



## MORPHOPHYSIOLOGICAL RESPONSE OF POTATO (*SOLANUM TUBEROSUM* L.) GENOTYPES TO PEG-INDUCED OSMOTIC STRESS IN VITRO

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### SUMMARY

This study evaluated the morphological and biochemical responses of potato genotypes to polyethylene glycol (PEG)-induced osmotic stress conditions. Using different concentrations of PEG solution assessed the adaptive response of genotypes in stress conditions. All potato genotypes exhibited reduced root and shoot length, a smaller leaf size, and decreased levels of photosynthetic pigments (chlorophyll a and b and carotenoids) under PEG treatments. However, an increase was evident in key indicators of oxidative and osmotic stress such as malondialdehyde (MDA), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), and proline levels. Among the tested cultivars, Telman showed the highest sensitivity to PEG-induced stress, while the cultivar Ugur maintained relatively stable productivity and exhibited adaptive traits. Despite considerable initial growth, the cultivar Vagif displayed a weaker adaptive response under prolonged stress conditions. A significant negative correlation was notable among the proline, MDA, and pigment contents, while a positive correlation was between the stem diameter and pigment levels. Among the traits, the highest coefficients of variation revealed genotypic differences in stress responses. The results enunciated that PEG-induced osmotic stress affects the physiological traits differently across the genotypes, providing valuable insights for identifying drought-tolerant potato cultivars. The findings provide a sound base for future breeding programs aimed at developing drought-resistant potato cultivars.

**Keywords:** Potato (*S. tuberosum* L.), polyethylene glycol, malondialdehyde, proline, hydrogen peroxide, photosynthetic pigments, morphological and biochemical traits

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**Key findings:** PEG-induced osmotic stress caused significant morphological and biochemical variations in potato (*S. tuberosum* L.) genotypes and revealed genotypic differences in drought response. The cultivar Ugur exhibited relatively stable adaptive traits, while the cultivar Telman was highly sensitive, and the genotype Vagif showed limited long-term tolerance. The results can provide a greater support in the selection of drought-tolerant potato genotypes.

## INTRODUCTION

Genetic resources provide a wide range of food and health products essential to human livelihoods, public health, and global economy (Hajiyeva *et al.*, 2024). However, population growth, climate change, ecological imbalances, and loss of biodiversity have raised the demand for food and agricultural crops (Guvendiyev *et al.*, 2025). Among the food crops, potato (*Solanum tuberosum* L.) is a globally important food crop, playing a vital role in food security worldwide (Birch *et al.*, 2012).

With its highest content of starch, proteins, vitamins, antioxidants, macro- and microelements, polyphenols, carotenoids, and tocopherols, potatoes are crucial in the human nutrition (Brown, 2005). However, the potato crop is drought-sensitive (Monneveux *et al.*, 2013), mainly due to its shallow root system that limits water uptake from deep soil layers, and being a cool-season crop, it is also sensitive to high temperatures (Gelmesa *et al.*, 2017). Abiotic stress factors, particularly drought and heat, severely affect its plant growth and development, which are the valid major threats to crop productivity and sustainable agriculture globally (Awasthi *et al.*, 2014).

Water deficit conditions lead to decreased biomass, disrupted metabolic activity, reduced water potential, loss of turgor, and damaged macromolecules (Aliyev *et al.*, 2014). It also impairs the plant's vital processes of photosynthesis, respiration, nutrient transport, and metabolism (Jaleel *et al.*, 2009). The plasma membrane is the first cellular structure affected by abiotic stress conditions; however, its stability has become a key indicator of drought and heat stress resistance. Membrane damage leads to leakage of cellular contents and, eventually, cell death (Salmanova *et al.*, 2002). In crop plants, the

malondialdehyde (MDA) accumulation widely serves as a marker for lipid peroxidation and membrane damage. Heat stress also leads to protein denaturation, membrane fluidity, enzyme inactivation, and reduced protein stability (Aliyev *et al.*, 2014).

Under drought conditions, one of the primary physiological responses in plants is osmotic stress. Water deficit reduces the cellular water potential, leading to dehydration at the cellular level. Coping with this condition makes plants accumulate osmolytes, such as proline, soluble sugars, and other compatible solutes, which help maintain cell turgor and stabilize cellular structures (Farooq *et al.*, 2009).

