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Comparison of Leaf Yield and Quality in Mulberry (*Morus alba* L.) Cultivars under Soil-Based and Soilless Greenhouse Cultivation

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ABSTRACT

The objective of this study was to compare the yield and leaf quality of three mulberry cultivars (Kenmochi, Ichinose, and Kanz) under soil-based and hydroponic cultivation systems in the research greenhouse of the University of Guilan. The experiment was arranged as a completely randomized design in factorial (2*3) arrangement. Two cultivation system (soil vs. soilless) and three mulberry cultivars (Kenmochi, Kanz, Ichinose) were evaluated in the present study. Each treatment was replicated four times, with four seedlings per treatment. Data were collected across three harvests and subjected to statistical analysis. Results indicated that the soil-based system was superior in volumetric traits (fresh leaf weight, leaf number, leaf area, and total dry weight). In contrast, the hydroponic system significantly improved leaf quality indices and water use efficiency. The highest nitrogen (10.4%) and protein content (25.63%) were recorded in Kanz under hydroponic cultivation. Ichinose maintained leaf quality under hydroponics without notable yield reduction, whereas Kanz exhibited the highest quantitative performance in soil. Kenmochi provided a balanced combination of leaf quality and growth stability under hydroponics. A key finding revealed that hydroponic cultivation enhanced water use efficiency by 36.7% compared to soil-based cultivation. Overall, hydroponics, combined with appropriate cultivar selection (Kanz for superior nutritional quality and Kenmochi for stability), represents the optimal approach for producing high-quality leaves with improved water efficiency for silkworm feeding. This study offers a practical model for advancing toward smart and sustainable agriculture in the sericulture industry.

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1. Introduction

The sericulture industry in Iran, particularly in northern provinces such as Guilan, plays a vital role in rural economies and the export of high-value products. A persistent challenge, however, is the shortage of mulberry (*Morus alba* L.) leaves with sufficient nutritional quality all around the year. Although soil-based cultivation remains the traditional practice, the rising demand for superior-quality leaves has underscored the need for advanced systems such as hydroponics. Studies conducted outside Iran have demonstrated that soilless cultivation can markedly improve the nutritional profile of mulberry leaves (Hosseini et al., 2021).

Mulberry leaves with nitrogen concentrations of approximately 3% and protein levels exceeding 18% are considered optimal for silkworm nutrition. While soil-based cultivation has historically dominated leaf production, increasing demand and constraints in natural resources such as water and land have shifted research toward modern approaches, including hydroponics. For example, evaluations of mulberry performance under cocopeat and hydroponic systems revealed that soilless cultivation significantly enhances leaf quality, particularly when nitrogen supply is optimized (Chen et al., 2023). These findings suggest that soilless systems may represent a viable alternative to conventional soil-based cultivation.

The composition of nutrient solutions and the pH of the growing medium are critical determinants of success in soilless systems, especially for mulberry production in the sericulture industry (Savvas & Passam, 2022). A comparative study in China reported that soil-based cultivation yielded superior volumetric traits (fresh leaf weight and leaf number), whereas soilless cultivation improved leaf quality (Zhang et al., 2021). Cultivar-specific responses have also been widely documented. In Japan, Li et al. (2019) found that the Ichinose cultivar exhibited the highest nitrogen and protein content under soilless conditions, making it particularly suitable for silkworm feeding. Conversely, Rahmani et al. (2021) observed that under soil-based conditions, the Kanz cultivar achieved greater volumetric performance but comparatively lower leaf quality.

In the present study, three common mulberry cultivars in Guilan Province (Kenmochi, Ichinose, and Kanz) were evaluated under soil-based and soilless cultivation systems. Quantitative traits (leaf weight, leaf number), physiological indices (SPAD chlorophyll index), biochemical parameters (nitrogen, protein, dry matter), and growth traits (stem length, stem diameter, and number of lateral branches) were measured. The primary objective was to identify the most suitable cultivar for each cultivation system, thereby enabling the production of nutritionally superior mulberry leaves under greenhouse conditions in Iran.

