Calculus in Plant Science

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FOREWORD

Phenomena in natural science that Georges L. Cuvier considered quantitatively inaccessible 200 years ago have today become scientifically untrustworthy if they are not backed by numbers. Today, based on solid physical and molecular foundations, Plant Biology has far surpassed the fundamentally descriptive stage. Now, the prediction and measurement of physical and chemical variables in cells, tissues, whole plants and their environment are essential to understand the plants, their functioning and evolution and their interactions with the environment. Current formalism explains most plant processes as a logical consequence of the properties of their components and of the physical and chemical peculiarities of the external medium. Therefore, the understanding of phenomena in Plant Science involves smooth conversion between molecular and macroscopic concepts. Explanations require that the student use quantitatively grounded concepts of physics, chemistry and molecular and cellular biology in a scenario well defined by taxonomy and evolution.

Experience has taught us that the resolution of problems in the classroom is a useful support to lectures and the laboratory. It provides the student with a necessary understanding of the physical and molecular bases of the plant processes. By solving problems, the students may assess their understanding of the matter and the professor gains a tool for the evaluation of the efficacy of explanations made in lectures. The problems stimulate questions, shape a scientific mentality and give the opportunity to critically discuss the possibilities and limits of different technologies.

The book includes 145 problems whose resolution has been experienced in the classroom and discussed with colleagues. Most problems were previously published in Spanish by the University of Alcalá (Spain). Although not exhaustive, the problems cover very diverse aspects of Plant Biology and are of variable degrees of difficulty. Usually within a group, the problems are ordered from low to high difficulty. However, within a topic, groups with various difficulties are ordered following the usual order of explanations in lectures. The International System (SI) of units is used thoroughly in the book, although sometimes other frequently used units are introduced to train the student on unit conversion exercises. Appropriate sections in appendixes provide supplementary data for the conversions.

Most problems are assembled in six specific topics, concluding with a last group where each problem deals with several topics of Plant Biology. The solution of each problem is explained in detail and accompanied, when appropriate, with figures. Sometimes, brief introductions or conclusions accompany the solutions to provide a perspective on the problem within the context of Plant Biology. Some problems dealing with transport, photosynthesis or metabolism require specific background on the mechanisms involved and their solutions are explained in extensive detail. The first topic "Cell and cell wall" commences with a subsection (1a. *Comparative number and size of molecules and cell structures*) on elementary calculations aimed as an introduction to comparison of sizes and number of cell components. The book includes several appendices on abbreviations, constants, units, conversion factors, useful formulas and reference data relevant to calculations in Plant Biology.

I hope that the book responds to a real necessity in graduate courses for Science, Agronomy, Ecology and Forestry and helps the future graduates to critically approach agricultural, energy and environmental challenges.

CHAPTER ONE

CELL AND CELL WALL

1a. *Comparative number and size of molecules and cell structures*

1.1.- The matrix of a typical mitochondrion is 1 μm^3 in volume and has a pH of 8.2. How many hydronium ions (H_3O^+) does one mitochondrion matrix have? Avogadro's number N = 6.024×10^{23} molecules/mole.

RESPONSE

The number of hydronium ions is the product of the volume x concentration x N.

Usually, volume is expressed in litres (L) and concentration in moles per litre (molar concentration, M) which for hydronium ions is related to pH by: $M = 10^{-pH}$. Then, the mitochondrial matrix volume is:

$1\ \mu m^3 = (10^{-5})^3\ dm^3$ (or L) = 10^{-15} L.

Therefore, the number of hydronium ion in one mitochondrial matrix is:

$10^{-15} \times 10^{-8.2} \times 6.024 \times 10^{23} = 6.024 \times 0.63096$,

Approximately **3.8 hydronium ions per mitochondrial matrix.**

Obviously, the number of hydronium ions must be an integer around 3 or 4. In fact, the aim of the calculation is to emphasize the low number of molecules or free ions of many compounds present in the small volume of cell organelles like mitochondria.

1.2.- The matrix of a typical mitochondrion has a volume of 1 μm^3 and 1 mM of citrate ion ($C_6H_6O_7^{=}$). How many citrate ions are in one

mitochondrial matrix? Avogadro's number N = 6.024×10^{23} molecules/mole.

RESPONSE

The number of citrate ions is the product of the volume x concentration x N.

Repeating the approach of the calculation in 1.1, volume is expressed in litres (L) and concentration in moles per litre (molar concentration, M). Then, the concentration of citrate is 1 mM = 10^{-3} M. Therefore, the number of citrate ions in one mitochondrial matrix:

$10^{-15} \times 10^{-3} \times 6.024 \times 10^{23} = 6.024 \times 10^{5} =$

602,400 citrate ions per mitochondrial matrix.

Compared with the number of hydronium ions calculated in 1.1, the mitochondrial matrix contains a huge number of citrate ions, but still low for the application of thermodynamic variables to a single mitochondrial matrix. Molar concentrations of most metabolites in the mitochondrial matrix range between 1 μM and 1 mM, that according to calculations like those for citrate, correspond to molecule numbers of each metabolite ranging between 600 and 600,000.

1.3.- The chloroplast stroma is typically 3 μm^3 and has a pH of 8.5. How many hydronium ions (H_3O^+) are in the stroma of one chloroplast? Avogadro's number N = 6.024×10^{23} molecules/mole.

