

Citation:

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Botany. — "*Contributions to the knowledge of the movement of water in plants.*" By Dr. K. ZIJLSTRA. (Communicated by Prof. J. W. MOILL).

(Communicated in the meeting of January 29, 1910).

For some time I have been occupied in the botanical laboratory at Groningen with the problem of the movement of water in plants and have carried out experiments of a somewhat diverse nature. Various circumstances have prevented me from continuing my experiments in this direction, so that the investigation has not been rounded off. I did not intend publishing it, but as I shall presumably have for some time no further opportunity of continuing my studies, I think I may be justified in publishing the data I have collected; possibly they may be of service to other investigators who have chosen for their researches the subject of the movement of water in plants.

The experiments referred to may be arranged under three heads, viz.:

1st. The trunk or stem of intact plants cooled to about 0° C.

2nd. The ascent of a dye solution in cut branches.

3^d. Interference with the movement of water in a tree-trunk by means of deep incisions.

First I propose to discuss the considerations which led to these experiments and the results obtained, and then I will give a more detailed account of the execution of the experiments.

1. *Trunk or stem of intact plants cooled to about 0° C.*

As is well known, GODLEWSKI (Zur Theorie der Wasserbewegung in den Pflanzen. Jahrb. f. Wiss. Bot. Bd. 15) attempts to find the cause of the movement of water in the activity of the living cells of the medullary rays and of the wood parenchyma; these cells would therefore have to act as it were as suction-pressure pumps. GODLEWSKI did not, however, adduce any direct experimental evidence in support of this theory. His theory is only made plausible with the aid of various data obtained by others, and it is urged that the theory does not conflict with the facts adduced by other investigators. Various botanists (JANSE, STRASBURGER, WEBER, URSPRUNG) have afterwards attempted to test the theory experimentally.

The most obvious method for such a test would be the following: to cut out the action of the living cells of medullary rays and wood parenchyma, and then to see whether the movement of water had become impossible.

This elimination of the action of living cells was most easily obtained by simply killing these cells by poisons or by a high temperature.

This method is, however, open to objection; such interference not only attains the elements which it is desired to put out of action; others also, especially the water-conducting vessels and tracheids will undoubtedly be affected, so that it is questionable whether the results of the experiments can only be attributed to the elimination of the activity of the living cells.

A method — already used by URSPRUNG but with a result opposite to mine — which meets this objection, is the cooling of the trunk or stem of the plant to about 0°. By this means it is possible to reduce the activity of the living cells to a minimum, while neither dead nor living elements undergo a permanent change. Moreover the advantage of being able to establish the original conditions after the conclusion of the experiment and therefore bring the plant back to normal conditions, should not be underrated. The experiment and its control can both be carried out on the same intact plant.

If by this means the plant could be made to fade, and to assume its original fresh appearance after the cooling had been stopped, GODŁEWSKI'S theory would receive considerable support.

According to this method I have myself carried out 3 experiments. The trunk of a small apple-tree, 2 stems of *Polygonum cuspidatum* and 2 stems of *Helianthus tuberosus* were cooled to about 0° C. over a length of 50 cm. The experiments lasted 6, 7, and 8 days, under conditions which were very favourable for a possible fading.

Nevertheless I have in no case been able to observe even incipient fading, although the transpiration from the leaves was strong, as shown by the cobalt test. Cut leafy branches, hung up near the plant, withered very rapidly.

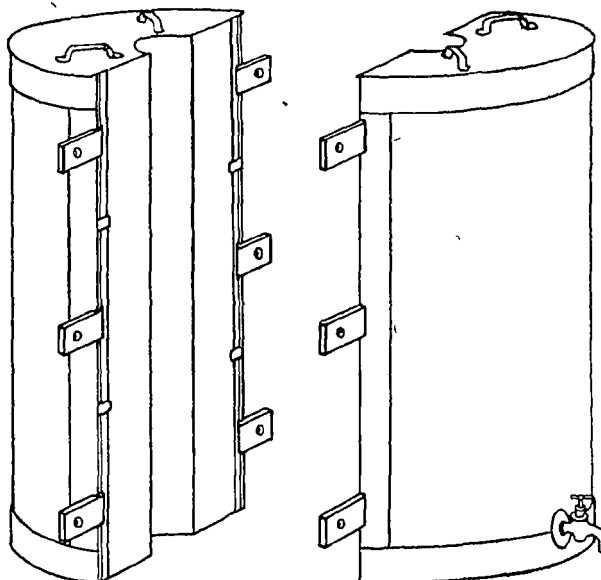
We may not, however, conclude from the negative result of these experiments that the living cells do not play a part in the movement of water. It is quite possible, even probable, that cooling a length of 50 cm. is not enough. This slight obstacle was perhaps easily overcome by differences of pressure present in the trunks. Had the results been positive it would have supported GODŁEWSKI'S theory. My negative results are, however, not able to oppose this theory.

