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Comparative analysis of water potential of plant tissues and its relation to Chlorophyll Stability Index and heat stress

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ABSTRACT

Plant products easily damage due to many reasons and one may be the susceptibility nature to diseases due high content of water. This research analyses the water potential of plant tissues and its relation to heat stress. Methods: Fresh, clean and healthy plants, Carrot (*Daucus carota subsp. Sativus*), Tomato (*Solanum lycopersicum*), Sweet potato (*Ipomoea batatas*), Capsicum (*Capsicum annuum*), Potato (*Solanum tuberosum*) and Cucumber (*Cucumis sativus*) were analyzed under different concentrations of sucrose solution for the calculation of water potential followed by the calculation of chlorophyll, carotenoid, and chlorophyll stability index for spinach, *Spinacia oleracea* as model for the study of heat stress using spectrophotometer. Results: Tomato tissue has lowest solute content and high-water potential and sweet potato tissue has highest solute content and lowest water potential. Spinach being leafy vegetable have very high solute content and high chlorophyll stability index. Conclusion: High water potential refers the availability of water in plant products such as fruits, vegetables etc. Solute concentration in tissues is inversely proportional to water potential, higher the solute concentration higher is the chlorophyll stability index and higher is the heat stress tolerance apart from the other factors affecting stress tolerance.

Introduction

Mostly, plants are distributed depending upon the availability of temperature, water content etc. [1] and these factors are most important for the full growth and development of plants apart from nutritional elements [2]. Water is driven by a water potential gradient from high to low water potential regions due to the cause by osmosis, gravitation, matrix potential etc. each alone or combined. However, plants have developed many mechanisms to hold water to survive under various stress conditions [3, 4], specifically superoxide dismutase as an anti-oxidant protector system for plants under water stress conditions [5] etc. Productivity of plants depend on water availability or deficit and the latter is caused by drought which is established due to the difference in water absorption from root and the water demand of the plant in aerial parts [6]. Moreover, water deficit directly affects the expression level of some of the responsible genes. Carbon starvation may take place if the plant is highly under water deficit for long time[7] and therefore, water is required for every growth phase of plant. Post-harvest fruits, vegetables, seeds hold water content for survival to maximum days for their daily metabolic activities such as photosynthesis, enzyme dilution etc. In this research, we have analyzed the role of water potential in different conditions of plant tissues.

Methodology

Fresh, clean and healthy six rooting and fruiting body of six plants, Carrot (*Daucus carota subsp. Sativus*), Tomato (*Solanum lycopersicum*), Sweet potato (*Ipomoea batatas*), Capsicum (*Capsicum annuum*), Potato (*Solanum tuberosum*) and Cucumber (*Cucumis sativus*) were collected for the calculation and comparison of water potentials in different tissues. The tissues were cut into small sizes of cylinders of 3 cm diameter weighing approximately in the range 1.5 gm to 4 gm. Sucrose solution of different concentrations were accurately prepared and labeled as 0.2M, 0.3M, 0.5M, 0.7M and 0.9M or other set of concentrations. Using digital analytical balance, a pair of cylinder of each plant tissue, for example Carrot (*Daucus carota subsp. Sativus*) was weighed nearest to 0.01 gm and immediately immersed into the already prepared and labeled respective concentration of sucrose solution, 0.2M, 0.3M, 0.5M, 0.7M and 0.9M and this is repeated for all plant tissues. After 45 minutes, the pair of cylinders from each concentration per plant was removed, quickly blotted and weighed. Repeated with other plant tissues and the data obtained were entered in the tables 2 to 7 under respective heads. From the data change of weight, percentage of change of weight and respective graphs were drawn taking concentration of sucrose on x-axis and percentage change of weight on y-axis. A regression best fit line was obtained with a respective equation. From the graph, the concentration of sucrose solution at which zero percentage of change of weight of tissue was found. This is because at this concentration solute potential of tissue and sucrose solution are equal which is also equal to water potential of the plant tissues. Similarly, spinach leaf tissue was taken for calculating water potential, chlorophyll content [8] and carotenoids [8]. Chlorophyll stability index was also calculated as per a standard protocol [9].For extraction, 80% acetone was used which was good for spectrophotometer extraction of chlorophyll [10] due to high accurate peak.

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Table 1. Ψ_w (MPa) of all six tissues.