In combating osmotic stress, plants accumulate osmolytes, such as proline—a non-protein amino acid that plays multiple protective roles during drought. Proline stabilizes proteins and membranes, scavenges reactive oxygen species (ROS), buffers pH, and improves the water uptake in crop plants (Ibrahimova, 2024). Increased proline levels correlate positively with drought tolerance, with documentations also in grape (Mehri *et al.*, 2015), pear (Javadi *et al.*, 2004), and wheat (Gholipour and Ebadie, 2017) crops under *in vitro* conditions. The use of polyethylene glycol (PEG) is common in simulating drought conditions in laboratory experiments by developing osmotic stress and limiting water uptake (Ranjbarfordoei *et al.*, 2000).

This study used PEG to simulate drought stress and assess its effects on three Azerbaijani potato genotypes, viz., Telman, Ugur, and Vagif, grown *in vitro*. Selected for differing adaptation levels, the potato genotypes entailed evaluation at early developmental stages (seven days post-germination) to examine physiological responses and identify the potential drought-tolerant biomarkers. The related study

analyzed key morphological traits (root and shoot length, stem thickness, and leaf width) and physiological markers (photosynthetic pigments, MDA, H<sub>2</sub>O<sub>2</sub>, and proline) in the potato genotypes. PEG-induced osmotic stress offers a cost-effective, scalable method for controlled screening, as well as linking stress biomarkers to morphological responses. Overall, this approach provides a promising platform for early detection of drought resistance and supports strategies to enhance the potato resilience under existing climate variability.

## MATERIALS AND METHODS

### Experimental procedure

This study examined the physiological responses of three potato (*S. tuberosum* L.) genotypes, viz., Telman, Ugur, and Vagif, to different polyethylene glycol (PEG) concentrations. The selection of potato cultivars Telman, Ugur, and Vagif was according to their wide cultivation under local agroecological conditions and their economic importance in regional potato production. These cultivars also exhibit distinct agronomic traits and differential stress response patterns, which make them suitable as comparative models for analysis under PEG-induced drought stress conditions. Seedlings grown under sterile conditions used the MS medium (Murashige and Skoog, 1962), and after the development of true leaves, the transfer of seedlings continued to MS medium supplemented with PEG concentrations of 0.009, 0.012, and 0.015 M, calculated according to the formula of Michel and Kaufmann (1973). The control group remained on the MS medium without PEG. Plants' incubation was at 25 °C, with 70% humidity, and a 16-h light/8-h dark photoperiod.

### Traits measurement

The collection of samples commenced after seven days of PEG treatment. Seven days after exposure, morphological traits, including root length, leaf length and width, stem length, and

stem diameter, reached measuring in 10 seedlings per genotype and treatment. Determining photosynthetic pigments (chlorophyll *a*, chlorophyll *b*, and carotenoids) was successful spectrophotometrically from 80% acetone extracts at 663, 645, and 470 nm by following Arnon (1949). Malondialdehyde (MDA) content measurement, indicating lipid peroxidation, used the thiobarbituric acid (TBA) method, with the absorbance recorded at 532 and 600 nm, as described by Heath and Packer (1968). Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) content determination was according to Sinha *et al.* (2005), with absorbance measured at 390 nm and results expressed as  $\mu\text{mol g}^{-1}$  fresh weight. Free proline content quantification utilized the colorimetric method of Bates *et al.* (1973) with ninhydrin reagent, measuring absorbance of the toluene phase spectrophotometrically. Proline concentration calculation employed a calibration curve.

### Statistical analysis

The study performed the correlation analysis and principal component analysis (PCA) using IBM SPSS Statistics (version 25.0), while the biplot analysis engaged the PAST software (version 4.11) (Hammer *et al.*, 2001; Jolliffe, 2002).

