Osmoregulation:

Stress Osmoregulation is the passive regulation of the osmotic pressure of an organism's body fluids, detected by osmoreceptors, to maintain the homeostasis of the organism's water content; that is, it maintains the fluid balance and the concentration of electrolytes (salts in solution) to keep the fluids from becoming too diluted or concentrated.

The protoplasm of living organisms has a high percentage of water, so without water, living organisms would die. Plants living in water, or those in hot, arid conditions where water is not readily available all the time, or in which there is a high concentration of solutes such as occurs in/near sea water, must adapt their structure and/or their various functions – or both – to ensure the conservation of needed water and prevent the upset of the osmotic balance of cell contents. Without the right osmotic balance – the plant dies.

Surviving the salt These Mangroves grow in wet, muddy soil at the sea -water's edge. If you look at the leaves, salt crystals are excreted on to their surfaces, and if you taste the sap – it's very salty! Surviving the salt Some mangroves are almost covered by salty sea water! Most trees cannot survive in water that has too much salt in it, but mangrove trees have a unique adaptation for dealing with the sea's salinity.

Surviving the salt: When they're submerged in sea water, warty growths on mangrove roots filter out most of the salt as they take water in through their roots. Some mangroves concentrate extra salt in old leaves (which turn yellow and die), and some are able to get rid of the salt by secreting it through the pores of special glands.

Surviving drought in contrast to mangroves, plants, such as these cacti and Acacia that live in places like along the Palisadoes strip or in the Hellshire area, grow in limited, dry, sandy soil, with little rainfall, a very high temperature and a hot, dry wind.

Some water conservation methods Succulent plant stem (Cactus) Succulent leaves of Sesuvium & Aloe

OSMOREGULATION IN PLANTS:

While there are no specific osmoregulatory organs in higher plants, the stomata are important in regulating water loss through evapotranspiration, and on the cellular level the vacuole is crucial in regulating the concentration of solutes in the cytoplasm. Strong winds, low humidity and high temperatures all increase evapotranspiration from leaves. Abscisic acid is an important hormone in helping plants to conserve water—it causes stomata to close and stimulates root growth so that more water can be absorbed.

Plants share with animals the problems of obtaining water but, unlike in animals, the loss of water in plants is crucial to create a driving force to move nutrients from the soil to tissues. Certain plants have evolved methods of water conservation.

Xerophytes are plants that can survive in dry habitats, such as deserts, and are able to withstand prolonged periods of water shortage. Succulent plants such as the cacti store water in the vacuoles of large parenchyma tissues. Other plants have leaf modifications to reduce water loss, such as needle-shaped leaves, sunken stomata, and thick, waxy cuticles as in the pine. The sand-dune marram grass has rolled leaves with stomata on the inner surface.

Hydrophytes are plants in water habitats. They mostly grow in water or in wet or damp places. In these plants the water absorption occur through the whole surface of the plant, e.g., the water lily.

Halophytes are plants living in marshy areas (close to sea). They have to absorb water from such a soil which has higher salt concentration and therefore lower water potential(higher osmotic pressure). Halophytes cope with this situation by activating salts in their roots. As a consequence, the cells of the roots develop lower water potential which brings in water by osmosis. The excess salt can be stored in cells or excreted out from salt glands on leaves. The salt thus secreted by some species help them to trap water vapours from the air, which is absorbed in liquid by leaf cells. Therefore, this is another way of obtaining additional water from air, e.g., glasswort and cord-grass.

Mesophytes are plants living in lands of temperate zone, which grow in well-watered soil. They can easily compensate the water lost by transpiration through absorbing water from the soil. To prevent excessive transpiration they have developed a waterproof external covering called cuticle.

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OSMOREGULATORY ADAPTATIONS:

The plants shown on the previous slides have adaptations that ensure osmoregulation. υ Osmoregulation is the active regulation of the osmotic pressure of an organism''s fluids to maintain the homeostasis (or constant unchanging balance) of the organism's water content; that is, it keeps the organism's fluids from becoming too diluted or too concentrated.

1. Plants such as mangroves develop structural and physiological adaptations to regulate the osmotic balance of their cell contents – i.e to carry out osmoregulation. ¬ The cacti and other plants living along the hot, dry scrubland of the Palisadoes strip also develop special adaptive features for osmoregulation.