S.No	Scientific Name of the Plant	Common Name	Part of the Plant	Value of C (mol/m^3)	Ψ_w (MPa)
1	*Solanum lycopersicum*	Tomato	Fruit	100	-0.25
2	*Capsicum annuum*	Capsicum,	Fruit	420	-1.04
3	*Cucumis sativus*	Cucumber,	Fruit	520	-1.30
4	*Solanum tuberosum*	Potato	Tuber	500	-1.24
5	*Daucus carota subsp. Sativus*	Carrot	Root	540	-1.34
6	*Ipomoea batatas*	Sweet potato	Tuber	1200	-3.00

Result

Three fruiting bodies and three rooting bodies were obtained for analysis to compare which part of the plant has more water potential and solute potential. The plants were selected according to the availability in the season and the feasibility for the experiment, suitability to cut into cylinders for the experiment etc. The respective concentration of sucrose (C) by which no more weight change takes place is obtained from the regression line graph when the best fit line cuts the x-axis. Using the C values, solute potential and water potential were calculated with the help of an equation, Equ.1, table 2 to 7 and figures 1 to 6. From all the tables, a comparative analysis is made to find the water potential of different tissues, table 1.

Table 2 .Tomato, Solanum lycopersicum.

	0.2M	0.3M	0.5M	0.7M	0.9M
Initial Weight	2.1	2.15	2.22	2.3	2.55
Final Weight	2	2.1	2.06	2.02	2.27
Change in Weight	-0.1	-0.05	-0.16	-0.28	-0.28
Percentage Change in Weight	-4.7619	-2.32558	-7.20721	-12.1739	-10.9804

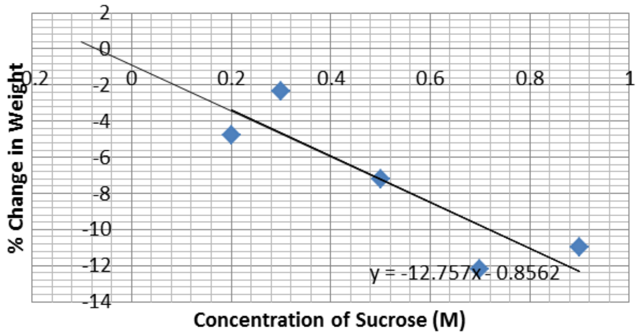

Figure 1. Tomato, Solanum lycopersicum, C= 0.1M, Ψ_w = -0. 25MPa.

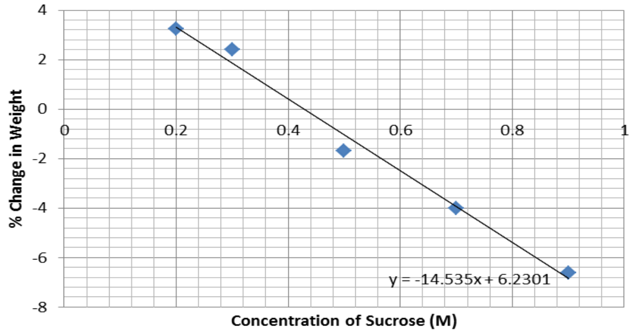

Figure 2. Capsicum, Capsicum annuum, C= 0.42M, Ψ_w = -1.04 MPa.

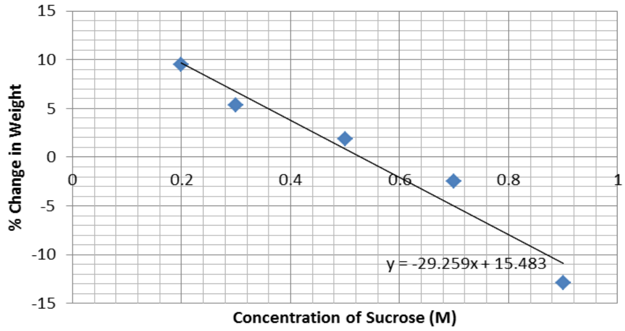

Figure 3. Cucumber, Cucumis sativus C= 0.52M, Ψ_w = -1.3 MPa.

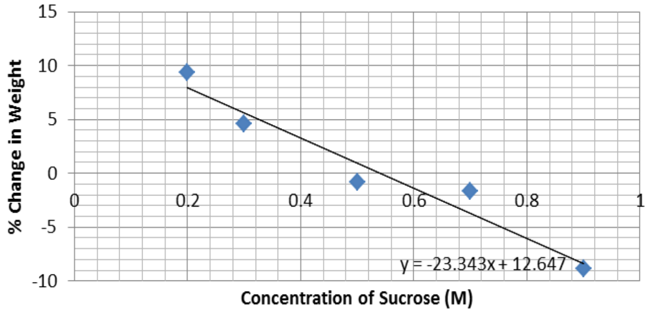

Figure 5. Carrot (Daucus carota subsp. Sativus),. C= 0.54M, Ψ_w = -1.34 MPa.