2. Materials and Methods

The experiment was conducted in the research greenhouse of the University of Guilan (Guilan Province, Iran) during the growth period of white mulberry (*Morus alba*) seedlings from spring to autumn 2025. The greenhouse environment was maintained at an average temperature of 22–28 °C with relative humidity ranging from 60–75%. Three mulberry cultivars were evaluated: Kenmochi (disease-resistant, vigorous growth), Ichinose (Japanese cultivar, high leaf quality), and Kanz (local cultivar, high volumetric yield). Seedlings originated from one-year-old tissue culture plants with an average height of 75 cm and stem diameter of 23.5 mm.

Seedlings were initially grown in pots containing cocopeat and perlite. They were carefully removed without root damage and transplanted either into soil or into pots containing cocopeat and perlite at equal volumetric ratios. Standard agricultural soil with loamy texture and adequate drainage was used for soil-based cultivation. Fertilization consisted of well-decomposed farmyard manure (10 t ha^{-1}) combined with 150 kg ha^{-1} chemical fertilizer (N-P-K; 20-20-20) applied in three stages. For soilless culture, pots were filled with cocopeat and perlite (1:1 v/v) and supplied with a standard nutrient solution (Table 1) formulated for mulberry (Ahmadi et al., 2022). Irrigation and nutrient application were performed manually at 24-hour intervals according to plant water requirements. Drainage water were collected and measured to calculate water use efficiency according field capacity of potes.

Measurements were recorded at two-month intervals and included morphological traits (leaf number, leaf area, fresh and dry weight of leaves and stems, number of lateral branches, plant height, and stem diameter), biochemical traits (nitrogen concentration, protein content, and dry matter percentage), and physiological indices (chlorophyll content assessed using a SPAD chlorophyll meter). Leaf area were measured using leaf area meter (G.C.L. BUBBLE ETCH TANKS). The digestion method and the Kjeldahl apparatus were used to measure the amount of nitrogen and multiply in 6.25 to calculate protein content (Emami, 1996).

The experiment was arranged as a completely randomized design in factorial (2*3) arrangement. Two cultivation system (soil vs. soilless) and three mulberry cultivars (Kenmochi, Kanz, Ichinose) were evaluated in the present study. Each treatment was replicated four times, with four seedlings per treatment. Data were analyzed statistically using SAS software after verification of normality. To calculate the dry weight of leaves and stems, each sample was placed in a paper envelope and an oven at 70°C for three days. SPAD measurements were reported as average of 3 readings per per plant.

Table 1. Composition and concentration of nutrients in the soilless culture solution used for white mulberry cultivation

Nutrient	Fertilizer source	Concentration (mg L^{-1})
Nitrogen (N)	Potassium nitrate (KNO_3)	200
	Calcium nitrate ($Ca(NO_3)_2 \cdot 4H_2O$)	180
Phosphorus (P)	Monopotassium phosphate (KH_2PO_4)	50
Potassium (K)	Potassium nitrate (KNO_3)	210
	Potassium sulfate (K_2SO_4)	100
Calcium (Ca)	Calcium nitrate ($Ca(NO_3)_2 \cdot 4H_2O$)	200
Magnesium (Mg)	Magnesium sulfate ($MgSO_4 \cdot 7H_2O$)	50
Sulfur (S)	Potassium sulfate (K_2SO_4)	45
	Magnesium sulfate ($MgSO_4 \cdot 7H_2O$)	20
Iron (Fe)	Iron chelate (Fe-EDDHA)	2
Manganese (Mn)	Manganese sulfate ($MnSO_4 \cdot H_2O$)	0.5
Zinc (Zn)	Zinc sulfate ($ZnSO_4 \cdot 7H_2O$)	0.05
Copper (Cu)	Copper sulfate ($CuSO_4 \cdot 5H_2O$)	0.02
Boron (B)	Boric acid (H_3BO_3)	0.5
Molybdenum (Mo)	Ammonium molybdate ($(NH_4)_6Mo_7O_{24} \cdot 4H_2O$)	0.1

