

STUDIES ON REHYDRATION OF VEGETABLES

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Rehydration capacities of cabbage, potato, turnip and onion, when dehydrated at 37°C to various levels of hydration, were studied. None of the vegetables, dehydrated to any degree, exhibited 100% rehydration. The greater the degree of dehydration, the less complete was the degree of rehydration. There seems to exist a critical point during dehydration of each vegetable beyond which a significant loss in rehydration capacity is apparent. These points for cabbage, potato, turnip and onion were respectively 43, 31, 63 and 45% of their fresh weight. Dehydration to more than 40% of the fresh weight inactivated the reducing enzyme of respiring system. Reasons for the failure of dehydrated tissue to regain the original weight have been discussed in terms of events that affect the structure and function of cellular components.

Key Words. Dehydration, Rehydration, Vegetable.

Introduction

In an earlier paper it was mentioned that desiccation conditions introduce a number of changes in the plant particularly at the sub-cellular level [1]. The cell wall extensibility factor m in Lockhart's equation (1) has been related to the degree of dehydration and hence to the amount of hydrogen bondable sites, ions and/or organic substances present in the cell:

$$\frac{dv}{dt} = m (\psi_p - Y) \dots\dots\dots (1)$$

According to the interpretation of Lockhart's equation by this theory, aging or reduction in growth induced by ionic and/or hormonal inter-action can be quantitatively related to a decrease in cell wall extensibility (m) or decrease in turgor pressure (ψ_p) or in an increase in the cell wall yield threshold (Y). It has been inferred that increase in ionic concentration or decrease in moisture content leads to the reduction in extensibility by introducing crosslinks in the proteinous cell wall accompanied by a loss of one water molecule at each such formative event. Reduction in moisture content under persistent desiccation conditions reduces the turgor pressure ψ_p with a consequent increase in Y , the cell wall yield threshold. It has therefore been suggested that dehydration would lower the hydrogen bondability of the cell wall by denaturation of proteins, through loss of enzymic activity induced by lowering of relative turgidity by increase of ionic concentration or by hormonal interaction.

The above interpretation of Lockhart's equation is quite simple and should explain the observation that dehydrated vegetables do not regain their original volume on rehydration. This mechanism, however, does not seem to have been clearly spelled out in the literature. It seems to provide the main factor responsible for the changes in physical and chemical properties leading to inhibition of the function to regain the original weight.

The mechanisms so far proposed to explain the structural changes introduced in plant tissues as a result of dehydration which prevents them from regaining their original weight and functionality include : collapse and over-lapping of cell wall [2], loss of viability [3], loss of osmotic properties and differential permeability of protoplasmic membrane [4,5], crystallization of polysaccharide gels in the cell wall [6,7] and coagulation of protoplasmic proteins [5]. These mechanisms can be interpreted by the present theory since it considers the formation of crosslinks, fusion of the lipid bilayer, loss of enzymic activity and changes of state of cellulose from amorphous to crystalline to be due to dehydration.

Dehydration as a general mechanism affects the integrity of cellular organelles. The metabolic functions which control the degree of hydration in the cell and its ionic balance by virtue of turgor pressure and osmotic potential are progressively reduced and ultimately lost. As a result the respiratory enzymic activity would also be reduced to zero. The phenomenon of rehydration would, under the stated conditions, be independent of Lockhart's equation and may not be governed by an increase of turgor pressure and/or osmotic potential but by simple absorption and adsorption processes. The degree of hydration at this stage would depend on the hydrogen bondable sites left over after the collapse of cell wall and fusion of the lipid bilayer etc. during dehydration.

Haas *et al.* [8] concluded that damage to tissue during dehydration becomes severe as drying progresses. No conclusion was, however, drawn about the point of no return after which the vegetables could be dehydrated without suffering from the inability to rehydrate completely. This study has been undertaken to demonstrate the existence of a critical point beyond which the cellular functionality is adversely affected. This was done for four vegetables by recording their degree of dehydration (expressed as % of fresh weight) at 37°C and correlating it with the regain of moisture (rehydrated weight).

Correlation was also made between degree of dehydration and percent relative turgidity and loss of the respiratory enzyme activity.

Experimental

Drying experiments were carried out on cabbage, onion, turnip and potato. They were purchased locally and their varieties were not known. Cabbage leaves were cut into pieces of (2-3) x (2-3) cm size while other three vegetables were peeled and sliced uniformly with an electrically driven circular blade. Thickness of each slice was 3 mm. During slicing of onion bulbs, attempt was made to obtain rings rather than slices of irregular shape. One hundred gram portions of cut/sliced vegetables were spread on wire-mesh trays and subjected to dehydration in a cabinet drier (Model : Mitchell Dryers No.6298/59 Manchester) at a dry bulb temperature of $37 \pm 2^\circ\text{C}$ to varying degree of desiccation. Original moisture of each sample was determined according to A.O.A.C. [9]. From the original moisture and the moisture loss at a particular stage of desiccation, the moisture content of the sample at a given stage of desiccation was calculated. "Degree of dehydration" was expressed as the percent loss in vegetable weight upon dehydration.

