Effect of Nodal Water Potential on the Germination of Sugarcane Buds

While the role of initial moisture content of sugarcane sett in the germination of bud has been investigated[1-4], its importance in terms of water potential has not been assessed. The significance of the term hydration of sett depends on whether the quantity factor (water content), or the intensity factor (water activity) is involved. The movement of water from the other regions of the sett towards the meristematic cells of the bud as observed by PANJE and RAO[4] is influenced more by latter than the former. The water potential of a nodal tissue generally is regarded as the sum of 2 components, pressure potential and osmotic potential. Alteration in any of the 2 components could be expected to result in shifts of water potential. The term water potential has been used in accordance with the recommendations of SLATYER and TAYLOR[5] and the free energy unit joules/kg has been used to express the different levels of energy status of water (101.3 joules/kg = 1 atmosphere)[6].

The initial water potential of fresh nodal tissues has been observed to range between -607.8 to -810.4 joules/kg. This value varies with changes in soil and environmental stress which in turn affect germination of the bud. Here an attempt has been made to determine the critical nodal water potential for germination of the bud.

Material and methods. Nodes of identical age having viable buds were selected from the top half portions of stalks of sugarcane variety Co. 453. These were divided longitudinally into 2 equal halves. Portions bearing the bud were taken for study. There were 10 lots of 10 nodes in each treatment. For variation in water potential in the nodes, the lots of fresh nodes with water potential of about -607.8 joules/kg were kept as control. The water potential of the second lots was raised to about -303.9 joules/kg by soaking in distilled water. The remaining lots were allowed to desiccate to about -861.0, -1063.6, -1266.2, -1468.8, -1722.8, -1823.4 and -2026.0 joules/kg by placing these under a fan for different lengths of time. Water potential of the nodal tissues in each lot was determined by the SCHARDAKOW[7] dye technique as modified by MANOHAR[8].

Five lots of nodal tissues from each treatment were then coated with beeswax, except the buds, to avoid any change in their initial water potential. The remaining lots were left uncoated. Both uncoated and coated lots were kept for germination at 30 ± 1 °C and 96% relative humidity in darkness in petri dishes with and without distilled water respectively. The results are given in the Table.

Results and discussion. The data indicate that in coated nodes germination started earlier and increased at a fast rate when its water potential was raised from -607.8 to -303.9 joules/kg. While a decrease in water potential from -607.8 to -1063.6 joules/kg resulted 90 to 18.44% germination, respectively. However, there was no germination beyond -1063.6 joules/kg. It may also be observed that drying of nodal tissue resulting in a decrease of water potential from -1063.6 to -1823.4 joules/kg did not cause permanent injury as evidenced by normal germination, though with slow rate, when kept under direct influence of external water. On the other hand further decrease in potential beyond -1823.4 joules/kg caused permanent damage to viability of the buds.

It appears from this study that buds can germinate up to -1063.6 joules/kg nodal water potential even in the absence of any external water intake, provided there is no loss of internal tissue moisture. With a reduction in water potential from -1063.6 to -1823.4 joules/kg, buds could germinate only on absorption of water by nodal tissues. This indicates that under field conditions, nodes desiccated up to -1823.4 joules/kg can be made to germinate if they are pre-soaked and planted in the soil with adequate moisture, while -2026.0 joules/kg which caused failure of germination even under optimum conditions may be considered as a sublethal water deficit for germination of the buds.

The physiological mechanism causing the revival of germination of a desiccated bud, when kept in water is not very clear. However, the internal osmotic tension which gets relieved due to intake of water seems to be the cause of inhibition of germination, probably due to

Influence of nodal water potential on the germination of sugarcane buds

Nodal water potentials (joules/kg)	Data transformed by sin-1/% germination (time in h)					
	30	40	60	80	100	120
— 303.9	45.00[a] (67.21)[b]	67.21 (90.00)	90.00 (90.00)	90.00 (90.00)	90.00 (90.00)	90.00 (90.00)
— 607.8	33.21 (63.44)	53.73 (90.00)	67.21 (90.00)	71.56 (90.00)	75.56 (90.00)	90.00 (90.00)
— 861.0	22.79 (63.44)	22.79 (90.00)	36.27 (90.00)	53.73 (90.00)	60.00 (90.00)	60.00 (90.00)
1063.6	0.00 (57.79)	12.92 (90.00)	18.44 (90.00)	18.44 (90.00)	18.44 (90.00)	18.44 (90.00)
—1266.2	0.00 (50.77)	0.00 (71.56)	0.00 (77.08)	0.00 (90.00)	0.00 (90.00)	0.00 (90.00)
—1468.8	0.00 (0.00)	0.00 (26.56)	0 00 (63.44)	0.00 (67.21)	0.00 (90.00)	0.00 (90.00)
—1722.1	0.00 (0.00)	0.00 (12.92)	0.00 (50.77)	0.00 (71.56)	0.00 (71.56)	0.00 (90.00)
—1823.4	0.00 (0.00)	0.00 (0.00)	0.00 (30.00)	0.00 (39.23)	0.00 (39.23)	0.00 (50.77)
—2026.0	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)

[a] The figures without parenthesis denote the % germination of node coated with beeswax kept as such in Petri dishes. [b] The figures in parenthesis denote the % germination of the uncoated nodes when kept in Petri dishes in distilled water on filter paper.

