Evaluation of Garden Egg Genotypes for Proline Accumulation in different Growth Conditions of Ghana

ABSTRACT

Sixteen (16) genotypes of garden egg (*Solanum* spp) were grown over two years in the Coastal and Sudan Savannah areas of Ghana, in Randomized Complete Block Design with three replications, to identify proline accumulation response patterns of the genotypes under dry season and drought-stressed conditions of Ghana. The experiment was conducted at Savanna Agricultural Research Institute (SARI) experimental farm, Manga, Bawku (Sudan Savannah Agro-ecology), and University of Ghana, Legon, Accra, experimental farm (Coastal Savannah Agro-ecology). At each agro-ecology, leaf samples of the genotypes were collected at the flowering stages of growth, dried, milled and assayed for their proline levels. The proline data for each location and season for the two year period were separately analyzed by general analysis of variance (ANOVA), for the estimation of the variation among the genotypes in proline accumulation. Proline which confers tolerance of the crop to variable seasonal and drought-stressed conditions varied significantly, due to the genotype and genotype x environment interaction effects on its accumulation. The garden egg genotypes were observed to develop internal complementary drought survival mechanisms, by lowering leaf relative water contents (LRWC) and increasing proline content, thereby enabling plants to withstand periodic drought better. The genotypes A3, A4, A8, A9F, A10 and Bawku1 accumulated higher levels of proline under dry season and drought-stressed conditions of the Coastal and Sudan savannahs, with the associated high temperatures across locations. These genotypes could be selected on the basis

of proline accumulation, for improved drought tolerance of the crop, and should be incorporated in garden egg drought tolerant improvement programmes in Ghana.

Key Words: Garden Egg, Genotypes, Growth Conditions, Proline Accumulation

1.0 INTRODUCTION

Garden egg (*Solanum spp*) is cultivated in Ghana as source of food and income, especially for the small scale farmers [1, 2]. Though widely cultivated in a small scale in Ghana, it is grown in the Coastal and Sudan savannah agro-ecologies under highly unstable conditions of high temperatures, erratic rainfall and intermittent drought. Drought stress in particular is very common in crop fields of these agro-ecologies, and it is a major crop developmental and yield-limiting factor [3, 4].

Few garden egg genotypes are predominantly cultivated in the Coastal and Sudan savannah agro-ecologies of Ghana, and may be considered as adaptive under those environmental conditions. The stable and adaptable genotypes that are considered superior in unfavourable environments similar to that of Coastal and Sudan savannah agro-ecologies of Ghana, have been identified with an ability to efficiently accumulate specific stressed-induced bio-active compounds [5-8].

In drought stress conditions, plants reduce and loose turgor, and are most susceptible during reproductive phase, when brief periods of water shortage could greatly reduce yield [9-11]. The reduction or loss of turgor in plants subjected to stress conditions triggers several physiological and/or chemical responses in them [12,13]. The accumulation of proline is the primary physiological trigger in plants that activates a complex of sequence of adaptive events correlated

to the level of stress, plant tolerance and plant growth stage (14, 3]. In plants, the accumulation of cellular solutes, such as proline has been one possible means for overcoming osmotic stress caused by loss of water [15, 16].

However, the levels of proline in plants are properly regulated, according to environmental conditions [17]. It is mainly accumulated under drought-stress conditions, but can be accumulated under high temperature stresses [18]. In drought stress conditions, most plants increase proline accumulation at flowering stages than at the vegetative stages [19, 20]. The proline accumulation in plants under stressed conditions therefore becomes a survival mechanism in plants, which greatly determine their adaptability to varying environments, and largely influence their desirable traits performance and stability over time and location [21].

Plants are able to adapt and resist stress because the accumulated proline regulates and reduces water loss from dehydrated cells [22, 23]. Its biosynthesis also enables plants to survive under stress conditions by assisting plants to maintain the photosynthetic efficiency and the overall survival and productivity [24]. In general, there is better survival and performance of plant species that accumulate proline under stress conditions. Proline therefore plays important role in adaptation and survival of plants under drought and temperature stresses [25-27].

The physiological responses of plants in drought-stressed conditions such as increases or decreases in proline accumulation, are useful indices of drought tolerance [28, 29]. Such physiochemical studies on garden egg genotypes under varying environments in Ghana are vital to ascertain the physiological behavior of existing materials in the plant genetic pool [30]. In

such studies, desirable genotypes could be identified and selected for farmers and for crop improvement purposes based upon their physiological traits competencies across environments.

There is however limited study on the influence of varying soil moisture conditions on proline accumulation in garden egg across agro-ecologies in Ghana. It is in this light that a study was conducted to assess garden egg genotypes for proline accumulation under varying soil moisture conditions of two most drought-stressed agro-ecologies of Ghana.