For rehydration, partially dried samples were soaked in excess tap water (3-8 fold on fresh weight basis) at room temperature (22°C). Weight of rehydrated vegetable samples was determined after draining the water and wiping off superficial moisture gently with a blotting paper. The samples were returned back to the drained water for further rehydration. Rehydrated weights were thus determined after 1,2,3,4,6 and 24 hr of rehydration.

The presence of respiratory activity in the vegetable tissues at various stages of dehydration was evaluated using an oxidation-reduction indicator, triphenyl tetrazolium chloride (TTC). An active system of respiratory reducing enzymes will result in the formation of the formazon derivative, a red stain, which can be observed in the tissue [8].

Percent relative turgidity was calculated as follows:

$$\text{Relative turgidity (\%)} = \frac{\text{Turgid wt.* of stressed (dehydrated) material}}{\text{Turgid wt. of unstressed (fresh) material}} \times 100$$

Results and Discussion

Results of dehydration of the vegetables to various degrees demonstrate that in all cases, rehydration to the level of fresh material is never achieved. (Fig. 1) rehydration curve of onion; the curves of cabbage potatoes and turnips have similar

* Rehydrated weight after 4hr soaking on water was taking as turgid wt.

pattern). However, the fresh material itself can be hydrated to a level above its original weight suggesting that the fresh material is not fully turgid. Similar results were also obtained by earlier workers [8] and therefore the data so far available suggest that the greater the degree of drying the less complete is rehydration. The curves correlating rehydration time and rehydrated weight have more or less, the same pattern as that of the curves of rehydration obtained by Haas [8] *et al.* Shimazu and Sterling [10] and Daud and Luu, [11] for green beans, bell peppers and carrots although the temperature at which the samples were dried $28 \pm 2^\circ\text{C}$. Based on all these data it is possible to say the rehydration values are different under different conditions of dehydration as well as rehydration and also for different varieties and stages of maturity of the vegetables.

The rehydrated weights (4 hr soaking) plotted against the degree of dehydration (expressed as % loss in fresh weight of the vegetable) are shown in Fig. 2 and 3, from where it would be noticed that the loss in rehydration capacity increases continuously with the degree of dehydration, and the actual rehydration line deflects away from the expected line of rehydration. This occurs up to a point whereafter there is further deflection to yield another straight line. The region of the points marking the second deflection seems to signify a 'critical point' of dehydration beyond which loss in rehydration capacity is substantially increased. The criticality of the phenomenon is observed in all the four vegetable and also for those studied but not identified by Hass *et al.* [8]. The critical point perhaps marks the stage of degeneration of the cell wall and is accompanied by cross-linkages leading to crystalliza-

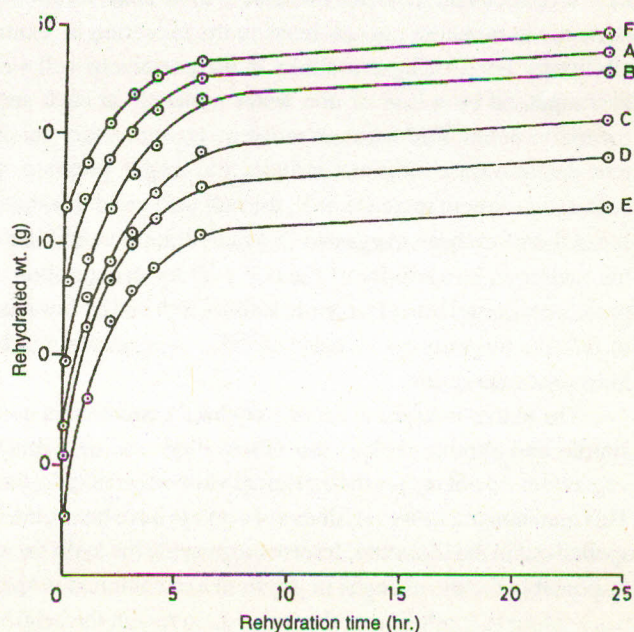


Fig. 1. Rehydration of partially dried onion.

tion of cellulose as observed by Shimazu and Sterling [10] and Shimazu *et al.* [12].