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6 101.3 joules/kg = 1.013×10^6 dynes cm^{-2} = 1.013 bar = 1033.2 cm water. Taking free water as the reference, the water potential in sugarcane tissues will always be negative, water will tend to flow from a region with low negative values to one with large negative values.
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altering the metabolic pathways leading to sprouting of the bud. Some more information is required to explain the mechanism of germination under water-deficient conditions[9].

Zusammenfassung. Das Keimen der Zuckerrohr-Achselknospen wurde als vom Knotenwasserpotential beeinflussbar beobachtet. Die Sprossentwicklung erfolgte noch bei so geringem Wasserpotential wie 1063,6 Joule/kg und selbst bei Fehlen jeglicher Wasseraufnahme. Ausgeschnittene Knospen mit einem Wasserpotential von 1063,5 bis 1823,4 Joule/kg konnten nur im Wasser gehalten austreiben, während ein Wasserpotential von 2026,0 Joule/kg tödlich war.

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[9] Thanks are due to Dr. KISHAN SINGH, Indian Institute of Sugarcane Research, for his valuable suggestions and kindly going through the manuscript.

Caffeine-Induced Contractures and Related Calcium Movements of Muscle in Hypertonic Media

Hypertonic solutions markedly reduce the mechanical responses of frog skeletal muscles to electrical stimuli while they leave the action potential normal[1–4]. The present paper describes effects of caffeine on contracture development and related ^{45}Ca movements in skeletal muscles in hypertonic media and media of normal tonicity and shows that ^{45}Ca still can be released by the action of caffeine on muscles in hypertonic media, although caffeine contracture development largely is blocked.

Methods. After excision the sartorii of the frog (*Rana pipiens*) were equilibrated for about 1 h in normal Ringer's solution before use. Normal Ringer's solution contained, in mmoles per liter of deionized water: 108 NaCl, 1.6 KCl, 1.0 $CaCl_2$, 2 *Tris* (hydroxymethyl) aminomethane-HCl buffer at pH 7.1, and 2×10^{-2} g/l of curare (D-tubocurarine chloride). Hypertonic Ringer's solution was prepared either by adding 330 m*M* sucrose to normal Ringer's or else by using 2.5 times as much NaCl and KCl as in normal Ringer's. Conventional massive stimulation[5] was used and isometric tension was recorded with a Statham ($\pm$ 4 oz) strain gauge and Sanborn amplifier and chart recorder. An atomic absorption spectrophotometer (Perkin-Elmer 303) was used for total calcium analyses[6]. ^{45}Ca movements were studied by standard techniques[6–8] and work was done at room temperature (about 23 °C). Caffeine (Eastman Organic Chemicals) was added to Ringer's solution as the free base.

Results. Exposure of muscles to the hypertonic sucrose Ringer's solution itself caused a contracture of 1–3 g (i.e. about 1–3% of maximal tetanic tention, P_0), which began within the first minute, reached a maximum within 3 min, and then started to decline after about 10 min (after 1 h the contracture tension was reduced by about half-maximum). After the completion of this work a paper by D. K. HILL[9] was published that also describes contractures of frog sartorii in hypertonic sucrose (although these are called 'changes of resting tension due to changes of the tonicity of the solution'). No such contractures were observed in the Na-K hypertonic Ringer's solution.

Table I shows that muscles in normal Ringer's solution exposed to caffeine (10 m*M*) develop a contracture tension of about 40% P_0, whereas muscles pre-soaked in either form of hypertonic Ringer's solution and then exposed to caffeine (10 m*M*) added to the hypertonic Ringer's develop much less contracture tension. This block by hypertonicity of caffeine contracture (and twitch and tetanic responses as well) appeared within 10 min after the muscles had been immersed in the hypertonic Ringer's and was almost complete by 1 h. Caffeine contractures of normal magnitude were obtained by returning a muscle to normal Ringer's solution after a soak in hypertonic Ringer's solution.

Using fresh frog sartorii, I tested for effects of hypertonicity on the total calcium content and the uptake and release of ^{45}Ca. Figures 1 and 2 demonstrate that hypertonic media (either with sucrose or extra Na and K) can cause a two- to three-fold increase in the rate of release of ^{45}Ca from frog sartorii. Control experiments show no such effect in the Achilles tendon of frog, so presumably in the whole muscle the ^{45}Ca release represents an effect on the muscle fibers per se. Furthermore, the effects of hypertonicity are shown in Figures 1 and 2 after about 120 min of washout of ^{45}Ca in normal Ringer's and thus represent an effect on essentially the slow component of ^{45}Ca release that is believed to reflect the rate of release of Ca from an intracellular locus across the surface membranes of the muscle fibers[8, 10]. This effect of hypertonicity on ^{45}Ca efflux is sustained for 1 h (Figures 1 and 2) and is somewhat better sustained in sucrose.

The addition of caffeine (10 m*M*; Figures 1 and 2) causes an increase in the rate of ^{45}Ca release in Ringer's of both normal and increased tonicity. In the latter case, the increase in ^{45}Ca release caused by caffeine appears additional to that caused by the hypertonicity itself.

Table II compares the total calcium content and the amount of ^{45}Ca uptake of frog sartorii that were exposed to ^{45}Ca for 10 min in either normal Ringer's solution (C) or in hypertonic Ringer's solution (E). Statistically there is a significant increase of 0.22 µmoles of calcium per

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