Mg^{2+} is added simultaneously with the thiol, dissociation does not occur and enzymatic activity is retained. Evidence to be published elsewhere indicates that Mg^{2+} not only protects the enzyme from inactivation by the thiol but also interacts with the dissociated subunits after all catalytic activity has disappeared.

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Received January 19; revised March 19, 1970.

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Tay-Sachs Disease is Probably not Increasing

RECENTLY, Shaw and Smith[1] have discussed the question of heterozygote advantage for the Tay-Sachs gene in Jewish populations with reference to our published computations[2] and more generally. Their estimate of 5·3 per cent advantage over normal homozygote, based on trial values in a discrete model, is better than our value of 4·6 per cent from a very rough continuous approximation. But they seem to imply that we also accepted another estimate of 1·3 per cent advantage; this value was mentioned only to indicate how little heterosis would be required simply to cancel deaths of the recessive homozygotes.

Just before their article appeared, we wrote to the authors to express our doubt that the Tay-Sachs gene frequency is still increasing, as they suggest. It seems likely that an actual increase of gene frequency under selection was occurring where the frequency of the disease was high—in Eastern Europe, especially in Poland. But there are no longer any Ashkenazi ghettoes where selection for heterozygotes presumably was strong, as actual reproductive data seem to show. And the presumed biological advantage is no longer demonstrable in young, American-born, families with Tay-Sachs disease, suggesting that the ecological circumstances which conferred such advantage no longer prevail[2]. Further, we have evidence that the frequency of Tay-Sachs disease was far from uniform in populations of European Ashkenazi Jews[3]. There is nothing to support the notion that heterozygotes have greater fitness in North America, Britain or Israel. The point might be worth investigating in Russia.

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Received May 5, 1970.

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Brain Damage in Infant Mice following Oral Intake of Glutamate, Aspartate or Cysteine

STRIKING degenerative changes in the infant mouse retina after subcutaneous treatment with monosodium glutamate (MSG) were reported by Lucas and Newhouse in 1957[1]. Other studies[2–6] established that the process of retinal degeneration induced by MSG treatment is a remarkably acute and irreversible form of neuronal pathology. Recently it was found that a similar process of acute neuronal necrosis occurs in several regions of the infant mouse brain after subcutaneous treatment with MSG, and that animals treated with high doses in infancy tend to manifest obesity and neuroendocrine disturbances as adults[7,8]. The arcuate nucleus of the hypothalamus is an area particularly vulnerable to glutamate induced damage in infant animals of several species (mice and rats[7], rabbits and chicks and the rhesus monkey[9]). In mice, which have been studied more extensively for MSG induced disturbances than other species, the infant animal suffered hypothalamic damage from a relatively low subcutaneous dose (0·5 g/kg of body weight)[7].

Table 1

Test compound	Dose (g/kg)	Number treated	Number affected	Necrotic hypothalamic neurones
Intubated, no treatment	—	10	0	0
MSG	0·25	10	0	0
MSG	0·50	23	12	7
MSG	0·75	16	13	13
MSG	1·00	19	19	25
MSG	2·00	7	7	40
L-Glutamic acid	1·00	4	4	23
Monosodium L-aspartate	1·00	4	4	26
L-Glutamate/L-aspartate	0·50/0·50	8	8	27
Monosodium-glutarate	3·00	4	0	0
NaCl	3·00	4	0	0
L-Glycine	3·00	2	0	0
L-Serine	3·00	2	0	0
L-Alanine	3·00	2	0	0
L-Leucine	3·00	2	0	0
DL-Methionine	3·00	2	0	0
L-Phenylalanine	3·00	2	0	0
L-Proline	3·00	2	0	0
L-Lysine	3·00	2	0	0
L-Arginine	3·00	2	0	0
L-Cysteine	3·00	4	4	57

Each of the listed compounds was given in 10 per cent aqueous solution except L-glutamic acid, L-leucine, DL-methionine and L-phenylalanine which were given in 2·5 per cent aqueous solution because of their poor solubility in water. Because a large volume of fluid was needed to deliver high doses of L-leucine, DL-methionine and L-phenylalanine, only half the dose was given orally and the remainder subcutaneously. All of the other compounds were given orally. Sources of L-glutamic acid and MSG were purity checked by thin layer chromatography. Figures in the necrotic hypothalamic neurone column represent averages for each dose level.