The nature of the results notwithstanding, I think it may be useful to bring them to the notice of others.

Description of the experiments.

The cooling of the trunk or stem was brought about by melting ice, which was placed in an apparatus indicated by the figure.

The apparatus consisted of two equal parts, i.e. of two semicircular tin-plate reservoirs with fixed bottom and loose lid. The two half-



cylinders had on the middle of the flat side a portion which was bent like a half cylinder, so that the two reservoirs when joined to form one cylinder, left in the centre a space for the passage of the trunk, which was to be cooled. The height of the apparatus was 50 cm., its diameter 30 cm.; the space left free for the trunk had a diameter of 10 cm. Each reservoir was provided at the bottom with a tap, through which superfluous water could run off. The cylindrical surface of each reservoir and also the bottom and the lid, were covered on the outside with a layer of felt, 15 mm. thick and over this there was a covering of asbestos paper, 2 mm. thick. In an experiment the two reservoirs were placed round the stem and screwed together, after a piece of felt had been placed between the two flat surfaces in apposition.

The reservoirs were filled with ice. The space through which the trunk passed, was closed off above and below round the stem by a solid plug of cotton wool through which a thermometer passed. The temperature in the annular air space surrounding the trunk varied between 0° and $+3^{\circ}$ C.

The apparatus was sufficiently protected by the felt and the asbestos against the heat of the surrounding atmosphere. Even on hot days it was only necessary to renew the ice twice in 24 hours.

During the experiment the apparatus rested on some bricks, so that it was about 20 cm. from the ground.

EXPERIMENT I.

Apple tree.

The ice apparatus was fixed round the trunk having a diameter of $3\frac{1}{2}$ cm., of a small apple tree, about $2\frac{1}{4}$ metres high, on July 21st 1904, at noon, the weather being hot and sunny. The apparatus was filled with ice and in the course of the afternoon the temperature in the space round the trunk fell to about 1° C.; not the slightest fading of the leaves could be detected, although such fading would have at once been noticeable by comparison with two other apple trees which stood next to the tree experimented on.

Nor could any change be observed on the following days. The temperature of the air space round the stem remained continuously between 0° and 3° .

The maximum temperature of the atmosphere on the days of the experiment oscillated between 23° and 29° .

On the sixth day, when the temperature of the atmosphere was 20° and that round the trunk 0° , an strong transpiration of the leaves was demonstrated by means of the cobalt test. On the seventh day the trunk was sawn through immediately above the ice apparatus; a hole was drilled in the portion of the trunk still inside the apparatus, and a thermometer was placed in it.

In this way I was able to show that the temperature *inside the trunk* was the same as that of the annular air space *round the trunk*, i.e. in the course of three hours it oscillated between 2° and 3° , while the temperature of the atmosphere was 24° to 25° .

EXPERIMENT II.

Polygonum cuspidatum.

The ice apparatus was fixed round two immediately adjoining stems, 2 metres in height, on July 6th 1905, at noon, and it was filled with ice. In the course of the afternoon the temperature in the air space round the stems fell to 0° , without withering taking place. The numerous stems surrounding the apparatus served as controls. Nor was any change noticeable during the following days.

The temperature round the stems remained continuously between 0° and 3° . The maximum temperature of the surrounding atmosphere

on the various days during the experiment oscillated between 19° and 30° . The experiment was stopped on the seventh day.

EXPERIMENT III.

Helianthus tuberosus.

