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PROLINE ACCUMULATION IN THE LEAVES OF FOUR POTATO CULTIVARS IN RESPONSE TO WATER STRESS

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Abstract

Influence of water stress on four potato cultivars (*i.e.* Kufri Chipsona-1, Kufri Pukhraj, Kufri Lauvkar and Desiree) was investigated with respect to three growth stages (*i.e* tuber initiation stage (T_2), tuber enlargement stage (T_3) and tuber maturation stage (T_4). In present study proline accumulation was found to be increased in potato leaves due to water stress at different growth stages. Much increase in proline content was found when water stress was imposed at tuber initiation stage (>75%) and at tuber enlargement stage (>85%) in comparison with well irrigated control T_1. Accumulation of proline may be ascribed as due to three metabolic causes *i.e. de novo* synthesis from glutamate, lower rate of proline catabolism because of inhibition activities of proline dehydrogenase and proline oxidase by water stress and slow incorporation of proline into protein. The cultivar Kufri Lauvkar recorded accumulation of maximum proline content due to water stress treatments T_2, T_3 and T_4 in comparison with well watered control T_1. Among growth stages, Kufri Lauvkar showed highest proline elevation (> two folds) when stress was given at tuber maturation stage. Kufri Chipsona-1 recorded minimum proline content as a result of all water stress treatments except T_3 where this cultivar was found second to lowest in proline accumulation during both the years during this study.

Key words: Proline, water stress, potato, tuber initiation, tuber enlargement, tuber maturation stage

Introduction

In the era of global climate change, sustainability of crop production is a serious challenging issue due to increasing incidences of both biotic and abiotic stresses in farmer's field (Muhammad Nadeem *et al.,* 2019). Enhancing water stress tolerance in plants through conventional breeding is a useful approach and a principal strategy for crop improvement (Farooq *et al.,* 2014). In mesophytic plants, proline accumulation has been reported as one of the marked responses to water stress by several authors (Keshav Dahal *et al.,* 2019, Paleg and Aspinall, 1981; Hanson and Hitz, 1982; Rhodes and Handa, 1989; Losch, 1991; Galiba, 1994). During water stress, proline plays an important role and act as a signaling compound to regulate mitochondria function and affect cell proliferation by means of activating particular genes, which are essential for stress recovery (Solanki *et al.,* 2015). Proline accumulation aids in retaining membrane integrity by decreasing oxidation of lipids through guarding cellular redox potential and scavenging free radicals (Shinde *et al.,* 2016). Proline accumulation is favoured by high leaf carbohydrate status and also by illumination (Hanson and Hitz, 1982). Naidu *et al.,* (1990) showed that a progressive water stress rather than a rapid water stress resulted in proline being the dominant accumulated compound. Accumulation of proline may be ascribed as due to three metabolic causes of which *de novo* synthesis from glutamate is considered to be the major factor (Hanson and Hitz, 1982; Venekamp *et al.,* 1989), other factors are lowered rate of proline catabolism because of inhibition of activities of proline dehydrogenase and proline oxidase by water stress (Venekamp *et al.,* 1989) and slower incorporation of proline into protein (Hanson and Hitz, 1982). Physiological significance of proline

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accumulation has long been a matter of dispute. However most of the workers are of view that such change is associated with drought tolerance. Proline can serve as a nitrogen and carbon source during recovery (Purvis and Yelenosky, 1982), as enzyme regulator (Stewart and Boggess, 1977), as osmoregulator and osmoprotectants (Shevyakava, 1984; Delauney and Verma, 1993). Free proline accumulation has been observed in leaf tissue of potato in response to water stress by several workers (Chen *et al.*, 1964; Singh *et al.*, 1972; Levy 1983, Keshav Dahal *et al.*, 2019,). Bansal and Nagarajan (1986) also noticed accumulation of free proline in potato leaves due to stress in all the genotypes. They found that proline accumulation in leaves showed a significant negative correlation with tuber weight (r = -0.729) and tuber number (r = -0.902) in stressed plants. In non stressed plants proline accumulation in leaves correlated negatively (r = -0.752) with tuber number. Accumulation was found least in Phulwa and G-2524. The low proline content as a result of water stress in a resistant cultivar, therefore suggest a negative correlation between the proline contents in leaves and the relative tolerance to stress. Levy (1983) pointed out the possible association of proline accumulation in tuber tissue with relative drought susceptibility. Similar proline accumulation in leaves in response to stress and drought susceptibility was obtained by Bansal and Nagaragen (1986). Singh *et al.*, (1972) showed a highly significant positive correlation between proline accumulation and drought injury in barley. Adaptation to water deficit stress may involve several different morphological and physiological characters of the plant. Balco (2002) studied some indirect selection characteristics for drought viz. water use efficiency, leaf water loss, chlorophyll fluorescence and proline accumulation in leaves of potato cultivars. Keeping above in view four potato cultivars were evaluated for leaf proline status when grown under water stress imposed at different stages of crop growth.