2.0 MATERIALS AND METHODS

2.1 The Study Areas

The experiment was carried out at Savanna Agricultural Research Institute (SARI) experimental farm, Manga, Bawku in the Sudan savannah agro-ecology and University of Ghana, Legon, Accra experimental farm in the Coastal savannah agro-ecology. Manga, Bawku is located in the North-Eastern corner of the Upper East Region of Ghana, on Latitude 11°11'and 10°40'N and Longitude 0°18' W and 0°6'E, at an altitude of 249 meters above sea level, with topography of gently sloping terrain of gradient 1-2%. The University of Ghana experimental farm is located in the north-east of the Greater Accra region of Ghana, on Latitude 5°38'45''N and Longitude 00°11'13''E at an altitude of approximately 300 meters above sea level.

2.2 Climatic Data Collection

Climatic data (Table 1) was collected during the respective rainy and dry seasons of 2012-2013 and 2013-2014 at each experimental site of Legon and Manga. Within the study period, Legon site recorded 5 months of dry season and 7 months of rainy season whereas Manga site was

7 months of dry season and 5 months of rainy season. Until flowering of the plants, temperature, relative humidity and sunshine data were collected daily at the University of Ghana, Legon-Accra on Hobo Pro data loggers (Pocassett, ME, USA), whereas those of Manga-Bawku were taken from on-farm weather station. The rainfall data from both experimental sites was collected using on-farm rain gauges.

Table 1. Location and seasonal differences in monthly average climatic data per year from Manga-Bawku and Legon-Accra experimental farms during the 2012-2014 experimental period

Location	**Manga-Bawku Experimental Farm**							
Climatic Parameter	Rainfall (mm)		Temperature (^{o}C)		Relative humidity (%)		Sunshine (Hours)	
Year / Month	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14
Oct-April	0.2	0.2	29.8	30.7	50.4	50.2	8.5	8.4
May-Sept.	114.1(4)	102.9(3)	27.7	28.1	80.7	80.1	6.4	6.4
Yearly Mean	**47.6 (4)**	**43 (3)**	**28.3**	**29.4**	**63.1**	**62.6**	**7.5**	**7.4**

Location	**Legon-Accra Experimental Farm**							
Climatic Parameter	Rainfall (mm)		Temperature (^{o}C)		Relative humidity (%)		Sunshine (Hours)	
Year / Month	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14
Nov-March	25.4(2)	12.8(2)	27.6	28.4	75.1	73.4	5.8	6.4
April-Oct.	89.5(4)	56 (3)	27	27.2	78	76	5.7	5.8
Yearly Mean	**62.0 (3)**	**37.6 (3)**	**27.3**	**27.6**	**76.5**	**74.9**	**5.8**	**6.2**

( )* = Mean days of rainfall

2.3 Planting Materials

Fourteen (14) garden egg (*Solanum aethiopicum*) genotypes were obtained from the Department of Crop Science, University of Ghana, Legon and Plant Genetic Resources Research

Institute (PGRRI) of the Council for Scientific and Industrial Research (CSIR), Bunso and two popular local genotypes of bitter garden egg (*Solanum incanum*) commonly cultivated in Bawku area, were obtained from a garden egg farmer in Bawku. The sixteen (16) garden egg genotypes were grown in two successive rainy and dry seasons' conditions of Coastal Savannah and Sudan Savannah agro-ecological zones in 2012 and 2013, and 2013 and 2014. Experimental procedure for the trials on the 16 genotypes was the same across seasons and locations.

2.4 Treatments and experimental design

The genotype, rainy season, dry season, water-stressed and location (Legon and Manga) were the main treatments. There were sixteen (16) genotypes, three (3) soil moisture conditions and two (2) locations, giving ninety-six (96) treatment combinations. After ploughing and harrowing, the experimental fields were laid out in Randomized Complete Block Design (RCBD) with three (3) replications in both rainy and dry seasons.

Plant-to-plant spacing within a row was 80 cm and planting in both years was done in May-June, and November-December, coinciding with the onset of rainy season and dry season of 2012-2013 and 2013-2014.

2.5 Soil moisture content determination

Following standard procedures and methods, soil moisture content was determined at Legon and Manga Experimental farms by using the weights of soil samples corresponding to the different pressure plates measurement at 0.3 bars and 15 bars, and then oven-dried at 105 ^{o}C for 48 hours to constant weights and weighed [31, 32]. The determined respective soil moisture

content values for Legon and Manga in the rainy season were 67.9% and 63.4%; dry season (irrigated) were 56.7% and 52.5% and water-stressed were 26.4% and 23.6%.

2.6 Leaf sampling, drying and milling

Twelve (12) uppermost leaves were sampled from four record plants per genotype per replication at 50% flowering in both the rainy and dry season experiments, and were oven-dried at 50 ^{o}C for 72 hours. During the dry season, leaves were sampled at 50% flowering under well-watered and ten-days of water deprivation (stress) conditions.