## RESULTS AND DISCUSSION

### Morphological traits

In this study, three potato (*S. tuberosum* L.) genotypes (Telman, Ugur, and Vagif) sustained evaluation under different PEG concentrations (Table 1). Overall, the genotypes exhibited the highest root and shoot growth under the control condition; however, a considerable decline at the higher PEG levels manifested. For instance, the cultivar Telman's root length decreased from 13 cm (control) to 7.5 cm (0.015 M PEG), and the cultivar Ugur's leaf width declined from 0.7 cm to 0.3 cm. The stem diameter and length also showed similar reductions, confirming PEG's inhibitory effect on growth traits. Among the potato genotypes,

**Table 1.** Effect of PEG-induced drought stress on morphological traits of potato genotypes under in vitro conditions.

Genotypes	Treatments	Concentration (mol)	Characters				
			Root length (cm)	Leaf width (cm)	Leaf length (cm)	Stem width (cm)	Stem length (cm)
Telman	Control	0	11	0.5	1.2	0.5	9
		0.009	10	0.5	1.1	0.4	8
	PEG	0.012	8	0.4	0.9	0.3	7.5
		0.015	7.5	0.3	0.8	0.25	7
Ugur	Control	0	13	0.7	0.79	0.5	10
		0.009	11.5	0.6	0.7	0.3	9.2
	PEG	0.012	10.2	0.4	0.65	0.29	8.8
		0.015	9.7	0.3	0.58	0.2	7
Vagif	Control	0	12.2	0.6	1.5	0.6	11
		0.009	11	0.55	1.3	0.45	10.2
	PEG	0.012	10.1	0.48	1.1	0.4	9
		0.015	9.4	0.4	0.9	0.37	8.5
SE			0.14	0.01	0.02	0.01	0.11
SD			1.54	0.12	0.27	0.11	1.21
CV%			14.89	24.35	28.31	29.86	13.73
Genotype			25.980***	7.030**	101.077***	18.242***	35.138***
Drought			79.268***	148.823***	17.530***	105.099***	54.239***

SE-standart error; SD-standard deviation; CV-coefficient of variation; \*\*  $p < 0.01$  (highly significant); \*\*\*  $p < 0.001$  (very highly significant).

the cultivar Ugur maintained relatively higher root and stem values under PEG-induced stress conditions, suggesting greater drought tolerance. The study results were greatly analogous to previous studies of Kanat *et al.* (2024), who reported that PEG-6000 reduced the morphological traits in most potato cultivars, while tolerant genotypes maintained their root and internode lengths.

The research further revealed potato genotypes and PEG treatments significantly ( $p < 0.001$ ) affect all morphological parameters. However, the leaf length was the most sensitive trait to genotypic variations, while the leaf width and stem diameter were more stress-responsive. Root and stem length showed the lowest variability, indicating greater stability under drought stress conditions. Overall, the relevant results confirmed that both genetic and environmental factors shaped the morphological adaptation under osmotic stress. Studies of Tican *et al.* (2021) and Barra *et al.* (2013) also classified potato genotypes based on PEG-induced growth reduction.

### Photosynthetic pigments

In this study, the PEG-induced osmotic stress significantly affects the photosynthetic pigments in the different potato (*S. tuberosum* L.) genotypes. Under the controlled condition, the cultivar Telman showed the highest levels of photosynthetic pigments (chlorophyll a: 30.6, chlorophyll b: 12.6, total chlorophyll: 43.2, and carotenoids: 7.5 mg kg<sup>-1</sup>). With increasing PEG, total chlorophyll decreased to 24.7–19.9 mg kg<sup>-1</sup> and carotenoids to 3.0 mg kg<sup>-1</sup>, indicating moderate tolerance. The cultivar Ugur, despite high control levels (total chlorophyll: 45.1, carotenoids: 6.9), these values declined sharply under PEG-stress conditions to 14.1 and 2.6 mg kg<sup>-1</sup>, respectively, showing the highest sensitivity. The cultivar Vagif had the maximum chlorophyll (45.9 mg kg<sup>-1</sup>) in the control, but it lowered to 9.3 mg, with its carotenoids to 1.7 mg kg<sup>-1</sup>, suggesting very low tolerance under PEG-stress conditions (Table 2). Drought conditions commonly reduce the chlorophyll and carotenoid contents by inhibiting their

**Table 2.** Effect of PEG-induced drought stress on photosynthetic pigment content in potato genotypes under in vitro conditions.

