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Dependence of Basipetal Polar Transport of Auxin Upon Aerobic Metabolism[1]

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Summary. The movement of IAA-^{14}C through coleoptile segments of *Avena* and *Zea* has been investigated under aerobic and anaerobic conditions. The results are as follows: *Zea.* Using a 5-mm segment and a 2-hour transport period anaerobic conditions reduced the total uptake of ^{14}C from an apical donor by 74 % and the proportion of the total found in the receiving block by at least 45 %. Anaerobic conditions reduced total uptake from a basal donor by 58 % but no ^{14}C reached the apical receiving block in either air or N_2. Uptake from apical and basal donor blocks in N_2 is closely similar.

The presence of ^{14}C in the basal receiving blocks, and its absence in the apical receiving blocks, in N_2 suggests that even in anaerobic conditions movement of IAA is polarized basipetally, although the movement occurs at only a fraction of the rate found in air.

Anaerobic conditions induced a similar reduction in basipetal movement of IAA in upper and lower 5-mm segments taken from the apical 10 mm of a *Zea* coleoptile.

Using 10-mm *Zea* segments no ^{14}C was recovered in the receiving blocks at the basal end of the segment after 2 and 4 hours in N_2 whereas large amounts were recovered in air.

Avena: Using 5-mm segments and a 2-hour transport period the total uptake of ^{14}C from an apical donor is reduced by 83 %. Movement of ^{14}C into the basal donor is totally inhibited in N_2. Total uptake of ^{14}C from a basal donor is reduced by 61 % in nitrogen and no ^{14}C reached the apical receiving blocks regardless of the atmospheric conditions.

A time course for the movement of ^{14}C into the basal and apical receiving blocks through 5-mm segments showed that in air the amount in the basal receivers increased for 4 hours and then remained approximately uniform. In N_2 no significant ^{14}C reached the receivers until 6 to 8 hours after the application of donors but even then the amounts were about 12 to 14 % of that in aerobic receivers. Movement of ^{14}C into apical receivers was similar in air and in nitrogen and even after 6 to 8 hours the amount of radioactivity barely reached significant levels.

The extent to which the polarity of auxin transport in shoots depends upon aerobic metabolism has been the subject of several investigations in which metabolic inhibitors and anaerobic conditions have been used. Inhibitors of metabolism such as DNP, KCN, NaN_3 and indoacetate have been shown generally to reduce the rate of basipetal transport of auxin (16, 18). On the other hand, exposure of tissues to low partial pressures of O_2 have yielded very conflicting results. It is reported, for example, that movement of natural auxin through woody shoots of apple is reduced by 95 % in atmospheres containing less than 5 % O_2 (12) and that auxin movement is unaffected when pine stem sections are placed in N_2 (4). In experiments with *Zea mays* coleoptiles, Hertel and Leopold (13) found that the amount of labelled IAA passing through a 5-mm segment is reduced by 66 % in an atmosphere of N_2, whereas Naqvi, Dedolph, and Gordon (15) used 10-mm segments and concluded that the rate of basipetal transport of labelled IAA is the same under aerobic and anaerobic conditions. Goldsmith (5) has found the basipetal transport of labelled IAA through 10-mm segments of *Avena sativa* coleoptiles to be totally inhibited under anaerobic conditions.

Recently, in experiments conducted simultaneously with those reported in this paper, Goldsmith (6, 7) found the rate of basipetal transport of IAA in *Zea* coleoptile sections to be about 15 mm hour^{-1} in air and about 2 mm hour^{-1} in N_2. Even under anaerobic conditions the slow movement of IAA appears to be polarized basipetally and independent of concentration gradients.

[1] Throughout this research project Mary Martin held a Science Research Council Postgraduate Studentship.

Both the reported rate of basipetal transport of IAA in air and the amount of IAA transported per unit time appear to be too great to be accounted for in terms of simple diffusion. In addition, it is difficult to imagine how either the movement of auxin into the receiving block against a concentration gradient, or the polar nature of the movement, can be achieved without the utilization of metabolic energy. Because of these difficulties and the contradictory nature of the results in the recent literature, we have reinvestigated the dependence upon aerobic metabolism of basipetal and acropetal movement of IAA in coleoptiles of both *Zea* and *Avena*, paying particular attention to the length of the segment employed and to the exclusion of O_2 from the atmosphere surrounding the tissue and the intercellular spaces within the tissue.

Materials and Methods

Plant Materials. Seeds of *Avena sativa* L. var. Svalof Victory I (husked) and *Zea mays* L. var. Giant Horse Tooth were soaked in distilled water for 3 hours and then sown in washed, moist vermiculite. The seedlings were grown in total darkness at 25°. The *Avena* seedlings were harvested after 5 days when the coleoptiles and mesocotyls were respectively 30 to 40 mm and 45 to 55 mm in length. The *Zea* seedlings were harvested after 6 days.

