Huygens Institute - Royal Netherlands Academy of Arts and Sciences (KNAW)

*Citation:*

Verschaffelt, E., The mechanism of the absorption of water by the seeds of the cucurbitaceae, in: KNAW, Proceedings, 13 I, 1910, Amsterdam, 1910, pp. 542-550

This PDF was made on 24 September 2010, from the 'Digital Library' of the Dutch History of Science Web Center (www.dwc.knaw.nl) > 'Digital Library > Proceedings of the Royal Netherlands Academy of Arts and Sciences (KNAW), http://www.digitallibrary.nl'

for leaves of *Rumex*, *Oxyria* and *Rheum*, yet they also eat a little from the leaves and flowers of *Begonia* (*manicata* Cels., *tuberosa* Lam., *ricinifolia* A. Dietr. and others). Leaves of *Oxalis* (*floribunda* Lehm., *Deppei* Lodd.) were on the other hand always refused. It should however be stated that on account of the far advanced season the latter experiments could only be done with the imago, which also rejected species of *Portulaca*, *Celosia*, and *Sedum*, and only eat slightly from *Polygonum* (*cuspidatum* Sieb et Zucc. for instance).

Also in the case of this insect direct proof can be adduced that the occurrence of a definite substance in the plants determines whether they shall or shall not serve as food. Leaves of *Lathyrus sylvestris* are never eaten fresh, but it was repeatedly observed that the beetles gnawed at them when for some time they had been immersed in a normal solution of oxalic acid. In the absence of material, through the disappearance of the last beetles, it was impossible to investigate further whether they are attracted by the acid reaction in general or whether they are exclusively dependent on oxalic acid. Next summer I hope to be able to decide this point and also to trace the behaviour of *Gastroidea* towards plants in which other organic acids (malic or citric) occur in considerable concentration.

**Botany.** — "*The mechanism of the absorption of water by the seeds of the Cucurbitaceae*". By Prof. ED. VERSCHAFFELT. (Communicated by Prof. J. W. MOLL.)

The seeds of the *Cucurbitaceae* are in general distinguished by the rapidity with which they take up water. This property is strongly developed in the ordinary cultivated varieties of the species *Cucurbita Pepo* L. and *Cucurbita maxima* Duch., our gourds and pumpkins. In the accompanying figure (fig. 1) one curve represents the increase in weight, during the first hours, of the air-dry seed of a variety of large yellow gourd, "Grosser gelber Zentner", when placed in water, and the other curve shows the same for a variety of *Vicia Faba* L., the Mazagan bean. The determination was made by taking the seeds out of water from time to time, drying them superficially with filterpaper, and weighing them. The estimation was made with a small number of seeds, so that, on account of the individual differences, a repetition of the experiment gives curves which do not agree completely with the one reproduced here. It is, however, easy to convince one's self that the general shape of the curv

remains unaltered, and that the remarkable difference between the curves of pumpkin-seeds and beans always appears.

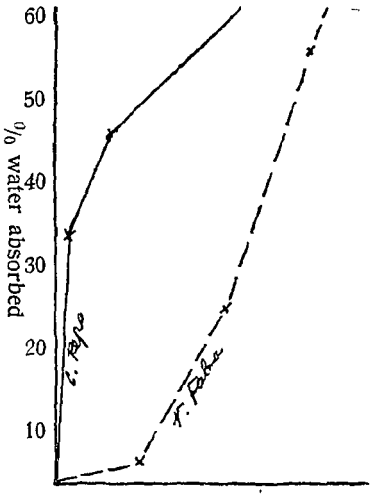

Fig. 1.

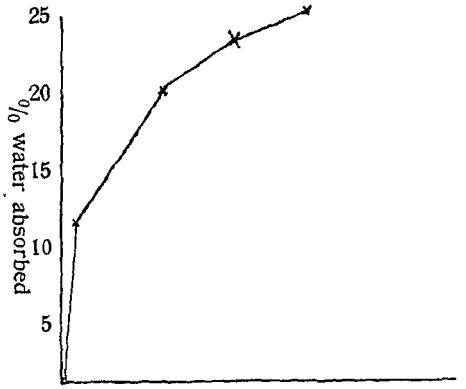

Fig. 2.