Genotypes	Treatments	Concentration	Characters			
			Chlorophyll a (mg kg <sup>-1</sup> )	Chlorophyll b (mg kg <sup>-1</sup> )	Chlorophyll a + b (mg kg <sup>-1</sup> )	Carotenoid (mg kg <sup>-1</sup> )
Telman	Control	0	30.6	12.6	43.2	7.5
		0.009	18.3	6.4	24.7	4.6
	PEG	0.012	16.4	6.4	22.8	3.3
		0.015	13.9	6	19.9	3
Ugur	Control	0	31.7	13.4	45.1	6.9
		0.009	11.9	5.2	17.1	2.8
	PEG	0.012	11.6	4.7	16.4	2.6
		0.015	10.1	4	14.1	2.6
Vagif	Control	0	30.6	15.3	45.9	7.2
		0.009	11.2	4.8	15.9	2.6
	PEG	0.012	7.3	3	10.3	1.8
		0.015	6.6	2.7	9.3	1.7
SE			0.81	0.37	1.18	0.19
SD			8.87	4.09	12.90	2.06
CV%			53.14	58.01	54.40	52.81
Genotype			4.682*	1.304 (ns)	3.367*	4.320*
Drought			329.885***	420.693***	382.698***	374.639***

SE-standart error; SD-standard deviation; CV-coefficient of variation; (ns) not statistically significant ( $p > 0.05$ ); \*  $p < 0.05$  (significant); \*\*\*  $p < 0.001$  (very highly significant).

biosynthesis, while chlorophyll degradation disrupts energy transfer and damages the photosynthetic apparatus (Gill and Tuteja, 2010; Ragab *et al.*, 2015).

The analysis revealed significant ( $p < 0.05$ ) genotype effects on chlorophyll a, total chlorophyll, and carotenoids but not for chlorophyll b. The highest coefficient of variation ( $> 50\%$ ) confirmed the considerable environmental and genetic influences, suggesting that chlorophyll and carotenoids were reliable biochemical markers of drought response. These results were consistent with the past findings of Zhang *et al.* (2014), where chlorophyll content was higher in irrigated than in drought-stressed plants.

### Proline content

The proline accumulation evaluated in potato (*S. tuberosum* L.) cultivars in this study, under PEG-induced stress conditions, varied (Table 3). In the cultivar Telman, the proline content increased from 9  $\mu\text{M g}^{-1}$  (control) to 25  $\mu\text{M g}^{-1}$  (at 0.009 PEG), peaking at 28  $\mu\text{M g}^{-1}$  at 0.012 PEG. However, it slightly dropped to 27  $\mu\text{M g}^{-1}$

at 0.015 PEG. This consistent accumulation suggests a considerable stress adaptation. Proline accumulation is a common response to abiotic stress factors such as drought and temperature fluctuations (Ibrahimova *et al.*, 2020). It helps maintain the cell turgor, stabilize membranes and proteins, and neutralize free radicals. Key enzymes in proline biosynthesis include P5CS and P5CR (Delauney and Verma, 1993).

The cultivar Ugur showed lower proline accumulation, ranging from 5  $\mu\text{M g}^{-1}$  (control) to 18, 19, and 18  $\mu\text{M g}^{-1}$  under different PEG concentrations, respectively. In response to stress conditions, its adaptation was less robust than the cultivar Telman. The cultivar Vagif initially accumulated the highest proline content (from 3 to 25  $\mu\text{M g}^{-1}$ ) but dropped to 15  $\mu\text{M g}^{-1}$  at the topmost PEG concentration, revealing the weakest response under severe stress conditions. These potato genotype-dependent variations for proline content align with previous findings, confirming PEG's considerable osmotic effect (Dorneles *et al.*, 2021). The analysis further exhibited the significant ( $p < 0.001$ ) genotype-by-

**Table 3.** Effect of PEG-induced drought stress on proline, MDA, and H<sub>2</sub>O<sub>2</sub> contents in potato genotypes under in vitro conditions.

Genotypes	Treatments	Concentration	Characters		
			Proline ( $\mu\text{M g}^{-1}$ )	MDA ( $\text{nM g}^{-1}$ )	H <sub>2</sub> O <sub>2</sub> ( $\mu\text{M g}^{-1}$ )
Telman	Control	0	9	2	2.5
		0.009	25	5	3.5
	PEG	0.012	28	6	4
		0.015	27	6.5	4.5
Ugur	Control	0	5	1.8	1
		0.009	18	5	4
	PEG	0.012	19	4	3
		0.015	18	6	1.5
Vagif	Control	0	3	2.1	0.5
		0.009	20	3.5	4.5
	PEG	0.012	25	3	3.2
		0.015	15	5	2.5
SE			0.73	0.15	0.12
SD			8.04	1.6	1.28
CV%			45.35	38.25	44.58
Genotype			11.747***	10.269***	12.688***
Drought			139.909***	119.089***	55.924***

