ENGINEERING Agriculture

Engineering Journals

RESEARCH ARTICLE

2027 / Volume 14 / Issue 1 / 27713

Effects of the Ca^{2+}/Mg^{2+} ratio on the growth and Mg absorption and distribution in cuttings of two grapevine cultivars (*Vitis vinifera*)

Qian LIU, Juan WANG (✉), Heqi ZHANG, Kunpeng TAN, Changnan YANG

College of Agriculture, Shihezi University/Xinjiang Oasis Eco-Agriculture Key Laboratory of the Corps, Shihezi 832003, China.

KEYWORDS

Absorption, Ca^{2+}/Mg^{2+} ratio, distribution, Ugni Blanc, Xinyu

HIGHLIGHTS

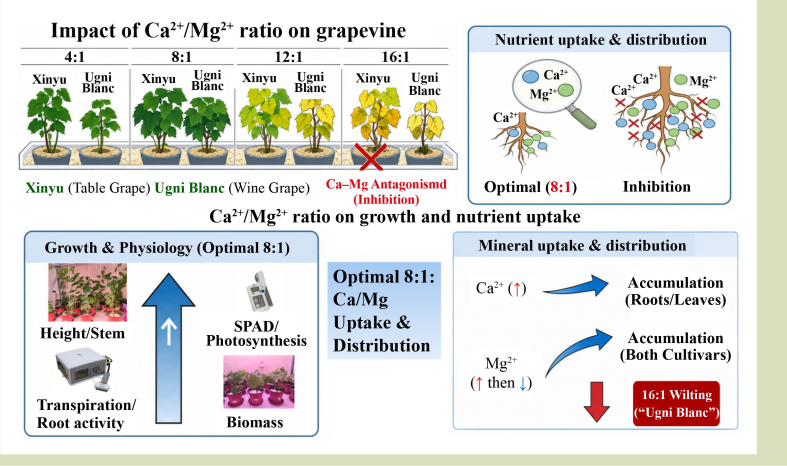
- Ca^{2+} to Mg^{2+} of 8:1 effectively promotes the growth and development of grapevines.
- This ratio increased plant biomass, root activity, leaf SPAD value, net photosynthetic rate and Mg^{2+} accumulation.
- Suboptimal Ca^{2+}/Mg^{2+} ratios (excessively low or high) reduced Mg availability via Ca–Mg antagonism, inhibiting plant growth.
- Grapevine cv. Xinyu cuttings were more adaptable than cv. Ugni Blanc to high Ca^{2+}/Mg^{2+} ratios.

Received December 30, 2025;
Accepted July 31, 2026.

Correspondence: symwj@163.com

Special Issue: Magnesium Nutrition in Soil–Plant Systems

GRAPHICAL ABSTRACT



ABSTRACT

To clarify the effects of the Ca^{2+}/Mg^{2+} ratio on the growth and mineral nutrient absorption and distribution of grapevine cuttings, a sand culture experiment was conducted using cv. Xinyu, a major table grape cultivar in Xinjiang, and cv. Ugni Blanc, a wine grape cultivar. Four Ca^{2+}/Mg^{2+} ratios (16:1, 12:1, 8:1 and 4:1) were set with a constant Mg^{2+} concentration of 1.25 mmol·L^{-1} to investigate the influence of suboptimal Ca–Mg ratio in calcareous soils. However, Ugni Blanc wilted and became necrotic at a 16:1 ratio; therefore, only the data from the other three treatments were analyzed. The results revealed that the biomass of both cultivars first increased then decreased with increasing Ca^{2+}/Mg^{2+} ratio, and the 8:1 ratio resulted in the optimal plant height, stem diameter, shoot dry weight, SPAD value and photosynthetic parameters. Ca and Mg ratio significantly affected the Ca and Mg concentrations in various plant parts (P < 0.05). With Ugni Blanc, the accumulations of Ca and Mg both increased to a peak and then declined. With Xinyu, Ca accumulation increased continuously, while Mg accumulation showed the same trend as that in Ugni Blanc. The 16:1 ratio caused obvious

Ca–Mg antagonism and reduced the Mg concentration in all the tissues, whereas the 8:1 ratio significantly increased the distribution rates of Ca and Mg. In conclusion, a Ca^{2+}/Mg^{2+} ratio of 8:1 was most favorable for the growth and Mg absorption and translocation of grapevine cuttings, and a suboptimal ratio inhibited plant growth through Ca–Mg antagonism.

© The Author(s) 2027. Published by Higher Education Press. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0)

1 Introduction

Magnesium is an essential plant nutrient. As the most abundant divalent cation in the cell cytoplasm, it is involved in a wide array of physiological and biochemical processes[1]. Its regulatory functions include ROS production and scavenging, respiration[2] and the tricarboxylic acid cycle[3]. Also, Mg, as the central atom of the chlorophyll porphyrin ring, is essential for photosynthesis[4], chlorophyll metabolism, and the transport of photoassimilates from source to sink organs[4,5]. Consequently, it directly influences crop yield formation and quality[6–8]. Grapevine (*Vitis vinifera*) is extensively cultivated in Xinjiang, China's largest grape-producing region[9]. With an annual output of 3.5 Mt, Xinjiang's vineyards account for 22% of total national production, underscoring the significant economic importance of crops[10]. Zlámalová et al.[11] demonstrated that the foliar application of Mg to grapevines, even in Mg-sufficient soil, can increase the fruit yield by up to 11.2%. Similarly, a study by Gu et al.[12] found that both drip irrigation and foliar application of Mg increased berry size and weight; increased yield; and promoted the accumulation of titratable acids, tannins, total phenols and anthocyanins, thereby enhancing both the visual quality and the intrinsic quality of the fruit.

The calcareous soils of Xinjiang are typically considered rich in available Mg (often > 300 $mg \cdot kg^{-1}$)[13], making Mg deficiency in plants relatively uncommon. However, recent field observations have shown that grapevines in the calcareous soil regions of southern Xinjiang can have subtle symptoms of Mg deficiency (e.g., interveinal chlorosis in old leaves), which may be attributed to high soil Ca^{2+} concentrations inhibiting Mg^{2+} uptake[13]. This phenomenon is consistent with reports of Mg deficiency in tomatoes[14] and cucumbers[15] in the calcareous soils of northern China, highlighting the need to investigate Ca–Mg interactions in grape production. Studies indicate that these deficiencies are not due to an absolute lack of Mg in the soil but rather a relative shortage[15]. High concentrations of Ca ions in calcareous soils inhibit plant uptake of Mg ions, diminishing their effectiveness.

Both Ca^{2+} and Mg^{2+} are absorbed by plant roots from the soil and translocated upward to the shoots via the xylem. A suboptimal ratio in their supply can lead to antagonistic interactions, inhibiting crop growth. This relationship is complex; for example, White et al.[16] reported a significant positive correlation between the Ca and Mg concentrations in plant shoots. Conversely, Yang[17] reported that high concentrations of either ion can induce antagonism, impairing normal plant development. The specific effects of these treatments appear to be crop dependent. Foliar application of Mg was found to reduce the Ca concentration in rice stems and leaves[18], whereas Mg fertilizer application significantly increased the yield, biomass, and accumulation of both Mg and Ca in wax gourd[19]. In grapevines, Mg application has been shown to increase Ca accumulation in berries under Mg deficiency stress[20].

Although recent research on the nutritional value of Mg in grapes has focused primarily on yield and quality improvements resulting from Mg fertilizer application[11,12,20], three critical knowledge gaps remain unaddressed. First, most studies have ignored Ca–Mg antagonism in calcareous soils, such as Xinjiang's Ca^{2+}/Mg^{2+}-dominant soil type, where high Ca^{2+} concentrations may reduce Mg^{2+} availability[13,15], yet the optimal Ca^{2+}/Mg^{2+} ratio for grapevine cuttings remains unclear. Second, existing research on Ca–Mg interactions has focused on other crops (e.g., rice[18] and wax gourd[19]) or has failed to consider cultivar-specific responses, despite significant differences in nutrient uptake between table and wine grapes. Third, the mechanisms underlying Ca–Mg antagonism in terms of nutrient translocation in grape (e.g., distribution patterns in roots, stems and leaves) have not been systematically elucidated, limiting targeted fertilizer application strategies.

In this study, cuttings of two major grapevine cultivars, cvs Ugni Blanc and Xinyu, were used as the experimental materials and a sand culture experiment was conducted to address the problem of a suboptimal Ca–Mg ratio in calcareous soils. Nutrient solutions with different Ca^{2+}/Mg^{2+} ratios were applied to investigate the effects of the Ca^{2+}/Mg^{2+} ratio on the uptake, accumulation, and distribution of Ca and Mg in grape cuttings. Also, the patterns of Ca–Mg interactions were revealed from the perspectives of cultivar specificity and organ nutrient distribution, providing a theoretical basis for coordinated nutrient management of grapevines under calcareous soil conditions.

2 Materials and methods

2.1 Plant materials and growth conditions

The experiments were conducted from July to October 2024 in a greenhouse at the College of Agriculture at Shihezi University. The facility was equipped with advanced environmental control systems that provided supplementary lighting, ventilation, and precise regulation of temperature and humidity. Throughout the experimental period, the conditions were maintained at an average temperature of 25 ± 1 °C (day) and 18 ± 1 °C (night) with a 16:8 h (light:dark) photoperiod. The plant materials consisted of one-year-old cuttings from two major Xinjiang grapevine cultivars: cv. Xinyu, a table grape, and cv. Ugni Blanc, a wine grape. Prior to transplantation, all the cuttings were selected for uniform growth: plant height (14.5 ± 2.0 cm), stem diameter (2.58 ± 0.57 mm) and weight (7.42 ± 0.46 g). The plants were cultivated using the sand culture method. The initial pH of the nutrient solution was adjusted to 6.5.

A modified Hoagland nutrient solution ($mmol \cdot L^{-1}$) was used as the basal formulation: $Ca(NO_3)_2$: 5.0, KNO_3: 5.0, KH_2PO_4: 1.0, $MgSO_4 \cdot 7H_2O$: 1.25, H_3BO_3: 4.6×10^{-2}, $CuSO_4$: 3.0×10^{-4}, $MnSO_4$: 1.9×10^{-2}, $ZnSO_4$: 7.6×10^{-4}, H_2MoO_4: 3.1×10^{-4}, and EDTA-Fe: 7.0×10^{-2}.

