

**AUXIN TRANSPORT IN COLEOPTILE SEGMENTS OF X-IRRADIATED  
*ZEA MAYS* SEEDLINGS<sup>1</sup>**

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**Abstract**

The effect of X-rays on the transport of <sup>14</sup>C—methylene labeled indoleacetic acid in sub-apical coleoptile segments of *Zea mays* L., was examined. Transport determinations were made of segments in vertical and horizontal orientation. X-irradiation had no significant effect on either the velocity or intensity of basipetal auxin transport in the segments oriented vertically after irradiation. However, irradiation did significantly reduce the amounts of auxin transported when the segments were oriented horizontally. In horizontal assemblies the lateral differences in auxin transport were accentuated by X-radiation. This action on transport can be correlated with the decreases of geotropic curvature in X-irradiated seedlings.

**Introduction**

Holmsen, Teas & Koch (1964), have examined the effect of gamma-irradiation on geotropic curvature of pea and corn seedlings. Using relatively high radiation exposures (40 kR) they found that irradiation decreased the rate of curvature development to continuous gravitational stimulus. Radiation-induced change in the auxin relationship of the seedling was inferred at least for the pea, since the decrease in georesponse could be reversed by exogenous auxin. Since there is increasing evidence that tropic response is in part mediated by lateral differences in rates of auxin transport (Naqvi & Gordon, 1964, 1966; Shen-Miller & Gordon, 1964) we wondered if the change in geosensitivity of a plant exposed to ionizing radiation might not be associated with a change in hormonal transport characteristics. Results show that exposure of the corn seedling to X-rays causes alterations in the rates of auxin transport in horizontally but not vertically oriented coleoptile sections.

**Materials and Methods**

Seeds of *Zea mays* L. (hybrid 64a x 22R, University of Wisconsin)† were soaked for 3 hours in tap water (60°C) and placed, embryo up, on moistened paper pads (2 ply "Kimpak") in plastic trays. The trays were covered and placed in the dark room. To inhibit mesocotyl growth, the germinating seeds were exposed to red light (Wratten filter 1A; incident irradiance ca. 750 μw/cm<sup>2</sup>) between the 56th and 68th hours and then returned to the dark for a total growth period of 96 hours. All experimental manipulations were performed under green "safe-light" at 25 ± 1°C. (Shen-Miller & Gordon, 1964).

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1. Work supported by the U.S. Atomic Energy Commission.
  2. Division of Biological and Medical Research, Argonne National Laboratory, Argonne, Illinois (U.S.A.) where the work was carried out.

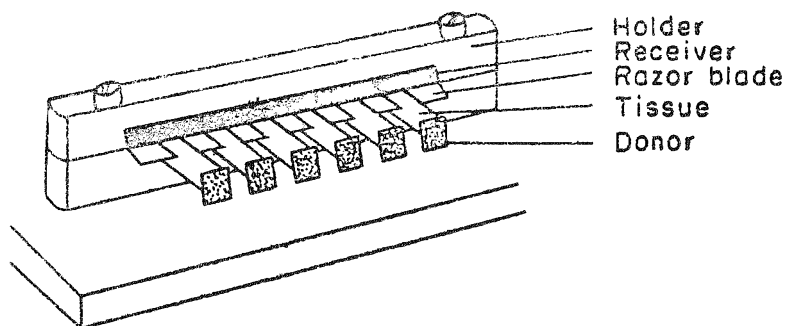


Fig. 1. Auxin-transport assembly, in horizontal orientation, for the determination of lateral differences in basipetal transport.

Seedlings were exposed to X-rays from a General Electric Company Maximar, driven at 250 kvp and 15 ma, filtration 0.5 mm cu. Exposure rates of either 25 or 60 roentgens (R) per minute were used. To examine transport in unsplit coleoptile segments in normal vertical orientation, single 7.0 mm sections cut 1 mm below the coleoptile tip were used. The base of the section rested on a "receiver" block of 1.5% agar (3.7 x 4 x 1 mm). The apical end of the section was covered with a "donor" block of agar (2.7 x 2.7 x 2 mm) containing  $^{14}\text{C}$ -methylene labelled indoleacetic acid ( $^{14}\text{C}$ -IAA, 13.3 curies/mole) with a concentration of 0.2 mg/1. Before use the auxin was purified by paper chromatography.