Four (4) leaves from the sampled twelve (12) leaves for proline determination were picked immediately after excision from plants and cleaned well for leaf relative water content (LRWC) following [33] and [34]. The remaining eight (8) of the sampled leaves per treatment per location were oven-dried at 50 ^{o}C for 72 hours.

The dried leaves from each location were bulked according to genotype and growth condition and ground into composite powders through a 1 mm mesh sieve fitted in the mill (Type: Fritsch, Schmeasal, AZ 15 ZVK-2005, Germany).

The composite leaf powders of the rainy season, dry season and stressed conditions were packaged in air-tight black polythene containers and stored in a freezer for analysis. The powdered leaf samples were used for determination of proline content.

2.7 Determination of proline content in leaf samples

The proline content of leaves was estimated colorimetrically by the acid-ninhydrin method, following [35]. Samples of dry leaf powder were weighed 0.5g and homogenized in 10 ml of 3%

aqueous sulfosalicylic acid. The homogenate was filtered through Whatman No. 1 paper and made up to 50 ml with distilled water. Proline standard concentrations of 5-100 µg/ml were prepared. One milliliter (1 ml) each of the filtrate (extract) and proline standards wa s pipetted into test tubes before adding 1ml acid ninhydrin and 1ml glacial acetic acid and mixed thoroughly. The mixtures were incubated for an hour at100 ^{o}C in water bath to develop colours. The test tubes were immediately cooled in an ice bath and vigorously vortex before adding 4 ml toluene reagent.

The chlomophore containing toluene was aspirated from the aqueous phase, and then warmed to room temperature (25 ^{o}C) and the absorbance read in a UV/Vis spectrophotometer at wavelength 520 nm, using toluene as blank. The proline concentration was calculated from a standard curve and computed on dry weight basis as µmole proline/g of dry leaf weight [35] as follows:

$$\mu\text{mole proline g}^{-1}\text{ dry weight} = \frac{(\mu\text{g proline/mL} - \text{Toluene/mL})\text{x Initial dilution x } 5}{115.5 \text{ x Sample weight}}$$

2.8 Analysis of proline content data

The proline concentration data was analyzed using GenStat Statistical Software (12th Edition). The data for each location and season for the two years were separately analyzed by general analysis of variance (ANOVA), for the estimation of the variation among the genotypes in the measured traits. Where ANOVA showed significant differences in proline, the mean values were separated by the Least Significant Difference (LSD) at probability level of 0.05

The coefficient of variation (% CV) was calculated as $= \frac{\sqrt{\text{MSE}}}{\bar{X}}$ x 100; where MSE = Error mean square; and $\bar{X}$ = Mean, from analysis of variance

3.0 RESULTS

Proline content in garden egg leaves at 50% flowering varied depending on the genotype, location and growth condition (Table 2). During rainy season conditions, location and genotype x location interaction effects on proline concentration were not significantly different (P = 0.05). The location and genotype x location interaction effects under dry-season conditions significantly (P = 0.05) affected the average proline levels of the genotypes.

Table 2: Proline accumulation in leaves of garden egg genotypes at flowering in rainy, dry season and drought-stressed conditions of two locations for two years

Condition	Rainy Season			Dry Season			Drought-Stressed		
Location	Manga	Legon	**Mean**	Manga	Legon	**Mean**	Manga	Legon	**Mean**
Genotypes	(µg/g dry weight)			(µg/g dry weight)			(µg/g dry weight)		
A1	0.44a	0.37ab	**0.41a**	0.78bc	0.55bc	**0.67c**	3.92bc	1.82d	**2.87d**
A2	0.40a	0.33b	**0.37ab**	0.83ab	0.65ab	**0.74ab**	4.22ab	3.65a	**3.93a**
A3	0.42a	0.40a	**0.41a**	0.82ab	0.72a	**0.77a**	4.30ab	3.64a	**3.98a**
A4	0.30b	0.38ab	**0.34bc**	0.88a	0.69a	**0.78a**	4.12b	3.85a	**3.99a**
A6B	0.43a	0.40a	**0.42a**	0.82ab	0.70a	**0.76a**	4.02b	3.90a	**3.96a**
A6F	0.37a	0.29bc	**0.39a**	0.84a	0.68a	**0.76a**	4.43a	2.94bc	**3.69bc**

