# Toward a Clearer Concept of Osmotic Quantities in Plant Cells[1]

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THE CONCEPT OF OSMOTIC PRESSURE and its role in the physiology of the plant cell has long been a stumbling block for plant physiologists. A good share of the confusion is due to the multiplicity and inadequacy of the symbols and definitions proposed by various workers. In more recent years some semblance of uniformity has arisen as a result of the improved terminology proposed by Meyer (*1, 2*). That even this system has not solved all the problems is indicated by the large number of attempts to alter his concepts and symbols (*3, 4, 5*). This paper is admittedly one of them.

If we were to show Meyer's classical equation $DPD = OP - TP$ to a physicist or chemist, he would say "I see. $D \times P \times D$ equals $O \times P$ minus $T \times P$. Now tell me what $D$, $P$, $O$, and $T$ stand for, and your equation will be quite clear to me." Even the student is likely to question the use of two and three symbols for a single quantity. On the other hand, if we were to use the Greek symbols of thermodynamics, as suggested by Broyer (*5*), most physiologists would be simply confused. The happy medium is to use one symbol for each quantity, and wherever possible to adopt the ordinary Latin letter used by physicists and chemists, e.g., $P$ or $p$ for pressures.

*Terminology.* Another difficulty has been the meaning of the term "osmotic pressure." This has been used in so many different senses that it seems advisable to drop it altogether as a specific term and to use it in a generic sense. A physical chemist once asked me, "What is this turgor pressure? Isn't it simply osmotic pressure?" And I was forced to admit that it is an osmotic pressure, since it seems perfectly logical to use the term for any pressure that arises as a result of osmosis. But every solution is capable of developing a definite maximum osmotic pressure (under standard, ideal conditions), which, according to Thoday (*6*), should be called the osmotic potential of the solution. In practice, this osmotic potential is determined by finding the applied pressure that is just sufficient to stop osmosis when the solution is separated from pure solvent by a semipermeable membrane. The whole system should be maintained at a standard temperature—e.g., 20° C. From the cellular point of view, it might be defined as the maximum pressure that would develop when the solution is enclosed in an ideal cell and immersed in pure solvent. The ideal cell would possess a perfectly semipermeable and perfectly rigid membrane.

The turgor pressure, on the other hand, is the actual hydrostatic pressure exerted on the membrane at any one time and tending to stretch it. It cannot logically be used in any calculations of water movement, since only pressures exerted on the water can have any effect on its movement. It can never be greater than the osmotic potential of the solution and is always, or nearly always, less. The osmotic potential of the cell solution in the above ideal cell can, in fact, be defined as the maximum turgor pressure it can develop. Crafts *et al.* state that since the semipermeable membrane is freely permeable to water, "it will not sustain a static pressure from water molecules." This would lead to the conclusion that turgor pressure is not a hydrostatic pressure. The argument is based on a confusion between the "static pressure" and the diffusion pressure of the water molecules. There is no reason why the water molecules on the inside cannot exert a greater pressure on the "partitions" in the membrane, though diffusing through the "pores" at the same rate in both directions. In fact, the diffusion pressure of the water molecules in the solution cannot be increased to the level of pure water unless they are subjected to a "static pressure," and they react with an equal (hydro)static pressure on the membrane—the so-called turgor pressure.

The wall pressure is the pressure exerted by the wall (or membrane) on the solution, causing an increase in diffusion pressure of the solvent and therefore affecting its movement into or out of the cell. According to Newton's third law of motion, it is at any one time exactly equal and opposite to the turgor pressure.

This is a point that has frequently been misunderstood. It has been stated (*4, 7*) that turgor pressure must be greater than wall pressure for cell growth to occur. Others (*6*) have recognized that such is not the case since, according to Newton's law, though the forces are oppositely directed, they are not acting on the same body, and therefore neither one is opposing the action of the other. The turgor pressure is exerted on the wall, the wall pressure on the solution. Whether the turgor pressure is sufficient to stretch the wall is dependent, not at all upon the wall pressure, but upon the tensile strength or modulus of elasticity of the wall. Thus (*8*):

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$M = \frac{\text{stress}}{\text{strain}}$ where $M$ = modulus of elasticity of the wall.
Since stress $= F/a$ and strain $= e/l$,
where $F$ = force
$a$ = area
$e$ = increase in length (perimeter) of wall
$l$ = original length (perimeter) of wall

$$M = \frac{F/a}{e/l}$$

Knowing the dimensions of a cylindrical cell and the turgor pressure, it is possible to calculate the total thrust on the end walls. This would give the value $F$, and $a$ would be the cross-sectional area of the wall. Thus the stress would be $\frac{pA}{a}$ where $A$ = area of end walls, $a$ = cross-sectional area of wall.