2.2 Experimental design

In the experiments, a sand culture method was used, and quartz sand with a 1–2 mm particle size was selected as the growth substrate for the plants. The pretreatment procedures included soaking in 3% hydrochloric acid to remove Ca impurities, repeated rinsing until no chloride ions remained, and autoclaving to obtain a pure, sterile and suitable substrate. The cuttings were rinsed with deionized water, and the surface moisture of their roots was blotted dry with filter paper before being weighed. On the basis of the growth status of both the shoot and root systems, cuttings with relatively uniform growth were selected again for transplantation into the pots. Plastic pots with an inner diameter of 18 cm and a height of 25 cm were used, each filled with 16 cm deep prerinsed quartz sand, and one cutting was transplanted into each pot. A modified Hoagland nutrient solution formula was used to maintain the original concentrations of all the other nutrients, and Ca and Mg concentrations in this solution were adjusted using $CaCl_2$ and $MgSO_4 \cdot 7H_2O$.

The Ca^{2+}/Mg^{2+} ratios in the nutrient solution were adjusted to 4:1, 8:1, 12:1 and 16:1. The Mg^{2+} concentration was maintained at 1.25 $mmol \cdot L^{-1}$ for all the treatments, whereas the Ca^{2+} concentrations were set to 5.0, 10.0, 15.0 and 20.0 $mmol \cdot L^{-1}$. $Ca(NO_3)_2$ solution was used for the 5.0 $mmol \cdot L^{-1}$ Ca^{2+} treatment, and $CaCl_2$ was used for the other treatments. $CaCl_2$ was selected for the medium- and high-Ca treatments to precisely regulate the Ca concentration without additional N input. The experiment consisted of four treatments, each with six replicates.

The experiment was established on 25 July 2024. Initially, the plants were irrigated with 1/2 strength Hoagland nutrient solution for a 2-week seedling establishment period. Starting from the 3rd week (10 August), each treatment was supplied with a full-strength nutrient solution corresponding to their respective Ca^{2+}/Mg^{2+} ratios until harvest on 10 October. During the cultivation period, each pot was supplied with 250 mL of the respective nutrient solution every 3 days (sufficient to saturate the quartz sand substrate without leaching). The pH was adjusted to 6.5 using 0.1 $mol \cdot L^{-1}$ NaOH and HCl solutions before each application. To prevent salt accumulation from affecting the experiment, the samples were leached with deionized water (2 L each time for thorough salt removal) every 10 days.

Following the initiation of treatments with different Ca^{2+}/Mg^{2+} ratios (starting from 10 August after seedling establishment), obvious wilting and necrosis symptoms were observed in Ugni Blanc cuttings during the second irrigation. These findings indicate that Ugni Blanc cannot adapt to a growth environment with an extremely high Ca^{2+}/Mg^{2+} ratio (16:1).

The different Ca^{2+}/Mg^{2+} ratios were established by fixing Mg^{2+} and varying the Ca^{2+} concentration. However, this approach inevitably resulted in differences in ionic strength among treatments, which is an inherent limitation of the experimental design. This variation may have influenced the root membrane potential, ion transport processes and nutrient diffusion, thereby potentially contributing to the observed effects in addition to the Ca^{2+}/Mg^{2+} ratio.

2.3 Determination items and methods

2.3.1 Determination of grapevine cutting growth indicators

Whole-plant sampling was conducted 60 days after treatment (10 October). The number of leaves per plant was recorded for each treatment. The plant height and stem diameter of each grapevine cutting were measured using a ruler and Vernier calipers, respectively. The roots, stems and leaves of each plant were subsequently separated, after which the fresh weights of the roots and aboveground parts (stems + leaves) were determined.

The roots, stems and leaves of the grapevines whose fresh weights were measured were separated again and placed in an oven for drying. After drying, the dry weights of the aboveground parts and roots were determined. The biomass of these parts of the grapevine plants was obtained on the basis of the dry weight and the sum of the biomass of the parts was considered the total plant biomass at the harvest stage.

2.3.2 Determination of photosynthetic physiological parameters and SPAD Values

On the 30th day (10 September) and 60th day (10 October) after the first application of the nutrient solution, leaves with a consistent light exposure direction and growth status were selected from each treatment on sunny days. The leaf relative chlorophyll concentration (SPAD value) was determined using a SPAD-502 chlorophyll meter (Konica Minolta, Tokyo, Japan). The net photosynthetic rate, intercellular CO_2 concentration, and transpiration rate of functional leaves were measured using a Yaxin-1102 Portable Photosynthesis and Transpiration Meter (Yaxin, Beijing, China).

2.3.3 Determination of root activity

The triphenyltetrazolium chloride (TTC) method was used for determination[21]. Exactly 0.3 g of the root tip segments of the grapevine cuttings was accurately weighed after the treatment period and placed in a 10 mL beaker. Phosphate buffer solution and 0.5% TTC solution were added to the beaker. After incubation at 37 °C in the dark for 3 h, triphenyl formazan was extracted by grinding with ethyl acetate and then brought to volume in a 10 mL volumetric flask for colorimetry.

TTC reduction intensity per unit root fresh weight was calculated as the TTC reduction amount / (root fresh weight × time).

$$I_{\mathrm{TTC}} = \frac{C_{\mathrm{TTC}} \times V}{W_{\mathrm{root}} \times t} \quad (1)$$

I_{TTC}: the TTC reduction intensity ($\mu g \cdot (g \cdot h)^{-1}$); C_{TTC}: the TTC concentration in the extract; V: the extract volume; W_{root}: the root fresh weight; t: the reaction time.

2.3.4 Determination of nutrient concentrations in grapevine tissues

After the stem, leaf and root samples were dried, ground and sieved, they were used for nutrient analysis. The Ca and Mg concentrations in the plants were determined by the dry ashing-inductively coupled plasma-optical emission spectrometry method. The nutrient accumulation in these plant parts was calculated as the product of the dry weight of each aboveground part and its corresponding nutrient concentration, and

$$D_{\mathrm{N}} = \frac{U_{\mathrm{organ}}}{U_{\mathrm{total}}} \times 100\% \quad (2)$$

D_{N}: the nutrient distribution (%); U_{organ}: the nutrient uptake of a specific organ; U_{total}: the total nutrient uptake per plant.

2.4 Data processing and analysis

The experimental data were statistically analyzed using SPSS 27.0 software and are given as mean ± standard error. The least significant difference test was used to test the significance of differences between different treatments within each cultivar at $P < 0.05$ and one-way analysis of variance was applied to evaluate the effects of treatments on plant growth indices, photosynthetic indices, and mineral ion concentrations in plant parts. Pearson correlation analysis was performed to clarify the correlations between the Ca and Mg concentrations in different plant parts under various Ca^{2+}/Mg^{2+} ratios. Differences were considered significant at $P < 0.05$.

3 Results

3.1 Effect of Ca^{2+}/Mg^{2+} ratio on the morphology

As given in Table 1, a Ca^{2+}/Mg^{2+} ratio of 8:1 had the most significant growth-promoting effect on both Ugni Blanc and Xinyu. With Ugni Blanc, the plant height in the 8:1 treatment was significantly greater than that in the 4:1 and 12:1 treatments by 2.7% and 43.5%, respectively. The stem diameter increased as the Ca^{2+}/Mg^{2+} ratio increased, with the 8:1 and 12:1 treatments giving significant increases compared to the 4:1 treatment by 58.6% and 65.1%, respectively; and the leaf number was greatest in the 4:1 treatment, with significant differences observed across all treatments. With Xinyu, plant height, stem diameter and leaf number all peaked at the 8:1 ratio being significantly lower in the 4:1, 12:1 and 16:1 treatments, with plant height reduced by 35.1% and stem diameter reduced by 36.7% in the 12:1 and 16:1 treatments, respectively, and the leaf number decreased by 22.6% in the 12:1 treatment.

Significant differences in cutting growth between Ugni Blanc and Xinyu were observed under different Ca^{2+}/Mg^{2+} ratios. Xinyu had a clear growth advantage, with greater plant height and more leaves, whereas a high Ca^{2+}/Mg^{2+} ratio mainly inhibited plant height development in cv. Ugni Blanc.

3.2 Effect of Ca^{2+}/Mg^{2+} ratio on the biomass

Comparisons of the effect of Ca^{2+}/Mg^{2+} ratio on the biomass of Ugni Blanc and Xinyu cuttings are given in Table 2. With Ugni Blanc, the root biomass in the 8:1 treatment was significantly greater than that in the 4:1 and 12:1 treatments by 20.9% and 8.1%, respectively. The stem biomass was the greatest in the 8:1 treatment, with biomass in the 4:1 and 12:1 treatments 12.5% and 15.0% lower, respectively. Leaf biomass was also the

Table 1 Effects of different Ca^{2+}/Mg^{2+} ratios on the morphological parameters of grapevine cuttings

Cultivar	Treatment	Plant height per plant (cm)	Stem diameter per plant (mm)	Number of leaves per plant
Ugni Blanc	4:1	44.3 ± 0.58a	4.13 ± 0.65b	16.3 ± 0.58a
	8:1	45.5 ± 0.71a	6.55 ± 0.04a	13.3 ± 0.58b
	12:1	31.7 ± 0.58b	6.82 ± 0.01a	15.0 ± 1.00a
Xinyu	4:1	53.0 ± 1.00b	4.28 ± 0.06b	15.7 ± 0.58b
	8:1	56.0 ± 1.00a	6.02 ± 0.09a	17.7 ± 0.58a
	12:1	36.3 ± 0.58d	4.19 ± 0.03b	13.7 ± 0.58c
	16:1	46.0 ± 1.73c	3.81 ± 0.05c	15.3 ± 0.58b

Note: Means followed by the same letter within each cultivar are not significantly different among Ca^{2+}/Mg^{2+} ratio treatments ($P < 0.05$).

