Botany. — *On the influence of glucose on auxin production by the root tip of Vicia Faba.* (Preliminary note.) By M. H. VAN RAALTE. (Communicated by Prof. J. C. SCHOUTE.)

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The problem of the occurrence of auxin in root tips has been largely solved by the work of BOYSEN JENSEN (1933). He demonstrated that, whereas root tips which were put on pure agar dit not give off any auxin, a considerable amount diffused into the agar if it contained 10 per cent of glucose. By using this glucose agar he was able to test directly and to confirm CHOLODNY's theory on the geotropism of roots (CHOLODNY 1927).

The present investigations concern the nature of this glucose action on the diffusion of auxin out of the root tip.

At first BOYSEN JENSEN (1933) thought that the root tip did not contain sufficient plastic materials to produce auxin. After he had discovered that mannite had the same influence as glucose, he changed his opinion: "Weil Mannit von der Wurzelspitze wohl kaum verarbeitet wird, darf man wohl annehmen, dasz die Bedeutung der Dextrose beim Abfangen des Wuchsstoffes physikalischer Natur ist; vielleicht ist der Kontakt zwischen Wurzelspitze und Agar besser, wenn in dem letzteren ein wasseranziehender Stoff, wie Dextrose oder Mannit, vorhanden ist." (1933, p. 348).

THIMANN (1934, p. 33) concludes from his experiments that "growth substance is not produced in the root tip, but merely accumulates there by the polarity of its transport". According to him, the action of the glucose is a purely osmotic one, the auxin being drawn backward in the direction opposite to the polarity in the root.

CHOLODNY (1934) states that root tips to which a little gelatine, containing glucose and diluted KNOP solution, has been applied still go on giving off auxin 5—6 hours after they have been cut, whereas the auxin production of tips without that gelatine has ceased by that time. From this fact he concludes that the glucose acts as a food substrate.

In the present paper will be shown that glucose can influence the amount of auxin diffusing out osmotically as well as by increasing the auxin concentration in the root tip.

1. *Influence of different glucose concentrations.*

A 3 per cent agar was mixed up with different amounts of glucose and cut into discs of the ordinary size ($8 \times 6 \times 0.9$ mm). On each disc 10 root tips of Vicia Faba were placed, which remained there for two hours.

Subsequently the tips were removed, the agar discs cut into 12 equal blocks, which were put on Avena coleoptiles for analysis. The results of four different experiments are shown in table 1.

TABLE 1.

Influence of different glucose concentrations on the auxin output of root tips. Length of tips: 3 mm. Each figure represents the mean curvature of 12 Avena plants.

Glucose concentration	Expt. 1	Expt. 2	Expt. 3	Expt. 4
0	1°	2°	0°	1°
1 per cent	3°	3°	5°	6°
3 per cent	6°	7°	8°	10°
5 per cent	7°	9°	11°	—
10 per cent	9°	13°	14°	14°
15 per cent	6°	8°	10°	13°

It is shown in table 1 that 3 per cent glucose already has a distinct influence on the amount of auxin yielded by the tips. Ten per cent proves to be the optimal concentration, whereas the curvature caused by the blocks containing 15 per cent of glucose is decidedly less. This latter phenomenon is probably due to the osmotic attraction of water out of the coleoptile by the high glucose concentration, since in a few blank experiments with agar containing 15 % of glucose without auxin positive curvatures were obtained. These experiments do not give any evidence in favour of CHOLODNY's, neither of THIMANN's opinion.

2. *Osmotic influence.*

If THIMANN's opinion that glucose attracts the auxin osmotically is right, it must be possible to obtain the same effect with inorganic salts.

Experiments were carried out with different salts, added to 3 % agar in such amounts that the concentrations were isotonic with 0.1, 1, 3, 5, or 10 per cent glucose. Tables 2 and 3 give some of the results.

Tables 2 and 3 show that more auxin diffuses into agar containing $BaCl_2$ or KCl than into pure agar. The experiments show rather large differences, according to the sensitivity of the Avena test plants changing daily (variations of the "Standart"; see KÖGL and HAAGEN SMIT). These variations, however, do not interfere with our results, as only the figures of each separate experiment (vertical row) have to be compared. These figures give clear evidence of the osmotic attraction effected by the salts upon the auxin in the tip. Similar results were obtained with agar containing a mixture of $Ca(NO_3)_2$ and KNO_3 in different concentrations.

TABLE 2.

Influence of KCl. 7 Root tips of Vicia were placed on agar. After 2½ hours the tips were removed and the agar was analysed. Length of the tips: 3 mm. Each figure represents the mean curvature of 12 Avena plants.