RESPONSE

Repeating the procedure used in 1.1 and 1.2:

The number of hydronium ions in the stroma of one chloroplast:

$3 \times 10^{-15} \times 10^{-8.5} \times 6.024 \times 10^{23}$.

Approximately **5.71 hydronium ions per chloroplast stroma.**

Obviously, the number of hydronium ions must be an integer around 5 or 6. Compared with hydronium ions in the mitochondrial matrix (1.1), the

higher number of hydronium ions in the stroma of one chloroplast is due to the higher volume, partially compensated for by the higher pH (lower concentration of hydronium ions) in the stroma than in the matrix. Again, this calculation emphasizes the low number of molecules of many chemical species in the small volume of cell organelles.

1.4.- A cotton thread weighs 0.2 mg and contains cellulose chains of around 10,000 glucosyl ($C_6H_{10}O_5$) groups as a mean. How many cellulose molecules are in the thread? Atomic masses: H, 1; C, 12; O, 16. Avogadro's number N = 6.024×10^{23} molecules/mole.

RESPONSE

The molecular mass of a cellulose chain is around:

10,000 x (6 x 12 + 10 + 5 x16) = 1,620,000 dalton.

Then, the thread contains 0.2×10^{-3} /1,620,000 moles of cellulose that must be multiplied by Avogadro's number to obtain the number of cellulose molecules:

$6.024 \times 10^{23} \times 0.2 \times 10^{-3}$ /1,620,000.

Approximately **7.5×10^{13} molecules of cellulose in a single thread.**

The result shows the high number of molecules in a cotton thread, barely visible without optical instrument, and emphasises the small size of molecules as large as cellulose when compared with objects within our visual perception.

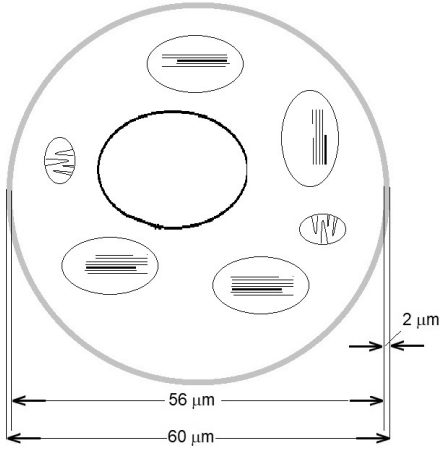
1b. *Cell composition and analysis*

1.5.- A typical mesophyll cell is approximately spherical with 60 μm total diameter, including 2 μm of wall width. Analyses show that water accounts for 82 % of total cell mass and 70 % of isolated cell wall mass. Assuming a cell density of 1 g/cm^3, what is the percentage of water in the mesophyll protoplasm (cell excluding wall)?

RESPONSE

By subtracting the wall width, diameter of the protoplasm sphere is:

60 - 2 x 2 = 56 μm (that is a radius of 28 μm).



MESOPHYLL CELL

Therefore, the wall volume is the difference between two spheres of respective radius 30 and 28 μm.

Total cell volume: $4 \times \pi \times 30^3/3 = 113{,}098\ \mu m^3$.

Protoplasm volume: $4 \times \pi \times 28^3/3 = 91{,}953\ \mu m^3$.

Wall volume = 113,098 - 91,953 = 21,145 μm^3.

As cell and water densities are 1 g/cm^3 and the water content of protoplasm is the difference between total cell and wall water content, for "*p*" the percentage of water in the protoplasm:

91,953 x *p* / 100 = (113,098 x 82 / 100) – (21,145 x 70 / 100); and:

p = (113,098 x 82 -21,145 x 70) / 91,953 = (approximately) **84.8 %**

Although the cell shape is idealised, this result is representative of the relative water contents in protoplasm and walls of mesophyll cells.

1.6.- Cellulose ($(C_6H_{10}O_5)_n$) accounts for 64 % of the dry mass of a sample of purified cell wall. Complete hydrolysis of 1 g of the same sample produces 0.75 g of glucose ($C_6H_{12}O_6$). What is the percentage of the total molecules of glucose in cell wall that are present as cellulose? Atomic masses: H, 1; C, 12; O, 16.

RESPONSE

Firstly, the molecular masses must be calculated, giving 180 for glucose ($C_6H_{12}O_6$) and 162 for glucosyl ($C_6H_{10}O_5$), a unit of cellulose.

Hence, of a total 0.75/180 moles of glucose obtained by hydrolysis of 1 g cell wall, 0.64/162 moles of glucose derive from cellulose. Therefore, the molar percentage of cell wall glucose that is present as cellulose is:

(0.64 / 162) x 100 / (0.75/180) = **94.8 %**

The remaining 5.2 % glucose should correspond to non-cellulosic cell wall polysaccharides.

Note the distinction between the use of glucose and glucosyl masses for proper calculations.

1.7.- A purified preparation of cell wall is composed (mass %) of: water 60 %, polysaccharides 22 %, pectic material 6 %, protein 5 %, lignin 5 % and minerals 2 %. In addition to the molecular mass of water (18), the following average molecular masses are estimated for: polysaccharides 100,000; pectic materials 10,000; proteins 40,000; lignin 500,000 and mineral ions 80. What is the molar proportion of the different components in the cell wall? Calculate the results as the number of molecules of each component per one million (1,000,000) water molecules.

