shaking with chloroform or acetone. If a "heterozygous" pattern results from this maneuver, then this pattern is due merely to low concentrations of antitrypsin globulin and is not the result of an "electrophoretically distinct molecule." In fact, we have observed that the electrophoretic mobility of the α_1-globulin on polyacrilamide gels varies with the dilution of serum protein.

The slight variation in antitrypsin pattern by which Kueppers *et al.* distinguish heterozygous α_1-antitrypsin deficiency from normal very likely does include all heterozygotes. However, this pattern may also occur with serums from individuals without the inherited deficiency whose α_1-antitrypsin nonetheless is normally low.

Kueppers *et al.* point out that the concentration of antitrypsin can be raised to the normal range in heterozygotes by conditions such as infection and pregnancy. However, they imply that the diagnostic variations of antitrypsin patterns seen on antigen-antibody crossed electrophoresis remain unchanged. No evidence for this statement is presented.

The difference between heterozygous deficient and normal individuals may depend less on the subject's minimum unstimulated antitrypsin and more upon differences in ability to increase this level during an inflammatory process (*3*).

JACK LIEBERMAN
Department of Respiratory Diseases, City of Hope Medical Center, Duarte, California 91010

References

1. F. Kueppers, R. Fallat, R. K. Larson, *Science* **165**, 899 (1969).
2. F. Kueppers and A. G. Bearn, *ibid.* **154**, 407 (1966).
3. J. Lieberman, *N. Engl. J. Med.* **281**, 279 (1969).

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On antigen-antibody crossed electrophoresis, the position of the different α_1-antitrypsin peaks characteristic for the different phenotypes remains the same over a wide range of concentrations. I have followed the α_1-antitrypsin pattern by antigen-antibody crossed electrophoresis of several heterozygotes for the deficiency gene, when their serum α_1-antitrypsin was low and when it was high (from 105 to 240 mg/100 ml of serum). The position of the peaks did not change; the height of the peaks varied with the concentration of α_1-antitrypsin. The precipitation pattern of a heterozygote can be explained by the presence of two groups of electrophoretically distinct but antigenically identical α_1-antitrypsin components (*1*). Although the α_1-antitrypsin bands in stained starch gels are usually weak or absent, homozygotes for the deficiency gene have a serum concentration of 25 ± 6 mg/100 ml (*2*).

As Lieberman suggests, heterozygotes respond differently to an inflammatory stimulus from normals. After an injection of typhoid vaccine normal individuals show an increase in serum of α_1-antitrypsin from approximately 200 mg to an average of 375 mg/100 ml, and heterozygotes show an increase from 112 mg to 196 mg/100 ml (*3*).

F. KUEPPERS
Institute of Human Genetics, 2 Hamburg 20, Martinistrasse 52, Germany

References

1. M. K. Fagerhol, *Scand. J. Clin. Lab. Invest.* **23**, 97 (1969); F. Kueppers, *Biochem. Genet.* **3**, 283 (1969).
2. F. Kueppers, *Humangenetik* **5**, 54 (1967).
3. ———, *ibid.* **6**, 207 (1968).

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Monosodium Glutamate: Specific Brain Lesion Questioned

Olney and Sharpe (*1*) administered, by subcutaneous injection, a single dose of monosodium glutamate (15 mmole per kilogram of body weight) to one newborn premature rhesus monkey and concluded that the lesion in the periventricular arcuate region of the hypothalamus was specifically induced by glutamate.

They stated "We have demonstrated susceptibility of a primate species to the mechanism of the glutamate effect" and "Presumably an elevated blood concentration of glutamic acid is an important requisite to the lesion formation." However, they did not supply data on concentrations of glutamic acid in the blood, nor did they supply control data for their experiment. A minimum requisite would have been the use of inorganic sodium salts administered in a way similar to that used for monosodium glutamate, the use of sodium salts of several other amino acids, and the administration of glutamic acid as the free acid. Perhaps most critical is the fact that they used only one animal.

An explanation as plausible as that presented is that the findings reflect a nonspecific solute effect. Finberg *et al.* (*2*) have shown that an intravenous or intraperitoneal dose of many univalent completely ionized salts (15 meq per kilogram of body weight) given to an infant cat reduces the cerebral spinal fluid pressure to zero in 180 minutes (the time interval used by Olney and Sharpe). Under these circumstances, there is always dilatation of capillaries and small vessels and occasionally, at this dosage, tearing of vessels in the Virchow-Robin space, with consequent hemorrhage. The specific lesion presumably varies with the conformation of the intracranial contents and skull of the animal model used.

