Cell Division And Elongation In The Decapitated Root Of *Vicia Faba* L.

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CELL DIVISION AND ELONGATION IN THE DECAPITATED ROOT OF *VICIA FABA* L.

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I. INTRODUCTION

In my previous papers I have given some accounts of the investigations on the relation between cell division and elongation (KOJIMA, 1928, 1931); and the present study, being done on the same subject, is the continuation of them.

The opinion has been entertained thoughtlessly by some that the differentiation of tissue in the root is accelerated, when the growth of the root is inhibited by embedding in gypsum; but in my previous investigation I have ascertained that the progress with regard to speed of cell differentiation in the root embedded in gypsum is also delayed by its pressure; and also I stated that the elongation and the division of cells are hindered at the same time by the pressure, mechanical or osmotic.

On the other hand, as in the case of the decapitated root, if the further elongation of the root tip is inhibited to some extent, by cutting off its tip, and the further growth, in length or thickness, of the root body is never hindered artificially, then in what relation do the two processes, namely the cell elongation and cell division, pass along? This question is the subject of my study described in this paper.

This investigation was made under the direction of Professor Dr. R. KÔKETSU; I wish to take this opportunity to express my very great indebtedness to him, for his guidance and interest. My cordial gratitude is due to Em. Professor Dr. K. FUJII, for his kind advice and unceasing encouragement.

¹ Contributions from the Botanical Laboratory, Kyushu Imperial University No. 47.

II. MATERIALS AND METHODS

As the material, young roots of seedling of *Vicia Faba* were used; the experiments were carried on in indoor temperature during the warm season, and in the green house during the winter season, so that the plant might grow in normal or nearly normal conditions. The seeds used in every experiment were nearly uniform in size and weight, and the seeds were got from the same habitat, though it can not be said that all of them were the same line.

Usually the seeds were soaked in water for about 24 hours and after the seed coat had swelled satisfactorily, they were sowed in a box filled with moist saw dust. When the root of seedlings grew up to suitable length, seedlings, with roots of nearly uniform length and size, were chosen out of many, and used as materials.

To decapitate the root tip a very sharp razor was used and the tip was cut off not in the air but in water.

In usual cases the decapitated roots were brought back to the box of saw dust and grown; namely the seedlings with decapitated roots were embedded carefully in wet saw dust, so that the roots were planted vertically and cotyledons were covered with the saw dust. In cases of culture in water, a wire-netting coated with paraffin, was used, on which the cotyledons were placed, and through the meshes the roots hung down vertically. The wire-netting was placed so as only the roots were dipt in water, and the cotyledons were held in the air.

In order to measure the elongation of the roots, the length was measured not as to the whole root length, but certain definite portions of the root which were determined at the beginning of the experiment. For this purpose black lines of indian ink were marked on the surface of the root respectively at the points 4 mm, 5 mm and 10 mm from the root apex (in *Vicia Faba* the boundary between the root cap and root proper is not remarkable, so I measured the distance from the root apex) and after certain pre-arranged time the distances between the marks were measured as exactly as possible. •

In abbreviation of the description I will use letters A, B and C hereafter: A means the portion between the root apex and the mark of 4 mm from the apex; B, the portion between the marks of 4 mm and 5 mm from the root apex; and C, the portion between the marks of 5 mm and 10 mm from the root apex.

Materials for microscopical observations were fixed in FLEMMING'S weaker solution²; paraffin sections of 10μ or 15μ , being parallel to the long axis of the root, were prepared and stained with HEIDENHAIN'S iron haematoxylin. The sections passing as near as possible to the central axis of the root were observed. ZEISS' microscope AA or DD $\times K 8$, with oc. micrometer $\frac{100}{100}$ mm was used.

It has been already mentioned that in the root tip of *Vicia Faba* mitotic figures are seen most abundantly in the portion within 2 mm from the apex (cf. PEKAREK, 1927), and the frequentness of the mitotic divisions is limited in the root tip within 4 mm from the apex (cf. SCHÜEPP, 1923); on the other hand the zone, where the elongation is most rapid, is not near the growing point but several millimeters apart from it basipetally (cf. PFEFFER'S Handbook, Vol. 2 p. 8).

For that reason, I studied the cell division chiefly on the portion A, namely the part within 4 mm from the root apex, and the elongation of the cell, on the basipetal portion from that point. Of course the cell division and cell elongation may take place on the same tissue in succession; it might be, therefore, most reliable to observe and compare these two phenomena on every part of the root having ability of growth, but it results naturally in the enlargement of the work; in order to simplify the work and so to realize the desired object I took the method above mentioned.

The cell length was measured on 4 or 5 sections of each root in the region of the cortex, near the epidermis—at the several portions corresponding to the points 4, 5 and 10 mm from the root apex at the time of decapitation. The length of the cell of each portion of a root was expressed by an average of length of more than 50 cells for each root; the number of length presented in this paper is the average of the lengths of several roots thus obtained, and it is expressed by observed value with micrometer scale; so, if necessary, we can get the real length in μ by multiplying this by $\frac{100}{69}$.

The frequency of cell division was observed on 10 sections (in some cases 5 sections) of each root³. The number of mitotic figures seen in the field—the belt-shaped field, across the long axis of the

² 1% chromic acid 25 cc + 1% glacial acetic acid 10 cc + 1% osmic acid 10 cc + water 55 cc.

³ The thickness of the section for this purpose was always 10 μ .

root, $\frac{25}{136}$ (mm $\div$ 184 μ) in width and having a length of diameter of the root section at that portion—was counted. The rate of frequency of cell division is expressed by the quotient, dividing the total number of mitotic figures thus found at similar given portions on several sections of a root, by the total amount of the area (calculated in qmm) of the fields examined; namely the rate of frequency seen in the tables in this paper is the number of mitotic figures found in 1 qmm of the field.

In connection with the growth of cell or tissue of the root the water content in the tissue was also considered. The root used for this purpose was previously cleared away from the saw dust and the moisture was blotted off from its surface. The portion, between the two marks made at 4 and 10 mm from the root apex at the beginning of the experiment, was cut off, then the fresh weight of that portion was measured; afterwards it was dried in an electric oven of about 100°C for about 24 hours, cooled in an exicator containing CaCl_2 for 1-2 hours, then the dry weight was weighed.

III. OBSERVATIONS ON THE ELONGATION OF DECAPITATED ROOT

a) Experiments in saw dust: The roots, respectively decapitated at 1, 2, 3 and 4 mm from the root apex, and intact roots were embedded in wet saw dust and grown for about 24 hours. Then the length of each portion respectively A, B and C, was measured, and the amount of the elongation was found. The results thus found are given in Tables 1 and 2, in which we can see that the elongation of the root, of which a large part of the root tip has been cut off, is generally speaking less than that of the root intact or the root of which a small part of the root tip has been cut off⁴. And comparing the intact root and the 1 mm decapitated root, the latter shows larger elongation than the former, in case of regions B and C, though there are some exceptional cases; and in A region, the elongation of the 1 mm decapitated root is certainly less. With regard to the total length of root, namely $A + B + C$ (basipetal the region C, there is

⁴ FIRTSCH, 1884, stated that, regarding the total length of root, the elongation of the root decapitated more than 2 mm from the root apex is rather pathologically hindered; and BÜNNING, 1928, found that roots decapitated more than 1.5 mm from the root apex show less elongation than the intact roots.