Table 2 Effect of Ca^{2+}/Mg^{2+} ratio on the biomass (dry weight) of grapevine cuttings

Cultivar	Treatment	Root per plant (g)	Stem per plant (g)	Leaf per plant (g)	Root-shoot ratio
Ugni Blanc	4:1	2.11 ± 0.05c	4.27 ± 0.13b	6.31 ± 0.08b	0.19 ± 0.00a
	8:1	2.55 ± 0.05a	4.88 ± 0.13a	7.88 ± 0.20a	0.20 ± 0.01a
	12:1	2.36 ± 0.10b	4.15 ± 0.06b	5.29 ± 0.10c	0.25 ± 0.01a
Xinyu	4:1	4.62 ± 0.19b	5.00 ± 0.07a	6.74 ± 0.08a	0.39 ± 0.01c
	8:1	5.36 ± 0.11a	5.18 ± 0.08a	6.88 ± 0.29a	0.44 ± 0.01b
	12:1	4.50 ± 0.05b	4.38 ± 0.09c	4.46 ± 0.02d	0.51 ± 0.00a
	16:1	4.72 ± 0.09b	4.61 ± 0.13b	5.92 ± 0.02c	0.45 ± 0.01b

Note: Means followed by the same letter within each cultivar are not significantly different among Ca^{2+}/Mg^{2+} ratio treatments ($P < 0.05$).

greatest in the 8:1 treatment being 19.9% and 32.9% lower in the 4:1 and 12:1 treatments, respectively. Root-shoot ratio peaked in the plants in the 12:1 treatment.

With Xinyu, the root biomass in the 8:1 treatment was significantly greater than that in the 4:1, 12:1, and 16:1 treatments by 16.0%, 19.1% and 13.6%, respectively. Stem biomass in the 8:1 treatment was greater than in the 4:1, 12:1 and 16:1 treatments by 3.6%, 18.3%, and 12.4%, respectively. The leaf biomass in the 8:1 treatment was the greatest, being 2.1%, 54.3%, and 16.2% greater than in the 4:1, 12:1 and 16:1 treatments, respectively. Root-shoot ratio also peaked in the 12:1 treatment.

In most Ca^{2+}/Mg^{2+} treatments, Xinyu had greater root and stem biomass than Ugni Blanc, whereas leaf biomass had cultivar-dependent responses. Xinyu had a greater root-shoot ratio, indicating a greater proportion of biomass allocation to the roots. In contrast, the growth of Ugni Blanc was significantly inhibited under high Ca^{2+}/Mg^{2+} conditions.

3.3 Effect of Ca^{2+}/Mg^{2+} ratio on the photosynthesis

3.3.1 Effect of Ca^{2+}/Mg^{2+} ratio on the SPAD values

With Ugni Blanc cuttings (Fig. 1(a)), the leaf SPAD values tended to increase then decrease as the Ca^{2+}/Mg^{2+} ratio increased at both measurement times, reaching a maximum in the 8:1 treatment. The leaf SPAD value at 60 days after treatment was slightly greater than that at 30 days after treatment. At 30 days after treatment, compared with those in the 8:1 treatment, the SPAD values in the leaves in the 4:1 and 12:1 treatments was 0.80% and 8.50% lower, respectively. At 60 days after treatment, the leaf SPAD values of the plants in the 4:1 and 12:1 treatments were 4.93% and 6.41% lower than those of the plants in the 8:1 treatment, respectively.

With Xinyu cuttings (Fig. 1(b)), the leaf SPAD values peaked in the 8:1 and 12:1 treatments at 30 days and 60 days after treatment, respectively, and increased with time. At 30 days after treatment, compared to the 8:1 treatment, the SPAD values in the 4:1, 12:1 and 16:1 treatments were 7.72%, 13.3% and 5.73% lower, respectively. At 60 days after treatment, compared with those in the 12:1 treatment, the SPAD values in the 4:1, 8:1 and 16:1 treatments were 4.56%, 2.22% and 9.82% lower, respectively.

3.3.2 Effect of Ca^{2+}/Mg^{2+} ratio on photosynthesis indicators

Table 3 gives the effect of Ca^{2+}/Mg^{2+} ratio on the photosynthetic gas exchange parameters of Ugni Blanc and Xinyu. For both cultivars, the net photosynthetic rate and

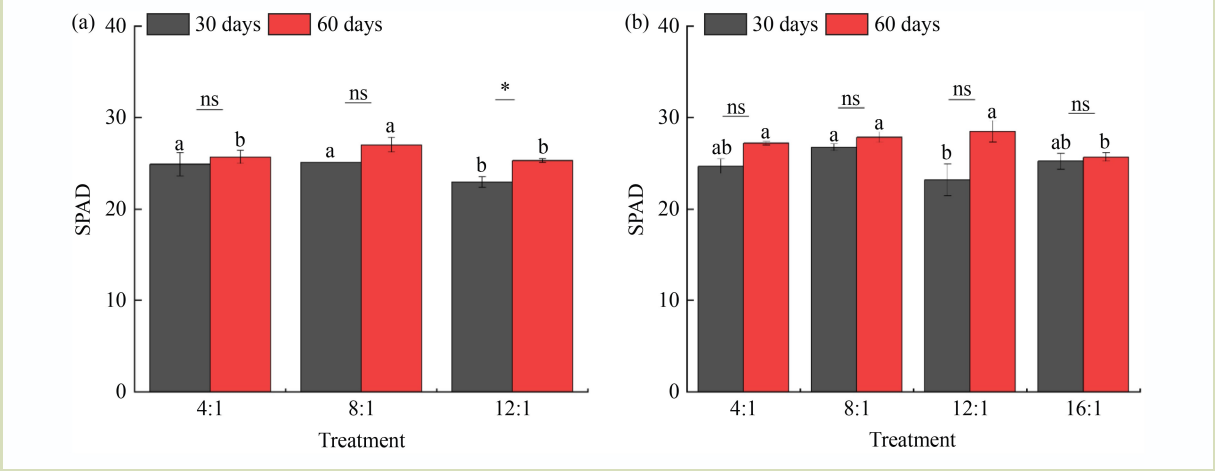


Fig. 1 Effect of Ca^{2+}/Mg^{2+} ratio on leaf SPAD values of the two grapevine cultivars at 30 and 60 days: (a) Ugni Blanc and (b) Xinyu. Values are given as mean ± standard error (n = 9). Data were analyzed by one-way analysis of variance, followed by the least significant difference test for multiple comparisons. The normality and homogeneity of variance were tested before analysis. Means with the same letter within sampling times are not significantly different and asterisks indicate significant differences between 30 and 60 days in the same treatment. *, $P < 0.05$ and ns, not significant.

Table 3 Effect of Ca^{2+}/Mg^{2+} ratio on the photosynthetic indicators of grapevine cuttings

Cultivar	Treatment	Net photosynthetic rate ($\mu mol \cdot m^{-2} \cdot s^{-1}$)	Intercellular CO_2 concentration ($\mu mol \cdot mol^{-1}$)	Transpiration rate ($mmol \cdot m^{-2} \cdot s^{-1}$)
Ugni Blanc	4:1	1.37 ± 0.07c	517 ± 23.0a	1.14 ± 0.09b
	8:1	1.95 ± 0.04a	562 ± 25.1a	1.93 ± 0.10a
	12:1	1.64 ± 0.09b	575 ± 22.1a	0.88 ± 0.04c
Xinyu	4:1	2.35 ± 0.95b	553 ± 59.6a	1.14 ± 0.04c
	8:1	4.95 ± 0.21a	525 ± 20.8a	2.63 ± 0.11a
	12:1	1.94 ± 0.66b	523 ± 1.4a	1.31 ± 0.08b
	16:1	2.47 ± 0.93b	493 ± 37.3a	1.15 ± 0.04c

Note: Means followed by the same letter within each cultivar are not significantly different among Ca^{2+}/Mg^{2+} ratio treatments ($P < 0.05$).

transpiration rate were greatest in the 8:1 Ca^{2+}/Mg^{2+} treatment. However, the significance of the treatment effects varied between the two cultivars. Specifically, compared with that of plants in the 8:1 treatment, the net photosynthetic rate of Ugni Blanc were 29.7% and 15.9% lower in the 4:1 and 12:1 treatments, respectively, whereas the transpiration rate was 40.9% and 54.4% lower, respectively.

Compared with those in the 8:1 treatment, the net photosynthetic rates in the 4:1, 12:1 and 16:1 treatments were 52.5%, 60.8% and 50.1% lower, respectively, and the transpiration rates were 56.7%, 50.2%, and 56.3% lower, respectively. In contrast, the intercellular CO_2 concentration tended to differ between the two cultivars, peaking for Ugni Blanc in the 12:1 treatment and for Xinyu in the 4:1 treatment; however, no significant differences were detected between treatments for either cultivar.

In the same Ca^{2+}/Mg^{2+} ratio treatments, the net photosynthetic rate and transpiration rate of Xinyu were greater than those of Ugni Blanc. In contrast, the net photosynthetic rate of Ugni Blanc was relatively stable across the Ca^{2+}/Mg^{2+} treatments, whereas its transpiration rate fluctuated considerably and decreased significantly in the 12:1 treatment. Overall, Ugni Blanc had weaker photosynthetic activity than Xinyu.

3.4 Effect of Ca^{2+}/Mg^{2+} ratio on root activity

For Ugni Blanc (Fig. 2(a)), root activity was slightly lower in the 4:1 and 12:1 treatments than the 8:1 treatment which was the highest. Compared to the 8:1 treatment, the root activity in the 4:1 and 12:1 treatments was 2.51% and 8.07% lower, respectively, but there were no significant differences between treatments.

With Xinyu cuttings (Fig. 2(b)), the root activity in the 8:1 treatment was greater than that in the other treatments. Compared to the 8:1 treatment, the root activity in the 4:1, 12:1, and 16:1 treatments were 6.15%, 16.2% and 5.20% lower, respectively. Compared with the other Ca^{2+}/Mg^{2+} treatments, the 12:1 treatment gave a significant reduction in root activity.

