Regeneration and its relation to traumatropism.

By

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(With 4 images in the text.)

It is a well known fact that plants are able to regenerate tissue at the root-tip, when such tissue has been destroyed, although the injury be so great that the entire tip be removed.[1]) It is equally well known that when root-tips are wounded in any manner such as burning with a hot glass rod, cutting etc., within the sensitive zone, that they respond to the stimulus, developing a traumatropic curve.[2]) The intensity of this curve varies within large limits. In some cases the roots describes two circles while in others the angle of divergence from the perpendicular is very small. Those roots which develop the extreme curves were injured very severely and die almost without exception, the plant depending upon the development of lateral roots.

The preceeding observations show that both regeneration and traumatropic curvature follow the same stimulus. On this point Spalding says,[3]) „the phenomena go hand in hand, and it seems impossible not to regard them as two different forms of Nachwirkung resulting from the same cause".

The relation of regeneration to traumatropic curvature as seen in the root-tip has not been, as far as I know, a subject of special consideration. Spalding refers only to the biological significance of the traumatropic curve when considered in connection with regeneration. He says, „In case of injury to the growing point of the root it is essential to the welfare of the plant that repair should take place as promptly and economically as possible. This is accomplished with remarkable rapidity by the process of regeneration. Meanwhile it is also important that while the work of repair is going on the root should avoid further contact with the source of injury . . . . . This is brought about by traumatropic curvature . . . . ."

In this same paper one division is devoted to a study of „Suspension of growth and its Relation to Traumatropic Curvature". Roots of Lupinus albus, Vicia Faba, and Zea Mais were wounded and placed in plaster-casts. The casts were then placed in moist saw-dust in such a position that the roots were perpen-

dicular. They were allowed to remain in this position in the casts for periods ranging from twenty two hours to more than eight days. At the end of this time they were released and grown in water or a damp atmosphere. In sixty to eighty minutes the roots showed the traumatropic curve. The reaction was the same as it would have been eight days before „had it not been hindered by mechanical means". Apparently the latent period was prolonged in the one case over eight days.

Two points of view were possible.[4]) Either the influence of the stimulus was conducted to the elongating part of the root and there held eight days or else, there was a continual stimulus. To determine this point the author cut off the wounded area after the root had been in the casts twenty-four hours and returned to their casts. They were finally removed from the casts and grown in water in a vertical position. Of eleven roots of Lupinus albus six showed a negative curve but this was not always at right angles to the first wound. Some were oblique. This method however is open to criticism in that a second stimulus is introduced the result of which it is impossible to separate from the result of the first. However this was only a side question to the theme in hand and the author dismisses it with these words,[5]) „that the latent period may by such artificial means be extended to more than a week is a face of sufficient physiological importance to warrant the more extended investigation which it is hoped may hereafter be given it".

It is with this question that the present paper deals.

Materials and methods.

The experiments recorded here were conducted in the winter and spring of 1902 when I was making a general study of the latent period on roots under the direction of Prof. F. C. Newcombe.

The experiments were conducted for the most part on the roots of Vicia Faba and Pisum sativum.

The wounding was done with a glass rod or by cutting. The first method was the one generally used. The glass rod was drawn to a fine tip which was heated to redness and then brought in contact with the growing root-tip one to one and one-half mm from the tip. A few minutes after burning a small brown spot could be seen which indicated the amount of tissue destroyed.

After wounding the plants were placed first, under conditions stopping both growth and curvature; second, others under conditions stopping curvature only; and still others under conditions allowing both.

In every case control plants were used and an effort was made to work only with strong growing plants. The results cited were obtained from work on a large number of roots but only a few will be noted in this paper.

Experiments.

A. Seedlings were allowed to remain in sawdust until the root was 2 or 3 cm long. They were then wounded by branding with a hot glass rod as described, and placed in a damp chamber in a vertical position. The temperature was about 22° C.

Seven peas were branded 1 mm from the tip.

No. 1. Described a circle and died.
„ 2. Curved at an angle of 100 degrees.
„ 3. „ „ „ „ „ 90 „
„ 4. „ „ „ „ „ 90 „
„ 5. „ „ „ „ „ 90 „
„ 6. „ „ „ „ „ 80 „
„ 7. „ „ „ „ „ 60 „
„ 8. Control. Grew straight.

The roots continued to grow at about the angles indicated for some time. Gradually they turned in response to gravity some of them at right angles while others never reached the vertical position. The length of time during which the root did not respond to gravity varied with different roots according to the severity of the wound. One root was especially noteworthy in that it grew at an angle of 80 degrees for six days. At the end of that time it turned down almost at right angles. The root grew well during all of this time. (Fig. 1).