Table .3. Capsicum, Capsicum annuum.

	0.2M	0.3M	0.5M	0.7M	0.9M
Initial Weight	1.23	1.25	1.19	1.25	1.21
Final Weight	1.27	1.28	1.17	1.2	1.13
Change in Weight	0.04	0.03	-0.02	-0.05	-0.08
Percentage Change in Weight	3.252033	2.4	-1.68067	-4	-6.61157

Table. 4. Cucumber, Cucumis sativus.

	0.2M	0.3M	0.5M	0.7M	0.9M
Initial Weight	3.397	2.963	3.235	3.744	3.378
Final Weight	3.719	3.122	3.296	3.65	2.943
Change in Weight	0.322	0.159	0.061	-0.094	-0.435
Percentage Change in Weight	9.478952	5.366183	1.885626	-2.51068	-12.8774

Table .5. Potato, Solanum tuberosum.

	0.2M	0.3M	0.5M	0.7M	0.9M
Initial Weight	2.66	2.93	3.12	2.67	1.97
Final Weight	2.99	3.19	3.1	2.4	1.65
Change in Weight	0.33	0.26	-0.02	-0.27	-0.32
Percentage Change in Weight	12.40602	8.87372	-0.64103	-10.1124	-16.2437

Table 6. Carrot, *Daucus carota* subsp. Sativus.

	0.2M	0.3M	0.5M	0.7M	0.9M
Initial Weight	3	2.81	2.32	2.35	2.26
Final Weight	3.28	2.94	2.3	2.31	2.06
Change in Weight	0.28	0.13	-0.02	-0.04	-0.2
Percentage Change in Weight	9.333333	4.626335	-0.86207	-1.70213	-8.84956

Table 7 .Sweet potato, Ipomoea batatas.

	0.2M	0.3M	0.5M	0.7M	0.9M
Initial Weight	2.78	2.62	2.46	2.9	3.33
Final Weight	2.97	2.75	2.53	2.98	3.38
Change in Weight	0.19	0.13	0.07	0.08	0.05
Percentage Change in Weight	6.834532	4.961832	2.845528	2.758621	1.501502

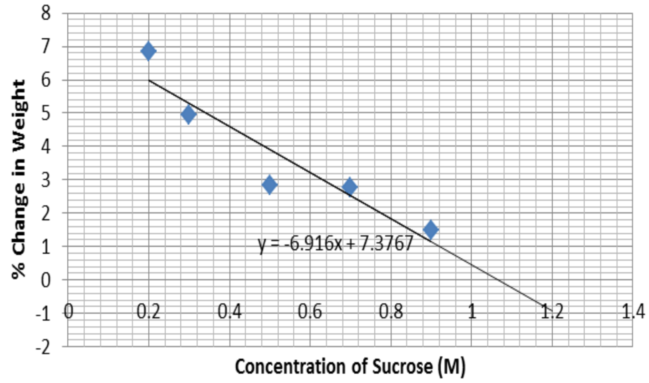

Figure 6. Sweet potato, Ipomoea batatas C= 1.2M, Ψ_w = -3.0 MPa.

Calculations of Ψ_w

Ψ_w = -CiRT ……………………………………..(Equation 1)

C = molarity

i = ionization constant = 1 for sucrose

R = gas constant = 8.31 J K^{-1} mol^{-1}

T = room temperature in K (ºC + 273 = K)

$$\Psi_w = \frac{-0.5 * 1000 \text{ mole}}{m^3 * 1000000} * \frac{1 * 8.31 \text{ J} * 298 \text{ K}}{\text{K} * \text{mol}} = -1.24 \text{ MPa}$$

(temperature is 25 degree Celsius)

Similar way, others were calculated.

Further experiment was done to analyze whether water potential depends on the size of tissues taken for experiment, for this, potato alone was used to find the water potential at different sizes such as large pieces and small pieces etc. and it was found that the water potential was independent of amount of tissue, size of tissue taken etc., data not attached.

Analysis on Spinach, *Spinacia oleracea*

Further analysis was done on spinach leaves for water potential which has 1.1M solute concentration with -2.74 MPa water potential. The high solute concentration may be due to the photosynthetic ability of the leaves. In addition to this, chlorophyll content, carotenoids were estimated to know the photosynthetic effect of the plant, table 9.

Table 8. Percentage change of weight of leaf disc of Spinach in Sucrose solution.

