Arginine protection against hydrazine toxicity

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BECAUSE of the increased use of hydrazine as a propellant fuel, considerable interest has arisen in prophylactic and therapeutic methods of counteracting its toxicity and consequently in the understanding of the basic mechanisms of its toxic action. Although it was thought originally that the toxicity of hydrazine, like that of thiosemicarbazide and various hydrazides, could be prevented by pyridoxal, experiments in our laboratory and others showed that pretreatment of animals with various forms of vitamin B_6 either had no effect on toxicity of hydrazine or even enhanced the seizure incidence and mortality from single doses over those found in animals receiving hydrazine alone. A survey of the published findings on hydrazine action suggested that a number of the effects could be rationalized if it were assumed that the primary action were on the structure of some essential membrane component or components in the affected tissues. Theoretical considerations (to be published in detail elsewhere) led to the suggestion that *one* of the modes of action might be the displacement by hydrazine of the guanidino group of an arginyl residue from interaction with two adjacent negatively charged phospholipid phosphate oxygen atoms, thus altering phospholipo-protein relationships in a membrane structure. Such a site also might be one of the loci of pharmacological action of ammonia and the various hydrazides that are known as psychic energizers.

From the above speculations it was suggested that arginine might exert a protective action against hydrazine toxicity, as it is known to do in the case of ammonia.[1] Of course, the present results in no way give direct support to the hypothesis that led to the testing of arginine.

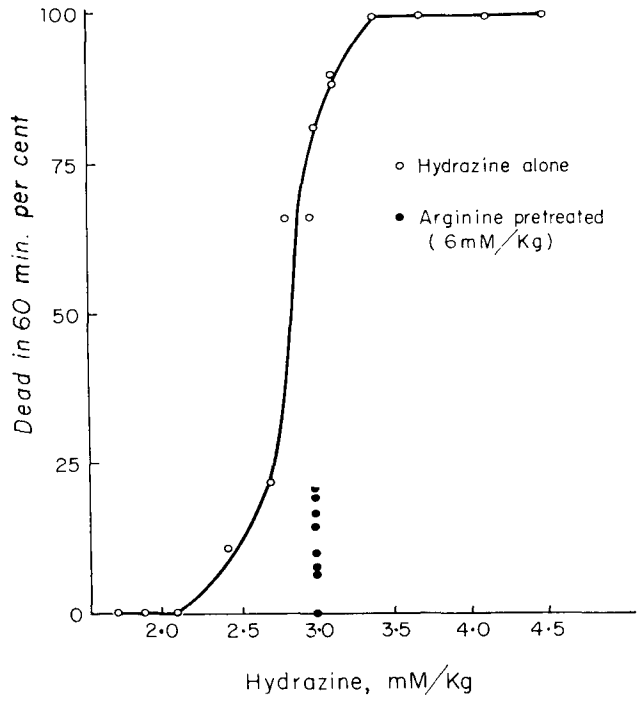

FIG. 1. Hydrazine dose–response curve. Protective effect of L-arginine.

Preliminary results showed that pretreatment of rats or mice with intraperitoneally administered arginine solutions significantly decreased the incidence of seizures and the lethality of single doses of hydrazine. The results in Fig. 1 show the hydrazine dose–response curve, with death in 60 min as endpoint, in male mice (25–35 g) of an inbred Swiss stock which had been starved for approximately

18 hr prior to a single intraperitoneal injection of approximately 0·1 ml of a freshly prepared hydrazine solution (pH 7·0). The injection of hydrazine was preceded by 30 min by the injection of 0·1 ml of physiological saline or by a similar volume of neutral test solution. It appears highly unlikely that the effects of arginine are attributable to any direct interaction with the hydrazine. Each point on the curve is for 10–20 mice. Also shown in Fig. 1 are the results of eight representative experiments of more than fifty performed over a period of a year in which similar groups of mice were given 6 mmoles of L-arginine/kg intraperitoneally prior to injection of 3 mmoles of hydrazine/kg. In addition to the greatly reduced mortality, there was a remarkable reduction in the total seizure incidence among the arginine-treated animals, usually less than 40 per cent of the latter group showing any seizure activity during the period of observation. It is not possible, of course, with the data from a single dose of arginine to determine the alteration in the nature of the entire dose–mortality curve of hydrazine. Further work on this point is in progress. In Fig. 2 are shown the results obtained with four different

