hol after 24 h. The 2×, 4×, 8× metaphases were scored from diploid and tetraploid treated root tips. For each interval three slides were prepared from three root tips and from each slide approximately 800–1000 cells were scored from Feulgen preparations.

Results and discussion. Mitotic cell cycle time was estimated as the interval between two successive divisions of a selected synchronous cell population, i.e. C-metaphases that emerge as tetraploid cell population at the time of the next division, seen as 4× cells in 2× roots and 8× cells in 4× roots. The percentage cells with C-metaphases was scored 1 h after treatment and subsequently at 2 h intervals until 32 h.

The proportion of C-metaphases in diploid pearl millet 1 h after treatment was 7.7%. A comparable percentage of 4× metaphases was observed only in the sample taken 22 h after the treatment. Thus the mitotic cell cycle time of diploid (IP 1475) pearl millet was estimated to be 21 h (22–1 h).

The proportion of C-metaphases in tetraploid pearl millet 1 h after treatment was found to be 7.8%. The highest proportion of C-metaphases at the 8× level was only 6.8%. This level of accumulation of 8× metaphases was found only in the sample taken 30 h after the treatment; therefore, the cell cycle time of tetraploids was estimated to be 29 h. Thus, the difference in mitotic cell cycle time of the diploid and its autotetraploid amounts to 8 h, i.e. an increase of 38%.

Comparison of mitotic cell cycle times in embryonic tissue of pearl millet diploids and tetraploids also revealed a small difference; the duration was longer in 2n embryos[10]. However, the same study using vg 212, IP 482, IP 482T and C_8 HF lines revealed no difference in mitotic cell cycle time in endosperm of diploid and tetraploid lines. They suggested that the effect of polyploidy can be different on different cell types within the same species. Results of the present experiment with root meristems, while agreeing with this general conclusion, further demonstrate that 1) the tetraploid root meristems show prolonged mitotic cell cycle compared to their related diploid and 2) that the cycle time in a more mature tissue (i.e. radicle) is longer than that in the embryonic tissue (early embryo)[11].

Frequencies of metaphases expressed as percentages at first and second divisions of root meristems of diploid (IP 1475) and tetraploid pearl millet

Sampling time (h)	Metaphases 2n		4n	
	2×	4×	4×	8×
1	7.70		7.80	
2	8.25		7.13	
4	8.50		1.40	
12	4.85		1.00	
14	1.90		0.50	
16	1.10	0.30	0.90	
18		0.90	0.60	
20		6.00	0.20	
22		7.38		
24		2.25		1.10
26		1.40		1.70
28		1.10		3.10
30				6.80
32				1.10

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Lack of mutagenicity of irradiated glucose in *Salmonella typhimurium* using host-mediated assay

M.B. Varma, K.P. Rao, S.D. Nandan and M.S. Rao

Department of Genetics, Osmania University, Hyderabad-500007 (India), 16 June 1982

Summary. Experiments were conducted to study the ability of irradiated glucose to induce reverse mutations in *S. typhimurium* by host-mediated assay. The results revealed no significant increase in the frequency of reverse mutations compared to controls.
Key words. Salmonella typhimurium; glucose, irradiated; mutations, reverse; host-mediated assay.

Preservation of food and food components by radiation offers several advantages in reducing spoilage. However, the suitability of the resulting products for human consumption is debatable in view of conflicting results on genetic hazards of irradiated food stuffs reported by several investigators[1–4]. Carbohydrates being one of the main components of food material, greater importance was attached to the studies on the effects of irradiation on sucrose and glucose in particular. Mutagenic effects of irradiated sugar solutions were first indicated by Ehrenberg[4] on barley. Subsequently, other investigators reported positive evidence of mutagenic effects in plant and mammalian cells after treatment with irradiated sugar solutions[6,7]. Host-mediated assay is a valuable test system which determines the ability of the host to detoxify the test compounds with regard to their mutagenic activity[8]. Using this test system, experiments were undertaken to assess the mutagenicity of irradiated glucose using mice as hosts and *Salmonella typhimurium* G-46 as indicator organism.

Materials and methods. D-Glucose powder (M/s Sarabhai Chemicals, Baroda, India) was packed in polythene bags and irradiated in the presence of air at room temperature (25 ± 1 °C) with a ^{60}Co gamma source (Bhabha Atomic Research Centre, Bombay) at a dose rate of 56–58 Gy/min. The dose rate was measured by ferrous sulphate dosimetry. In all the experiments random-bred Swiss male mice were used.