3. Results

The analysis of variance (Table 2) demonstrated that cultivar, cultivation system, and their interaction exerted significant effects on leaf number across all three harvests as well as the cumulative total, at the 1% probability level. Mean separation using Tukey's test revealed that the Kanz cultivar under soil cultivation exhibited the highest leaf production capacity, with 61 leaves per plant in the first harvest, 58 in the second, 55 in the third, and a total of 174 leaves per plant (Table 3). This performance reflects the strong genetic potential of Kanz for lateral bud initiation. In contrast, the Ichinose cultivar in soil cultivation produced the lowest leaf numbers, averaging 25.08 leaves in the first harvest, 24 in the second, 23 in the third, and a cumulative total of 72.08 leaves per plant (Table 3). These results suggest that Ichinose tends to allocate assimilates toward the maintenance of existing foliage rather than the formation of new lateral branches.

Table 2. Analysis of variance (ANOVA) results for the effects of cultivar and cropping system on mulberry leaf number across different harvests

Source of variation	df	Leaf number – 1st harvest	Leaf number – 2nd harvest	Leaf number – 3rd harvest	Total leaf number
Cultivar	2	7849.55**	7200.34**	6800.45**	122.07**
Cropping system	1	9800.23**	9200.45**	8900.56**	74.87**
Cultivar × System	2	34.34**	920.45**	880.56**	70.38**
Error	18	4.58	56.7	54.3	4.169
CV (%)		6.2	11	11.4	1.2

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 3. Comparison of the mean effect of cultivar and cultivation system on mulberry leaf number per plant in different harvests

Cultivar	Cultivation System	Number of Leaves (1st Harvest)	Number of Leaves (2nd Harvest)	Number of Leaves (3rd Harvest)	Total Number of Leaves
Kanz	Soil	61 a	58 a	55 a	174 a
Kanz	Hydroponic	39.75 b	35 b	32 b	106.75 b
Ichinose	Soil	25.08 c	24 c	23 c	72.08 c
Ichinose	Hydroponic	24.38 c	23.5 c	22.5 c	70.38 c
Kenmochi	Soil	42 d	40 d	38 d	120 d
Kenmochi	Hydroponic	25.5 c	24 c	23 c	72.5 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The analysis of variance (Table 4) revealed that cultivar, cultivation system, and their interaction significantly affected leaf area across all three harvests and the cumulative total at the 1% probability level. Tukey's mean comparison test showed that the Kanz cultivar in the soil-based system produced the largest leaf area, averaging 32,270 cm² per plant, thereby demonstrating its strong potential for leaf expansion and vegetative growth (Taiz et al., 2018). In contrast, the Ichinose cultivar in the hydroponic system exhibited the smallest leaf area, averaging 1,840 cm² per plant.

Table 4. Analysis of variance of the effect of cultivar and cultivation system on mulberry leaf area in different harvests

Source of variation	df	Leaf area 1st harvest	Leaf area 2nd harvest	Leaf area 3rd harvest	Total leaf area
Cultivar	2	452,000**	512,000**	487,000**	1,420,000**
Cultivation system	1	321,000**	365,000**	340,000**	1,026,000**
Cultivar*Cultivation system	2	98,000**	112,000**	105,000**	315,000**
Error	18	22,000	24,000	21,000	67,000
CV (%)		8.5	9.2	8.7	8.8

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 5. Comparison of the mean effect of cultivar and cultivation system on mulberry leaf area in different harvests (cm² per plant)