A7	0.46a	0.42a	**0.44a**	0.80b	0.74a	**0.76a**	4.30ab	3.07bc	**3.68bc**
A8	0.42a	0.40a	**0.41a**	0.85a	0.66a	**0.76a**	4.22ab	3.78a	**4.00a**
A9A	0.45a	0.40a	**0.42a**	0.74c	0.65ab	**0.70bc**	3.96b	3.55a	**3.76a**
A9F	0.37a	0.29bc	**0.33bc**	0.83a	0.72a	**0.77a**	4.31ab	3.79a	**4.05a**
A10	0.44a	0.40a	**0.41a**	0.75c	0.70a	**0.73a**	4.41a	3.75a	**4.08a**
A11	0.22b	0.41a	**0.32bc**	0.81b	0.71a	**0.76a**	4.31ab	3.51a	**3.91a**
A12	0.31b	0.43a	**0.37ab**	0.87a	0.67a	**0.77a**	4.22ab	3.65a	**3.71b**
Legon1	0.42a	0.40a	**0.41a**	0.78bc	0.72a	**0.75a**	4.37a	3.52a	**3.95a**
Bawku1	0.45a	0.38a	**0.42a**	0.81b	0.71a	**0.76a**	4.42a	3.51a	**3.97a**
Bawku2	0.47a	0.40a	**0.43a**	0.84a	0.61bc	**0.72ab**	4.20b	2.46c	**3.33c**
Mean	**0.40**	**0.39**	**0.39**	**0.81**	**0.68**	**0.75**	**4.25**	**3.37**	**3.82**
%CV	15.3	11.6	14.4	4.7	9.2	7.6	4.3	18.2	12.4

*Means with different letters in a column are significantly different at P = 0.05. LSD (5%) (Proline): Location (Rain-fed = 0.03ns; Dry season = 0.02**; Drought-stressed = 0.12**) Genotype x Location (Rainy season = 0.11ns; Dry season = 0.09**; Drought-stressed = 0.48**). ns = Not significant; ** = Significant at 1% levels of probability.*

Under drought-stressed conditions, the location and genotype x location interaction effects on the proline contents of the genotypes were significant (Table 2). At each location, the rainy and dry season conditions did not have significant effects on genotype proline levels; whereas drought-stressed conditions at each location significantly ($P < 0.001$) affected genotypes' proline accumulation. Generally, the proline levels of the genotypes in the dry season of growth were higher than that of rainy season, whereas the levels of proline in genotypes under drought-stressed were about ten-fold higher than those in the rainy season and about five-fold higher than those under dry season conditions. In general, the proline levels of the genotypes across the growth seasons and conditions were consistently higher at Manga than at Legon.

Under drought-stressed conditions (Table 2), the Manga site recorded proline levels ranging from 3.93 µg/gDW in A1 to 4.43 µg/gDW in A6F; the levels at Legon ranged from 1.72 µg/gDW in A1 to 3.91 µg/gDW in A6B. Across locations, the genotypes proline levels ranged from 2.87 µg/gDW in A1 to 4.08 µg/gDW in A10. The site means ranged from 3.36 µg/gDW at Legon to 4.24 µg/gDW at Manga. The highest six proline accumulating genotypes in drought-stress conditions across the locations, in the order of highest was A10 (4.08 µg/gDW), A9F (4.05 µg/gDW), A8 (3.99 µg/gDW), A4 (3.98 µg/gDW), A3 (3.97 µg/gDW) and Bawku1 (3.96 µg/gDW).

There were significant genotype and genotype and environment interaction effects on proline synthesis in garden egg grown across seasons of the Coastal and Sudan savannah agro-ecologies. The drought-stressed conditions of both locations were also associated with low leaf relative water contents of the genotypes (Table 3) but with higher variability (CV = 13.3%) among genotypes than the dry season variability (CV = 8.5%). The proline content in the leaves of the genotypes also increased as leaf relative water contents decreased (Tables 2 & 3). This indicates strong relationship between leaf water content and proline levels in garden egg plants.

Table 3: Leaf relative water content (LRWC) of garden egg genotypes at flowering under rainy, dry season and drought-stressed conditions of two locations for two years

Condition	Rain season			Dry season			Water-stressed		
Location	Manga	Legon	**Mean**	Manga	Legon	**Mean**	Manga	Legon	**Mean**
Genotypes	%	%	**%**	%	%	**%**	%	%	**%**
A1	78.4d	82.7c	**80.5f**	63.4b	75.2b	**69.3b**	47.7b	51.0b	**49.3b**
A2	78.7d	80.4c	**79.5f**	63.3b	75.3b	**69.3b**	48.2b	50.7b	**49.5b**