Indole-3-Acetic Acid. The movement of IAA in the coleoptile sections was followed by using carboxyl labelled IAA-^{14}C of specific activity 32.1 c/mole (U.K. Atomic Energy Authority, Amersham, Berks, U.K.). The manufacturer's analysis of this material indicated a purity greater than 98 %. Chromatographic autoradiographic analysis of the stock solution at the end of these experiments revealed 2 radioactive spots: a major spot having the same R_F as a pure sample of twice re-crystallized IAA (Light and Co., U.K.) and a minor spot having a slightly lower R_F than IAA. Although this impurity is obviously undesirable, it does not affect the validity and conclusions of this study since Miss Bridget Parkes has recently shown by chromatographic autoradiographic analysis in this laboratory that even when the slightly impure radioactive IAA solution is applied in agar blocks to the apical end of coleoptile sections, IAA is the only radioactive molecule to emerge into a receiving block at the basal end. The movement of radioactivity into the receiving blocks is thus a true reflection of the movement of IAA.

The IAA was applied to the coleoptile sections at a standard concentration of 5 μM in blocks of 2 % agar, and was collected in receiver blocks of plain 2 % agar in contact with the opposite ends of the sections. The blocks were prepared from "Difco" Bacto-agar and distilled water.

Anaerobic Atmosphere. The most critical part of this investigation was to achieve strictly anaerobic conditions in the tissue and then to apply the donor blocks to the tissue without the strictness of these conditions being impaired. These problems were overcome by constructing a transparent perspex chamber which could be repeatedly evacuated and flushed with N_2 after the tissue and the donor and receiving blocks had been placed inside. When anaerobic conditions had been attained the tissue could be brought into contact with the donor blocks containing the IAA without reopening the chamber. The design of the chamber is shown in figure 1.

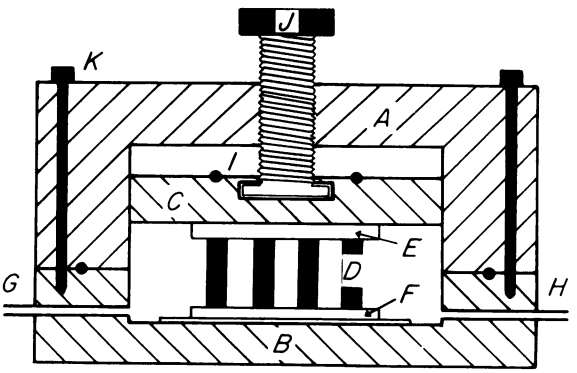

FIG. 1. Vertical section through chamber used to achieve anaerobic conditions in the tissues. For explanation see text.

It comprises 2 hollow cylinders (A and B) each closed at one end. The open ends of the cylinders fit together to form a gas-tight joint on a rubber O-ring. In the upper half there is a closely fitting piston (C) having a device to prevent its rotation while being raised and lowered. Raising and lowering of the piston is achieved by turning the screw (J) which is free to rotate in the top of the piston. When fully raised the piston makes a gas-tight seal with the top of the cylinder by compressing the rubber O-ring (I). In the bottom half of the chamber (B) 2 plastic tubes (G and H) are fitted to facilitate evacuation and flushing of the chamber. When the chamber was closed by screwing together the 2 halves (A and B) with screws (K) it was completely gas tight and would hold a partial vacuum (33 cm of Hg, the maximum applied) for several hours. The arrangement of the chamber in relation to the N_2 supply, which was humidified to prevent drying out of the sections, and to the vacuum pump and manometer, is shown in figure 2.

In all cases O_2-free N_2 or argon (British Oxygen Co.) from commercial tanks was used to flush the chamber. The 2 gases yielded closely similar results.

Experimental Procedure. Sections 5 mm and 10 mm in length were excised 1 mm below the apex of the coleoptile from which the primary leaf had previously been removed. The sections were prepared and the experiments conducted at 25° under continuous dim red irradiation in the spectral band from 620 nm to 700 nm with peak irradiation at

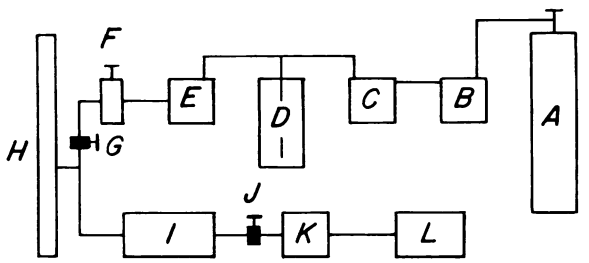

FIG. 2. Arrangement of the plant chamber (I) in the gas flow system used to achieve anaerobic conditions in the tissues. A) Nitrogen tank; B, C) humidifiers; D) constant head pressure and safety device; E) water trap; F) pressure regulating valve; G) tap; H) manometer; I) plant chamber; J) tap; K) CO_2 trap containing KOH; and L) vacuum pump.

