scientific reports

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Multilevel evaluation of *Prunus cerasifera* Ehrh. rootstock candidates on nutritional and biochemical networks in 'Hacıhaliloğlu' apricot

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Understanding rootstock-mediated physiological and biochemical traits is critical for selecting stress-resilient apricot genotypes. However, integrated assessments of mineral nutrition, oxidative balance, hormone profiles, and carbohydrate metabolism in grafted systems remain limited. We evaluated mineral uptake, oxidative stress responses, antioxidant enzyme activity, hormone content, and sugar profiles in ungrafted apricot rootstock candidates and in 'Hacihaliloglu' cultivar grafted onto 13 *Prunus cerasifera* genotypes. Genotypic variation was significant across all parameters. Particularly, 63B69, 66B14 and 63B16 and its grafted combination (H/63B69, H/63B14 and H/63B16) consistently exhibited superior nutrient accumulation, lower oxidative damage (H_2O_2, MDA), higher antioxidant enzyme activity (CAT, SOD), and favorable hormonal and sugar profiles. Multivariate and correlation analyses revealed that grafting reorganized the physiological network, enhancing integration between mineral nutrition, antioxidants, and hormones. Notably, H/63B69, H/63B14 and H/63B16 formed a distinct cluster with high values for beneficial traits, suggesting efficient nutrient uptake and stress mitigation capacity. These findings indicated the critical role of genotype-specific rootstock selection in enhancing the physiological, biochemical, and nutritional performance of apricot trees, particularly under adverse environmental conditions such as drought and heat stress. The consistent superiority of certain genotypes (e.g., 63B69, 66B14, 63B16 and its grafted combination H/63B69, H/63B14 and H/63B16) across multiple functional traits, including nutrient uptake efficiency, antioxidant defense capacity, osmoprotectant accumulation, and hormonal balance, demonstrates that rootstock choice is not merely a supporting factor, but a decisive determinant of overall plant resilience and adaptability. Therefore, strategic utilization of well-characterized rootstock genotypes tailored to specific stress profiles represents a promising approach for improving orchard sustainability and productivity in the face of climate-induced challenges.

Keywords Antioxidant enzymes, Hormonal profiling, Graft compatibility, Carbohydrate metabolism, Genotypic variation

Apricot (*Prunus armeniaca* L.) cultivation in Türkiye relies heavily on grafting, where scions are united with genetically distinct rootstocks to optimize performance under diverse environmental conditions. This technique, despite its ancient origins[1], continues to play a crucial role in fruit tree propagation as it allows different characteristics, such as fruit quality and stress tolerance, to be combined within a single plant. In the grafted system, the interaction between scion and rootstock genotypes ultimately shapes the physiological and biochemical profile of the composite organism[2]. The rootstock is known to modulate the uptake and

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translocation of water, minerals, and hormones to the scion, thereby playing a decisive role in its growth and productivity under abiotic stress[3,4]. The selection of suitable rootstock genotypes is therefore a key factor in improving resource-use efficiency, particularly in arid regions. On the other hand, physiological responses to rootstock, scion interactions manifest at multiple levels of organization, from hydraulic regulation and mineral transport to sugar metabolism and hormone signaling[5–7]. These interactions can modify stomatal behavior, nutrient assimilation, and carbohydrate distribution, all of which influence leaf development and photosynthetic efficiency. Although the precise mechanisms underlying these modifications remain elusive, studies have shown that differences in root hydraulic conductivity, xylem architecture, and phloem connectivity may be responsible for variations in scion vigor and fruit yield[8,9]. For example, invigorating rootstocks have been associated with higher stem water potential and improved gas exchange, while dwarfing rootstocks may restrict growth through reduced solute flow and hormone production[10,11].

A comprehensive approach that integrates anatomical, physiological, and biochemical metrics is essential for understanding graft compatibility beyond visual or morphological assessments. Leaf nutrient profiles, sugar accumulation patterns, and endogenous hormone concentrations serve as sensitive indicators of rootstock influence and overall scion health[2]. Furthermore, genotype × environment interactions complicate compatibility assessments, as physiological responses may vary depending on environmental stressors, soil conditions, and management practices[3,12]. These complexities are especially relevant in arid and semi-arid growing regions, where rootstock-mediated differences in water and nutrient use efficiency can significantly impact productivity and long-term orchard sustainability. Yet, there is a lack of integrative studies that assess compatibility through physiological and biochemical parameters in controlled conditions, especially using local genetic resources. As such, identifying compatible and physiologically synergistic rootstocks is not only a matter of graft union success, but also a strategic response to increasing climatic uncertainty and resource limitations in Turkish apricot production.

Despite substantial advances in the physiological and molecular understanding of rootstock–scion interactions, most studies have focused on clonal or commercial rootstocks and neglect the genetic diversity found in seedling-origin *Prunus cerasifera* populations[13]. In particular, there is limited integrative research combining anatomical, physiological, and biochemical indicators to assess graft compatibility in apricot. Similar integrative approaches have recently been applied in wild olive, where morphological, anatomical and histological analyses were combined to diagnose graft compatibility and to support rootstock selection under Mediterranean conditions[14]. This is a critical gap, as effective compatibility between scion and rootstock ensures successful vascular reconnection and efficient resource allocation[2]. The biochemical composition of scion leaves, including macro- and micronutrient content, hormone levels, and sugar profiles, can serve as a proxy for compatibility, yet such data remain scarce for regionally adapted genotypes. Moreover, the responses of different combinations may be context-dependent, influenced by both genotype and environment[3,12]. In Türkiye, the Hacihaliloglu apricot cultivar holds exceptional economic and cultural importance, particularly for dried fruit production Korkmaz et al.[15]. Yet, the physiological and biochemical interactions between Hacihaliloglu and locally selected *P. cerasifera* rootstocks remain poorly understood. Previous work on these genotypes has been limited to phenotypic screening, with little emphasis on nutrient dynamics or leaf biochemistry under controlled environmental conditions. From this perspective, we aimed (I) to assess the compatibility and physiological behavior of Hacihaliloglu when grafted onto 13 genetically distinct *P. cerasifera* rootstock candidates, (II) to identify rootstocks that offer both high compatibility and improved physiological performance through a comprehensive analysis involving mineral content, sugar levels, hormonal balance, and anatomical features of the graft union, (III) to support sustainable apricot production in semi-arid ecosystems.

Material and methods

Plant material selection

The study began with the identification of 155 *Prunus cerasifera* genotypes in the Middle Euphrates region of Türkiye, selected based on their general morphological traits. Following more detailed evaluations of plant growth and morphological characteristics, the number of genotypes was narrowed down to 79. Hardwood cuttings were collected from these genotypes in February 2021[13]. Among them, the 40 genotypes that exhibited the highest rooting performance were selected for further evaluation regarding their potential semi-dwarf and dwarf growth habits. Ultimately, 13 genotypes were identified as promising rootstock candidates. For comparative purposes, Myrobolan 29C was used as the standard rootstock[13].

Experimental design and grafting

The experiment followed a randomized design with 10 replicates per genotype, each replicate consisting of 10 plants (n = 100 plants per genotype). Prior to grafting, the buds on the stems of selected plants were removed. In early October 2021, the 'Hacihaliloglu' apricot cultivar [*P. armeniaca* (L.)] was T-budded onto 14 plum rootstock candidates: "63B11, 63B14, 63B16, 63B33, 63B43, 63B61, 63B62, 63B63, 63B69, 63B72, 63B76, 63B78, 63H66," along with Myrobolan 29C as a control. In addition, ungrafted rootstock plants were maintained separately as controls. This two-stage approach was designed to allow for a more robust comparison by assessing rootstock traits both independently and in grafted combinations.

Grafting process and growing conditions

All rooted one-year-old plants were potted in 21-L containers and placed in a controlled greenhouse environment at Harran University's Agricultural Research Station (Şanlıurfa), Türkiye.Grafting was carried out under greenhouse conditions using one-year-old rootstocks. After grafting, the plants were irrigated every three days, depending on soil moisture levels, until the end of the growing season. Environmental conditions were regulated to ensure optimal growth for all grafted combinations.

Sample collection for analysis

In our previous study[13,15], we conducted a comprehensive evaluation of *Prunus cerasifera* rootstock candidates grafted with the 'Hacihaliloglu' apricot cultivar, focusing on growth vigor, grafting success, and compatibility characteristics. The genotypes exhibited notable differences across all parameters. In terms of growth vigor, the rootstocks were ranked from strongest to weakest as follows: Myrobolan 29C, 63B76, 63H66, 63B72, 63B43, 63B78, 63B11, 63B14, 63B33, 63B69, 63B16, 63B62, 63B63, and 63B61. Regarding grafting success with the 'Hacihaliloglu' scion, genotype 63B11 showed the highest success rate, followed by Myrobolan 29C, 63B76, 63H78, 63B62, 63B16, 63B33, 63B63, 63H66, 63B69, 63B14, 63B61, 63B43, and 63B72. Compatibility based on the starch score placed 63B69 at the top, followed by 63B14, 63B16, 63H66, 63B78, 63B11, Myrobolan 29C, 63B61, 63B43, 63B33, 63B62, 63B63, 63B76, and finally 63B72. A similar trend was observed in histological assessments, with the highest scores recorded for 63B69, 63B14, 63B16, and 63H66, while the lowest belonged to 63B76 and 63B72. Leaf samples were collected from each individual plant in mid-July 2023. In the present study, leaf samples from the grafted combinations were collected and analyzed for biochemical properties, hormone levels, nutrient element concentrations, and sugar content.

Mineral analysis

To determine the mineral nutrient composition of the leaf samples, a series of well-established laboratory procedures were followed. Initially, leaves were carefully washed to remove surface contaminants and then air-dried to eliminate residual moisture. The dried samples were further oven-dried to a constant weight, ensuring consistency across all measurements. Once completely desiccated, the leaves were ground into a fine powder using a mortar and pestle, allowing for accurate and uniform analysis. Leaf nitrogen content was determined using the classical Kjeldahl method. This involved digesting the powdered samples in concentrated sulfuric acid (H_2SO_4), followed by distillation and titration to quantify the nitrogen levels. The K, Ca, Mg, Mn, Fe and Zn concentrations were measured using atomic absorption spectrophotometry. For this analysis, powdered leaf samples underwent acid digestion using a mixture of nitric acid (HNO_3) and hydrochloric acid (HCl). The digestion process was carried out by adding a known volume of the acid mixture to the samples and heating them on a hot plate or digestion block. This step effectively broke down the organic matrix and released the target elements into solution. After digestion, the mixtures were cooled and diluted with deionized water to a suitable concentration before analysis. The prepared solutions were then analyzed with an atomic absorption spectrophotometer equipped with element-specific hollow cathode lamps. Phosphorus (P) levels were assessed using spectrophotometry with a Shimadzu UV-1700 spectrophotometer. The absorbance of this complex was measured at a defined wavelength, and phosphorus content was calculated based on a calibration curve generated from standard solutions. All nutrient analyses were performed according to the protocols described by Kacar and Inal[16], which are widely recognized for their reliability in plant tissue analysis.