While therefore in *Vicia Faba* the increase of weight is at first a very gradual one, it begins in *Cucurbita Pepo* so rapidly, that already at the end of the first hour water to the extent of 34% of the dry weight was taken up, in the experiment of fig. 1; this is a third of the total quantity which the seed was found able to retain.

Even in the first few minutes the absorption is specially marked. It is represented graphically (fig. 2) for the same yellow variety of gourd ("gelber Zentner"). As will be seen, in twenty minutes water was taken up to the extent of more than 25% of the air-dry weight of the seed.

It is natural to hold the seed-coat responsible for such a rapid and considerable absorption of water, and we find indeed that the seed-coat of *Cucurbita*, in contradistinction to that of many other seeds, especially that of *Vicia*, is immediately wetted. If a drop of water be placed on the seed, one recognises by the darkening of the white surface round about the drop of water, that the liquid has been sucked up by the tissue, as if by blotting paper.

This is therefore the cause of the *rapid* imbibition. But that such a *large* quantity of water is taken up in a short time, is also a result of the properties of the seed coat, as is evident from a comparison between the absorption of water by a pumpkin-seed as a whole and that of the interior of the seed, freed from its hard testa. The

former can easily be dissected out and then consists principally of the germ surrounded by a thin greyish green membrane, which is firmly connected with it.

Such seeds of the large yellow gourd, after being deprived of their testa, absorbed:

| during | 1h | 4h | 24h | 50h |
|---|---|---|---|---|
| water | 8% | 20% | 36% | 42% |

whereas in the same times, the entire seed took up:

| | | | |
|---|---|---|---|
| 34% | 48% | 86% | 96% |

and afterwards this considerable difference was maintained. If we consider that the testa only constitutes $^1/_6$ to $^1/_5$ of the weight of the seed, it follows from this proportion that the testa must be very rich in water. Direct determinations indeed showed that a testa, separated off and placed in water, finally underwent an increase in weight of 194%. This was in the variety Courge gaufrée of *Cucurbita Pepo*, a variety which takes up less water than the large yellow gourd ("gelber Zentner") previously experimented on, where the increase in the testa was 228%. On comparison of the absolute quantity of water taken up by the seed freed from its testa, with that absorbed by the testa alone, the latter quantity is found to be the larger. In Courge gaufrée, where some seeds took up on the average 0.25 gram of water, the testa of a single seed contained on the average 0.16 gram of water.

We will now consider by what peculiarities of the anatomical structure the testa in Cucurbita is able to hold so much water. For this purpose the accurate description by F. v. HÖHNEL (*Sitzungsber. K. Akad. Wiss. Wien*, Math. Nat. Cl. Bd. 73 Abt. I. 1876. p. 297) can be used. As this paper and the plates attached to it can be referred to for the details, here only the anatomical arrangements which may have significance with regard to absorption, will be dealt with.

The thin transparent pellicule of which single pieces are frequently found still attached to the seed and which represents the inner layer of the fruit wall can here remain unnoticed, because it is so easily loosened and is without importance in the imbibition of the seed.

On the other hand the structure of the epidermis is in a high degree adapted to increase the absorption of water. This layer consists of elongated prismatic cells without cuticle or contents, whose walls swell very greatly in water, so that according to v. HÖHNEL's measurements the length of these cells in a dry condition is only 30 $\mu$ while in water it increases to 300 $\mu$.

The presence of this epidermis explains how the seed so easily

becomes moistened. Another question is moreover, whether all tissues which compose the testa in like measure take part in this storing up of water.

Apart from the innermost thin membrane, consisting of inner integument, perisperm and endosperm, which surrounds the germ and is firmly fixed to it, the testa of *C. Pepo* and *maxima* is divided into five layers. The two outer ones we are now acquainted with. Next within these there is a tissue, 4 to 6 cells thick, built up out of small elements with fairly thick walls (layer III). Layer IV is the schlerenchyma-layer, composed of a single row of hard, thick-walled and very irregularly formed cells; this gives hardness to the testa. Finally layer V is a strongly developed spongy tissue composed of cells filled with air, separated by numerous intercellular spaces.

This spongy tissue at once gives the impression of being specially fitted by capillarity to hold a large quantity of water. In how far this and the remaining tissues of the testa partake in this phenomenon, we shall endeavour to determine quantitatively.