Table 1. Elongation of the roots (in saw dust) about 24 hours after decapitation

Experiment	Time between the operation and the observation (in hours)	Portion of which the elongation is observed ¹	Roots decapitated at the several points respectively from the root apex:					
			Intact roots					
			Observed value (in mm)	Relative value	1 mm	2 mm	3 mm	4 mm
I	24	A	26.34(8) ²	100	86.0(10)	49.5 (6)	8.8 (9)	—
		B	4.87 (8)	100	91.0(10)	76.2 (6)	61.8 (9)	13.6 (9)
		C	5.30(10)	100	126.4(10)	84.3 (9)	79.8 (9)	44.3 (9)
II	24½	A	23.27 (7)	100	91.1 (7)	60.2 (5)	13.3 (7)	—
		B	3.34 (7)	100	126.3 (7)	93.4 (5)	66.5 (7)	17.7(10)
		C	3.65 (7)	100	120.8 (7)	87.8 (5)	80.3 (6)	48.8(10)
III	22½-23	A	20.22(10)	100	76.6 (9)	59.8(11)	11.6(11)	—
		B	2.10(10)	100	153.8 (8)	125.7(11)	68.6(11)	16.7(12)
		C	2.30(10)	100	145.2(11)	124.3(12)	97.4(11)	57.0(12)
IV	24½-25	A	26.32(11)	100	89.0(14)	49.0(14)	7.4(14)	—
		B	3.75(10)	100	101.3(13)	73.3(14)	52.0(14)	9.9(14)
		C	4.16(13)	100	90.4(14)	80.5(14)	63.5(14)	32.9(14)
V	23½	A	26.88 (8)	100	85.8 (8)	39.5 (8)	4.6 (3)	—
		B	3.80 (8)	100	91.1 (8)	73.2 (8)	47.9 (7)	9.5 (8)
		C	4.52 (8)	100	111.3 (8)	70.4 (3)	53.8 (8)	38.3 (8)
Average		A	24.61	100	85.7	51.6	9.1	—
		B	3.57	100	112.7	88.4	59.4	13.5
		C	3.99	100	118.8	89.5	76.1	44.3

¹ A is the portion from the root apex or cut surface to the point 4 mm from the apex; B is the portion between the two points respectively 4 and 5 mm from the root apex; and C is the portion between the two points respectively 5 and 10 mm from the root apex.

² The number of the roots observed is given in brackets.

no elongation practically), the elongation of the 1 mm decapitated root is also less than that of the intact root⁵. The fact that the elongation of regions B and C is somewhat accelerated in the 1 mm decapitated root is possibly related to the irritation or hormone action caused by cutting the root tip, yet this is beside the question here.

Table 2. Elongation of the roots (in saw dust) 24 hours after decapitation

Experiment	Time between the operation and the observation (in hours)	Portion of which the elongation is observed	Intact roots		Roots decapitated at the several points respectively from the root apex :	
			Observed value (in mm)	Relative value	1 mm	4 mm
VI	24	A	2.65 (8)	100	85.7 (8)	—
		B	2.20 (8)	100	107.3 (8)	15.0 (8)
		C	6.42 (8)	100	103.3 (8)	40.7 (8)
VII	24	A	5.95 (4)	100	75.6 (4)	—
		B	3.57 (4)	100	93.8 (4)	9.8 (4)
		C	6.80 (4)	100	113.2 (4)	26.0 (4)
VIII	24	A	7.10 (14)	100	88.7 (14)	—
		B	4.12 (14)	100	103.3 (14)	31.3 (14)
		C	5.84 (14)	100	118.3 (14)	70.4 (14)
IX	24	A	18.62 (11)	100	71.4 (10)	—
		B	3.63 (11)	100	109.1 (10)	31.1 (12)
		C	4.81 (11)	100	104.6 (10)	58.8 (12)
Average		A	8.58	100	80.4	—
		B	3.38	100	104.6	21.8
		C	5.97	100	109.9	49.0

⁵ FIRTSCH, 1884, observed that roots decapitated at a point within 2 mm from the root apex elongate to same extent or often a little less than the intact roots; MOLISCH, 1883, stated that the 1 mm decapitated root elongates always less than the intact root; WIESNER, 1881, reported that the root, of which the growing point has been cut off, elongates slower than the intact root.

The above mentioned facts are the cases 24 hours after the decapitation, yet the fact that roots of which the greater portion of the tip has been decapitated elongate slower than the 1 mm decapitated roots or intact roots, is also seen in the material 7 hours after the decapitation (Table 3).

Table 3. Elongation of the roots (in saw dust) about 7 hours after decapitation

Experiment	Time between the operation and the observation (in hours)	Portion of which the elongation is observed	Intact roots		Roots decapitated at the several points respectively from the root apex:			
			Observed value (in mm)	Relative value	1 mm	2 mm	3 mm	4 mm
X	7½	A	1.57 (7)	100	100.0 (9)	57.3(7)	3.9 (9)	—
		B	1.03 (8)	100	93.1 (9)	95.4(7)	59.3 (9)	13.5 (9)
		C	3.40(10)	100	123.8(10)	93.8(9)	76.5 (9)	38.5 (9)
XI	7	A	0.41(13)	100	97.6(12)	—	2.4(14)	—
		B	0.50(13)	100	96.0(12)	—	34.0(14)	3.0(13)
		C	1.65(13)	100	93.9(12)	—	53.9(14)	45.5(13)
XII	7	A	0.94 (9)	100	79.8(10)	—	4.3(10)	—
		B	0.84 (9)	100	91.7(10)	—	42.9(10)	7.1(10)
		C	2.91 (9)	100	92.1(10)	—	58.1(10)	28.9(10)
XIII	7	A	1.75(12)	100	73.7(12)	—	10.3(12)	—
		B	1.24(12)	100	80.6(12)	—	42.7(12)	10.5(12)
		C	2.37(12)	100	102.8(12)	—	73.5(12)	39.0(12)
XIV	7	A	0.80(12)	100	93.3(12)	—	—	—
		B	0.53(12)	100	115.1(12)	—	—	33.9(12)
		C	2.54(12)	100	79.1(12)	—	—	43.4(12)
Average		A	1.09	100	90.0	—	6.5	—
		B	0.84	100	96.3	—	44.7	15.6
		C	2.67	100	93.3	—	65.5	40.1

But the difference between the elongation of the intact root and the 1 mm decapitated root is not always alike in C region, in B region

the intact root elongates rather more rapidly; and in A region, of course, intact root elongates more rapidly.

Now we consider the experiments with decapitated roots observed 4 hours after the operation; we can see also in this case that the roots of which the larger portion of the tip has been cut off stretch slower than the intact roots or the 1 mm decapitated root. There is not a decided difference between the elongation of the intact root and that of the 1 mm decapitated root, yet, speaking in general, the elongation of the intact root is likely to be larger than that of others (Table 4).