To determine the effect of orientation on transport similar sections, in groups of 6 were mounted on a razor blade which penetrated about 1 mm into the base of the section along its narrow axis. Each half of the bisected base rested on a receiver block of agar (35 x 2.5 x 1.5 mm). The donor blocks of agar plus auxin described above were placed on the apical ends of the sections. These assemblies, illustrated in Figure 1, were oriented so that the longitudinal axis of the coleoptile segments were either horizontal or vertical.

At termination of the transport period radioactivities in the sections and in the donor and receiver blocks were determined by liquid scintillation counting using Naqvi's scintillant (Naqvi, 1963). Each datum given is the mean of 3 replicates. Curves were fitted by the method of least squares. Analysis of variance was used to evaluate statistical significance.

## Results and Discussion

Seedlings were exposed to 0, 200, 300 and 400 R while vertically oriented. Auxin transport through coleoptile sections of these seedlings was then determined, care being taken to maintain vertical orientation throughout the transport period. The results,

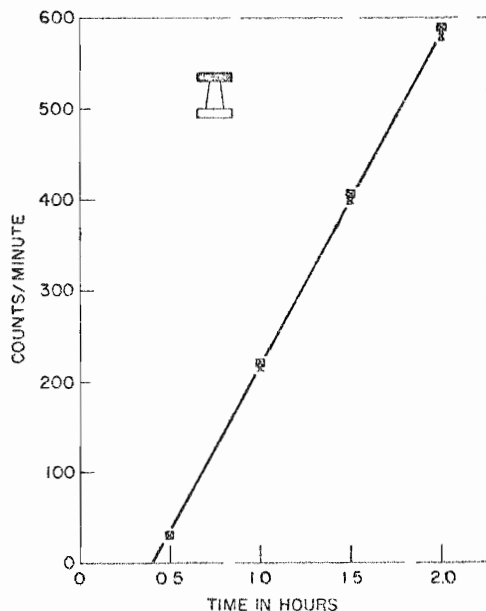


Fig. 2. Effect of X-ray exposure (0,200, 300, 400 R) on the velocity and intensity of  $^{14}\text{C}$ —IAA transport in vertically oriented sections. The ordinate represents the radioactivity found in the receiver block after various transport intervals.

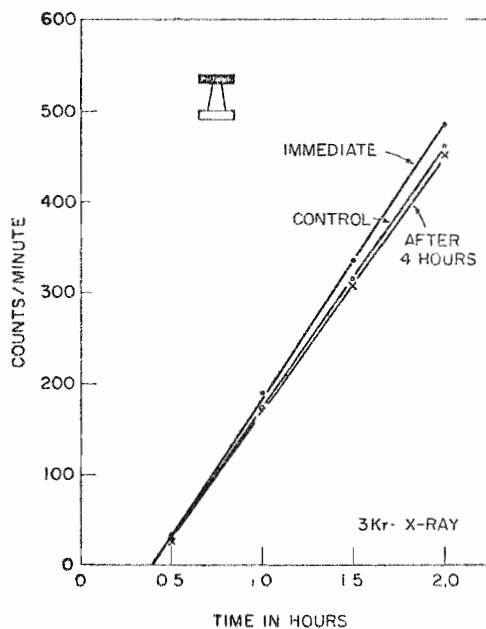


Fig. 3.  $^{14}\text{C}$ —IAA transport in segments of coleoptiles exposed to 3kR X-rays. Transport determinations were made immediately and 4 hours later after irradiation.

shown in Figure 2, indicate that these X-rays exposures have no significant effect on either the intensity or velocity of transport of  $^{14}\text{C}$ —IAA. Our observation on the lack of effect of ionizing radiation on the rate of auxin transport in vertically oriented monocot tissues supports the observations of Skoog (1935) with nonetiolated dicot plants. He also found no effect of X-rays on the amount of IAA basipetally transported in vertically oriented *Pisum* and *Helianthus* seedling segments.

