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# Potential impacts of amino acids, putrescine and glycine betaine on productivity and fruit attributes of "Le-Conte" pear trees grown under water scarcity stress

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# **ABSTRACT:**

The problem of water shortage worries many countries in the world because its negative impact on many economic crops. In this study, the foliar application by amino acids 3 cm/l, putrescine 1.03 g/l, glycine betaine 1.35 g/l, betaine 0.45 g/l and proline 1.28 g/l. added to reduce stress These compounds were sprayed twice, the first spraying one month after fruit set, the second spraying two months after fruit set on "Le-Conte" pear trees that grow under conditions of water scarcity stress (60% of water requirements) and control (60% of water requirements) with any these compounds treatments. Water scarcity stress caused a reduction in vegetative parameters, macro nutrients, soluble proteins, lowest productivity per tree and fruit weight, fruit valium, fruit length and fruit width, TSS, and fruit attribute. However, the contents of proline, full phenol, and antioxidant enzyme activity were augmented in pear trees grown under water scarcity stress. application of amino acids, putrescine, and glycine betaine on stressed trees showed different responses and improvements in the yield, fruit attribute, morphological traits, macronutrients, reduction of oxidative stress and antioxidant enzyme activity. a Foliar spray amino acids 3 cm/L on "Le-Conte" pear trees grow under conditions of water scarcity stress (60% CWR) during after fruit set stage to harvest gave the greatest yield per tree, fruit weight, fruit valium, fruit length, fruit diameter, TSS and vegetative growing compared to control (60% CWR) in two seasons. It could be recommended that foliar spray of amino acids, putrescine and glycine betaine, especially amino acids, could be commercially intended for the stimulation of pear trees cultivated under water scarcity stress.

**KEYWORDS:** pear, Le-Conte, amino acids, putrescine, glycine betaine, proline, crop water requirement, yield, fruit weight, vegetative growth. antioxidant

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# **1. INRODUCTION:**

Many economic crops face many risks and stress, such as drought, especially in light of climatic changes. In this regard, many investigators throughout the world have turned to try to improve plant physiological immunity against stress (Shahzad, et al. 2021). Pears are among the most popular fruits farmed globally. Regarding the cultivated area, it comes in sixth. The primary variety of pear grown in Egypt is "Le Conte." It was the off spring of Pyrus communis (L.) and Pyrus serotina (Rehd). The farmed area reached 11772 feddans, producing roughly 79206 tonnes with a standard output of 6.73 tonnes/feddan (Ministry of Agriculture, 2020), in comparison to the global average of 7.94 tons/feddan (Fao stat, 2020).

Drought is one of most predominant biotic factors controlling the global production of agriculture (Gholipoor et al., 2012; Ihsan et al., 2016), especially in arid or semiarid regions of the planet (Bodner et al., 2015). which are intensively affected by climate change (Wassmann et al. 2009) Water shortages are answerable for the paramount crop losses, and they are expected to worsen (Comas et al., 2019). 60-70% of the productivity losses in agriculture are estimated to be attributable to abiotic stresses (Rouphael et al., 2017; Yakhin et al., 2017). It also affects growth and development due to the reduction of turgor in the leaves, resulting in a reduced acquisition of nutrients from the soil by the plant (Luo et al. 2011). Drought reduces fruit yield and attribute (Alam et al., 2021). Water deficiency has an impact on a variety of physiological, biochemical, metabolic, and molecular processes in apple trees (Massonnet et al., 2007Zu et al., 2017;). Lack of water in plants induces oxidative stress (Lei et al., 2006; Noctor et al., 2014). Drought can cause a significant reduction

and damage in photosynthesis and chlorophyll degradation (Viljevac et al., 2013; Bhusal et al., 2019).

Amino acids aid in the development of trees and their defense against abiotic stresses. These substrates may affect how plants physiologically. They function are necessarv for the biosynthesis of chlorophyll and the metabolism of nitrogen (Bulgari et al., 2019). Plants can withstand stress better thanks to amino acids' role in the detoxification of reactive oxygen species, control, pН and osmotic adjustments (Khan et al., 2020). Proteins and other organic compounds, such as nucleic acids, which play an active role in how plants respond to various stresses, are mimicked by amino acids, which also serve as precursors to those compounds. These molecules can function as signaling and regulatory agents (Dondoni et al., 2006). Numerous crops have shown growthpromoting effects after receiving exogenous amino acid treatments (Mohammadipour and Souri 2019a). Under challenging environmental circumstances like water stress, the growth-stimulating effect of exogenous amino acids is especially stronger. It regulates membrane permeability and ion uptake, allowing plants to endure extreme stress (Khan, et al., 2020). Additionally, various amino acids function in plants in a variety of ways, including as an osmolyte, an enzyme regulator, a modulator of stomatal opening, a regulator of ion transport, and a heavy metal detoxifier (Torres and Dangl, 2005). Due to the presence of bioactive peptides (Colla et al., 2014, 2015), amino acids may also interfere with hormonal functions. Thereby promoting root and shoot growth and, consequently, crop productivity (Colla et al., 2014; Lucini et al., 2015).

Putrescine is a good plan to adapts crops to harsh environments and meet the demands of a complex global environment (Liu et al., 2007). Putrescine is one of the polyamines, a new class of plant growth regulators, which are thought to be growth substances and are involved in a wide range of physiological procedures like embryogenesis, cell partition. morphogenesis, and enlargement (Bais and Ravishankar 2002; Liu et al. 2006). According to Ahmed et al. (2017), putrescine plays a crucial role in numerous processes of plant growing and enlargement, in adding to playing roles as an antisenescence and anti-stress agent. It strengthens the stability of cell membranes and serves as an antioxidant (Li et al. 2015). It influences how plants react defensively to various environmental stresses, such as drought stress (Ahmed et al. 2013; Khorshidi and Hamedi 2014). It has been optional that putrescine acting a role in the building of other metabolites, e.g. proline, and that it can play as an osmolyte to decreased water loss from cells and help plants maintain higher leaf water content under stressful circumstances (Kotakis et al., 2014).