3.5 Effect of Ca^{2+}/Mg^{2+} ratio on the concentration, accumulation and distribution of Ca and Mg in different plant parts

3.5.1 Ca and Mg concentrations

The effect of Ca^{2+}/Mg^{2+} ratio on the Ca and Mg concentrations in the roots, stems and leaves of Ugni Blanc and Xinyu are given in Table 4. With Ugni Blanc, leaf Ca concentration was highest in the 8:1 treatment being 34.3% higher than in the 4:1 treatment. Stem Ca concentration was highest in the 4:1 treatment being 69.1% higher than in the 12:1 treatment. Leaf Mg concentration was highest in the 12:1 treatment, being 72.5% higher than in the 4:1 treatment. With Xinyu, the root Ca concentration was highest in the 16:1 treatment being 14.1% higher than in the 8:1 treatment. Stem Ca concentration was highest in the 12:1 treatment and leaf Ca in the 16:1 treatment, with the latter 134.6% higher than in the 12:1 treatment. Stem Mg concentration was highest in the 8:1 treatment.

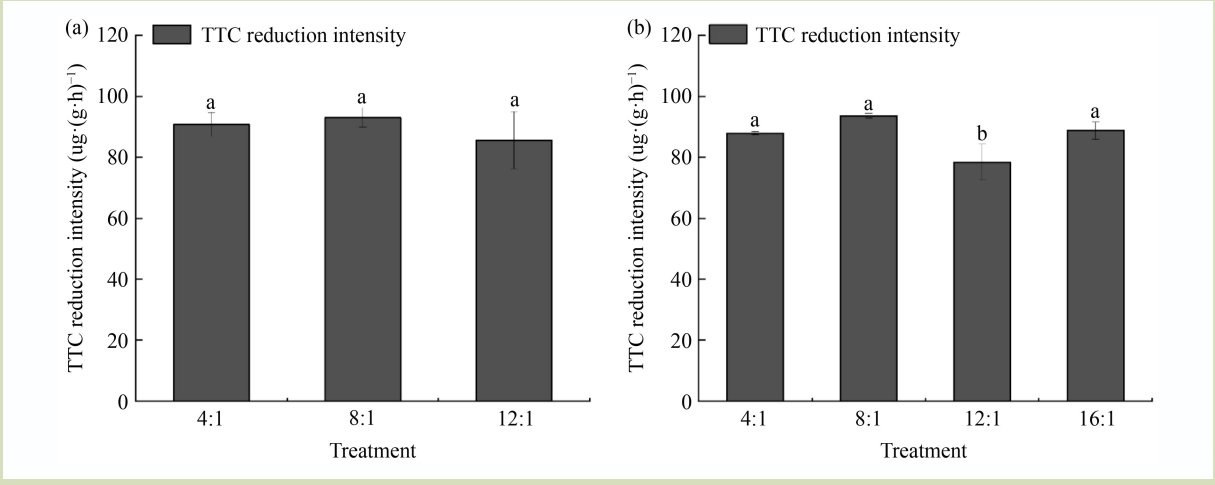


Fig. 2 Effect of Ca^{2+}/Mg^{2+} ratio on the root activity of two grapevine cultivars measured as the TTC reduction intensity: (a) Ugni Blanc and (b) Xinyu. Values are given as mean ± standard error (n = 3). Data were analyzed using one-way analysis of variance and means were compared using the least significant difference test. Prior to analysis, normality and homogeneity of variance were examined. Means with the same letter within columns are not significantly different between Ca^{2+}/Mg^{2+} ratio treatments ($P < 0.05$).

Table 4 Effect of Ca^{2+}/Mg^{2+} ratio on Ca and Mg concentrations in different parts of cuttings of two grapevine cultivars ($mg \cdot g^{-1}$)

Cultivar	Treatment	Ca			Mg		
		Root	Stem	Leaf	Root	Stem	Leaf
Ugni Blanc	4:1	14.9 ± 1.48a	27.4 ± 1.00a	25.4 ± 1.79b	3.83 ± 0.33a	4.51 ± 0.03a	4.04 ± 0.17b
	8:1	12.7 ± 0.66b	18.4 ± 0.43b	34.1 ± 0.59a	3.24 ± 0.03a	4.64 ± 0.13a	4.07 ± 0.13b
	12:1	16.4 ± 1.80a	16.2 ± 0.13c	32.8 ± 0.50a	2.77 ± 0.10b	2.80 ± 0.18b	6.97 ± 0.03a
Xinyu	4:1	18.1 ± 0.98ab	4.18 ± 0.23c	9.85 ± 0.34b	4.15 ± 0.13a	2.62 ± 0.07c	4.21 ± 0.07b
	8:1	17.0 ± 1.40b	7.91 ± 0.50b	8.14 ± 0.63c	3.31 ± 0.29b	4.95 ± 0.21a	2.98 ± 0.20d
	12:1	17.1 ± 0.41b	18.4 ± 0.44a	7.29 ± 0.24d	3.29 ± 0.16b	3.71 ± 0.17b	3.76 ± 0.23c
	16:1	19.4 ± 0.71a	4.02 ± 0.09c	17.1 ± 0.49a	2.88 ± 0.16c	2.13 ± 0.16d	4.49 ± 0.20a

Note: Means followed by the same letter within columns are not significantly different among treatments within a cultivar ($P < 0.05$).

3.5.2 Ca and Mg accumulation and distribution in Ugni Blanc

As the Ca^{2+}/Mg^{2+} ratio increased, the total Ca accumulation in Ugni Blanc (Fig. 3(a)) tended to first increase then decrease, being the highest in the 8:1 treatment. Compared to the 8:1 treatment, the total Ca accumulation in the 4:1 and 12:1 treatments were 14.6% and 26.2% lower, respectively.

Total Mg accumulation (Fig. 3(b)) also tended to first increase then decrease, peaking in the 8:1 treatment. Compared to the 8:1 treatment, the total Mg accumulation in the 4:1 and 12:1 treatments were 7.25% and 8.15% lower, respectively.

Ca and Mg in Ugni Blanc were mainly in the leaves, followed by the stems and roots. With increased Ca^{2+}/Mg^{2+} ratio, Ca accumulation (Fig. 4(a)) in leaves tended to increase then decrease, whereas Ca accumulation in stems and roots first decreased then increased. Consistent with the trend of Ca accumulation in leaves, the proportion of Ca in leaves also peaked in the 8:1 treatment. These findings indicate that an appropriate Ca^{2+}/Mg^{2+} ratio can promote the translocation and distribution of Ca to leaves, whereas an excessively high ratio may reduce the proportion of Ca in leaves.

In contrast to the distribution pattern of Ca, Mg accumulation

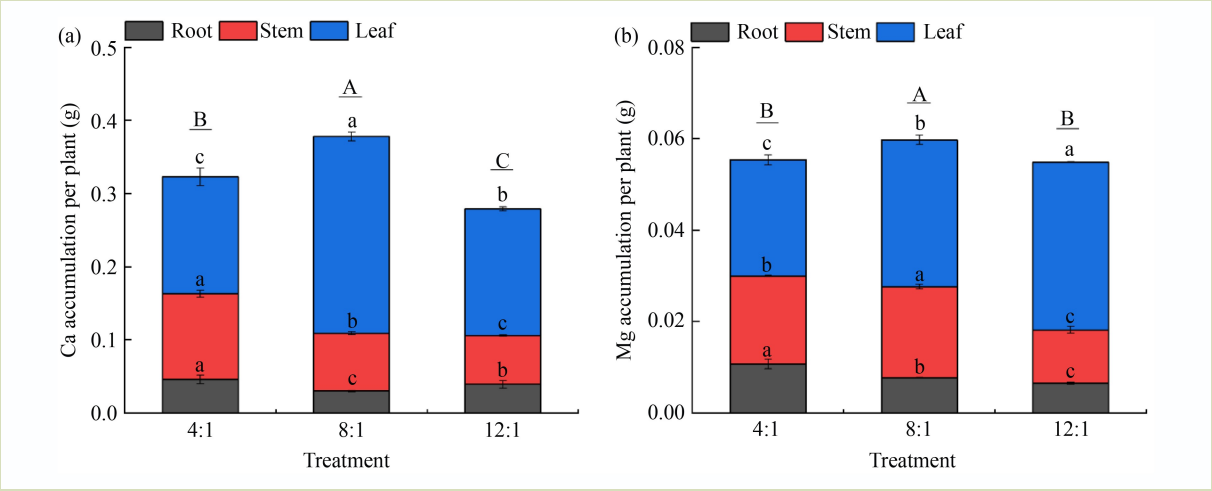


Fig. 3 Effect of Ca^{2+}/Mg^{2+} ratio on Ca and Mg accumulation in different parts of grapevine cv. Ugni Blanc: (a) Ca accumulation and (b) Mg accumulation. Values are given as mean ± standard error (n = 6). Data were analyzed using one-way analysis of variance and means were compared using the least significant difference test. Prior to analysis, the normality and homogeneity of variance were examined. Means followed by the same lowercase letter within columns are not significantly different between tissue types within Ca^{2+}/Mg^{2+} ratio treatments and those with the same uppercase letter are not significantly different in total accumulation between Ca^{2+}/Mg^{2+} ratio treatments ($P < 0.05$).

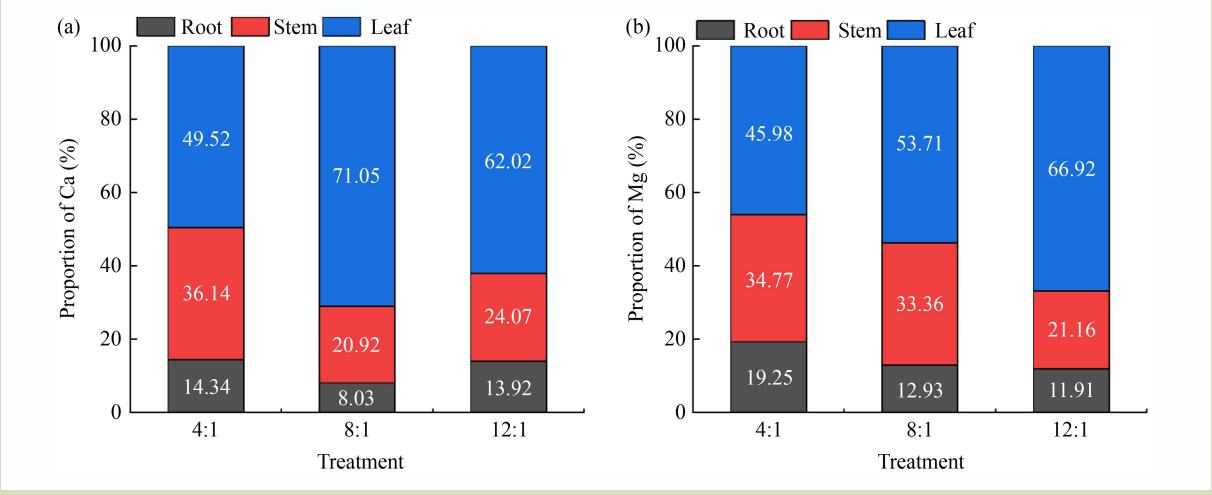


Fig. 4 Distribution patterns of Ca and Mg in different parts of grapevine cv. Ugni Blanc grown with different Ca^{2+}/Mg^{2+} ratios: (a) Ca proportion and (b) Mg proportion. Gray, red and blue represent the proportions in root, stem and leaf, respectively. Numbers within the bars indicate the proportion of Ca or Mg in to each plant part.