By scraping with a knife, layers II and III are easily removed from the seed: thereupon the brownish schlerenchyma comes into view. If this is done partly with air-dry seeds partly with seeds soaked in water, then the change in weight will show how much water these two layers together take up. In the same way the spongy tissue (layer V) can be removed from a pealed testa and by a comparison of the loss of weight in the dry and soaked testa the water content can be deduced. Finally also layer IV, the schlerenchyma must be isolated and its absorption of water determined separately.

With seeds of the variety Courge gaufrée I obtained in this way the following results.

| Layers. | Weight when dry. | Weight of water absorbed. |
|---|---|---|
| II—III. | 0.02 gr. | 0.02—0.03 gr |
| IV. | 0.02—0.03 gr. | 0.02 gr. |
| V. | 0.01 gr. | 0.08—0.10 gr. |

The great importance of the spongy tissue as a water-reservoir is thus demonstrated.

The greater part of this water is absorbed by capillarity, while the air which fills the cells and the intercellular spaces is expelled.

Connected with this is the fact that the seed of the pumpkin, when thrown into water air-dry, floats, but after some time sinks to the bottom, when a sufficient quantity of air has been expelled from the spongy tissue. In Courge gaufrée this is the case as

soon as the quantity of water absorbed amounts to 50—60% of the air-dry seed.

Now its seems very likely *a priori* that the air finds an outlet through the hilum of the seed. There is indeed an opening by means of which the intercellular spaces communicate with the outside air; the schlerenchyma is interrupted at the level of the hilum (v. HÖHNEL l. c. p. 315). Bubbles of air are seen to escape from the hilum of a seed immersed in water, although slowly and not very profusely. Thus F. NOLL has also pointed out that this arrangement may assist the water-absorption in the seeds of *Cucurbita* (Landw. Jahrb. Bd. 30, Erg. Bd. III, 1902 p. 150; footnote). But in how far this actually takes place remained to be investigated experimentally.

For this purpose a comparison was made between the increase of weight in water of seeds with the hilum open or opened still further where necessary, and seeds in which the hilum had been closed by means of sealing wax or bees wax. In this way it was found that the significance of the hilum is not specially great. The water-absorption was but little retarded by the closing of the hilum; the differences between seeds treated in this way and those not so treated frequently falls within the limits of individual variations; and also in the case of carefully selected and apparently comparable seeds the difference is indeed generally in favour of those with open hilum; but it is always relatively slight.

A single example is quoted:

3 seeds of Courge gaufrée untreated, weigh air-dry 0.76 gr.
„ „ whose hilum was closed with beeswax weigh air dry 0.77 „
after lying in water for 24 hours they weigh respectively 1.17 and 1.15 „
„ „ „ „ „ 48 „ „ „ „ 1.35 and 1.24 „
Thus there is water absorption after 48 hours of respectively 77.6 and 61.0 %

The difference which occurs here is among the most notable of those observed.

It is thus in the highest degree probable that the air, which is driven out of the spongy tissue, finds an outlet through the testa. This is also undoubtedly the reason why seeds with closed hilum do not in general remain floating longer than seeds which are not so treated. Moreover one can observe directly that water does not penetrate through the opening in the hilum to an appreciable extent. Even if this mouth is made somewhat wider, and it is immersed in a solution of a colouring matter, for instance 0.1% methylene blue in water,

so that the spongy tissue may come into contact with the fluid at the hilum, the fluid is only imbibed extremely slowly by capillarity. After 24 hours it has scarcely risen in the testa. On the other hand the coloured solution is quickly taken up by the surface of the seed, and it also diffuses very soon in the deeper layers.

It is different if the seed is immersed with the hilum in a fluid whose surface-tension is less than that of water, as for instance in alcohol. Such a fluid is soaked up much more quickly by capillarity (See inter alia L. Errera, Bull. Soc. belg. micr. t. 13, 1886, no. 3 and Rec. Inst. botan. Univ. Bruxelles, t. 2, 1906, p. 111). A solution of methylene blue in alcohol penetrates almost immediately into the hilum and in a very short time fills the whole spongy tissue. This can be seen very well if in a dry seed a piece of the testa is cut away from the extremity opposite to the hilum, and the latter is held in the solution. After a few seconds the blue fluid penetrates to the top, while the outer layer remains uncoloured and dry.