Table 4. Elongation of the roots (in saw dust) 4 hours after decapitation

Experiment	Time between the operation and the observation (in hours)	Portion of which the elongation is observed	Intact roots		Roots decapitated at the several points respectively from the root apex:		
			Observed value (in mm)	Relative value	1 mm	3 mm	4 mm
XV	4	A	0.23 (11)	100	23.6 (13)	10.7 (14)	—
		B	0.25 (12)	100	34.0 (13)	28.0 (14)	12.0 (16)
		C	1.37 (12)	100	38.3 (13)	56.2 (14)	35.8 (16)
XVI	4	A	0.50 (10)	100	70.0 (10)	2.0 (10)	—
		B	0.22 (10)	100	127.3 (10)	27.3 (10)	27.3 (10)
		C	1.09 (10)	100	99.1 (10)	58.7 (10)	34.9 (10)
XVII	4	A	0.45 (14)	100	95.6 (16)	6.7 (16)	—
		B	0.28 (14)	100	100.0 (16)	39.3 (16)	17.9 (14)
		C	1.50 (14)	100	104.7 (16)	40.7 (16)	36.7 (14)
XVIII	4	A	0.39 (11)	100	110.3 (9)	10.3 (11)	—
		B	0.40 (11)	100	80.0 (9)	52.5 (11)	20.0 (11)
		C	1.60 (11)	100	100.0 (11)	60.6 (11)	37.5 (11)
Average		A	0.41	100	76.1	7.4	—
		B	0.29	100	97.3	36.8	19.3
		C	1.39	100	98.0	54.1	36.2

b) Experiment in water: The similar experiments as above mentioned have been done, not in saw dust but in water. The roots,

decapitated as well as intact, are grown in water for about 24 hours (Table 5); under these several experiments the results of observation can not be said to be exactly parallel, as seen in the cases in saw dust. Concerning C region, sometimes the 4 mm decapitated roots show less elongation than the intact roots, but in other cases, on the contrary; yet generally speaking there is likely to be no clear difference between them (the average elongation of the intact root : that of the 4 mm decapitated root = 100:104). Regarding C region, in

Table 5. Elongation of the roots (in water) 24 hours after decapitation

Experiment	Time between the operation and the observation (in hours)	Portion of which the elongation is observed	Intact roots		Roots decapitated at the several points respectively from the root apex :		
			Observed value (in mm)	Relative value	1 mm	3 mm	4 mm
XIX	24	A	12.36 (19)	100	120.1 (22)	32.0 (21)	—
		B	3.39 (19)	100	117.7 (22)	105.3 (21)	64.9 (20)
		C	4.10 (20)	100	110.7 (20)	104.2 (20)	103.5 (20)
XX	24	A	7.24 (9)	100	102.5 (9)	37.6 (9)	—
		B	4.08 (9)	100	82.8 (9)	68.1 (9)	62.7 (16)
		C	4.36 (14)	100	133.0 (15)	116.7 (14)	141.3 (16)
XXI	24	A	4.01 (16)	100	133.2 (18)	96.3 (14)	—
		B	2.66 (16)	100	107.9 (18)	103.8 (14)	67.3 (16)
		C	3.86 (16)	100	102.6 (18)	117.9 (15)	92.2 (16)
XXII	24	A	8.99 (14)	100	112.8 (14)	—	—
		B	3.91 (14)	100	117.1 (14)	—	63.2 (15)
		C	5.85 (15)	100	109.6 (14)	—	81.2 (15)
XXIII	24	A	10.82 (13)	100	110.7 (12)	—	—
		B	3.70 (13)	100	86.5 (12)	—	78.4 (11)
		C	5.61 (13)	100	75.8 (12)	—	96.4 (11)
Average		A	8.63	100	115.9	55.3	—
		B	3.55	100	102.4	92.4	67.3
		C	4.76	100	106.3	112.9	103.9

general, the 3 mm decapitated roots elongate rather more rapidly than the intact roots. And comparing the 1 mm decapitated root with the intact root there are not such definite differences between their elongations as regards C region and B region, but within A region the 1 mm decapitated root shows larger elongation. Hence as to the sum of these 3 regions (A + B + C) the 1 mm decapitated roots elongate more rapidly⁶.

Table 6. Elongation of the roots (in water) 7 hours after decapitation

Experiment	Time between the operation and the observation (in hours)	Portion of which the elongation is observed	Intact roots		Roots decapitated at the several points respectively from the root apex :			
			Observed value (in mm)	Relative value	1 mm	2 mm	3 mm	4 mm
XXIV	7	A	0.67(14)	100	143.3(14)	—	70.1(14)	—
		B	0.53(14)	100	147.2(14)	—	154.7(14)	105.7(15)
		C	2.80(14)	100	103.9(14)	—	130.4(14)	120.7(15)
XXV	7	A	0.70(12)	100	101.4(12)	—	—	—
		B	0.43(12)	100	109.3(12)	—	—	90.7(13)
		C	2.16(12)	100	104.6(12)	—	—	106.5(13)
XXVI	7	A	0.43(12)	100	146.5(13)	114.0(12)	—	—
		B	0.43(12)	100	86.0(13)	100.0(12)	—	86.0(12)
		C	1.90(12)	100	106.8(13)	93.7(12)	—	95.3(12)
Average		A	0.60	100	130.4	—	—	—
		B	0.46	100	114.2	—	—	94.1
		C	2.29	100	105.1	—	—	107.5

The experiments, in water for about 7 hours, show that, in general, the 4 mm decapitated root has a little more rapid elongation than the intact root, in C region; and the former elongates in less degree than the intact root as regards B region (Table 6). The 1 mm decapitated root elongates more rapidly than the intact root, in general, in every region.

⁶ This fact coincides with the results of WIESNER (1934), who stated that the 1 mm decapitated root cultured in water elongates far more rapidly than the intact root by maize root.

Roots cultured in water for 4 hours were also examined. As a whole, the rate of elongation of the 3 mm decapitated roots as well as the 4 mm decapitated roots in C region is larger; and as to B region, with some exceptions, it is also larger. The elongation of the 1 mm decapitated root is always larger than the intact root as regards the total length of the root (Table 7).

Table 7. Elongation of the roots (in water) 4 hours after decapitation

Experiment	Time between the operation and the observation (in hours)	Portion of which the elongation is observed	Intact roots		Roots decapitated at the several points respectively from the root apex:			
			Observed value (in mm)	Relative value	1 mm	2 mm	3 mm	4 mm
XXVII	4	A	0.42(12)	100	142.9(13)	—	69.0(13)	—
		B	0.25(12)	100	120.0(13)	—	133.2(13)	124.0(12)
		C	10.7(11)	100	117.8(13)	—	112.1(12)	119.6(12)
XXVIII	4	A	0.30(14)	100	140.0(14)	—	50.0(14)	—
		B	0.24(14)	100	104.2(14)	—	137.5(14)	87.5(14)
		C	1.30(14)	100	117.7(14)	—	108.5(14)	109.2(14)
XXIX	4	A	0.14(13)	100	207.1(13)	—	142.9(13)	—
		B	0.18(13)	100	122.2(13)	—	150.0(13)	127.8(13)
		C	1.17(13)	100	111.1(13)	—	118.8(13)	111.1(13)
XXX	4	A	0.53(12)	100	120.8(13)	126.4(13)	—	—
		B	0.35(12)	100	128.6(13)	122.9(13)	—	102.9(12)
		C	1.73(12)	100	100.6(13)	97.1(13)	—	102.9(12)
Average		A	0.35	100	152.7	—	87.3	—
		B	0.26	100	118.7	—	140.2	110.6
		C	1.32	100	111.8	—	113.1	110.7

c) Comparing the above mentioned two series of experiments, we can find that in the series in saw dust, the results of each experiment are almost parallel, namely, it may be said that the more largely decapitated roots, either in A, B or C, elongate less; while in the cases of series in water the results of individual experiments are

variable, and in these cases, for example, the 3 mm and 4 mm decapitated roots 4 hours after the cutting, in regions B and C, the 4 mm decapitated root 7 hours after the decapitation, in C region, the 3 mm decapitated root 24 hours after the decapitation, in C region, etc., the elongation of the decapitated roots is rather greater. And the 1 mm decapitated root cultured in saw dust shows greater elongation than that of the intact root in regions B and C, and in A region the former is less in the rate of elongation, while the 1 mm decapitated root cultured in water always elongates to a larger extent (either in A, B or C).

