THE ENTRANCE OF WATER INTO CUT LEAFY SHOOTS UNDER CONDITIONS WHICH PREVENT TRANSPIRATION

by

F. VAN DER PAAUW.

CONTENTS.

I. Introduction.

Dixon (2) communicated experiments, indicating that cut, leafy shoots, submersed in water, continue to take up water through the cut surface, notwithstanding the fact that transpiration is prevented. This was concluded from the ascent of dyes in the xylem of the shoot.

Bose (1) demonstrated that shoots continue to take up water for several hours after transpiration has been prevented by covering them with a thin layer of vaseline or by placing them in air saturated with water.

These investigators believe that the entrance of water under these circumstances can be explained by assuming that the cells of the leaf actively give off water and continue to do so after transpiration has stopped. The water deficit of the cells is covered by water from the xylem.

Schmucker (7) also, as a result of his narcosis experiments, came to the conclusion that the mesophyll cells actively influence the movement of the sapstream, in particular when transpiration is obstructed.

Smith, Dustman and Shull (9), using Dixon's method, found that this entrance of water takes place during a short period only, after which it ceases completely. The intake during the first hours after submersion has to be regarded as the suction of a not yet fully saturated shoot.

Sen and Blackman (8), who investigated the cause of impregnation with water of submersed leaves, could also observe only a slight intake of water through the petiole.

The following communication contains the results of potometer experiments on this same problem. In contradiction to the results obtained by Smith, Dustman and Shull (9) it will be shown that the entrance of water into submersed shoots can continue for several days and that a considerable intensity can be reached. Experiments with dyes are probably less suitable to demonstrate a slowly moving sapstream than potometer experiments, since dye substance will be absorbed while in contact with the walls during a long period, thus reducing the concentration of the dye solution, which results in a loss of staining effect at some distance from the surface of the cut.

II. Experimental observations on the intake of water by submersed shoots.

The cut shoots were cut again under water, fixed onto the potometer, and placed in inversed position in a cylindrical jar containing about 3 liters of ordinary tapwater. The water in the potometer was brought to the same level as that in the glass jar. The potometer consisted principally of a horizontal glass tube of 1 or 2 mm bore.

The experiments were carried out at a distance of about 1 meter behind a window facing north. The temperature of the laboratory room varied but little.

The following specimens were experimented upon: Shoots of *Eucalyptus globulus*, (juvenile stage with opposite, sessile leaves), *E. citriodora, E. resinifera, Nerium Oleander, Sparmannia africana, Swietenia Mahagoni, Cinnamonum pedunculatum, Prunus Laurocerasus, Ilex Aquifolium, Thuja occidentalis, Taxus baccata, Pinus maritima, Canavallia ensiformis* (young plant with two primary leaves), *Acer platanoides, Tilia vulgaris, Populus pyramidalis* and leaves of *Aspidistra elatior* and *Pteris tremula.*

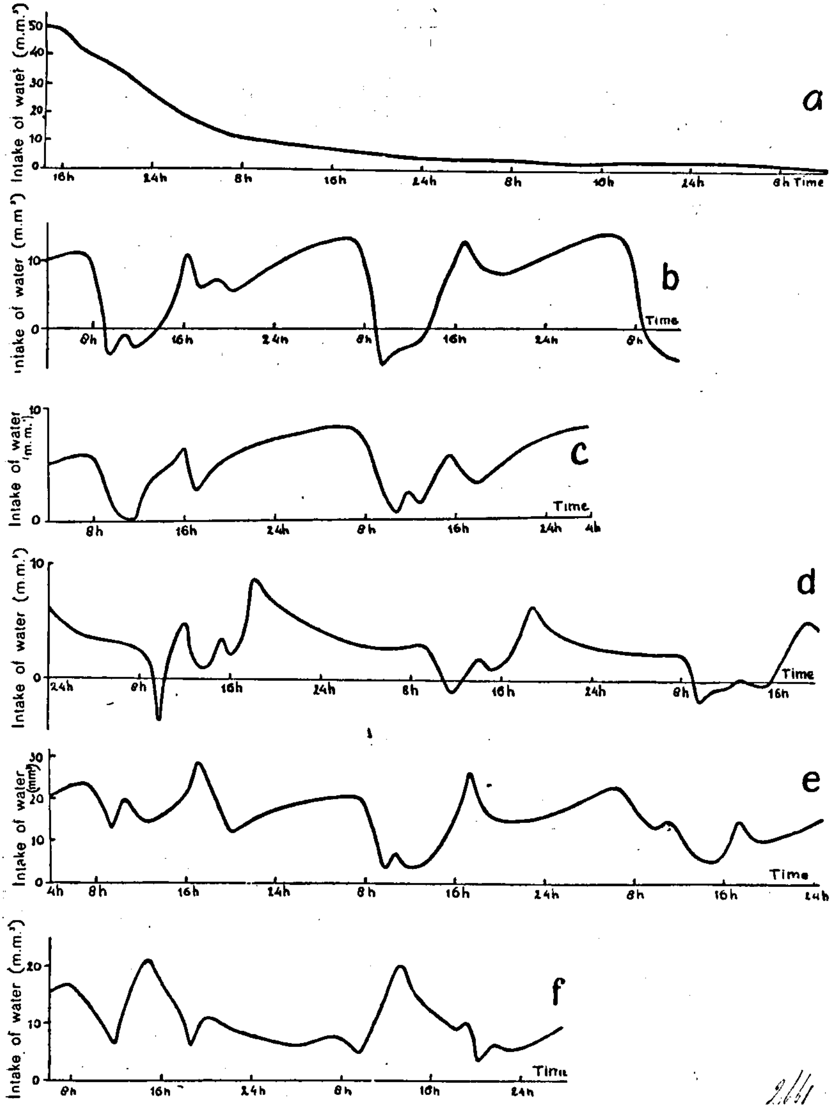
Only little water entered into the leaf of *Pteris,* (date 31-1-33) [1]. The intake of water by *Acer platanoides* was strong at first, but diminished quickly after some hours (14-6-33). All other plants showed a considerable intake of water, which lasted for days.