For the same reason a much quicker stream of air bubbles escapes from the hilum of a seed thrown into alcohol than when it is thrown into water. Also the weighings show important differences. A gourd-seed held with the hilum only in alcohol takes up in one minute 0.04 – 0.05 gr. of alcohol, whilst in the same time barely 0.01 gr of water is absorbed, and then moreover partly by imbibition of the outer layer. The outermost tissues of the testa only slightly imbibe alcohol; a seed immersed in alcohol so that only the hilum protrudes from the fluid, takes up in one minute 0.01—0.02 gr.; in water, under the same conditions about 0.05 gr. is taken up in the same time.

Along the narrow border and the whole way round on the outermost edge of the spongy tissue in the testa of *C. Pepo* and *C. maxima* there runs a canal filled with air that on its inner side contains the vascular bundle and opens out at the hilum, on the left and right (v. HÖHNEL l. c. p. 317).

In the experiment with coloured alcohol described above it is along this canal that the fluid first rises, in order to spread thence through the spongy tissue. It is therefore conceivable that the circumambient canal plays a dominant role in the capillary ascent. In order to decide this question a piece of the testa was removed half way up on both sides in such a way that the canal was interrupted on the left and right. As in the former experiment a piece of the testa was also removed from the round end of the seed and the hilum immersed in blue-coloured alcohol. Although now the fluid could penetrate to the top only through the spongy-tissue it did this about as quickly

as in seeds in which the canal was uninterrupted: no important differences could be observed.

In order to give an idea of the rate at which water is imbibed by seeds of other *Cucurbitaceae*, the increase in weight is given in the following table for a number of species, expressed as a percentage of the dry weight, which the seeds undergo when they lie in water for one hour:

| | |
|---|---|
| *Acanthosicyos horrida* Welw. | 14.0 % |
| *Momordica Charantia* L. | 9.6 „ |
| *Luffa cylindrica* Roem. | 4.1 „ |
| *Citrullus vulgaris* Schrad. (with black seed) | 25.6 „ |
| „ „ „ (with red seed) | 14.4 „ |
| „ „ „ (American lemon) | 16.4 „ |
| *Cucumis sativus* L. (Excelsior) | 13.9 „ |
| „ „ „ (Indian giant net-cucumber) | 24.4 „ |
| *Cucumis Melo* L. ("cantaloup" from Algiers) | 20.4 „ |
| *Benincasa cerifera* Savi. | 71.4 „ |
| *Lagenaria clavata* | 26.1 „ |
| *Trichosanthes Anguina* L. | 13.8 „ |
| *Cucurbita Pepo* L. ("gelber Zentner") | 33.7 „ |
| „ „ „ ("Miracle") | 18.2 „ |
| „ *argyrosperma* | 82.5 „ |
| „ *melanosperma* A. Br. | 16.3 „ |

It is seen from this table that the imbibition-velocity of different species varies considerably. The peculiarities of the structure of the testa, which explain these differences, will not be dealt with here for all the species mentioned. Attention will only be called to a few of the more notable cases. Only two species equal *Cucurbita Pepo* as regards the rate of absorption of water, viz. *Benincasa cerifera* and *Cucurbita argyrosperma*. Both these seeds finally take up very considerable quantities of water; after 4 or 5 days *Benincasa* increases in weight by 130%, and *C. argyrosperma* even by 150% of the original weight.

The seeds of *Benincasa* are found on microscopic investigation to possess an exceptionally thick layer of spongy tissue of the same structure as that observed in *C. Pepo* [1]). The spongy tissue in this case is most strongly developed on the *outer* side of the schlerenchyma instead of on the *inner* side; it is, in other words, layer III of

[1]) Compare H. A. Lothar. Anatomie comparée des Cucurbitacées Lille 1881 p. 215.