In the decapitated surface the cells being cut are exposed, so the conditions of the tissues near the cut are quite contrary to those of the tissues in the normal root. The influence of the outside conditions upon the water content of the tissue of the root is, therefore, quite different in this case. As a matter of fact, the cut surface of the root cultured in saw dust is adjoined to moisture or grains of saw dust, as the case may be; and in other and more common cases, the cut surface is exposed to the air with moisture, kept at saturation point, contained in the space among the grains of saw dust. On the other hand, in the cases of the roots, cultured in water, it goes without saying that the cut surfaces are always covered with water. It must be possible to attribute the majority of these discrepancies between the results, obtained in the experiments in saw dust and in water, to the influence of water. The fact that the decapitated root in water showed greater elongation may be considered as the results of superfluous elongation caused by the absorption of excess of water through the cut surface. On this account, we have seen that the regions B and C the decapitated root, in water, elongated rapidly within 4 hours after cutting, but decreased its rate with progress of time, namely 7 hours and 24 hours after decapitation; and it perhaps reveals to us the truth that the rapidity of elongation within 4 hours is not the growth, in real sense, but only the symptom of growth disturbed by the superfluous elongation. In the same manner, we may think that the rapid elongation in A region of the 1 mm decapitated root is due to the excess of water.

Of the two methods of experiment above mentioned, the one has a defect in the loss of water through the cut surface, and the other, a superfluous elongation by excess of water; both of them have advantages and disadvantages, yet we can see from these experiments, the state of elongation of the decapitated roots even though it is not

so exact. Consequently, the relation between the elongation of the root and that of the cell can be seen by either of the two methods of experiment; and in order to observe this point of question I used the materials cultured in saw dust.

IV. OBSERVATION OF CELLS IN THE GROWING ZONE OF THE DECAPITATED ROOT

As I have already mentioned in § 2 (on the methods), the observation on the elongation of individual cells was made on the basipetal portions from the point 4 mm from the root apex.

Selecting several roots out of the materials of Exp. II, Table I (i.e. roots 24½ hours after the decapitation), that had elongated at a nearly average rate of elongation in that experiment of growth, and fixing and preparing them for the microscopic use, I measured the length of the cells (Table 8, Series 1). And it was found that the 1 mm decapitated root and the intact root had nearly the same length of cell; the length of cell of the 3 mm decapitated root at the portions corresponding to the points 4 mm and 5 mm from the root apex at the time of the operation, is a little shorter; and the length of cell of the 4 mm decapitated root, in the portion corresponding to the point 5 mm from the root apex at the time of the operation, is very short, nearly the half of that in the roots, being decapitated at 1 mm or intact, at the corresponding portion.

Table 8. Length of cell in the roots (in saw dust) about 24 hours after decapitation

Series 1.

	Intact roots		Roots decapitated at the several points respectively from the root apex:		
	Observed value ¹	Relative value	1 mm	3 mm	4 mm
No. of roots observed	7		5	5	6
Cell lengths, measured 24½ hours after the decapitation, at the points corresponding to the points apart from the apex of root at the time of the operation, respectively:	4 mm 14.32(707) ²	100	101.5(323)	68.4 (649)	—
	5 mm 14.19(634)	100	101.8(353)	75.9 (666)	56.2 (464)
	10 mm 14.73(634)	100	98.1(349)	91.1 (535)	98.5 (367)

Series 2.

		Intact roots		Roots decapitated at the several points respectively from the root apex :		
		Observed value	Relative value	1 mm	3 mm	4 mm
No. of roots observed		5		6	7	6
Cell lengths, measured 22½-23 hours after the decapitation, at the points corresponding to the points apart from the apex of root at the time of the operation, respectively :	4 mm	14.87(309)	100	94.4 (361)	57.2 (440)	—
	5 mm	13.37(309)	100	96.5 (352)	76.4 (416)	50.5 (367)
	10 mm	14.37(303)	100	95.8 (361)	93.0 (410)	86.0 (362)

1 The unit of cell length is $\frac{100}{69} \mu$

2 No. of cells observed is given in brackets.

The results obtained by measuring on the cell length in several roots, selected out of the seedlings of Exp. III, Table 1 (i.e. materials about 23 hours after the decapitation), having the rate of elongation nearly equal to the average elongation, are nearly identical to those above mentioned (Table 8, Series 2).

In the same manner I studied the materials (out of the roots of Exp. XII, Table 3) 7 hours after decapitation. The length of cells in the 3 mm decapitated root or the 4 mm decapitated root is smaller to some degree than that of the intact root or the 1 mm decapitated root (Table 9), in this respect the result is identical to that of experiments 24 hours after the decapitation.

Table 9. Length of cell in the roots (in saw dust) 7 hours after the decapitation

		Intact roots		Roots decapitated at the several points respectively from the root apex :		
		Observed value	Relative value	1 mm	3 mm	4 mm
No. of roots observed		4		5	5	5
Cell lengths, measured 7 hours after the decapitation, at the points corresponding to the points apart from the apex of root at the time of operation, respectively :	4 mm	3.14 (206)	100	112.7(267)	77.4 (280)	76.4 (255)
	5 mm	4.61 (197)	100	120.2(243)	81.9 (285)	84.2 (277)
	10 mm	11.29 (196)	100	102.2(245)	89.4 (240)	94.3 (254)

In the materials 4 hours after the decapitation (out of the material of Exp. XV, Table 4), there is not such a remarkable difference between the cells of the different kind of roots (Table 10) (except in the smallness of cell length of the 3 mm decapitated root, at 5 mm point) as seen in the materials of the experiments 24 hours and 7 hours after the decapitation. This is explained by the reason that as the time of elongation is too short, the differences of the length are not remarkable enough to be measured.

Table 10. Length of cell in the roots (in saw dust) 4 hours after the decapitation

	Intacts roots		Roots decapitated at the several points respectively from the root apex :		
	Observed value	Relative value	1 mm	3 mm	4 mm
No. of roots observed	5		5	5	5
Cell lengths, measured 4 hours after the decapitation, at the points corresponding to the points apart from the apex of root at the time of the operation, respectively :	4 mm 1.70 (250)	100	110.0 (272)	96.5 (250)	103.3 (248)
	5 mm 3.32 (256)	100	102.4 (273)	69.6 (252)	90.4 (250)
	10 mm 9.73 (255)	100	97.6 (246)	92.3 (249)	63.5 (249)

From the observations on the cell length, taking into consideration the results of experiments in saw dust as well as in water, we find that the difference between the cell length of the different kind of roots is parallel to the difference in the elongation of the roots.

V. OBSERVATIONS OF CELL DIVISION IN THE ROOT TIP OF THE DECAPITATED ROOT

Observation of cell division was made mainly in the portion within 4 mm from the root apex as above mentioned. Measuring the frequency of cell division in the root, 4 hours after the decapitation (material of Exp. XV, Table 4, selected by their rate of elongation, namely the roots elongated in a rate nearly equal to that of average elongation of the whole roots of the experiment) we can get the results shown in Table 11. In the intact root the frequency of cell division is maximum at the portion 1.0 mm from the root apex, and from that

point upward the rate decreases gradually; in the 1 mm decapitated roots the maximum of the rate is at 0.5 mm from the decapitated surface—and the absolute value of the maximum rate in the 1 mm decapitated root is far less than that of the intact root—and from that point the rate decreases; and in the 3 mm decapitated root the frequency of cell division in the portion near the cut surface as well as that of the basipetal is always small; the region observable in this root is 3 mm from the root apex to the basipetal and as there is no more frequency of cell division, it is natural that there arises no remarkable variation of the frequency of cell division, by the cutting.