660 nm. The radiant flux at the level of the plants, measured with a Kipp-Zonen Thermopile and a Hewlett-Packard D.C. Null Voltmeter (Model 419A), was 128 ergs cm^{-2} sec^{-1}. No significant difference could be detected between the amounts of IAA transported through sections kept under continuous red light and those prepared under red light (20-min exposure) and kept in darkness during the 1-hour flushing period and the 2-hour period of transport. However, this finding should not be interpreted as indicating that the latter coleoptiles transport IAA at the same rate as those which have never been exposed to red light.

Twenty sections were used in each experiment, 10 being placed in anaerobic conditions in the chamber previously described and 10 in a normal aerobic atmosphere in an identical chamber. Three receiving blocks each 3 × 19 × 1 mm in dimensions were placed on the surface of the piston of each chamber (fig 1, E). A batch of 5 coleoptile segments was placed on 2 of the receiving blocks (fig 1, D), the one batch having their apical ends, and the other batch their basal ends in contact with the block. The third receiving block was not placed in contact with any tissue and served merely as a check for possible contamination of the receiving blocks from the atmosphere within the chamber. The radioactivity of this block at the end of the transport period never exceeded the normal background count.

The donor block of dimensions 13.5 × 19 × 1 mm was placed on an inverted planchet on the floor of the chamber (fig 1, F). The piston was raised to its maximum height and the chamber was then closed; the sections remained suspended from the receivers and were not in contact with the donor block. The effect on IAA transport of both length of flushing time and number of evacuations has been investigated. Flushing times of 1 to 3 hours were examined, in each case the chamber was evacuated and refilled with N_2 3 or 6 times during the first half-hour of the flushing period. No significant difference could be found in the radioactivity in the sections and in the receivers with these various procedures. A 1-hour flushing period and three successive evacuations thus appeared to remove all traces of O_2 from the tissue. The following standard procedure was therefore adopted: 1 of the chambers was flushed with N_2 for 1 hour and during the first half-hour of this flushing period the chamber was evacuated to 33 cm of mercury and refilled with O_2-free N_2 6 times at 5-minute intervals. At the end of the 1-hour flushing period the piston was lowered by adjusting the screw (J, fig 1) to bring the sections into contact with the donor blocks. Nitrogen continued to flow through the chamber during the transport period. The sections in normal air were allowed to remain suspended from the receiving blocks for 1 hour before being lowered on to the donor block. Goldsmith (6) has shown that sequential partial evacuation of sections has no effect on the ability of the tissues to transport IAA.

At the end of the transport period the chambers were opened and the receiving blocks placed on separate planchets. The donor block was bisected and each half placed on a separate planchet. These planchets were then stored in darkness at −20° until they were prepared for counting. The sections were divided into upper and lower halves and stored under the same conditions as the blocks until they were prepared for counting.

Preparation of Material for Counting. The procedures were similar to those used by Goldsmith (5, 6, 8). The aluminum planchets were boiled in 1% KOH for 3 minutes, washed in distilled water and dried. The agar blocks were prepared for counting by adding a drop of distilled water to the planchet which was then gently warmed on an electric hotplate. The molten agar was spread evenly over the planchet with a needle and then dried.

The plant sections were dried in an oven at 80° for 1 hour. The 5 apical or basal half sections in a particular treatment were pooled and ground together in 0.5 ml of chloroform. The suspension was then transferred quantitatively to a planchet and the chloroform evaporated. One drop of distilled water was then added and the ground plant material spread evenly over the surface of the planchet which was then dried on a hotplate.

Sample Counting. Radioactivity was counted with a thin window, low background, Nuclear-Chicago gas flow counter (Model C115; Scaler Model 8703) in which sample changing was automatic. The background count of this instrument was consistently between 2 and 3 cpm. Samples were counted for either 2000 counts or 60 minutes. Self absorption by the plant material was estimated by the method of Goldsmith and Thimann (8) and the radioactivity data in this paper are given in cpm per section corrected for both background and self absorption.

Each experiment was carried out at least twice. In cases where sufficient data are available standard

errors are shown, and in other cases the number of individual experiments contributing to the mean data in the figures is stated.

Results

Movement of IAA Through Zea *Coleoptiles During a 2-Hour Period.* The basipetal and acropetal movement of IAA through a 5-mm segment of *Zea* coleoptile was investigated under aerobic and anaerobic conditions. The amounts of ^{14}C found in the upper and lower halves of the coleoptile segments and in the receiving blocks are shown in figures 3 and 4, in which the data are the means of 16 and 14 independent experiments respectively. When the donor is applied at the apical end of the section (fig 3) the total ^{14}C found in the tissue and receiver block was 173 cpm under aerobic conditions and 45 cpm under anaerobic conditions. Uptake of radioactivity from an apical donor is thus reduced by 74 % under anaerobic conditions. Of the total radioactivity taken up by the segments, 17 % was found in the receiving blocks under aerobic conditions but only 9 % under anaerobic conditions. Since radioactivity in the receiving block is confined to the IAA molecule these data show that under anaerobic conditions not only is the uptake of IAA reduced by 74 % but the amount of IAA moving through the section into the receiving block is reduced by approximately 45 %.