OSMOREGULATION AND STRESS PARADIGM IN PLANTS:

Drought and salinity stress are the major causes of historic and modern agricultural productivity losses throughout the world. υ Both drought and salinity result in osmotic stress that may lead to inhibition of growth. Salinity causes additional ion toxicity effects mainly through perturbations in protein and membrane structure. υ In contrast to animals, which rely on Na1/K1-ATPases for the expulsion of osmotic plants rely on plasma membrane and endosomal ATPase activities to generate proton gradients to drive ion extrusion and intracellular sequestration.

Osmoregulatory adaptations:

Types of plants Depending on their habitat, plants can be grouped into four different types according to the osmoregulatory adaptations that they show either in their structure, functions, or both. υ Groups are as follows: θ Halophytes θ Hydrophytes θ Xerophytes θ Mesophytes

Examples (a) mangrove = halophyte (b) Catus = xerophyte (c) an ackee tree = mesophyte (d) water lily = hydrophyte The leaves float on the water surface and numerous stomata are present on the upper surface of the leaves facing the atmosphere to promote loss of water. The surface area of these leaves is very large to enable excessive water loss by transpiration

Why is osmoregulation important to plants?

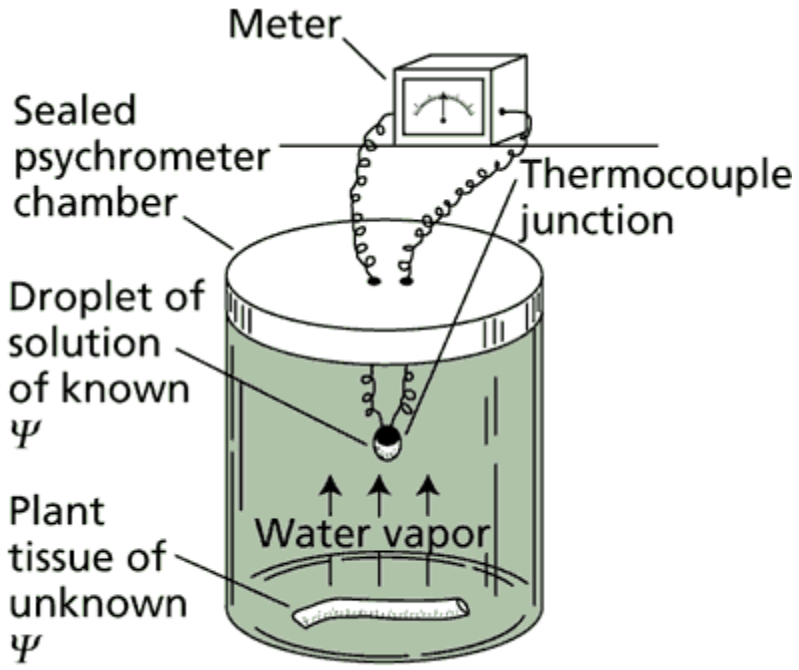
1) Enables the plant to grow, develop, carry on respiration, photosynthesis and survive, even if: θ the habitat is dry, hot and desert-like. θ sandy/rocky soil does not hold much water. θ rainfall is scarce or only at certain times. θ adequate water is not available for photosynthesis and θ hydration of the cell contents. θ habitat is completely aquatic. θ salinity of the habitat is high. 2) It regulates and balances the uptake and loss of water and solutes so maintains homeostasis.

Measuring Water Potential

Plant scientists have expended considerable effort in devising accurate and reliable methods for evaluating the water status of a plant. Four instruments that have been used extensively to measure Ψ, Ψ_s, and Ψ_p are described here: psychrometer, pressure chamber, cryoscopic osmometer, and pressure probe.