RESPONSE

Let be:

Po: polysaccharide molecules per 1,000,000 water molecules.
Pe: pectic molecules per 1,000,000 water molecules.
Pr: protein molecules per 1,000,000 water molecules.
Li: lignin molecules per 1,000,000 water molecules.
Io: mineral ions per 1,000,000 water molecules.

Hence, per 1,000,000 water molecules:
1,000,000 x 18 = 18,000,000 g water that make 60 % of the total mass
Po x 100,000 g polysaccharides that make 22 % of the total mass
Pe x 10,000 g pectin that make 6 % of the total mass
Pr x 40,000 g protein that make 5 % of the total mass
Li x 500,000 g lignin that make 5 % of the total mass
Io x 80 g ions that make 2 % of the total mass

If 60 % of the total mass is 18,000,000 g (for water), direct proportional calculations show that 22, 6, 5, 5 and 2 % of the total mass must be respectively, 6,600,000, 1,800,000, 1,500,000, 1,500,000 and 600,000 g. Then, dividing by the corresponding molecular masses, the numbers of molecules are:

Po = 6,600,000 / 100,000 = **66** polysaccharide molecules per million water molecules.
Pe = 1,800,000 / 10,000 = **180** pectic molecules per million water molecules.
Pr = 1,500,000 / 40,000 = **37.5** protein molecules per million water molecules.
Li = 1,500,000 / 500,000 = **3** lignin molecules per million water molecules.
Io = 600,000 / 80 = **7,500** mineral ions per million water molecules

The results emphasize that water molecules are by far the most abundant in biological materials.

This math exemplifies the conversion of weight data, obtained experimentally, to molar data that is commonly used in structural and metabolic studies.

1.8.- How many moles of ATP are consumed for the synthesis of one mole of a cellulose composed of 7,500 glucosyl units ($(C_6H_{10}O_5)_{7500}$)? How many molecules of cellulose are in 1 g? Atomic masses: H, 1; C, 12; O, 16. Avogadro's number N = 6.024×10^{23} molecules/mole.

RESPONSE

For the first question. The incorporation of one mole of glucose consumes 2 moles of ATP; one to form glucose-6-phosphate and another, indirectly, for conversion of glucose-1-phosphate to UDP-glucose according to the sequence of reactions:

ATP + glucose ⟶ ADP + glucose-6- phosphate
Glucose-6-phosphate ⟶ glucose-1- phosphate
UTP + glucose-1-phosphate ⟶ PP + UDP-glucose
UDP-glucose + $(glucosyl)_n$ ⟶ UDP + $(glucosyl)_{n+1}$
(enlargement of the cellulose chain)
UDP + **ATP** ⟶ UTP + ADP

Therefore, the 7,500 moles of glucose required to form one mole of cellulose consume 7,500 x 2 = **15,000 moles of ATP**.

For the second question. One glucosyl unit ($C_6H_{10}O_5$) has 162 g molecular mass and the molecular mass of cellulose is 7,500 x 162 = 1,.215 x 10^6 g. This mass contains an Avogadro's number of cellulose molecules.

Therefore, 1 g of cellulose contains (6.024 x 10^{23}) / (1.215 x 10^6) = **4.96 x 10^{17} cellulose molecules**.

A small correction to the ATP consumption and cellulose molecular mass values should be considered because the number of glucose bonds is 7,500 - 1 = 7,499. However, in this case the correction has been omitted due to low impact.

1.9.- By assuming that the consumption of one mole of glucose by glycolysis and respiration produces 36 moles of ATP, and remembering that the incorporation of one glucose molecule into cellulose consumes 2 moles of ATP, how many g of glucose are required in total for the synthesis of 1,000 g of cellulose? Atomic masses: H, 1; C, 12; O, 16.

RESPONSE

Glucose is required for both the generation of ATP and its incorporation into cellulose. For calculations, molecular masses of glucose ($C_6H_{12}O_6$), 180, and of glucosyl units ($C_6H_{10}O_5$), 162, of cellulose must be distinguished and used properly.

Hence, the number of moles of glucosyl in 1,000 g of cellulose is obtained by dividing 1,000 /162, and is also equal to the moles of glucose used as the carbon backbone of cellulose.

One mole of glucose incorporated into cellulose consumes 2 moles of ATP. Therefore, 2 x 1,000 / 162 moles of ATP are required for the biosynthesis of 1,000 g of cellulose. As one mole of glucose must be

degraded by glycolysis and respiration to produce 36 moles of ATP, in addition to those used as the carbon backbone of cellulose, 2 x 1,000 / (162 x 36) moles of glucose are required to synthesize 1,000 g of cellulose. Then, the total moles of glucose required are:

((1,000 / 162) + 2 x 1,000)/ (162 x 36), that multiplied by 180 are the total g consumed of glucose:
180 x ((1,000 / 162) + 2 x 1,000) / (162 x 36)) =

1,172.8 g of glucose are necessary for the synthesis of 1,000 g of cellulose.