Underneath water scarcity conditions, proline (Pro), fundamental organic solute, is accumulated and increased in plants (Gill and Tuteja, 2012; Hayat et al., 2012; Anjum et al., 2016). In accordance with the authors, proline and protein building are critical in protecting cellular structures from stress caused by a water deficit. The facility of plants to live under stress from dehydration has been attributed to the activation of this synthesis of collective osmolytes and proteins (Molaei et al., 2012). Furthermore, it can hunt some free extremists produced under stressful circumstances (Liang et al., 2013). Under

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conditions. it harsh acts as an osmoprotectant (Khan et al., 2018a). Amassing of glycine betaine (GB) protects against a variety of environmental factors, such as drought (Chen and Murata, 2008). GB is a low-molecular-weight substance with high solubility and low viscosity. It can build up significantly in chloroplasts and plastids, the organelles responsible for photosynthesis when abiotic stresses are present (Fariduddin et al., 2013). As a result, GB functions as an essential osmolyte and encourages parental priminginduced drought tolerance (Wang et al., 2018). Due to increased accumulation of leaf GB and Pro hydro and osmo-priming and improved drought tolerance. These osmolytes improved leaf area and water relation parameters in plants under stress and reduced Na+ content (Tabassum et al., 2017). By maintaining the structure and activity of macromolecular complexes, GB controls the water balance between a plant cell and its surroundings (Khan et al., 2020).

The purpose of the study is to provide 40% of the water needed and lessen the negative effects of water scarcity on pear trees via specific ingredients and their effects on fruit productivity and quality by enhancing the physiological resistance of the trees. Amino acid foliar spray is regarded as a nutrient that protects against stress factors, particularly water deficiency.

# **2. MATERIALS AND METHODS:**

# Experimental conditions and plant material:

The present experiment was performed during 2020 and 2021 at a private orchard located in El-Khatatba district, Minufiya governorate. Mature "Le-Conte" pear trees budded on *Pyrus communis* rootstock, spaced 5 x 5 m, vase trained and subjected to cultural practices recommended by the Ministry of Agriculture and Soil

Reclamation, with an average height of 3.5 m and ground cover about 85% were adopted. Trees were drip irrigated using two drip irrigation lines for each row. Soil physical and chemical properties were determined in the laboratory of the Soil, **Table 1. Physical properties of the orchard soil.** 

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Water, and Environmental Res. Inst. according to the methods described by Jackson (1973), and the results are summarized in Table 1.

		Soil sample depth			
	Parameter	0-30 cm	30-60 cm		
		Value			
Physical	Fine sand %	40.43	39.28		
properties	Coarse sand %	45.18	48.00		
	Silt %	5.66	3.35		
	Clay %	8.73	9.37		
	Texture class	L. Sand	L. Sand		

# The experimental trees were divided into two groups:

1- Trees irrigated at a water level of 100% of the crop water requirement (CWR)

2- Watering trees at a water level of 60% CWR (stressed trees) control

Trees exposed to their water requirement during the after fruit set (fruitlets) to harvest (10/05 to 15/08/2020 season and 20/5 to 20/8/2021 season). This is the most perceptive phenological stage to water defect in this period, which has the peak rate of evapotranspiration in two seasons.

The stressed trees (Group 2) were sprayed with the following compounds to reduce the stress on the trees: Commercial amino acids (20% free amino acid) 3 cm/l (S + T1), putrescine 1.03 g/l (S + T2), glycine betaine 1.35 g/l (S + T3), betaine 0.45 g/l (S + T4), and proline 1.28 g/l (S + T5), These compounds were sprayed twice, the first spraying one month after fruit set, the second spraying two months after fruit set on "Le-Conte" pear trees

Note: the concentration of the previous materials was chosen according to the

recommendations of previous research to choose the best concentration.

The experimental design of each irrigation treatment consisted of four standard experimental plots distributed randomly in blocks. The standard plot was made up of 15 trees, organized into 4 adjacent rows. The 3 central trees of the middle row were devoted to assessments (each tree acting as a replicate), and the other 12 trees were guard trees.

# **Irrigation treatments:**

The applied levels of irrigation were calculated as daily crop water requirements (liters/tree/day), as follows:

1 – The 1st irrigation level (optimum rate) = 100% of the crop water requirement (CWR). This amount of water was calculated theoretically from the "TAHRIR" meteorological data of the planting region.

2-The 2nd irrigation level (low rate) is equal to 60% of the CWR.

The relative requirements were applied by changing the discharge of emitters used. Water requirements were calculated as elucidated by **Karmeli and Keller (1975)**:

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IR =	(Se.SL.E	To.ŀ	Kc.Kr/Ea)*(1/1-Lr)
	IR	=	Daily irrigation requirements
	Se. SL	=	Plant area (Plant distance on lateral* between laterals)
	ЕТо	=	Daily reference evapotranspiration on mm/day
	Kc	=	coefficient factor for pear trees (Allen, et al., 1998).
	Kr	=	Reduction coefficient Gc/0.85
	Gc	=	Ground cover (area of tree canopy)
	Ea	=	Efficiency of irrigation system (80-90%)
	Lr	=	Leaching requirements = Eci/Ecd
	Eci	=	Electrical conductivity of irrigation water
	Ecd	=	Electrical conductivity of drainage water

The ETo value, was calculated using data from the El-Khatatba district's atmospheric conditions. Crop irrigation requirements were scheduled weekly according to daily ETo. Since, Penman Monteith method was used to calculate ET crop for pear trees in the district during the 2020 and 2021 seasons of study using the CROPWAT model (**Smith 1991**).

FT <sub>a</sub> -		$0.408 \Lambda(Bn - C) + \gamma [900/(T + 273) II2 (es-eg)]$
E I O		$\frac{1}{1+0.24} = \frac{1}{2} =$
		$\Delta + \gamma (1 + 0.34 \text{ U2})$
ЕТо	=	reference evapotranspiration, mm/day
Rn	=	Net radiation (MJm-2d-1)
G	=	Soil heat flux (MJm-2d-1)
$\Delta$	=	slope vapor pressure and temperature curve (kPa °C-1)
Γ	=	psychrometric constant (kPa °C-1)
U2	=	wind speed at 2 m height (ms-1)
es-ea	=	vapor pressure deficit (kPa)
Т	=	Daily air temperature at 2 m height (°C)

Crop coefficient (KC) value was used for quantifying crop water use. It was calculated from the equation: KC = ETc /ETo; where ETc is ETe/ETo the actual water consumptive use and ETo is the reference (potential evapotranspiration).