(Fig. 4(b)) in leaves gradually increased as the Ca^{2+}/Mg^{2+} ratio increased. The proportion of Mg in stems and roots decreased as the Ca^{2+}/Mg^{2+} ratio increased. In roots, the proportion of Ca first decreased then increased as the Ca^{2+}/Mg^{2+} ratio increased, whereas the degree of Mg accumulation in roots decreased progressively as the Ca^{2+}/Mg^{2+} ratio increased.

3.5.3 Ca and Mg accumulation and distribution in Xinyu

Total Ca accumulation in Xinyu (Fig. 5(a)) increased as the Ca^{2+}/Mg^{2+} ratio increased and reached a maximum in the 16:1 treatment. Compared to the 16:1 treatment, the total Ca accumulation in the 4:1, 8:1 and 12:1 treatments were 19.2%, 15.2% and 10.3% lower, respectively. Total Mg accumulation

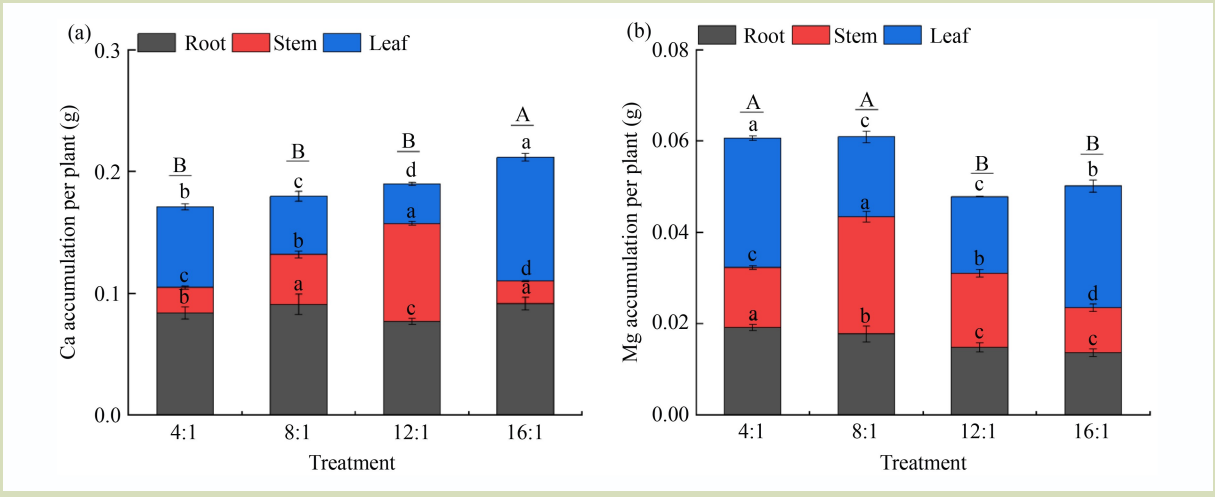


Fig. 5 Effect of Ca^{2+}/Mg^{2+} ratio on Ca and Mg accumulation in different parts of cv. Xinyu: (a) Ca accumulation and (b) Mg accumulation. Values are given as mean ± standard error (n = 6). Data were analyzed using one-way analysis of variance and means were compared using the least significant difference test at $P < 0.05$. Prior to analysis, the normality and homogeneity of variance were examined. Means followed by the same lowercase letter within columns are not significantly different between tissue type within Ca^{2+}/Mg^{2+} ratio treatments and those with the same uppercase letter are not significantly different in total accumulation between Ca^{2+}/Mg^{2+} ratio treatments.

(Fig. 5(b)) tended to first increase then decrease as the Ca^{2+}/Mg^{2+} ratio increased, peaking in the 8:1 treatment. Compared to the 8:1 treatment, total Mg accumulation in the 4:1, 12:1 and 16:1 treatments was 0.46%, 21.5%, and 17.7% lower, respectively.

As the Ca^{2+}/Mg^{2+} ratio increased from 4:1 to 16:1, the proportion of Ca (Fig. 6(a)) in roots was relatively high among the plant organs in most of the Ca^{2+}/Mg^{2+} ratio treatments and reached the maximum value in the 8:1 treatment. The proportion of Ca in the stems first increased then decreased as

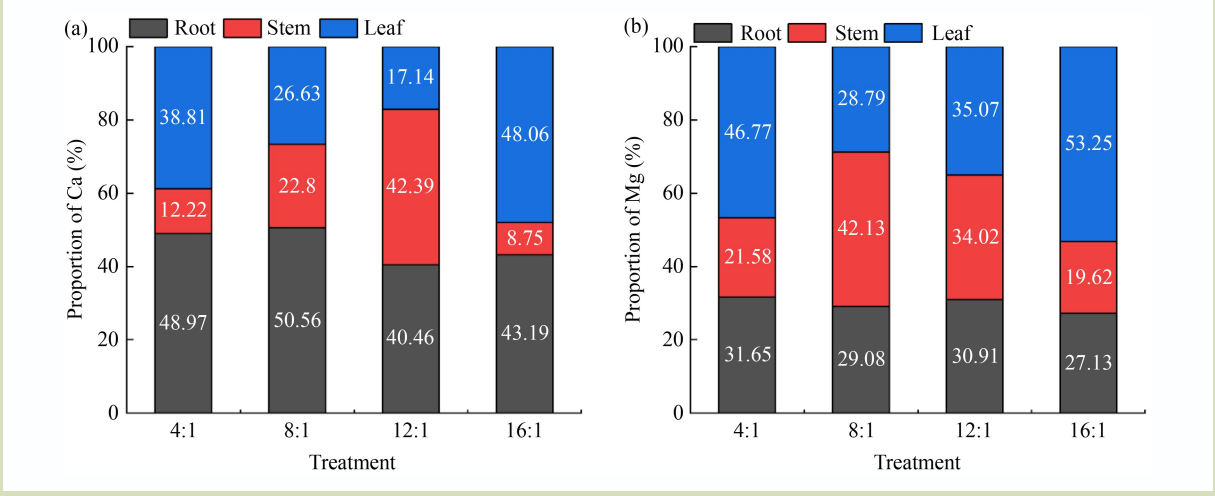


Fig. 6 Distribution patterns of Ca and Mg in different parts of grapevine cv. Xinyu grown in different Ca^{2+}/Mg^{2+} ratios; (a) Ca proportion and (b) Mg proportion. Gray, red and blue represent the proportions in root, stem and leaf, respectively. Numbers within the bars indicate the proportion of Ca or Mg in to each plant part.

the Ca^{2+}/Mg^{2+} ratio increased, peaking in the 12:1 treatment. In contrast, the distribution of Ca in the leaves tended to be opposite to that in the stems, with the highest value observed in the 16:1 treatment. For the distribution of Mg (Fig. 6(b)), Mg in the stems first increased then decreased as the Ca^{2+}/Mg^{2+} ratio increased, reaching a maximum in the 8:1 treatment. The proportion of Mg in leaves initially decreased then increased with increasing Ca^{2+}/Mg^{2+} ratios, peaking in the 16:1 treatment.

3.6 Correlation analysis between Ca and Mg concentrations in different parts of the grapevines and their growth and photosynthetic parameters

Pearson correlation analysis revealed clear differences between the two cultivars with respect to the relationships of Ca and Mg concentrations in different organs with growth and photosynthetic parameters. In Xinyu (Fig. 7(a)), root Ca was strongly positively correlated with leaf Ca, whereas stem Mg was strongly negatively correlated with leaf Mg. Also, stem Mg was positively correlated with Pn, Tr and stem diameter, whereas leaf Mg was mostly negatively correlated with these parameters. In Ugni Blanc (Fig. 7(b)), stem Ca was strongly negatively correlated with leaf Ca and root Mg was strongly positively correlated with stem Mg, whereas both were strongly negatively correlated with leaf Mg. In addition, leaf Ca and Mg were positively correlated with Pn, whereas root Mg was strongly positively correlated with plant height.

4 Discussion

4.1 Effect of Ca^{2+}/Mg^{2+} ratio on the growth, biomass and root activity

Ca and Mg are secondary macronutrients for plants and contribute to cell structure, photosynthesis and substance transport. For both Ugni Blanc and Xinyu (Table 1) cuttings, the plant height and stem diameter generally improved in the 8:1 Ca^{2+}/Mg^{2+} ratio treatment. This ratio is consistent with the recommended Ca^{2+}/Mg^{2+} ratio threshold (below 8:1) reported in cassava field trials[22], which has been proven to optimize nutrient uptake and increase yield. When the Ca^{2+}/Mg^{2+} ratio increased to 12:1 or 16:1, the plant height and number of leaves decreased significantly, indicating that a high-Ca environment may inhibit Mg absorption.