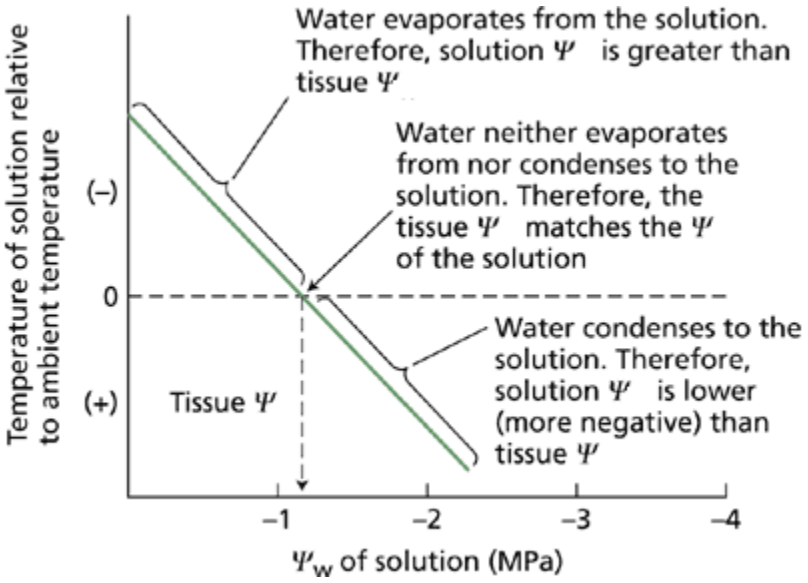
Psychrometer (Ψ measurement)

Psychrometry (the prefix "psychro-" comes from the Greek word *psychein*, "to cool") is based on the fact that the vapor pressure of water is lowered as its water potential is reduced. Psychrometers measure the water vapor pressure of a solution or plant sample, on the basis of the principle that evaporation of water from a surface cools the surface.



One psychrometric technique, known as *isopiestic psychrometry*, has been used extensively by John Boyer and coworkers (Boyer and Knipling 1965). Investigators make a measurement by placing a piece of tissue sealed inside a small chamber that contains a temperature sensor (in this case, a thermocouple) in contact with a small droplet of a standard solution of known solute

concentration (known Ψ_s and thus known Ψ). If the tissue has a lower water potential than that of the droplet, water evaporates from the droplet, diffuses through the air, and is absorbed by the tissue. This slight evaporation of water cools the drop. The larger the difference in water potential between the tissue and the droplet, the higher the rate of water transfer and hence the cooler the droplet. If the standard solution has a lower water potential than that of the sample to be measured, water will diffuse from the tissue to the droplet, causing warming of the droplet. Measuring the change in temperature of the droplet for several s olutions of known Ψ makes it possible to calculate the water potential of a solution for which the net movement of water between the droplet and the tissue would be zero signifying that the droplet and the tissue have the same water potential.



Psychrometers can be used to measure the water potentials of both excised and intact plant tissue. Moreover, the method can be used to measure the Ψ_s of solutions. This can be particularly useful with plant tissues. For example, the Ψ of a tissue is measured with a psychrometer, and then the tissue is crushed and the Ψ_s value of the expressed cell sap is measured with the same instrument. By combining the two measurements, researchers can estimate the turgor pressure that existed in the cells before the tissue was crushed ($\Psi_p = \Psi - \Psi_s$).

A major difficulty with this approach is the extreme sensitivity of the measurement to temperature fluctuations. For example, a change in temperature of 0.01°C corresponds to a

change in water potential of about 0.1 MPa. Thus, psychrometers must be operated under constant temperature conditions. For this reason, the method is used primarily in laboratory settings. There are many variations in psychrometric technique; interested readers should consult Brown and Van Haveren 1972, Slavik 1974, and Boyer 1995.

Pressure chamber (Ψ measurement)

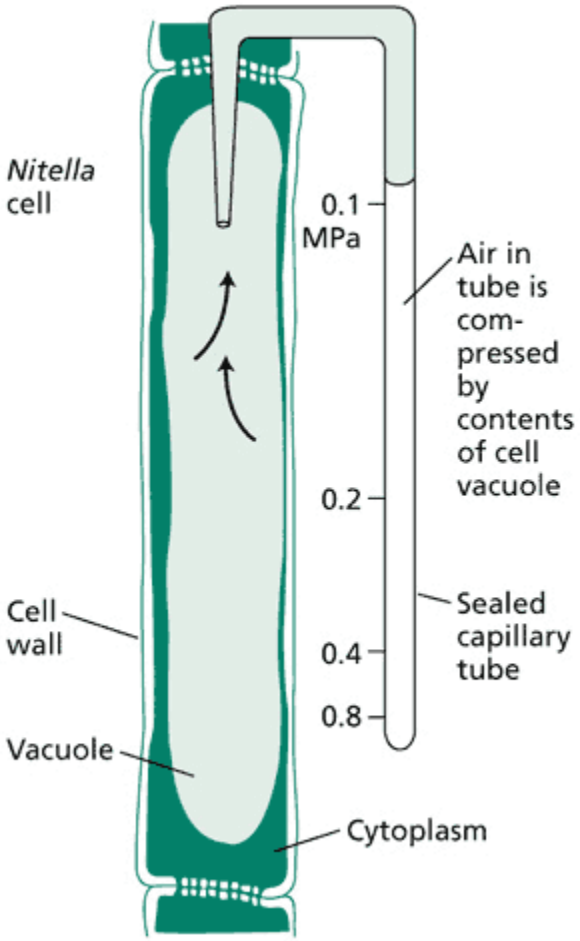
A relatively quick method for estimating the water potential of large pieces of tissues, such as leaves and small shoots, is by use of the **pressure chamber** . This method was pioneered by Henry Dixon at Trinity College, Dublin, at the beginning of the twentieth century, but it did not come into widespread use until P. Scholander and coworkers at the Scripps Institution of Oceanography improved the instrument design and showed its practical use (Scholander et al. 1965).