VON HÖHNEL which in respect of thickness as of power of imbibition, is the of most importance in this seed. A result of this is, that if the seeds are peeled down to the schlerenchyma and are then placed in water, they then take up much less water than seeds not so treated:
3 entire seeds weighing 0.18 gr. took up in 24 hours 0.13 gr. of water or 72%
3 peeled „ „ 0.15 „ „ „ „ „ „ 0.05 gr. of water or 33%

The great quantity of air present in the intercellular spaces is the reason why the seeds of *Benincasa* remain floating for a longer time on water than those of *Cucurbita Pepo*. Of ten *Benincasa*-seeds which were placed in water, two were still floating after 5 days, notwithstanding the increase in weight of the whole batch then already amounted to 132%.

The seeds of *Cucurbita argyrosperma* are very light and spongy to the touch and are specially characterised by the great development of the projecting edge of the seed, which, just as in *C. Pepo* and *maxima*, is divided by a groove from the body of the seed.

Especially this edge, which has a grey-blue colour while the rest of the seed is white, has a spongy structure, which appears very well adapted for taking up much water. Under the microscope it is found to consist of the same tissue with strongly developed air-spaces which constitutes layers III and V in *C. Pepo*. The schlerenchyma is not however present in the edge, which must therefore be considered as an excrescence of the testa which is formed by layer III and runs round the seed.

Its importance with regard to absorption is clear from the following observation:
3 entire seeds, weighing 1.22 gr. in 19 hours took up 1.19 gr. of water or 97%
3 seeds without edge, weighing 1.05 gr. in 19 hours took up 0.58 gr. of water or 55%.

The edge of the 3 seeds last mentioned was cut away beforehand. And this difference could still be observed three days later when the seeds which had been deprived of their edge had only taken up 78% of water, while on the other hand the entire seeds had taken up 131%.

When we now turn to the *Cucurbitaceae* whose seeds saturate themselves less thoroughly with water than those of *C. Pepo*, our attention is specially claimed by *Luffa cylindrica*, whose seeds, as will be seen from the above table, take up in the first hour eight times less water than those of the pumpkin and twenty times less

than the seed of *C. argyrosperma*. Nevertheless the structure of the testa of *Luffa* is not so strikingly different from that of *Cucurbita* as might be expected from the above-mentioned fact. The testa of *Luffa* possesses, under a hard and thick schlerenchyma layer, a fairly well-developed spongy tissue. [1]). This contradiction is only apparent; the seed of *Luffa* indeed takes up water very slowly, but in the long run it absorbs a fairly large quantity of water, more than 80% of its own dry-weight.

The difference in the rate of imbibition between *Cucurbita* and *Luffa* is explained by a considerable difference in the structure of the outermost layer of the seed. This has a strongly developed cuticle, as appears on treatment with strong sulphuric acid, and undoubtedly on this account becomes moistened relatively slowly. The same reason does not account for the slighter absorption of *Cucumis sativus* and *C. Melo*. These seeds become quickly moistened, but they cannot store up much fluid, because the spongy parenchyma is more or less completely absent from the testa. [2]) Indeed the total quantity of water absorbed by the species of *Cucumis* mentioned does not exceed 60 % of the dry weight.

One word finally about the variety of *C. Pepo* included in the above list as "miracle", which possesses a low capacity for imbibition. The seeds of this variety are distinguished by their testa consisting only of a thin, soft, grey-green membrane that very easily becomes saturated with water, yet can only store up a small quantity of it. Spongy tissue and schlerenchyma are both entirely absent from this membrane, and the "miracle" seeds, when thrown into water, sink at once to the bottom. Finally their increase in weight amounts to no more than 50 %; it is therefore comparable with that of the peeled seeds of *Curcurbita Pepo*.

**Microbiology.** — *"Individuality and heredity in a lower mould fungus, Trichophyton albiscicans"*. By Prof. A. W. NIEUWENHUIS. (Communicated bij Prof. M. W. BEIJERINCK).

*Introduction.*

One of the most striking observations arising from a prolonged residence among the still primitive tribes of the East-Indian Archipelago, for example among the Dajaks of Borneo, is certainly that our dark fellow-men are endowed with so good mental dispositions, of the same nature as ours, that they may in general well be compared

[1]) Compare LOTHAR l. c. p. 219 and K. G. BARBER, Bot. Gaz. vol. 47, 1909, p. 305.
[2]) Description and figures in VON HÖHNEL, LOTHAR, and BARBER.