1.10.- The analysis of a preparation of the cell wall protein extensin reveals that 8 % of its amino acids are tri-arabinosylated and 5 % are tetra-arabinosylated. How many arabinose molecules does that extensin contain per 100 total amino acids of the molecule?

RESPONSE

Of 100 amino acids, 8 have 3 linked arabinoses and 5 amino acids have 4 linked arabinoses. In total:

8 x 3 + 5 x 4 = **44** units of arabinose for each 100 amino acids.

Arabinoses are linked to hydroxyprolil amino acid units by glycosyl bonds.

1.11.- The hydrolysis of 0.1 g of a preparation of extensin with proteinases produces 4 mg of proline ($C_5NO_2H_9$) and 3 mg of hydroxyproline ($C_5NO_3H_9$). The hydrolysis of 0.1 g of the same preparation of extensin with a mixture of proteinases and glycosidases produces 4 mg of proline and 12 mg of hydroxyproline. What are the molar proportions of proline, hydroxyproline and arabinosylated hydroxyproline in the extensin? Atomic masses: H, 1; N, 14; C, 12; O, 16.

RESPONSE

From atomic masses, molecular masses of proline (115) and hydroxyproline (131) are easily calculated.

The hydrolytic reactions in the assays are

$$\text{extensin (0.1 g)} \xrightarrow{\text{proteinases}} \text{proline (4 mg)} + \text{hydroxyproline (3 mg)} + \text{arabinosylated hydroxyproline} + \text{other amino acids} \quad [1]$$

In moles:

$$\text{extensin (0.1 g)} \xrightarrow{\text{proteinases + glycosidases}} \text{proline (0.004/115)} + \text{hydroxyproline (0.012/131)} + \text{other amino acids} + \text{arabinose} \quad [2]$$

Where proline and hydroxyproline production are indicated in mg and moles.

From reaction [1] it is obvious that extensin contains 0.003/131 mole of non-arabinosylated hydroxyproline per 0.004/115 mole of proline. The difference between reactions [2] and [1] also indicates that (0.012 - 0.003)/131 of hydroxyproline is hydroxylated in extensin. Then, the molar proportion is: 0.004/115 proline per 0.003/131 of non-arabinosylated hydroxyproline and 0.009/131 arabinosylated hydroxyprolines.

To express the results as natural numbers, we first divide all values by the lowest, 0.003/131, obtaining:

1.52 prolines per 1 non-arabinosylated hydroxyproline and 3 arabinosylated hydroxyprolines.

Then, multiplying by 2 and discarding the second decimal, the results show that the analysed extensin contains:

for 3 prolines, 2 non-arabinosylated hydroxyprolines and 6 arabinosylated hydroxyprolines.

1.12.- A cell wall xyloglucan is composed of 30 glucoses, 20 xyloses, 5 galactoses and 3 fucoses. What specific radioactivity (GBq/mole) will be reached when it is synthesized from glucose equally labelled with ^{14}C at its six carbons (1 GBq/mole) and when it is synthesized from glucose with only the carbon at position 6 labelled with ^{14}C (1 GBq/mole)?

RESPONSE

After their synthesis from glucose, all structural units of the xyloglucan except xylose, conserve all six carbons of glucose. However, the synthesis of xylose involves the loss of the carbon at position 6 of the glucose unit.

Thus, the specific radioactivity (GBq/mole) in each structural block will be:

	Specific radioactivity (GBq/mole) in:			
Synthesised from↓	Glucose	Galactose	Fucose	Xylose
Uniformly labelled glucose	1	1	1	5/6
Glucose labelled at position 6	1	1	1	0

Therefore, depending of the label of the precursor glucose, one mole of xyloglucan reaches a radioactivity (specific radioactivity):

Starting from uniformly labelled glucose: 30 + 20 x 5/6 + 5 + 3 = 54.7 GBq.
Starting from glucose labelled at carbon 6: 30 + 5 + 3 = 38 GBq.

1.13.- An *Acer pseudoplatanus* xyloglucan contains 42 units of glucose (180), 30 of xylose (149), 5 of arabinose (149), 7 of galactose (180) and 7 of fucose (164) (molecular masses in parentheses). What is the molecular mass of the xyloglucan? What specific radioactivity will it have (Bq/mole and Bq/g) when it is synthesized from glucose labelled with ^{14}C on C-6 with a specific radioactivity of 1 GBq/mmole and when it is uniformly labelled with the same specific radioactivity?

RESPONSE

The formation of the glycoside bond between two contiguous carbohydrate units implies the loss of one water molecule. The number of glycoside bonds in the xyloglucan is equal to the total number of carbohydrate units minus one. Thus, the molecular mass of the xyloglucan is obtained by summation of the product of the number of each kind of carbohydrate by their molecular mass, and subtraction of the product of the total number of carbohydrate units except one by the molecular mass of water (18). The total number of carbohydrates is:

42 + 30 + 5 + 7 + 7 = 91; and the number of glycoside links 91 - 1 = 90.