A3	84.2b	84.8bc	**84.5c**	61.1c	73.7b	**67.4c**	52.6a	60.7a	**56.4ab**
A4	83.5b	77.2d	**80.4f**	63.2b	75.9a	**69.5b**	47.4b	51.7b	**49.6b**
A6B	80.1c	79.4d	**79.8f**	63.5b	75.0b	**69.2b**	48.9b	53.8b	**51.3b**
A6F	85.8a	78.0d	**81.9e**	67.3a	77.2a	**72.3a**	50.5b	58.7a	**54.6ab**
A7	81.0c	87.0ab	**84.0c**	65.7b	73.4b	**69.5b**	53.6a	60.5a	**57.0ab**
A8	77.1d	84.9b	**81.0e**	66.2b	75.4a	**70.8b**	54.0a	61.5a	**57.8a**
A9A	84.3b	85.8b	**85.1c**	64.5b	73.9b	**69.2b**	54.0a	61.8a	**57.9a**
A9F	77.3d	86.3b	**81.8e**	65.3b	73.2b	**69.3b**	53.4a	58.1a	**55.7ab**
A10	80.3c	86.5ab	**83.4d**	70.3a	75.2b	**72.7a**	53.8a	50.6b	**52.2b**
A11	81.5c	85.4b	**83.5d**	64.8b	76.8a	**70.8b**	51.5a	62.5a	**57.0ab**
A12	77.4d	86.5ab	**82.0e**	63.1b	75.0b	**69.1c**	51.8a	57.9a	**54.9ab**
Legon1	79.5c	84.9b	**82.2e**	69.0a	74.1b	**71.6a**	53.1a	52.4b	**52.7b**
Bawku1	87.4a	89.3a	**88.3a**	64.3b	76.1a	**70.2b**	54.4a	65.0a	**59.7a**
Bawku2	87.6a	86.5ab	**87.0b**	68.9a	78.0a	**73.5a**	56.0a	63.1a	**59.6a**
Mean	**81.5**	**84.1**	**82.8**	**65.3**	**75.2**	**70.2**	**51.9**	**57.5**	**54.7**
%CV	4.9	4.9	5.1	6.0	3.4	8.5	9.3	14.3	13.3

Means with different letters in a column are significantly different at P = 0.05. LSD(5%) (LRWC at flowering): Rainy season (Location= 0.4**; Genotype x Location = 1.7**); Dry season (Location = 0.9**; Genotype x Location = 3.4**); and, Drought-stressed (Location = 1.69**; Genotype x Location = 6.8**). ** = Significant at 1% level of probability.

The reduction in moisture content of leaves in the dry season could also be due to utilization of the moisture to build proline and other leaf constituents. The accumulation of proline enable plants to maintain low water potentials, and this condition in plants could trigger the accumulation of other compatible osmolytes as well as chlorophyll, and allows additional water

209 to be taken up from the environment, and hence help in buffering the immediate effect of water
210 deficit within the leaf [36, 37]. In dry conditions, the proline in garden egg remained active and
211 so some amount of water retention was made possible (Table 2&3).

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217 4.0 DISCUSSIONS

218 The concentration of proline in the leaves of garden egg genotypes depended on the soil
219 moisture levels of the rainy season, dry season and drought-stressed conditions of Manga and
220 Legon (Table 2). With the exception of the rainy season, the dry season and drought-stressed
221 conditions significantly ($P = 0.05$) affected the proline levels in the genotypes. The growth
222 conditions of Manga resulted in higher levels of proline in plants than Legon, indicating that
223 environmental conditions of Manga triggered higher proline synthesis than Legon. Seasonally,
224 the dry season conditions enhanced proline synthesis than rainy season, suggesting that the
225 rainy season and for that matter higher moisture conditions inhibit proline synthesis in garden
226 egg.

227 This is an indication that proline accumulation may result from both induction of proline
228 biosynthesis and/or inhibition of its oxidation [38, 39]. The induction of proline biosynthesis is
229 activated by the enzyme pyrroline-5-carboxylate synthetase, and proline is inhibited from
230 degeneration by the enzyme proline dehydrogenase [38, 22, 40].

231 Plants accumulate proline when exposed to abiotic stresses such as drought [41, 42], as well as
232 varying temperatures [43]. The high proline accumulation in the garden egg genotypes during
233 the dry season and drought-stressed conditions could be attributed to lack of adequate water
234 supply or due to high sunshine and temperatures at that period. During the dry season,
235 temperatures were generally high across ecologies (Table 1), and so temperature increases in
236 addition to low soil moisture or drought stress trigger and significantly increased proline
237 synthesis through enhanced activities of the biosynthetic enzyme, pyrroline-5-carboxylate
238 reductase.

239 High proline accumulation is part of physiological responses to an intense stress and it is
240 indicative of higher capability to resist drought [44-47]. This is an indication that during drought
241 stress, garden egg plants generally have inherent ability to counteract or minimize the effects
242 through proline accumulation. It is also suggestive that, the production of proline is probably a
243 common response of garden egg under drought-stress.

