The effect upon mice of intraventricular injection of excitant and depressant amino acids

(*Received* 29 *August* 1963; *accepted* 19 *September* 1963)

A SHORT report has recently been published by Kita *et al.*[1] on the effects of intraventricular injections of some amino acids upon mice. The relative potencies of the substances which caused convulsions were similar to those of the same substances as excitants of spinal neurones of the cat[2, 3] and toad[4], and also of cortical neurones of the cat.[5] It was therefore of interest to extend the results of these workers to include other amino acids known to have strong excitatory and depressant actions on mammalian and amphibian neurones.

Small groups of 4-week-old mice of both sexes were used. The amino acids were dissolved in a mammalian Ringer solution and the range of concentration used for each substance is shown in Table 1. The solution to be tested (0·05 ml) was injected through the skull of unanaesthetised mice according to the method of Haley and McCormick.[6] Controls, in which the same quantity of a suspension of Indian Ink was similarly injected, were also carried out. Post-mortem sections performed

TABLE 1. (A) EXCITANT AMINO ACIDS

Group*	Amino Acids	No. of mice	Dose (μg/0·05 ml)	Effect
A	N-Methyl-D-aspartic acid	4	10	L
		9	2–5	+ + +
		4	1	+ (+)
B	N-Methyl-DL-aspartic acid	4	20–50	L
		6	5–10	+ + +
		5	2	+ +
D	N-Methyl-L-aspartic	4	50	+ +
		2	20	+
C	D-Homocysteic acid	8	20–50	+ + +
		2	10	+ +
C	DL-Homocysteic acid	8	25–50	+ + +
		4	10	+ (+)
E	L-Homocysteic acid	6	20–50	+
D	N-Methyl-DL-glutamic acid	4	50	+ + (+)
		2	20	+
E	D-Aspartic acid	5	50	+
E	L-Aspartic acid	5	50	+
F	L-Glutamic acid	2	150	+
		6	50–100	Nil

(B) DEPRESSANT AMINO ACIDS

Group*	Amino Acids	No. of mice	Dose (μg/0·05 ml)	Effect
A^1	3-Amino-1-propane-sulphonic acid	8	100	− − −
		4	50	− −
		4	20	−
B^1	γ-Amino-n-butyric acid	6	150–250	− − −
		6	100	− −
		4	50	Nil
	Taurine	13	100–150	− −
		6	20–50	Nil

* The symbols A, B . . . F and A^1, B^1 are intended to convey relative ranking in decreasing order of potency. It is, however, not possible for accurate quantitative comparisons to be made from these data.

immediately after these animals had been killed with ether showed that in four out of twenty control mice some of the injected fluid was located outside the ventricles. After injection of mammalian Ringer or Indian Ink suspension, the animals tended to be subdued in their behaviour for periods of 15–60 sec, but occasionally slight twitching or incoordination of movement was noted, presumably due to trauma produced by the injection.

In large doses, amino acids known to be strong excitants of single neurones[3] produced clonic flexor convulsions followed by spasm (usually of extensor type) and death. In Table 1 the effects of these lethal doses are denoted by "L".

Lower dosages caused gross hyperactivity, with running, leaping and violent scratching at the head or body. Flexor convulsions were frequently observed as part of this response. During recovery the animal was usually subdued and its responses to being disturbed remained sluggish over long periods (10 min to over half an hour). This pattern of effects is denoted "+ + +". Still smaller doses cause moderate (+ + or +) excitation, usually with somewhat incoordinated attempts at grooming, or a continued circling around the cage at a walking pace. During recovery, which took place after 1–5 min, the animals were subdued.

Depressant amino acids produced loss of muscle tone and gross incoordination of movement. The animals were unable to right themselves if placed on their back or sides, and tended after a short period to fall asleep. Recovery from these effects, denoted "— — —", was very slow (greater than 30 min). Smaller doses of the depressants produced incoordination of movement and transient flaccidity, after which gradual recovery occurred. Lethal dosages of the depressant amino acids were not determined and although 200 μg of 3-amino-propane-sulphonic acid impaired respiration in two of the four mice tried, one of which subsequently died, this result is of doubtful significance.

The relative potencies of the amino acids in both series is very similar to that found on other preparations and by other methods of drug administration. N-Methyl-D-aspartic acid was the most potent substance, but the minimum lethal dose found for this compound was about ten times greater than that reported by Kita *et al.*[1] It is interesting also that taurine had a depressant effect not greatly lower than that of γ-amino-*n*-butyric acid. This result is in accordance with those of Curtis and Watkins[5] on cat cerebral cortical neurones, although Purpura *et al.*[7] have reported that taurine has no action on cat cortical cells.

There would appear to be little justification for the suggestion of Kita *et al.*,[1] that N-methylaspartic acid may have a transmitter function in the central nervous system. As yet, there is no report in the literature of the occurrence of N-methyl-aspartic acid in nervous tissue, nor indeed in any biological tissue. In an investigation of the free acidic amino acids of sheep brain, no N-methyl-aspartic acid was found.[8] Further, the L-form, which would be the more likely enantiomorph to occur naturally in mammals (including rodents), is not significantly stronger than either L-aspartic or L-glutamic acids, both of which occur abundantly in nervous tissue. L-Cysteic and L-homocysteic acids are also potent excitants[3] and could well be considered if an L-amino acid is found to have a role as an excitatory transmitter in the central nervous system. However, in discussing this question, Curtis and Watkins[2, 3] have been led to conclude that a transmitter function of amino acids is unlikely.

Acknowledgement—The author wishes to thank Dr. J. C. Watkins for samples of amino acids.

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