The correction coefficient for ground cover was according to Fereres and Goldhmaer (1990).

To unify the applied nutrients, applications were done manually every week.

A crop coefficient (KC) value was used for quantifying crop water use. It was calculated from the equation: KC =ETc/ETo, where ETc is ETe/ETo, the actual water consumptive use, and ETo is the reference (potential evapotranspiration). According to Fereres and Goldhmaer (1990), the correction coefficient for ground cover. To unify the applied nutrients, applications were done manually weekly.

# **Experiment measurements:**

# 1. Yield

When fruits reach the maturity stage as said by **El-Azzouni et al. (1975)**, the number of fruits born on each tree was counted and multiplied by the middling fruit weight born on that specific tree, taken from a representative sample of ten fruits.

# 2. Fruit characteristic

Twenty mature fruit per tree were examined at the picking date from the experimented trees to determine fruit physical and chemical characteristics and the following were assessed:

Fruit weight (gm) was determined using a digital scale, fruit firmness (lb/inch2) was determined using a pressure tester, juice TSS% was determined using a hand refractometer, and juice titratable acidity% was determined using **A.O.A.C** (1990).

# 3. Vegetative growth parameters

A random sample of ten current year's shoots was tagged at the end of October for each tree devoted to morphological determinations. Average shoot length (cm), shoot diameter (cm) by using a vernier caliper, number of leaves/shoot first by counting the number of leaves per each shoot, and leaf area (cm2) was measured on leaves (the 3 and 4 leaves from the base) by using a leaf area metre.

## 4. Leaf content of macronutrients

From each of the replicates that were chemical devoted to analysis, а representative sample of thirty leaves born in the current season growth was taken in mid-July of each season, and the leaves were washed with tap water and oven dried at 60 °C. 0.5 gram of the dried samples was digested using  $H_2SO_4$  and  $H_2O_2$ as previously described by Cottenie (1980). The extract was used to conclude the following minerals:

Nitrogen content (%) in the digested solution by the modified microkjeldahl method as described by **Plummer (1971)**. Phosphorous content was determined calorimetrically according to the method **of Jackson (1958)**. Potassium content (%) against a standard using a flame-photometer (**Piper, 1950**).

# **3. RESULTS AND DISCUSSION:**

Amino acid at 60% of the crop water requirement (CWR) on stressed trees through fruitlets to harvest (F-H), yield per tree and fruit weight, valium, length, and Leaf ash percentage: ash content was determined using the method described in A.O.A.C. (1995). The extract ash sample was used to determine the following:

According to Jackson and Ulrich (1959) and Yoshida (1976), the Perking Elmer Atomic Absorption Spectrophotometer Model 2380 A1 was used to conclude macro-elements Mg% and micro-elements (Fe, Zn, Mn, and Cu (ppm).

# 5. Chemical analysis

At the end of the consideration stage, a sample of leaves from each replicate was collected for leaf chemical content determination. Total water-soluble protein content was determined according to the method of **Lowery** *et al.* (1951) using casein as a standard protein. The colorimetric method of Folin-Denis as described by **Daniel and George (1972).** 

Free proline content (mg/g d.wt) was determined according to the method described by **Bates** *et al.* (1973). The terminal buds, in addition to the first and second young leaves, were also used for the estimation of peroxidase (POX) and polyphenol oxidase (PPO) enzyme contents. Witch assayed as said by (Matta and Dimond 1963, Bergmeyer 1965, Shin, Bhandari et al. 2020) respectively.

# Statistical analysis

The analysis of variance (ANOVA) was done for each season separately as a splitsplit design according to procedures reported by **Gomez and Gomez (1984)**.The differences between mean values of treatments were compared by the L.S.D test at 0.05 level of probability

diameter were added to statically achieve their highest magnitudes in both seasons(Table 1). The lowest of the previously evaluated parameters was

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devoted to lowering the applied water regime to 60% CWR (control) during the F-H stage in two seasons.

The difference between the yields of the two seasons is very large, due to the lack of

fulfillment of the cold needs for the second year (total chilling in 2019/2020=712 hours: while 2020/2021 = 423 hours), which led to a delay and irregular in flowering, and increased fruit drop.

Table 1. Effect of treatments on yield/tree and average fruit weight, valium, length and diameter.

	Yi	ield	F. Weight		F. Va	F. Valium		ngth	f. diameter	
Treatments _	(kg/	'tree)	(gn	(gm)		(cm3)		m)	(cm)	
-	2020	2021	2020	2021	2020	2021	2020	2021	2020	2021
- -	154.44	67	330	336	317	327	9.80	9.93	8.27	8.47
$S + T_1$	$\pm 0.44$ <sup>a</sup>	$\pm 0.75^{a}$	±0.91 ª	±2.4 a	±2.6 ª	±2.5 ª	$\pm 0.40$ <sup>a</sup>	$\pm 017$ <sup>a</sup>	$\pm  0.54^{a}$	$\pm 0.29$ <sup>a</sup>
C I T	104.83	46	224 f	230	223	230	8.57	8.93	7.01	7.57
S + 1 <sub>2</sub>	$\pm 0.78$ f	$\pm 0.44^{e}$	$\pm 3$	$\pm 2.2$ f	$\pm 2.8$ f	$\pm 1.4$ f	$\pm 0.57$ bcd	$\pm 0.53$ bc	$\pm 0.21$ <sup>cd</sup>	$\pm 0.23$ <sup>d</sup>
C I T	108.58	49	232	246	233	247	8.70	8.90	7.11±	7.63
$5 + 1_3$	±0.55 °	$\pm 0.50^{d}$	$\pm 1^{e}$	±1.5 °	$\pm 1^{e}$	±1.6 °	$\pm 0.30^{bc}$	$\pm 0.33$ bc	0.28 bcd	$\pm 0.67^{cd}$
C I T	113.26	53	242	265	240	267	8.73±	8.97	7.70	7.73
$5 + 1_4$	$\pm 0.25^{d}$	$\pm 0.80^{\circ}$	$\pm 2.5$ d	±2 °	$\pm 3$ d	±2.72 °	0.27 <sup>bc</sup>	$\pm  0.23^{b}$	$\pm 0.54^{abc}$	±0.79 °
S I T	121.21	52	259	260	260	262	8.36±	$8.37\pm$	$7.47\pm$	7.77±
5 + 15	±0.70 °	$\pm 0.90^{\circ}$	±0.75°	$\pm 3.1^{d}$	±0.9°	$\pm 2.9$ d	0.35 <sup>cd</sup>	0.37 <sup>cd</sup>	0.26 bcd	0.33c
$C_{ontrol}(600/)$	$81.90 \pm$	39±	175±	195±	179±	$200\pm$	$7.88\pm$	$8.09\pm$	$6.90\pm$	7.26
	0.40 g	$0.80^{\mathrm{f}}$	1.5 <sup>g</sup>	1.3 <sup>g</sup>	1.7 <sup>g</sup>	1.9 <sup>g</sup>	0.22 d	0.41 <sup>d</sup>	0.30 <sup>d</sup>	±0.44 °
1000/ CWD	129.64	56	277	279	276	273	9.13±	9.15	7.88	8.02
100% CWR	$\pm 0.35$ <sup>b</sup>	$\pm 1.5$ <sup>b</sup>	$\pm 3.5$ <sup>b</sup>	$\pm 1.4$ <sup>b</sup>	$\pm 1.4$ <sup>b</sup>	$\pm 1.3$ <sup>b</sup>	0.63 ab	$\pm \ 0.19$ $^{b}$	$\pm 0.77$ ab	$\pm 0.84$ <sup>b</sup>
I SDat 0.05			1.35	3.73	3.65	3.74	0.73	0.60	0.79	0.19
LSDat 0.05	0.92	1.53								