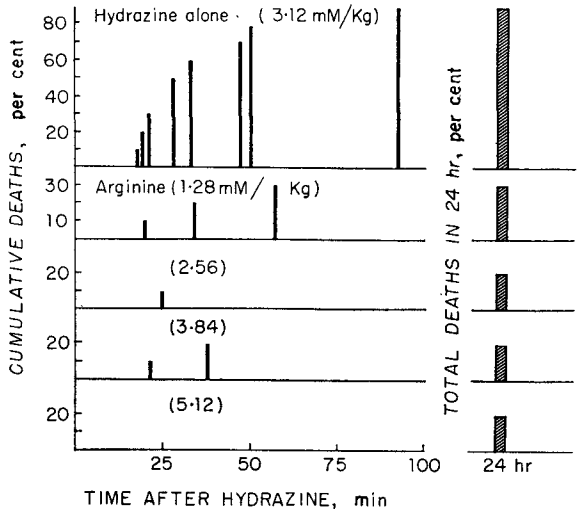

FIG. 2. Influence of four levels of L-arginine on extent of protection against hydrazine. Each line is drawn at the time of death of one or more mice, the height of the line representing the cumulative deaths expressed in percentage of total animals employed.

dosage levels of arginine. There was a marked protective effect in all groups, and at the highest dose of arginine, 5·12 mmoles/kg, no deaths occurred and no convulsions were noted for 110 min after the hydrazine injection, at which time 90 per cent of the control animals had died in convulsion. The effect of L-arginine appears to be stereospecific, since D-arginine* showed no protection (Fig. 3). The α-amino and α-carboxyl groups also appear to be necessary. Pretreatment with N-acetyl-L-arginine actually accelerated the rate at which hydrazine-treated mice died. L-Arginine ethyl ester, which itself was toxic at 6 mmoles/kg, also increased the rate of dying in tonic-extensor seizures and the percentage mortality at the level employed.

A detailed report will be forthcoming in which results with many more compounds as well as related metabolic experiments will be reported. However, the above results already may have important practical applications. It has been found that arginine solutions administered intravenously to

* The D-arginine, N-acetyl-L-arginine, and L-arginine ethyl ester were kindly donated by Dr. MiltonWinitz.

rabbits and cats *after* normally fatal doses of hydrazine can prevent or stop seizures and allow the animals to survive.[2] These results would appear to be applicable immediately to any cases of human exposure to hydrazine, since sterile pyrogen-free solutions of arginine approved for human use are available commercially. Intravenous infusions containing 20 g of L-arginine hydrochloride have been

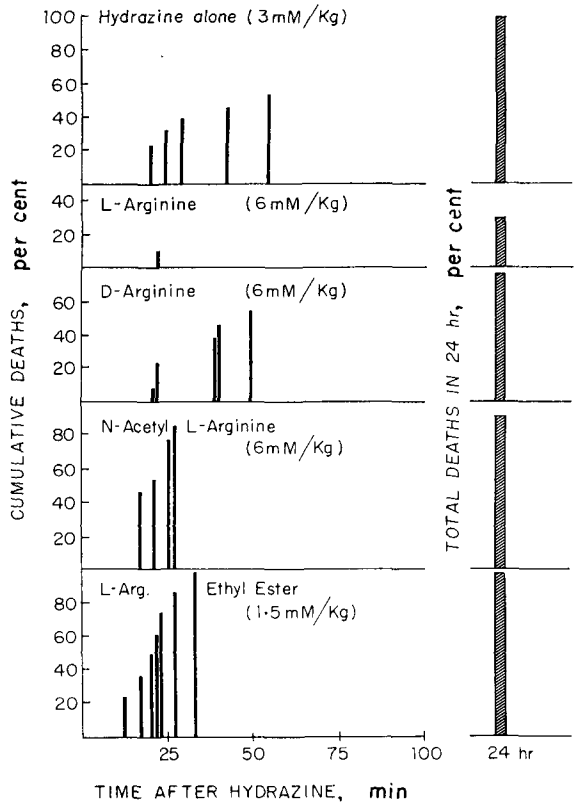

FIG. 3. Comparison of protective effects against hydrazine of L-arginine, D-arginine, N-acetyl-L-arginine, and L-arginine ethyl ester.

given to human patients with no ill effects.[3] Also it might be useful from a prophylactic point of view to give arginine-containing capsules *per os* routinely in situations where exposures to small amounts of hydrazine are unavoidable.

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Department of Biochemistry,
Medical Research Institute,
City of Hope Medical Center,
Duarte, Calif., U.S.A.

EUGENE ROBERTS
DAISY G. SIMONSEN
ETHEL ROBERTS

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