The implication of a risk of brain damage as a result of human infants' consuming infant food products is unsupportable. Infant food products on the market, until recently, contained between 150 and 180 mg of monosodium glutamate per jar. A few contained as much as 600 mg per jar. A 5-kg infant would have had to consume, at one sitting, 20 jars of an infant food product containing 600 mg per jar to obtain the amount of monosodium glutamate used by Olney and Sharpe. The controversy surrounding monosodium glutamate in infant food products has aroused fear on the part of mothers that their infants would become mentally retarded as a result of their having been fed commonly available infant food products. There obviously was no such risk and no basis in fact that this could have occurred.

The issue of whether or not monosodium glutamate should be in infant food products is a different one. The amino acid serves no useful nutritional purpose either as a flavoring agent or as a supplement to an infant diet already high in glutamate as a result of milk and wheat content. Furthermore, the addition of glutamate probably increases the price of the product, and under these circumstances inclusion cannot be condoned. However, this issue is completely different from the proposition that a specific neurological lesion occurs in infants receiving these foods, and publishing information based on an experiment in a single animal with no controls is only confounding.

CHARLES U. LOWE
National Institute of Child Health and Human Development, Bethesda, Maryland 20014

References

1. J. W. Olney and L. S. Sharpe, *Science* **166**, 386 (1969).
2. L. Finberg, C. Lutrell, H. Redd, *Pediatrics* **23**, 46 (1959); L. Finberg, *Advan. Pediat.* **16**, 325 (1969).

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Olney and Sharpe administered to mice, presumably by subcutaneous injection, 0.5 g of monosodium glutamate per kilogram of body weight. They gave, by the same route, 2.7 g per kilogram of body weight to one newborn monkey. The monkey is characterized as being similar to a premature infant, but it is not clear whether the monkey was, in terms of development, itself premature. During a 3-hour period when the monkey was observed, there was no manifestation of a central nervous system disturbance. The animal was then anesthetized with Sernylan, and the brain and some other organs were removed.

There is no indication of previous observations on monkeys of similar age, no controls to indicate observation of animals handled in identical fashion but without the use of monosodium glutamate, and no quantitative relation to the dose of monosodium glutamate that could be expected to be given to the human infant.

Unless the above questions can be answered, and information can be provided about the effect of monosodium glutamate on passage through the gastrointestinal tract, the report of Olney and Sharpe has no meaning. The fears engendered among mothers with infants in arms cannot be calculated.

MITCHELL R. ZAVON
Kettering Laboratory, College of Medicine, University of Cincinnati, Cincinnati, Ohio 45219

Reference

1. J. W. Olney and L. S. Sharpe, *Science* **166**, 386 (1969).

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Lowe and Zavon raise questions concerning specificity of glutamate-induced brain damage, doses, routes of administration, and absorption of glutamate from the gastrointestinal tract. We share their interest in these questions and in order to explore them (*1*, *2*) have conducted experiments in several hundred infant mice. Although not gathered on infant monkeys, these data may be of value in interpreting our infant monkey experiment (*3*) and in guiding future glutamate research with other preciously scarce primate infants.

In studies designed to investigate specificity and mechanism of action (*2*), mice (10 days old) were given (by feeding tube) chromatographically pure glutamic acid (*4*, *5*) (1 g/kg); this dose consistently induced a process of necrosis in hypothalamic neurons which was indistinguishable by electron microscopic examination from the neuronal pathology observed in the infant rhesus monkey treated with monosodium glutamate (*3*). Doses of sodium chloride as high as 8 g/kg (25 times the sodium content of monosodium glutamate at 1.0 g/kg) rendered infant mice nearly prostrate over a 4-hour observation period but resulted in no pathological alterations in hypothalamic or retinal neurons. Monosodium glutarate, which differs from monosodium glutamate only in that it has no amino group, induced no changes in hypothalamic or retinal neurons at 5 g/kg (*5*). The majority of amino acids tested induced no detectable changes in hypothalamic or retinal neurons at 3 g/kg (L-serine, L-glycine, L-alanine, DL-methionine, L-phenylalanine, L-proline, L-leucine, L-arginine, and L-lysine). Striking lesions did appear, however, in both the hypothalamus and inner retina after either oral or subcutaneous administration of sodium L-aspartate, sodium DL-α-aminoadipate, and L-cystiene. These findings suggest that the phenomenon of glutamate-induced brain damage to the immature central nervous system is not exclusively specific to glutamate but may be specific to a select group of compounds identified by others as having neuroexcitatory properties in vivo (*6*) and the ability to depolarize neural membranes in vitro (*7*).