Determination of proline, malondialdehyde (MDA), H_2O_2 content, and antioxidant enzyme activities

Proline content in leaf samples was determined following the method described by Bates et al.[17]. For each measurement, 0.5 g of fresh leaf tissue was homogenized in 10 mL of 3% sulfosalicylic acid. The homogenate was centrifuged at 10,000 × g for 10 min. From the supernatant, 2 mL was mixed with 2 mL of acid-ninhydrin solution and 2 mL of glacial acetic acid. This mixture was incubated at 90 °C for one hour, then extracted with 5 mL of toluene. The chromophore-containing toluene phase was collected, and absorbance was read at 520 nm using a Shimadzu UV-1700 spectrophotometer. Proline concentrations were calculated against a standard curve prepared with L-proline and expressed as μg g^{-1} fresh weight. Malondialdehyde (MDA) levels, indicative of lipid peroxidation, were assessed using a modified version of the protocol by Velikova et al.[18]. A 0.5 g portion of frozen leaf tissue was homogenized in 0.1% trichloroacetic acid (TCA) and centrifuged at 10,000 × g for 5 min. The supernatant was mixed with thiobarbituric acid (TBA) in 20% TCA and incubated at 95 °C for 45 min. After rapid cooling, samples were centrifuged again at 10,000 × g for 15 min. Absorbance readings were taken at 532 nm and corrected at 600 nm. MDA content was calculated using an extinction coefficient and expressed as nmol g^{-1} fresh weight. Hydrogen peroxide (H_2O_2) concentrations were measured following the method of Kaya and Köse[19]. Leaf tissues were extracted in cold acetone containing 25 mM H_2SO_4. After centrifugation, the supernatant was reacted with the eFOX reagent. Following incubation, absorbance was measured at 550 nm and 800 nm. The H_2O_2 content was quantified using a standard curve based on known H_2O_2 concentrations. The activities of key antioxidant enzymes, superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD), were analyzed using protocols adapted from Beauchamp and Fridovich[20], Aebi[21], and Cvikrová et al.[22], respectively. Leaf tissue (0.5 g) was homogenized in 3 mL of 50 mM phosphate buffer (pH 7.0), and the homogenate was centrifuged at 15,000 × g for 15 min at 4 °C. The supernatant was immediately stored at – 80 °C until analysis. For enzyme extraction, frozen tissue was ground in phosphate buffer (pH 7.8) containing 0.5% polyvinylpyrrolidone (PVP), 1 mM phenylmethylsulfonyl fluoride (PMSF), and 1 mM EDTA. Enzyme activities were quantified spectrophotometrically: SOD at 560 nm, POD at 470 nm, and CAT at 240 nm. One unit (U) of SOD activity was defined as the amount of enzyme required to inhibit photoreduction of nitroblue tetrazolium (NBT) by 50%. One unit of CAT or POD activity was defined as the enzyme amount causing an absorbance change of 0.01 per minute.

Hormone analysis

The extraction and quantification of key plant hormones, cytokinin, indole-3-acetic acid (IAA), gibberellic acid (GA_3), and abscisic acid (ABA), were conducted with slight modifications to the methodology outlined by Bolat et al.[23]. To begin the extraction, 1 g of fresh leaf tissue was homogenized in 80% methanol (v/v) at –40 °C for 10 min. The resulting homogenate was centrifuged, and the supernatant was filtered to remove debris. The

filtrate was then evaporated at 35 °C to concentrate the hormone content. Dried residues were dissolved in 0.1 M potassium dihydrogen phosphate (KH_2PO_4) buffer (pH 8.0) to prepare for chromatographic separation. For purification, the samples were passed through Sep-Pak C18 cartridges (Waters Corp., Milford, MA, USA), which effectively removed interfering substances and isolated the hormones of interest. Quantification was carried out using high-performance liquid chromatography (HPLC) equipped with a UV detector (Agilent Technologies, Santa Clara, CA, USA). Absorbance was measured at 265 nm to detect the target hormone compounds. This methodology, with the described modifications, has been successfully applied in previous studies, including those focusing on stress tolerance enhancement in plant rootstocks[23].

Sugar analysis

Sugar analyses were conducted on leaves to determine the concentrations of glucose, fructose, and sucrose, following the optimized HPLC–RI method described by Filip et al.[24]. Standards for glucose, fructose, and sucrose were obtained from Aldrich (Milwaukee, USA). Analytical grade water (18.2 MΩ cm) was prepared using a Milli-Q Ultrapure water purification system (Millipore, USA), and ethanol was purchased from Chimopar (Bucharest, Romania).

Statistical analysis

Growth-related data from each graft combination were subjected to analysis of variance (ANOVA) using SPSS software version 21.0 (SPSS Inc., Chicago, IL, USA). To determine differences among means, Duncan's multiple range test was applied, with significance considered at $p \leq 0.001$. Prior to ANOVA, all datasets were tested for normality using the Shapiro–Wilk test and for homogeneity of variances using Levene's test. To further explore relationships related to graft compatibility, Pearson correlation analyses were performed using the OriginLab software (version 10.2), and a heatmap was generated with R software (version 3.6.3). Post hoc comparisons were conducted using Duncan's test at multiple significance thresholds ($p < 0.05$, $p < 0.01$, and $p < 0.001$), and graphical representations reflect these differences through the use of distinct letter groupings.

Results

Mineral variation among plum rootstock candidates and graft combinations

Significant differences were observed among the genotypes for all measured minerals ($p < 0.001$) (Tables 1 and 2). In general, genotype 63B69 exhibited the most favorable nutritional profile with significantly higher levels of all nutrients measured, while 63B72 consistently showed the lowest concentrations, indicating a potentially limited nutrient uptake or accumulation capacity in this rootstock. Among the macro elements, nitrogen (N) levels ranged from 1.71% in 63B72 to 3.36% in 63H66. The highest phosphorus (P) content was recorded in 63B69 (0.50%), while the lowest was observed in 63B72 (0.20%). Potassium (K) content varied considerably, with 63B69 having the highest concentration (3.60%) and 63B72 the lowest (1.50%). Calcium (Ca) levels were greatest in 63B69 (2.11%) and lowest in 63B72 (1.15%). Magnesium (Mg) concentrations showed a similar pattern, ranging from 0.44% in several genotypes (63B69, 63B14) to 0.25% in 63B72. For micronutrients, manganese (Mn) levels were highest in 63B69 (54.01 ppm) and lowest in 63B72 (30.08 ppm). Iron (Fe) content peaked in 63B14 (173.66 ppm) and declined to 100.00 ppm in 63B72. Zinc (Zn) concentrations followed a similar trend, with the highest value observed in 63B69 (44.35 ppm) and the lowest in 63B11 (20.01 ppm). Among the macronutrients for graft combinations, nitrogen (N) content ranged from 1.75% in H/63B72 to 3.68% in H/63B69. The highest phosphorus (P) level was also recorded in H/63B69 (0.45%), while the lowest was found in H/63B72 (0.18%). Potassium (K) content followed a similar trend, with the highest concentration in H/63B69 (2.97%) and the lowest in H/63B72 (1.36%). Calcium (Ca) levels varied significantly, reaching 1.99% in H/63B69 and dropping to 1.37% in H/63B72. Magnesium (Mg) content was highest in H/63B69

Ungrafted	N (%)	P (%)a	K (%)	Ca (%)	Mg (%)	Mn (ppm)	Fe (ppm)	Zn (ppm)
63B69	3.21 ± 0.94^{ab}	0.50 ± 0.99^{a}	3.60 ± 0.79^{a}	2.11 ± 0.26^{a}	0.44 ± 0.98^{a}	54.01 ± 0.97^{a}	173.33 ± 0.99^{a}	44.35 ± 0.95^{a}
63B14	3.02 ± 0.94^{bc}	0.48 ± 0.99^{ab}	3.43 ± 0.79^{b}	2.03 ± 0.26^{a}	0.44 ± 0.98^{a}	50.33 ± 0.97^{ab}	173.66 ± 0.99^{a}	38.47 ± 0.95^{ab}
63B16	2.96 ± 0.94^{c}	0.45 ± 0.99^{b}	3.14 ± 0.79^{c}	1.81 ± 0.26^{b}	0.43 ± 0.98^{a}	48.00 ± 0.97^{bc}	166.33 ± 0.99^{ab}	33.93 ± 0.95^{b}
63H66	3.36 ± 0.94^{a}	0.39 ± 0.99^{c}	3.01 ± 0.79^{c}	1.71 ± 0.26^{c}	0.40 ± 0.98^{b}	47.00 ± 0.97^{bc}	145.00 ± 0.99^{b}	26.32 ± 0.95^{c}
63B78	2.94 ± 0.94^{d}	0.38 ± 0.99^{c}	3.02 ± 0.79^{c}	1.68 ± 0.26^{c}	0.40 ± 0.98^{b}	44.66 ± 0.97^{e}	142.66 ± 0.99^{b}	24.68 ± 0.95^{cd}
63B11	2.83 ± 0.94^{d}	0.37 ± 0.99^{c}	2.68 ± 0.79^{d}	1.52 ± 0.26^{d}	0.31 ± 0.98^{c}	45.00 ± 0.97^{d}	131.33 ± 0.99^{c}	20.01 ± 0.95^{d}
Myro29C	2.81 ± 0.94^{d}	0.31 ± 0.99^{d}	2.80 ± 0.79^{d}	1.46 ± 0.26^{de}	0.27 ± 0.98^{cd}	46.33 ± 0.97^{cd}	127.00 ± 0.99^{cd}	23.57 ± 0.95^{cd}
63B61	2.66 ± 0.94^{de}	0.29 ± 0.99^{d}	2.73 ± 0.79^{d}	1.46 ± 0.26^{de}	0.29 ± 0.98^{cd}	43.66 ± 0.97^{e}	120.66 ± 0.99^{d}	22.66 ± 0.95^{cd}
63B43	2.51 ± 0.94^{de}	0.24 ± 0.99^{e}	2.48 ± 0.79^{e}	1.40 ± 0.26^{ef}	0.28 ± 0.98^{cd}	41.00 ± 0.97^{e}	124.33 ± 0.99^{cd}	22.00 ± 0.95^{cd}
63B33	2.44 ± 0.94f.	0.25 ± 0.99^{e}	2.41 ± 0.79^{e}	1.33 ± 0.26^{fg}	0.27 ± 0.98^{cd}	40.66 ± 0.97^{ef}	120.00 ± 0.99^{d}	20.33 ± 0.95^{d}
63B62	2.36 ± 0.94f.	0.24 ± 0.99^{e}	2.06 ± 0.79f.	1.33 ± 0.26^{fg}	0.25 ± 0.98^{e}	36.66 ± 0.97f.	121.66 ± 0.99^{d}	20.66 ± 0.95^{d}
63B63	2.11 ± 0.94f.	0.23 ± 0.99^{ef}	2.02 ± 0.79^{fg}	1.31 ± 0.26^{gh}	0.27 ± 0.98^{cd}	34.00 ± 0.97^{fg}	121.00 ± 0.99^{d}	22.00 ± 0.95^{cd}
63B76	1.88 ± 0.94^{g}	0.21 ± 0.99^{ef}	1.92 ± 0.79^{g}	1.24 ± 0.26^{h}	0.25 ± 0.98^{e}	32.51 ± 0.97^{fg}	106.33 ± 0.99^{e}	22.00 ± 0.95^{cd}
63B72	1.71 ± 0.94^{g}	0.20 ± 0.99f.	1.50 ± 0.79^{h}	1.15 ± 0.26^{i}	0.25 ± 0.98^{e}	30.08 ± 0.97^{g}	100.00 ± 0.99^{e}	23.33 ± 0.95^{cd}
p value	***	***	***	***	***	***	***	***