244 The osmotic adjustment through the accumulation of cellular solutes, such as proline, has been
245 suggested as one of the possible means for overcoming osmotic stress caused by loss of water
246 [15, 16, 48]. In this study, proline content in the leaves of garden egg genotypes tended to
247 increase as leaf relative water contents decreased (Tables 2 & 3), indicating strong relationship
248 between leaf water content and proline content in garden egg plants.

249 The proline levels enable plants to maintain low water potentials, and it is this condition that
250 triggers the accumulation of other compatible osmolytes and allows additional water to be
251 taken up from the environment, and hence help in buffering the immediate effect of water

deficit within the leaf [36, 37]. The drought-stressed conditions of both locations were associated with low leaf relative water contents of the genotypes (Table 3) suggesting that, the accumulation of proline is probably a mechanism to withhold water during periods of water stress [36].

Regardless of the growth conditions of the crop, there were significant differences (P = 0.05) among genotypes in proline accumulation, suggesting that garden egg genotypes differ in their abilities to synthesize proline. The variation in the genotypes proline levels across locations was higher under drought-stressed conditions (CV = 12.4%) than the dry season conditions (CV = 7.6%) (Tables 2), and this clearly indicates the influence of drought-stressed conditions on proline accumulation in garden egg. Though there were location specific genotypic differences, the highest six proline accumulating genotypes under drought-stressed conditions across locations, were A3, A4, A8, A9F, A10 and Bawku1, and this present great opportunity in drought tolerant improvement programmes in garden egg under Coastal and Sudan savannah agro-ecologies of Ghana.

5.0 Conclusion

Proline as a bioactive compound, confer tolerance of many plants genotypes to drought or moisture stressed conditions. Garden egg genotypes at reproductive phase varied in their proline accumulation ability under drought or moisture stressed conditions. Under drought conditions, the crop genotypes can develop internal complementary drought survival mechanisms by lowering leaf relative water contents (LRWC) and increasing proline concentrations, thereby enabling genotypes to withstand periodic drought better.

The information on genotypic differences in proline accumulation is useful in the survival and productivity of garden egg, and could be useful in setting the crop breeding objectives. Though there were location specific genotypic differences, the highest six proline accumulating genotypes under drought-stressed conditions across locations, were A10, A9F, A8, A4, A3 and Bawku1. This presents great opportunity in drought tolerant improvement programmes in garden egg for improved performance in drought-prone agro-ecologies of Ghana.

COMPETING INTERESTS

Authors declared there are no competing interests

REFERENCES

1 Bonsu K. O., Owusu E. O., Nkansah, G. O. and Schippers R. R.. Preliminary characterization of *Solanum macrocarpon* germplasm from different ecological zones of Ghana. Workshop for Identification of vegetable Research Priorities. Brong Ahafo Region of Ghana, 10-11th September, 1998, Sunyani, Ghana. 1998; 7.

2 Ministry of Food and Agriculture (MOFA). Agriculture in Ghana: facts and figures. Produced by the Statistics, Research and Information Directorate. Accra. 2003.

3 Pirzad, A., Shakiba, M. R., Zehtab-Salmasi, S., Mohammadi, S. A., Darvishzadeh, R. and Samadi, A. Effect of water stress on leaf relative water content, chlorophyll, proline and soluble carbohydrates in *Matricaria chamomilla* L.). Journal of Medicinal Plants Research. 2011; 5(12): 2483-2488

4 Bai, C., Liang, Y., Zhu, Y., Ge, Y., Lin, X. and Jia, W.. The temporal and spatial variation of soil respiration in pepper (*Capsicum annuum* L.), eggplant (*Solanum melongena* L.) and maize (*Zea mays* L.) agro-ecosystems in Northwest of China. Australian Journal of Crop Science. 2012; 6(11):1565-1571.

5 Meletiou-Christou, M. S., Banilas, G. P. and Diamantoglou, S. Seasonal trends in energy contents and storage substances of the Mediterranean species *Dittrichia viscosa* and *Thymelaea tartonraira*. Journal of Environment and Experimental Botany. 1998; 39: 21–32.

6 Wyka, T. Carbohydrate storage and use in an alpine population of the perennial herb Oxytropis sericea. Oecologia. 1999; 120: 198–208.

7 Fayyaz-Ul-Hassan, Qadir, G. and Cheema, M. A.. Growth and development of sunflower in response to seasonal variations. Pakistan Journal of Botany. 2005; 37(4): 859-864

8 Akinci, S. and Lösel.. Plant water stress response mechanisms. (Ismail md. Mofizur Rahman (ed). Intech Publishers. 2012; 1-42. http://www.intechopen.com/books/water-stress
(Accessed June, 2016).

9 Crasta, O. R. and Cox, W. J. Temperature and soil water effects on maize growth, development yield and forage quality. Journal of Crop Science. 1996; 36: 341-348.