The ice apparatus was fixed round two immediately adjoining stems of plants, $1\frac{1}{4}$ M. in height, on July 14th 1905 at noon, and was filled with ice. In the course of the afternoon the temperature of the air space round the stems fell to 0° . No fading could be observed; several specimens of the same species, standing next to the plants experimented upon, served for comparison. Cut leaves, hung up on the plants were completely withered in a few hours. Nor was withering observable on the experimental plants on following days.

The temperature in the air space round the stems remained about 0° . The maximum temperature of the atmosphere in the days of the experiment oscillated between 17° and $26\frac{1}{2}^{\circ}$. The experiment was stopped on the eighth day.

2. *Ascent of a dye solution in living and dead cut branches.*

When cut branches, with a freshly cut surface, are placed with this surface in a dye solution, the liquid will in general ascend into the branches for some distance, and thus may be easily traced by cutting them across at different levels. Various elements of the wood are then found to have been stained. It matters little whether one takes for this experiment living or dead branches, with or without leaves; the fluid always ascends in the branches, even when these are upside down, i. e. are placed in the solution with their cut apex. I generally carried out such experiments with twigs 30—40 cm. long; sometimes with pieces of a branch, which had also been cut at its upper end. After some days the stain shewed itself on the surface of the upper section of these latter branches.

Although the dye ascends in all branches, the *way* in which the various elements are stained is not the same in living and dead branches. A sharp difference is observable.

In comparing living branches with dead ones, it was of course necessary to use a harmless stain; the experiments of STRACKE: investigation of the immunity of the higher plants towards their own poison (Dissertation), led me to choose *Säureviolett* of GRÜBLER. I used this stain in a $\frac{1}{10}\%$ aqueous solution. The twigs were placed

separately in a small bottle with the solution of the stain, the neck of the bottle being closed with a plug of cotton wool to prevent evaporation.

After the experiment the twigs were examined at different levels by microscopic sections. Transverse, radial and tangential sections were examined in *oil of cloves*, a medium in which Säureviolett is insoluble, so that the stain remained properly localized. The sections were cut without the use of any liquid and were at once placed in the oil of cloves. The slight water content of these preparations did not interfere. After a very short time the oil had thoroughly permeated. This method had moreover the advantage, that after most of the clove-oil had been wiped away, the preparations could be very well enclosed in Canada balsam, without further treatment.

A comparison of the behaviour of the xylem elements of living and dead branches brought out the following differences:

<i>living branch</i>	<i>dead branch</i>
a. torus of the closing membrane of the bordered pits deeply stained.	a. torus not stained, or only very slightly.
b. adjoining the lumen, a thin layer of the wall in the border of the pits is stained. The walls of vessels and fibres are only stained in a very thin layer, which is immediately adjacent to the lumen.	b. the walls of the vessels, fibres and parenchymatous cells are stained uniformly.
c. contents and wall of the cells of medullary rays and wood parenchyma are unstained.	c. contents of the cells are coloured.

The deep staining of the tori in living branches was especially noticeable, also in transverse sections, the more so because the staining of the layer next to the lumen in the walls of vessels and fibres was often difficult to see and because the living cells of the medullary rays and parenchyma were quite colourless.

In the wood of *Salix* and of *Fagus*, in which the tori cannot otherwise be seen at all, they were made very obvious by this staining of living branches.

The staining of the tori by eosine in a living branch of *Ginkgo* was already mentioned by JANSE in "Die Mitwirkung der Markstrahlen bei der Wasserbewegung im Holze" (Jahrb. f. Wiss. Bot. 1887 Bd. XVIII). In this case also the stain had ascended the branch:

the "primäre Wandlamelle" of the medullary ray cells was according to JANSE all that had been stained.

In my experiments with living branches the staining extended not only to the tori of the vessels and fibres, but also to those of the *half* bordered pits between the medullary ray cells on the one side and the vessels and fibres on the other side; the *contents* of the medullary ray cells however remained colourless, as stated above.

The results of other experiments carried out by me, agree well with these facts. Instead of taking dead branches, I caused to ascend in living branches a $\frac{1}{10}\%$ solution of Säureviolett in strong alcohol, and also a $\frac{1}{10}\%$ solution in water containing 4% formaldehyde. As controls I employed living branches in a $\frac{1}{10}\%$ solution of Säureviolett in water.