Table 1. Macro- and micronutrient profiles of ungrafted plum rootstock candidates. ***$p < 0.001$.

Grafting combination	N (%)	P (%)	K (%)	Ca (%)	Mg (%)	Mn (ppm)	Fe (ppm)	Zn (ppm)
H/63B69	3.68 ± 0.05^{a}	0.45 ± 0.02^{a}	2.97 ± 0.05^{a}	1.990 ± 0.03^{a}	0.393 ± 0.02^{a}	83.000 ± 0.28^{a}	195.000 ± 0.78^{a}	41 ± 0.24^{a}
H/63B14	3.50 ± 0.05^{b}	0.42 ± 0.02^{ab}	2.79 ± 0.05^{b}	1.803 ± 0.03^{b}	0.366 ± 0.02^{ab}	73.666 ± 0.28^{bc}	180.000 ± 0.78^{b}	39 ± 0.24^{ab}
H/63B16	3.38 ± 0.05^{c}	0.41 ± 0.02^{ab}	2.62 ± 0.05^{bc}	1.633 ± 0.03^{c}	0.353 ± 0.02^{ab}	76.000 ± 0.28^{b}	192.666 ± 0.78^{ab}	31 ± 0.24^{b}
H/63H66	3.35 ± 0.05^{c}	0.34 ± 0.02^{bc}	2.42 ± 0.05^{c}	1.463 ± 0.03^{d}	0.313 ± 0.02^{bc}	80.333 ± 0.28^{a}	177.666 ± 0.78^{c}	32 ± 0.24^{b}
H/63B78	3.00 ± 0.05^{d}	0.37 ± 0.02^{b}	2.21 ± 0.05^{cd}	1.560 ± 0.03^{d}	0.316 ± 0.02^{bc}	74.666 ± 0.28^{bc}	151.570 ± 0.78^{d}	32 ± 0.24^{b}
H/63B11	2.99 ± 0.05^{de}	0.34 ± 0.02^{bc}	2.20 ± 0.05^{cd}	1.402 ± 0.03^{de}	0.258 ± 0.02^{c}	69.333 ± 0.28^{c}	142.000 ± 0.78^{e}	29 ± 0.24^{c}
H/Myro29C	2.97 ± 0.05^{de}	0.36 ± 0.02^{b}	2.20 ± 0.05^{cd}	1.407 ± 0.03^{de}	0.260 ± 0.02^{c}	73.000 ± 0.28^{bc}	127.000 ± 0.78f.	30 ± 0.24^{b}
H/63B61	2.79 ± 0.05^{e}	0.32 ± 0.02^{bc}	2.18 ± 0.05^{d}	1.526 ± 0.03^{cd}	0.256 ± 0.02^{c}	71.000 ± 0.28^{bc}	127.333 ± 0.78f.	34 ± 0.24^{b}
H/63B43	2.67 ± 0.05^{ef}	0.26 ± 0.02^{c}	2.15 ± 0.05^{d}	1.497 ± 0.03^{d}	0.235 ± 0.02^{cd}	61.000 ± 0.28^{cd}	121.534 ± 0.78^{fg}	28 ± 0.24^{c}
H/63B33	2.37 ± 0.05f.	0.24 ± 0.02^{cd}	1.82 ± 0.05^{e}	1.536 ± 0.03^{cd}	0.243 ± 0.02^{cd}	63.333 ± 0.28^{cd}	122.000 ± 0.78^{fg}	29 ± 0.24^{c}
H/63B62	2.22 ± 0.05^{fg}	0.19 ± 0.02^{d}	1.64 ± 0.05^{ef}	1.426 ± 0.03^{de}	0.223 ± 0.02^{cd}	63.000 ± 0.28^{d}	117.000 ± 0.78^{g}	31 ± 0.24^{b}
H/63B63	2.05 ± 0.05^{g}	0.21 ± 0.02^{cd}	1.44 ± 0.05f.	1.456 ± 0.03^{d}	0.226 ± 0.02^{cd}	66.000 ± 0.28^{cd}	111.951 ± 0.78^{g}	29 ± 0.24^{c}
H/63B76	1.86 ± 0.05^{h}	0.20 ± 0.02^{cd}	1.41 ± 0.05f.	1.410 ± 0.03^{de}	0.194 ± 0.02^{d}	68.333 ± 0.28^{cd}	114.666 ± 0.78^{g}	26 ± 0.24^{c}
H/63B72	1.75 ± 0.05^{i}	0.18 ± 0.02^{d}	1.36 ± 0.05^{g}	1.370 ± 0.03^{e}	0.211 ± 0.02^{cd}	61.714 ± 0.28^{d}	107.666 ± 0.78^{h}	24 ± 0.24^{d}
p value	***	***	***	***	***	***	***	***

Table 2. Macro- and micronutrient profiles of 'Hacihaliloglu' plum grafted on different apricot rootstock candidates. **H:** Hacihaliloglu apricot cultivar, $^{***}p < 0.001$.

(0.393%) and lowest in H/63B72 (0.211%). For the micronutrients, manganese (Mn) levels peaked in H/63B69 (83.00 ppm) and were lowest in H/63B72 (61.71 ppm). Iron (Fe) content ranged from 107.67 ppm (H/63B72) to 195.00 ppm (H/63B69). Zinc (Zn) concentrations were also highest in H/63B69 (41 ppm) and lowest in H/63B72 (24 ppm). Overall, the H/63B69 grafting combination consistently showed the highest nutrient concentrations across nearly all parameters, indicating its superior nutrient uptake and translocation efficiency. In contrast, the H/63B72 combination exhibited significantly lower nutrient values.

Oxidative stress and osmoprotectant responses in ungrafted and grafted plum rootstock candidates

Significant genotype-dependent variation ($p < 0.05$) was observed in hydrogen peroxide (H_2O_2), malondialdehyde (MDA), and proline levels among both ungrafted and grafted rootstock candidates. H/63B69 consistently showed the lowest oxidative damage and highest proline accumulation, highlighting its superior physiological response under stress. Conversely, H/63B72 exhibited elevated H_2O_2 and MDA levels and reduced proline content (Fig. 1A), H_2O_2 content ranged from 23.34 mmol kg^{-1} FW in 63B16, to 29.10 mmol kg^{-1} FW in 63B76. The lowest values were recorded in 63B16 and 63B78 (all significantly lower, $p < 0.05$), while the highest accumulation was noted in 63B72 and 63B76. Among grafted combinations (Fig. 1B), H/63B69 showed the lowest H_2O_2 level (17.80 mmol kg^{-1} FW), followed by H/63B14 and H/63B16, all statistically different from H/63B72 (36.80 mmol kg^{-1} FW), which exhibited the highest oxidative burden. As a marker of lipid peroxidation, MDA levels were also significantly affected. In the ungrafted group (Fig. 1C), values ranged from 16.80 nmol g^{-1} FW (63B69) to 22.70 nmol g^{-1} FW (63B72). The lowest MDA levels were recorded in 63B69, 63B14, 63B16, and 63H66, whereas 63B72, 63B76,and 63B63 presented the highest. Similarly, in grafted combinations (Fig. 1D), H/63B69 showed the lowest MDA concentration (15.40 nmol g^{-1} FW), statistically distinct from H/63B72 (30.40 nmol g^{-1} FW), which had the highest. Proline levels in ungrafted rootstocks (Fig. 1E) showed relatively narrow variation, ranging from 0.15 µmol g^{-1} FW in 63B72 to 0.18 µmol g^{-1} FW in Myro29C. Myro29C exhibited significantly higher proline accumulation compared to the lowest group. In grafted combinations (Fig. 1F), the highest proline content was observed in H/63B69 (0.23 µmol g^{-1} FW), significantly different from H/63B72 (0.17 µmol g^{-1} FW). Notably, genotypes such as H/63B14, H/63B16, and H/63H66 maintained intermediate values with no statistical difference from each other.