We may distinguish here two kinds of behaviour with respect to the intake of water. Firstly, though the experiment lasted for several days, the intake decreases in intensity and gradually approaches zero, illustrated by *Nerium Oleander* (14-12-32), fig. 1a, and *Pinus maritima* (7-3-33), and found once only in a shoot of *Eucalyptus resinifera* (28-11-32). Secondly, the intake of water fluctuates, usually showing a daily rhythm which varies between a diurnal small intake and a nocturnal large one, e.g. *Eucalyptus globulus* (18-12-33), fig. 1b. The entrance of water shows a maximum rate at about 1 to 2 hours after sunset, followed by a decrease, while a few hours afterwards an increase takes place again. On the whole, each plant shows its own special type of behaviour. Experiments with *Eucalyptus globulus* were performed at various times in the course of a year, with always about the same result as illustrated in fig. 1b. It could repeatedly be seen that water was forced back into the potometer during exposure to light.

The rate of intake of water proved to be considerable, as shown by an experiment (16-11-32), selected at random for calculation. This experiment was performed with a shoot of *E. globulus*, carrying 12 leaves. Its weight amounted to 19 g and the intake of

---

[1] The date indicates the day on which the experiment was started.



2661

Fig. 1. Intake of water through the cut surface of submersed shoots in daylight expressed in mm³/hr.

a. *Nerium Oleander*, weight 15.6 g, 16 leaves, date 14-2-33, temp. 16°.7—18°.8.
b. *Eucalyptus globulus*, date 18-12-33, temp. 15°.6—18°.1.
c. *Eucalyptus citriodora*, weight 9.5 g, leaves separately 6.3 g, date 12-2-33, temp. 14°.1—18°.1.
d. *Aspidistra elatior* (leaf), length of lamina 40 cm, weight 6.9 g, date 12-2-33, temp. 13°.8—16°.8.
e. *Ilex Aquifolium*, weight 44.6 g, leaves separately 27.3 g, date 7-3-33, temp. 14°.8—17°.1.
f. *Thuja occidentalis*, weight 11.5 g, date 15-3-33, temp. 15°.5—18°.3.

water during 24 hrs. amounted to 1212 mm^3, at a temperature of 16—18° C., the largest hourly intake being 69 mm^3 (64 hrs. after submersion).

After concluding the experiment the total area of the cross sections of the wood-vessels was measured microscopically. Assuming that all vessels are of equal importance for the transport of water, and using the maximum hourly intake, the calculated velocity of the sapstream would amount to 18 cm/hr. In another instance 15 cm/hr was calculated in this manner. Since probably not all vessels function as organs for transport, the real maximum velocity is presumably greater.

Some other examples of rhythmic intake of water are shown in fig. 1. Fig. 1c illustrates the entrance of water into a shoot of *Eucalyptus citriodora* (21-2-33). At the conclusion of the experiment the weight amounted to 9.5 g after drying with filter paper; the shoot carrying 20 leaves of normal size and 4 smaller ones. The leaves together weighed 6.3 g; 132 mm^3 water were taken in during 24 hrs.

Another example is a leaf of *Aspidistra elatior*, fig. 1d. (12-2-33). The lamina, 40 cm in length, finally weighed 6.9 g. The intake of water in 24 hrs. amounted to 83 mm^3.