SE-standart error; SD-standard deviation; CV-coefficient of variation; \*\*\* p < 0.001 (very highly significant).

environment interaction effects. High SD (8.04) and CV% (45.35) further reflected the trait's sensitivity. Thus, proline emerged as a reliable biochemical marker of drought adaptation. Among the potato genotypes, the cultivar Telman demonstrated better accumulation and the highest tolerance, while the cultivar Vagif showed limited resilience under severe stress conditions (Table 3). Previous studies also confirmed proline as a widely recognized indicator of drought tolerance in potatoes (Amini et al., 2017).

### MDA content

Malondialdehyde (MDA) use is common in assessing lipid peroxidation and membrane damage under abiotic stress conditions. In this study, the MDA levels increased with PEG-induced stress conditions in potato (*S. tuberosum* L.) genotypes (Table 3). In the cultivar Telman, the MDA rose steadily from 2 nM g<sup>-1</sup> (control) to 6.5 nM g<sup>-1</sup> (0.015 PEG), indicating severe oxidative damage. However, the cultivar Ugur showed fluctuations, starting at 1.8 nM g<sup>-1</sup> (control), rising to 5 nM g<sup>-1</sup> (0.009 PEG), decreasing to 4 nM g<sup>-1</sup> (0.012 PEG), and increasing again to 6 nM g<sup>-1</sup> (0.015

PEG). This suggests temporary adaptation at moderate stress conditions but limited protection at higher levels. Potato cultivar Vagif was more stable, increasing from 2.1 nM g<sup>-1</sup> (control) to 5 nM g<sup>-1</sup> (0.015 PEG), reflecting better membrane stability and oxidative stress tolerance. Drought induces oxidative damage through the accumulation of reactive oxygen species (ROS), which attack lipids, proteins, and nucleic acids, destabilizing membranes and causing cytosolic leakage (Munns and James, 2003; Jiang and Hung, 2001).

The analysis also confirmed significant (p < 0.001) genotypic effects on MDA levels under drought stress conditions. Moderate variability was noticeable, with SD = 1.60 and CV% = 38.25. These results revealed that MDA is a reliable biochemical marker for oxidative damage in drought-stressed plants.

### Hydrogen peroxide content (H<sub>2</sub>O<sub>2</sub>)

In this study, measuring the H<sub>2</sub>O<sub>2</sub> content ensued in three potato (*S. tuberosum* L.) genotypes under PEG-induced drought stress (Table 3). In the cultivar Telman, H<sub>2</sub>O<sub>2</sub> increased steadily from 2.5  $\mu\text{M g}^{-1}$  (control) to

4.5  $\mu\text{M g}^{-1}$  (0.015 PEG), revealing a considerable but stress-prone oxidative response. Concerning the cultivar Ugur, the  $\text{H}_2\text{O}_2$  level rose from 1  $\mu\text{M g}^{-1}$  (control) to 4  $\mu\text{M g}^{-1}$  (0.009 PEG) and later decreased to 3 and 1.5  $\mu\text{M g}^{-1}$  at higher PEG concentrations, respectively. It suggests early oxidative signaling followed by effective regulation and moderate stress adaptation. The cultivar Vagif showed a sharp initial increase, from 0.5  $\mu\text{M g}^{-1}$  (control) to 4.5  $\mu\text{M g}^{-1}$  (0.009 PEG), and then decreased to 3.2 and 2.5  $\mu\text{M g}^{-1}$ , indicating activation of adaptive mechanisms to reduce the oxidative stress. Hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) acts both as a damaging ROS at the highest levels and a signaling molecule at optimal concentrations, regulating plant growth, metabolism, redox balance, and stress tolerance (Abd-Elhady *et al.*, 2021).