# Stress 60% CWR (S), amino acids (T<sub>1</sub>), putrescine (T<sub>2</sub>), glycine betaine (T<sub>3</sub>), betaine (T<sub>4</sub>), proline (T<sub>5</sub>), crop water requirement (CWR) and means±standard deviation

Data in Table 2, the trees through which putrescine was applied at 60% CWR during the F-H stage in both seasons produced statistically the firmest fruits. As opposed to that, utilizing the greatest irrigation dose (100% CWR) during that period in two seasons resulted in noticeably the slightest fruit firmness.

In two seasons, the F-H stage was the exclusive time to apply amino acids at a rate of 60% CWR for the trees, which caused the greatest statistically meaningful

TSS%. Instead, the use of 100% CWR during F-H in two seasons caused the TSS% to be much lower than the mean (Table 2).

Agreeing to statistics, the usage of glycine betaine at a slight regime on trees during the F-H stage in two seasons was the cause of the maximum fruit juice acidity. During the same stage in two seasons, raising the utilized water to the excessive regime significantly lessened the acidity of the fruit juice (Table 2).

Table 2. Effect of treatments on average fruit firmness, TSS and fruit acidity.									
Treatments _	Firmness	s lb/inch <sup>2</sup>	TS	S	Aci	dity			
	2020	2021	2020	2021	2020	2021			
	20.35	20	14	15	0.020	0.025			
$S + T_1$	$\pm 0.40$ bc	$\pm 0.55$ <sup>cd</sup>	$\pm0.1^{\mathrm{a}}$	$\pm 0.3$ a	±0.02 °	$\pm 0.001$ d			
	21.72	21.92	11	12	0.024	0.024			
$S + T_2$	±0.52 ª	$\pm 0.8$ <sup>a</sup>	$\pm 0.05$ d	$\pm 0.2$ d	$\pm 0.00$ °	±0.001 °			
	21.20	20.96	13	14	0.027	0.029			
$S + T_3$	$\pm 0.18$ <sup>a</sup>	$\pm 0.14$ <sup>b</sup>	±0.7 b	$\pm 0.5$ <sup>b</sup>	$\pm 0.001$ a	$\pm 0.001$ <sup>a</sup>			
	19.55	19.40	12	13	0.027	0.026			
$S + T_4$	$\pm 0.38$ <sup>cd</sup>	$\pm 0.33$ de	$\pm0.5$ °	±0.1 °	±0.002 a	$\pm 0.00$ °			
	21.15	19.83	12.50	13	0.026	0.027			
$S + T_5$	$\pm 0.51$ ab	$\pm 0.17$ <sup>d</sup>	$\pm 0.4$ bc	±0.6 °	$\pm 0.001$ <sup>b</sup>	$\pm 0.001$ <sup>b</sup>			
	20.27	20.59	14	15	0.022	0.024			
Control (60%)	±0.72 °	$\pm 0.4$ bc	±0.2 ª	±0.3 a	$\pm 0.002$ d	±0.00 °			
	19.31	19.06	11	12	0.018	0.022			
100% CWR	$\pm 0.34$ d	±0.55 °	$\pm 0.4$ d	±0.2 <sup>d</sup>	±0.00 °	$\pm 0.001$ f			
LSD at 0.05	0.81	0.64	0.69	0.62	0.002	0.001			

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Stress 60% CWR (S), amino acids (T<sub>1</sub>), putrescine (T<sub>2</sub>), glycine betaine (T<sub>3</sub>), betaine (T<sub>4</sub>), proline (T<sub>5</sub>), crop water requirement (CWR) and means±standard deviation

The procedure of 100% CWR during theF-H stage was greatest effective in producing statistically the longest shoots, the broadest shoot thickness; the highest total of leaves each shoot, and the maximum leaf area followed by amino acid treatment on stressed trees (60% CWR) in both years. As opposed to that, agreeing to the double years, the shortest shoots, narrowest shoot diameter, fewest leaves, and smallest leaf area were committed to 60% CWR (control) (Table 3).