Materials and Methods

The field experiments were conducted at Research Farm of ICAR-CPRI Campus, Modipuram, Meerut (29_40N, 77_460E, 237 masl) during *rabi* season. Forty eight plots were used in a split plot design for accommodating 4 treatments. Field trials were conducted in split plots with three replicates employing the 4 varieties namely Kufri Chipsona-1, Kufri Pukhraj, Kufri Lauvkar and Desiree having treatments, T_1 : Control (well watered plants), T_2:Water stress at tuber initiation stage, T_3:Water stress at tuber enlargement stage, T_4:Water stress at tuber maturation stage. T_1 control (well watered) plots were irrigated at 6 DAP (days after planting), 27 DAP, 42 DAP, 63 DAP and 80 DAP during the year 1st year and at 8 DAP, 25 DAP, 44 DAP, 67 DAP and 83 DAP during the 2nd year. The water stress was imposed by withholding water in T_2, T_3 and T_4 treatments at different growth stages. The growth stage was identified and confirmed by uprooting the plants and by examining the stage of tuber development. All the curtivars were screened for proline by taking composite samples on 4th leaf from top in 3–4 plants on peak stress day in all the treatments. Proline content of the leaf was estimated according to the method of Bates *et. al.* (1973). Experimental plots were dehaulmed at 90 DAP and harvesting was done 10-15 days after of dehaulming so that tuber skin is matured.

Results and Discussion

Mean values of treatments in table 1 showed that proline content increased significantly due to water stress treatments at different growth stages in comparison with well watered control (T_1). Maximum proline content (439 and 454 µg/g fw during 1styear and 2nd year, respectively) was recorded due to water stress treatment at tuber initiation stage (T_2) whereas minimum increase in proline content (133 and 135 µg/g fw during 1styear and 2nd year, respectively) was recorded when water stress was imposed at tuber maturation stage (T_4). Maximum percent increase (92% and 86% in respective years) in proline content due to water stress was found at tuber enlargement stage (T_3) whereas minimum percent increase (42% and 43% in respective years) was found at tuber maturation stage (T_4). It is clear from Table 1 that Kufri Lauvkar recorded accumulation of maximum proline content due to water stress treatments T_2 (685 and 703 µg/g fw during 1styear and 2nd year, respectively), T_3 (343 and 344 µg/g fw during Ist year and IInd year respectively) and T_4 (154 and 158 µg/g fw during Ist year and IInd year respectively) in comparison with well watered control $T_{1.}$ Kufri Chipsona-1 recorded minimum proline content as a result of all water stress treatments except T_3 where this cultivar was found second to lowest in proline accumulation during both the years.

The cultivar Kufri Pukhraj recorded maximum percent increase (120% and 126% in respective years) in proline content whereas cultivar Kufri Chipsona-1 recorded minimum percent increase (62% in both the years) in proline content when water stress was imposed at tuber initiation stage (T_2). As a result of water stress at tuber enlargement stage (T_3), the cultivar Desiree recorded maximum percent increase in proline content (142% and 161% in respective years) whereas cultivar

Kufri Chipsona-1 recorded minimum percent increase (27% and 29% in respective years) in proline content in comparison with respective well irrigated control. When water stress was imposed at tuber maturation stage (T_4) the cultivar Kufri Lauvkar recorded maximum percent increase in proline content (103% and 105% in respective years) whereas cultivar Kufri Pukhraj recorded minimum percent increase (10% and 11% in respective years) in proline content. The interaction between cultivar and treatments was also found significant.

Proline is a basic amino acid present in high percentage in basic proteins. It is accumulated as compatible solutes in plants under water stress (Muhammad Abid *et al.,* 2018). Water deficit increases plant antioxidant enzyme activity, ion leakage and proline content, but reduces total antioxidant capacity (Xin Lia *et al.,* 2019). Elevated levels of proline have been reported as one of the marked responses to water stress by several

Table 1: Effect of water stress on leaf proline content (µg/g fresh weight) at various growth stages of potato cultivars.