	0.2M	0.3M	0.5M	0.7M	0.9M
Initial Weight	0.225	0.36	0.249	0.285	0.263
Final Weight	0.362	0.49	0.318	0.35	0.302
Change in Weight	0.137	0.13	0.069	0.065	0.039
% change	60.9	36.1	27.7	22.8	14.8

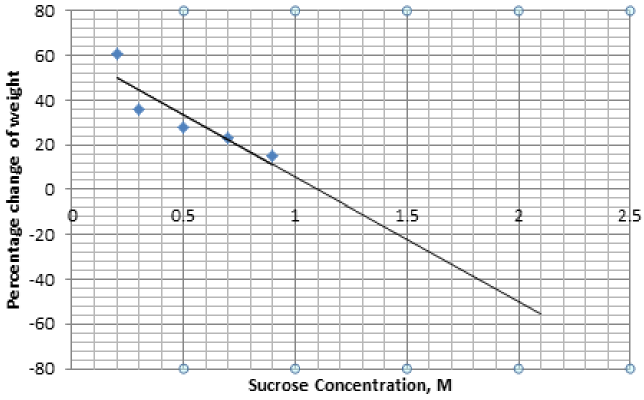

Figure 7. Spinach, Spinacia oleracea C= 1.1M, Ψ_w = -2.74 MPa.

Table 9. Chlorophyll, Carotenoid and anthocyanin content of Spinach leaf tissue.

Total Chlorophyll content (mg/gm)	1.426852
Chlorophyll a content (mg/gm)	0.686686
Chlorophyll b content (mg/gm)	0.672812
Total carotenoids (mg/gm)	0.037

The chlorophyll stress was calculated after exposing the tissues to heat and again measured and the absorbance at 660 nm was 0.491 (40.84%)under heat stress against 0.830 at normal making the chlorophyll index as 0.34 which reflects a better stability of chlorophyll content, which is equal to 40.84%.

Discussion

The vegetables easily damage due to the high-water content **[11],** high water potentials. Tomato tissues have low, 0.1M solute concentration with the water potential -0.25MPa (Mega Pascals), sweet potato tissue has highest, 1.2 M solute concentration with the water potential -3.0 MPa etc. table 1. Similary all the tissues of the plants have water potential or solute concentration between these two range table1. The water potential of tomato in this research is lower than other different studies [12] because water potential depends on many other factors [13]. When tomato fruit tissues is kept for more than three days, it becomes susceptible to fungi, *Rhizopus sps*. and gets damaged in most of the seasons due to high water content which support the growth of the fungi, data not shown. On the other hand, sweet potato, potato etc. remain for some more days without damage due to the more accumulation of solute content and this is in-agreement with many other studies [14, 15]. Moreover, sweet potato, potato, carrot etc. have high solute concentration [15] in their tissues so that they are not so susceptible to the microorganisms because when single celled organisms infect such edible fruiting bodies (soil bacteria etc.), their cells get plasmolyzed by ex-osmosis and the tissues may be alive for long time.

The spinach tissues look flexible and soft like tomato, capsicum etc. and the analysis shows that spinach leaf has high solute concentration 1.1M with water potential -2.74 which is higher than the soft vegetables, table 8. This is because of high photosynthetic effect of the chlorophyll and carotenoid content and which is estimated to be as total chlorophyll content of 1.4 mg/gm and total carotenoid 0.037mg/gm. This research result is same as the others [16] and the other pigments, chlorophyll a, b etc. are also in line with the standard as per the other researches however, due to shade, stress etc. the content of chlorophyll a, b and carotenoids may vary, table 9. Moreover, the stability of chlorophyll of spinach under heat stress showed betted result in this research.

This result is same as in other studies and it has been shown that spinach is highly salt tolerant which is only possible due to the high solute content [17].

The high chlorophyll content determines the chlorophyll stability index under stress condition [18]. The chlorophyll stability index of spinach is 40.84% under heat stress which is in agreement with many results even in hybrids [19], salinity as stress and during stress period plants have some adjustment in osmotic potentials [20]. This is the indication that spinach can survive under scarce conditions of water and many nutrients due to better chlorophyll stability index. The carotenoid content helps in photosynthesis along with chlorophyll [21] and the variation of carotenoids and chlorophyll content is the indication of environmental pollution [22].

Conclusion

High water potential refers the availability of water in plant products such as fruits, vegetables etc. Solute concentration in tissues is inversely proportional to water potential, higher the solute concentration higher is the chlorophyll stability index and higher is stress tolerance apart from the other factors affecting stress tolerance.

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