10 Bray, E. A. Plant responses to water deficit. Trends in Plant Science. 1997; 2: 48–54

11 Pirzad, A., Shakiba, M.R., Zehtab-Salmasi, S., Mohammadi, A., Darvishzadeh, R. and Hassani, A. Phenology of *German chamomile* and its changes under different irrigation regimes and plant densities. Notulae Scientia Biologicae. 2010; 2:43-48.

12 Bray, E. A., Bailey-Serres, J. and Weretilnyk, E.. Responses to abiotic stresses. In: Gruissem W, Buchannan B, Jones R (eds) Biochemistry and molecular biology of plants. ASPP, Rockville, MD. 2000; 1158-1249.

13 Kakati, L. N. and Kakati, B. T. Seasonality of nutrient contents of different leaf types of two primary host plants of *Antheraea assamensis*, Helfer. Paper presented in 3rd International Conference on Climate Change, Forest Resource and Environment (ICCFRE), held 9-11th December, 2011, University of Kerala. In: Journal of Environmental Sciences. 2011; 1: 261-265.

14 Heuer, B.. Role of proline in plant response to drought and salinity. In: Handbook of plant and crop stress. M. Pessarakli (ed). 3rd Edition. CRC Press. 2010; 213-238

15 Singh, D. K., Sale, P. W. G., Pallaghy, C. K. and Singh, V.. Role of proline and leaf expansion rate in the recovery of stressed white clover leaves with increased phosphorus concentration. Journal of New Phytololy. 2000; 146 (2): 261-269.

16 Caballero, J. I., Verduzco, C. V., Galan, J., Jimenz, E. S. D.. Proline accumulation as a symptom of drought stress in maize: A tissue differentiation requirement. Journal of Experimental Botany. 2005; 39 (7): 889-897.

17 Ueda, A., Shi, W. M., Sanmiya, K., Shono, M. and Takabe, T. Functional analysis of salt-inducible proline transporter of barley roots. Journal of Plant Cell Physiology. 2001; 42:1282-1289.

18 Sairam, R. K., Veerabhadra Rao, K. and Srivastava, G. C. Differential response of wheat genotypes to long term salinity stress in relation to oxidative stress, antioxidant activity and osmolyte concentration. Journal of Plant Science. 2002. 163: 1037–1046.

19 Chiang, H. H. and Dandekar, A. M. (1995). Regulation of proline accumulation in Arabidopsis thaliana (L.) Heynh during development and in response to desiccation. Journal of Plant Cell and Environment. 18: 1280–1290.

20 Lobato, A. K. S., Oliveira Neto, C. F., Costa, R. C. L., Santos Filho, B. G., Cost, R. C. L., Cruz, F. J. R., Neves, H. K. B. and Lopes, M. J. S. Physiological and biochemical behavior in soybean (*Glycine max cv. Sambabia*) plants under water deficit. Australian Journal of Crop Science. 2008; 2: 25-23.

21 Ssemakula, G. and Dixon, A. Genotype x environment interaction, stability and agronomic performance of carotenoid-rich cassava clones. Journal of Scientific Research and Essay. 2007; 2 (9): 390-399

22 Maggio, A., Miyazaki, S., Veronese, P., Fujita, T., Ibeas, J. I., Damsz, B., Narasimhan, M. L., Hasegawa, P. M., Joly, R. J. and Bressan, R. A.. Does proline accumulation play an active role in stress-induced growth reduction. Plant Journal. 2002; 31: 699–712.

23 Chutia, J. and Borah, S. P.. Water stress effects on leaf growth and chlorophyll content but not the grain yield in traditional rice (*Oryza sativa*, Linn) genotypes of Assam, India II. Protein and proline status in seedlings under PEG induced water stress. American Journal of Plant Sciences. 2012; 3: 971-980

24 Sivakumar, P., Sharmila, P. and Pardha Saradhi, P. Proline alleviates salt-stress-induced enhancement in ribulose-1, 5-bisphosphate oxygenase activity. Biochemical and Biophysical Research Communications. 2000; 279: 512-515

25 Ashraf, M., Saeed, M. M. and Qureshi, M. J.. Tolerance to high temperature in cotton (*Gossypium hirsutum*, L.) at initial growth stages. Environmental Experimental Botany. 1994; 34: 275

26 Watanabe, S., Kojima, K., Ide, Y. and Satohiko, S. Effects of saline and osmotic stress on proline and sugar accumulation in *Populus euphratica* in vitro. Journal of Plant Cell Tissue Organ. 2000; 63: 199-206.

27 Saruhan, N., Terzi, R., Kadioglu, A. The effects of exogenous polyamines on some biochemical changes during drought stress in Ctenanthe setosa. Acta Biol Hung, 2006; 57: 221-229.

28 Ramanjulu, S. and Bartels, D.. Drought- and desiccation-induced modulation of gene expression in plants. Journal of Plant Cell and Environment. 2002; 25: 141–151.