When 10-day-old mice were fed monosodium glutamate in 10 percent aqueous solution by tube (*2*), hypothalamic damage occurred in 54 percent of 24 animals treated at doses of 0.5 g/kg and in 100 percent of 19 animals receiving 1.0 g/kg. In the same feeding experiments, a 0.5 g/kg dose of glutamate in combination with a 0.5 g/kg dose of aspartate resulted consistently in more severe hypothalamic damage than that induced by a 0.5 g/kg dose of either compound alone. Blood glutamate curves monitored in 23 tube-fed infant mice (*8*) indicate that oral intake of monosodium glutamate in doses of 0.5 to 1 g/kg produces high concentrations of glutamate in the blood, often exceeding 50 mg/100 ml by 15 to 30 minutes but returning to base-line values, in the range of 5 mg/100 ml, within 2 hours.

Lowe infers that 2.7 g/kg, the high dose of monosodium glutamate that we administered to test susceptibility in an infant monkey, represents an estimate of lowest effective dose. The only data on lowest effective dose that we are aware of are those for the mouse (*2*). Lowe's 20-jar margin of safety would become much narrower if he were to base his calculations on the lowest effective dose that we found in the infant mouse (0.5 g/kg). Lowe correctly points out that, until recently, some glutamate-rich strained baby food (4½ oz. jars) was supplemented with glutamate (615 mg) (*9*). In his calculation of risk, however, Lowe considered only the added glutamate. Moreover, it is uncertain, because unstudied, whether food with a high natural glutamate content (or glutamate plus aspartate content) imparts enhanced risk, and therefore calculations which ignore such fundamental considerations may not be valid. The weakness in Lowe's "obviously . . . no . . . risk" position, and in the position of others who have made similar pronouncements (*10*), is in the complete absence of supporting relevant experimental evidence. Glutamate was established years ago as a safe food additive by "expert" opinion and was introduced into baby foods without any safety tests being run on infant animals of any species. The question of safety would not have become an issue if experimental evidence rather than a priori assumption had been relied upon in the first place.

JOHN W. OLNEY
LAWRENCE G. SHARPE
Department of Psychiatry, Washington University School of Medicine, St. Louis, Missouri 63110

References and Notes

1. J. W. Olney, *Science* **164**, 719 (1969).
2. ———, in preparation.
3. ——— and L. G. Sharpe, *Science* **166**, 386 (1969).
4. Glutamic acid (Fisher) used in this experiment and two sources of monosodium glutamate used in others (*1*, *2*, *5*) (Sigma and Nutritional) were tested in our laboratories and found to contain no impurities detectable by thin layer chromatography. We thank Dr. Blake Moore for testing these compounds chromatographically.
5. J. W. Olney, *J. Neuropathol. Exp. Neurol.* **28**, 455 (1969).
6. D. R. Curtis and J. C. Watkins, *J. Neurochem.* **6**, 117 (1960); K. Krnjevic and J. W. Phillis, *J. Physiol.* **165**, 274 (1963); F. A. Steiner and K. Ruf, *Helv. Physiol. Pharmacol. Acta* **24**, 181 (1966); K. Kishida and K. Nada, *Science* **156**, 650 (1967).
7. H. F. Bradford and H. McIlwain, *J. Neurochem.* **13**, 1163 (1966).
8. R. D. Feigin and J. W. Olney, unpublished results.
9. Gerber Products, Inc., in *Appendix to Hearings before the Select Committee on Nutrition and Human Needs of the United States Senate*, Part 13A, 4170 (July 1969).
10. F. R. Blood, B. L. Oser, P. L. White, *Science* **165**, 1028 (1969).

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