

CORRESPONDENCE

Effects of cysteamine given before and after X-irradiation under different temperature conditions on mortality of the fish, *Oryzias latipes* †

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1. Introduction

In the hibernating dormouse, *Glis glis*, Künkel, Höhne and Maass (1957), and Künkel and Schubert (1958) have reported that cysteine is able to give full protective action even 21 days after x-irradiation, if the animals are in the hibernating state during the irradiation and for the following 21 days. Since these interesting reports, several investigators have tried this kind of work with other vertebrates but no conclusive results have been published (Bacq 1965).

In a series of experiments with the small fish, *Oryzias latipes*, it has been well established that animals irradiated with lethal doses at 4°C and maintained under low temperature conditions (2-6°C) can survive for a long period, until the fish are brought to a higher temperature (e.g. 23°C) (Egami 1969 a, Etoh and Egami 1965, Hyodo-Taguchi and Egami 1969). From these experiments, the author has concluded that the development of radiation lesions in the fish is almost completely inhibited in the cool thermal environment, and the situation in fish seems to be similar to that in hibernating mammals. Therefore, experiments along the same line to Künkel's works were attempted with the fish. The results will be briefly dealt with in this report.

2. Materials and methods

Three series of experiments were carried out with adults of the red variety of *Oryzias latipes*, weighing 600-800 mg. Schemes of the experiments are given in figures 1-3. Before the experiments fish had been kept in water adjusted at 23°C (Groups 1, 2, 3, 7, 8, 13 and 14) or at 4°C (Groups 4, 5, 6, 9, 10, 11, 12, 15 and 16) for about a month. In all groups, fish were irradiated with x-rays (200 kV, 20 mA, 0.5 mm Cu and 0.5 mm Al filters) at 23° or 4°C in a small Lucite vessel containing water, exposure rate being 360 R/min. Total doses given in Series 1, 2 and 3 were 2.8 kR, 3.0 kR and 2.5 kR, respectively. Each fish of the cysteamine-treated groups was given an intraperitoneal injection of 0.02 ml. of Ringer's solution containing 20 mg of cysteamine per 10 ml. Fish injected with the same volume of Ringer's solution without cysteamine served as controls. Cysteamine solution was made immediately before the injection. The injection was also done under low or high temperature conditions, as shown

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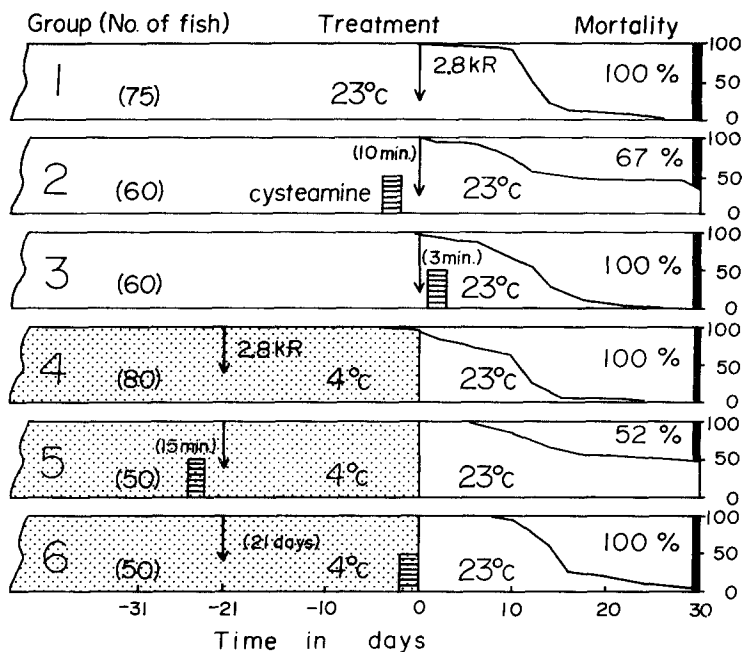


Figure 1. Cysteamine effects on mortality (Series 1). Diagram showing treatment and results. Cysteamine injection is shown as a hatched column. Vertical arrow indicates irradiation; dotted area, cooling period; length of vertical solid column, percentage of dead fish on the thirtieth day.

