

Table 1. FREQUENCY OF CHLOROPHYLL MUTATIONS IN  $X_2$  PROGENIES OF BARLEY SEEDS, IRRADIATED WITH ULTRA-VIOLET RADIATION

Treatment	No. of $X_1$ families studied	% of families segregating for mutations	No. of $X_2$ plants scored	% of $X_2$ plants mutated
Control	135	0	9,722	0
Oxygen pressure (4 h)	50	0	3,450	0
Ultra-violet (4 h)	80	2.5	6,042	0.07
Oxygen—4 h + ultra-violet 4 h	65	6.2	4,262	0.14
Ultra-violet—4 h + immediate oxygen saturation	60	0	4,560	0
Soaking in water—4 h + ultra-violet (4 h)	127	7.1	7,877	0.35

of ultra-violet light is mainly through the excitation of the molecules and this may set in a prolonged free-radical reaction in the presence of oxygen<sup>9</sup>. This may be the probable explanation for the enhanced effect of ultra-violet light in the present case. Such reactions are short-lived and hence are not available for post-irradiation oxygen effect, unlike X- or  $\gamma$ -irradiated barley seeds, which respond to post-irradiation oxygen treatment<sup>10</sup>. The increased effect of ultra-violet light on soaked seeds appears to be due to one or more of the following three causes: (1) a better penetration of ultra-violet light in hydrated systems<sup>9</sup>; (2) the initial metabolism set in the seeds due to soaking, making them a more susceptible system; (3) the effects of ultra-violet light on water molecules which are in excess in soaked seeds and which can act indirectly on the genetic material. The failure to induce mutations with ultra-violet irradiation alone in hexaploid wheat<sup>4,5</sup> may be attributed to its high polyploid nature as well as the chromosomal origin of most of the induced mutations in this crop.

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### In vivo Investigation of the Removal of Trace Elements from Nucleic Acids of Yeast by Ionizing Radiation

INVESTIGATIONS by several authors<sup>1,2</sup> have demonstrated that paramagnetic metal ions produce a certain amount of resistance to radiation in cells. This fact is due partly to the redox effect of these ions<sup>3</sup>, and partly to the facilitation of forbidden transitions of excited electron states<sup>4</sup>. Since different nucleic acids contain trace elements in significant amounts<sup>5</sup>, and since the biological activity of nucleic acids is dependent to a large extent on their trace-element content<sup>6-8</sup>, it seemed essential to us to investigate the trace-element content of those most important macro-molecules of the cell before and after irradiation. It was also shown that by roentgen irradiation removal of  $Ca^{++}$  and  $Mg^{++}$  from nucleohistone is possible, whereby at the same time reduction in DNA occurs<sup>9</sup>.

Determinations of the trace-element content were carried out by means of neutron activation analysis<sup>10</sup>. The removal of trace elements from nucleic acids was determined between 0 and 24,000 rads of cobalt-60 irradiation. Part of the trace elements was detected in the low

molecular cell fraction after irradiation in this dose-range. Another part might be bound by metal binding molecules, or else might be re-bound at ionizing points caused by the irradiation. In this way the decrease of the trace element content at doses above 8,000 rads could be explained. Yet it seems that in the investigation range of dose no diffusion of trace elements occurs through the cell wall owing to changes in permeability.

Yeast was suspended in aqua bidest., and air was bubbled through for 5 h. For the irradiation carried out at 23° C the centrifugated yeast was re-suspended in aqua bidest. (20 ml./g yeast). Then the yeast was centrifugated (distribution of trace elements in this centrifugate is given in Table 3), the low molecular compounds were extracted with 60 per cent ethanol and the nucleic acids with 1.0 M ammonium formate as recently shown<sup>11</sup>.

Table 1. TRACE ELEMENT CONTENT ( $\mu g$  IONS PER SAMPLE) OF NUCLEIC ACID OF YEAST AFTER DIFFERENT IRRADIATION DOSES (MEAN VALUE OF SIX DIFFERENT CULTURES)

Dose (rad)	Cu	Mn	Ni	Zn	p
0	30.1 ± 3.0	5.9 ± 1.2	32.3 ± 6.0	130 ± 27	3,870 ± 190
3,000	27.7 ± 2.7	7.3 ± 1.3	30.0 ± 5.9	87 ± 18	3,720 ± 180
8,000	13.1 ± 1.4	3.7 ± 0.8	14.9 ± 3.1	66 ± 14	3,950 ± 200
24,000	15.9 ± 2.0	1.7 ± 0.7	10.1 ± 2.2	27 ± 9	3,637 ± 180

Table 2. TRACE ELEMENTS ( $\mu g$  IONS PER SAMPLE) IN THE ALCOHOL SOLUBLE 'LOW MOLECULAR' FRACTION OF THE YEAST AFTER DIFFERENT IRRADIATION DOSES

Dose (rad)	Cu	Mn	Ni	Zn	p
0	4.4 ± 0.4	1.6 ± 0.3	< 2.0	5.2 ± 1.0	581 ± 30
3,000	7.5 ± 0.7	1.2 ± 0.2	< 2.0	6.2 ± 1.1	530 ± 25
8,000	9.8 ± 1.0	2.2 ± 0.5	6.8 ± 1.5	8.8 ± 1.7	517 ± 26
24,000	8.4 ± 0.9	1.1 ± 0.2	15.6 ± 3.0	5.4 ± 1.1	528 ± 24

Table 3. RELEASE OF TRACE ELEMENTS FROM THE CELL CAUSED BY IRRADIATION IN  $\mu g$  PER SAMPLE

Dose	Cu	Mn	Ni	Zn	p
0	0.34 ± 0.04	0.14 ± 0.03	2.4 ± 0.5	0.4 ± 0.1	15.4 ± 1.6
3,000	0.36 ± 0.05	0.15 ± 0.04	2.0 ± 0.4	0.5 ± 0.1	17.2 ± 1.8
8,000	0.38 ± 0.05	0.18 ± 0.04	2.2 ± 0.5	0.5 ± 0.1	19.1 ± 1.0
24,000	0.36 ± 0.05	0.16 ± 0.03	2.3 ± 0.5	0.4 ± 0.1	16.2 ± 1.7

The metal ions liberated from nucleic acids may cause a diminution of the biological activity of the macromolecules (eventual change of the tertiary structure). Moreover, they may cause slight damage (early stages) to the cell by operating as inhibitors or stimulators of enzyme systems<sup>12</sup>.

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## BIOLOGY

### A Barley Endosperm Bioassay for Gibberellins

SINCE the review of bioassay techniques for gibberellins by Phinney and West<sup>1</sup>, several new techniques have been proposed<sup>2-4</sup>. These are based on the increased elongation of plant tissues induced by gibberellins. Recent work has defined other biological characteristics of gibberellic acid ( $GA_3$ ), notably a stimulation of the hydrolytic enzymes in germinating cereal grains<sup>5</sup>. Paleg<sup>6-8</sup> demonstrated that this response resides in the cereal endosperm, and the bioassay reported here was developed from procedures used in those studies.