## Table 3. Effect of treatments on shoot length, shoot diameter, number of leaves and leaf area.

Treatments	Shoot Length (cm)		Shoot Diameter (cm)		Number o	f leave/shoot	Leaf area (cm2)	
-	2020	2021	2020	2021	2020	2021	2020	2021
с <u>-</u> т.	32±	32.67±	$0.60\pm$	$0.62\pm$	16.50±	$17.50 \pm$	29.13±	$30.50\pm$
5 + 11	0.7 <sup>b</sup>	0.32 <sup>b</sup>	0.06 <sup>ab</sup>	0.01 <sup>ab</sup>	0.40 <sup>b</sup>	0.49 <sup>b</sup>	$0.47$ $^{ab}$	0.63 <sup>b</sup>
S   T	25.50	27.50	0.55	0.58	12.62	$12.86 \hspace{0.1cm} \pm \hspace{1cm} 0.14$	28.80	29.50
5 + 12	±0.40 °	$\pm 0.80$ °	$\pm 0.04$ <sup>ab</sup>	$\pm 0.03$ <sup>ab</sup>	$\pm 0.37$ <sup>d</sup>	d	$\pm 0.29$ <sup>b</sup>	±0.41 °
	23.38±	25.75	$0.52\pm$	$0.55\pm$	13.47±	$14.16 \pm$	26±	27±
$S + T_3$	0.30 <sup>d</sup>	$\pm 0.24$ d	0.03 <sup>ab</sup>	0.04 <sup>ab</sup>	0.53 °	0.34 °	0.9 °	0.15 <sup>d</sup>
S   T	$22.33 \pm$	$22.50\pm$	$0.54\pm$	$0.58\pm$	$14.14\pm$	14.86±	$24.50\pm$	25±
5 + 14	0.20 <sup>e</sup>	0.29 °	0.06 <sup>ab</sup>	0.02 <sup>ab</sup>	0.37 °	0.24 °	0.45 <sup>d</sup>	0.84 e
C   T	23.50±	25±	$0.56\pm$	$0.59\pm$	$11.97\pm$	12.83±	$26.25\pm$	$26.58\pm$
5 + 15	0.60 <sup>d</sup>	$0.60^{\text{d}}$	$0.05$ $^{\rm ab}$	0.04 <sup>ab</sup>	0.23 de	0.17 <sup>d</sup>	0.65 °	0.42 <sup>d</sup>
Control (00/	19±	20±	$0.45\pm$	$0.48\pm$	$11.40\pm$	12±	22±	23±
Control 00%	$0.20^{\rm f}$	0.45 f	0.06 °	0.07 °	0.59 °	0.51 °	0.28 <sup>e</sup>	0.44 f
1000/ CWD	33.25±	35±	$0.62\pm$	$0.64\pm$	17.71±	18.33±	$30\pm$	31.71±
100% CWK	0.35 <sup>a</sup>	0.55 <sup>a</sup>	0.06 <sup>a</sup>	0.06 <sup>a</sup>	0.29 <sup>a</sup>	0.67 <sup>a</sup>	0.51 <sup>a</sup>	0.29 a
LSD at 0.05	0.76	0.88	0.087	0.075	0.72	0.71	0.95	0.87

Stress 60% CWR (S), amino acids (T<sub>1</sub>), putrescine (T<sub>2</sub>), glycine betaine (T<sub>3</sub>), betaine (T<sub>4</sub>), proline (T<sub>5</sub>), crop water requirement (CWR) and means±standard deviation

The uppermost leaf nitrogen % was statistically caused by the appliance of glycine betaine and amino acid on treed trees (60% CWR) at the F-H stage in two years. The same watering strategy deprived of any interventions at the F-H stage caused in much lower nitrogen levels in two seasons (table 4).

When trees were exposed to the least irrigation routine through the F-H stage, phosphorus concentration was statistically at its largest magnitude. The least P contented was produced in two seasons when proline was applied on treed trees.

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Data in Table (4) shows that the lowest irrigating schedule (60% CWR) at F-H stage and then amino acid treatment at an equivalent previous watering level significantly increased leaf K leaves. An extensive decrease in content occurred in two years when water amounts were increased to 100% CWR. The massive leaf Mg was greatly increased in twin seasons by applying putrescine on

stressed trees. The lowest level was significantly caused by the excessive water regime at the F-H stage (Table 4).

 Table 4. Effect of treatments on leaf nitrogen, phosphorus potassium and magnesium content.

Treatments	Ν	%	Р	%	K	%	Mg %	
	2020	2021	2020	2021	2020	2021	2020	2021
- • • • •	$3.26 \pm$	2.15 ±	$0.29 \pm$	$0.22 \pm$	$1.65 \pm$	$1.85 \pm$	$0.510 \pm$	$0.406 \pm$
$5 + 1_1$	0.04 <sup>a</sup>	0.30 <sup>a</sup>	0.01 <sup>b</sup>	0.07 <sup>b</sup>	0.24 <sup>ab</sup>	0.16 <sup>ab</sup>	0.009 ab	0.009 <sup>ab</sup>
S + T	$3.01 \pm$	2 ±	$0.27 \pm$	$0.21 \pm$	$1.29~\pm$	1.25±	$0.533 \pm$	$0.427\pm$
5 + 12	0.19 <sup>ab</sup>	0.19 <sup>ab</sup>	0.04 <sup>b</sup>	0.04 <sup>b</sup>	0.12 °	0.07 <sup>d</sup>	0.01 a	0.008 a
$\mathbf{S} \perp \mathbf{T}$	$3.28 \pm$	$2.18 \pm$	$0.29 \pm$	$0.22 \pm$	$1.51 \pm$	$1.48 \pm$	$0.513 \pm$	$0.409 \pm$
5 + 13	0.07 <sup>a</sup>	0.11 a	0.01 b	0.03 <sup>b</sup>	0.22 <sup>abc</sup>	0.13 <sup>cd</sup>	0.012 <sup>ab</sup>	0.01 <sup>ab</sup>
$\mathbf{S} + \mathbf{T}$	$2.76 \pm$	$1.81 \pm$	$0.30 \pm$	$0.23 \pm$	$1.59\pm$	$1.56 \pm$	$0.510 \pm$	$0.406 \pm$
5 + 14	0.13 <sup>bc</sup>	0.18 abc	0.03 <sup>b</sup>	$0.07$ $^{\rm ab}$	0.09 <sup>ab</sup>	0.25 bc	0.018 <sup>ab</sup>	0.015 <sup>ab</sup>
$S \perp T_{-}$	$2.51 \pm$	$1.67 \pm$	$0.21\pm$	0.19±	$1.37 \pm$	$1.34\pm$	$0.491 \pm$	$0.395\pm$
5 + 15	0.16 °	0.10 bc	0.03 °	0.025 <sup>b</sup>	0.14 <sup>bc</sup>	0.16 <sup>cd</sup>	0.028 <sup>b</sup>	0.026 bc
Control (60%)	$2.01 \pm$	$1.52 \pm$	$0.43~\pm$	$0.30\pm$	$1.70 \pm$	$1.89\pm$	$0.521\pm$	$0.406 \pm$
	0.19 <sup>d</sup>	0.08 °	0.02 <sup>a</sup>	0.01 a	0.2 ª	0.16 <sup>a</sup>	0.017 <sup>ab</sup>	0.014 <sup>ab</sup>
100% CWD	$2.13 \pm$	$1.60 \pm$	$0.31\pm$	$0.23\pm$	$1.28 \pm$	$1.25\pm$	$0.454 \pm$	$0.379\pm$
10070 CWK	0.25 <sup>d</sup>	0.37 °	0.005 <sup>b</sup>	$0.07$ $^{ab}$	0.08 °	0.19 <sup>d</sup>	0.02 °	0.011 °
LSD at 0.05	0.28	0.38	0.042	0.077	0.29	0.29	0.030	0.025