The amounts of IAA reaching the receiving block under aerobic and anaerobic conditions do not necessarily give a true picture of the extent to which the active basipetal transport is reduced under anoxic conditions since some radioactivity might reach the receiving block under anaerobic conditions by simple diffusion. If this is so the percent reduction in anaerobic conditions will depend upon the transport period and the length of section. The actual decrease in the active basipetal transport of IAA through these 5-mm sections must thus be at least 45 % and may be much greater.

When sections are supplied with donor blocks at their basal ends (fig 4) the total radioactivity taken up into the sections and receivers is 109 cpm in air and 46 cpm in anaerobic conditions. Uptake is thus reduced by 58 %. Under anaerobic conditions the total radioactivity taken up from the donor is the same regardless of whether the donor is applied at the apical or basal end of the segment. In air, on the other hand, the uptake from a basal donor is only 63 % of that from an apical one.

Under anaerobic conditions the distribution of ^{14}C in the section and receiver is somewhat but not exactly similar when the donor is applied at the apical and basal ends. The half section next to the donor contains about 30 to 40 cpm, the further half section about 4 to 8 cpm and the receiving block 0 to 4 cpm. The similarity in acropetal and basipetal movement as judged from the distribution of ^{14}C in the segments and receivers after a 2-hour transport period under anaerobic conditions strongly suggests that the movement is due principally to diffusion. Nevertheless, the amount of ^{14}C in the receiver blocks of sections having an apical donor was significantly higher than that in the receiver blocks of sections having a basal donor. Moreover, there appears to be slightly more ^{14}C in the half section next to the donor and less in the half section furthest from the donor when the donor is supplied at the basal end of the section than when it is applied at the apical end.

These facts suggest that even under anaerobic conditions the movement of IAA basipetally, while occurring at only a fraction of the rate observed in aerobic conditions, is slightly greater than acropetal movement. It is possible that in tissues deprived of O_2 just enough energy is made available by anaerobic metabolism to enhance slightly the rate of basipetal movement of IAA above the rate expected from diffusion alone.

Under aerobic conditions the total amount of ^{14}C taken up from a basal donor is only 63 % of that taken up from an apical donor. Moreover, the distribution of radioactivity within the sections and receivers is quite different. There is the same amount of radioactivity in the half section next to the donor regardless of whether the donor is applied to the apical or basal end. In contrast, the further half section and the receiver block have a large amount of radioactivity with apical donors but little or none with basal donors. These facts suggest that cells at the apical and basal end of

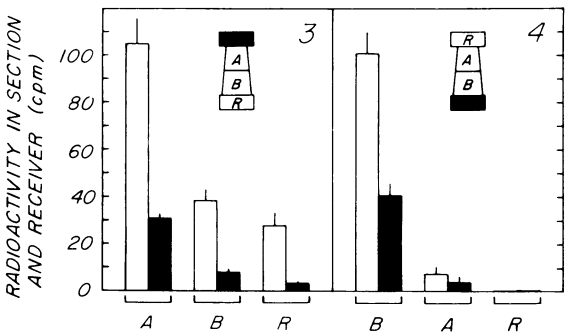

FIG. 3. Basipetal movement of IAA in 5-mm *Zea* coleoptile segments after 2 hours in air (white columns) and in anaerobic conditions (black columns). Radioactivity on a per section basis is given for the apical (A) and basal (B) half of the section and for the receiver (R) as indicated in the inset diagram. Donor block shaded. Vertical line at the top of each column shows the standard error of the mean. Data shown are the mean of 16 individual experiments.

FIG. 4. Acropetal movement of IAA in 5-mm *Zea* coleoptile segments after 2 hours in air (white columns) and in anaerobic conditions (black columns). Arrangement of donor is shown in the inset diagram. Data shown are the mean of 14 individual experiments and the vertical line at the top of each column shows the standard error of the mean. Notation as in figure 3.

the segment have an equal capacity to take up IAA. The basipetal transport of the IAA away from the apical half of the segment most probably accounts for the greater total uptake of ^{14}C by sections supplied with apical donors. This point is discussed further in relation to the *Avena* data.

Movement of IAA Through 5-mm Segments Taken From Different Regions of Zea *Coleoptiles.* The basipetal movement of ^{14}C was compared in 5-mm segments taken 1 mm below the apex (upper segment) and 6 mm below the apex (lower segment) of *Zea* coleoptiles under both aerobic and anaerobic conditions. The results are shown in figures 5 and 6 in which the data are the means of 3 independent experiments.