Treat-ments*	1st year			2nd year		
	Growth stage**			Growth stage		
	TI	TE	TM	TI	TE	TM
Kufri Chipsona-1						
T_1	164	149	101	167	151	103
T_2	265 (+62%)***	96	92	270 (+62%)	100	93
T_3	163	189 (+27%)	88	165	195 (+29%)	89
T_4	160	150	122 (+21%)	162	152	123 (+19%)
Mean	188	146	101	191	150	102
Kufri Pukhraj						
T_1	214	82	120	221	86	121
T_2	471 (+120%)	143	110	500 (+126%)	146	111
T_3	218	176 (+115%)	107	220	180 (+109%)	108
T_4	215	180	132 (+10%)	218	181	134 (+11%)
Mean	280	145	117	290	148	119
Kufri Lauvkar						
T_1	409	207	76	414	211	77
T_2	685 (+68%)	277	144	703 (+70%)	276	145
T_3	411	343 (+66%)	84	415	344 (+63%)	85
T_4	414	200	154 (+103%)	411	210	158 (+105%)
Mean	480	257	114	486	260	116
Desiree						
T_1	199	114	91	201	124	92
T_2	337 (+69%)	178	122	341 (+70%)	181	123
T_3	200	298 (+161%)	122	205	300(+142%)	122
T_4	197	115	123 (+35%)	202	117	125 (+36%)
Mean	233	176	114	237	181	115
Mean values of treatments						
T_1	246	138	97	251	143	98
T_2	439 (+78%)	174	117	454 (+81%)	176	118
T_3	248	252 (+92%)	100	251	255 (+86%)	101
T_4	247	162	133 (+42%)	248	165	135 (+43%)
CD at 5%						
Cultivar(C)	15	8	6	14	8	5
Treatment (T)	12	7	4	11	7	4
C × T	23	15	8	22	14	8

**Growth stages: TI = Tuber initiation, TE =Tuber enlargement and TM = Tuber maturation

*Treatments: T_1 = -Control (well watered), T_2 = water stress at tuber initiation, T_3 = water stress at tuber enlargement and T_4 = water stress at tuber maturation stage

***Figures in parenthesis are percent (%) change in proline content of leaf due to water stress treatment T_2, T_3 and T_4 as compared with respective control.

authors (Paleg and Aspinall, 1981; Losch, 1991, Kumar and Minhas, 2013). In present study proline accumulation was found to be increased in potato leaves due to water stress at different growth stages table 1. Mean proline content indicated much increase in proline content was found when water stress was imposed at tuber initiation stage (>75%), at tuber enlargement stage (>85%) and > 40 % at tuber maturation stage in comparison with well irrigated control. Accumulation of proline may be ascribed as due to three metabolic cases *i.e. de novo* synthesis from glutamate, lower rate of proline catabolism because of inhibition activities of proline dehydrogenase and proline oxidase by water stress (Venekamp *et al.*, 1989) and slow incorporation of proline into protein (Hanson and Hitz, 1982). The cultivar Kufri Pukhraj recorded maximum increase in proline content whereas the cultivar Kufri Chipsona-1 recorded minimum increase in proline content due to water stress. On perusal of varietal behavior at different growth stages for proline content, it was observed that Kufri Lauvkar had highest proline enhancement under stress at tuber maturation stage as compared to other three cultivars. Physiological significance of proline accumulation is associated with drought tolerance, as it is known to protect the tissue system either as osmoprotectant (Hanson, 1993) and prevention of denaturation of proteins and enzymes or both (Schwab and Gaff, 1990). These results indicated the stage specific response for proline enhancements in response to drought event in different potato cultivars. Findings may have larger implications in varietal selection needed for growing in drought affected situations.

References

Balco, C. (2002). Drought tolerance in potatoes: Variability and indirect selection criteria. *Zuchtungsforschung-Bundesanstalt fur Zuchtungsforschung an Kulturpfl-anzen,* **8:** 115–118.

Bansal, K.C. and S. Nagarajan (1986). Leaf water content, stomatal conductance and proline accumulation in leaves of potato (*Solanum tuberosum* L.) in response to water stress. *Indian J. Plant Physiol.*, **29(4):** pp. 397-404.

Bates, L.S., R.P. Waldren and I.D. Teare (1973). Rapid determination of free proline for water stress studies. *Plant Soil,* **39:** 205-207.

Chen, D., B. Kessler and S.P. Monselise (1964). Studies in water regime and nitrogen metabolism of citrus seedlings grown under water stress.*Plant Physio*l, **39:** 379-386.

Delauney, A. and D.P.S. Verma (1993). Proline biosynthesis and osmoregulation in plants. *Plant J.*, **4:** 215-233.

Farooq, M., M. Farooq, M. Hussain and K.H.M. Siddique (2014). Drought stress in wheat during flowering and grain filling periods. *Crit. Rev. Plant Sci.*, **33:** 331–349.

Galiba, G. (1994). *In vitro* adaptation for drought and cold hardiness in wheat. In: J. Janick, ed. Plant Breeding Reviews vol. 12 John Wiley & Sons Inc. New York pp. 115-162.