The roots, used as the material of this observation, elongated during 4 hours after decapitation, in an average, as follows: the intact root 0.28 mm, the 1 mm decapitated root, 0.08 mm and the 3 mm decapitated root, 0.03 mm. So not taking into consideration the elongation of each of the different kind of roots, we may rearrange the curves as in Fig. 1, namely placing the portion most contiguous to the cut surface of the 1 mm decapitated root (i.e. 0) to the point 1.0 mm from the root apex in the intact root, and the points 0 of the

Fig. 1. Frequency of cell division of the decapitated roots (4 hours after decapitation), in comparison with that of the intact root. Ordinates represent the frequency of cell division; abscissa, distance (in mm) from the apex of the intact root, at the time of measurement. Curve a, the intact root; b, the 1 mm decapitated root; c, the 3 mm decapitated root.

Table 11. Frequency of cell division at the several points in the root (in saw dust) 4 hours after decapitation, indicated by the number of mitotic figures found in the field of 1 qmm (The area of the field examined is given in brackets (in qmm))

	Frequency of cell division at the distance from the root apex or cut surface (at the time of observation) of:										
	0	0.5 mm	1.0 mm	1.5 mm	2.0 mm	2.5 mm	3.0 mm	3.5 mm	4.0 mm	5.0 mm	6.0 mm
Intact root <i>Average of 5 roots</i>	—	8.06 (3.81)	136.86 (4.27)	122.14 (4.86)	82.24 (5.37)	53.42 (5.74)	45.56 (5.95)	26.28 (6.03)	19.90 (6.08)	8.78 (6.09)	3.22 (6.08)
1 mm decapitated root <i>Average of 5 roots</i>	39.60 (4.64)	100.32 (5.23)	70.20 (5.75)	52.90 (6.06)	38.72 (6.28)	26.20 (6.31)	20.78 (6.28)	16.06 (6.30)	15.38 (6.32)	8.66 (6.28)	5.88 (6.34)
3 mm decapitated root <i>Average of 5 roots</i>	15.44 (5.30)	17.68 (5.58)	20.84 (5.75)	18.20 (5.81)	11.16 (5.82)	—	5.84 (5.80)				

Table 12. Frequency of cell division at several points in the root (in saw dust) 7 hours after decapitation, indicated by the number of mitotic figures found in the field of 1 qmm (The area of the field examined is given in brackets (in qmm))

	Frequency of cell division at the distance from the root apex or cut surface (at the time of observation) of:												
	0	0.5 mm	1.0 mm	1.5 mm	2.0 mm	2.5 mm	3.0 mm	3.5 mm	4.0 mm	5.0 mm	6.0 mm	7.0 mm	8.0 mm
Intact root <i>Average of 4 roots</i>	—	21.88 (3.54)	104.38 (4.54)	119.25 (5.19)	77.95 (5.76)	52.40 (5.86)	48.00 (6.17)	25.90 (6.28)	22.53 (6.25)	11.43 (6.22)	4.50 (6.14)	6.58 (6.19)	3.30 (6.89)
1 mm decapitated root <i>Average of 5 roots</i>	39.48 (4.80)	82.08 (5.69)	75.50 (6.44)	65.36 (6.82)	33.62 (6.99)	34.04 (7.01)	20.14 (6.98)	—	9.94 (6.99)	7.12 (7.13)	4.32 (7.46)		
3 mm decapitated root <i>Average of 5 roots</i>	22.14 (5.25)	11.16 (5.63)	6.94 (5.68)	6.94 (5.81)	6.86 (5.81)	—	3.94 (5.83)						

3 mm decapitated root to the point 3.0 mm in the intact root. Comparing these three curves it is seen that in the 3 mm decapitated root and the 1 mm decapitated root, the frequency of cell division is very small at the portions most contiguous to the surface of decapitation (namely 0), as a matter of course, and also at the portion 0.5 mm from the cut surface it is small.

In the next place I studied the frequency of cell division of the roots 7 hours after decapitation (as materials the roots, showing an intermediate rate of the elongation in Exp. XII, Table 3, were chosen). In this case (Table 12) the maximum point of the frequency of cell division in the intact root moved to the point 1.5 mm from the apex; as in the case of the roots 4 hours after decapitation, in the 1 mm decapitated root the frequency in the portion nearest to the cut surface is small and the maximum point of frequency is at the point 0.5 mm from the cut surface; and in the 3 mm decapitated root, the portion nearest to the surface is the maximum of frequency, though the absolute value of it is very short.

The average elongation of the portion within 4 mm from the root apex, before the decapitation, during 7 hours after the decapitation, are as below: in the intact root, 1.1 mm; in the 1 mm decapitated root, 0.9 mm and in the 3 mm decapitated root, 0.2 mm. We may, therefore, put the point of 4.0 mm of the 1 mm decapitated root to the point of 5.0 mm of the intact root; and the point of 1.0 mm of the 3 mm decapitated root, to the point 5.0 mm of the intact root, as shown in Fig. 2.

Comparing these three curves, we find that in the 1 mm decapitated root, the frequency of the point nearest to the cut surface is of course small, and the value at the maximum point is also less than that at the corresponding point in the intact root. It must be, therefore, recognized that the influence of cutting extended to this point namely 0.5 mm from the cut surface. The 3 mm decapitated root shows a curve nearly similar to the corresponding part of the curve of the intact root in this figure. But the majority of the results of the elongation of the region within 4 mm from the apex (at the beginning of the experiment) of the intact root, during 7 hours, is attributable rather to the elongation of the basipetal part of that region, namely near the portions 4 or 5 mm from the root apex, measured 7 hours after the operation. Therefore, the position of 0 of the 3 mm decapitated root must be removed more to the leftward than shown in

Fig. 2. Frequency of cell division of the decapitated roots (7 hours after decapitation), in comparison with that of the intact root. Ordinates represent the frequency of cell division; abscissa, distance in mm from the apex of the intact root, at the time of measurement. Curve a, the intact root; b, the 1 mm decapitated root; c, the 3 mm decapitated root.

the figure; consequently, perhaps it may be true that the frequency of cell division in the root is less than that of the other two.

I did not test the frequency of cell division in the preparations of material more than 7 hours after the cutting, as the real state of the effect of cutting is likely to be fully seen in the two experiments above mentioned, also in fear that in a long time after the operation, besides the direct influence of cutting, there may appear the secondary factor as regeneration, which might disturb the observation.

VI. COMPARISON OF THE CELL DIVISION AND THE CELL ELONGATION

The elongation of the decapitated roots are retarded by the decapitation; on the other hand, there is found no tendency to accelerate the frequency of cell division by the decapitation, but, in general, it tends also rather to decrease; especially at the point nearest the cut surface the frequency is very minute.

As regards the influence upon the elongation, in the case 4 hours

after the cutting, as the method of observation is to compare the length, which is the final result of elongation during 4 hours, the difference of elongation is still so small and not so remarkable.

Yet in the experiments more than 7 hours after the decapitation, we can recognize the influence of cutting more clearly.