Therefore

$$M = \frac{\frac{pA}{a}}{e/l}$$

and

$$e/l = \frac{\frac{pA}{a}}{M} = \frac{pA}{aM}.$$

Similar equations have been worked out in greater detail by Haines (*9*).

Since $e/l$ is really a measure of cell growth, this means that the growth of the cell may be increased either by an increase in turgor pressure or by a decrease in the modulus of elasticity of the cell wall. Unfortunately, however, this is an oversimplification. The relationship holds only within the elastic limit of the cell wall, and only if the cell wall obeys Hooke's law (*6*). If the turgor pressure is too large, it will stretch the wall beyond its elastic limit, and cell enlargement will be greater than calculated from the above relationship (*10*). In other words, the above equation gives the minimal cell growth for a specific turgor pressure and tensile strength of the wall, and it clearly shows that wall pressure is not a factor.

Thus turgor pressure affects cell enlargement, wall pressure affects the diffusion pressure of the cell water, and the two are always numerically equal.

The problem as to which of the two equal and opposite pressures is the "initial one" will depend on the conditions. If water enters a flaccid cell (as normally happens), it perhaps may be stated that the hydrostatic pressure ("turgor pressure") is the "initial one." If, instead, the cell walls of the flaccid cells were suddenly to contract (a doubtful occurrence), the wall pressure might be considered the "initial one." But the question is somewhat academic since the two pressures must arise simultaneously—the one cannot exist even momentarily without the other.

*Symbols.* In the system to be followed below, several simple principles are borrowed from standard usage by physicists and chemists: (1) One letter is to be used for one quantity. (2) Since pressures are vector quantities, the direction is to be indicated by the sign. (3) Subscripts will be used where a further characterization of the quantity is needed. Thus:

Diffusion pressures of the solvent are represented by $D$, the real mechanical pressures (e.g., turgor pressure and wall pressure) are represented by $p$; and osmotic potentials are represented by $O$.

Pressures that tend to send water into the cell are preceded by a negative sign; those that tend to send water out of the cell are preceded by a positive sign. (The reason for this choice will be explained.) The following are the main symbols:

$D_c$ is the diffusion pressure of the solvent in a cell.

$D_e$ is the diffusion pressure of the solvent in a solution external to the cell.

$D_{H_2O}$ is the diffusion pressure of pure water.

$O_c$ is the osmotic potential of a cell solution.

$O_e$ is the osmotic potential of an external solution.

$p_w$ is the wall pressure of a cell (determined in sections or tissue strips).

$p_e$ is the pressure on the cell by the surrounding tissues.

$p_t$ is the turgor pressure.

*Equations.* The equations are developed from the simple thermodynamics of osmotic pressure (*11*). In order to remove one mole of solvent (having a volume $V_1$) from a much larger volume of solution (in an artificial cell), a pressure ($P$) infinitesimally larger than the osmotic potential ($O$) would have to be applied (e.g., by means of a semipermeable piston), and the free energy change would have to be equal to the work done:

$$\Delta F = PV_1. \tag{1}$$

This basic equation establishes the significance of the sign. A positive pressure on the solvent molecules increases their escaping tendency. Thus $P$ must also equal the increase in diffusion pressure of the solvent molecules. It is obvious that the opposite relation must also hold (e.g., when the piston pressure is removed):

$$-\Delta F = -PV_1 = -OV_1, \tag{2}$$

where $-\Delta F$ is the reduction in free energy of the solvent molecules below that of pure solvent, and since $-O = -P$, $-O$ is the reduction in diffusion pressure of the solvent molecules due to the presence of solute. In other words, if it takes a pressure $P$ to raise the diffusion pressure of the solvent molecules to that of pure water, the diffusion pressure must have been lowered by the same amount due to the presence of solute. Thus, when the solvent is water and there is no applied pressure:

$$D_c - D_{H_2O} = -O_c. \tag{3}$$

The minus sign indicates that the diffusion pressure difference tends to send water into the solution, with a pressure equal to the osmotic potential of the solution.