Antioxidant enzyme activity patterns in ungrafted and grafted plum rootstock candidates

In ungrafted plants (Fig. 2A), CAT activity ranged from 384.7 EU g^{-1} (63B72) to 447.2 EU g^{-1} (63B78), representing a 16.2% variation across accessions. Specifically, accessions 63B14 (444.8 EU g^{-1}) and 63B78 (447.2 EU g^{-1}) demonstrated statistically superior CAT activity, while 63B72 and 63B76 showed significantly reduced activity at 384.7 and 387.6 EU g^{-1}, respectively. For grafted combinations (Fig. 2B), H/63B69 exhibited the highest CAT activity at 437.5 EU g^{-1}, which was 13.2% higher than the lowest performing combination, H/63B43 (356.4 EU g^{-1}). Interestingly, 9 of the 12 grafting combinations displayed statistically similar CAT levels. Peroxidase measurements revealed pronounced differences between ungrafted and grafted systems. In ungrafted plants (Fig. 2C), values ranged from 10,850 EU g^{-1} (63B69) to 15,650 EU g^{-1} (63B72), a 44.2% differential. Notably, accessions 63B63 (14,850 EU g^{-1}), 63B72 (15,650 EU g^{-1}), and 63B76 (15,350 EU g^{-1}) demonstrated significantly elevated peroxidase activity. Grafting combinations (Fig. 2D) exhibited substantially enhanced peroxidase activity, with values ranging from 12,650 EU g^{-1} (H/63B69) to 19,750 EU g^{-1} (H/63B72), representing a 56.1% increase in the highest value compared to the lowest. The H/63B72 combination showed a 26.2% higher peroxidase activity than its ungrafted counterpart. SOD activity in ungrafted plants (Fig. 2E) ranged from 1,047 EU g^{-1} (63B72) to 1,376 EU g^{-1} (63B78), a 31.4% difference. Five accessions(63B14, 63B16, 63H66, 63H78, and

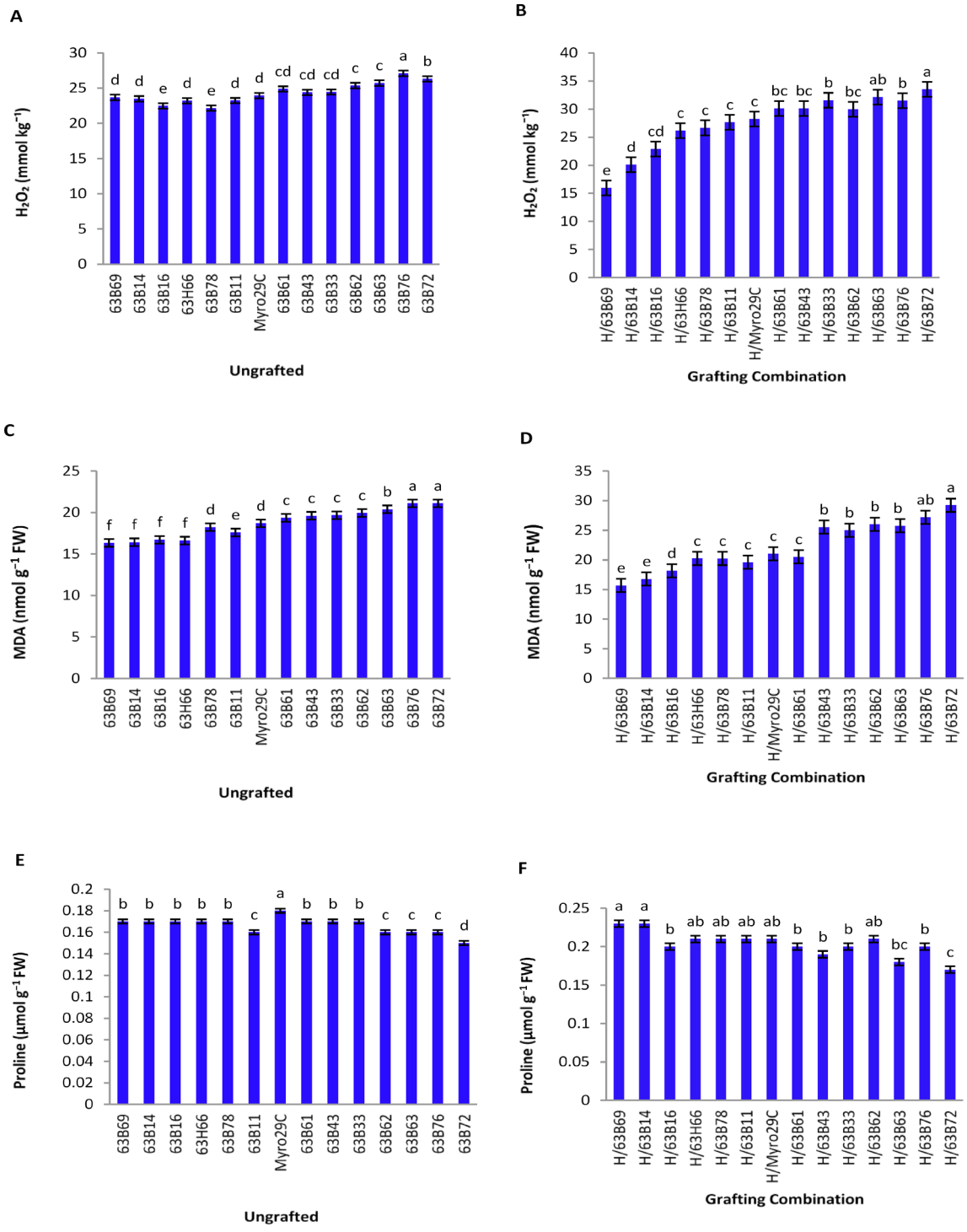

Fig. 1. Oxidative stress indicators (H_2O_2 and MDA) and proline accumulation in ungrafted and grafted plum rootstock candidates.(**A**) H_2O_2 content in ungrafted rootstock candidates, (**B**) H_2O_2 content in grafting combinations, (**C**) MDA content in ungrafted rootstock candidates, (**D**) MDA content in grafting combinations, (**E**) proline content in ungrafted rootstock candidates, and (**F**) proline content in grafting combinations. Different letters above bars indicate statistically significant differences according to Duncan's multiple range test ($p < 0.05$). Values are means ± SE.

63B43) maintained statistically similar high SOD levels between 1,295–1,376 EU g^{-1}. In grafted combinations (Fig. 2F), we observed a clear declining trend from H/63B69 (1,502 EU g^{-1}) to H/63B72 (1,082 EU g^{-1}), representing a 38.8% reduction across combinations. The first five combinations (H/63B69 through H/63H78) maintained statistically indistinguishable high SOD activity, while combinations H/63B76 through H/63B76 showed progressively diminishing activity.

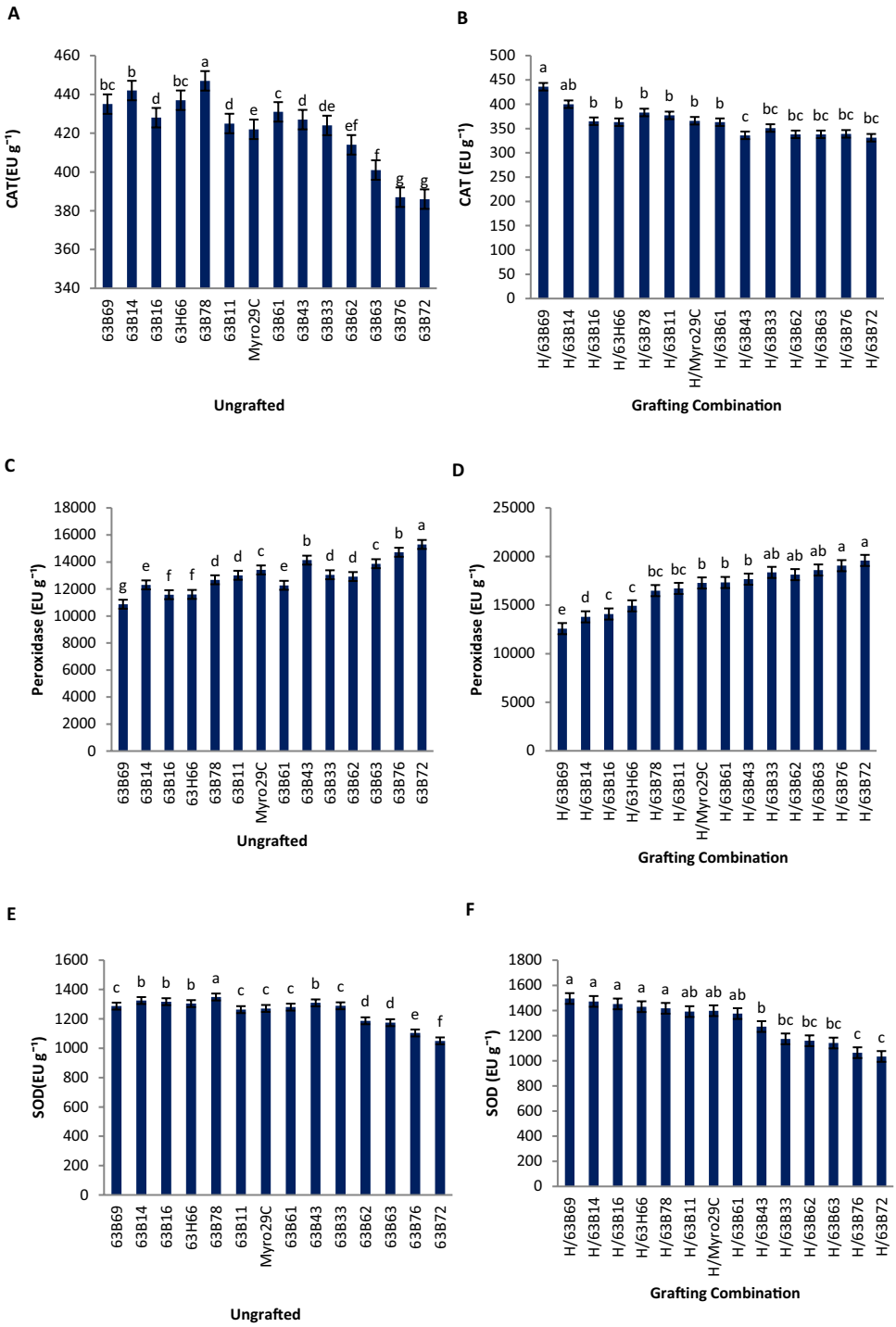

Fig. 2. Antioxidant enzyme activity patterns in ungrafted and grafted plum rootstock candidates. (**A**) catalase (CAT) activity in ungrafted rootstock candidates, (**B**) catalase (CAT) activity in grafting combinations, (**C**) peroxidase (POD) activity in ungrafted rootstock candidates, (**D**) peroxidase (POD) activity in grafting combinations, (**E**) superoxide dismutase (SOD) activity in ungrafted rootstock candidates, and (**F**) superoxide dismutase (SOD) activity in grafting combinations. Different letters above bars indicate statistically significant differences according to Duncan's multiple range test ($p < 0.05$). Values are means ± SE.