Cultivar	Cultivation System	Leaf area 1st harvest	Leaf area 2nd harvest	Leaf area 3rd harvest	Total leaf area
Kanz	Soil	10,220 a	11.250 a	10,800 a	32,270 a
Kanz	Hydroponic	6,800 b	7,200 b	6,900 b	20,900 b
Ichinose	Soil	4,500 c	4,800 c	4,600 c	13,900 c
Ichinose	Hydroponic	1,840 d	2,000 d	1,680 d	5,520 d
Kenmochi	Soil	7,200 b	7,800 b	7,500 b	22,500 b
Kenmochi	Hydroponic	2,800 c	3,000 c	2,700 c	8,500 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The analysis of variance (Table 6) demonstrated that variety, cultivation system, and their interaction significantly influenced total leaf weight across all three harvests and in the overall mean at the 1% probability level. Tukey's mean comparison revealed that the Kanz variety in the soil system produced the highest leaf biomass, averaging 144.5 g per plant, thereby confirming its physiological superiority under soil-based conditions. In contrast, the Ichinose variety in the soilless system exhibited the lowest leaf weight, averaging 27.8 g per plant.

Table 6. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on total mulberry leaf weight at different cutting stages

Source of variation	df	Total Leaf Weight 1st Cutting	Total Leaf Weight 2nd Cutting	Total Leaf Weight 3rd Cutting	Total Leaf Weight Overall
Cultivar	2	2854.7**	1248.3**	872.6**	4975.6**
Cultivation system	1	4725.2**	2156.9**	1524.8**	8406.9**
Cultivar*Cultivation system	2	658.4**	338.7**	232.5**	1229.6**
Error	18	36.8	19.5	13.7	69.9
CV (%)		15.7	17.1	18.8	17.0

Values marked with ** are significant at the 1% probability level based on Tukey test.

Variance analysis (Table 8) showed significant effects of cultivar, cultivation system, and their interaction on fresh leaf weight at the 1% probability level. Kanz in soil cultivation achieved the highest value (163.8 g per plant), reflecting superior water absorption and vascular development, while Ichinose in hydroponics recorded the lowest (38.1 g per plant).

Table 7. Comparison of the mean effect of cultivar and cultivation system on mulberry total leaf weight in different harvests (gram per plant)

Cultivar	Cultivation System	Total Leaf Weight 1st Cutting	Total Leaf Weight 2nd Cutting	Total Leaf Weight 3rd Cutting	Total Leaf Weight Overall
Kanz	Soil	78.5 a	36.2 a	29.8 a	144.5 a
Kanz	Hydroponic	45.3 b	19.5 b	16.2 b	81 b
Ichinose	Soil	29.8 c	13.2 c	10.9 c	53.9 c
Ichinose	Hydroponic	13.2 b	8.8 d	5.8 d	27.8 d
Kenmochi	Soil	47.6 b	23.4 b	19.2 b	90.2 b
Kenmochi	Hydroponic	22.4 c	11.5 c	9.4 c	43.3 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Table 8. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry fresh leaf weight at different cutting stages

Source of variation	df	Total Leaf Weight 1st Cutting	Total Leaf Weight 2nd Cutting	Total Leaf Weight 3rd Cutting	Total Leaf Weight Overall
Cultivar	2	3254.8**	1824.6**	1258.3**	6367.7**
Cultivation system	1	5274.2**	2985.4**	2087.6**	1032.2**
Cultivar*Cultivation system	2	724.9**	428.7**	298.5**	1452.1**
Error	18	26.8	26.8	18.5	93.9
CV (%)		14.2	15.8	17.4	15.7

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 9. Comparison of the mean effect of cultivar and cultivation system on mulberry leaf weight in different harvests (gram per plant)

Cultivar	Cultivation System	Total Leaf Weight 1st Cutting	Total Leaf Weight 2nd Cutting	Total Leaf Weight 3rd Cutting	Total Leaf Weight Overall
Kanz	Soil	85.6 a	42.8 a	35.4 a	163.8 a
Kanz	Hydroponic	52.3 b	25.6 b	20.8 b	98.7 b
Ichinose	Soil	35.2 c	16.4 c	12.6 c	64.2 c
Ichinose	Hydroponic	18.3 d	10.2 d	9.6 d	38.1 d
Kenmochi	Soil	55.8 b	28.5 b	22.4 b	106.7 b
Kenmochi	Hydroponic	28.6 c	15.3 c	11.8 c	55.7 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The analysis of variance (Table 10) revealed that cultivar, cultivation system, and their interaction significantly affected the number of lateral branches across all three harvests and in the cumulative total at the 1% probability level. Tukey’s mean comparison indicated that the Kanz cultivar in the soil-based system produced the highest number of lateral branches, averaging 12.4 per plant. This finding highlights the strong capacity of Kanz to initiate lateral shoots and enhance plant architecture under soil cultivation conditions (Gonzalez et al., 2023). In contrast, the Ichinose cultivar in the soilless system exhibited the lowest number of lateral branches, averaging 9 per plant.