Suppose a pressure less than $O_c$ is exerted on the cell solution (e.g., by the semipermeable piston). Using the symbol $p_w$ for this pressure,

$$D_c - D_{H_2O} = p_w - O_c. \tag{4}$$

This $p_w$ is obviously the wall pressure, and we have the equivalent of the expression, $DPD = OP - TP$. But there is one difference—the terms on the right side of

the equation are reversed. We now see that it should be wall pressure minus osmotic potential, in order to yield the correct sign, and to indicate that water would tend to go into the cell as long as $p_w$ is less thas $O_c$. $p_w$ will have a negative value in cells under tension—e.g., in wilted plants—thus increasing the value of $D_c - D_{H_2O}$.

If, instead of pure solvent, there is a solution outside the cell, then the equation for this solution is

$$D_e - D_{H_2O} = -O_e. \quad (5)$$

Subtracting (5) from (4):

$$(D_c - D_{H_2O}) - (D_e - D_{H_2O}) = p_w - O_c - (-O_e),$$
$$\text{or } D_c - D_e = p_w - O_c + O_e. \quad (6)$$

This equation enables us to determine not only the net pressure but also whether it tends to send the water into (−) or out of (+) the cell.

$p_w$ should really represent the net wall pressure—i.e., the algebraic sum of all pressures exerted on the wall. Thus in a tissue this would include the pressure exerted by the surrounding cells on the wall of the cell under consideration. However, if (as is usually the case) $p_w$ is determined on sections or strips of tissue in which the cell is no longer subjected to the tissue pressure ($p_e$), then a correction must be made if we are to obtain a true picture of the new pressure when the cell is in its normal environment. Thus

$$D_c - D_e = p_w + p_e - O_c + O_e, \quad (7)$$

where $p_w$ is the wall pressure of the cell in a section or tissue strip, and $p_e$ is the additional tissue pressure exerted on the cell in its normal environment. The turgor pressure ($p_t$) will, of course, also equal $p_w$ in the section or strip but will equal $p_w + p_e$ in the cell subjected to normal tissue pressures. Whether or not $p_e$ is small enough to be neglected is difficult to determine.

The terminology for the expression $D_c - D_e$ is not easy to decide. European workers may still prefer to call it "suction tension" and American physiologists, "net diffusion pressure deficit." It should, however, be emphasized that the quantity may be positive (e.g., when a cell is transferred to a hypertonic solution) and therefore is not necessarily a deficit. For the sake of clarity, a longer expression is needed—e.g., the diffusion difference between the cell and the surrounding solution. Similarly, $D_c - D_{H_2O}$ is the diffusion difference between the cell and pure water. It seems superfluous to lengthen the term diffusion difference to "water diffusion pressure difference," though purists may prefer to do so. The objections to any term, including "suction," have been clearly presented (*1, 2*).

The methods used to measure these values are well known. The physical chemist determines the osmotic potential of a solution by finding the counter pressure that is just sufficient to stop osmosis. The physiologist calculates $O_c$ from freezing point determinations on the sap or equates it to the osmotic potential of the solution causing incipient plasmolysis. Since these two values may differ somewhat from each other, they may be designated $O_{c(\Delta)}$ and $O_{c(i)}$ respectively. $D_c - D_{H_2O}$ is determined by finding the numerically equal osmotic potential ($O_e$) of the solution that fails to cause any change in size of the cell. This depends on the fact that $-O_e = D_e - D_{H_2O}$ (equation 5) and, if no change in size of the cell occurs, $D_e = D_c$; therefore $D_c - D_{H_2O} = -O_e$. In most cases, $p_w$ must be calculated from the above two results. Any existing imbibitional forces are included in $O_c$ and $O_e$ unless the methods of measurement change them.