Fig. 1e illustrates the intake of water of a holly shoot. (7-3-33). Considerable fluctuation was noted here. The final weight of the shoot was 44.6 g, that of the leaves 27.3 g, and the intake of water in 24 hrs. was 385 mm^3.

A pronounced daily rhythm was observed in *Eucalyptus resinifera* (5-12-32 and 21-12-32). The diverging result of another experiment with this plant has been mentioned above.

Distinct rhythms, though less pronounced, were observed with *Swietenia Mahagoni* (19-2-33) and *Populus pyramidalis* (19-6-33).

A very distinct difference between day- and night- intake was shown by *Canavallia ensiformis* (25-4-33). After 36 hrs., however, only little water was entering.

Taxus baccata (14-3-33) showed, the day following the submersion, a minimum of intake while exposed to light, followed by a maximum in darkness, after which the fluctuations became irregular.

A rather irregular rhythm was shown by *Cinnamonum pedunculatum* (25-3-33), though the lowest minimum fell during daytime and the highest maximum at night. Another very faint rhythm was found in *Prunus Laurocerasus* (15-3-33).

Thuja occidentalis behaved quite differently (15-3-33), showing during the daytime two minima, with a very high maximum in between. During the night very little water entered, even less than during the day. This is illustrated by fig. 1f. In two later experiments (14-6-33 and 10-4-33 resp.) the behaviour of *Thuja* was quite different; during the night the intake was twice that of the daytime, a minimum was observed about noon, and a maximum about sunset.

Sparmannia africana (16-1-33) showed irregular fluctuations, and only slight difference could be observed between day and night.

In *Tilia vulgaris* (14-6-33) the intake was rhythmic only in that a slight increase could be observed in the morning; otherwise this plant should be placed in the first category of plants on account of its gradually decreasing intake of water.

III. Experimental observations on the intake of water by shoots, the transpiration of which is prevented in some other manner.

The following experiments prove that the entrance of water occurs also when transpiration is prevented by other means. In experiment 14 (10-1-33) with *E. globulus* (a shoot with 12 leaves, final weight 16.5 g) the ventral side of the leaves was covered with water-free vaseline and fixed onto the potometer; in experiment 15 (10-1-33) with a shoot with 8 small leaves, final weight 6.3 g, the dorsal side was covered as well, likewise in experiment 18 (31-1-33) with a shoot with 9 leaves. Expts. 14 and 15, fig. 2, show a complete agreement; the observed rhythm is exactly that of the submersed leaves, viz. a minimum in the forenoon and a maximum after sunset. The intake, however, decreases gradually overnight and no second maximum can be noted. In expt. 14 the branch was submersed during the second day without this influencing the character of the intake. In expt. 18 the rhythm was faint.

In two following experiments transpiration was prevented by placing the shoot under a bell-jar which had its base in a basin of water. The atmosphere inside the jar was kept moist with aid of strips of filterpaper against the inside wall. In the first experiment (31-1-33) with a shoot of 10 leaves, final weight 16.1 g, the results were the same as with a greased shoot: a distinct minimum in light, and a maximum after sunset. The difference was slight, however, and the rhythm faint. In the second experiment (16-1-33) with a shoot having 10 large leaves, final weight 16.7 g, the result was the opposite: the maximum intake of water took place at about noon. The intake was of the same nature as that of a shoot

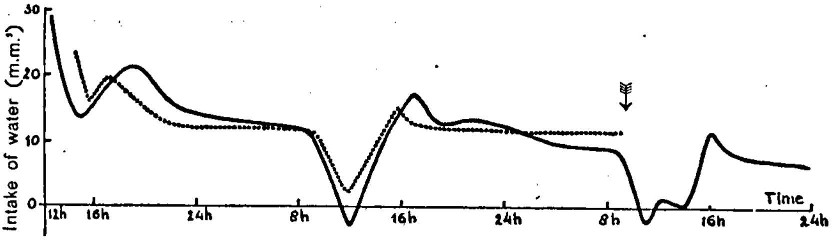


Fig. 2. Intake of water by shoots of *Eucalyptus globulus*, in mm³/hr.
a. drawn line, ventral side of leaves covered with vaseline. ↧ Shoot submersed in water. Weight 16.5 g, 12 leaves.
b. dotted line, both sides of leaves covered with vaseline. Weight 6.3 g, 8 leaves.
Date of both experiments 10-1-33.

in dry air. The maximum was not pronounced and it is probable that in this experiment transpiration had not stopped altogether.

IV. On the causes of the entrance of water.

Having demonstrated in the foregoing pages that the intake of water by leafy shoots takes place even after transpiration has been stopped, the causes of this phenomenon may now be discussed.