However, regarding the frequency of cell division, we can see remarkable differences between them, already in the materials 4 hours after the cutting. The influence of cutting upon elongation extends far from the cut (in the 3 mm decapitated root we could see the result of influence in the portion far more than 2 mm from the cut surface); on the contrary, the influence upon the cell division extends not so far: within 0.5 mm it is seen clearly but at a distance from the cut end of 1.0 mm less distinctly.

In short, the cutting decreases the frequency of cell division and at the same time hinders the elongation of cell. This fact is similar to the result of the inhibition upon the elongation of the root by means of gypsum, showing the retarding of cell division as well as cell elongation.

• Now we must consider the various influences from the conditions introduced by the operation of decapitation, upon the cell division, cell elongation or elongation of root tissue. It is hard to research the results of influences upon the cell division but it is relatively easy to examine some influences concerning the elongation of the cell or that of the root tissue. In the first place we must consider the influence resulting from the change of water content of the tissue.

VII. MEASUREMENT ON WATER CONTENT

In the nature of the study, the measurement on water content was done in the region basipétal 4 mm from the root apex, namely in regions B and C.

The material cultured in saw dust for 24 hours (Exps. VIII and IX, Table 2) was weighed (Table 13) and it was seen that there was no remarkable difference between the 1 mm decapitated root and the intact root in fresh weight, water content and also the ratio $\frac{\text{water content}}{\text{dry weight}}$, and that the 4 mm decapitated root showed less values as to those points. It can be said, therefore, that the 4 mm decapitated

roots were in the state of want of water. However, the materials discussed here are those 24 hours after the decapitation and show only the state of that time. In reality, the elongation of the cell is influenced in the majority of cases by the state of the root prior to that time. So we must research, in the next plan, the materials 7 hours after decapitation.

Table 13. Average water content of the decapitated root (in saw dust) measured about 24 hours after the operation (The unit of weight is mg)

		Intact roots		Roots decapitated at the several points respectively from the root apex:	
		Observed value	Relative value	1 mm	4 mm
Series 1	No. of roots observed	14	100	14	14
	Fresh weight	41.5	100	100.7	90.6
	Dry weight	2.32	100	101.2	99.1
	Water content	39.2	100	100.5	88.5
	$\frac{\text{Water content}}{\text{Dry weight}}$	16.88	100	99.3	89.2
Series 2	No. of roots observed	11		10	12
	Fresh weight	38.2	100	109.4	91.6
	Dry weight	2.43	100	108.6	97.1
	Water content	35.7	100	109.5	91.3
	$\frac{\text{Water content}}{\text{Dry weight}}$	14.72	100	100.7	94.0
Average	Fresh weight	39.9	100	105.1	91.1
	Dry weight	2.38	100	104.9	98.1
	Water content	37.5	100	105.0	89.9
	$\frac{\text{Water content}}{\text{Dry weight}}$	15.80	100	102.0	91.6

Measuring the materials 7 hours after decapitation (Exp. XI, XIII and XIV in Table 3) we can find that regarding water content and the ratio $\frac{\text{water content}}{\text{dry weight}}$ the 1 mm decapitated roots are nearly equal

to the intact root or a little less than that, and the 4 mm decapitated root and 3 mm decapitated root are less than the intact root (Table 14).

Table 14. Average water content of the decapitated root (in saw dust) measured 7 hours after the operation (The unit of weight is mg)

		Intact roots		Roots decapitated at the several points respectively from the root apex:		
		Observed value	Relative value	1 mm	3 mm	4 mm
Series 1	No. of roots observed	13		12	14	13
	Fresh weight	15.1	100	94.0	90.7	87.4
	Dry weight	1.07	100	93.5	100.0	107.5
	Water content	14.0	100	94.3	90.0	86.4
	$\frac{\text{Water content}}{\text{Dry weight}}$	13.08	100	100.6	90.3	80.2
Series 2	No. of roots observed	12		12	12	12
	Fresh weight	17.0	100	105.3	94.1	81.2
	Dry weight	1.02	100	111.8	102.0	89.2
	Water content	15.9	100	105.0	94.3	81.1
	$\frac{\text{Water content}}{\text{Dry weight}}$	15.59	100	94.1	92.6	90.6
Series 3	No. of roots observed	12		12	—	12
	Fresh weight	16.1	100	87.0	—	82.6
	Dry weight	1.08	100	86.1	—	88.0
	Water content	15.0	100	87.3	—	82.0
	$\frac{\text{Water content}}{\text{Dry weight}}$	13.87	100	101.2	—	93.7
Average	Fresh weight	16.1	100	95.4	92.4	83.7
	Dry weight	1.06	100	97.1	101.0	94.9
	Water content	15.0	100	95.5	92.2	83.2
	$\frac{\text{Water content}}{\text{Dry weight}}$	14.18	100	98.6	91.5	88.2

In the materials 4 hours after decapitation (Exp. XVI, XVII and XVIII in Table 4), also we can see the smallness in the value of water content and its ratio to the dry weight of the 3 mm decapitated roots and the 4 mm decapitated roots; the 1 mm decapitated roots are also small in a slight degree (Table 15).

Table 15. Average water content of the decapitated root (in saw dust) measured 4 hours after the operation (The unit of weight is mg)

		Intact roots		Roots decapitated at the several points respectively from the root apex :		
		Observed value	Relative value	1 mm	3 mm	4 mm
Series 1	No. of roots observed	10		10	10	10
	Fresh weight	9.9	100	100.0	88.9	89.9
	Dry weight	0.75	100	101.3	93.3	100.0
	Water content	9.2	100	98.9	88.0	89.1
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.20	100	98.4	94.4	89.3
Series 2	No. of roots observed	14		16	16	14
	Fresh weight	13.4	100	98.5	84.3	84.3
	Water content	0.98	100	101.0	95.9	96.9
	Dry weight	12.4	100	90.3	83.9	83.9
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.66	100	97.1	87.3	86.2
Series 3	No. of roots observed	11		9	11	11
	Fresh weight	16.1	100	93.8	87.6	87.6
	Dry weight	1.24	100	98.4	98.4	101.6
	Water content	14.8	100	93.9	86.5	87.2
	$\frac{\text{Water content}}{\text{Dry weight}}$	11.89	100	95.4	88.0	85.6
Average	Fresh weight	13.1	100	97.4	86.9	87.3
	Dry weight	0.99	100	100.2	95.9	99.5
	Water content	12.1	100	94.3	86.1	86.7
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.25	100	97.0	89.9	87.0

The above mentioned measurements show that the decapitated roots are in want of water. The lack of water probably has its influence intensely upon the cell elongation, thenceforth.

Next, I studied the materials of experiments in water. Materials 24 hours after decapitation (Exp. XX, XXI, XXII and XXIII in Table 5) do not show any definite visible differences among the different kinds of roots, except that the 4 mm decapitated root is a little less in the ratio $\frac{\text{water content}}{\text{dry weight}}$, compared to the others (Table 16).