The critical stage of dehydration could serve as a useful guide to which a vegetable should preferably be dehydrated with no appreciable loss of rehydration characteristics, particularly for preservation such as by dehydro-freezing and freeze-drying. For cabbage, potato, turnip and onion, these particular points were 43, 31, 63 and 45% of fresh weight respectively (Fig. 2 and 3). Although the curves shown were based on 4 hour rehydration weights (representing 70-90% of total water absorbed in 24 hr), similar results were obtained if

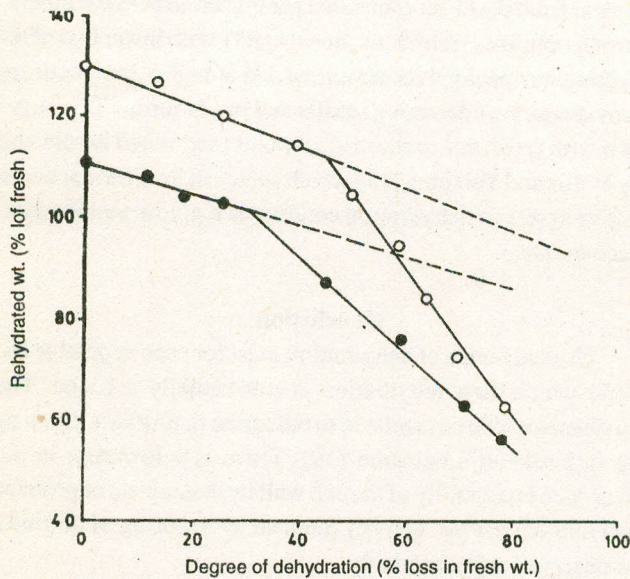


Fig. 2. Effect of the degree of dehydration of cabbage (—o—) & potatoes (—●—) on rehydration capacity.

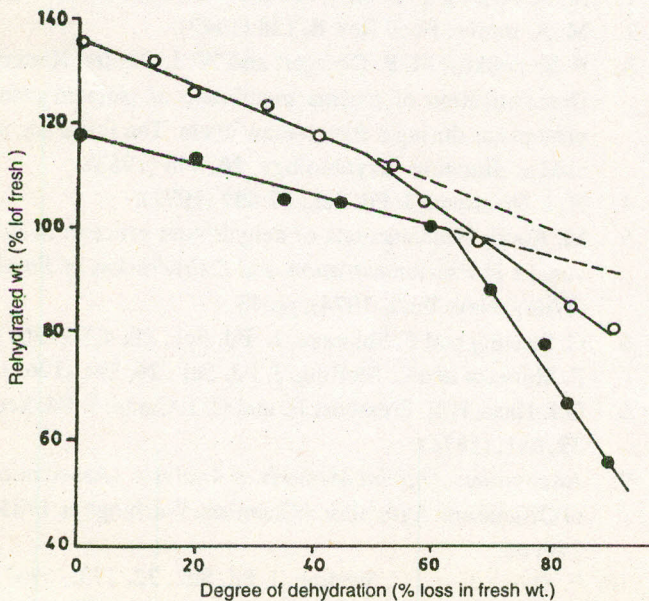


Fig. 3. Effect of the degree of dehydration of onion (—o—) and turnips (—●—) on rehydration capacity.

6 hr or 24 hr rehydration weights were plotted; the points might alter if the drying conditions are changed.

The change in percent relative turgidity with the degree of dehydration is shown in Fig.4 (shown only cabbage and turnips). As expected, graphs have similar shape as in Fig.3, i.e. two straight lines intersecting at the respective critical point mentioned above. The values of relative turgidity for cabbage, turnips, potato and onion corresponding to their critical points stated above, are 86, 84, 94 and 89% respectively. It is observed from the graph that the decline of relative turgidity during the initial stages is much less than the decline beyond the critical points of dehydration. This enhanced loss of turgidity is evidently due to the adverse effects of drying on the physico-chemical structure and function of cellular organelles which control the degree of hydration and ionic balance in the cell.

Activity of the reducing enzyme of the respiring system of the four vegetables at two levels of dehydration is given in Table 1. All the four vegetables exhibit different behaviour so far as enzyme activity Vs. degree of dehydration is concerned. Cabbage lost the activity somewhere in between 40 and 52% degree of dehydration while turnips did so between 60 and 83% (Table 1).