I now found that the living branches in the poisonous solutions were stained practically in the same way as the dead branches in innocuous ones, only not so completely. It was clear that the alcohol and the formaldehyde only gradually exercised their fatal action on the plant. The tori were always unstained; only a few were stained very faintly. The walls generally showed a uniform staining; the medullary ray and parenchyma cells with contents were coloured dark blue.

Finally I may add that microscopical transverse sections through living branches, which sections were afterwards placed for 20 hours in an aqueous Säureviolett solution of $\frac{1}{10}\%$, were stained quite uniformly dark blue, exactly in the same way as those sections made after the stain had ascended in *dead* branches; the colour was only somewhat more intense. The transverse sections through control branches, which had previously stood in the same solution for 4 days, on the other hand showed, as was to be expected, a staining quite similar to that which was described above for living branches.

Description of the experiments.

EXPERIMENT IV.

Fagus silvatica.

A living leafy twig, about 4 mm. thick at its base, stood for 9 days in a solution of Säureviolett. The stain ascended to the top and into the leaves. The bark, the cambium and the pith remained quite unstained; the staining was limited to the wood and here the stain was only in the inner layer (adjoining the lumen) of the walls of vessels and fibres; the tori of the bordered pits were stained a very

deep blue-violet, and this was also the case with the half bordered pits between medullary ray cells and fibres. The medullary rays and the xylem parenchyma were quite unstained, both as regards wall and contents.

EXPERIMENT V.

Larix decidua.

A living leafy twig, 6 mm. thick at its base, stood for 5 days in the solution, after which the stain had penetrated to the apex. Staining completely limited to the wood, but no stain in the oldest of the 6 annual rings.

The stain only taken up by a very thin layer of the wall, adjoining the lumen of the tracheids and the cavities of the pits. Torus of the pits deep blue-violet, also in the half bordered pits between medullary rays and tracheids. For the rest everything unstained.

EXPERIMENT VI.

Salix spec.

Two living leafless branches, provided at either end with a cut surface, both 30 cm. long and more than $\frac{1}{2}$ cm. thick, stood for 2 days in the aqueous solution of Säureviolett; one of the branches had its lower end in the solution, the other its upper end.

The stain ascended readily, and *in the two branches simultaneously*. The stain only present in a thin layer of the wall adjoining the lumen of the vessels and fibres and the cavities of the pits. Tori deep blue-violet.

EXPERIMENT VII.

Fagus silvatica. Taurus baccata.

Of each of these plants two similar 3—5 year old leafless branches were placed with the cut surface in the aqueous Säureviolett solution for 3 days. One of the branches of each species was alive, the other had been treated as follows. It had stood for $1\frac{1}{2}$ hours in boiling water. Then water was sucked through the boiled branch by means of a filter pump in order to remove possible obstructions, finally a fresh surface was cut.

After 3 days the stain had almost reached the top in all the four branches.

In the living branches staining was scarcely visible against the walls of the vessels and tracheids. The tori, including those of the half bordered pits were deeply stained. Medullary ray- und parenchyma cells quite colourless.

In the boiled branch of *Fagus* the walls of the libriform fibres and of the vessels were a uniform pale blue. Against the walls of the vessels in the spring wood a darker layer. Nowhere however coloured tori. The medullary rays also proved to be colourless.

In the boiled branch of *Taxus* the walls of many tracheids were stained a uniform pale blue; towards the inside against the walls a darker layer. The tori unstained. The medullary rays dark blue,

EXPERIMENT VIII.

Taxus baccata.

Two living branches were taken. One was placed with its cut surface in a solution of 0.1 gram of Säureviolett in 100 c.c. of *water*; the other in a solution of 0.1 gram of Säureviolett in 100 c.c. of *alcohol*.

Both branches remained standing in the solution for 43 hours, after which time sections were made through both at a height of 7 cm. The staining was as follows:

Branch in *aqueous solution*: staining only in the secondary xylem. A very thin blue layer against the walls of the tracheids, and of the cavities of the bordered pits. Tori dark blue, including those of the half bordered pits. Medullary rays unstained.