Stress 60% CWR (S), amino acids (T<sub>1</sub>), putrescine (T<sub>2</sub>), glycine betaine (T<sub>3</sub>), betaine (T<sub>4</sub>), proline (T<sub>5</sub>), crop water requirement (CWR) and means±standard deviation

Owing to the extreme regime (100% CWR) at the F-H stage in dual seasons, the leaf manganese was the greatest. While the significantly lower manganese a level was produced by reducing the employed water to a little regime (Table 5).

The peak leaf zinc level in dual years was greatly increased by applying betaine on stressed trees. The lowest level was significantly affected by the excessive water regime appliance at F-H stage (Table 5).

Regarding iron level in leaves, trees yielded with amino acids on tree that taken 60% CWR thru F-H stage in dual seasons had statistically the upper iron content. However, the leaf iron levels that were much lower were used to apply the maximum regime (100% CWR) at F-H stage in twofold seasons (table 5).

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The info in Table (5) shows that the lowly regime (60% CWR) of trees that application betaine at F-H stage was in charge of the finest statistical leaf copper ppm followed by amino acid treated. Whereas, raising the

applied amounts to 100% CWR in twin years the statistically least amount of those nutrients that had a detrimental impact.

Treatments	Mn (p	pm)	Zn (j	ppm)	Iron (	(ppm)	Cu (	Cu (ppm)	
	2020	2021	2020	2021	2020	2021	2020	2021	
S + T	$26.83 \pm$	21±	39±	12.95±	$195 \pm$	$140.5 \pm$	$34 \pm$	28.64	
<b>S</b> + <b>I</b> <sub>1</sub>	0.17 <sup>b</sup>	0.39 <sup>b</sup>	0.18 <sup>d</sup>	0.14 <sup>e</sup>	0.99 <sup>a</sup>	1.02 ª	0.62 <sup>b</sup>	$\pm 0.10$ <sup>b</sup>	
S   T	$21.96\pm$	$17 \pm$	$40\pm$	$14.07\pm$	$131 \pm$	$91 \pm$	$30 \pm$	$26 \pm$	
5 + 12	0.14 <sup>d</sup>	0.25 e	0.45 °	0.1 °	0.33 <sup>d</sup>	0.00 °	0.45 <sup>d</sup>	0.18 <sup>d</sup>	
S + T	$21.79 \pm$	$17 \pm$	$41.18\pm$	$14.46~\pm$	$137 \pm$	94.5±	$33 \pm$	$28 \pm$	
5 + 13	0.11 <sup>d</sup>	0.13 e	0.12 <sup>b</sup>	0.16 <sup>b</sup>	0.51 °	0.4 <sup>d</sup>	0.37 °	0.40 °	
S + T.	$26.64 \pm$	20±	$43.92 \pm$	$15.43 \pm$	$176 \pm$	$119.9\pm$	36.33	$30.19\pm$	
5 + 14	0.26 <sup>b</sup>	0.46 °	0.18 a	0.24 ª	0.30 <sup>b</sup>	0.26 °	$\pm 0.30$ a	0.19 a	
S   T	$24.02 \pm$	$19 \pm$	$38.85 \pm$	$13.65 \pm$	$188 \pm$	$121.55 \pm$	$30 \pm$	$25.93 \pm$	
5 + 15	0.19 °	0.55 <sup>d</sup>	0.15 <sup>d</sup>	0.00 <sup>d</sup>	$0.47$ $^{ab}$	0.36 <sup>b</sup>	$0.75^{\text{ d}}$	0.12 <sup>d</sup>	
Control (00/ CWD	$21 \pm$	16±	$36.61 \pm$	$12.86 \pm$	$116 \pm$	89 ±	$25.2 \pm$	$24.19\pm$	
	0.28 <sup>e</sup>	0.61 f	0.26 <sup>e</sup>	0.14 <sup>e</sup>	0.18 <sup>e</sup>	0.11 f	0.80 °	0.31 <sup>e</sup>	
1000/ CWD	$30 \pm$	$23.5\pm$	34±	$11.88 \pm$	$91 \pm$	$70 \pm$	$19.84\pm$	16.17±	
100% CWK	0.54 ª	0.51 ª	0.09 <sup>f</sup>	0.21 <sup>f</sup>	0.29 <sup>f</sup>	0.21 <sup>g</sup>	0.45 f	0.28 f	
LSD at 0.05	0.49	0.77	0.41	0.28	1.3	0.80	0.98	0.43	

# Table 5. Effect of treatments on leaf manganese, zinc , iron and copper content.

# Stress 60% CWR (S), amino acids (T<sub>1</sub>), putrescine (T<sub>2</sub>), glycine betaine (T<sub>3</sub>), betaine (T<sub>4</sub>), proline (T<sub>5</sub>), crop water requirement (CWR) and means±standard deviation

Table's 6 results show demonstrate a minimal water supply system (60% CWR) at F-H stage induced the highest levels of phenol concentration, proline, and polyphenol oxidase (PPO), and peroxidase (Pro) activation. For the same phenological the aforementioned activities stage, decreased when applied water amounts were increased (100% CWR), followed by amino acid treated on tree stressed (60% CWR).