A heavier exposure of X-rays without filter, 3 kR was then tried. Transport determinations were made immediately and 4 hours after X-ray exposure. The results, given in Figure 3, again show no radiation effect on transport of the auxin in vertically oriented sections.

The effect on auxin transport of horizontal orientation of the tissues from X—irradiated seedlings was then determined. Assemblies (Fig. 1) were made with sections from seedlings exposed to 0, 200, 300 and 400 R. During the transport period, 1.5 hours, the sections were maintained in either horizontal or vertical orientation. The transport data are given in Table I. The following inferences may be drawn from the Table. First, orientation of the organ segments had no significant effect on the absorption of auxin by the tissues. Residual activities in the donor blocks were the same. Second, horizontal orientation decreased the amount of auxin transported through

TABLE I. Effect of X irradiation on basipetal transport of  $^{14}\text{C}$ -IAA (CPM) in corn coleoptile sections, transport time: 1.5 hours.

Assembly parts	TREATMENTS												
	Horizontal						Vertical						
	C	200 R	300 R	400 R	400 R	C	200 R	300 R	400 R	400 R	C	200 R	
Donor block	1230a	1260a	1230a	1260a	1260a	1220a	1220a	1230a	1230a	1240a			
Tissues	340a	410b	395b	387b	387b	312c	308c	321c	321c	321c			
Receiver blocks	514a	448b	437b	429b	429b	561c	572c	527c	527c	555c			
Receiver block halves	U	L	U	L	U	L	R	L	R	L	R	L	R
	719a	395b	77c	371d	75c	362d	77c	352d	278e	283e	288e	284e	264e
Ratio	Lower	3.3a	4.8b	4.8b	4.6b	Upper							

Means within rows across the table not followed by the same subscript differ at odds greater than 99:1.

U Upper, L Lower, R Right, L Left.

the tissues. All of the activities in the receiver blocks are less in the horizontal assemblies. This impairment of transport in horizontal segments is significantly enhanced by X—irradiation, which does not affect the auxin transport in vertically oriented tissues. Third, the impairment of transport by both orientation and irradiation is also manifest as an increased retention of activity in the tissues. Fourth, splitting of the tissue bases did not in itself result in a material difference in the transport occurring through the adjacent lateral portions of the organ segment; the activities in the receiver block halves were essentially the same in the vertical assemblies. However, horizontal orientation enhanced the auxin transport in the lower halves and reduced it in the upper tissues. Both of these changes in transport were accentuated by irradiation; the ratios between lower and upper halves change from 3.3 to about 4.8. No significant difference between X—ray exposures was noted in either absorption or translocation of the auxin.

To check the effect of X—irradiation on the geotropic response, intact corn seedlings were oriented horizontally, immediately after exposure (0, 200, 300, 400 R) for 1.5 hours. The seedlings were then shadowgraphed and their curvatures, the angle between lines bisecting the tip apex and the coleoptile base, were determined. Table 2 shows that irradiation reduced the response to continuous geostimulation for 1.5

TABLE 2. Effect of various doses of X-irradiation on curvature response of *Zea mays* seedlings (n = 30)

Treatments	Degree curvature $\pm$ S.E.
Control	20.70 $\pm$ 0.69
200 R	15.10 $\pm$ 0.43
300 R	15.80 $\pm$ 0.43
400 R	15.20 $\pm$ 0.59

hours by 1/4. It may be suggested, therefore, that the impairment of geotropic response in the irradiated plants in the work of Holmsen *et. al.*, (1964), was a consequence of radiation-induced lateral differences in the basipetal transport of auxin.

We stress, then, that the X—irradiation effect on auxin transport was manifest only when the tissues were horizontally reoriented. It may be inferred, therefore, that the action of the X-rays on transport occurs at a locus activated by geotropic stimulation. It may also be suggested that this locus has a relatively high radiosensitivity; the X-ray response was already maximum at 200 R in the restricted exposure range examined. It would be of interest to determine the dosage-response relations of this sensitivity.

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