The following experiments were all performed with *E. globulus*.

a. *The daily periodicity is caused by the daily alternation of light and dark.*

From the foregoing experiments, all performed in normal light, it appears that the daily rhythm is very probably due to the periodic changes of light and dark. Experiments with artificial light support this point of view. In the dark much water is taken up, while in the light little water enters or passes out throug the cut surface. With light during the night and darkness during the day-time there is a corresponding change in the direction of the sap-stream. No rhythm could be observed when the plants were kept in the dark for a considerable period (61 hrs.); the intake decreased gradually. After exposure to normal daylight the rhythm reappeared immediately.

In connection with the experiments with artificial light it should be mentioned that an approximately equally strong effect could be obtained with red light of a wave length of > 600 mμ in place of white light. The red light was obtained by means of a Wratten filter 71 Aβ.

b. *The water which has entered through the cut surface stays in the shoot.*

The following questions arise: 1. Do the shoots show an increase in weight as a result of the entrance of water through the cut surface, or is this water again given off by the leaves? 2. Do the submersed leaves take up water through their outer surface?

A small shoot is weighed quickly, fixed to the potometer and submersed. After this has been kept in the dark for some hours the quantity of water taken up through the cut surface is read from the potometer tube. The shoot is then dried superficially between filterpaper and weighed again. The difference between the second and the first weighing gives the gain or loss of water. The difference between the weight-difference and the potometer reading gives the gain (or loss) of water by the leaves.

After the second weighing the shoot is again attached to the potometer and submersed, but this time is kept in the light. Thus one shoot is used several times, alternately in dark and light. The results of two experiments (21-2-33 and 7-3-33) are tabulated below. The first shoot had 6 leaves and weighed at first 6.335 g. The stem without leaves had a final weight of 1.934 g. A shoot with 7 small leaves was used in the second experiment, original weight 6.155 g.

Table I.

Gain and loss of water through the cut surface and the leaf-surface in periods of light and dark.

Conditions		Length of period in hrs.		Intake in mg through cut surface		Intake in mg through leaf surface		Total intake of water in mg	
dark		11		47		13		60	
	light		7		31		19		50
dark		16¼		124		1		125	
	light		7½		2		—8		—6
dark		16		331		19		350	
	light		7¾		—14		—27		—41
dark		20¼		493		1		494	
dark	**light**	63½	22¼	995	19	34	—16	1029	3
	light		7½		30		—15		15
dark		16		311		32		343	
	light		7¾		6		—22		—16
dark		15¾		291		39		330	
	light		7½		13		—18		—5
dark		20½		165		— 10		155	
dark	**light**	52¼	22¾	767	49	61	—55	828	—6

Taking into consideration the fact that during the weighing the shoots rapidly lose weight and that a slight error is thus introduced, the result is convincing. Water enters nearly exclusively in the dark. The water is taken up principally through the cut surface of the shoot; the intake of water by the leaves themselves is always slight, whereas no water is given off. In the light usually a small quantity enters through the cut surface, while the leaves give off a very small quantity. We may state therefore that the water which has entered through the cut surface in the dark is not given off but stays in the shoot.

c. *The water taken up passes into the intercellular spaces.*

It was of interest to find out whether the water taken up entered the cells of the leaves or that it passes into the intercellular spaces. For this purpose the length, width, and thickness of the leaves were measured before and after a prolonged stay under water, under alternating light and dark conditions. In order to have fully water-saturated cells with the very first weighing, the leaves were submersed a few hours beforehand. The results of a few experiments proved that the dimensions of the leaves do not change to any appreciable extent. For example:

before the expt: length 149.4, width 75.0, thickness 0.250 mm
after 3 days : „ 149.4, „ 75.1, „ 0.250 mm.

Hence the volume did not change. On the other hand the weight increased from 2010 mg to 2413 mg, i.e. 20%.

We may conclude from this that the cells of the leaf do not take up any water, but that a volume equal to that of the water which has entered through the cut surface passes into the intercellular spaces.

This conclusion may be arrived at in another way. The volume of the intercellular system can be measured by the method of Nius (3). According to this method small disks of tissue are cut out, weighed, impregnated with water and re-weighed. The relation between the weight-difference and the weight of the impregnated disks approximately equals the relation between the air-volume and the total volume of the leaf.