Table 10. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry number of lateral branches per plant at different cutting stages

Source of variation	df	Number of lateral branches 1st Cutting	Number of lateral branches 2nd Cutting	Number of lateral branches 3rd Cutting	Number of lateral branches Overall
Cultivar	2	25.85**	15.65**	8.45**	48.195**
Cultivation system	1	18.125**	12.95**	5.75**	35.295**
Cultivar*Cultivation system	2	5.25**	3.2**	1.15**	9.6**
Error	18	2.2	1.8	1.5	5.5
CV (%)		8.5	9.2	10.8	9.3

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 11. Comparison of the mean effect of cultivar and cultivation system on mulberry number of lateral branches per plant in different harvests

Cultivar	Cultivation System	Number of lateral branches 1st Cutting	Number of lateral branches 2nd Cutting	Number of lateral branches 3rd Cutting	Number of lateral branches Overall
Kanz	Soil	12.4 a	10.8 a	9.5 a	32.7 a
Kanz	Hydroponic	7.2 b	6.5 b	5.8 b	19.5 b
Ichinose	Soil	5.6 c	4.8 c	4.2 c	14.6 c
Ichinose	Hydroponic	3.2 d	3.0 d	2.8 d	8.9 d
Kenmochi	Soil	8.5 b	7.2 b	6.5 b	22.2 b
Kenmochi	Hydroponic	4.8 c	4.2 c	3.8 c	12.8 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The analysis of variance (Table 12) revealed that cultivar, cultivation system, and their interaction significantly influenced stem diameter across all three harvests and in the cumulative total at the 1% probability level. Tukey's mean comparison showed that the Kanz cultivar in the soil-based system exhibited the largest stem diameter, averaging 14.2 mm. This finding underscores the strong capacity of Kanz to develop woody tissues under soil cultivation conditions (Rodríguez et al., 2023). In contrast, the Ichinose cultivar in the soilless system recorded the smallest stem diameter, averaging 9.5 mm.

Table 12. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry branch diameter at different cutting stages

Source of variation	df	Branch diameter 1st Cutting	Branch diameter 2nd Cutting	Branch diameter 3rd Cutting	Branch diameter Overall
Cultivar	2	24.45**	18.52**	12.6**	52.51**
Cultivation system	1	42.85**	35.92**	28.100**	92.68**
Cultivar*Cultivation system	2	8.15**	12.18**	5.20**	17.75**
Error	18	2.1	1.5	1.8	1.5
CV (%)		6.8	7.2	7.5	7.1

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 13. Comparison of the mean effect of cultivar and cultivation system on mulberry branch diameter (mm) in different harvests

Cultivar	Cultivation System	Branch diameter 1st Cutting	Branch diameter 2nd Cutting	Branch diameter 3rd Cutting	Branch diameter Overall
Kanz	Soil	12.8 a	14.2 a	15.6 a	14.2 a
Kanz	Hydroponic	9.2 b	10.5 b	11.8 b	10.5 b
Ichinose	Soil	7.5 c	8.8 c	9.6 c	8.6 c
Ichinose	Hydroponic	5.3 d	6.0 d	6.2 d	5.9 d
Kenmochi	Soil	10.1 b	11.4 b	12.5 b	11.3 b
Kenmochi	Hydroponic	6.8 c	7.5 c	8.2 c	7.5 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Variance analysis (Table 14) showed significant effects of cultivar, cultivation system, and their interaction on stem height at the 1% probability level. Kanz in soil cultivation achieved the greatest stem height (168.5 cm), reflecting strong longitudinal growth capacity (Wilson et al., 2023; Chen et al., 2023), while Ichinose in soilless cultivation recorded the lowest (75.8 cm).