Because of the widespread practice of weaning human infants on foods which are not only rich in natural glutamate content but may contain substantial quantities of glutamate (MSG) added for flavouring[10,11], it is important to establish whether damage to the infant central nervous system could follow from oral as well as from parenteral administration of glutamate[12]. We describe here experiments which demonstrate hypothalamic damage in infant mice following relatively low oral doses of glutamate, and also report that orally administered aspartate and cysteine can induce retinal and hypothalamic damage.

Seventy-five Webster Swiss albino mice, 10 to 12 days old, were given single oral doses of a 10 per cent aqueous solution of MSG at one of 5 dose levels (0·25, 0·5, 0·75, 1·0 or 2·0 g/kg). Ten control animals were intubated but given no treatment, and an additional 46 were given single oral doses of other test compounds, as shown in Table 1. Accurate dosage control was ensured by use of an improvised flexible gastric tube inserted gently through the mouth and oesophagus into the stomach. About 5 h after

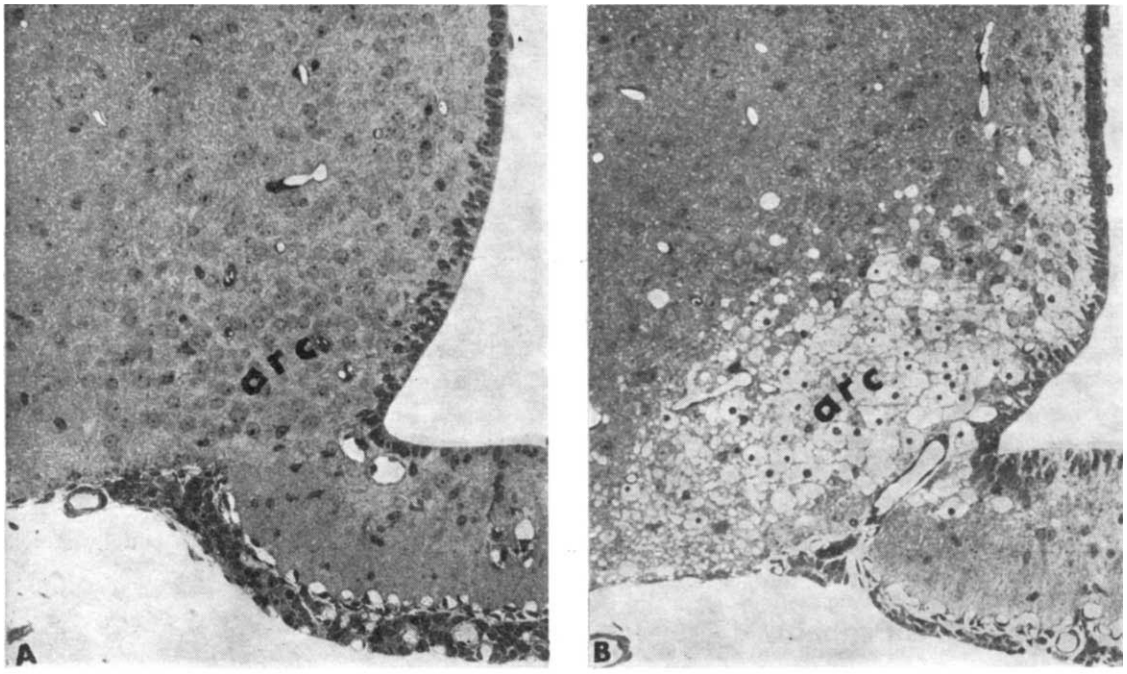

Fig. 1. *A*, Section through arcuate nucleus (arc) of hypothalamus from untreated 10 day old mouse. The tissue is well preserved by perfusion fixation and no signs of cytopathology are evident. Empty spaces are blood vessels dilated by perfusion fixation (×150). *B*, Section through arcuate nucleus (arc) from 10 day old mouse treated orally with MSG, 1 g/kg. There are approximately 33 necrotic cells in the arcuate region of this section (×150).

treatment, each animal was anaesthetized with chloral hydrate and killed by perfusion fixation of the central nervous system with 1·5 per cent glutaraldehyde and 1 per cent paraformaldehyde in 0·1 M cacodylate buffer. After 15 min of perfusion, the retinas and brain areas of interest were further fixed in osmium tetroxide and processed by a technique described elsewhere[9] which permits alternative examination of any specimen by either light or electron microscopy. To provide a rough estimate of the severity of brain damage at various dose levels, the hypothalamus of each animal was sectioned from its rostral (pre-optic) to caudal (post-infundibular) extent and necrotic neurones were counted in a representative section (1 μm thick) cutting across the arcuate nucleus at its level of maximal damage.