The analysis confirmed significant ( $p < 0.001$ ) for the genotypes and PEG treatments' effect on  $\text{H}_2\text{O}_2$  content. High sensitivity to genetic and environmental factors manifested in  $\text{SD} = 1.28$  and  $\text{CV}\% = 44.58$ . In conclusion, the cultivar Telman revealed a vigorous and considerable oxidative response to water stress conditions, as evidenced by consistently higher  $\text{H}_2\text{O}_2$  levels. The cultivar Ugur showed an early rise in  $\text{H}_2\text{O}_2$  level; however, later it moderates them, indicating a mild level of adaptation. The cultivar Vagif enunciated an initial spike in  $\text{H}_2\text{O}_2$  level, followed by a sharp decline, signifying activation of stress mitigation mechanisms. These results confirmed that oxidative balance and response mechanisms to water stress differed among the potato cultivars. Thus, the analysis of  $\text{H}_2\text{O}_2$  content serves as an important indicator for assessing the drought resistance in potato cultivars.

### Correlation analysis

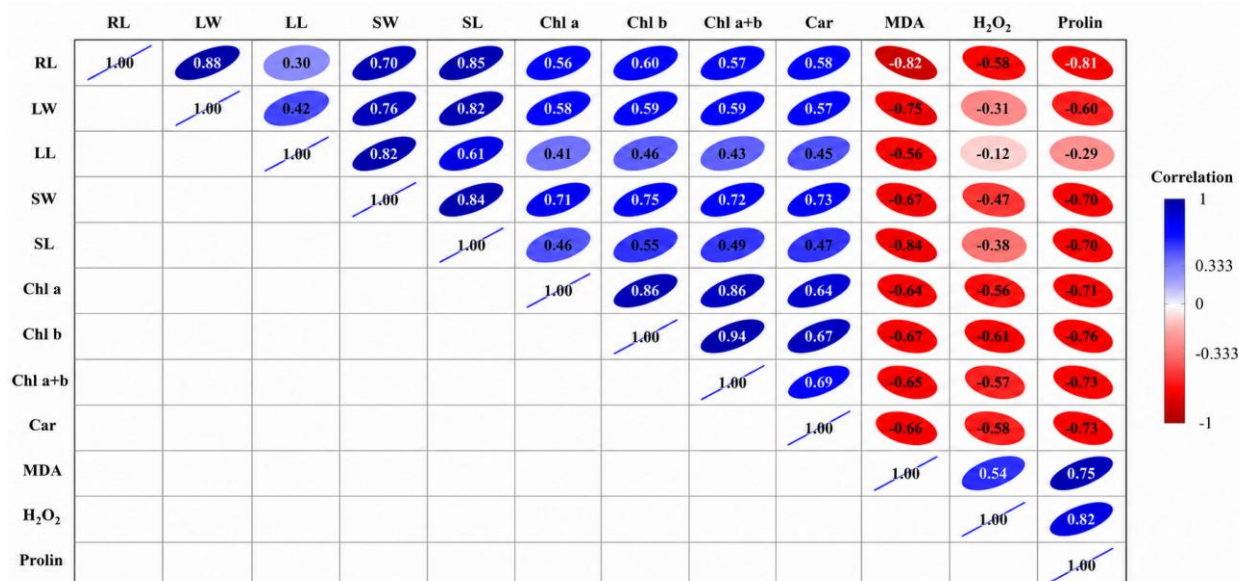
Using Pearson's correlation coefficient ( $r$ ), the study sought to determine the strength of a linear relationship between the traits (Pearson, 1900). Considerable positive correlations were evident among the potato morphological features, such as root length and stem length ( $r = 0.85$ ), and moderate correlation between

leaf width and length ( $r = 0.42$ ), indicating coordinated leaf development (Figure 1). This study assessed the effects of PEG on the morphological and physiological characteristics of three potato (*S. tuberosum* L.) genotypes through analyzing traits relationships (Norman *et al.*, 2011).

Photosynthetic pigments appeared as highly positively correlated, with values of chlorophyll a and chlorophyll b at  $r = 0.95$  and chlorophyll a and total chlorophyll at  $r = 0.99$ , suggesting a synergistic role in photosynthesis. Carotenoids also have a positive correlation with chlorophyll ( $r = 0.87$ ), supporting their vital role in light absorption and oxidative protection. Morphological traits expressed a moderately positive association with photosynthetic pigment (chlorophyll a) and root length ( $r = 0.56$ ), indicating that structural growth parallels photosynthetic capacity. Various stresses reduced the leaf area, stem area, and plant height, thereby decreasing photosynthetic activity (Allen and Scott, 1980).