In this technique, the organ to be measured is excised from the plant and is partly sealed in a pressure chamber. Before excision, the water column in the xylem is under tension. When the water column is broken by excision of the organ (i.e., its tension is relieved allowing its Ψ_p to rise to zero), water is pulled rapidly from the xylem into the surrounding living cells by osmosis. The cut surface consequently appears dull and dry. To make a measurement, the investigator pressurizes the chamber with compressed gas until the distribution of water between the living cells and the xylem conduits is returned to its initial, pre-excision, state. This can be detected visually by observing when the water returns to the open ends of the xylem conduits that can be seen in the cut surface. The pressure needed to bring the water back to its initial distribution is called the *balance pressure* and is readily detected by the change in the appearance of the cut surface, which becomes wet and shiny when this pressure is attained.

Pressure probe (Ψ_p measurement)

If a cell were as large as a watermelon or even a grape, measuring its hydrostatic pressure would be a relatively easy task. Because of the small size of plant cells, however, the development of methods for direct measurement of turgor pressure has been slow. Using a micromanometer, Paul Green at the University of Pennsylvania developed one of the first direct methods for measuring

turgor pressure in plant cells (Green and Stanton 1967). In this technique, an air-filled glass tube sealed at one end is inserted into a cell). The high pressure in the cell compresses the trapped gas, and from the change in volume one can readily calculate the pressure of the cell from the ideal gas law (pressure × volume = constant). This method works only for cells of relatively large volume, such as the giant cell of the filamentous green alga *Nitella.* For smaller cells, the loss of cell sap into the glass tube is sufficient to deflate the cell and this yields artifactually low pressures.



For higher plant cells, which are several orders of magnitude smaller in volume than *Nitella,* a more sophisticated device, the **pressure probe** , was developed by Ernest Steudle, Ulrich Zimmermann, and their colleagues in Germany (Husken et al. 1978). This instrument is similar to a miniature syringe. A glass microcapillary tube is pulled to a fine point and is inserted into a cell. The microcapillary is filled with silicone oil, a relatively incompressible fluid that can be readily distinguished from cell sap under a microscope. When the tip of the microcapillary is first

inserted into the cell, cell sap begins to flow into the capillary because of the initial low pressure of that region. Investigators can observe such movement of sap under the microscope and counteract it by pushing on the plunger of the device, thus building up a pressure. In such fashion the boundary between the oil and the cell sap can be pushed back to the tip of the microcapillary. When the boundary is returned to the tip and is held in a constant position, the initial volume of the cell is restored and the pressure inside the cell is exactly balanced by the pressure in the capillary. This pressure is measured by a pressure sensor in the device. Thus the hydrostatic pressure of individual cells may be measured directly.