Table 16. Average water content of the decapitated root (in water) measured about 24 hours after the operation (The unit of weight is mg)

		Intact roots		Roots decapitated at the several points respectively from the root apex :		
		Observed value	Relative value	1 mm	3 mm	4 mm
Series 1	No. of roots observed	8		7	8	15
	Fresh weight	28.5	100	98.2	102.8	103.8
	Dry weight	1.65	100	97.0	104.8	112.7
	Water content	26.8	100	98.5	102.6	108.6
	$\frac{\text{Water content}}{\text{Dry weight}}$	16.25	100	108.6	97.4	96.1
Series 2	No. of roots observed	16		18	14	16
	Fresh weight	31.1	100	95.8	103.9	87.5
	Dry weight	2.02	100	93.6	102.0	89.6
	Water content	29.1	100	95.9	104.1	87.3
	$\frac{\text{Water content}}{\text{Dry weight}}$	14.41	100	102.5	102.1	97.6
Series 3	No. of roots observed	13		14	—	15
	Fresh weight	35.1	100	104.3	—	81.8
	Dry weight	2.16	100	104.2	—	86.6
	Water content	33.0	100	103.9	—	81.5
	$\frac{\text{Water content}}{\text{Dry weight}}$	15.25	100	99.9	—	93.2

Series 4	No. of roots observed	13		12	—	11
	Fresh weight	30.9	100	91.3	—	97.1
	Dry weight	1.97	100	91.9	—	98.5
	Water content	28.9	100	91.3	—	96.9
	$\frac{\text{Water content}}{\text{Dry weight}}$	14.68	100	99.4	—	98.6
Average	Fresh weight	31.4	100	97.4	103.4	93.8
	Dry weight	1.95	100	96.7	103.4	96.9
	Water content	29.5	100	97.4	103.4	93.6
	$\frac{\text{Water content}}{\text{Dry weight}}$	15.15	100	102.6	99.8	96.4

Results of measurements on the materials 7 hours after decapitation (Exp. XXIV, XXV and XXVI in Table 6) is shown in Table 17. As we can see there, in two cases, the 4 mm decapitated root is rather larger than others in the ratio $\frac{\text{water content}}{\text{dry weight}}$; it seemingly promises the superfluous or real elongation of the root, thenceforth.

Table 17. Average water content of the decapitated root (in water) measured 7 hours after the operation (The unit of weight is mg)

		Intact roots		Roots decapitated at the several points respectively from the root apex:		
		Observed value	Relative value	1 mm	3 mm	4 mm
Series 1	No. of roots observed	14		14	14	15
	Fresh weight	16.0	100	107.5	109.2	103.1
	Dry weight	1.11	100	103.4	99.1	95.5
	Water content	14.9	100	107.4	108.1	103.4
	$\frac{\text{Water content}}{\text{Dry weight}}$	13.39	100	103.9	103.2	107.8
Series 2	No. of roots observed	12		12	—	13
	Fresh weight	14.3	100	100.7	—	100.0
	Dry weight	1.05	100	96.2	—	95.2

Series 3	Water content	13.3	100	100.8	—	100.0
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.63	100	105.2	—	105.5
	No. of roots observed	12		13	12	12
	Fresh weight	16.1	100	100.0	98.8	91.9
	Dry weight	1.18	100	100.0	101.7	93.2
	Water content	14.9	100	100.7	98.7	91.9
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.61	100	100.2	97.3	99.0
	Average Fresh weight	15.5	100	102.7	104.0	96.3
	Dry weight	1.11	100	99.9	100.4	94.6
	Water content	14.4	100	103.0	103.4	98.4
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.86	100	103.1	100.3	104.1

Materials 4 hours after decapitation (Exp. XXVII, XXVIII and XXIX in Table 7) were weighed, and the results are shown in Table 18. In two cases of these, the 4 mm decapitated roots are large in the ratio $\frac{\text{water content}}{\text{dry weight}}$. This must be also favourable for the roots to grow superfluously, henceforward.

Table 18. Average water content of the decapitated root (in water) measured 4 hours after the operation (The unit of weight is mg)

		Intact roots		Roots decapitated at the several points respectively from the root apex :		
		Observed value	Relative value	1 mm	3 mm	4 mm
Series 1	No. of roots observed	11		13	12	12
	Fresh weight	13.3	100	104.5	108.3	103.8
	Dry weight	1.01	100	106.9	105.9	95.0
	Water content	12.3	100	104.1	103.1	104.9
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.13	100	97.0	102.6	110.3

Series 2	No. of roots observed	14		14	14	14
	Fresh weight	12.6	100	107.9	106.3	107.1
	Dry weight	0.95	100	108.4	104.2	108.4
	Water content	11.7	100	106.8	106.0	106.3
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.27	100	99.2	101.7	99.0
Series 3	No. of roots observed	13		13	13	13
	Fresh weight	13.5	100	103.0	102.2	104.4
	Dry weight	1.03	100	104.9	102.9	101.9
	Water content	12.4	100	103.2	102.4	104.3
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.07	100	98.8	99.4	102.5
Average	Fresh weight	13.1	100	105.1	105.6	105.1
	Dry weight	1.00	100	106.7	104.3	101.8
	Water content	12.1	100	104.7	105.5	105.5
	$\frac{\text{Water content}}{\text{Dry weight}}$	12.17	100	98.3	101.2	103.9

As a whole, the 4 mm decapitated roots, in saw dust, show the ratio $\frac{\text{water content}}{\text{dry weight}}$ in small amount; but as to those in water, it is not necessarily the case, namely, in some cases the ratio $\frac{\text{water content}}{\text{dry weight}}$ in the 4 mm decapitated root is larger than the intact roots.

Now, we see that there are some differences in regard to the water between the materials in saw dust and in water. Therefore, when we discuss the length of the cell, measured in the decapitated root, we ought to consider, beside the primary effect of the cutting, also the secondary influence of the water absorbed upon the growth of the cell. At any rate, the fact is sure that the shortness of elongation of the decapitated root is due to the shortness of the elongation of the cell.

VIII. EFFECTS OF PRICKING ON THE ROOT TISSUE

In order to give an unusual condition upon the root, instead of cutting, I pricked it and examined the result of the pricking influenced

upon the elongation of the normal part of the root. The method applied here is as follows: after pricking the roots uniformly about 15 times in the respective regions with a fine glass needle (diameter:

Table 19. Elongation of the pricked root during 24 hours after the operation

	Intact roots		Roots pricked at the several portions between the respective points from the root apex :				
	Observed value (in mm)	Relative value	0-4 mm	4-5 mm	5-10 mm	10-15 mm	
No. of roots observed	8		9	9	10	9	
Portion of which the elongation is observed	A	14.45	100	19.9	54.2	37.2	21.2
	B	3.94	100	98.0	20.1	45.9	33.3
	C	5.63	100	77.4	52.2	10.5	26.8

Table 20. Elongation of the pricked root during 24 hours after the operation

		Intact roots		Roots pricked at the several portions between the respective points from the root apex :			
		Observed value (in mm)	Relative value	0-1 mm	1-2 mm	3-4 mm	5-6 mm
Series 1							
No. of roots observed		13		9	12	13	12
Portion of which the elongation is observed	A	9.30	100	77.7	90.6	39.6	65.5
	B	3.71	100	96.5	111.3	37.3	53.2
	C	5.51	100	93.3	101.1	36.2	34.3
Series 2							
No. of roots observed		10		11	11	11	11
Portion of which the elongation is observed	A	10.99	100	93.8	75.6	37.2	63.1
	B	3.72	100	94.4	102.4	63.3	51.9
	C	4.73	100	97.0	108.9	33.4	21.8
Average							
Portion of which the elongation is observed	A	10.15	100	88.3	83.1	38.4	66.8
	B	3.72	100	95.5	106.9	73.1	55.1
	C	5.12	100	95.2	105.0	37.3	26.3

ca. $\frac{20}{100}$ mm), they were planted in moist saw dust. After 24 hours, the elongation of each region was measured (Table 19 and 20). The results of the experiments are similar, in general, to those of decapitated root; the elongation of the portions near the position of pricking are short. After all, this must be attributed to the distraction of some cells and the change of turgor of irritated cells by pricking, and it is the similar cause as in the case of the decapitation. In this case the influence of pricking does not extend to so far a distance as in the case of cutting, and the reason of it may depend on the smallness of the change of water relation in this, in comparison to that case.