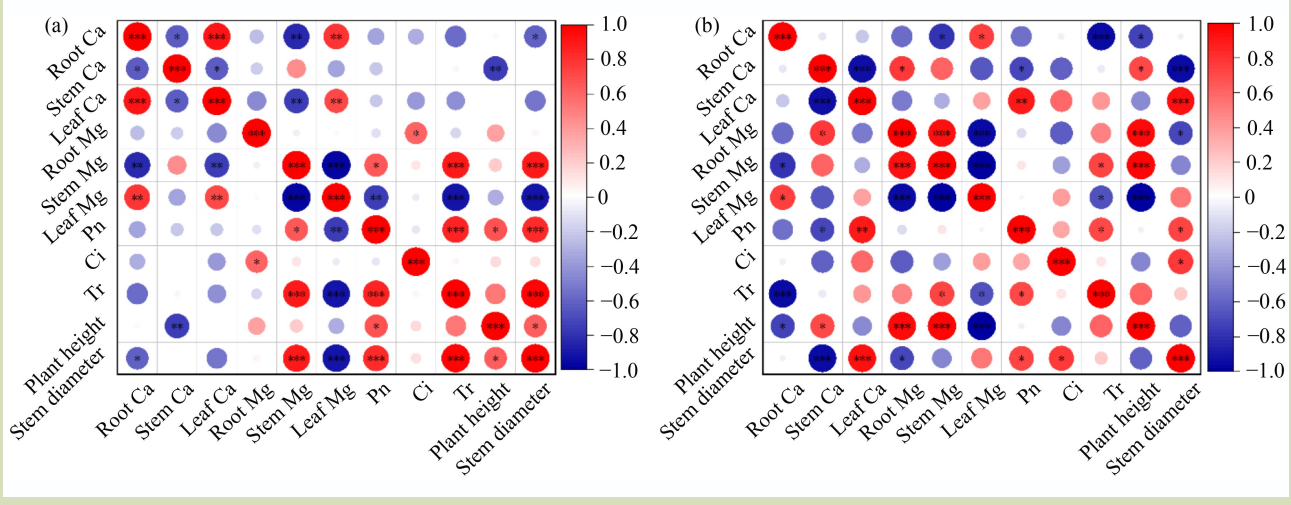


Fig. 7 Pearson correlation analysis between Ca and Mg concentrations in different parts of grapevine cuttings and their growth and photosynthetic parameters: (a) Xinyu and (b) Ugni Blanc. The rows and columns represent Ca and Mg concentrations in roots, stems and leaves, and photosynthetic parameters including net photosynthetic rate (Pn), intercellular CO_2 concentration (Ci) and transpiration rate (Tr), together with the growth traits plant height and stem diameter. Circle color indicates the direction and magnitude of the Pearson correlation coefficient (*r*), with red and blue indicating positive and negative correlations, respectively. Color intensity corresponds to the correlation coefficient ranging from –1 to 1. Circle size represents the absolute strength of the correlation, with larger circles indicating stronger correlations. *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$; and ns, not significant.

For both Ugni Blanc and Xinyu (Table 2) cuttings, the biomass (dry weight) of all the plant parts reached a maximum after the 8:1 Ca^{2+}/Mg^{2+} ratio treatment. An excessively high Ca^{2+}/Mg^{2+} ratio, especially 12:1, reduced root biomass in both cultivars, which may be associated with the antagonistic effect between Ca^{2+} and Mg^{2+} during nutrient absorption. When the Ca^{2+}/Mg^{2+} ratio is too high, excess Ca^{2+} may compete for Mg^{2+} uptake sites, resulting in relative Mg deficiency. This may impair chlorophyll synthesis and photosynthetic efficiency, ultimately restricting biomass accumulation[23]. These results are consistent with the findings of Meng et al.[24], who reported significantly reduced biomass in rice (*Oryza sativa*) and cucumber (*Cucumis sativus*) under low Mg conditions (0.01 mmol·L^{-1}). However, the biomass of the Xinyu cuttings was slightly better in the 16:1 Ca^{2+}/Mg^{2+} ratio treatment. Previous studies have demonstrated that the serpentine ecotype of *Erythrina crista-galli* can maintain normal cellular physiological functions by sequestering excess Mg^{2+} into vacuoles, thereby tolerating an extremely low Ca^{2+}/Mg^{2+} molar ratio (0.02)[25]. Although this scenario is opposite to the high Ca^{2+}/Mg^{2+} ratio condition in the present study, it similarly indicates that plants possess active physiological regulatory mechanisms to cope with ionic stress.

Ca and Mg interact with each other in plants, competing for certain intracellular transporters and transport proteins and thereby affecting plant growth[26]. Root activity is a key indicator of the ability of a plant to absorb nutrients from the soil. It not only directly reflects the absorption efficiency of mineral nutrients by roots but also indirectly reflects the overall growth and development status of plants[27]. In this experiment, root activity in both Ugni Blanc and Xinyu cuttings (Fig. 2) was relatively high in the 8:1 treatment. Both lower and higher Ca^{2+}/Mg^{2+} ratio treatments decreased the root activity in the grapevine cuttings, indicating that an appropriate Ca^{2+}/Mg^{2+} supply ratio is beneficial for maintaining vigorous root activity and thereby promoting water and nutrient uptake. These results are consistent with the findings of Ding et al.[28], who reported that a moderate Mg concentration (1.0 mmol·L^{-1}) was most beneficial for increasing root activity and bleeding sap intensity in rice, whereas both excessively high and low Mg concentrations had inhibitory effects.

4.2 Effect of Ca^{2+}/Mg^{2+} ratio on photosynthesis

The chlorophyll concentration directly affects the light-harvesting capacity of plant leaves, and a relatively high chlorophyll concentration facilitates the absorption and conversion of light energy[29,30]. Different Ca^{2+}/Mg^{2+} ratios affected the chlorophyll concentration of the plants (represented by SPAD values). The SPAD values of Ugni Blanc and Xinyu cuttings measured at different time points (Fig. 1) increased with increasing cultivation time, but no significant differences were detected between treatments. Although the relative chlorophyll concentration of both cultivars was generally greater in the 8:1 treatment, the differences between treatments were not always significant at all time points. However, the relative chlorophyll concentration decreased with higher Ca^{2+}/Mg^{2+} ratios. This pattern indicates that excessively high Ca^{2+}/Mg^{2+} ratios may induce relative Mg deficiency, thereby reducing leaf SPAD values. A study on betel nut (*Areca catechu*) seedlings revealed that SPAD values significantly decreased by 30.8% under Mg-deficient conditions[31], which is consistent with the findings in banana (*Musa acuminata*)[32] and faba bean (*Vicia faba*)[33].

Net photosynthetic rate is closely associated with the intercellular CO_2 concentration and transpiration rate[34,35]. In the 8:1 Ca^{2+}/Mg^{2+} ratio treatment, both grapevine cultivars generally have relatively high net photosynthetic and transpiration rates. However, the responses of the intercellular CO_2 concentration to changes in the Ca^{2+}/Mg^{2+} ratio differed between the two cultivars.

For Ugni Blanc (Table 3), the intercellular CO_2 concentration increased as the Ca^{2+}/Mg^{2+} ratio increased. Previous studies have shown that under stressful conditions, the net photosynthetic rate decreases significantly, whereas the intercellular CO_2 concentration increases, indicating that the limitation is mainly non-stomatal[36]. With Xinyu (Table 3), the intercellular CO_2 concentration decreased as the Ca^{2+}/Mg^{2+} ratio increased. In the high Ca^{2+} and Mg^{2+} treatments, the net photosynthetic rate gradually decreased, and the intercellular CO_2 concentration also tended to decrease, indicating that the photosynthetic response of this cultivar may differ from that of Ugni Blanc. This may be because the Ca^{2+} and Mg^{2+} concentrations in the nutrient solution were not high enough to cause antagonistic absorption of K^+, thereby inhibiting stomatal opening[37].

4.3 Effect of Ca^{2+}/Mg^{2+} ratio on the concentration, accumulation and distribution of Ca and Mg

As essential divalent cations in plants, the antagonistic effects

and synergistic balance between Ca and Mg are crucial for regulating plant nutrient use efficiency and growth and development[38]. Different Ca^{2+}/Mg^{2+} ratios significantly affected the distribution patterns of Ca and Mg in the two cultivars. As given in Table 4, in Ugni Blanc, the leaf Ca concentration was the highest at an 8:1 ratio, whereas at a 12:1 ratio, Ca was retained in the roots, and the leaf Mg concentration increased in a compensatory manner. In contrast, Xinyu was characterized mainly by Ca enrichment in the roots, and the overall Mg concentration decreased with increasing Ca^{2+}/Mg^{2+} ratio. Overall, under the present experimental conditions, the 8:1 ratio resulted in a better integrated effect and was more favorable for maintaining the relative balance of Ca and Mg uptake and distribution. However, this ratio should be regarded as a relatively suitable proportion on the basis of the trade-off among multiple indicators rather than the absolute optimum for all physiological processes.

Further analysis of the physiological responses of Ugni Blanc under different Ca^{2+}/Mg^{2+} ratios revealed that when the Ca^{2+}/Mg^{2+} ratio in the nutrient solution was 8:1, the total accumulation of Ca and Mg in Ugni Blanc (Fig. 3) reached the highest level among the tested treatments. Additionally, the distribution of Ca and Mg among the various organs followed a consistent pattern: leaves > stems > roots. This finding indicates that within the range of treatments used in the present study, the 8:1 ratio was most favorable for the absorption, translocation and distribution of the two nutrients in a coordinated manner.

When the Ca^{2+}/Mg^{2+} ratio in the nutrient solution increased moderately (from 4:1 to 8:1), the ability of Ca^{2+} to compete for absorption sites on the root surface increased, and its absorption and translocation to the aboveground parts subsequently increased, as manifested by the initial increase in leaf Ca accumulation[39]. However, when the Ca^{2+}/Mg^{2+} ratio is excessively high (16:1), excess Ca^{2+} may trigger feedback inhibition and potentially disrupt the permeability of root cell membranes, ultimately leading to a decrease in Ca^{2+} absorption and translocation efficiency. These findings are consistent with the phenomenon observed in which leaf Ca accumulation decreased from its peak. These results are consistent with findings in flue-cured tobacco (*Nicotiana tabacum*), where when the rhizosphere Ca^{2+} concentration exceeded a certain threshold, it significantly inhibited Mg absorption, resulting in a reduction in the leaf Mg concentration[40]. In the present study, the leaf Mg concentration of Ugni Blanc peaked at an 8:1 Ca^{2+}/Mg^{2+} ratio then decreased, indicating enhanced Ca–Mg antagonism under a higher Ca supply. In contrast, for Xinyu (Fig. 5), as the Ca^{2+}/Mg^{2+} ratio increased, the total plant Ca accumulation increased significantly, with Ca being distributed mainly in the roots. The total plant Mg accumulation tended to increase initially then decrease, and root Ca accumulation peaked at a Ca^{2+}/Mg^{2+} ratio of 8:1. This may be related to competition between Ca^{2+} and Mg^{2+} for absorption sites on the root surface. At this moderate ratio, the inhibitory effect of Mg on Ca absorption was weakened, and the absolute supply of Ca was not high enough to induce significant negative feedback regulation, thereby promoting the efficient absorption of Ca by the roots. Also, the relatively high Mg concentration in the stems at the 8:1 ratio also indicated that Mg translocation to aboveground parts and its temporary allocation in stems were more active at this supply ratio.