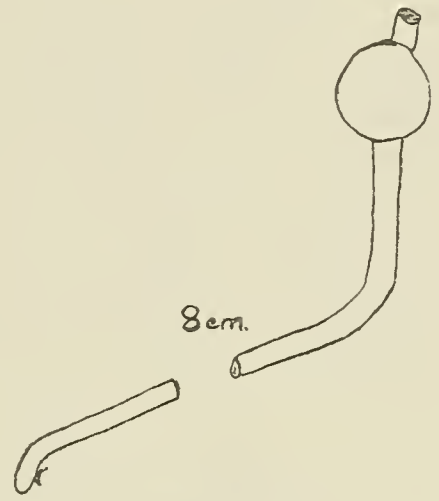

Fig. 1.

One important observation was made on this set of seedlings which led to the solution of the problem. It was noticed that about the time that the root-tips turn down the wounded tissue either had disappeared or remained as a little brown disc on the root. This was easiliy removed with a needle. As this subject receives careful consideration in a following paragraph further mention at this place will be omitted.

B. Seedlings whose roots were 2 or 3 cm long were wounded and placed in plaster-cats. This was best done by placing the root to be imbedded on a small glass plate 1 by 3 cm, adding a very little plaster and finally a second glass plate the same size as the first. After the preparations had hardened a little they were tied firmly together. They were then placed in a vertical position in damp sawdust and allowed to remain from two to eight days. At the end of this time they were removed from the casts, which is very easily done without injury to the tips and placed in a damp chamber or on the klinostat. The results were the same as those recorded by Spalding. The roots developed a traumatropic curve. (Fig. 2.) Apparently the latent period has been prolonged by this mechanical means.

Fig. 2.

C. In this set of experiments the roots after wounding were allowed to grow but the traumatropic curve was prevented. For this purpose glass tubes of small bore were used. After being wounded 1 mm from the tip with the hot glass rod, the roots were inserted into the glass tubes and the preparations were kept in a dampchamber or in water. The tubes varied in length from 3 to 8 cm. The roots then must grow from 1 to 6 cm before they can produce the traumatropic curve.

1. Eight roots were wounded and placed in tubes 8 cm long. May 20th. 1902.

No. 1. Came out and gave curve of 45 degrees.
„ 2. „ „ „ „ „ „ „ 60 „
„ 5. „ „ „ grew straight.
The other five died in the tubes.

2. Roots wounded as in the previous experiment, May 16th. at 10 A. M.

	Distance root must grow in tube.	Amount of growth May 17th. at 8 P. M.	Angle of curvature May 19th.
No. 1.	6 cm	4 cm	60 degrees.
„ 2.	4 „	3 „	20 „
„ 3.	5 „	4.5 „	45 „
„ 4.	5 „	4.2 „	10 „
„ 5.	5 „	4.5 „	90 „
„ 6.	3 „	out	60 „
„ 7.	4 „	„	60 „

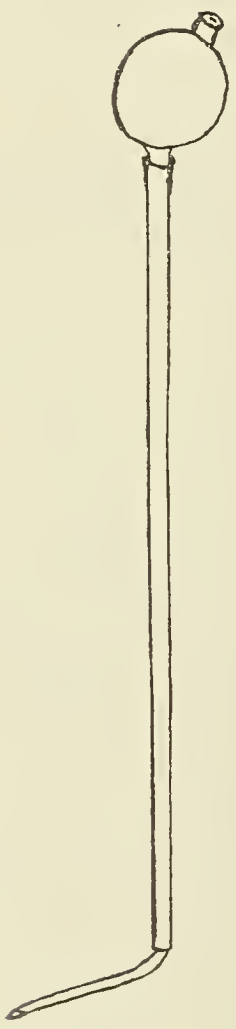

Fig. 3.

These roots were allowed to grow either in water or in a damp chamber and in a short time all turned down in answer to gravity.

A number or experiments were set up to determine the length of time a root can hold the influence of the geotropic stimulus. This was done for the sake of comparison. Uninjured plants whose roots were from 2 to 3 cm in length were placed in plaster-casts and then left in damp sawdust in a horizontal position from 30 minutes to 15 hours. At the end of this time some were reversed so that gravity worked in the opposite direction from what it did at first, others were placed in a vertical position. and still others were taken from the horizontal position and placed on the klinostat. They were left under these conditions from one to seven hours when they were removed from the casts and all placed on the klinostat in a damp chamber. The results are shown in the following table.