In comparison to trees that have not been treated, leveled (60%) CWR at F-H stage, all usages on trees subjected to a lack of water level showed a decrease in proline, phenol concentration, polyphenol oxidase (PPO), and peroxidase (Pro) activation. The aforementioned activities decreased when usage water volume were risen (100% CWR), consequently, amino acids treated on trees stressed (60% CWR).

Peroxic	dase activi	ty						
Treatments	Pro (mg/10	line Dg d.wt)	Ph (mg/10	Phenol (mg/100g d.wt)		PO g f.wt)	PRO (u/g f.wt)	
	2020	2021	2020	2021	2020	2021	2020	2021
C   T	$1.66 \pm$	$1.45$ $\pm$	1.47	1.22	1.03	0.95	$0.044\pm$	0.043
$S + I_1$	0.08 °	0.25 <sup>b</sup>	$\pm 0.07$ <sup>d</sup>	$\pm 0.02$ d	$\pm 0.15$ <sup>b</sup>	$\pm 0.07$ bc	0.007 <sup>b</sup>	$\pm 0.003$ <sup>b</sup>
C I T	$1.11 \pm$	$1.05$ $\pm$	$2.08$ $\pm$	1.73±	$1.11 \pm$	$0.99\pm$	$0.045 \pm$	0.044
$S + 1_2$	0.10 e	0.14 <sup>d</sup>	0.21 °	0.10 c	0.10 <sup>b</sup>	0.13 <sup>b</sup>	0.004 <sup>b</sup>	$\pm 0.005$ b
S   T	$1.01 \pm$	$0.97$ $\pm$	$1.41 \pm$	1.09	$0.91\pm$	$0.84\pm$	$0.011\pm$	0.009±
5 + 13	0.20 f	0.18 e	0.40 <sup>d</sup>	$\pm 0.07$ d	0.09 bcd	0.42  bcd	0.045 °	0.001 <sup>d</sup>
S   T	$1.57 \pm$	$1.38$ $\pm$	$2.55$ $\pm$	2.12±	$0.78\pm$	$0.64\pm$	$0.037 \pm$	0.031±
5 + 14	0.13 <sup>d</sup>	0.30 °	0.31 <sup>b</sup>	0.18 <sup>b</sup>	0.22 <sup>cd</sup>	0.04 <sup>cd</sup>	0.015 <sup>b</sup>	0.009 °
S   T	$2.63 \pm$	$2.32 \pm$	$2.81 \ \pm$	2.36±	$0.95\pm$	$0.87\pm$	$0.021\pm$	0.013±
5 + 15	0.10 <sup>b</sup>	0.02 a	0.19 <sup>b</sup>	0.20 <sup>b</sup>	0.16 bc	0.14  bcd	$0.002^{bc}$	0.001 <sup>d</sup>
Control 600/	$2.85 \pm$	$2.35~\pm$	$3.46$ $\pm$	$3.26\pm$	$1.95 \pm$	$1.75\pm$	0.071	$0.081\pm$
Control 00%	0.14 ª	0.05 a	0.15 ª	0.24 <sup>a</sup>	0.05 a	0.16 <sup>a</sup>	$\pm 0.011$ <sup>a</sup>	0.006 <sup>a</sup>
100% CWR	0.84	0.79	1.39	1.08	0.71	0.58	0.009	0.007
	$\pm 0.16$ g	$\pm 0.11$ f	$\pm 0.07$ <sup>d</sup>	$\pm 0.37$ d	$\pm 0.09$ d	$\pm 0.07$ <sup>d</sup>	$\pm 0.00$ °	$\pm 0.02$ d
LSD at 0.05	0.009	0.006	0.39	0.34	0.23	0.32	0.017	0.0083

Mohamed A. Abd El-wahab and Naglaa. Shakweer FJARD VOL.38,NO.1.PP 15-35 (2024) Table 6. ETable 6. effect of treatments on Proline, phenol content, polyphenol oxidase and

# Stress 60% CWR (S), amino acids (T<sub>1</sub>), putrescine (T<sub>2</sub>), glycine betaine (T<sub>3</sub>), betaine (T<sub>4</sub>), proline (T<sub>5</sub>), crop water requirement (CWR) and means±standard deviation.

A lack of water at the phase of phenology at fruitlets set to harvest is the most vulnerable to a lack of water. During this time, when evapotranspiration is at its highest, plant cells are subjected to oxidative explosions, which have a negative a result size of fruits and crop productivity.

According to the study findings, stressed trees had greatly diminished vegetative growth, which had a negative influences production and fruiting attributes. These findings are in agreement with Li et al. (2020) and Dietz et al. (2021). The use of the tested stimulants (amino acids, glycine betaine, putrescine, and proline) increased the quantity of the yield and improved the features of fruits and vegetative development in pear trees cultivated in water-scarce environments of 60% CWR providing strong evidence of plant resistance to water stress.

Our findings verified that yield per tree and average fruit weight, valium, length, and diameter were statistically at their highest levels. An amino acid beneficial effects on growth and some chemical elements at 60% CWR on trees through after fruit set to harvest in twin seasons are verified. These outcomes correspond to Khattab et al., 2014; Colla et al., 2017; Radkowski and Radkowska 2018

Usually, plants being a biotical stressed show signs of amino acids (Ferreira et al., 2018). Plants can use amino acids as replacement substrates for mitochondrial respiration in situations where there aren't enough carbohydrates available because photosynthesis rates have decreased, which typically happens under dry soil stress (Hildebrandt, 2018).

These findings are explicable by **Rizwan et al. (2017); Hussain et al. (2018)**, who noted that amino acids possess a range of

significant biological functions in plant cells, including the cleansing from toxins; optimizing nutrient uptake, translocation, and metabolic; vitamin manufacture; growth biostimulating; along with in the creation and synthesizing of amino-chelate fertilizers (**Sharma and Dietz 2006; Souri and Hatamian 2019**). Additionally, the presence of amino acids promotes stress tolerance by adjusting osmotic pressure and regulating intracellular pH. (**Rizhsky et al., 2004**).

Similar to how plant hormones control numerous biological and physiological processes, peptides, which are composed of chains of amino acids, play a significant role to withstand harsh conditions (Matsubayashi, 2014; Oh et al., 2018; Fletcher, 2020; Jeon et al., 2021; Kim et al., 2021).