Two series of studies were carried out, namely short-term and long-term feeding studies, using mice of 7–8 and 4–5 weeks of age, respectively.

In the short-term studies, irradiated glucose (0.2 or 50 kGy) was dissolved immediately after irradiation in distilled water. The concentrations of the glucose solutions were 10%. 2 ml of these solutions were administered daily to the mice in the test groups over a period of 7 days. 2 ml of distilled water or a 10% solution of nonirradiated glucose were given to the mice in the two control groups. Each group consisted of six animals.

In the long-term studies two sets of experiments were carried out (1) with irradiated glucose (0.2, 2.0, 20 or 50 kGy) dissolved in distilled water immediately after irradiation and (2) with irradiated glucose stored for 2 months at room temperature before use. In these experiments the concentrations of the glucose solutions were again 10% and 2 ml of these solutions were administered daily to the mice in the test groups over 8 weeks. The animals in the two control groups received 2 ml of distilled water or a 10% solution of nonirradiated glucose in a similar manner.

Following treatment, the test was performed using a modified procedure of Gabridge and Legator[8]. 2 ml of saline suspension of log phase bacteria at a concentration of 3×10^8 cells/ml were intraperitoneally injected into each mouse. The bacteria were allowed to grow for 3 h. Animals were sacrificed and 1 ml of physiological saline solution was injected into the peritoneum of each mouse and the bacteria recovered. The bacteria of all six animals of a group were pooled and plated on selective media plates[9]. The plates were incubated at 37 °C for 48 h, and scored for the number of revertants and total bacterial counts to determine the mutation frequency. The significance of differences in the number of revertants between contol and treated groups was tested following the method of Goldstein[10] under the asssumption that the number of revertants corresponds to a Poisson distribution.

Frequency of reverse mutations in *S. typhimurium* with irradiated glucose

Experiment	No. of animals used	Average number of induced reversion mutants (mean ± SE)
1. Short-term studies		
(irradiated glucose, used immediately after treatment)		
Control 1 (distilled water)	6	9.00 ± 1.06
Control 2 (nonirradiated glucose)	6	7.56 ± 0.62
0.2 kGy	6	9.12 ± 0.79
50 kGy	6	8.13 ± 1.29
2. Long-term studies		
a) Irradiated glucose, used immediately after treatment		
Control 1 (distilled water)	6	8.11 ± 0.82
Control 2 (nonirradiated glucose)	6	7.96 ± 1.22
0.2 kGy	6	8.37 ± 0.98
2.0 kGy	6	9.36 ± 1.45
20.0 kGy	6	9.17 ± 1.86
50.0 kGy	6	8.73 ± 2.06
b) Irradiated glucose, stored		
Control 1 (distilled water)	6	8.45 ± 0.49
Control 2 (nonirradiated glucose)	6	8.26 ± 1.97
0.2 kGy	6	7.32 ± 1.73
2.0 kGy	6	8.78 ± 2.36
20.0 kGy	6	9.48 ± 1.08
50.0 kGy	6	8.50 ± 2.97

$p > 0.05$.

Results and discussion. The results obtained in the short-term study showed no increase in the frequency of revertants in the treated groups as compared to the control groups. Similarly, the data obtained from the long-term studies with irradiated glucose used immediately after treatment or after a storage time failed to show any mutagenic effect compared to the controls. The results of the present investigation indicate the inability of irradiated glucose to induce mutagenic effects in *S. typhimurium* by host-mediated assay.

In our earlier studies, irradiated glucose failed to induce sex-linked recessive lethals in *Drosophila melanogaster*[11], dominant lethal mutations and micronuclei in mice[12,13]. However, Aiyar and Rao[14] reported that irradiated sugar solutions were mutagenic towards *S. typhimurium* in vitro.

It is known that radiation-induced hydrogen peroxide plays a precursor role in the ultimate toxicity of irradiated carbohydrates. Irradiation of water or of aqueous solutions of carbohydrates produces hydroperoxide radicals (HO_2) and hydrogen peroxide (H_2O_2), which can react with carbohydrates to produce toxic substances like glyoxal[15]. If solid carbohydrates are irradiated and then dissolved in water, production of HO_2 and H_2O_2 and subsequently toxic substances is not likely.

In this study, the experiments were carried out using irradiated glucose in powder form and no toxicity was observed. This suggests that reaction products of HO_2 and H_2O_2 with organic substances may have been responsible for the toxic effects observed in earlier studies.

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