## Amoebic Meningoencephalitis

The report by Červa and Novák concerning 16 human fatalities associated with amoebic meningoencephalitis (*1*) should ring a warning bell among members of the health professions. Amoebas are being cultivated with remarkable frequency from human sources such as the upper respiratory tract and from serum during searches for viruses thought to cause various respiratory symptoms, fevers, and hepatitis (*2*). Speculations on how free-living amoebas gain access to the human host, whether by inhalation from the air or via the oral route, and whether they gain entrance in the amoeboid or cyst form, are fascinating from an epidemiological point of view, and since no one yet knows the overall occurrence of these potentially important amoebas in the nasal-pharyngeal regions of human populations, it would seem prudent to undertake at once an extensive survey of their occurrence. Moreover, it would seem wise for medical practitioners to include the possibilities of amoeba in the unexplained meningoencephalitides that they encounter. In light of Červa and Novák's paper, someone should also immediately attempt to isolate these protozoans from public and private swimming pools at large since some amoebas (for example, *Entamoeba histolytica*) are known to be resistant to the common methods of chlorination used in water treatment.

JOHN I. PAYNE
*Department of Biology,*
*Georgia State College, Atlanta*

**References**

1. L. Červa and K. Novák, *Science* **160**, 92 (1968).
2. J. A. Armstrong and M. S. Pereira, *Brit. Med. J.* 1, 212 (1967); T. H. Dunnebacke and R. C. Williams, *Proc. Nat. Acad. Sci. U.S.* **57**, 1363 (1967); D. C. Warhurst and J. A. Armstrong, *J. Gen. Microbiol.* **50**, 207 (1968); J. J. Callicott, *Amer. J. Clin. Pathol.* **49**, 84 (1968); R. S. Chang, I. Hung Pan, B. J. Rosenau, *J. Exp. Med.* **124**, 1153 (1966).

18 April 1968

## Endovitamins and Orthomolecular Environment

In his article on the role of "orthomolecular" concentrations of essential nutrilites in mental health (*1*), Pauling quotes evidence showing that when an essential metabolite is exogenously supplied auxotrophic mutants have an evolutionary advantage over prototrophic strains (*2*); he discusses, among other examples, evolutionary loss of the ability to synthesize ascorbic acid by primates and a few other species.

An interesting aspect of this story, which is relevant to Pauling's contention that different tissues may require different amounts of an essential nutrilite, is the fact that in those species which do not require ascorbic acid in the diet, the vitamin is not synthesized by all cells, but only by one organ which provides for the rest of the organism. Curiously enough, the site of synthesis changes with evolution (*3*). It is localized in the kidney in amphibia and reptiles and in the liver in mammals. Birds show a transition pattern; early species, such as the chick, synthesize ascorbate in the kidney, and late species, such as the Passeriformes, synthesize it in the liver (*3*).

The missing step in those species and organs unable to synthesize ascorbic acid has been tracked down to a single enzyme, gulonolactone oxidase (GLO), which is the last one in the synthetic pathway (*3*). As far as we know this enzyme belongs exclusively to this pathway so that it can be dropped without derangement of related paths such as glucuronate metabolism.

In addition to evolutionary variations, GLO is also subject to temporal and spatial programming in ontogenic development. In the chick embryo, which makes all its ascorbic acid *de novo* (*4*), GLO activity is initially spread throughout the blastoderm, including the extra-embryonic mesoderm, and then gradually disappears from differentiating cells. The liver and stomach still retain some activity at the middle of development, but later GLO is found solely in the nephrogenic tissues where it becomes highly concentrated (*5*); in the hatched bird the enzyme is present only in the kidney (*3, 5*). In chick, therefore, as in most higher animals, ascorbic acid may be considered an "endovitamin" in the sense that most organs depend symbiotically on a group of specialized cells for its supply.

In connection with the problem of ascorbic acid requirement of brain tissue, it is remarkable that embryonic brain, in chick at any rate, has one of the highest ascorbic acid titers in the organism and yet has no synthetic activity (*4, 5*). This puts the developing brain in this species under severe dependence from synthesizing tissues. In the embryo of man and other primates, the central nervous system presumably depends entirely on maternal supplies of vitamin C, although preliminary evidence suggests that leakage of GLO activity in early development may occur even in ascorbic acid–requiring species (*6*).

Besides the mental manifestations of scurvy, mentioned by Pauling, the requirement of ascorbic acid as a cofactor in the beta-hydroxylation of sympathomimetic and hallucinogenic amines (*7*) provides more direct evidence of its involvement in brain function. The foregoing considerations emphasize the possibility that brain damage due to ascorbic acid deficiency may occur as a consequence of metabolic imbalance during embryonic development without ostensible dietary shortage or even indeed in species which do not require ascorbic acid as an external vitamin.

The mechanisms underlying the epigenetic regulation of ascorbic acid synthesis remain obscure. Since GLO requires no cofactors, and diffusible inhibitors were ruled out (*5*), the loss of synthetic activity in differentiating tissues may be attributed to actual restriction of enzyme synthesis. The differential expression of the GLO phenotype in different organs and developmental stages and even perhaps in different species and phyla could conceivably reflect a differential gain in gene activity controlled by cytoplasmic factors at the level of replication, transcription, or translation. The resulting equilibrium could affect, in turn, the penetrance of other genes. Thus, the shedding of unnecessary synthetic mechanisms may not only confer an evolutionary advantage but may also serve an economical purpose in epigenetic differentiation to spare free energy for the synthesis of more specific molecules or the performance of higher integrative functions. The case of ascorbic acid may not be unique.

LUIS M. RINALDINI
*Department of Biology, Brooklyn College of the City University of New York, Brooklyn, New York 11210*

**References**

1. L. Pauling, *Science* **160**, 265 (1968).
2. See also the early work of A. Lwoff, "L'Evolution physiologique. Etudes des pertes de fonctions chez les microörganismes," *Actualités Sci. Ind.* (Hermann, Paris, 1943), pp. 1–308.
3. J. J. Burns, *Nature* **180**, 553 (1957); A. P. Grollman and A. L. Lehninger, *Arch. Biochem.* **69**, 458 (1957); I. B. Chatterjee, N. C. Kar, N. C. Ghosh, B. C. Guha, *Ann. N.Y. Acad. Sci.* **92**, 36 (1961).
4. L. M. Rinaldini, *J. Embryol. Exp. Morphol.* **8**, 527 (1960).
5. S. P. Fabro and L. M. Rinaldini, *Develop. Biol.* **11**, 468 (1965).
6. S. P. Fabro, thesis, University of Cordoba, Argentina (1966).
7. M. Goldstein, *Pharmacol. Rev.* **18**, 77 (1966).

5 May 1968