Stress markers, such as MDA,  $\text{H}_2\text{O}_2$ , and proline content, showed negative correlations with morphological (root length) and photosynthetic traits like MDA ( $r = -0.82$ ), demonstrating that oxidative stress impairs plant growth. Positive correlations among stress markers MDA and  $\text{H}_2\text{O}_2$  ( $r = 0.54$ ) and  $\text{H}_2\text{O}_2$  and proline ( $r = 0.82$ ) revealed their simultaneous enhancement in stress conditions. Although root length has a linkage to drought tolerance, the correlations were weak and insufficient to explain the seasonal variations in potato drought tolerance (Hill *et al.*, 2021).

The results further indicate a decreased morphological development in stress appeared to be closely associated with reduced photosynthetic pigments. Elevated proline may aid osmotic regulation; however, persistently higher levels reflect the physiological limits under chronic stress conditions. Overall, morphological and physiological traits disclosed an interconnection, and monitoring both is essential to improve genotype resilience in stressful conditions.



**Figure 1.** Pearson's correlation analysis of potato genotypes' response to PEG-induced drought stress under in vitro conditions.

### Principal component analysis

In the principal component analysis (PCA), PC1 explained 68.99% of the total variance, with the highest positive loadings for root length (0.833), leaf width (0.798), leaf length (0.579), stem thickness (0.909), and stem length (0.804). These indicate their crucial roles in the vegetative development and stress response of the potato (*S. tuberosum* L.) genotypes (Table 4). Photosynthetic pigments, such as chlorophyll a (0.878), chlorophyll b (0.909), total chlorophyll (0.891), and carotenoids (0.888), also loaded considerably on PC1, emphasizing their involvement in plant adaptation and antioxidant activity. PC2 showed only stem length (0.547) with a significant loading, suggesting morphological variation, with PC3 mainly defined by leaf length (0.623). The results signified the selected components reflect the key physiological and morphological traits related to growth and stress tolerance. Kanat *et al.* (2024) also reported similar trends in PCA-differentiated control and PEG-6000-treated potato cultivars. The conduct of principal component analysis distinguishes the underlying data structures, integrating both

quantitative and qualitative variables (Jolliffe, 2002).

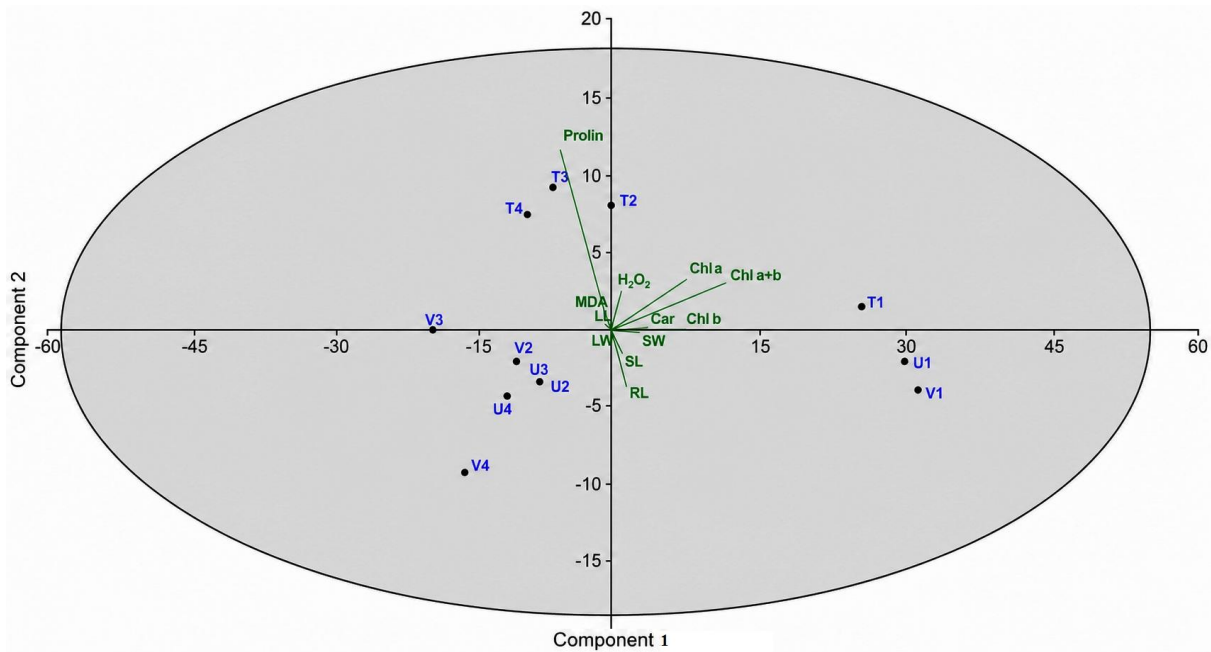
In the concerned study, the potato morphological parameters, such as plant height, root length, leaf number, and internode number, incurred significant reductions under osmotic stress. These results align with previous studies and support the identification of genotypes with enhanced phenotypic and physiological traits for breeding and selection programs (Tajaragh *et al.* 2022). The results will help the researchers in identifying genotypes with pronounced phenotypic and physiological traits and provide a basis for pinpointing priority targets in future breeding and selection programs.