All of these roots curved immediately on leaving the tubes except number 6 which grew straight 2 mm and then developed a traumatropic curve. I am not able to account for this fact.

No. of seeds.	Time in horizontal position in casts.	Time in vertical position in casts.	Time in reversed horizontal position.	Time on klinostat in casts.	Results when removed from casts and revolved on klinostat.
20	10 hrs.		1 hr.		Responded to first stimulus.
15	15 hrs.		1 hr.		ditto.
20	15 hrs.		2 hrs.		Responded to the second stimulus.
37	5 hrs.	7 hrs.			Grew straight.
10	1 hrs.			6 hrs.	ditto.
10	10 min.			1 hr.	9 grew straight 1 developed the curve.

A study of this table shows that roots are able to hold the influence of a geotropic stimulus for a very short time only. It would be interesting to determine the exact length of time but from the standpoint of this paper it is not essential.

Anatomical studies.

In the foregoing experiments it has been noted that the roots turned down in answer to gravity after a certain length of time. Roots were killed and sectioned longitudinally to study their structure at the time when this change took place. Those roots were selected on which the brown mass of dead tissue still remained. This usually dropped off during the process of imbedding but by using a large part of the root it was not hard to get sections through that part of the root which had been wounded. A study of these sections showed no trace of the wound. The process of regeneration was completed.

A series of sections was now made from roots which had been wounded and which still showed a marked traumatropic curve. Figure 4 shows part of the longitudinal section of a root which was killed 48 hours after wounding. At the time it was killed it was growing at an angle of 90 degrees. The figure shows that the wound has not been regenerated but that process is still going on.

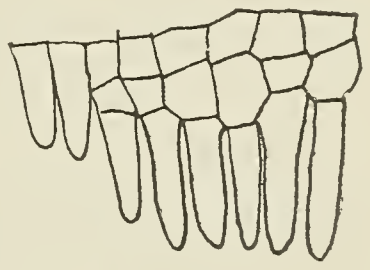

Fig. 4.

Discussion and conclusions.

A review of the facts obtained by the experimental work cited in the preceding pages show that the two phenomena, regeneration and traumatropism, are very closely connected in the case of the root-tips examined.

The first set of experiments, taken in connection with the anatomical work, show clearly that as long as wounded tissue is found, the root will form a traumatropic curve. Or in other words, when the process of regeneration is complete the stimulus causing the curve is removed. This seems to show then, that there is a constant irritant and that the latent period is not

prolonged as was apparently shown by the second set of experiments.

The other question finds its solution, as I believe, in the third set of experiments. Is the influence of the stimulus conducted to the elongating zone and there held until the root is free to respond to it? This experiment with the glass tubes is free from the objections cited before against the method of cutting away the wounded tissue. Roots wounded and grown in tubes grew as high as 6 cm and responded to the stimulus over two days after they were wounded. If now the influence of the stimulus was conducted to the elongating area immediately and there held it must have returned down the root as it elongated. This seems highly improbable although we have some evidence that the traumatropic stimulus may be conducted toward the tip as well as in the opposite direction.

The work seems to justify the following conclusions:

1. That the influence of the stimulus is not conducted to the elongating zone and there held from one to eight days.
2. That the latent period is not prolonged by mechanical means.
3. That the wounded tissue forms a constant irritant.
4. That this irritant is removed when regeneration is complete.

University of Michigan, January 4, 1904.

Literature.

1) Prantl, Untersuchungen über die Regeneration des Vegetationspunktes an Angiospermenwurzeln. (Arbeiten des Bot. Inst. in Würzburg. Bd. 1. p. 546. (Older literature is cited here.
Lopriore, Über die Regeneration gespaltener Wurzeln. (Nova Acta Leop. Carol. 66. No. 5.) His other papers are cited here.
2) Spalding, On the traumatropic curvature of roots. (Annals of Botany. Vol. 8. 1894.)
3) „ l. c. p. 448.
4) „ l. c. p. 438.
5) „ l. c. p. 450.

Explanation of figures.

No. 1. Pisum sativum six days after branding. The wounded tissue has been regenerated and the root is turning down.

No. 2. Pisum sativum. Radical six hours after release from plaster-cast in which it had been confined 72 hours after previous branding.

No. 3. Pisum sativum. This radical, after being wounded, grew 4 cm in the tube and developed a traumatropic curve on leaving it.

No. 4. Pisum sativum. Small part of a longitudinal section of a root-tip which was killed 48 hours after wounding. The process of regeneration is not complete.