Thus, the molecular mass of the hemicellulose is:

(42 x 180) + (30 x 149) + (5 x 149) + (7 x 180) + (7 x 164) - (90 x 18) = **13,563 g.**

Synthesis with glucose labelled at C-6
In the synthesis of xyloglucan with glucose labelled with ^{14}C at C-6, structural blocks of glucose, galactose and fucose, but not those of pentoses xylose and arabinose (that lack the radioactive carbon at position 6 of the original glucose), maintain the radioactivity due to the ^{14}C labelling of the glucose position 6. Thus, 42 + 7 + 7 = 56 blocks account for 56 GBq per mmole of xyloglucan. Hence, the *Acer pseudoplatanus* xyloglucan synthesized from C-6 labelled glucose has a per mmole radioactivity:

56 GBq/mmole.

As one millimole is 13.563 g, the specific radioactivity expressed per g is:

56/13.563 = (approximately) **4.13 GBq/g.**

Synthesis with uniformly labelled glucose
Again, each structural block of glucose, galactose and fucose provides to synthesized xyloglucan 1 GBq per mmole. Now, each pentose block provides to xyloglucan 5/6 GBq per mmole. Hence, total radioactivity per xyloglucan mmole is:
42 + 7 +7 + (30 + 5) x 5/6 = **85.17 GBq/mmole**.

Expressed per g of xyloglucan, 85.170/13.563 = **6.28 GBq/g.**

1.14.- After subcellular fractionation, marker activities of chloroplast (glycolate-phosphatase, GPase) and mitochondria (isocitrate dehydrogenase, IDH) were determined in fractions of partially purified chloroplast and mitochondria obtaining:

	Activity (U/g protein)	
	GPase	IDH
Chloroplast fraction	94	6
Mitochondria fraction	21	84

If there is no other contaminant fraction, calculate the percentage of chloroplast and mitochondrial protein present in, respectively, mitochondrial and chloroplast fractions.

RESPONSE

As pure chloroplast does not contain IDH and pure mitochondria does not contain GPase, data indicate that both chloroplast and mitochondria fractions are not pure. Let "*x*" be the percentage of mitochondria protein in chloroplast fraction, "*y*" be the percentage of chloroplast protein in mitochondria fraction, "*g*" the specific activity (unit/mg protein) of GPase in hypothetical pure chloroplasts, and "*i*" the specific activity of IDH in hypothetical pure mitochondria. According to the data in the table, *x*, *y*, *g* and *i* are related by:

$g = 94 \times 100 / (100 - x)$ [1]
$i = 84 \times 100 / (100 - y)$ [2]
$21 = g \times y / 100$ [3]
$6 = i \times x / 100$ [4]

Substituting of *g* and *i* in, respectively, [3] and [4] by their expressions in [1] and [2]:

$21 = 94 \times x / (100 - y)$
$6 = 84 \times x / (100 - y)$

Or the equivalent:

$2{,}100 = 94 \times y + 21 \times x$
$600 = 6 \times y + 84 \times x$

Solving the system of equations for *x* and *y*, one obtains the results:

***x* (contamination by mitochondria in the chloroplast fraction) = 5.64 % protein**

***y* (contamination by chloroplasts in the mitochondrial fraction) = 21.1 % protein.**

From these results and equations [3] and [4], *g* and *i* can also been calculated with the results:

g (specific activity of GPasa in pure chloroplast) = 99.5 U/g protein

i (specific activity of IDH in pure mitochondria) = 106.4 U/mg protein.

CHAPTER TWO

WATER RELATIONS IN THE CELL AND THE WHOLE PLANT: ABSORPTION, CIRCULATION AND LOSS OF WATER AND NUTRIENTS

2.1.- At 300 K, the cells of a tissue show incipient plasmolysis when exposed to a solution of 200 g of sucrose per litre of water, and a piece of the same tissue does not gain or lose weight when exposed to a solution of 120 g of sucrose per litre of water. Disregarding other components of water potential (Ψ) and cell volume difference between incipient plasmolysis and water equilibrium, calculate the values of Ψ_π (osmotic potential) and Ψ_P (pressure potential). R (gas constant) = 8.3 J mole^{-1} K^{-1}; molecular mass of sucrose 342.

RESPONSE

When the changes to cell volume are ignored, the cell osmotic potential ($\Psi_{\pi c}$) is equal to the external osmotic potential ($\Psi_{\pi e}$, due to the dissolution of 200 g of sucrose per litre of water) at incipient plasmolysis. When cells neither lose nor gain weight they are in water equilibrium with the external environment, and the water potentials of the cells and the external solution are equal ($\Psi_c = \Psi_e$). The only component of Ψ_e is its osmotic potential due to the dissolution of 120 g of sucrose per litre of water. Then, by calculation in the appropriate units (sucrose concentration must be expressed as mole/m^3):

$\Psi_{\pi c}$ = - R x (concentration of sucrose for plasmolysis) x T =
- 8.3 x (200/342) x 1,000 x 300 = - 1,456,140 Pa = - 14.56 bar

Ψ_c = - R x (concentration of sucrose for water equilibrium) x T =
- 8.3 x (120/342) x 1,000 x 300 =- 873,684 Pa = - 8.74 bar

Remembering that $\Psi_c = \Psi_{\pi c} + \Psi_{Pc}$; reordering: $\Psi_{Pc} = \Psi_c - \Psi_{\pi c}$ = - 8.74 - (- 14.56); Ψ_{Pc} = 5.82 bar

In summary, the requested values of cell Ψ_π and Ψ_P are:

Ψ_π = - 14.56 bar and **Ψ_P = 5.82 bar.**

2.2.- The water potential Ψ of a leaf at 300 K is - 3.5 bars. At the same temperature, leaf cells show incipient plasmolysis when exposed to a solution of 114 g of sucrose per litre of water. Disregarding other components of the water potential and differences in volume, calculate the values of osmotic (Ψ_π) and pressure (Ψ_P) potentials in those cells. R (gas constant) = 8.3 J mole^{-1} K^{-1}; molecular mass of sucrose 342.