Hanson, A.D. (1993). Accumulation of quaternary ammonium andtertiary sulfonium compounds. In '*Plant Responses to Cellular Dehydration during Environmental Stress'* (T.T. Close and E.A.Bray, Eds.) pp.30-36. *Amer. Soc.Plant Physiologists. Rockhill, M.D.*

Hanson, A.D. and W.D. Hitz (1982). Metabolic responses of mesophytes to plant water deficits. *Annu. Rev. Plant Physiology,* **33:** 163-203.

Keshav Dahal, Xiu-Qing Li, Helen Tai, Alexa Creelman and Benoit (2019). Improving Potato Stress Tolerance and Tuber Yield Under a Climate Change Scenario – A Current Overview. *Frontiers in Plant Science*, www.frontiersin.org **10:** 563.

Kumar Devendra and J.S. Minhas (2013). Evaluation of indigenous potato varieties, advanced clones and exotic genotypes against water deficit stress under sub-tropical environment. *Indian J. Plant Physiology,* **18(3):** 240-249.

Levy, D. (1983). Varietal differences in the response of potatoes to repeated short periods of water stress in hot climates. 1 Turgor maintenance and stomatal behavior, *Potato Res.*, **26:** 303-313.

Losch, R. (1991). Plant water relations. In H.D. Behnke K. Esser, K. Kubitzki, M. Runge, H. Ziegler eds Progress in Botany. **52:** springer-verlag Berlin pp. 35-60.

Muhammad Abid, Shafaqat Ali, Lei Kang Qi1, Rizwan Zahoor, Zhongwei Tian, Dong Jiang, John L. Snider and Tingbo Dail (2018). Physiological and biochemical changes during drought and recovery periods at tillering and jointing stages in wheat (*Triticum aestivum* L.) *Scientific Reports* **8:**4615 doi:10.1038/s41598-018-21441-7 www.nature.com/scientificreports.

Muhammad Nadeem, Jiajia Li, Muhammad Yahya , Alam Sher, Chuanxi Ma, Xiaobo Wang and Lijuan Qiu (2019). Research Progress and Perspective on Drought Stress in Legumes: A Review *Int. J. Mol. Sci*., **20:** 2541; doi:10.3390/ijms20102541 www.mdpi.com/journal/ijms.

Naidu, B.P., L.G. Paleg, D. Aspinall, A.C. Jennings and G.P. Jones (1990). Rate of imposition of water stress alters the accumulation of nitrogen containing solutes by wheat seedlings. *Aust. J. Plant Physiol*., **17:** 653-664.

Paleg, L.G. and D. Aspinall (1981). The physiology and biochemistry of drought resistance in plants. Academic Press, Sydney New York.

Purvis, A.C. and G. Yelenosky (1982). Sugar and proline accumulation in grapefruit flavedo and leaves during cold hardening of young trees. *J. Am. Soc. Hort. Sci.,* **107:** 222-226.

Rhodes, D. and S. Handa (1989). Amino acid metabolism in relation to osmotic adjustment in plant cells. In J.H. Cherry, ed. Environmental stress in Plants. Springer-verlag Berlin pp 41-61.

Schevyakava, N.I. (1984). Metabolism and physiological role of proline in plants under conditions of water and salt stress. *Sov. Plant Physiol.*, **31:** 597-608.

Schwab, K.B. and D.F. Gaff (1990). Influence of compatible solutes on soluble enzymes from desiccation tolerant *Sporobolous stafianus* and desiccation sensitive *Sporobolous pyranidalis. J. Plant Physiol.*, **137(2):** 208-215.

Shinde, S., J.G. Villamor, W. Lin, S. Sharma and P.E. Verslues (2016). Proline co-ordination with fatty acid synthesis and redox metabolism of chloroplast and mitochondria. *Plant Physiol.*, **172:** 1074–1088.

Singh, T.N., D. Aspinall and L.G. Plaeg (1972). Proline accumulation and varietal adaptability to drought in barley. A potential metabolic measure of drought resistance. *Nature New Biol.*, **236:** 188-190.

Solanki, J.K. and S.K. Sarangi (2015). Effect of drought stress on proline accumulation in peanut genotypes. *Int. J. Adv. Res.*, **2:** 301–309.

Stewart, C.R. and S.F. Boggess (1977). The effect of wilting on the conversion of arginine, ornithine and glutamine to proline in bean leaves. *Plant Lett,* **8:** 147-153.

Venekamp, J.H. and J.E.M. Lampe (1989). Organic acids as sources for drought-induced proline synthesis in field bean plants, *Vicia faba L. J. Plant Physiol.*, **133:** 654-659.

Xin Lia, David A. Ramíreza, Junhong Qinc, Richard Dormateya, Zhenzhen Bia, Chao Suna, Han Wanga and Jiangping Baia (2019). Water restriction scenarios and their effects on traits in potato with different degrees of drought tolerance *Scientia Horticulturae,* **256:** 108525.