It is of interest to examine some of the more recent suggestions with the aid of these equations. Burström (*4*) states that turgor pressure is "of the same nature as . . . Meyer's diffusion pressure deficit . . . but relative to the surrounding medium instead of to distilled water." His equation for turgor pressure is: $T = O - E$, where $O$ is the above-described osmotic potential of the cell sap, and $E$ is the osmotic potential of the external solution. Using the symbols developed above:

$$O - E = O_c - O_e.$$

But from equation (6)

$$O_c - O_e = p_w - (D_c - D_e).$$

Thus, when wall pressure is zero, Burström's "turgor pressure" is actually the diffusion difference (with the sign reversed) between the cell and its surrounding solution ($D_c - D_e$). On the other hand, if diffusion equilibrium exists ($D_c - D_e = 0$), then his quantity does actually represent the turgor pressure (since $p_t = p_w$). Broyer (*12*) has, in fact, used the expression in the latter sense—i.e., he assumes that diffusion equilibrium or near-equilibrium conditions prevail.

*Active absorption of water.* If an active (i.e., a "nonosmotic") uptake of water occurs in consequence of the expenditure of energy by the cell system, this would be equivalent to decreasing the free energy of the cell water molecules relative to the external molecules. Thus, if the resulting active (i.e., metabolically induced) decrease in diffusion pressure is $-A_c$:

$$D_c - D_e = p_w - A_c - O_c + O_e. \quad (8)$$

The importance of such an "active osmotic pressure" has been the subject of controversy in recent years. Many papers have attempted to prove that it is quantitatively much higher than the osmotic potential of the cell sap. If this were true, the above equations would not be of much value, since the "active," or "nonosmotic," component, if it exists, may conceivably be altered by the methods used to determine $D_c - D_e$ and $O_c$. But it has been shown that the evidence for a "nonosmotic" absorption is based on incorrect interpretation of results and that, from thermodynamic considerations, an "active" osmotic pressure, if it exists, can only be of the order of one atmosphere (*10*). True, Bennet-Clark (*13*), though accepting these thermodynamic calculations, has attempted to justify his theory by suggesting that the experimentally de-

termined value for the permeability of cells to water is 100–1,000 × too high, as used in these calculations.

The sole basis for this suggestion is an observation by Huber and Höfler (*14*). They noted that when the long, cylindrical cells of the aquatic plant *Salvinia* were plasmolyzed, they frequently separated into two protoplast fragments. In nearly all such cases the smaller fragment plasmolyzed more rapidly than the larger, approximately the same amount of water leaving the smaller one per unit of time as the larger one. Huber and Höfler calculated the specific surface of the two parts and found very little difference between them. Consequently, they concluded that practically all the water was leaving the two protoplast fragments through the free convex ends, and very little through the adherent sides.

Although he does not say so, Bennet-Clark interprets this unproved hypothesis of Huber and Höfler in direct opposition to their own interpretation. He maintains that the protoplasm detached from the wall must therefore have a permeability of 100–1,000 × that of the normal adherent protoplasm. Huber and Höfler, on the other hand, emphatically point out that, if their hypothesis is correct, it cannot be explained by a difference in permeability. Their evidence for this statement is twofold: (*a*) other workers (Fitting and Bärlund) have failed to detect any difference in permeability to solutes between plasmolyzed and unplasmolyzed cells; (*b*) their own results with the slowly plasmolyzing cells of *Spirogyra nitida* yielded a smooth curve, though measurements of shrinkage were made both before and after plasmolysis. Their conclusion is that nearly all the water leaves the plasmolyzing cells of *Salvinia* through the convex menisci, not because of any such impossible difference in permeability, but simply because of freer access to the plasmolyzing solution.

All the results of other workers also disprove Bennet-Clark's contention. Thus, Levitt *et al.* (*15*) showed that the permeability of free protoplasts of onion scale tissue was the same as that of plasmolyzed protoplasts still inside their cell walls and therefore partially adherent to them. Resühr's (*16*) results with naturally free protoplasts (*Fucus* eggs) agree well with the above value for artificially freed onion protoplasts. In fact, anyone who has made measurements on shrinking cells cannot doubt that the rate is just as rapid in the case of unplasmolyzed cells as in plasmolyzed cells, as long as free access to the plasmolyzing solution is maintained.