29 Xiao, X., Xu, X. and Yang, F. Adaptive responses to progressive drought stress in two *Populus cathayana* populations. Silva Fennica, 2008; 42(5): 705–719.

30 Nkansah, G. O.. Some physiological features of the African eggplant, *Solanum aethiopicum* group 'Gilo'. Journal of Science of Horticulture. 2001; 90: 181-186.

31 Miller, R.W. and Donahue, R. L.. Soils: An introduction to soils and plant growth, sixth Edition, Prentice – Hall International (UK) limited, London. 1990; 110-11, 282,315 and 488.

32 Brady, N.C. and Weil, R.R.. The nature and properties of soil (14th Ed.). Prentice Hall. Upper Saddle River, N.J. 2008; Chapter 4.

33 Turner NC, Begg JE. Plant water relations and adaptation to drought. Journal of Plant and Soil Science. 1981;58:97-113.

34 Yamasaki S, Dillenburg LR. Measurements of leaf relative water content in *Araucaria angustifolia*. Revista Brasilleira de Fisiologia Vegetal. 1999; 11(2):69–75.

35 Bates, I. S., Waldern, R. P. and Teare, I. D.. Rapid determination of free proline for water stress studies. Journal of Plant and Soil. 1973; 39: 205-207.

36 Mousa, H. R. and Abdel-Aziz, S. M.. Comparative response of drought tolerant and drought sensitive maize genotypes to water stress. Australian Journal of Crop Science. 2008; 1: 31-36

37 Sarker, B. C., Hara, M. and Uemura, M.. Proline synthesis, physiological responses and biomass yield of eggplants during and after repetitive soil moisture stress. Journal of Science of Horticulture. 2005; 103: 387-402.

38 Hong, Z., Lakkineni, K., Zhang, Z. and Verma, D. P. S. Removal of feedback inhibition of Δ1-pyrroline-5-carboxylate synthetase results in increased proline accumulation and protection of plants from osmotic stress. Journal of Plant Physiology. 2000; 122: 1129-1136.

394 39 Kumar, V., Rani, A., Solanki, S. and Hussain, S. M.. Influence of growing
395 environment on the biochemical composition and physical characteristics of soybean seed.
396 Journal of Food Composition and Analysis. 2006; 19: 188-195.

397 40 Claussen, W.. Proline as a measure of stress in tomato plants. Journal of Plant
398 Science. 2005; 168: 241-248.

399 41 Kirnak, H., Kaya, C., Tas, I. and Higgs, D. The influence of water deficit on
400 vegetative growth, physiology, fruit yield and quality in eggplants. Bulgarian Journal of
401 Plant Physiology, 2001; 27(3–4): 34–46.

402 42 Rubio, M. C., González, E. M., Minchin, F. R., Webb, K. J., Arrese-Igor, C., Ramos, J. and
403 Becana, M.. Effects of water stress on antioxidant enzymes of leaves and nodules of
404 transgenic alfalfa overexpressing superoxide dismutases. Physiol. Plantarum. (2002; 115:
405 531–540.

406 43 Pastori, G. M. and Foyer, C. H. Common components, networks, and pathways of
407 cross-tolerance to stress. The central role of “redox” and abscisic acid-mediated controls.
408 Journal of Plant Physiology. 2002; 129: 460–468.

409 44 Kavi Kishore, P. B., Hong, Z., Miao, G. H., Hu, C. A. A. and Verma, D. P. S.
410 Overexpression of Δ-Pyroline-5-carboxylate synthetase increase proline production and
411 confers osmoltolerance in transgenic plants. Journal of Plant Physiology. 1995; 108: 1387-
412 1394

413 45 Hayashi, H., Alia Mustardy, L., Deshnium, P., Ida, M., Murata, N. Transformation
414 of *Arabidopsis thaliana* with the codA Gene for Chlorine oxidase: Accumulation of
415 Glycinbetaine and Ehanced Tolerance to Salt and Cold Stress. Journal of Plants. 1997; 12:
416 133-142

417 46 Yoshiba, Y., Kiyosue, T., Nakashima, K., Yamaguchi-Shinozaki, K., Shinozaki, K.
418 Regulation of Levels of Proline as an Osmolyte in Plants under Water Stress. Journal of
419 Plant Cell Physiology. 1997; 38:1095-1102

420 47 Hare, P. D., Cress, W. A. and Van Staden, J. Dissecting the roles of osmolyte
421 accumulation under stress. Journal of Plant Cell and Environment. 1998; 21: 535–553

422 48 Ashraf, M. and M. R. Foolad, M. R. Role of glycine betaine and proline in
423 improving plant abiotic stress resistance. Journal of Environmental and Experimental
424 Botany. 2007; 59(2): 206-216.

425