No evidence of cellular pathology was detected in the arcuate nuclei of any of the ten untreated control animals or in any of the ten animals treated with MSG at 0·25 g/kg (Table 1). Of the twenty-three animals given 0·5 g/kg doses of MSG, twelve (52 per cent) suffered hypothalamic damage; and of sixteen animals treated at 0·75 g/kg, thirteen (81 per cent) were affected. Nineteen animals (100 per cent) treated at 1 g/kg and seven (100 per cent) treated with 2 g/kg developed arcuate lesions. Necrotic neurone counts reflected considerable individual variability in response to MSG at any given dose level. But a comparison of average counts at each dose level revealed a consistent dose response relationship (Table 1). Brain sections from an untreated control animal and from an animal treated with MSG at 1 g/kg are illustrated in Fig. 1*A* and *B*.

We also found that a 1 g/kg dose of glutamic acid destroyed approximately the same number of hypothalamic neurones as a comparable dose of MSG, but neither sodium chloride nor sodium glutarate affected hypothalamic neurones at 3 g/kg. Most amino-acids tested (see Table 1) also failed to produce hypothalamic damage at 3 g/kg. Aspartate and cysteine, however, were striking exceptions because each animal treated with these compounds developed both retinal and hypothalamic lesions which seemed identical to those which are usually found after treatment with MSG. The possibility that glutamate and aspartate are additive in their toxic effect was suggested by the observation that every one of eight animals treated orally with a mixture of MSG (0·5 g/kg) and sodium aspartate (0·5 g/kg) developed a degree of hypothalamic damage characteristically seen in animals treated with either agent at 1 g/kg (Table 1).

Curtis[13] and others have found that glutamate, aspartate and cysteine comprise a select group of amino-acids (the "neuroexcitatory" amino-acids) which can depolarize nerve membranes. Whether the striking ability of this select group of compounds to induce neuronal necrosis in the immature central nervous system relates to their ability to depolarize nerve membranes needs further study.

Because glutamate is a naturally occurring constituent of dietary protein there has been little tendency to question its safety for human infant consumption. But, in our experiments, both glutamate and aspartate are toxic to the infant mouse at relatively low levels of oral intake and, when taken together, these common amino-acids have an additive brain damaging effect. Contrary to conclusions which others have reached from studies on adult animals[12] these experiments with tube fed infant animals raise serious questions concerning the advisability of supplementing the human infant diet with MSG.

This work was supported by grants from the National Institutes of Mental Health, US Public Health Service.

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Received January 5; revised April 16, 1970.

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Monosodium Glutamate and the Chinese Restaurant Syndrome

It has been suggested[1-4] that monosodium glutamate (MSG) is responsible for the "Chinese restaurant syndrome"—a burning sensation in the back of the neck spreading to the forearms and to the anterior thorax, accompanied by a feeling of infraorbital pressure, tightness and substernal discomfort. But no study of this phenomenon involved a double blind technique, or any other experimental condition which can be used to assess the significance of subjective reactions.

We have carried out a study on twenty-four healthy volunteers (seventeen males and seven females) aged between 18 and 34 (mean 25), using double blind and crossover techniques. MSG was administered at lunch time in doses of 3 g/subject in 150 ml. of beef broth. This was followed by other dishes (meat, vegetables and fruit).

Subjective and objective evaluations were carried out every 20 min during the following 3 h. The subjects, randomly divided into two groups, were told they were going to receive three different kinds of broth, one of which would contain MSG (the L isomer). Observations were carried out in two experimental sessions. In the first, eight subjects received MSG in the broth and sixteen had normal beef broth—in the second session, two days later, the eight subjects who had previously received MSG were given normal beef broth, and the other sixteen were given MSG in the broth.

The subjects were asked to specify every twenty minutes any particular feeling or sensation they were experiencing. They were supplied with a form which listed possible symptoms (headache, burning feeling, flushing, facial pressure, chest pressure, weeping, gastric distress, nausea, perspiration, tiredness), and there was space to record additional symptoms. The symptoms had to be scored as mild, moderate or severe. These parameters were considered to be subjective symptoms. At the same time, arterial blood pressure (max and min), pulse and respiration rate were recorded. These parameters were considered to be objective symptoms. The data were subjected to the χ^2 test, Bartlett-box test and variance analysis.

No differences in subjective symptoms were observed between the subjects given MSG and those who had normal beef broth (Tables 1 and 2). There was also no significant difference in the number of times each single symptom occurred in the 3 h period (Table 1) nor in the number of subjects who experienced symptoms at each time (Table 2). There was a tendency in both groups to experience more symptoms during the first hour.