For the upper and lower segments in air, both the total amounts of ^{14}C taken up into the section and receiving blocks (119 cpm and 109 cpm respectively), and the amounts recovered in the receiving blocks (18.5 % and 23 % of the total taken up respectively) were closely similar. However, the amount of ^{14}C in the apical half of the upper 5-mm segment was more than the amount in the apical half of the lower segment, and the amount in the basal half of the upper segment was less than that in the basal half of the lower segment. Since the total ^{14}C in the upper and lower segments is closely similar, it appears that the cells in the apical half of the upper segment retain more ^{14}C than those of the apical half of the lower segment. Movement of ^{14}C taken up apically towards the basal end of the segment seems to occur more readily in the lower segment in air.

Under anaerobic conditions the total ^{14}C taken up from donors is 41 cpm for the upper segment and 49 cpm for the lower segment. This corresponds to reductions of 65 % and 56 % of the amounts taken up in air respectively. Of the total amounts taken up under anaerobic conditions, the amounts of ^{14}C recovered from the receiving blocks of the upper and lower segments are 6.2 % and 5.1 % respectively (cf. 18.5 % and 23 % respectively in air).

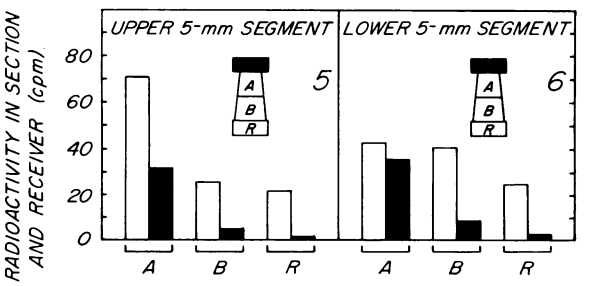

FIG. 5 and 6. Basipetal movement of labelled IAA in 5-mm segments taken 1 mm (fig 5) and 6 mm (fig 6) below the tip of *Zea* coleoptiles after 2 hours in air (white column) and in anaerobic conditions (black column). Mean of 3 independent experiments. Arrangement of donor blocks (shaded) and sections is shown in the inset diagrams. Notation as in figure 3.

Thus in the upper and lower segments anaerobic conditions lead to a closely similar reduction both in uptake of IAA-^{14}C from the donor and in the amounts moving basipetally into the receiving blocks. On the basis of these results it would be predicted that under anaerobic conditions no significant ^{14}C would reach the receiving block in a 2-hour transport period when a 10-mm segment is used. This prediction has been tested experimentally.

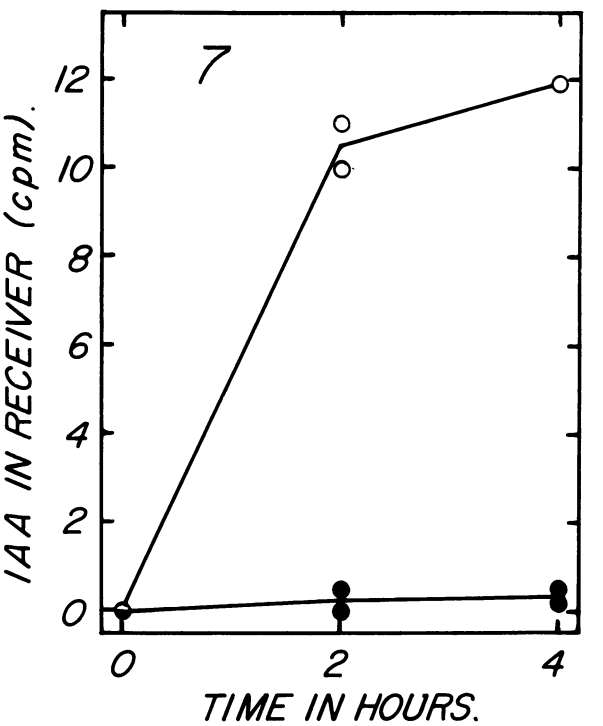

FIG. 7. Time course for the basipetal movement of labelled IAA into the receiving block at the basal end of a 10-mm *Zea* coleoptile segment in air (—○—○—) and in anaerobic conditions (—●—●—). Each point is the mean of 5 coleoptiles, the data being expressed on a per coleoptile basis.

Transport Through a 10-mm Segment of Zea *Coleoptile.* The amounts of ^{14}C reaching the receiving block at the basal end of a 10-mm segment taken 1 mm below the apex of a *Zea* coleoptile has been determined after transport periods of 2 and 4 hours in both aerobic and anaerobic conditions (fig 7). No significant ^{14}C was found in receivers under anaerobic conditions after 2 or 4 hours whereas considerable amounts of ^{14}C occurred in receivers under aerobic conditions.