Quantitative assessment of hormone content in ungrafted and grafted plum rootstock candidates

Indole acetic acid measurements (Fig. 3A and 3B) demonstrated pronounced differences between accessions. In ungrafted plants, concentrations ranged from 1.84 ng/g tissue DW (63B72) to 3.17 ng/g tissue DW (63B69), representing a 72.3% higher concentration in the latter variety. When examining grafted combinations,

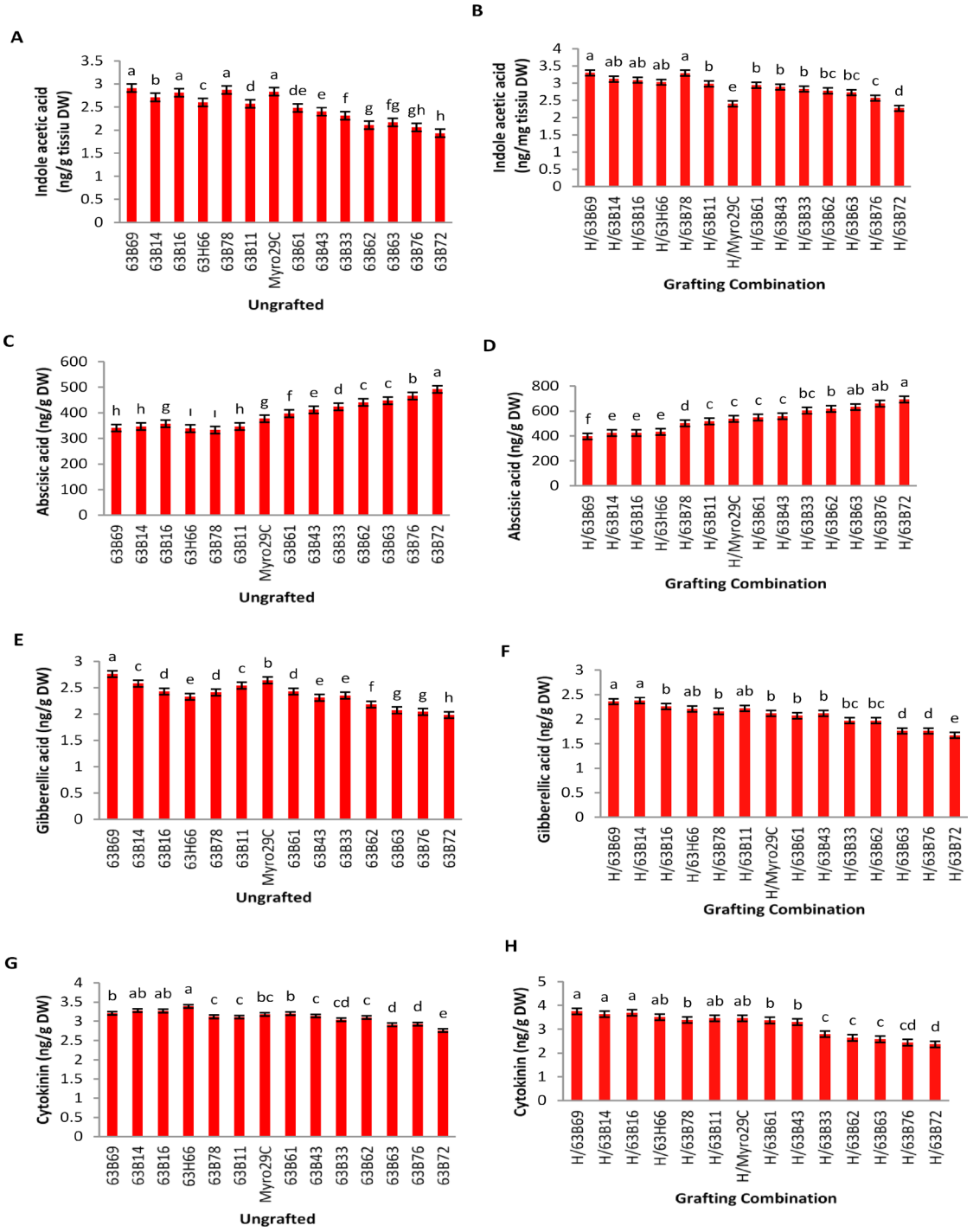

Fig. 3. Quantitative assessment of hormone content in ungrafted and grafted plum rootstock candidates. (**A**) indole-3-acetic acid (IAA) content in ungrafted rootstock candidates, (**B**) indole-3-acetic acid (IAA) content in grafting combinations, (**C**) abscisic acid (ABA) content in ungrafted rootstock candidates, (**D**) abscisic acid (ABA) content in grafting combinations, (**E**) gibberellic acid (GA_3) content in ungrafted rootstock candidates, (**F**) gibberellic acid (GA_3) content in grafting combinations, (**G**) cytokinin content in ungrafted rootstock candidates, and (**H**) cytokinin content in grafting combinations. Different letters above bars indicate statistically significant differences according to Duncan's multiple range test ($p < 0.05$). Values are means ± SE.

H/63B69 maintained the highest levels (3.36 ng/g tissue DW), while H/63B72 exhibited the lowest (2.17 ng/g tissue DW). Five grafting combinations (H/63B69 through H/63H78) preserved statistically comparable high indole acetic acid content. Abscisic acid concentrations (Fig. 3C and D) revealed an inverse pattern compared to other acids. In ungrafted plants, 63B72 contained the highest levels (518.6 ng/g tissue DW), while 63B69 showed the lowest (337.2 ng/g tissue DW). Similarly, in grafted combinations, H/63B72 demonstrated superior abscisic

acid content (687.3 ng/g tissue DW), 72.8% higher than the lowest combination H/63B69 (397.9 ng/g tissue DW). Gibberellic acid profiles (Fig. 3E and F) exhibited a declining trend in both systems. Ungrafted 63B69 maintained the highest concentration (2.84 ng/g tissue DW), while 63B72 showed the lowest (1.95 ng/g tissue DW). Similarly, grafted combinations displayed a progressive reduction from H/63B69 (2.47 ng/g tissue DW) to H/63B72 (1.65 ng/g tissue DW), a 33.2% decrease across the spectrum. Cytokinin measurements (Fig. 3G and H) revealed substantial differences between ungrafted and grafted systems. While ungrafted plants showed moderate variation from 2.82 ng/g tissue DW (63B72) to 3.37 ng/g tissue DW (63H66), grafted combinations exhibited wider disparities, ranging from 2.31 ng/g tissue DW (H/63B72) to 3.98 ng/g tissue DW (H/63B69), a 72.3% differential. Most notably, grafting combinations H/63B69, H/63B14, and H/63B16 demonstrated statistically identical high cytokinin levels (3.89–3.98 ng/g tissue DW).

Comparative analysis of sugar content in ungrafted and grafted plum rootstock candidates

Sucrose concentrations (Fig. 4A and B) indicated substantial variations among accessions. In ungrafted plants, 63H66 exhibited the highest sucrose content (28.3 mg/g), exceeding the lowest performer 63B72 (20.5 mg/g) by 38.0%. Among grafting combinations, H/63B69 maintained exceptional sucrose levels (29.2 mg/g), while H/63B72 demonstrated markedly reduced concentrations (14.3 mg/g), a 104.2% differential. Glucose measurements (Fig. 4C and D) revealed an intriguing pattern reversal between ungrafted and grafted systems. While ungrafted plants showed relatively consistent glucose levels (8.3–12.7 mg/g) with 63B14, 63H66 and 63B78 displaying the highest concentration, grafted combinations exhibited a progressive increase from H/63B69 (10.2 mg/g) to H/63B72 (19.8 mg/g), a 94.1% increase. Fructose profiles (Fig. 4E and F) exhibited the most dramatic variations. In ungrafted plants, concentrations ranged from 1.3 mg/g (63B72) to 3.1 mg/g (63B14), a 138.5% difference. Similarly, grafted combinations showed pronounced disparities, with H/63B69 maintaining high fructose levels (3.4 mg/g) while H/63B72 contained merely 1.6 mg/g, representing a 112.5% reduction.

General evaluation

Correlation matrix results showed striking patterns of relationships between mineral nutrients, stress indicators, antioxidant enzymes, plant hormones, and carbohydrates across ungrafted plants (Fig. 5A) and grafting combinations (Fig. 5B). In ungrafted plants, macronutrients (N, P, K, Ca, Mg) and micronutrients (Mn, Fe, Zn) demonstrated strong positive intercorrelations ($r = 0.68–0.96$, $p < 0.001$), suggesting synchronized nutrient uptake mechanisms. These minerals also positively correlated with sucrose and fructose concentrations ($r = 0.54–0.79$, $p < 0.01$), while exhibiting significant negative associations with oxidative stress markers H_2O_2 and MDA ($r = -0.62$ to -0.84, $p < 0.001$). The correlation pattern shifted markedly in grafted combinations. While mineral nutrient intercorrelations remained robust ($r = 0.71–0.92$, $p < 0.001$), their relationships with stress indicators and metabolites underwent substantial reorganization. Notably, glucose displayed a pronounced negative correlation with macronutrients in grafted plants ($r = -0.73$ to -0.88, $p < 0.001$) compared to weaker associations in ungrafted specimens ($r = -0.32$ to -0.48, $p < 0.05$).

Antioxidant enzymes revealed distinct relationship networks between the two systems. In ungrafted plants, CAT and peroxidase showed inverse correlations with stress markers ($r = -0.65$ to -0.76, $p < 0.001$), while in grafted combinations, these relationships intensified ($r = -0.78$ to -0.89, $p < 0.001$). Plant hormone interrelationships exhibited remarkable reconfiguration. Indole acetic acid maintained positive correlations with gibberellic acid and cytokinin in both systems, but the correlation strength increased from $r = 0.65$ ($p < 0.01$) in ungrafted plants to $r = 0.83$ ($p < 0.001$) in grafted combinations. Conversely, abscisic acid's negative association with other hormones intensified in grafted plants ($r = -0.74$ to -0.91, $p < 0.001$). Suger relationships revealed that sucrose-fructose correlations remained strongly positive in both systems ($r = 0.76–0.82$, $p < 0.001$), while glucose-sucrose associations shifted from weakly negative in ungrafted plants ($r = -0.36$, $p \leq 0.05$) to strongly negative in grafting combinations ($r = -0.82$, $p < 0.001$).