Accumulation of osmolytes (protein and proline) supplies energy to plant cells, maintains cell vitality, protects cells from oxidative scorches, traps free radicals, and governs cell redox equilibrium (Shahzad, et al. 2021).

the buildup of proline as osmolytes, plant phenols serving as antioxidants, along with the increased activity of PPO and PRO enzymes in the leaves of pears, all of which have been outlined as indicators of Plants' ability to withstand abiotic stress was influenced by the irrigation schedule using 60% CWR in the present study. In accordance with research, using all inducers helped to lessen the impact of water scarcity, which had a negative consequence on growth and production.

The minerals make up (K and Fe) and antioxidant activities improved vegetation development in the present study and increased productivity, fruit weight, valium, length, diameter, and soluble solids by used amino acids. These findings are consistent with those made by **Colla et al. (2017)**,

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Lucini et al. (2015), Colla et al. (2014), and Etani et al. (2009), who found using amino acids as a tool to increased total soluble solids, K, and Mg content. Banana plants have significantly increased their protein, total phenolics, flavonoids, and antioxidant activity (Gurav and Jadhav, 2013), phenolic substance of red grape's (Parrado et al., 2007). On pomegranate fruits' increased TSS, TSS acid ratio, and total sugar content (Khattab et al., 2014). Through metal chelation and binding (Sharma and Dietz, 2009; Sytar et al., 2013), amino acid use on stressed plants improves potassium and proline concentrations. leaf Fe content increases and enhances net CO<sub>2</sub> assimilation, and photosynthates (i.e., soluble sugars) translocation via the phloem to potential sinks (Colla et al., 2017).

The spraying of putrescine on stressed plants in this investigation was interesting it not only improved because the productivity and vegetative traits but also increased anti-stress factors like proline, phenol, and antioxidant enzymes when compared to untreated. These results confirm the results of Verma and Mishra (2005), Li et al. (2015), and Ahmed et al. (2017), which the fact that external appliance of putrescine improved water status, chlorophyll, proline, amino acids, and soluble sugar concentration in plants exposed to water stress.

Putrescine reduced  $H_2O_2$ and lipid peroxidation also influencing the work of multiple antioxidant enzymes. Hassan et al. (2020) provided evidence that putrescine spraying resulting in obvious decreases in electrolyte leakage, Na+/K+ ratio, and ROS ROS markers. reduced levels. and malondialdehyde (MDA) levels. When leaves are senescent and flowers along with fruit ripening, putrescine and ethylene biosynthesis have adverse consequences on

compounds that serve as a forerunner to methionine (Pandey et al. 2000; Wi and Park 2002).

In this regard, putrescine action works by stabilizing membranes, scavenging free radicals, affecting nucleic acid, protein synthesis, RNase, protease, and different enzyme functions, and interacting with hormones, phytochrome, and ethylene biosynthesis (Bias and Ravishankar 2002; Kaur-Sawhney et al., 2003). It included Cell death, DNA and protein synthesis, cell differentiating and multiplying, and cell division (Igarashi and Kashiwagi 2000; Childs et al. 2003; Seiler and Raul 2005). This resulted in increased plants' height, leaf area, and productivity, along with improved adaptation to stress in several plant species studied (Todorova et al., 2015).

When higher plants are impacted to circumstances like osmotic, hydric, and oxidative stress, the osmolytes glycine betaine (GB), and pro, which aid in water conservation and safeguard proteins and biological membranes, play a significant influence (Ashraf and Foolad, 2007; Farooq et al., 2009a, 2009b). Spray of glycine has favorable outcomes and stimulatory effect on plant quality and growth (Noroozlo and colleagues, 2019). More effective in reducing water stress's consequences and increasing productivity is foliar of GB at the stage of vegetation (Iqbal et al. 2005). This K+ efflux was decreased by GB at a concentration of 5 mM. (Wei et al., 2017).

use of GB on leaf tissue, which effectively stabilizes the quaternary structure of proteins and enzymes, oxygen-evolving photosystem II (PSII), and component of the photosynthetic machinery like ribulose-

# **CONCLUSION:**

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1,5-biphosphate carboxylase/oxygenase (RuBisCO), can safeguard the intricate membrane structures (**Murata et al., 2007**). Capacity for withstand abiotic stress. In chaperone-induced protein disaggregation, glycine betaine discovered to be effective (**Hasanuzzaman, et al., 2019**).

External equipment appliance of GB was reported by **Banu et al. (2010)** to lower the amount of methyl-glyoxal in tobacco cells cultured in osmosis (as a toxic that severely inhibits cell growth in plants and is a product of glycolytic metabolism (**Borysiuk et al., 2018**).

As a result of glycine betaine's ability to scavenge ROS, balance cell redox status, act as osmo-protectants, stabilize cytosolic pH, proteins, enzymes, membranes and act as a potential supplier of carbon and nitrogen for plants both under stress conditions (Kumar and Khare, 2015).

Proline enhanced kiwifruit productivity, including fruit length, diameter, width, and weight (**Mehran et al. 2013**). Exogenous proline at 200 mg L-1 under stress given the highest N,P,K, proline, and soluble sugar content (**Sakr et al., 2012**). Proline functions as an osmolyte, a scavenger of reactive oxygen species (ROS), and can stabilize sub-cellular structures, thereby modulating the oxidative homeostasis of the cell. It also operates as a signal molecule and energy source that interacts with other metabolic pathways during times of stress (**Szabados and Savoure, 2010; Sharma et al., 2011).** 

The physiological effects of the materials employed during the test caused trees to encounter water lack, and as a result, they performed better than stressed trees (control) with the same level of deficiency.

Dry spell tension led to oxidative damage, which then resulted in stunted growth, decreased physiological function, and a

marked drop in production. Dry tension danger was minimized by applying amino acids to a pear tree that was stressed. Aside from this, stressed trees treated with amino acids demonstrated improved production, fruit attribute, development and accumulation of osmo-protectants, phenols, and antioxidant systems that also work as a scavenging tool to eliminate the extra ROS under draught stress.

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The observations confirm the study existence of 40% CWR during the fruitlets harvesting demonstrate and the to protection of pears from the negative repercussions of a lack of water by injecting amino acids on their leaves, which increases yield and improves fruit attribute. As a result, foliar treatment in cultivated localities is an option. This study confirms the beneficial appliance of amino acids in safeguarding pear trees under water scarcity stress.

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