The movement of IAA into receiving blocks at the basal ends of 10-mm *Zea* coleoptile segments is therefore totally inhibited during 2 and 4 hour transport periods under anaerobic conditions. This finding confirms the prediction made on the basis of the data in the previous section.

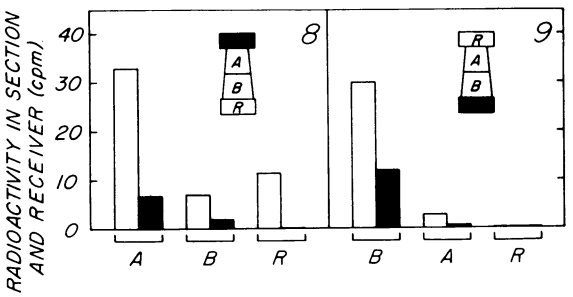

FIG. 8 and 9. Basipetal (fig 8) and acropetal (fig 9) movement of labelled IAA in 5-mm segments of *Avena* coleoptiles after 2 hours in air (white columns) and in anaerobic conditions (black columns). Arrangement of donor blocks and sections is shown in the inset diagrams. Notation as in figure 3. Data presented are the mean of 3 individual experiments.

Movement of IAA Through Avena *Coleoptile Segments During a 2-Hour Period.* The basipetal and acropetal movement of IAA through 5-mm segments of *Avena* coleoptiles was determined under aerobic and anaerobic conditions. The amounts of ^{14}C in the apical and basal halves of the segments and in the receiving blocks are shown in figures 8 and 9.

In sections supplied with apical donors the total ^{14}C in the sections and receiving blocks was reduced from 52 cpm in air to 9 cpm in anaerobic conditions, a reduction of 83 %. Of the total ^{14}C taken up from the donor, 22.4 % was found in the receiving block in air and none in the receiving blocks under anaerobic conditions. There is therefore a total inhibition of IAA movement into receiving blocks at the basal end of 5-mm segments of *Avena* coleoptiles which are deprived of oxygen during the 2-hour transport period.

When sections were supplied with donors at the basal end the total radioactivity taken up from the donor was 34 cpm in air and 13 cpm in N_2; a reduction of 61 %. In air, closely similar amounts of ^{14}C were found in the half sections next to the donor regardless of whether the donor was at the basal or apical end, but little or no ^{14}C moved into the further half section and receiver in sections with a basal donor (fig 9). This polarity of movement probably accounts for the decreased total uptake (66 %) from basal donors in air. The half-section next to the donor appears to have the capacity to take up a limited amount of IAA and when this limit is reached further uptake from the donor depends upon the transportation of IAA from this zone, a phenomenon which occurs only when the donors are at the apical end of the segment. This situation in *Avena* is almost identical with that found in *Zea* (figs 3 and 4).

Under anaerobic conditions the total amounts of ^{14}C taken from apical and basal donors in *Zea* were identical. In *Avena* under anaerobic conditions the amount taken up from an apical donor was about 33 % less than that from a basal one but the actual difference in the total number of counts taken up from the apical and basal donors was only 4 cpm. Since the *Avena* data in figures 8 and 9 are means of 3 individual experiments a difference of 4 cpm is too close to the experimental error to establish a significant difference between the total uptake from an apical and a basal donor under anaerobic conditions.

The amount of ^{14}C in the basal half of *Avena* sections supplied with basal donors is reduced by 60 % in anaerobic conditions. This is in close agreement with the corresponding value for *Zea*, namely 61 %.

In the 2-hour transport period in air the polarity of IAA movement into the receiving block is absolute; no ^{14}C reaches the apical receiver. In the absence of O_2, however, no ^{14}C reaches the receiver regardless of its position. Within the tissue, however, there appears to be a slight tendency for more ^{14}C to move into the further half section, and less to remain in the half section next to the donor, when the donor is at the apical end of the segment. This tendency was also noticed in the *Zea* coleop-