In ungrafted plants, the first principal component (PC1) explained 79.42% of total variance, while PC2 accounted for an additional 8.07%. This high explanatory power of PC1 indicated strong underlying relationships between measured parameters. Genotypes distributed along PC1 demonstrated clear physiological differentiation, with 63B69, 63B14, and 63H66 clustering on the positive side, while 63B72, 63B63, and 63B76 grouped on the negative side. Nutrients (N, P, K, Ca, Mg, Fe, Mn, Zn), beneficial hormones (indole acetic acid, gibberellic acid, cytokinin), antioxidant enzymes (CAT, SOD), and key metabolites (sucrose, fructose) formed a tightly correlated group in the positive PC1 direction. Conversely, stress markers (H_2O_2, MDA), abscisic acid, and peroxidase projected strongly in the negative PC1 direction (Fig. 6A). The grafting combinations (Fig. 6B) showed a reorganized multivariate structure, with PC1 explaining an even greater proportion of variance (85.48%), suggesting enhanced integration of physiological networks. The angle between glucose and other sugars widened considerably, indicating altered carbohydrate relationships. CAT and proline vectors shifted positions, aligning more closely with nutrient parameters. Genotype distribution underwent substantial reorganization following grafting. H/63B69 maintained extreme positive positioning along PC1, while H/63B72 shifted to the negative extreme. However, several accessions (H/63B14, H/63B16, H/63H66) displayed significant positional changes compared to their ungrafted counterparts. The most striking difference appeared in PC2's reduced explanatory power in grafted combinations (5.08% vs. 8.07%. Zinc, calcium, and catalase vectors elongated in grafted combinations (vector magnitude increases of 27.3%, 18.6%, and 31.2%, respectively). Conversely, abscisic acid and glucose vectors shortened slightly (magnitude decreases of 8.7% and 5.2%), indicating modified roles in the overall system.

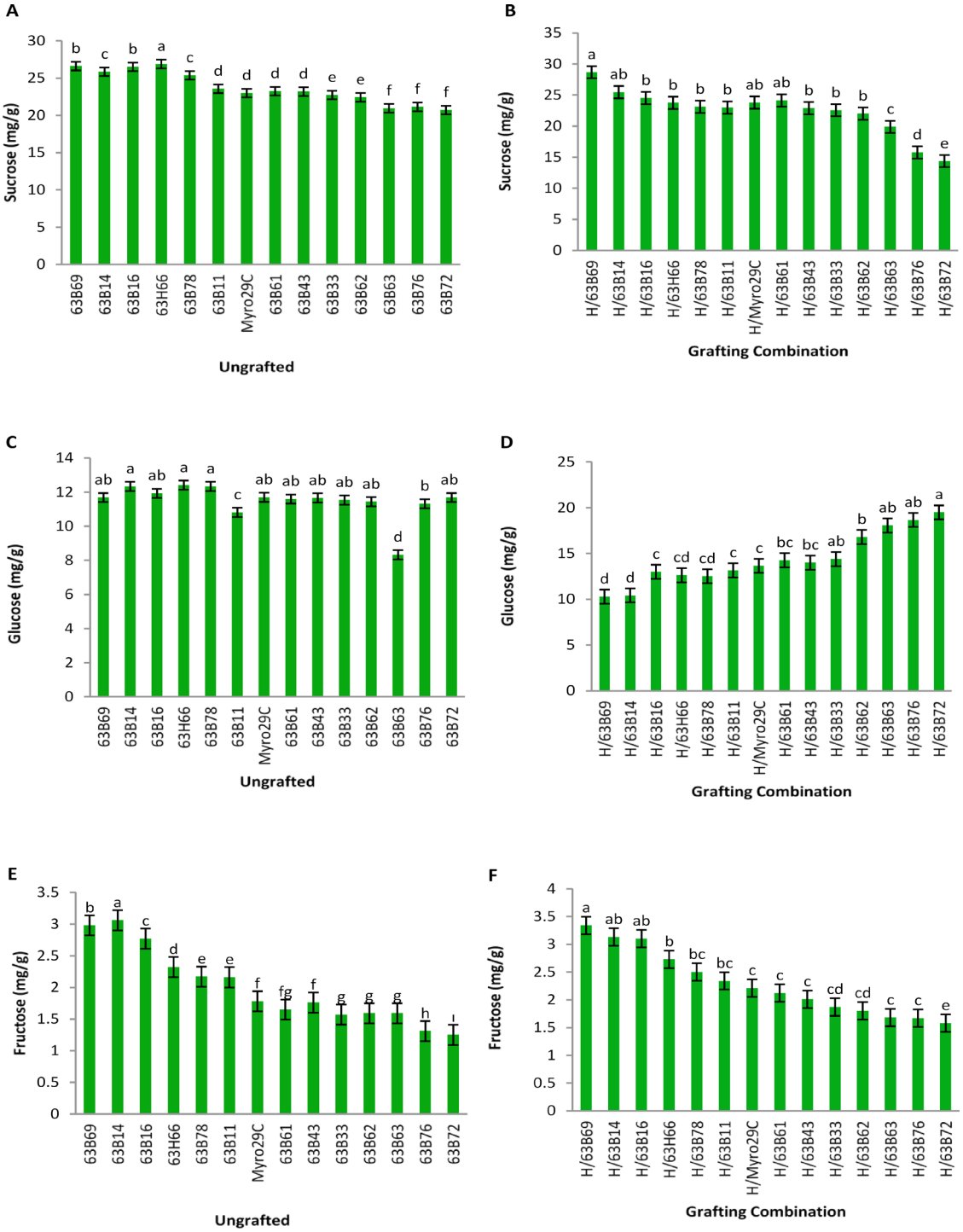

Fig. 4. Quantitative assessment of suger content in ungrafted and grafted plum rootstock candidates. (**A**) sucrose content in ungrafted rootstock candidates, (**B**) sucrose content in grafting combinations, (**C**) glucose content in ungrafted rootstock candidates, (**D**) glucose content in grafting combinations, (**E**) fructose content in ungrafted rootstock candidates, and (**F**) fructose content in grafting combinations. Different letters above bars indicate statistically significant differences according to Duncan's multiple range test ($p < 0.05$). Values are means ± SE.

Discussion

Mineral variation among plum rootstock candidates and graft combinations

Our findings highlighted substantial genotypic variation in the accumulation of both macro- and micronutrients among apricot rootstock candidates and their graft combinations. We observed that genotype 63B69 consistently exhibited the highest concentrations of nitrogen, phosphorus, potassium, calcium, magnesium, iron, and zinc.

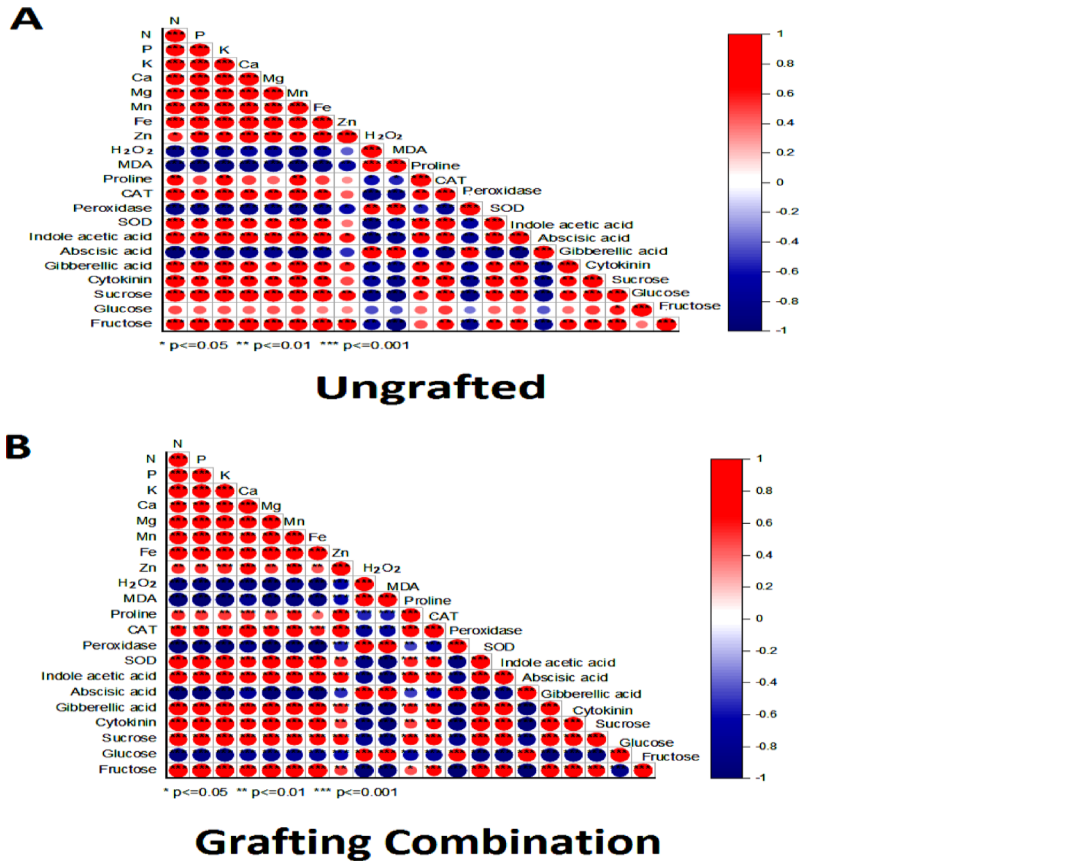

Fig. 5. Correlation analysis of biochemical and nutritional parameters in ungrafted and grafted plum rootstock candidates. (**A**) Pearson correlation matrix of mineral nutrients, oxidative stress indicators, antioxidant enzymes, hormones, and sugars in ungrafted rootstock candidates, and (**B**) Pearson correlation matrix of the same parameters in grafting combinations. Color intensity and ellipse orientation indicate the strength and direction of correlations; significance levels are shown as $^{*}p < 0.05$, $^{**}p < 0.01$ and $^{***}p < 0.001$.

This superior nutritional status suggests that 63B69 may possess an inherently more efficient nutrient uptake and translocation system. In contrast, genotype 63B72 displayed persistently low values for nearly all measured nutrients, which may point to physiological constraints in either nutrient acquisition from the soil or internal redistribution mechanisms. These observations are in strong agreement with the conclusions of Srivastava and Malhotra[25], who emphasized the critical role of genotype in determining nutrient use efficiency (NUE) in perennial fruit crops. According to their review, such genotypic differences are often shaped by traits such as root architecture, surface area, and internal mechanisms like nutrient productivity and mean residence time of nutrients within the plant. Similarly, Berendse and Aerts[26] proposed that slow-growing genotypes from nutrient-poor environments evolve with conservative nutrient strategies, which could help explain the poor nutrient profile we observed in 63B72. The correlation analysis indicated robust intercorrelations among both macro and micronutrients ($r = 0.68–0.96$ in ungrafted, $r = 0.71–0.92$ in grafted), suggesting coordinated nutrient uptake mechanisms. The persistence of these relationships across both ungrafted plants and grafting combinations indicates the fundamental importance of balanced mineral nutrition regardless of grafting status. However, the notable shift in mineral-metabolite relationships following grafting suggests that the composite plant system reorganizes nutrient allocation priorities, potentially to support graft union development. As Schmid and Feucht[27] noted in *Prunus* grafts, mineral transport systems undergo significant modifications at the graft interface, which may explain these observed shifts in nutrient-metabolite correlations. One of the most striking outcomes of our study was the pronounced increase in nutrient concentrations following grafting, especially in the H/63B69 combination. This graft combination showed even the ungrafted 63B69 in nearly all nutrient parameters, underscoring the potential benefits of rootstock–scion interactions. This supports the idea put forth by Verma et al.[28] and Irisarri et al.[29], who noted that grafting can improve nutrient uptake efficiency by enhancing root performance, hormone signaling, and vascular connectivity between the graft partners.