### Biplot analysis

The biplot analysis of the potato (*S. tuberosum* L.) genotypes and quantitative traits based on the first two principal components (PC1 and PC2) obtained through morphological and physiological data appears in Figure 2. Morphological traits, such as root length (RL), leaf width (LW), leaf length (LL), stem width (SW), and stem length (SL), cluster positively on PC1, reflecting overall growth.

**Table 4.** Principal component analysis (PCA): Eigenvalues and variance explained by the major axes for morphological and physiological traits in potato genotypes.

Traits	PC1	PC2	PC3
Root length	0.833	0.292	-0.402
Leaf width	0.798	0.386	-0.101
Leaf length	0.579	0.373	0.623
Stem width	0.909	0.265	0.242
Stem length	0.804	0.547	-0.083
Chlorophyll a	0.878	-0.415	0.179
Chlorophyll b	0.909	-0.359	0.15
Chlorophyll a + b	0.891	-0.4	0.169
Carotenoid	0.888	-0.397	0.181
MDA	-0.882	-0.279	0.066
H <sub>2</sub> O <sub>2</sub>	-0.656	0.316	0.489
Proline	-0.868	0.099	0.373
Eigenvalue	8.279	1.55	1.124
Variance (%)	68.994	12.919	9.367
Σ variance (%)	68.994	81.913	91.28



**Figure 2.** Analysis of the biplot of evaluated traits (T1–T4, V1–V4), and U1–U4 represent potato genotypes. Treatments: 1 = control, 2 = PEG 0.009, 3 = PEG 0.012, and 4 = PEG 0.015.

Photosynthetic pigments like chlorophyll a (Chl a), chlorophyll b (Chl b), total chlorophyll (Chl a+b), and carotenoids (Car) also grouped on PC1, indicating synergistic activity. Stress indicators malondialdehyde (MDA), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), and proline aligned along PC2, separated from growth traits, showing stress acts independently. Biplot PCA is a

powerful tool for visualizing complex multivariate datasets, displaying quantitative variables and samples on a two-dimensional plane to clarify data structure (Johnson and Wichern, 2014). By reducing dimensionality through principal components, PCA highlighted the variables contributing most to variance.

Vectors indicate variable contributions, while point proximity shows sample similarity.

Genotypes 'T1' and 'V1' position positively on PC1, representing healthy, photosynthetically active plants, whereas 'T3,' 'T4,' 'V3,' and 'V4' cluster near stress indicators on PC2, reflecting stress and reduced growth. Overall, PCA biplot effectively illustrates genotypic differences in growth, photosynthetic, and stress traits, providing insights into adaptation and developmental potential under varying environmental conditions. The PCA proved suitable for continuous numerical variables; however, it was unsuitable for categorical data, which can distort results (Wold et al., 1987).

## CONCLUSIONS

Increasing PEG concentrations significantly reduced root and shoot growth, leaf and stem size, and photosynthetic pigments in potato (*S. tuberosum* L.) genotypes (Telman, Ugur, and Vagif) under in vitro conditions to evaluate the drought tolerance. Stress indicators, including malondialdehyde (MDA), hydrogen peroxide, and proline, increased notably in stress conditions. The cultivar Telman was the most sensitive genotype; Ugur showed better tolerance; and the cultivar Vagif declined sharply despite potential growth under controlled conditions. The results highlighted genotype-specific responses to drought and the importance of genetic variability in breeding drought-tolerant potatoes.

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