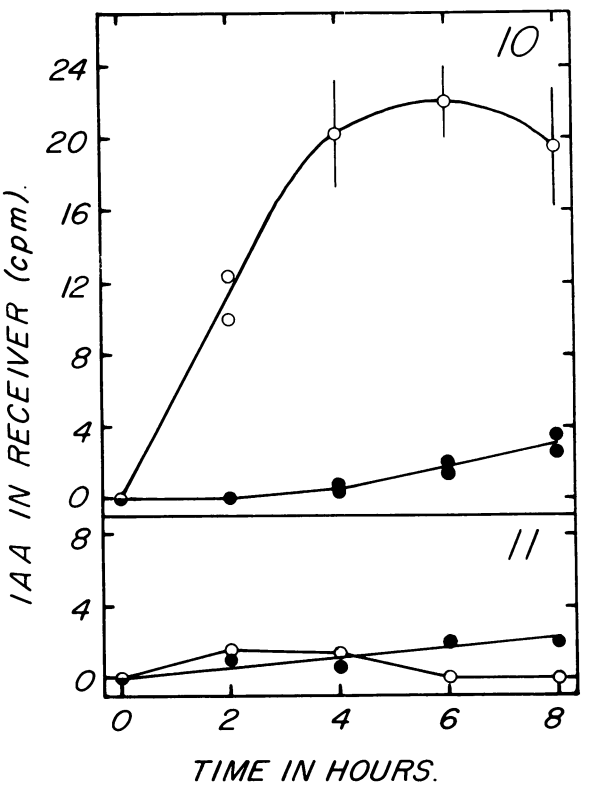

FIG. 10 and 11. Time courses for the basipetal (fig 10) and acropetal (fig 11) movement of labelled IAA into a receiving block through a 5-mm segment of *Avena* coleoptiles in air (—○—○—) and in anaerobic conditions (—●—●—). In figure 10 the vertical lines show twice the standard error in the means, otherwise the points indicate the mean values from 5 coleoptiles in independent experiments. In figure 11 the radioactivity in the receiver never exceeded 2 to 3 cpm and for clarity the mean values for the 2 independent experiments are shown.

tile segments (figs 3 and 4) in which, unlike *Avena*, it was supported by the appearance of significant amounts of IAA in basal receiving blocks under anaerobic conditions.

In air there is more radioactivity in the receiving block than in the basal half of the segment supplied with an apical donor.

Time Course for IAA Movement in Avena *Segments.* The amounts of ^{14}C moving acropetally and basipetally into receiving blocks through 5-mm *Avena* coleoptile segments were determined as a function of time both in air and in N_2. The results are shown in figures 10 and 11.

With apical donors (fig 10) the amount of ^{14}C reaching the receiver in air increased with time for the first 4 hours and then remained relatively uniform at about 20 cpm. In N_2, on the other hand, the amount of ^{14}C in the receiver reached only about 2 cpm after 6 hours and about 3 to 4 cpm after 8 hours. With basal donors (fig 11) the amount of ^{14}C in the receiver never exceeded 2 cpm regardless of the atmospheric conditions. In nitrogen the values are similar to those found when the receiver is at the basal end of the segment, while in air they appeared to be even lower after 6 and 8 hours, a finding which might be accounted for by the activity of the basipetal transport system. The data in the previous section tend to suggest that even under anaerobic conditions basipetal movement of auxin might be slightly greater than acropetal movement. However, even after 8 hours in N_2 the ^{14}C reaching the receiver is low and not significantly different for acropetal and basipetal movement.

The similarity in the values for acropetal and basipetal movement in nitrogen strongly suggests that movement is due principally to diffusion. Polarity of movement of ^{14}C into the receiver blocks is lost completely under anaerobic conditions. The percent reduction of basipetal transport under anaerobic conditions clearly depends upon the duration of the transport period.

Discussion

In discussing the extent to which auxin movement is inhibited by anaerobic conditions it is necessary to specify A) the length of the section, B) the transport period and C) whether it is the movement within the section or into the receiving block that is being considered. Failure to specify these points can lead to confusion and artificial differences between the results of different laboratories.

Movement in Avena *Coleoptiles.* On the basis of the amount of ^{14}C reaching the receiving block, our results show that the basipetal movement of IAA through a 5-mm *Avena* coleoptile segment is totally inhibited by anaerobic conditions during a 2 or 4 hour transport period. In these times no significant amount of ^{14}C reached a receiving block at the apical end of a 5-mm segment supplied with a basal donor either in the presence or absence of O_2.

Our results with 5-mm *Avena* coleoptile segments are in close agreement with those of Goldsmith (5) who used 10-mm segments and found no ^{14}C in the basal receiving block after transport periods of 0.5 to 8 hours under anaerobic conditions. Goldsmith also determined the amounts of ^{14}C in different zones of the coleoptile segment. After 2 hours in anaerobic conditions ^{14}C had reached the second but not the third 1-mm zone, while even after 8 hours all the ^{14}C was confined to the 5-mm half section next to the donor. With our 5-mm segments most of the ^{14}C was confined to the 2.5-mm half section next to the donor after 2 hours in anaerobic conditions, although a little was recovered from the further half section. Not until 6 to 8 hours after the application of donors under anaerobic conditions did we recover any ^{14}C from the receiving blocks at the ends of the 5-mm segments. The time courses for the movement of ^{14}C into apical and basal receivers are similar under anaerobic conditions, thus suggesting that movement is due principally to diffusion. The amount recovered in basal receivers even after 8 hours was only about 12 % of that recovered in air.

We find the total uptake of IAA from an apical donor in 2 hours is reduced by 84 % under anaerobic conditions; Goldsmith's data for a 2-hour transport period under anaerobic conditions revealed uptake to be reduced by 75 to 80 % (5).