RESPONSE

At incipient plasmolysis the cell osmotic potential (Ψ_π) is equal to the osmotic potential (or -π) of external solution which is related to the external concentration of sucrose by the formula:

-π = - R x T x concentration of sucrose (mole m^{-3}) =
- 8.3 x 300 x (114 x 1,000 / 342) = 830,000 Pa = - 8.3 bar

As the cell water potential (Ψ) is - 3.5 bar:

$\Psi = \Psi_P + \Psi_\pi$ or, reordering, $\Psi_P = \Psi - \Psi_\pi$ = - 3.5 - (-8.3) = 8.3 - 3.5 = 4.8 bar.

Then, the requested values are:

Ψ_π = - 8.3 bars and **Ψ_P = 4.8 bar.**

2.3.- At 300 K, an osmotic potential of -8.0 bar is estimated for some plant cells. When exposed to water solutions containing different concentrations of sucrose, the weight of the cell mass does not change (they are in hydric equilibrium) with 34.2 g of sucrose per litre of water. Disregarding other components of water potential and cell volume differences, calculate the values of the water potential (Ψ) and pressure potential (Ψ_P). R (gas constant) = 8.3 J mole^{-1} K^{-1}; molecular mass of sucrose 342.

RESPONSE

The concentration of sucrose in the external solution for hydric equilibrium is: 34.2/342 mole/litre = 34,200/342 mole m^{-3} = 100 mole m^{-3}.

When the cells are in hydric equilibrium, their water potential is equal to that of the external solution that has no other component than the osmotic potential. Thus:

Ψ = - R x T x sucrose concentration = - 8.3 x 300 x 100 = - 249,000 Pa = - 2.49 bar.

Ignoring differences in cell volume between incipient plasmolysis and hydric equilibrium:

$\Psi = \Psi_\pi + \Psi_P$; or: $\Psi_P = \Psi - \Psi_\pi$ = -2.49 - (-8.0) = 5.51 bar.

Then, values are:

Ψ = -249.000 Pa = **-2-49 bar** and **Ψ_P = 551,000 Pa** = **5.51 bar**.

2.4.- The water potential of the pith of a carrot root at 300 K is - 4.5 bar. Under the same conditions, the pith is at incipient plasmolysis in a solution of 135 g of sucrose per litre of water. Disregarding other components of water potential and volume changes, calculate the values of potential osmotic (Ψ_π) and pressure (Ψ_P) in the pith cells. R (gas constant) = 8.3 J $mole^{-1}$ K^{-1}; molecular mass of sucrose 342.

RESPONSE

The concentration of sucrose in the external solution required for incipient plasmolysis is: 135/342 mole/litre = 135,000/342 = mole m^{-3}.

At incipient plasmolysis the cellular osmotic potential is equal to the external osmotic potential: - R x T x concentration of sucrose = - 8.3 x 300 x 135,000/342 = - 982,895 Pa; approximately equal to - 9.83 bar.

Ignoring cell volume differences between incipient plasmolysis and equilibrium balance, and remembering $\Psi = \Psi_\pi + \Psi_{Pc}$:

$\Psi_{Pc} = \Psi - \Psi_\pi$ = -4.5 - (-9.83) = 5.33 bar.

Then, **Ψ_π = -9.83 bar** and **Ψ_P = 5.33 bar.**

2.5.- As determined by incipient plasmolysis, the osmotic potential (Ψ_π) of the pith of carrot root at 300 K is -9.0 bar. At this temperature, the pith is in hydric equilibrium (no weight change) after exposure to a solution of 68.4 g of sucrose per litre of water. Disregarding other components of water potential and volume changes, calculate the values of water potential (Ψ) and pressure potential (Ψ_P) in the pith cells. R (gas constant) = 8.3 J $mole^{-1}$ K^{-1}; molecular mass of sucrose 342.

RESPONSE

The concentration of sucrose in the external solution for hydric equilibrium is: 68.4/342 mole/litre = 68,400/342 mole m^{-3}, and its Ψ_π (the single component of its water potential), which is equal to the water potential of the pith, is:

$\Psi_{\pi(\text{of the external solution for hydric equilibrium})} = \Psi_{(\text{pith})} =$
-R x (sucrose concentration) x absolute temperature =
-8.3 x (68,400 / 342) x 300 = -498,000 Pa = -4.98 bar.

On the other hand, $\Psi_P = \Psi - \Psi_\pi$ = -4.98 - (-9) = 4.02 bar.

Therefore, values are:

Ψ = - 4.98 bar and **Ψ_P = 4.02 bar.**

2.6.- At 300 K, some cells are at incipient plasmolysis when exposed to a solution of 171 g of sucrose per litre of water and neither gain nor lose water when exposed to a solution of 114 g of sucrose per litre of water. Disregarding other components of water potential and volume changes, calculate the values of water potential (Ψ) and pressure potential (Ψ_P) in the cells. R (gas constant) = 8.3 J $mole^{-1}$ K^{-1}; molecular mass of sucrose 342.