There is thus no reason to doubt the validity of the experimentally determined value for water permeability. In fact, the excellent agreement between the values obtained by different workers, using different kinds of cells and different methods of measurement, speaks well for the accuracy of the published values. Consequently, the above conclusion still stands—i.e., an "active" osmotic pressure, if it exists, can only be of the order of one atmosphere.

It is interesting to note that the most recent results indicate little or no "nonosmotic" absorption by roots. The best evidence to date for active "nonosmotic" absorption has been produced by van Overbeek (*17*). Van Nie *et al.* (*18*) repeated his experiments and obtained a value of only 0.5 atmosphere that could possibly be ascribed to "nonosmotic" absorption. This is about one atmosphere less than van Overbeek's value, and even it could be just as logically explained by a loss of salts or a gain of water as the sap is transported to the stump.

The system of symbols and terminology for osmotic quantities proposed has the following advantages over the accepted ones:

1) The expression $D_c - D_{H_2O}$ gives the diffusion difference between the cell and pure water. This is more explicit than *DPD* because it clearly indicates that the diffusion pressure of the cell's water is being compared with that of pure water. *DPD,* on the other hand, gives rise to the question "deficit with respect to what?" Even "net *DPD*" is not as explicit as $D_c - D_e$ because the latter again indicates that the cell is being compared with the external solution.

2) *DPD* is applicable only when there is a deficit. $D_c - D_e$ can be used in all cases, whether a diffusion pressure deficit or a diffusion pressure excess is involved.

3) The direction of water movement as a result of the diffusion gradient is clearly indicated by the sign. When $D_c - D_e$ is negative, a negative gradient exists, and water will move into the cell; when it is positive, water will move out.

4) The accepted symbol for pressure ($p$) is used only for real pressures (e.g., turgor pressure and wall pressure).

5) The symbol $O$ is used for the osmotic potential of the cell solution.

6) The term "osmotic pressure," frequently used in different senses, is here regarded as a generic term applying to all pressures resulting from osmosis (turgor pressure, wall pressure, and the potential pressure of the solution).

### References


1. MEYER, B. S. *Botan. Rev.,* **4**, 531 (1938).
2. ———. *Plant Physiol.,* **20**, 142 (1945).
3. CRAFTS, A. S., CURRIER, H. B., and STOCKING, C. R. *Water in the Physiology of Plants.* Waltham, Mass.: Chronica Botanica (1949).
4. BURSTROM, H. *Physiologia Plantarum,* **1**, 57 (1948).
5. BROYER, T. C. *Botan. Rev.,* **13**, 1 (1947).
6. THODAY, D. *Ann. Botany,* **14**, 1 (1950).
7. BROYER, T. C. *Plant Physiol.,* **25**, 135 (1950).
8. STEWART, O. M. *Physics, a Textbook for Colleges.* Boston: Ginn (1931).
9. HAINES, F. M. *Ann. Botany,* **14**, 385 (1950).
10. LEVITT, J. *Plant Physiol.,* **22**, 514 (1947).
11. GETMAN, F. H., and DANIELS, F. *Outlines of Theoretical Chemistry.* New York: Wiley (1937).
12. BROYER, T. C. *Plant Physiol.,* **25**, 420 (1950).
13. BENNET-CLARK, T. A. *Discussions of the Faraday Soc.,* **3**, 134 (1948).
14. HUBER, B., and HOFLER, K. *Jahrb. wiss. Botan.,* **73**, 351 (1930).
15. LEVITT, J., SCARTH, G. W., and GIBBS, R. D. *Protoplasma,* **26**, 237 (1936).
16. RESUHR, B. *Ibid.,* **24**, 531 (1935).
17. VAN OVERBEEK, J. *Am. J. Botany,* **29**, 677 (1942).
18. VAN NIE, R., HELDER, R. J., and ARISZ, W. H. *Proc. Kon. Ned. Akad. van Wet.,* **53**, 3 (1950).