, the conditions employed (heat and mild oxidation) promote the condensation of two molecules, so that 3,4,3′,4′-tetrachloroazoxybenzene (TCAOB), or 3,4,3′,4′-tetrachloroazobenzene (TCAB) are formed as unwanted contaminants. Several outbreaks of acne (chloracne) have occurred among workers in chemical plants manufacturing 3,4-dichloroaniline or its deriva-