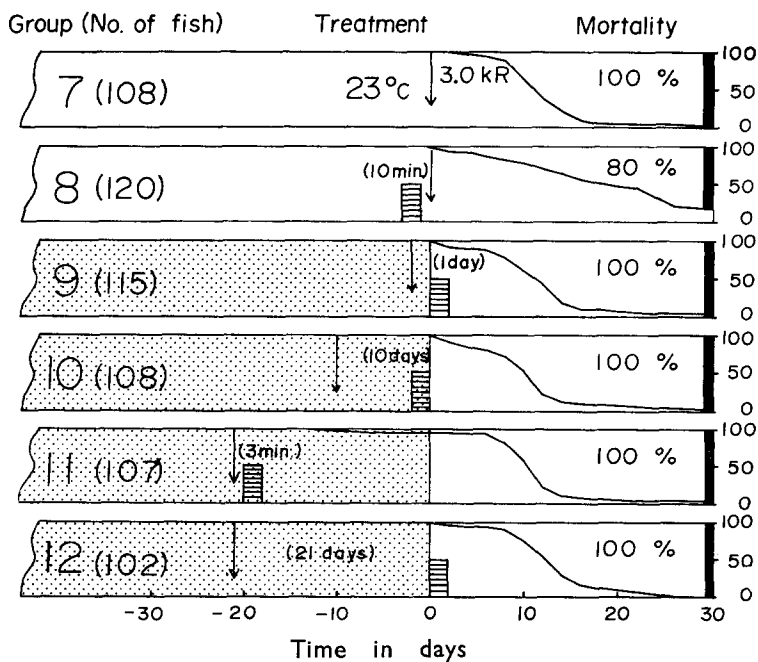


Figure 2. Cysteamine effects on mortality (Series 2).

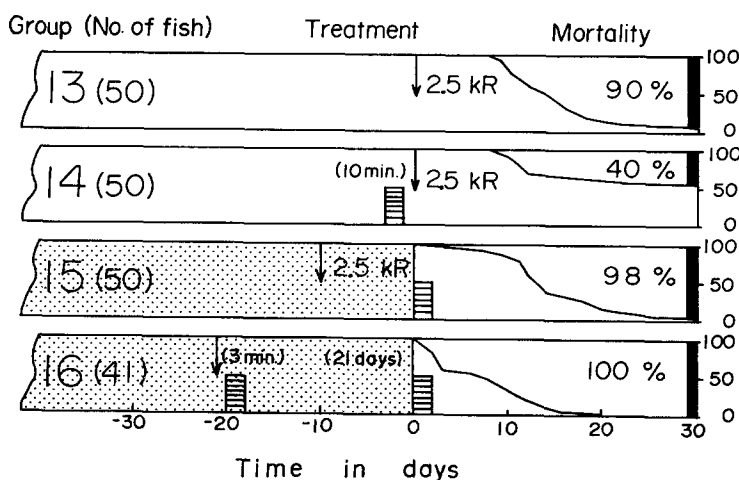


Figure 3. Cysteamine effects on mortality (Series 3).

in figures 1–3. After these experimental procedures at 4° or 23°C, every fish was transferred singly to a special small box kept at 23°C for a period of 30 days to examine the mortality rate.

3. Results and discussion

The daily mortality figures are summarized in figures 1, 2 and 3.

All fish irradiated with 2.8 kR of x-rays at 23°C and kept at the same temperature died within 30 days (Group 1). However, one-third of the irradiated fish survived for 30 days if cysteamine was given 10 min before the same dose of radiation (Group 2). This result indicates protective action by cysteamine against x-rays, and is in good agreement with previous experiments (Egami 1969 b). On the other hand, no radioprotective action of cysteamine was recognizable, if the chemical was applied 3 min after irradiation (Group 3).

In Group 4, fish were irradiated and kept at 4°C for 3 weeks after irradiation, and then transferred to 23°C. No fish died before the transfer, and a majority of fish died within 15 days after the transfer. In Group 5, fish were injected with cysteamine 15 min before irradiation at 4°C, and radioprotection by the chemical was clearly demonstrated. In Group 6, cysteamine was injected 21 days after irradiation, just before the moment when the fish were transferred from 4° to 23°C. The result shows that treatment with cysteamine after irradiation was without significant effect on the survival rate of the fish, no fish being alive on the thirtieth day after transfer to 23°C.

The second series of experiments was designed to confirm the results of the first series with many individuals of fish and with different radiation doses. Essentially the same results were obtained. Comparison of Groups 7 and 8 reveals typical radioprotective action of cysteamine, as in Groups 1 and 2. In Group 9 fish were kept at 4°C for 1 day after irradiation, and injected with cysteamine immediately after transfer to 23°C. In Group 10, cysteamine was given to irradiated and cooled fish on the tenth day after irradiation and then transferred to 23°C. In Group 11, cysteamine injection was done only 3 min

after irradiation under cool conditions and the fish were subsequently kept at 4°C. In Group 12, fish were maintained at 4°C for 21 days after irradiation and cysteamine was given immediately after transfer to 23°C. In these four groups (Groups 9–12), all fish died within 30 days after transfer to 23°C, and no indication of protection by cysteamine against x-rays was demonstrated.

From the third series of experiments, consisting of four groups, it is evident that the lethal effect of x-rays was weakened only when cysteamine was given before irradiation even at low temperature: the results of Groups 13, 14 and 15 were in agreement with those of the preceding series. In Group 16, cysteamine was twice given after irradiation, and cysteamine could not weaken lethal effects of x-rays. On the contrary, the substance exerted toxic effects on the fish.

So far as the present results are concerned, the protective action of cysteamine was very marked, if fish were treated with cysteamine before being irradiated. However, cysteamine was not effective if applied to fish after irradiation even at low temperature. More experiments of this kind with cysteine and non-SH protectors are now in progress.

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