IX. EFFECTS OF PRESSURE GIVEN ON A PART OF THE ROOT

Instead of cutting or pricking, I gave pressure to a root so as to change the turgor of that portion.

A piece of fine thread was tied around a distance 3 mm from root apex, and tightened for a while (but not too tightly or the root would be cut off at that portion); that portion was seen to be depressed a little and at the same time the surface of that part moistened slightly, then the thread was untied. Beside these roots, roots decapitated at

Table 21. Elongation of the squeezed root during 24 hours after the operation

Portion of which the elongation is observed	Intact roots		Squeezed roots	3 mm decapitated roots
	Observed value (in mm)	Relative value		
B	5.21 (7) ¹	100	85.6 (8)	67.0 (10)
C	7.27 (11)	100	89.8 (12)	74.3 (12)

¹ The figure given in brackets is the number of roots observed.

3 mm from the root apex, and the intact roots were embedded in saw dust; after 23½ hours the elongation of these roots were measured. The results are as shown in Table 21, namely in C region and B region, the squeezed root elongates to less extent⁷ than the intact root, though the elongation of the former is not so little as the 3 mm

⁷ JANSE (1926, 1927) hindered the elongation of the root by pressing it between wet fingers.

decapitated root shows. Perhaps the smallness of elongation of the squeezed root may be attributed to the destruction or the diminution of turgor of the cells.

X. GENERAL CONCLUSIONS

The principal object of the study is to see the correlation between the cell division and cell elongation, and not to know the detail of the cell division and elongation in every portion of the given material. So, as it is convenient to simplify the work yet to fulfil the expected purpose, I divided the root tip at 4 mm from the apex, and in the portion from that point up to basipetal, I studied mainly the elongation of the cell and the tissue, and in the portion from the point to the apex, I observed the cell division.

To sum up the results, we can find that the cutting diminishes the frequency of cell division, and at the same time shortens the elongation of the cells. But there must be consideration on the results of the investigation, whether those facts are the result influenced directly by the inhibition of further elongation of the root by cutting of the tip, or not. Cutting with even the best sharpened razor, pressure on the portion to be cut can not be avoided. When the root is cut in water, as I did, it can not be seen, but if it is being done in the air we can see that the gloss is off the surface of the portion of root most contiguous to the cut surface and it becomes dull and transparent, and the surface becomes moistened to a slight degree. These phenomena are seen also in the root pricked, namely on the surface near the point of pricking, there appears a small fine strip, which the gloss is taken off and becomes dull and transparent, running parallel to the long axis of the root. And in the squeezed root, as in the case of the cut root, the taking off of the gloss, turning dull and transparent and the moistening of the surface are seen⁸. On the other hand, by cutting we can see that the surface of the cut is wet with moisture from the tissue of the root. Perhaps those abnormal phenomena may also happen when the root is cut in water, as in the cases of our experiments. Moreover, from the measuring the water content and the difference between the results of experiments in saw dust and in water, we can generally say that the de-

⁸ cf. JANSE, 1926, 1927.

capitated roots in saw dust are losing the water from the cut surface. Putting these facts together we may consider that the cutting or injuring causes the change of water content in the tissue or of the turgor. Therefore, at least a part of the cause of the diminution of the elongation of tissue or cells of decapitated roots is due to the inhibition of cell elongation by the diminution of turgor. But it is questionable in this case whether it is the whole of the cause of that, or not. On the other hand it was observed that the cell division is hindered by the cutting, on the portion near the cut surface. I have observed in my previous study (1931) that the frequency of cell division was hindered by diminution of turgor of the root by the use of sugar solution. Probably also in this case, it must be one of the causes of the hindering of cell division, that the diminution of turgor has influenced it either directly, or indirectly, through the effect of it upon the cell elongation. Namely the presence of conditions diminishing the turgor of the cell seems to cause the hindering of the cell division. Yet it is not of importance to discuss here whether the cause effects directly or indirectly, or in these two ways side by side.

In short, in case of inhibiting the elongation of the root tip by decapitation, the cell division and cell elongation are hindered at the same time, as in the cases of inhibiting the growth of the root by means of gypsum or sugar solution.

It is obvious that after a long time there may be the formation of a callus or adventive root at the place of cutting or that in the neighbourhood to it, for the sake of that, the regenerative activity of tissues must induce the cell division; but this must be considered rather the appearance of secondary phenomenon caused by injuring. The point of view of this study is to see the direct relation of cell division and elongation in the root tissue itself, inhibited from the growth; therefore we are to deal with only the primary results of cutting, in our case. In other words, the correlation of growth and cell division or tissue differentiation is mingled in complicated connection of several direct or indirect phenomena, therefore, these problems must be considered carefully and analytically, lest one should reach an unexpected and conflicting conclusion. It is regrettable that one is liable to get the true phase of phenomenon, concerning these problems of differentiation of tissue and growth, from considering only the final state which has resulted from the phenomenon during a long time. One must inquire into the problems as analytically as possible.

XI. SUMMARY

1) The writer observed the relation between cell division and cell elongation in the decapitated roots. And in order to simplify the study the cell division was studied chiefly in portions within 4 mm from the root apex and the elongation in the portions basipetal from that point.

2) Measuring the roots cultivated in saw dust, it was found that the elongation of the portions basipetal from the 4 mm point of the roots decapitated at 3 mm or 4 mm from the root apex is less than the intact root; and that the 1 mm decapitated roots, 24 hours after decapitation, elongate rather longer than the intact roots (control), yet the 1 mm decapitated roots, 4 hours or 7 hours after the cutting, elongate in a still lesser degree, compared with the intact roots.

Next, measuring the portion within 4 mm from the root apex, it was seen that the elongation of all the decapitated roots was always less than the intact root.

In the cases of experiments of materials cultured in water, it often happened that decapitated roots elongate longer than the intact roots, especially concerning the portion within 4 mm from the root apex, the elongation of the 1 mm decapitated root was always longer than that of the intact root. However, this is probably to be attributed to the disturbance of the normal elongation, by superfluous stretching, caused by the excess of water.

3) Putting together the results of measuring the water content of the portion of root, on which the elongation was observed, and other several experiments, it is likely to be assumed that by cutting or by other analogous operations the water content or turgor of the portion is mindered, and this must be one of causes inhibiting the elongation.

4) The cell lengths measured in the positions basipetal from the 4 mm point, run parallel to the rate of elongation of corresponding portions.

5) Diminution of the frequency of cell division is seen at the portion near the cut surface.

6) What is the cause of the diminution of cell elongation and cell division, in the decapitated roots, is beside the question in our case, yet the diminuent of the frequency of cell division as well as retardation of elongation of cells go together side by side. And this

result is parallel with the cases of inhibition of growth of the root treated by gypsum or sugar solution.

7) In considering the final results only of such experiments one may come to an erroneous conclusion, because there are several related indirect phenomena such as the formation of calli or adventive roots etc., besides the direct effects of cutting upon the division and elongation of cells, which is the only question which should be analysed here.

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