Movement Through Zea *Coleoptiles.* Turning now to the results with *Zea* coleoptiles, we find that basipetal movement of IAA into receivers at the end of a 10-mm segment is totally abolished under anaerobic conditions during transport periods of 2 and 4 hours. With 5-mm segments we find that a small but significant amount of ^{14}C reaches a basal receiving block whereas none reaches an apical block after a 2-hour transport period under anaerobic conditions. The latter result suggests that basipetal movement in *Zea* coleoptiles under anaerobic conditions is still slightly greater than acropetal movement. It has already been pointed out in the Results section that this suggestion is supported by the distribution of ^{14}C in the sections of *Zea* and also in *Avena* after a 2-hour transport period, although no ^{14}C emerged into the receiving blocks with the latter tissue.

These findings are in close agreement with Goldsmith's (6,7) recent results obtained with a completely different technique. Goldsmith (6,7) monitored the movement of a pulse of labelled IAA, supplied to the apical end of a 20-mm *Zea* coleoptile segment, under anaerobic and aerobic conditions, and found the rate of movement to be 15-mm $hour^{-1}$ in air and about 2mm $hour^{-1}$ in the absence of O_2. Even under anaerobic conditions the movement was polarized basipetally. Our data with 5-mm and 10-mm segments of *Zea* are con-

sistent with these rates, and in addition, we find with 5-mm segments under anaerobic conditions that the slow movement of IAA is polarized basipetally.

Our data, and those of Goldsmith (6, 7), stand in complete contrast to those recently published by Naqvi, Dedolph, and Gordon (15) who find that under anaerobic conditions the amount of ^{14}C reaching the receiving block at the basal end of a 10-mm *Zea* coleoptile segment is reduced by only 50 % when anaerobic conditions prevail during a 2-hour transport period. They attribute this reduction entirely to the inhibition of uptake of IAA from the donor by 50 % in the absence of O_2 and maintain that the rate of basipetal transport is unaffected. The serious discrepancy between the results of Naqvi et al. (15) on the one hand, and those of Goldsmith (6, 7) and ourselves on the other, can hardly be ascribed to slight differences in technique such as the use by Naqvi et al. (15) of methyl-labelled IAA. The most likely explanation for the discrepancy is that the conditions used by Naqvi et al. (15) were not strictly anaerobic. Although the gas flowing over the tissues was found to have an O_2 concentration of below 0.01 %, it is highly probable that a much higher concentration of O_2 persisted in the cavities of the coleoptiles, since these cavities were closed throughout the flushing period by the receiving blocks at one end of the sections and the "keeper" blocks at the other. No evacuation procedures were used to ensure the removal of residual O_2 from the coleoptile cavity and tissues.

Hertel and Leopold (13) have also reported a smaller reduction in basipetal movement of IAA in *Zea* coleoptile segments under anaerobic conditions than that found either by ourselves or by Goldsmith (5, 6). This discrepancy is again probably due to the fact that Hertel and Leopold (13) did not use evacuation procedures to ensure the removal of O_2 from the tissues.

Relationship Between Auxin Transport and the Geoelectric Effect. There is now convincing evidence that the geoelectric effect is due to the establishment of a lateral concentration gradient of IAA in the shoot tissues (1, 2, 3, 10, 14, 19). This gradient is set up by the lateral polar transport of auxin from the upper to the lower half of the shoot (9, 17). Grahm and Hertz (11) and Woodcock and Wilkins (unpublished) have shown that the geoelectric effect does not occur under anaerobic conditions. Naqvi et al. (15) claim that the lateral transport of IAA during geotropic stimulation occurs as readily under anaerobic conditions as it does in air. They therefore argue that the geoelectric effect cannot be a mandatory consequence of the lateral transport of IAA. Lateral transport of auxin in geotropically stimulated coleoptiles is strictly polarized (9) and can occur against a concentration gradient (9). In these respects it is similar to longitudinal polar transport. Since our data and those of Goldsmith (6, 7) show that longitudinal polar transport of IAA in *Zea* coleoptiles is dependent to a great extent upon metabolic energy it seems highly likely that lateral polar transport in geotropically stimulated coleoptiles would show a similar dependence upon aerobic metabolism.

The strictness of the anaerobic conditions under which Naqvi et al. (15) conducted their experiments is open to serious question because of the divergence between their results for longitudinal transport of IAA and those of both Goldsmith (5, 6, 7) and ourselves. Their data for the dependence of lateral transport of IAA upon aerobic metabolism must therefore be viewed with reservation until independent confirmation is available. The argument of Naqvi et al. (15) that the geoelectric effect cannot be a mandatory consequence of lateral transport of IAA in shoots is, therefore, on the basis of present evidence, somewhat tenuous: it would have been much stronger had they actually determined whether or not the geoelectric effect developed in the coleoptile sections from which their IAA transport data were obtained.

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