Further analysis indicated that an appropriate Ca^{2+}/Mg^{2+} ratio not only affects the competitive uptake of these two nutrients but is also related to the coordinated demand for Ca and Mg in leaves, which are key functional organs. Under an appropriate Ca^{2+}/Mg^{2+} ratio, both cultivars tended to exhibit coordinated accumulation of Ca and Mg in leaves (Figs. 3 and 5(b)), indicating that leaves, as important organs for maintaining cell wall stability[41], preserving the core structure of chlorophyll and supporting related enzyme activation, may have a relatively high demand for the coordinated balance of Ca and Mg. When the Ca^{2+}/Mg^{2+} ratio exceeded this range, plant growth began to decline, which may have occurred because excess Ca both weakened root ion uptake through competitive inhibition and induced relative Mg deficiency, thereby disturbing Mg-dependent energy metabolism. An insufficient adenosine triphosphate (ATP) supply may further affect the energy-dependent active transport of Ca^{2+} and reduce its ability to be transported to functional leaves[42].

Notably, Mg is an important regulator in the redistribution of Ca within plants. Guo et al.[43] reported that Mg can affect Ca absorption and its subcellular distribution in citrus plants, thereby regulating the Ca–Mg nutritional balance. The results of the present study are generally consistent with this view, indicating that a balanced Mg supply helps maintain effective Ca translocation to the leaves. Owing to the poor mobility of Ca in plants, its long-distance transport and transmembrane redistribution are active, energy-consuming processes dependent on ATP. As Mg is an activator of key enzymes

involved in energy metabolism, an adequate Mg supply can ensure ATP synthesis, thereby providing the energy required for the active transport of Ca[44]. Ding et al.[28] reported that Mg application in lettuce significantly increased the concentrations of various mineral nutrients, including Ca, which further supports the important role of Mg in promoting overall nutrient metabolism and energy supply. Also, the magnesium transporter (MGT) gene identified by Ge et al.[45], as an important component of high-affinity Mg^{2+} uptake in grapevines, may help alleviate the antagonistic effect of Ca^{2+} on Mg^{2+} absorption. By maintaining intracellular Mg^{2+} homeostasis, this process may reduce the accumulation of reactive oxygen species (ROS)[46] and help sustain the activities of Mg^{2+}-dependent enzymes such as ATP synthase and hexokinase, thereby supporting normal energy metabolism. Therefore, an excessively high Ca^{2+}/Mg^{2+} ratio may further restrict effective Ca transport and redistribution by intensifying competition between Ca^{2+} and Mg^{2+} at uptake and transport sites. However, these mechanistic explanations regarding ATP supply, ROS homeostasis, and transporter involvement are inferred mainly from previous studies and the response patterns observed in the present study rather than from direct measurements of gene expression, ATP metabolism, ROS, or transporter activity in this study.

4.4 Differential responses of grape cultivars to suboptimal Ca–Mg ratios and their adaptive mechanisms

This study demonstrated that two grapevine cultivars respond differently to suboptimal Ca–Mg ratios, with Xinyu being more adaptable to high-Ca conditions. Compared with Ugni Blanc, Xinyu maintained higher root activity (Fig. 2) and a greater root-to-shoot ratio (Table 2) with elevated Ca levels, indicating a better capacity to sustain belowground growth and nutrient uptake under stress. This enhanced tolerance may be associated with its genetic background, root physiological activity and strategies for mineral nutrient uptake and distribution. As a high-quality table grape cultivar that is independently bred in China, Xinyu is typically cultivated in relatively fertile soils, with the goal of breeding focuses more on fruit quality[47]. On this basis, the Casparian strip in its root endodermis may be more well developed, thereby partially restricting excessive Ca transport to the shoot and alleviating toxicity[48]. Correlation analysis (Fig. 7(a)) further revealed that, in Xinyu, the distribution of Mg between different organs was more closely related to growth and photosynthetic parameters. In particular, stem Mg was significantly positively correlated with the Pn, Tr and stem diameter, whereas leaf Mg was mostly negatively correlated. These findings indicate that Xinyu may maintain better physiological status and enhance adaptation to high-Ca environments by regulating Mg redistribution between transport organs and functional organs.

In contrast, Ugni Blanc had relatively weaker adaptability to high-Ca conditions. As an important wine grape cultivar, Ugni Blanc is more suited to nutrient-poor soils, and its buffering and regulatory mechanisms under high-Ca stress may be less effective than those of Xinyu. Notably, in the 16:1 treatment, Ugni Blanc plants were severely damaged and thus excluded, which itself indicates a relatively high sensitivity to extremely high Ca^{2+}/Mg^{2+} ratios. Correlation analysis (Fig. 7(b)) revealed that under high-Ca conditions, Ugni Blanc relies more heavily on mineral accumulation in leaves and Mg uptake by roots. Specifically, its photosynthetic activity is influenced mainly by leaf Ca and Mg status, while its growth is closely related to the root Mg supply. Under a suboptimal Ca–Mg ratio, once nutrient distribution among organs is disrupted, both growth and physiological functions are more likely to be constrained.

The present study used a sand culture system and precisely regulated the Ca^{2+}/Mg^{2+} ratios through nutrient solutions, thereby partially eliminating the interference caused by complex ion compositions and interactions in soil. This approach enabled the physiological responses of grapevine cuttings to different Ca^{2+}/Mg^{2+} ratios to be characterized under controlled conditions. The results indicated that under the conditions of this experiment, a Ca^{2+}/Mg^{2+} ratio of 8:1 resulted in relatively favorable growth and physiological metabolism, indicating that this ratio represented a suitable ratio of Ca and Mg supply under controlled experimental conditions.

Notably, this ratio was obtained as a theoretical physiological reference value in a controlled sand culture system and primarily reflects the response patterns of grapevine cuttings to different Ca^{2+}/Mg^{2+} ratios rather than the optimal application ratio under field production conditions. Nevertheless, this study provides a theoretical basis for further elucidating the mechanisms underlying Ca and Mg uptake and distribution in grapevine roots. Also, the findings provide preliminary insights and for subsequent field validation, Ca–Mg nutrient regulation studies under fertigation systems, and the further optimization of nutrient management strategies during grapevine nursery cultivation.

5 Conclusions

This study demonstrated that an appropriate Ca^{2+}/Mg^{2+} ratio, particularly 8:1, can effectively promote the growth and development of grapevines. Although not all the measured traits reached their maximum values under this ratio, the 8:1 treatment resulted in relatively favorable overall growth across multiple physiological indicators, including plant biomass, root activity, leaf SPAD value, net photosynthetic rate and Mg accumulation. In contrast, suboptimal Ca^{2+}/Mg^{2+} ratios (either excessively high or low) reduced Mg availability because of the antagonistic interaction between Ca^{2+} and Mg^{2+}, thereby inhibiting normal plant growth. Additionally, compared with Ugni Blanc, Xinyu had better adaptability to higher Ca^{2+}/Mg^{2+} ratios. Therefore, under the conditions of this experiment, Ca^{2+}/Mg^{2+} of 8:1 may be regarded as a favorable ratio on the basis of the integrated response of multiple traits rather than the absolute optimum for all physiological processes.

Acknowledgements

This research was supported by the National Natural Science Foundation of China (32360791).

Compliance with ethics guidelines

Qian Liu, Juan Wang, Heqi Zhang, Kunpeng Tan, and Changnan Yang declare that they have no conflicts of interest or financial conflicts to disclose. This article does not contain any studies on human or animal subjects performed by any of the authors.

REFERENCES

1. Ishfaq M, Wang Y Q, Yan M W, Wang Z, Wu L Q, Li C J, Li X X. Physiological essence of magnesium in plants and its widespread deficiency in the farming system of China. *Frontiers in Plant Science*, 2022, **13**: 802274
2. Jin X L, Ma C L, Yang L T, Chen L S. Alterations of physiology and gene expression due to long-term magnesium-deficiency differ between leaves and roots of *Citrus reticulata*. *Journal of Plant Physiology*, 2016, **198**: 103–115
3. Xie R R, Gao J J, Wu C Z, Guo J P, Cheng Q, Wang H L, Liang T M, Lu J J, Zheng C Y, Wu L Q, Chen L S, Chen S B, Li W Q. Magnesium deficiency limits electron transport efficiency and photosynthesis, and induces oxidative stress in tobacco. *Plant Science*, 2026, **362**: 112844
4. Toor M D, Adnan M, Ur Rehman F, Tahir R, Saeed M S, Khan A U, Pareek V. Nutrients and their importance in agriculture crop production: a review. *Indian Journal of Pure & Applied Biosciences*, 2021, **9**(1): 1–6
5. Guo W L, Nazim H, Liang Z S, Yang D F. Magnesium deficiency in plants: an urgent problem. *The Crop Journal*, 2016, **4**(2): 83–91
6. He Z B, Wang Z, Hao J X, Wu Y F, Liu H J. The use of magnesium fertilizer can improve the nutrient uptake, yield, and quality of rice in Liaoning Province. *Agronomy*, 2024, **14**(3): 639
7. Ahmed N, Zhang B G, Bozdar B, Chachar S, Rai M, Li J, Li Y Q, Hayat F, Chachar Z, Tu P F. The power of magnesium: unlocking the potential for increased yield, quality, and stress tolerance of horticultural crops. *Frontiers in Plant Science*, 2023, **14**: 1285512
8. Santos M, Pereira S, Ferreira H, Sousa J R, Vilela A, Ribeiro C, Raimundo F, Egea-Cortines M, Matos M, Gonçalves B. Optimizing sweet cherry attributes through magnesium and potassium fertilization. *Horticulturae*, 2024, **10**(8): 881
9. Liu K H, Zhang R, Gao Y T, Zhang H J, Wen J, Xue L, Li Q Q. Present situation analysis and development countermeasures of grape industry in China. *China Fruits*, 2024, (7): 132–138 (in Chinese)
10. National Bureau of Statistics of China. China Statistical Yearbook. Beijing: *China Statistics Press*, 2024 (in Chinese)
11. Zlámalová T, Elbl J, Baroň M, Bělíková H, Lampíř L, Hlušek J, Lošák T. Using foliar applications of magnesium and potassium to improve yields and some qualitative parameters of vine grapes (*Vitis vinifera* L.). *Plant, Soil and Environment*, 2015, **61**(10): 451–457
12. Gu C F, Zhang L, Liu S Q, Tian C, Wang J, Wang R. Study on application effect and economic benefit of magnesium fertilizer on wine grape. *Soil and Fertilizer Sciences in China*, 2022, (2): 128–133 (in Chinese)
13. Bai Y L, Jin J Y, Yang L P. Study on the content and distribution of soil available magnesium and foreground of magnesium fertilizer in China. *Soils and Fertilizers*, 2004, (2): 3–5 (in Chinese)