Applying this method, and using 4 disks of a diameter of 5 mm for each determination, using also a torsion balance to obtain immediate weighing results, it was found that the intercellular air-volume had decreased considerable after every period in the dark, while hardly any change was noticeable after a period of light; in the latter case even a slight increase could be observed at times. The results of one experiment (11-9-33) are illustrative. The volume of air after a dark period was 19% of the total volume of the leaf. After 7 hours exposure to light it amounted to 20%, after another 16¾ hours of darkness only 8%. This fully supports our conclusion: The water that is taken up by the leaves passes into the intercellular spaces.

d. *The entrance of water can take place only if the intercellular spaces are filled with air.*

In connection with the hypothesis of Dixon and Bose mentioned in the introduction, another question arises, namely, is it essential that the intercellular system into which the water passes contains air, or can the sapstream take place equally well if the air is replaced by water? According to the hypothesis of Dixon and Bose the latter will be true. From our experiment it appeared, however, that the entrance of water can take place only in case intercellular air-spaces do exist inside the leaves.

Two very similar shoots were used for comparison. One was treated in the usual manner, and the other one previously impregnated with water. This was kept under water in vacuo, so that the air might escape. When air was let in again water was forced into the intercellulars. It should be realized that it is nearly impossible to remove all air completely in this manner.

Fig. 3 shows the results of the experiment. Both shoots take up water, but little water enters into the impregnated shoot. The loss during the daytime and the intake overnight approximately counterbalance.

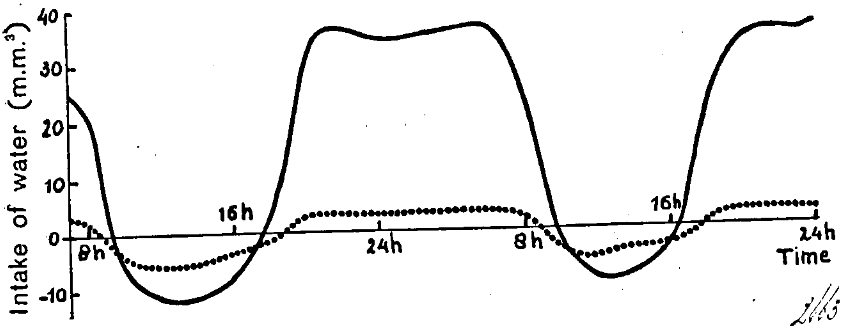


Fig. 3. The intake of water by shoots of *Eucalyptus globulus* in mm^3/hr.
a. drawn line: normal submersed shoot, with air in the intercellulars.
b. dotted line: shoot impregnated with water.
Normal daylight, date 18-9-33.

e. *According to Sen and Blackman a pressure deficit is developed during respiration, which causes the injection with water.*

The investigations of Sen and Blackman (8) on the injection of water into submersed leaves have already been mentioned. These investigators conclude that a pressure deficit in the intercellulars causes water to enter into the leaves. The deficit develops during respiration, since gaseous oxygen is absorbed in the dark and no equivalent volume of gaseous carbondioxide can develop because the latter gas is more soluble in water than oxygen. In the light the effect of photosynthesis outweighs that of respiration and no water enters.

Their results agree very well with ours, with the difference that they did not observe any noticeable entrance of water through the cut surface and therefore they could reject an explanation according to the hypothesis of Dixon and Bose. How far the theory of Sen and Blackman holds for our experiments will now be discussed.

f. *Small pressure-changes in the intercellular spaces cause a movement of the sap.*

1. *Influence of heat.* Sudden heating may develop a pressure above the normal in the intercellular spaces. In a few experiments hot water was added to the water in the jar. The water was stirred all the time by an air-current bubbling through the water and thorough mixing took little time. As a result of the rise in temperature water suddenly passed out through the cut surface. An experiment which was performed in the dark illustrates this. The temperature rose from 22° to 26°.2 C. Before the hot water was added, water entered at a rate of 24 mm^3/ hr. During the first 5 minutes after the addition of hot water 4 mm^3 passed out, i.e. an average speed of 48 mm^3/hr. During the next 5 minutes water neither entered nor passed out, and thereafter the rate of intake rapidly increased to the original one.

Immediately after the hot water was added the rate of transport was at its maximum, certainly appreciably more than 48 mm^3/hr., presumably about 80 mm^3/hr. By raising the temperature by 4°.2 the pressure in the intercellulars could at most have increased by 4.2/273 atm. = 11.7 mm Hg. A slight overpressure thus can force the water in opposite direction through the cut surface.

The same branch had been used in a previous experiment in the light. Here water passed out at a maximum rate of 6—7 mm^3/hr.