Concentration in agar	Expt. 1	Expt. 2
KCl isotonic with 0.1% glucose	0°	2°
KCl isotonic with 1 % glucose	5°	1°
KCl isotonic with 3 % glucose	8°	9°
KCl isotonic with 5 % glucose	12°	—
KCl isotonic with 10 % glucose	7°	7°
1% glucose	5°	5°
5% glucose	16°	10°

TABLE 3.

Influence of $BaCl_2$. 7 Root tips of Vicia remained on agar during 2 hours or 2½ hours. Length of the tips: 3 mm. Each figure represents the mean curvature of 12 Avena plants.

Concentration in agar	Expt. 1	Expt. 2	Expt. 3	Expt. 4
$BaCl_2$ isot. with 1% glucose	—	9°	6°	9°
$BaCl_2$ isot. with 2% glucose	3°	10°	11°	—
$BaCl_2$ isot. with 3% glucose	5°	15°	0°	5°
$BaCl_2$ isot. with 5% glucose	11°	5°	0°	6°
$BaCl_2$ isot. with 7% glucose	5°	11°	2°	3°
$BaCl_2$ isot. with 9% glucose	6°	9°	—	5°
Pure agar	2°	6°	1°	1°

3. *Glucose increases the auxin concentration in the root tip.*

Although the above-described experiments seem to explain the glucose effect satisfactorily, there were two phenomena which gave rise to the idea that this explanation could not be exhaustive. Firstly, in some experiments, in which the influence of salt and glucose were compared, the latter substance proved to have a greater effect than salt agar of an isotonic concentration. The second phenomenon can be seen from table 4. Here the amounts of auxin are compared, which diffuse out of root tips remaining on glucose agar during the first and the second hour after they have been cut off.

TABLE 4.

Tips of Vicia roots were cut off. On each agar disc 12 tips were put. These were removed after one hour and placed on a new agar disc, containing the same glucose concentration as the first. After another hour the auxin content of the first and the second agar disc were compared. Each figure represents the mean curvature of 12 Avena plants.

Concentration of glucose in the agar	Auxin diffusing into agar during:	
	first hour	second hour
0	2°	1°
1%	2°	4°
3%	4°	6°
5%	3°	8°
10%	2°	8°
15%	1°	6°

The amount of auxin obtained during the second hour proves to be considerably higher. This, however, could hardly be the case if the glucose acted only by osmotic attraction. This action would start immediately after the tips had come into contact with the agar, and one would expect the auxin concentration in the tip to decrease as the auxin was diffusing out. Then, however, the amount of auxin in the second block could at most be equal, but by no means more than in the first.

In order to decide whether or not the auxin concentration in the tips decreased during the time they remained on agar, ether extractions were made. The technique is similar to that described by KÖGL and HAAGEN SMIT (1931) and by THIMANN (1934).

In short the procedure is as follows: root tips were put on pure agar or on agar containing 10 % of glucose. After a few hours the tips were removed. Different numbers were immersed in peroxide-free ether, acidified with HCl and thoroughly ground together with sand. The ether was separated off and evaporated on a water bath. The residue was dissolved in 0.3 cc of water. Two agar discs were immersed in this solution and remained there sufficiently long to allow the auxin to diffuse through equally. Then the auxin content of the discs was tested with Avena plants. Table 5 shows the results. From column 3 can be seen that of the tips which had to be compared not the same number was extracted. The reason of this lies in the restricted range of auxin concentrations which can be analysed with Avena plants. If the concentration is too high, an increase of it does not give a corresponding increase of the curvature (limit angle of F. W. WENT). In order to avoid this complication, in each experiment different numbers of root tips were extracted, the extracts obtained, in this way always giving at least one result below the limit angle. The latter is

given in column 4. The figures in the fifth column are derived from those in columns 3 and 4. They represent the ratio between the auxin content of tips from glucose agar and from pure agar, calculated for equal numbers of tips.

TABLE 5.

Extraction with ether of root tips, which had remained on agar containing 10 % of glucose or on pure agar. Length of the tips: 5 mm.

Expt.	Tips from	Number of tips extracted	Mean curv. of test plants	Ratio
1	glucose agar	10	5.7	
	pure agar	50	6.9	4.1
2	glucose agar	15	6.4	
	pure agar	25	3.5	3.0
3	glucose agar	30	10.8	
	pure agar	50	8.0	2.3
4	glucose agar	30	4.0	
	pure agar	75	2.7	3.7
5	glucose agar	15	3.2	
	pure agar	75	8.1	2.0
6	glucose agar	15	2.7	
	pure agar	50	1.9	4.5

Column 5 clearly shows that in the tips from glucose agar the auxin content is much higher. This fact makes it very probable that in the root tips of Vicia Faba the auxin does not merely accumulate, as was accepted by THIMANN, but that new auxin may be actually produced.

Whether this also occurs in salt agar, is still to be investigated. The experiments are still being continued; a more detailed report will be published later on.

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