However, it is clear that in the case of all four vegetables the critical point i.e. the degree of dehydration beyond which rehydration capacity is grossly affected lies in between the points where the enzyme test was positive and negative. Under the experimental conditions, however, the exact point or stage of dehydration when the enzyme became non-functional is not known. It is possible that the stage may coincide with or may precede/follow closely the critical point. In case of onion, for example, the enzyme was active at 30% but inactive at 52% degree of dehydration. The transformation of the enzyme to the inactive form might, in effect, have taken place within the

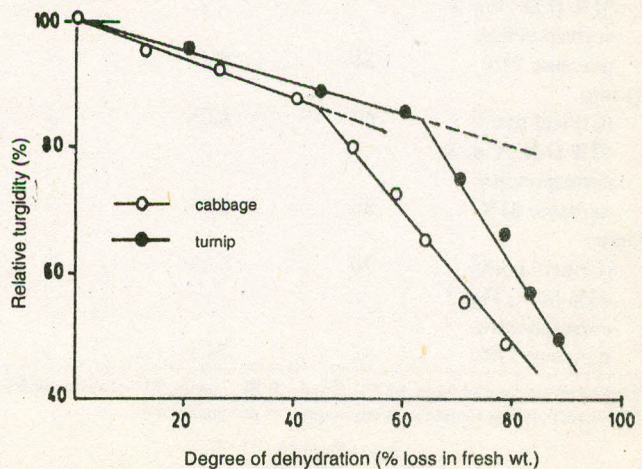


Fig. 4. Relative turgidity (%) vs degree of dehydration.

span of these two points which could actually be the same or close to the critical point i.e. 45%.

The loss of enzymic activity seems to be related to the loss of dehydration capacity because it is known that respiration is a combined function involving not only gas exchange but also loss of water. Dehydration beyond the critical point may be assumed to manifest changes which may have occurred at the cellular level. Activity of several enzymes is known to have strict requirement of membrane integrity which depends as much on the degree of hydration [1] as on the potential difference in osmotic pressures across the membrane [13, 14]. Water is drawn across the membrane by the potential difference across the plasma membrane [15].

Removal of water from the cytoplasm gives rise to changes which are responsible for the failure of cells to take up water and regain the original shape. Such a change, noted among certain nematodes during anhydrobiosis, has been explained in terms of fusion of lipid bilayers [16]. The same mechanism i.e. dehydration, perhaps leads to compression of plasma membrane and when loss of moisture exceeds the critical level, the packing ability of the phospholipids and proteins which make up the membrane, plasma and the tonoplast, is

TABLE 1. ACTIVITY OF THE REDUCING ENZYME AT TWO LEVELS OF DEHYDRATION OF CABBAGE, POTATO, TURNIP AND ONION.*

Material	Degree of dehydration (D.D.)	Corresponding % moisture**	Enzyme*** activity
Cabbage (Critical point; 43% D.D.; Fig. 2 corresponding moisture 85%)	40	85.5	+
	52	82.0	-
Potato (Critical point; 31% D.D.; Fig. 2, corresponding moisture 71%)	28	72.2	+
	54	56.5	-
Turnip (Critical point; 63% D.D.; Fig. 3, corresponding moisture 81%)	60	82.5	+
	83	58.5	-
Onion (Critical point; 45% D.D.; Fig. 3, corresponding moisture 77%)	30	82.3	+
	52	74.1	-

* Initial moisture: Cabbage; 81.5%; Potato: 80%; Turnip, 93%; Onion, 84.6%.

** Corresponding moisture at any degree of dehydration =

$$\frac{\text{INITIAL MOISTURE (\%)} - \text{D.D.}}{100 - \text{D.D.}}$$

*** + appearance of red stain on reaction with 0.6% solution of TTC. - no stain formation i.e. nil activity.

greatly disturbed. The extensibility of the cell wall is reduced and so also the modulus of elasticity in tissues as observed by Willis and Teixeira [17]. Dehydration similarly leads to protein denaturation and loss of differential permeability due to solute concentration as observed by Curtis & Clark [18]. Shimazu and Sterling [7] and Neumann [4] and increase in cell wall crystallinity owing to increase in polysaccharide - polysaccharide bonds replacing polysaccharide-water bonds as observed by Sterling and Shimazu [6] and Shimazu and Sterling [10].

Temperature plays an important role in these changes as is clear from the observation that sample dried at low temperature (lyophilized) rehydrate more rapidly with lower loss of rehydration capacity than samples dried at higher temperatures provides some interesting results and needs further investigation with reference to the critical point mentioned herein and by Willis and Teixeira, [17] which seems to be different under different sets of dehydration conditions e.g. low temperature, vacuum etc.

Conclusion

Critical points of dehydration exist for each vegetable beyond which their rehydration is substantially reduced. The mechanism offered is related to reduction in turgidity given by Ψ_p in Lockhart's equation [19]. There is a lowering in the hydrogen bondability of the cell wall by denaturing of proteins and loss of enzymic activity induced by lowering of turgidity as observed experimentally.

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