RESPONSE

The cell osmotic potential ($\Psi_{\pi c}$) is equal to the external osmotic potential ($\Psi_{\pi e}$) (due to the dissolution of 171 g of sucrose per litre of water) at incipient plasmolysis because at incipient plasmolysis pressure potential is 0 in the cells and the external environment. When cells neither lose nor gain water they are at water equilibrium with the external solution ($\Psi_{\pi c}$ =

$\Psi_{\pi e}$). In the latter case the sole component of $\Psi_{\pi e}$ is osmotic potential due to the dissolution of 114 g of sucrose per litre of water.

Disregarding the differences of cell volume between incipient plasmolysis and water equilibrium and expressing equations in appropriate units (SI, International System; E.g.: the concentration of sucrose in mole m^{-3}):

$\Psi_{\pi c}$ = -R x (sucrose concentration for incipient plasmolysis) x T = -8.3 x (171 / 342) x 1,000 x 300 = -8.3 x (1 / 2) x 1,000 x 300 = -1,245,000 Pa = -12.45 bar.

Ψ_c = -R x (sucrose concentration for hydric equilibrium) x T = -8.3 x (114/342) x 1,000 x 300 = -8.3 x (1 / 3) x 1,000 x 300 = - 830,000 Pa = -8.3 bar.

As $\Psi_c = \Psi_{\pi c} + \Psi_{Pc}$; then: $\Psi_{Pc} = \Psi_c - \Psi_{\pi c}$ = -8.3 - (-12.45) = 4.15 bar = 415,000 Pa.

In conclusion:

Ψ_π = -1,245,000 Pa = -12.45 bar, and **Ψ_P = 415,000 Pa = 4.15 bar.**

2.7.- At 300 K, water potential (Ψ) and matric potential (Ψ_τ) of some cells are - 0.40 MPa and - 0.5 bar, respectively. These cells show incipient plasmolysis when exposed to a solution of 136.8 g of sucrose per litre of water. Disregarding volume changes and other components of water potential, calculate the values of the osmotic water potential (Ψ_π) and pressure potential (Ψ_P) in these cells. R (gas constant) = 8.3 J $mole^{-1}$ K^{-1}; molecular mass of sucrose 342.

RESPONSE

In the International System of units (SI), the concentration of sucrose for incipient plasmolysis is 136.8 x 1,000 / 342 mole m^{-3} = 400 mole m^{-3}.

According to the extended formula for water potential:

$$\Psi = \Psi_P + \Psi_\pi + \Psi_\tau$$

where: Ψ_τ = -0.5 bar = -50,000 Pa and Ψ = -0.4 MPa = -400,000 Pa.

The summation of the cell $\Psi_\pi + \Psi_\tau$ must equal to the Ψ_π of the solution at which the cells show incipient plasmolysis, because under this condition Ψ_P is 0 inside and outside of the cells. Then:

$\Psi_{\pi cell}$ - 50,000 = -R x T x (sucrose concentration for incipient plasmolysis) = -8.3 x 300 x 136.8 x 1,000 / 342 = -996,000 Pa, and

$\Psi_{\pi cell}$ = -996,000 + 50,000 = -946,000 Pa = -9.46 bar, and

$\Psi_{cell} = \Psi_{Pcell} + \Psi_{\pi cell} + \Psi_{\tau cell}$; or: $\Psi_{Pcell} = \Psi_{cell} - \Psi_{\pi cell} - \Psi_{\tau\text{-celular}}$ = -400,000 - (-946,000) - (- 50,000) = 596,000 Pa = 5.96 bar.

Summarising:

$\Psi_{\pi cell}$ = -946,000 Pa = -9.46 bar and **Ψ_{Pcell} = 596,000 Pa = 5.96 bar**.

2.8.- At 300 K, the cells of a certain plant tissue show incipient plasmolysis when exposed to a solution of 136.8 g of sucrose per litre of water and are in hydric equilibrium balance (neither gain nor lose weight) when exposed to another solution of 102.6 g of sucrose per litre of water. Estimating the matric potential (Ψ_τ) at about -50,000 Pa and disregarding the changes in volume and value of other components of water potential, calculate the values of Ψ, Ψ_π and Ψ_P in those cells. R (gas constant) = 8.3 J mole^{-1} K^{-1}; molecular mass of sucrose 342.

RESPONSE

Expressed in SI units, the concentrations of sucrose are:

for incipient plasmolysis: 136.8 x 1,000 / 342 mole m^{-3}.
for hydric equilibrium: 102.6 x 1,000 / 342 mole m^{-3}.

According to equation for water potential: $\Psi = \Psi_P + \Psi_\pi + \Psi_\tau$, cell Ψ is equal to that of the solution for hydric equilibrium which has no other component than its osmotic potential:

$\Psi_{cell} = \Psi_{\pi eq\text{-}hi}$ = -R x T x (sucrose concentration for hydric equilibrium) = -8.3 x 300 x 102.6 x 1,000 / 342 = -747,000 Pa = -7.47 bar.