Estimating the maximum rate of the sapstream in the heated shoot at 80 mm³/hr., and assuming the temperature of the air in the intercellulars at that time to already have been raised by 4°.2, — which is almost impossible within such a short time —, we note that for every mm Hg overpressure 80/11.7 mm³ water is pressed out. *An overpressure of 1 mm in the intercellulars suffices to explain the fact that in the light water passes out through the cut surface at a rate of 6—7 mm³/hr, as observed.*

2. *Influence of cold.* Cooling, effected by the addition of small pieces of ice, causes a pressure deficit in the intercellulars. This is illustrated by an experiment performed in the light. Before cooling, water entered at a rate of 8.5 mm³/hr. During the previous dark period a maximum intake of 49.2 mm³/hr. had been observed. The temperature fell from 23°.2 to 18°.2. During the first 7 minutes after the addition of ice 15.8 mm³ entered, or 135 mm³/hr. During the following 10 minutes 8.0 mm³ entered. The temperature in the meantime had been rising slowly to 18°.7. Hereafter the intake decreased rapidly.

A drop in temperature of 5° can cause a pressure drop of at most 13.9 mm Hg. As the ice melted slowly the average decrease in temperature during the first 7 minutes was certainly less than 5°; yet the water entered at a maximum rate, estimated at 200 mm³/hr. at least. *A pressure deficit of less than 49.2/200 × 13.9 mm = 3.5 mm therefore suffices to explain the maximum intake of water observed in the dark.*

3. *Influence of Hydrogen.* The effect of the exchange of gas on the movement of water in the shoot could also be demonstrated by passing hydrogen through the water. This readily diffusable gas must cause an overpressure inside the leaves. The experiment was performed in daylight at 2 p.m. The original intake of water amounted to 2 mm³/hr. In the forenoon a maximum loss of 15.5 mm³/hr was observed. When the hydrogen was passed through the sapstream during the first 7 minutes was nil, in the next period 6 mm³ passed out, in the following 10 minutes 19.5 mm³ and thereafter 22.5 mm³, i.e. 135 mm³/hr.

The movement of the sapstream was reversed practically instantaneously when the hydrogen current was replaced by one of air; after 33 minutes a maximum rate of 83 mm³/hr. was reached.

4. *Influence of submersion in boiled water.* The water in which a shoot was submersed at night was replaced by boiled water of the same temperature. It was to be expected that the air in the intercellulars would dissolve and cause a pressure deficit.

The intake at first was 58 mm³/hr. and immediately increased to 112 mm^3/hr. within the first 30 minutes. A still further increase followed. From 1—3 hours after the exchange the average amounted to 157 mm^3/hr.

g. *The intake of water and the gas exchange are closely related.*

If the entrance of water into submersed shoots can be explained wholly or for the greater part by the theory of Sen and Blackman, we may expect a close relation between the intake of water and the exchange of gas. These processes were therefore investigated simultaneously.

The intake of water by a shoot was measured at constant temperature in the dark and in artificial light. The shoot was placed in a dilute solution of $NaHCO_3$ and Na_2CO_3, so that more CO_2 was available than in ordinary tapwater. This did not affect the normal behaviour.

One of the leaves of this shoot was cut off beforehand and the wound covered with lanoline. The exchange of gas of this leaf was studied with the apparatus of van der Paauw (4,5), in which it was kept submersed in the same carbonate-bicarbonate solution. The CO_2 pressure in this apparatus is constant; changes in the oxygen volume only are measured. The leaf and the shoot were exposed to light of about equal intensity while at the same temperature. (19°.6).

The results are shown in fig. 4. There is a close parallelism between intake of water and gas-exchange. Shortly after exposure to light photosynthesis is at its maximum and the intake of water at its minimum. Photosynthesis gradually decreases, while the water intake increases. Shortly after the light was switched off respiration and intake of water showed a maximum and thereafter gradually decreased again.

No water passed out during the experiment; this, however, does not always happen. Not all leaves were equally exposed to light. Furthermore, in the carbonate-bicarbonate solution, unlike tapwater, carbon-dioxide when used up was supplied by the bicarbonate, and very likely gaseous carbondioxide was present all the time in the intercellulars. Assimilation of gaseous carbondioxide does not cause a change in pressure, because an equivalent quantity of gaseous oxygen is produced. When, on the contrary, shoots are submersed in tapwater, the carbondioxide which is present in the mesophyll cells, either dissolved or chemically bound, will first be used up, and gaseous oxygen is evolved, which causes an overpressure in the intercellulars.