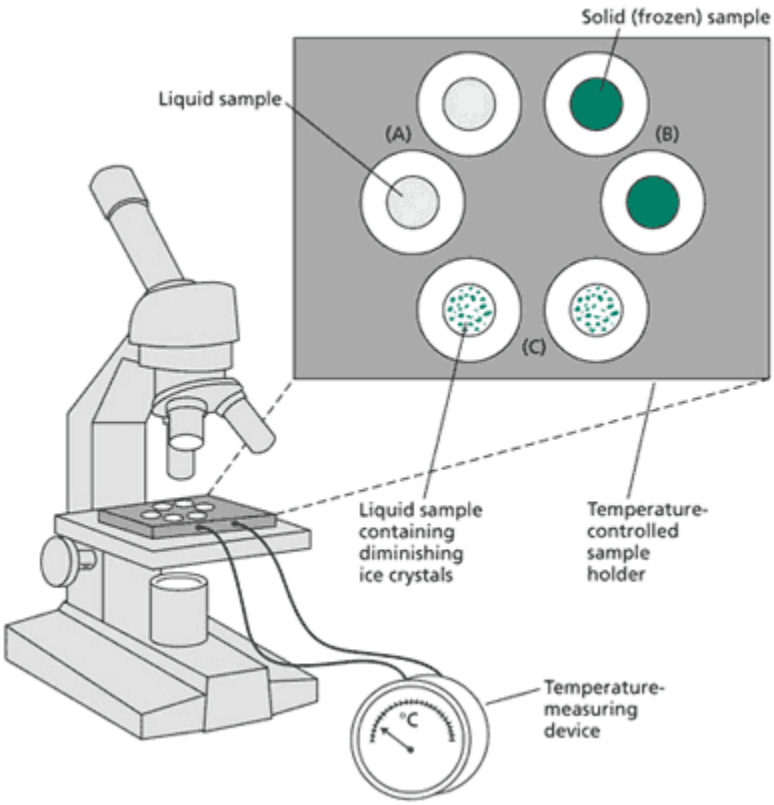
This method has been used to measure Ψ_p and other parameters of water relations in cells of both excised and intact tissues of a variety of plant species (Steudle 1993). The primary limitation of this method is that some cells are too small to measure. Furthermore, some cells tend to leak after being stabbed with the capillary, and others plug up the tip of the capillary, thereby preventing valid measurements. The pressure probe has also been adapted to measure positive and negative values of Ψ_p in the xylem (Heydt and Steudle 1991). However, technical problems with cavitation (see textbook Chapter 4) limit the measurement of negative Ψ_p by this technique.

Cryoscopic osmometer (Ψ_s measurement)

The **cryoscopic osmometer** measures the osmotic potential of a solution by measuring its freezing point. Solutions have *colligative* properties that collectively depend on the number of dissolved particles and not on the nature of the solute. For example, solutes reduce the vapor pressure of a solution, raise its boiling point, and lower its freezing point. The specific nature of the solute does not matter. One of the colligative *properties* of solutions is the decrease in the freezing point as the solute concentration increases. For example, a solution containing 1 mol of solutes per kilogram of water has a freezing point of –1.86°C, compared with 0°C for pure water.

Various instruments can be used to measure the freezing-point depression of solutions (for two examples, see Prager and Bowman 1963, and Bearce and Kohl 1970). With a cryoscopic osmometer, solution samples as small as 1 nanoliter (10^{-9} L) are placed in an oil medium located on the temperature-controlled stage of a microscope. The very small sample size allows sap from single cells to be measured and permits rapid thermal equilibration with the stage. To prevent evaporation, the investigator suspends the samples in oil-filled wells in a silver plate (silver has

high thermal conductivity). The temperature of the stage is rapidly decreased to about –30° C, which causes the sample to freeze. The temperature is then raised very slowly, and the melting process in the sample is observed through the microscope. When the last ice crystal in the sample melts, the temperature of the stage is recorded (note that the melting and freezing points are the same). It is straightforward to calculate the solute concentration from the freezing-point depression; and from the solute concentration (c_s), Ψ_s is calculated as $-RTc_s$.This technique has been used to measure droplets extracted from single cells (Malone and Tomos 1992).



Pressure-volume curve:

Pressure-volume curve describe the relationship between total water potential (Ψt) and relative water content (R) of living organisms. These values are widely used in research on plant-water relations, and provide valuable information on the turgor, osmotic and elastic properties of plant tissues.

According to the Boyle–v'ant Hoff Relation, the product of osmotic potential and volume of solution should be a constant for any given amount of osmotically active solutes in an ideal osmotic system.

$\psi_0 (V)$ = A constant is osmotic potential

ψ_0 and (V) is volume of solution.

This can then be manipulated to a linear relation which describes the ideal situation:

$$\psi_0 = \frac{1}{V} \times \text{A constan}$$