Branch in *alcoholic solution*: the stain had also penetrated into the cambium and the innermost layers of the cortex parenchyma, where both walls and contents were dark blue. In the secondary xylem the tracheid walls light blue; against the walls also clearly a blue layer, further in the cavities of the pits. Tori unstained. Medullary rays dark blue, both as regards walls and contents. The walls also coloured in the primary xylem.

EXPERIMENT IX.

Taxus baccata.

A living branch was placed with the cut surface in a solution of 0.1 gram of Säureviolett in 100 c.c. of a 4% *formaldehyde solution* (diluted formalin).

After 3 days the branch was examined; the stain had already reached the apex.

Staining only in the secondary xylem. Against the walls of the tracheids there was a thin blue layer, also in the cavities of the bordered pits. Tori colourless. Of the medullary rays both the walls and the protoplasm dark blue.

EXPERIMENT X.

Salix spec.

A living twig was placed with its cut surface in a solution of 0.1 gram of Sänrevioletti in 100 c.c. of a 4% formaldehyde solution. After 3 days the stain had penetrated to the apex and the twig was examined.

Staining only in the secondary xylem. The walls of the vessels coloured light blue with an indication of a somewhat darker layer adjoining the lumen. Tori practically colourless. The medullary ray cells, which adjoined the vessels, are coloured blue.

3. *Interference with the movement of water in a tree-trunk by means of deep incisions.*

EXPERIMENT XI.

An experiment with a small willow tree in the Botanic Gardens at Groningen showed, that in a trunk in which the transpiration current had been largely prevented or perhaps completely cut off as a result of transverse incisions on both sides at various heights, measures were taken in course of time which ultimately led to a complete recovery of this current.

The experiment was carried out as follows.

The trunk of the tree, 11½ cm. thick, was sawn into transversely to slightly beyond the centre at four places, alternately on either side of the trunk. The incisions were 22 cm. apart, and the lowest was 1.25 Metres from the ground. They were prevented from closing up again by the insertion of tin plates, which in future remained in position. At these four places the water current was therefore irreparably interrupted.

As the trunk had of course been greatly weakened by this operation the tree was supported by four iron wires, which were attached high up to the trunk and also to four pegs driven into the ground at some distance round the tree.

This experiment was started on July 14th 1908; the incisions were ready and the plates were pushed in at 9.30 a.m. At 10 a.m. the leaves were already drooping and they remained so throughout the day.

In the course of the five following days, in cool dry weather, the leaves gradually recovered. On the 7th day of the experiment the foliage began to wither from the top downwards; many yellow leaves also appeared in the crown. In all these days the temperature had not risen above 18° in the neighbourhood of the tree. On the 9th day the temperature rose in the afternoon to more than 26°, and probably as a result of this the number of yellow leaves now increased rapidly. Those leaves which had remained green also began to droop again. The tops of the branches in the upper part of the crown withered completely.

The 3 following days were warm and sunny with temperature maxima of 27° and 28°. Most of the leaves now fell off, while in the upper half of the crown the foliage withered completely.

After this time cooler weather supervened and the few remaining green leaves recovered and remained in good condition until the autumn.

That the tree had not suffered greatly however from the incisions, was shown in the following summer, for then the foliage developed as well as before the experiment, and remained fresh throughout the entire season.

Wageningen, Dec. 13th 1909.

Physics. — "*The magnetic separation of absorption lines in connexion with Sun-spot spectra.*" (I). By Prof. P. ZEEMAN and Dr. B. WINAVER.

1. As a consequence of the intimate connexion between emission and absorption, there exists closely corresponding to the magnetic separation of emission lines, a magnetic division of absorption lines. The dark lines which appear in a continuous spectrum, if a beam of white light traverses an absorbing flame, are divided and polarized under the influence of magnetic forces in exactly the same way as the emission lines. This correspondence between emission and absorption was shown to exist already in some of the first experiments on the subject by one of the present authors. Our knowledge of emission spectra under magnetic influence has since been extended considerably. The experimental study however of the inverse effect i. e. the magnetic division of absorption lines has less advanced.