14. Yan B. Mechanism of Magnesium Deficiency and the Effects of Magnesium on Tomato in Greenhouse That Built on Calcareous Soil. Yangling: *Northwest A&F University*, 2015 (in Chinese)
15. Li H X. The Reason of Magnesium Deficiency in Tomato Grown in Calcareous Soil under Solar Greenhouse. Yangling: *Northwest A&F University*, 2018 (in Chinese)
16. White P J, Broadley M R, El-Serehy H A, George T S, Neugebauer K. Linear relationships between shoot magnesium and calcium concentrations among angiosperm species are associated with cell wall chemistry. *Annals of Botany*, 2018, **122**(2): 221–226
17. Yang Z Q. Effects of Ca-magnesium fertilizer on tomato yield, quality and nutrient absorption. *Soils and Fertilizers*, 1994, (2): 14–18 (in Chinese)
18. Hao J X. Effects of Spraying Magnesium Fertilizer on Nutrient, Yield and Quality of Rice. Shenyang: *Shenyang Agricultural University*, 2022 (in Chinese)
19. Lian Y X, Li J, Li J L, Chang J J, Li Z, Chen L, Zhang B G. Effects of magnesium fertilizer dosage on yield and the Ca, Mg absorption and distribution in wax gourd [*Benincasa hispida* (Thunb.) Cogn.]. *Journal of Plant Nutrition and Fertilizers*, 2025, **31**(8): 1631–1643 (in Chinese)
20. Ma X L, Wang J, Guan S, Lü X L, Xiang P W, Ren J Q. Effects of magnesium application on the mineral nutrition absorption by grapes with magnesium deficiency. *Soil and Fertilizer Sciences in China*, 2017, (6): 117–121 (in Chinese)
21. Zhu X Y, Liang M, Ma Y. A review report on the experiments for the determination of root activity by TTC method. *Guangdong Chemical Industry*, 2020, **47**(6): 211–212 (in Chinese)
22. Nyein E Z, Sasirat M, Kheoruenromme I, Anusontpornperm S, Thanachit S. A response of cassava to magnesium supplementation and three-year applications of gypsum in a tropical loamy sand soil supplementation. *Communications in Soil Science and Plant Analysis*, 2025, **56**(20): 2906–2918
23. Tian X Y, Cao H R, Li Q R, Wu X, Liu B S, Guo Z L, Zhong X B, Chen Z C. OsCAX1a-dependent Ca-Mg balance is required for optimal growth in rice. *Rice*, 2025, **18**(1): 108
24. Meng X S, Bai S, Wang S Y, Pan Y H, Chen K H, Xie K L, Wang M, Guo S W. The sensitivity of photosynthesis to magnesium deficiency differs between rice (*Oryza sativa* L.) and cucumber (*Cucumis sativus* L.). *Frontiers in Plant Science*, 2023, **14**: 1164866
25. Palm E, Guidi Nissim W, Colasurdo G, Van Volkenburgh E. Inducible tolerance to low Ca: Mg in serpentine ecotype of *Erythranthe guttata*. *Journal of Plant Physiology*, 2024, **303**: 154355
26. Liu Y, Lei J, Hu C X, Tan Q L, Zhuang M L, Sun X C, Wu S W. Effects of combined application of calcium and magnesium on yield, quality and calcium and magnesium nutrients of 'Guanxi' Pomelo. *Journal of Southern Fruits of China*, 2023, **52**(3): 15–21 (in Chinese)
27. Wang Y S, Thorup-Kristensen K, Jensen L S, Magid J. Vigorous root growth is a better indicator of early nutrient uptake than root hair traits in spring wheat grown under low fertility. *Frontiers in Plant Science*, 2016, **7**: 865
28. Ding Y C, Jiao X Y, Nie D, Li L J, Wang S T. Effects of application of nitrogen and magnesium fertilizers on yield and quality of lettuce. *Northern Horticulture*, 2012, (12): 167–169 (in Chinese)
29. Ye X L, Gao Z Y, Xu K, Li B L, Ren T, Li X K, Cong R H, Lu Z F, Cakmak I, Lu J W. Photosynthetic plasticity aggravates the susceptibility of magnesium-deficient leaf to high light in rapeseed plants: the importance of Rubisco and mesophyll conductance. *The Plant Journal*, 2024, **117**(2): 483–497
30. Ma Y Y, Wang Z, Zhou B Y, Yang W H, Wang Y P. Salicylic acid improving salinity tolerance by enhancing photosynthetic capacity, osmotic adjustment and maintenance of Na^+/K^+ homeostasis in faba bean seedlings. *Chemical and Biological Technologies in Agriculture*, 2025, **12**(1): 89
31. Li J, Cao X M, Liu L Y, Niu Q X. Effects of different magnesium nutrition levels on photosynthetic characteristics and chloroplast ultrastructure of areca palm seedlings. *Journal of Plant Nutrition and Fertilizers*, 2019, **25**(11): 1949–1956 (in Chinese)
32. Kan B L, Yang Y, Du P M, Li X P, Lai W J, Hu H Y. Chlorophyll decomposition is accelerated in banana leaves after the long-term magnesium deficiency according to transcriptome analysis. *PLoS One*, 2022, **17**(6): e0270610
33. Parisa D, Repnik U, Abdalla M A, Mühling K H. Leaf anatomical adaptation and chloroplast ultrastructure changes upon magnesium foliar application of faba bean (*Vicia faba* L.) grown under drought stress. *Journal of Plant Nutrition and Soil Science*, 2025, **188**(1): 78–91
34. Ye M, Wu M, Zhang Y, Wang Z Y, Zhang H, Zhang Z J. Physiological factors limiting leaf net photosynthetic rate in C_3 crops like rice and approaches for improving it. *Agronomy*, 2022, **12**(8): 1830
35. Rogiers S Y, Greer D H, Moroni F J, Baby T. Potassium and magnesium mediate the light and CO_2 photosynthetic responses of grapevines. *Biology*, 2020, **9**(7): 144
36. Weng X H, Li H, Ren C S, Zhou Y B, Zhu W X, Zhang S Z, Liu L Y. Calcium regulates growth and nutrient absorption in poplar seedlings. *Frontiers in Plant Science*, 2022, **13**: 887098
37. Feng T Y, Jiang H, Kong X, Zhao Z Y, Ma Y T, Zhai B N. Diagnosis of maladjustment of calcium and magnesium in young apple trees and its effect on growth and development. *Northern Horticulture*, 2021, (20): 18–26 (in Chinese)
38. Tang R J, Luan S. Regulation of calcium and magnesium homeostasis in plants: from transporters to signaling network. *Current Opinion in Plant Biology*, 2017, **39**: 97–105

39. Kwon T R, Shaheed Siddiqui Z, Harris P J C. Effects of supplemental calcium on ion accumulation, transport and plant growth of salt Sensitive *Brassica Rapa* Landrace. *Journal of Plant Nutrition*, 2009, **32**(4): 644–667
40. Feng X W, Huang Y, Wu G L, Gou J Y, Peng Y L. Effects of different calcium concentrations on growth and magnesium absorption of flue-cured tobacco. *Crops*, 2021, (3): 190–194 (in Chinese)
41. Gao Q Y, Xiong T T, Li X P, Chen W X, Zhu X Y. Calcium and calcium sensors in fruit development and ripening. *Scientia Horticulturae*, 2019, **253**: 412–421
42. Chen Z C, Peng W T, Li J, Liao H. Functional dissection and transport mechanism of magnesium in plants. *Seminars in Cell & Developmental Biology*, 2018, **74**: 142–152
43. Guo J X, Jiao Y L, Wang Y W, Hu W L, Jia Y M, Huang Z R, Yang L T, Chen L S. Regulation of magnesium and calcium homeostasis in citrus seedlings under varying magnesium supply. *Plant Physiology and Biochemistry*, 2023, **204**: 108146
44. Cakmak I, Kirkby E A. Role of magnesium in carbon partitioning and alleviating photooxidative damage. *Physiologia Plantarum*, 2008, **133**(4): 692–704
45. Ge M Q, Zhong R, Sadeghnezhad E, Hakeem A, Xiao X, Wang P P, Fang J G. Genome-wide identification and expression analysis of magnesium transporter gene family in grape (*Vitis vinifera*). *BMC Plant Biology*, 2022, **22**(1): 217
46. Livigni S, Lucini L, Sega D, Navacchi O, Pandolfini T, Zamboni A, Varanini Z. The different tolerance to magnesium deficiency of two grapevine rootstocks relies on the ability to cope with oxidative stress. *BMC Plant Biology*, 2019, **19**(1): 148
47. Wang J F, Bai L, Jiang B, Ju Y L, Li R, Zhu P P, Wang L J. Evaluation of the introduction performance of Yongyou No.1 and Xinyu grape varieties in the Weinan area of Shaanxi Province. *Anhui Agricultural Science Bulletin*, 2025, **31**(21): 35–39 (in Chinese)
48. Li Y H, Tian S F, Ma C, Niu L L, Yuan Y, Li Q Y. Analysis on root anatomical structure and hydraulic characteristics in different grape varieties. *Journal of Fruit Science*, 2021, **38**(5): 714–724 (in Chinese)