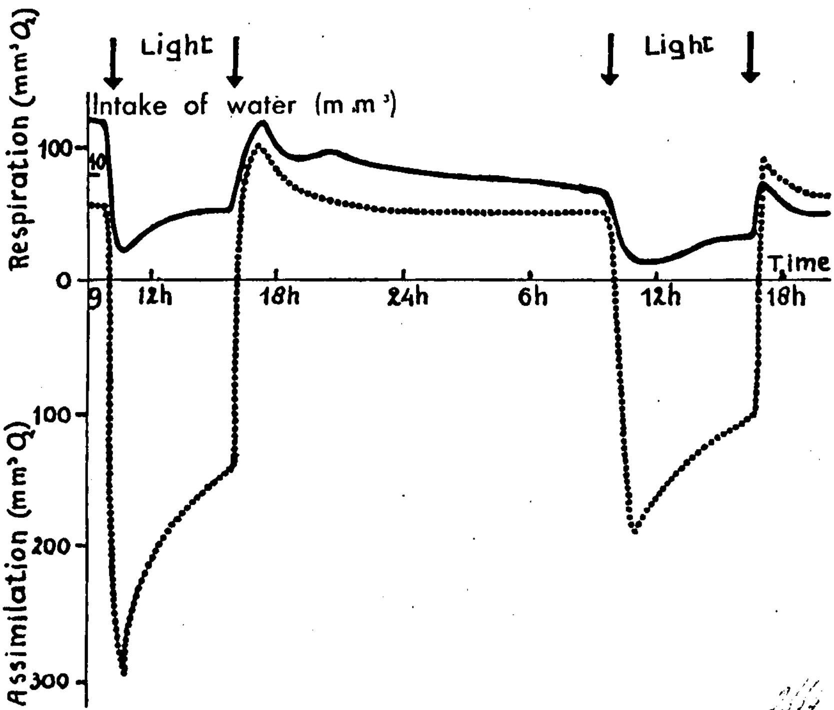


Fig. 4. Intake of water and gas-exchange of *Eucalyptus globulus.*
a. drawn line: intake of water by submersed shoots in mm^3/hr.
b. dotted line: gas-exchange of cut leaf of same branch in mm^3 O_2/hr.
Artificial light, constant temp. 19°.6, date 7-3-34.

V. Discussion.

We may now discuss the results obtained with *Eucalyptus globulus.* It has been demonstrated that the entrance of water through the cut surface of a submersed shoot depends on the simultaneous entrance of water into the intercellulars. In this way a sapstream originates. This movement can take place only in case the intercellulars contain air. It is very probable that the passing of water into the intercellulars is always caused by a slight pressure deficit inside the leaf, due to respiration. In support of this point of view we cite the observed fact that no water enters when CO_2 is assimilated by the plant in the light. All variations in the exchange of gases are followed by corresponding variations in the intake of water. Pressure deficits of probably much less than 3.5 mm Hg and overpressures of at most 1 mm Hg are sufficient to explain the variations in sap movement.

It is very probable that such pressure variations actually do occur, considering the relatively high intensity of the gas-exchange in the plant. This will be illustrated by the results of an experiment with a leaf weighing 452 mg. We may safely assume that after the leaf had been under water for several days the air volume did not exceed 20% of the total volume of the leaf. The leaf then contained $\frac{1}{4} \times 452$ mm^3 $= 113$ mm^3 of air. The highest respiratory intensity of this leaf was found to be 98 mm^3/hr. A large part of this quantity of oxygen must have been obtained from the air in the intercellulars. The stomata were closed (since only a negligible quantity of water was taken up by the leaves), allowing a pressure deficit to develop readily.

Vaseline-covered shoots and those in saturated air take up water in the dark like submersed leaves. The above explanation for submersed leaves probably is true for these also; the pressure deficit develops because carbondioxide, which is generated during respiration, remains in the plant — at least partially — in a dissolved state.

Sen and Blackman (8) were the first to state the opinion that the leaves become impregnated with water as a result of a pressure deficit caused by respiration. The difference between their results and the ones of the writer is principally that they observed the entrance of water through the leaf-surface, while in the latter experiments nearly all water entered through the cut surface of the shoot. Continuous entrance of water through the cut surface could also be explained by a regular active excretion of water by the cells of the leaf, as maintained by Dixon and Bose. Though Sen and Blackman and the writer have not disproved such an activity of the leaf, it is of subordinate importance for the explanation of the entrance of water into cut shoots, under conditions which prevent transpiration. This has been sufficiently demonstrated as dependent largely or completely on changes of air-pressure.

The experiments with *Eucalyptus globulus* do not prove that in all other plants the movement of sap is caused by one and the same factor. It is very probable, however, for all those plants which show a daily periodicity in the intake of water.

Where this periodicity was not observed but the sapstream continued for several days we may assume that photosynthesis was inhibited by unfavourable conditions, while the respiration rate was not affected. Another explanation is that the shoots were not fully saturated with water (Smith, Dustman and Shull).

This, however, is not very probable as the shoots have been kept submersed for long periods. The explanation of Dixon and Bose might be used here, although it does not appear very probable.