It should be emphasized that nobody, either in the control or in the MSG group, experienced the burning feeling which is typical of the Chinese restaurant syndrome. Also, there was no difference in the intensity of the symptoms experienced by each group. Only one subject, a female 24 years old, experienced a panic-like syndrome after being given MSG. This lasted for about 20 min, but it was not associated with any significant modification of objective parameters such as arterial pressure, pulse or respiration rate.

There was no significant variation in objective parameters ($P > 0.05$). There was some tendency towards an increase in maximum blood pressure in those given MSG, but there was no variation in minimum values (Table 3). In fact, by variance analysis, there was a significant difference ($P < 0.01$) in the overall variation. It should be noted that all the blood pressure variations are within the physiological modifications which take place during a normal digestive process. Previous authors[2] have reported that MSG is responsible for the Chinese restaurant syndrome in a very small number of subjects. The same authors later studied the acute pharmacology of MSG using an experimental procedure which was not fully described[4]. No double blind trials or statistical analysis were reported. The high dose used and the route of administration were also very questionable.

From our observations, there is no evidence that Chinese restaurant syndrome follows ingestion of MSG in a beef broth. The only subject who described a panic-like syndrome did not experience any burning feeling. Moreover, many of the subjective symptoms considered to be part of the Chinese restaurant syndrome may also be present during a normal digestive process. We therefore believe that MSG administered orally in relatively high doses does not provoke any symptoms of Chinese restaurant syndrome.

We thank Dr S. Paglialunga for the statistical

Table 1. INCIDENCE OF THE VARIOUS SUBJECTIVE SYMPTOMS IN THE 3 H OBSERVATION PERIOD

Symptoms	Number of subjects experiencing the symptom		Number of times (observations) in which the symptom was experienced in the three hours	
	Control	MSG	Control	MSG
Headache	1	2	3	5
Burning feeling	0	0	0	0
Flushing	3	1	4	2
Facial pressure	0	1	0	2
Chest pressure	4	2	10	3
Weeping	0	1	0	1
Fainting feeling	1	2	2	4
Gastric distress	2	2	6	3
Nausea	0	1	0	2
Perspiration	1	1	1	2
Tiredness	5	4	15	17
Total	17	17	41	41

No difference between control and MSG groups was statistically significant ($P > 0.05$).

Table 2. INCIDENCE OF SYMPTOMS AT VARIOUS TIMES AFTER ADMINISTRATION

Time (min)	Number of subjects experiencing symptoms		Total number of symptoms experienced	
	Control	MSG	Control	MSG
20	5	6	7	14
40	6	5	7	7
60	5	4	7	5
80	4	1	6	1
100	4	1	4	1
120	2	2	3	1
140	2	1	1	1
160	1	0	2	0
180	1	0	1	0
		Total	38	30

No difference between control and MSG groups was statistically significant ($P > 0.05$).

Table 3. CHANGES IN BLOOD PRESSURE AFTER MSG

Time (min)	Maximum values		Minimum values	
	Control	MSG	Control	MSG
20	−1·54 ± 1·50	+0·67 ± 1·61	+2·96 ± 1·92	+1·71 ± 2·18
40	−3·29 ± 1·90	+1·08 ± 2·02	+1·58 ± 2·49	+4·00 ± 2·82
60	−5·71 ± 2·34	−0·87 ± 2·01	+0·79 ± 2·58	+0·96 ± 3·22
80	−4·38 ± 2·38	−1·42 ± 1·83	−3·67 ± 2·52	−1·04 ± 2·92
100	−5·29 ± 2·45	−2·29 ± 2·15	−6·83 ± 2·56	−0·67 ± 3·06
120	−5·25 ± 1·91	−0·37 ± 2·16	−6·08 ± 2·36	−3·12 ± 2·96
140	−3·79 ± 2·15	−1·54 ± 2·48	−4·79 ± 1·98	−3·83 ± 3·04
160	−3·75 ± 2·22	−3·92 ± 1·62	−0·17 ± 2·03	−0·67 ± 3·30
180	−4·83 ± 1·82	−0·79 ± 1·72	−2·50 ± 2·24	−1·04 ± 3·05

The figures show the mean differences (mm Hg) ± s.e. from the basal values of blood pressure. Basal values in control session = 114 ± 2 (max); 67 ± 1 (min); basal values in MSG session = 111 ± 2 (max); 69 ± 2 (min).