Effect of a Serotonin Antagonist on Radiation Lethality.* (28642)

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Recently Field, Mireles, and Dolendo(1) demonstrated that the serotonin antagonist 1-(N-methyl-piperidyl-4')-3-phenyl-4-benzyl-pyrazolone-5 (KB-95) possessed a significant ability to reduce the mortality produced by the nitrogen mustard HN_2. Because nitrogen mustard is in certain respects radiomimetric, it was considered of interest to test the effects of KB-95 on the lethal effects of whole-body X-irradiation.

Materials and methods. Animals used were CF-1 female mice, 7-8 weeks of age, and Badger male rats, 6 weeks of age. The KB-95 was kindly provided by Dr. Leonard B. Achor, of Sandoz Pharmaceuticals. The serotonin used was a product of the Upjohn Co. All injections were given intraperitoneally, in volumes of 0.01 ml/g of body weight. KB-95 was administered as a suspension in 5% gum arabic. Controls were given an equivalent volume of 5% gum arabic. X-Irradiation was done with a GE Maximar machine, operating at 250 KV and 15 ma, with 0.5 mm copper and 3 mm Bakelite filtration. The half value layer was 1.72 mm of copper. Mice were irradiated at approximately 38 r/min; rats, at 114 r/min. Doses of drugs and radiation are given with individual experiments.

Results. Table I presents the pooled results of 2 experiments, indicating that prior injection of 400 mg KB-95 per kg is significantly protective against 600 r of whole-body X-irradiation, although survival time of those mice that succumbed within 30 days increased by only 0.9 day. In single experiments, not listed, it was found that (a) administration of KB-95 2 hours after irradiation slightly increased per cent survival (from 33 to 50%) and slightly decreased mean survival time (from 11.3 to 10.6 days), and (b) administration of the KB-95 both before, and again 2 hours after, the irradiation slightly worsened the survival situation as compared to animals pretreated only (from 56% to 44%). However, none of these stated effects are statistically significant.

TABLE I. Effect of KB-95 on Lethality of Whole-body X-Irradiation of Mice.

Treatment	No. of mice	Survival No.	Survival %	Median survival time (days)
5% gum arabic, then 600 r	25	4	16	12.2
KB-95, 400 mg/kg then 600 r	26	16	62	13.1
KB-95, 400 mg/kg no X-ray	26	26	100	—

Table II lists the LD_{50} of whole-body X-irradiation, and the dose reduction factor (DRF) of KB-95, for both mice and rats. It is evident that the agent, though protective, is only mildly effective.

TABLE II. Whole-body X-Irradiation Dose Reduction by KB-95.

Species	$LD_{50(30)}$, r Control	$LD_{50(30)}$, r Treated	Dose reduction factor
Mice	615	687	1.12
Rats	650	710	1.09

A total of 72 rats and 170 mice was used for LD_{50} determinations.

Table III represents an attempt to determine whether or not the protective effect of KB-95 is due to its serotonin antagonism. The results again show protection by KB-95, even better protection by serotonin, and a bit less protection by the drug combination than by either drug alone.

TABLE III. Effect of Combined Serotonin and KB-95 on Lethality of Whole-body X-Irradiation to Mice.

Treatment	No. of mice	Survival No.	Survival %	Mean survival time, days
Saline, then X-ray	10	1	10	16.2
KB-95, then serotonin, then X-ray	10	7	70	13.7
KB-95, then X-ray	10	8	80	17.5
Serotonin, then X-ray	10	10	100	—
KB-95, then serotonin, no X-ray	5	5	100	—

Doses administered: X-rays, 700 r; KB-95, 500 mg/kg; serotonin, 100 mg/kg.

* This work was performed under the auspices of U. S. Atomic Energy Commission.

Discussion. The protective effect of the KB-95 against ionizing radiation differs from that against HN_2(1) in at least one important respect, namely that its protective effects appear limited to pre-irradiation treatment, whereas in the case of HN_2, the KB-95 actually appears more protective if given *after* the HN_2 than if administered prior to the toxic agent.

The experiment reported in Table III suggests that KB-95, an established antagonist of serotonin in the usual tests,† also somewhat antagonizes the radioprotective ability of the serotonin. This is in interesting contrast to the experiments of van den Brenk and Elliott (2), who found that several antagonists and anti-metabolites of serotonin were not themselves at all radioprotective and, furthermore, completely negated the radioprotective ability of the serotonin when given together with that agent.

Summary. The serotonin antagonist 1-(N-methyl-piperidyl-4')-3-phenyl-4-benzyl-pyrazolone-5 (KB-95) shows mildly protective effects against 600 r whole-body X-irradiation in mice, if administered prior to irradiation. The compound exhibits a dose-reduction factor of approximately 1.1 in both rats and mice.

† Unpublished data from laboratories of Sandoz, Inc.

1. Field, J. B., Mireles, A., Dolendo, E. C., PROC. SOC. EXP. BIOL. AND MED., 1962, v111, 1.
2. van den Brenk, H. A. S., Elliott, K., *Nature,* 1959, v182, 1506.

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Protective Effect of Neonatal Thymectomy on Mouse LCM Infection. (28643)

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A number of features of lymphocytic choriomeningitis (LCM) infection of mice indicate that the mechanism by which the cerebral form of infection induces fatal convulsions involves some form of tissue hyperreactivity(1,2). Symptomatology is closely correlated with lymphocytic infiltration of the meninges and choroid plexus, which is probably elicited by a hypersensitivity reaction. It is not clear whether the stimulus for induction of convulsions is the inflammatory infiltrate itself or some other hyperreactivity response such as cerebral vasospasm or edema. Agents which produce both lymphopenia and suppression of cell-mediated immune responses, such as delayed hypersensitivity and homograft rejection, protect against the disease manifestations of infection, although not suppressing growth of virus. These include total body x-irradiation(3), cortisone(4), and various antileukemic drugs, particularly folic acid antagonists(5,6).

Intraperitoneal (i.p.) infection of mice with a viscerotropic LCM strain rarely produces disease of the central nervous system, but produces illness and death primarily by massive pleural effusion(1). The pleural reaction is completely prevented in irradiated mice (3), suggesting a hypersensitivity component in the pathogenesis of this form of the disease as well. The prime importance of lymphocytic reaction is also indicated by the uniform association of the pleural effusion with lymphocytic infiltration of the mediastinal and pulmonary pleura(7).

The precise form of tissue hyperreactivity involved is not clear, although delayed-type hypersensitivity and the Shwartzman reaction have been considered. However, present understanding of the fundamental pathogenetic mechanisms of the diverse hyperreactivity syndromes is too incomplete to warrant equating the LCM mechanism with any particular one.