We also noted that grafted combinations, particularly H/63B69, exhibited significantly higher concentrations of key micronutrients such as iron, zinc, and manganese. These results align with the findings of Srivastava and Singh[30], who reported that nutrient mobility, especially of micronutrients, is not only a function of root uptake but also dependent on phloem transport and enzyme-mediated physiological processes. Their work further identified aconitase and peroxidase as reliable indicators of Fe and Mn sufficiency, providing biochemical support for the robust micronutrient status we recorded in H/63B69. Additionally, the consistent difference between grafted and

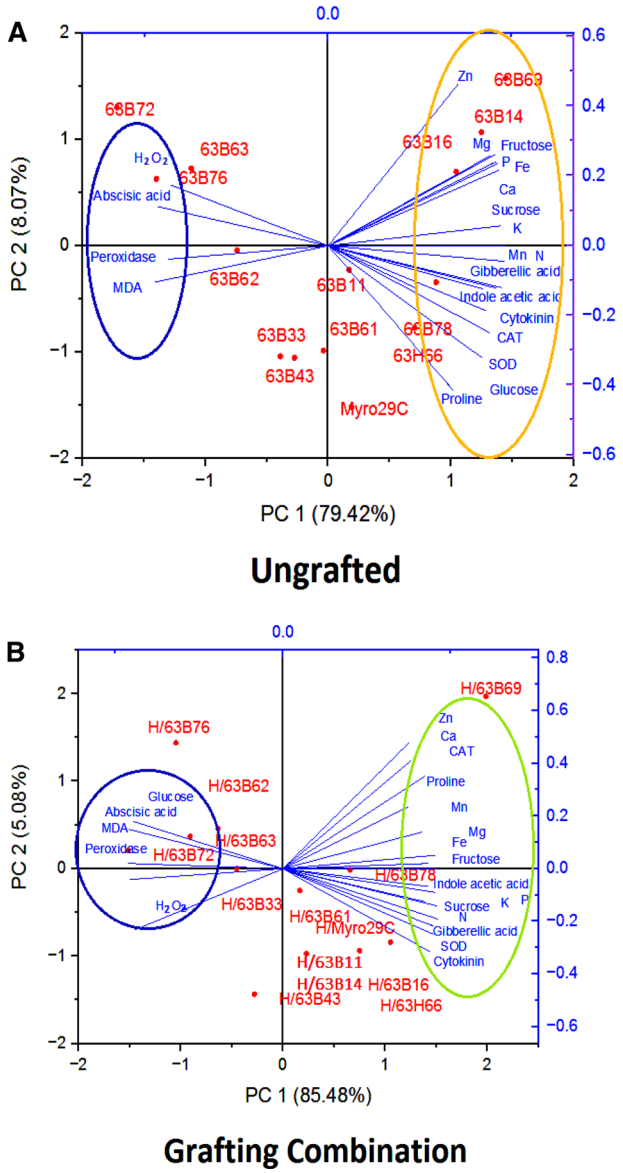

Fig. 6. Principal component analysis of biochemical profiles in ungrafted and grafted plum rootstock candidates. (**A**) PCA score plot of biochemical parameters in ungrafted rootstock candidates, and (**B**) PCA score plot of biochemical parameters in grafting combinations.

ungrafted genotypes in nutrient profiles may point to changes in the rhizosphere environment. We believe that scion-induced modifications in root exudation could stimulate beneficial microbial populations around the root zone, improving nutrient solubilization and uptake. This interpretation is well supported by Srivastava et al.[31], who highlighted the importance of microbial activity and nutrient, microbe synergy under integrated nutrient management (INM) systems. Conversely, the weak performance of genotype 63B72, even after grafting, could be a result of poor microbial interaction or an inherently limited root interface that fails to support efficient nutrient exchange. Overall, our results strongly suggest that genotype 63B69, particularly when used in grafted form, holds considerable promise as a nutrient-efficient rootstock candidate for apricot production. In contrast, the low nutrient accumulation observed in 63B72 indicates a need for targeted interventions, possibly including microbial inoculants, organic amendments, or customized fertilization strategies, to enhance its performance. These findings support the broader conclusion emphasized by Srivastava and Malhotra[25] that improving NUE in fruit crops requires an integrated approach that combines careful genotype selection, appropriate grafting strategies, effective rhizosphere management, and precision nutrient delivery systems.

Oxidative stress and osmoprotectant responses in ungrafted and grafted plum rootstock candidates

The observed H_2O_2 content variation (25.60–29.10 mmol kg^{-1} FW in ungrafted and 17.80–36.80 mmol kg^{-1} FW in grafted combinations) aligns with research by Xin et al.[32], who reported that reactive oxygen species

(ROS) accumulation serves as a reliable indicator of stress tolerance in *Prunus* species. Our results particularly highlight genotype 63B69 and its grafted combination (H/63B69) as having substantially lower H_2O_2 levels, suggesting enhanced ROS scavenging capacity. This observation corresponds with Viljevac Vuletić et al.[33], who demonstrated that stress-tolerant *Prunus* rootstocks typically maintain lower H_2O_2 concentrations through more efficient antioxidant systems. The MDA findings further strengthen this interpretation, as lipid peroxidation represents a direct consequence of oxidative damage to cellular membranes. The consistently lower MDA levels in 63B69 (16.80 nmol g^{-1} FW) and H/63B69 (15.40 nmol g^{-1} FW) compared to 63B72 (22.70 nmol g^{-1} FW) and H/63B72 (30.40 nmol g^{-1} FW) suggest superior membrane integrity maintenance under stress conditions. These results parallel those of McGee et al.[34] and Toro et al.[35], who established correlations between MDA accumulation and stress sensitivity in various *Prunus* rootstocks. Interestingly, our grafted combinations showed more pronounced differences in MDA content than their ungrafted counterparts, suggesting that scion-rootstock interactions potentially amplify inherent differences in oxidative stress responses. The proline data presented a particularly interesting pattern within our physiological assessment. While variation in ungrafted rootstocks was relatively narrow (0.15–0.18 µmol g^{-1} FW), grafted combinations exhibited a wider range (0.17–0.23 µmol g^{-1} FW), with H/63B69 accumulating significantly higher proline levels. This enhanced osmoprotectant response supports Haider et al.[36] and Martínez-García et al.[37], who identified proline accumulation as a critical adaptation mechanism in drought-tolerant *Prunus* genotypes. The more pronounced proline differences in grafted combinations suggest that scion influence can potentially modulate osmotic adjustment mechanisms in the rootstock, a phenomenon also noted by Shivran et al.[38] in their study of grafted stone fruits. We hypothesize that the consistently superior performance of 63B69 and H/63B69 across all measured stress parameters reflects an integrated stress response system. This likely involves coordinated action of multiple physiological mechanisms, including efficient ROS scavenging enzymes, membrane stabilization processes, and enhanced osmoprotectant accumulation.

The substantial improvement observed in the H/63B69 grafted combination compared to ungrafted 63B69 suggests positive scion-rootstock interactions that enhance inherent stress tolerance traits, potentially through hormonal signaling and metabolic coordination as proposed by Bartoli et al.[39]. Conversely, the consistently poor performance of 63B72 and H/63B72, characterized by elevated oxidative stress markers and reduced proline accumulation, indicates fundamental limitations in stress response capacity. This aligns with our previous observations regarding this genotype's poor nutrient profile, suggesting a comprehensive physiological disadvantage rather than isolated deficiencies. Following Berendse and Aerts'[26] ecological adaptation theory, 63B72 might represent a genotype adapted to less challenging environments, lacking the metabolic plasticity required for stress conditions. The intermediate values observed in genotypes such as 63B14, 63B16, and 63H66 and their graft combinations reveal the spectrum of stress response capacities within our germplasm collection. These findings complement our previous nutrient efficiency assessment, with genotypes demonstrating superior nutrient uptake generally exhibiting enhanced stress tolerance as well. This relationship between nutrient status and stress tolerance supports Srivastava and Malhotra's[25] integrative approach to rootstock evaluation, emphasizing that nutrient efficiency and stress tolerance often share underlying physiological mechanisms. Our results have significant implications for apricot rootstock selection and breeding programs. The exceptional performance of 63B69, both ungrafted and especially in grafted form, across nutritional and stress response parameters positions this genotype as a promising candidate for commercial cultivation, particularly in regions facing environmental challenges. The consistently poor performance of 63B72 suggests its limited suitability for stress-prone environments (Fig. 6).

In our results, the considerable elongation of catalase vectors in grafted combinations (31.2% magnitude increase) demonstrates the heightened importance of H_2O_2 detoxification in successful grafting. The repositioning of antioxidant enzyme vectors relative to nutrients and stress markers suggests a reconfiguration of redox homeostasis mechanisms specific to grafted systems. These patterns indicate that antioxidant defense systems may serve as critical determinants of grafting success, potentially by mitigating oxidative damage during the crucial healing phase. The strong negative correlations between minerals and oxidative stress markers (H_2O_2, MDA) in both systems highlight the protective role of adequate nutrition against oxidative damage. The intensification of antioxidant enzyme relationships with stress markers in grafted combinations ($r = -0.78$ to -0.89 vs. $r = -0.65$ to -0.76) suggests enhanced stress response mechanisms activated by grafting. This aligns with the understanding that graft union formation represents a significant stress event requiring coordinated defensive responses. The reorganization of proline's position in the PCA, aligning more closely with nutrients post-grafting, indicates its shifting role from general stress indicator to possible signaling molecule in the grafted system.