As $\Psi_{\tau cel}$ = -50,000 Pa, the value of $\Psi_{\pi cell} + \Psi_{\tau cel}$ is the same as the Ψ_π of the solution for incipient plasmolysis, when $\Psi_P = 0$ inside and outside

cells. Therefore: $\Psi_{\pi cell} + \Psi_{\tau cell} = \Psi_{\pi}$ of the solution for incipient plasmolysis. That is:

$\Psi_{\pi cell} - 50{,}000 = -R \times T \times$ (sucrose concentration for incipient plasmolysis) $= -8.3 \times 300 \times 136.8 \times 10^3 / 342 = -996{,}000$ Pa; and: $\Psi_{\pi cell} = -996{,}000 + 50{,}000 = -946{,}000$ Pa $= -9.46$ bar.

As $\Psi_{cell} = \Psi_{Pcell} + \Psi_{\pi cell} + \Psi_{\tau cell}$; $\Psi_{Pcell} = \Psi_{cell} - \Psi_{\pi cell} - \Psi_{\tau cell} = -7.47 - (-9.46) - (-0.5) = 2.49$ bar.

Summarising:

Ψ_{cell} = -747,000 Pa = -7.47 bar, $\Psi_{\pi cell}$ = -946,000 Pa = -9.46 bar, Ψ_{Pcell} = 249,000 Pa = 2.49 bar.

It must be noted that the inclusion of $\Psi_{\tau cell}$ in Ψ_{cell} does not affect the calculation of Ψ_{Pcell} from the experimental determination of Ψ_{cell} and $\Psi_{\pi cell}$.

2.9.- At 300 K, a plant tissue has a water potential of -4.0 bar and it is at incipient plasmolysis when exposed to a solution of 120 $\pm$ 3 g of sucrose per litre of water. Disregarding other components of water potential, calculate values ($\pm$ standard error) of $\Psi_{\pi c}$ and Ψ_{pc} of the cells of the tissue. R (gas constant) = 8.3 J mole^{-1} K^{-1}; molecular mass of sucrose 342.

RESPONSE

Disregarding the cell volume contraction when the cell is subjected to incipient plasmolysis, the osmotic potential of the cell ($\Psi_{\pi c}$) equals the external osmotic potential due to the 120 $\pm$ 3 g sucrose per litre of water. Therefore:

$\Psi_{\pi c} = -$ R x (sucrose concentration for incipient plasmolysis) x T $= -8.3 \times [(120 \pm 3)/342] \times 1{,}000 \times 300 = -873{,}684.2 \pm 21{,}842.1$ Pa = (approximately) $= 8.737 \pm 0.218$ bar.

As $\Psi_c = \Psi_{\pi c} + \Psi_{Pc}$, and Ψ_c is the water potential of the tissue: $\Psi_{Pc} = \Psi_c - \Psi_{\pi c} = -4 - (-8.737 \pm 0.218) = 4.737 \pm 0.218$ bar.

Summarising:

$\Psi_{\pi c}$ = - 8.737 ± 0.218 bar and **Ψ_{Pc} = 4.737 ± 0.218 bar.**

2.10.- At 300 K, cells of some plant tissue show incipient plasmolysis when exposed to a solution of 130 ± 5 g of sucrose per litre of water and are in hydric equilibrium (neither gain nor lose weight) when exposed to another solution of 90 ± 3 g of sucrose per litre of water. Disregarding the other components of water potential and volume differences, calculate the values (± SE) of Ψ, Ψ_π y Ψ_P. R (gas constant) = 8.3 J $mole^{-1}$ K^{-1}; molecular mass of sucrose 342.

RESPONSE

Ignoring differences of cell volume between incipient plasmolysis and hydric equilibrium, the cell osmotic potential ($\Psi_{\pi c}$) equals the external osmotic potential ($\Psi_{\pi e}$, due to dissolution of 130 ± 5 g of sucrose per litre of water) in incipient plasmolysis when the pressure potential is 0 inside and outside of the cells. In the other case, when cells are in hydric equilibrium with the external solution, values of the water potential of the cells (Ψ_c) and of the external solution (Ψ_e) are equal ($\Psi_c = \Psi_e$). Under the last condition the only component of Ψ_e is its osmotic potential due to the dissolution of 90 ± 3 g of sucrose per litre of water.

Expressed with appropriate units:

$\Psi_{\pi c}$ = -R x (sucrose concentration for incipient plasmolysis) x T = -8.3 x ((130±5) / 342) x 1,000 x 300 = -946,491±36,403 Pa = (approximately) -9.465 ± 0.365 bar.

Ψ_c = -R x (sucrose concentration for hydric equilibrium) x T = -8.3 x ((90±3) / 342) x 1,000 x 300 = -655,263±21,842 Pa= (approximately) -6.553 ± 0.218 bar.

As both summation and subtraction add errors and remembering $\Psi_c = \Psi_{\pi c} + \Psi_{Pc}$,

then $\Psi_{Pc} = \Psi_c - \Psi_{\pi c}$ = -6.553 - (-9.465) ± (0.364 + 0.218) = 2.912 ± 0.582 bar.

Summarising:

Ψ_π = -9.465 ± 0.364 bar and **Ψ_P = -2.912 ± 0.582 bar.**