The result of expt. 42 with *Thuja occidentalis* (fig. 1 f) is not yet explained and does not fit in with the others. Here the intake of water reached a maximum in the light, though preceded by a minimum which was not very pronounced. A solution of this problem might be arrived at if we could assume that in this case the rate of photosynthesis had been reduced and the respiration rate accelerated under the influence of light. As has been demonstrated by the writer (4), respiration can sometimes be intensified by light [1]. Two other experiments with *Thuja* gave results more in agreement with the normal behaviour.

It is possible that variations in the air-pressure in the intercellulars do not constitute the only cause of the entrance of water. It is probable that the force of suction shows an increase due to the formation of sugars during photosynthesis. More water consequently would enter during the daytime. A long period of exposure to light might in this way be at least partly the cause of the increase in intake of water, but probably this is of little importance.

The air-pressure in the intercellulars may vary also because of a change in the surrounding water, as was noted in the experiments with boiled water and with a hydrogen current bubbling through the water. The oxygen concentration of the water decreases after a long period of respiration. The absorbed oxygen is replaced at a lesser rate and the pressure deficit increases, which again leads to an increase in the rate of absorption. It has been noted frequently in the experiments that the intake of water increased after a long period in the dark (fig. 16, 1c, 1e). This fact can not be explained by assuming an increase in respiration intensity, since no increase could be observed.

If the entrance of water is really caused by pressure changes, it must be possible to draw conclusions as to the rate of gasexchange from the intensity of the sapstream.

From the potometer experiments with *Eucalyptus globulus* it may be deduced that in submersed shoots:

a. respiration shows a maximum shortly after sunset, and decreases thereafter,

b. on exposure to light carbondioxide assimilation shows a

---

[1] Parija and Saran (6) recently stated that light stimulates respiration in starving leaves.

maximum at the beginning and decreases thereafter, probably due to the exhaustion of the CO_2 supply.

Measurements of the exchange of gas support these views.

The cited case where intake of water increased during a long dark period demonstrates that great care must be taken when drawing conclusions, since many factors are involved that may lead one astray.

VI. Summary.

Potometer experiments prove that cut leafy shoots, when transpiration is prevented by submersion, a covering with vaseline, or their being placed in saturated moist air, continue for days to take up water through the cut surface, which is contradictory to the observations of Smith, Dustman and Shull.

Frequently the shoots, when exposed to the daily changes of light and dark, show a rhythmical intake of water, as especially observed in *Eucalyptus globulus.* In the dark the intake is large, in the light it is reduced, or water may even pass out.

The entrance of water is caused by a pressure deficit in the intercellular spaces in the leaves, as a result of respiration (Theory of Sen and Blackman). This pressure deficit develops as a result of the difference in solubility of absorbed oxygen and produced carbondioxide, which leads to a reduced quantity of gas. The water which has entered through the cut surface passes into the intercellular spaces.

Photosynthesis takes place in the light and the deficit disappears; the sapstream practically stops.

Only little water directly enters or passes out through the leaf surface.

The theory of Dixon and Bose that the sapstream of *submersed* shoots is caused by the activity of the leaf cells excreting water, is not confirmed.

These investigations were carried out in the Laboratory of Technical Botany of the Technical University at Delft, Netherlands. I should like to express my sincere thanks to Professor Dr. G. van Iterson for his interest and helpful criticisms.

LITERATURE.

1. Bose J. C.: Physiology of the Ascent of Sap. London 1923.
2. Dixon H. H.: Transpiration and the Ascent of Sap in Plants. London, 1914.

3. Nius E.: Untersuchungen über den Einflusz des Interzellularvolumens und der Öffnungsweite der Stomata auf die Luftwegigkeit der Laubblätter. Jahrb. f. wiss. Bot. *74*, 33 (1931).
4. Paauw F. van der: The indirect Action of external Factors on Photosynthesis. Rec. trav. bot. néerl. *29*, 497 (1932).
5. Paauw F. van der: Zur Methodik der Kohlensäureassimilationsmessung. Planta *22*, 393 (1934).
6. Parya P. and A. B. Saran: The Effect of Light on the Respiration of Starved Leaves. Ann. Bot. *48*, 347 (1934).
7. Schmucker Th.: Uber den Einflusz narkotischer Stoffe auf Transpiration und Wasserleitung. Jahrb. f. wiss. Bot. *68*, 771 (1928).
8. Sen P. K. and V. H. Blackman: On the Conditions leading to the Injection with Water of Leaves submerged in Water. Ann. Bot. *47*, 663 (1933).
9. Smith F., Dustman and Ch. A. Shull: Ascent of Sap in Plants. Bot. Gaz. *91*, 395 (1931).