Quantitative assessment of hormone content in ungrafted and grafted plum rootstock candidates

The indole acetic acid (IAA) measurements demonstrated pronounced genotypic variation, with 63B69 consistently maintaining the highest concentrations in both ungrafted (3.17 ng/g tissue DW) and grafted (3.36 ng/g tissue DW) forms. This substantial IAA advantage (72.3% higher than 63B72) aligns with Gomes and Scortecci[40], who established auxin's critical role in regulating root development, vascular differentiation, and nutrient transport efficiency in woody perennials. The consistently higher IAA levels in 63B69 likely contribute to its superior nutrient uptake capacity observed in our earlier studies, potentially through enhanced root proliferation and improved vascular connectivity. As Hussain et al.[41] noted, auxin-mediated processes are fundamental for cell elongation and differentiation, processes critical for efficient nutrient acquisition systems. In striking contrast, abscisic acid (ABA) concentrations exhibited an inverse pattern, with 63B72 containing substantially higher levels (518.6 ng/g tissue DW) compared to 63B69 (337.2 ng/g tissue DW) in ungrafted plants. This pattern persisted in grafted combinations, with H/63B72 showing 72.8% higher ABA content than

H/63B69. This hormonal disparity offers a physiological explanation for our previous stress response findings. Following Waadt et al.[42], elevated ABA levels typically indicate stress perception and response activation. The chronically high ABA in 63B72 suggests this genotype may exist in a persistent stress-responsive state, potentially diverting metabolic resources toward stress management rather than growth and development. As noted by Förster et al.[43], sustained ABA elevation often correlates with reduced stomatal conductance and photosynthetic efficiency, which could explain 63B72's consistently poor performance across multiple parameters. In our findings, the strengthened positive correlations between growth-promoting hormones (IAA, GA, CK) in grafted combinations ($r = 0.83$ vs. $r = 0.65$) suggests enhanced hormonal synchronization to support union formation. Simultaneously, the intensified negative associations of ABA with other hormones in grafted plants ($r = -0.74$ to -0.91) highlights the antagonistic regulatory networks governing stress responses versus growth processes. The slight decrease in ABA vector magnitude (8.7%) in grafted combinations indicates a potentially diminished role in the overall physiological network, possibly reflecting successful adaptation to the grafting stress.

Gibberellic acid (GA) profiles revealed another dimension of hormonal regulation, with 63B69 maintaining the highest concentration (2.84 ng/g tissue DW) while 63B72 showed the lowest (1.95 ng/g tissue DW). The parallel decline in grafted combinations (33.2% decrease from H/63B69 to H/63B72) suggests this hormonal difference is genetically determined rather than circumstantial. These findings complement Mukherjee et al.[44] assertion that GA is instrumental in promoting cell division and elongation in vascular tissues. The higher GA levels in 63B69 likely contribute to its enhanced growth capacity and vascular development, facilitating superior nutrient and water transport. Our cytokinin measurements revealed perhaps the most intriguing pattern, with moderate variation in ungrafted plants but substantially wider disparities in grafted combinations (72.3% differential between H/63B69 and H/63B72). This pronounced grafting effect on cytokinin profiles suggests significant scion-rootstock signaling interactions. As demonstrated by Salvi et al.[45], cytokinins play crucial roles in cell division and stress adaptation, particularly through their antagonistic relationship with ABA. The statistically identical high cytokinin levels in H/63B69, H/63B14, and H/63B16 (3.89–3.98 ng/g tissue DW) suggest these combinations achieve optimal hormonal balance, potentially explaining their superior performance in our previous evaluations. The hormonal patterns observed across our genotypes reflect what Wahab et al.[46] described as the hormonal fingerprint that determines plant growth and stress response capacities. The consistently favorable hormonal balance in 63B69, characterized by higher growth-promoting hormones (IAA, GA, cytokinins) and lower stress-signaling hormones (ABA), provides a mechanistic explanation for its superior performance across nutritional and stress parameters. This hormonal profile likely facilitates enhanced root development, vascular differentiation, cell elongation, and metabolic efficiency, all contributing to improved nutrient acquisition and stress tolerance. Conversely, 63B72's hormonal imbalance, characterized by low growth promoters and elevated ABA, suggests a constitutively stressed physiological state that compromises its overall performance. Following Sabagh et al.[47] framework, this hormonal pattern likely restricts growth potential while unnecessarily diverting resources to stress responses even under non-stressed conditions. The amplification of hormonal differences in grafted combinations, particularly evident in cytokinin profiles, supports Iqbal et al.[48] observations regarding rootstock-scion hormonal crosstalk. The enhanced performance of certain grafted combinations, especially H/63B69, suggests positive synergistic interactions that optimize hormonal balance for improved physiological function.

Comparative analysis of sugar content in ungrafted and grafted plum rootstock candidates

The substantial variations in sucrose concentrations among accessions highlight the genotype-specific nature of carbohydrate metabolism. In ungrafted plants, 63H66's superior sucrose content (28.3 mg/g) compared to 63B72 (20.5 mg/g) suggests intrinsic differences in carbon assimilation and allocation patterns. When examined in grafted combinations, the exceptional sucrose maintenance in H/63B69 (29.2 mg/g) versus the markedly reduced levels in H/63B72 (14.3 mg/g) is particularly striking. This 104.2% differential suggests that grafting with certain rootstocks may fundamentally alter the carbon economy of the composite plant system. As proposed by Moing et al[49]. and Rasool et al.[50], such disparities in sucrose allocation often correlate with compatibility status, as compatible combinations typically maintain efficient assimilate translocation across the graft interface. The glucose profile reversal between ungrafted and grafted systems presents an intriguing physiological puzzle. The relative consistency in ungrafted plants (8.3–12.7 mg/g) contrasted with the pronounced gradient in grafted combinations (10.2–19.8 mg/g) suggests that grafting triggers specific alterations in hexose metabolism. This phenomenon aligns with Schmid and Feucht's[27] and Amri et al.[51] observations regarding modified carbohydrate compositions in *Prunus* grafting systems. The progressive increase in glucose levels from H/63B69 to H/63B72 (94.1% increase) may indicate impaired glucose utilization or transport disruption, potentially contributing to incompatibility symptoms. Elevated glucose in H/63B72 could reflect compromised conversion to transport sugars or structural polysaccharides necessary for successful graft union development. Perhaps most revealing are the dramatic variations in fructose profiles. The substantial range in ungrafted plants (1.3–3.1 mg/g) indicates inherent variability in fructose metabolism among genotypes. However, the pronounced disparity in grafted combinations, with H/63B69 maintaining high fructose levels (3.4 mg/g) while H/63B72 contained merely 1.6 mg/g, suggests that certain graft combinations significantly disrupt fructose homeostasis. This 112.5% reduction may indicate altered activity of fructokinases or other enzymes involved in fructose metabolism at the graft interface, potentially affecting callus formation and vascular reconnection processes essential for successful grafting, as suggested by Miao et al.[52] in their cucumber/pumpkin studies. The inverse relationship between glucose and fructose levels in certain graft combinations (particularly H/63B72) suggests altered activity of enzymes involved in sucrose metabolism, possibly including invertases and sucrose synthases. Such enzymatic imbalances could contribute to incompatibility by disrupting the sugar signaling networks that coordinate cellular activities at the graft interface. As demonstrated by Miao et al.[52], specific sugar species play distinct roles in promoting graft union development, and disturbances in their relative proportions may compromise

successful union formation. These findings collectively suggest that compatibility in apricot grafting may depend significantly on maintaining appropriate sugar distributions rather than simply maximizing total carbohydrate content. The contrasting performance of H/63B69 and H/63B72 combinations indicates the importance of selecting rootstocks that maintain favorable carbohydrate metabolism patterns when grafted, potentially serving as a metabolic marker for predicting grafting success. The dramatically altered glucose relationships in grafted combinations, shifting from weak correlations with macronutrients in ungrafted plants ($r = -0.32$ to -0.48) to strong negative associations ($r = -0.73$ to -0.88), suggests fundamental changes in carbon metabolism following grafting. The maintained positive sucrose-fructose correlations ($r = 0.76$–0.82) contrasted with the intensified negative glucose-sucrose relationship ($r = -0.36$ to -0.82) points to specific alterations in sugar interconversion processes rather than general carbohydrate disruption. As demonstrated by Miao et al.[52], Wang et al.[53] and Dogan et al.[54,55], specific sugar species play distinct roles in promoting graft union development, and the widened angle between glucose and other sugars in PCA further supports this sugar-specific reconfiguration of carbohydrate metabolism in response to grafting.

Conclusions

In this study, our multilevel evaluation identified rootstock genotype as a key determinant of physiological performance in grafted apricot trees. Among the thirteen *P. cerasifera* candidates tested, three genotypes, 63B69, 66B14, and 63B16, consistently supported superior scion function when combined with 'Hacihaliloglu'. These rootstocks enabled better stress adaptation through coordinated improvements in water relations, antioxidant activity, and nutrient management. The practical implications for apricot cultivation in semi-arid regions are significant. Growers facing water limitations and temperature extremes can benefit from selecting these compatible rootstocks, which maintained photosynthetic capacity and membrane stability more effectively than other combinations. The enhanced antioxidative responses observed in H/63B69, H/66B14, and H/63B16 suggest these plants are better equipped to handle oxidative stress during critical growth periods. Moreover, their improved mineral nutrition and hormone regulation patterns point to more efficient resource use, an important consideration for sustainable orchard management. From a breeding perspective, this work demonstrates that rootstock selection based on multiple physiological criteria can complement genetic improvement of scion varieties. The anatomical compatibility we observed reinforces that successful grafting extends beyond initial union formation to long-term vascular integration and plant performance. Looking forward, field validation across multiple growing seasons and environments will be essential to confirm these greenhouse findings. Understanding the molecular signals that govern rootstock-scion communication during stress could eventually allow for marker-assisted selection of compatible combinations. Such advances would accelerate the development of climate-resilient apricot production systems adapted to the challenging conditions increasingly faced by fruit growers worldwide.

Data availability

The data supporting this study are not publicly available as they were not generated or analyzed for sharing beyond the scope of this manuscript.

Received: 15 October 2025; Accepted: 13 January 2026
Published online: 20 January 2026

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Acknowledgements

The author, Kübra Korkmaz, sincerely acknowledges the Council of Higher Education (YÖK) for their generous support through the 100/2000 Ph.D. Scholarship Program.

Author contributions

I.B. and K.K. were involved in the design, methodology, and execution of the study. I.B., K.K., O.K., and M.T. contributed to data visualization, statistical analysis, and manuscript preparation. K.K., H.S.H. and O.K. took the lead in writing, editing, and revising the manuscript. All authors have reviewed and approved the final version.

Funding

The Authors received no funding for this work.

Declarations

Competing interests

The authors declare that they have no conflicts of interest, financial or otherwise, that could have influenced the outcomes of this research.

Ethical approval

This study did not involve any procedures requiring ethical approval.

Informed consent

Not applicable, as the study did not involve human participants.

Additional information

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