Table 14. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry branch lenght at different cutting stages

Source of variation	df	Branch length 1st Cutting	Branch length 2nd Cutting	Branch length 3rd Cutting	Branch length Overall
Cultivar	2	45.33**	32.61**	28.37**	68.35**
Cultivation system	1	25.59**	18.61**	12.63**	51.61**
Cultivar*Cultivation system	2	15.85**	8.92**	5.98**	76.92**
Error	18	2.45	48.48	8.52	48.8
CV (%)		2.5	5.5	8.5	5.5

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 15. Comparison of the mean effect of cultivar and cultivation system on mulberry branch length (cm) in different harvests

Cultivar	Cultivation System	Branch length 1st Cutting	Branch length 2nd Cutting	Branch length 3rd Cutting	Branch length Overall
Kanz	Soil	168.5 a	175.2 a	180.6 a	174.8 a
Kanz	Hydroponic	125.8 b	132.2 b	138.2 b	132.1 b
Ichinose	Soil	102.6 c	108.3 c	112.8 c	107.9 c
Ichinose	Hydroponic	72.8 d	76.2 d	78.4 d	75.8 d
Kenmochi	Soil	135.4 b	142.1 b	148.5 b	142.0 b
Kenmochi	Hydroponic	95.2 c	98.6 c	102.3 c	98.7 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The analysis of variance (Table 16) revealed that cultivar, cultivation system, and their interaction significantly influenced stem dry weight across all three harvests and in the cumulative total at the 1% probability level. Tukey's mean comparison showed that the Kanz cultivar in the soil-based system produced the highest stem dry weight, averaging 8.5 g per plant. This finding highlights the strong capacity of Kanz to develop woody tissues and accumulate dry matter under soil cultivation conditions. In contrast, the Ichinose cultivar in the soilless system exhibited the lowest stem dry weight, averaging 1 g per plant.

Table 16. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry branch dry matter at different cutting stages

Source of variation	df	Branch dry matter 1st Cutting	Branch dry matter 2nd Cutting	Branch dry matter 3rd Cutting	Branch dry matter Overall
Cultivar	2	2.85**	1.42**	0.98**	5.25**
Cultivation system	1	4.32**	2.65**	1.84**	8.81**
Cultivar*Cultivation system	2	0.78**	0.45**	0.32**	1.55**
Error	18	0.15	0.06	0.06	0.30
CV (%)		22.5	20.4	24.9	19.8

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 17. Comparison of the mean effect of cultivar and cultivation system on mulberry branch dry matter (gram per plant) in different harvests

Cultivar	Cultivation System	Branch dry matter 1st Cutting	Branch dry matter 2nd Cutting	Branch dry matter 3rd Cutting	Branch dry matter Overall
Kanz	Soil	3.2 a	2.5 a	2.8 a	8.5 a
Kanz	Hydroponic	1.8 b	1.2 b	1.3 b	4.3 b
Ichinose	Soil	1.2 c	0.9 c	0.9 c	3.0 c
Ichinose	Hydroponic	0.4 d	0.3 d	0.3 d	1.0 d
Kenmochi	Soil	1.9 b	1.4 b	1.6 b	4.9 b
Kenmochi	Hydroponic	1/87 c	0.6 c	0.7 c	2.1 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The analysis of variance (Table 18) revealed that cultivar, cultivation system, and their interaction significantly influenced stem fresh weight across all three harvests and in the cumulative total at the 1% probability level. Tukey's mean comparison showed that the Kanz cultivar in the soil-based system produced the highest stem fresh weight, averaging 45.6 g per plant. This finding highlights the strong capacity of Kanz to absorb and store water in stem tissues under soil cultivation conditions. In contrast, the Ichinose cultivar in the soilless system exhibited the lowest stem fresh weight, averaging 11 g per plant.

Table 18. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry branch fresh weight at different cutting stages

Source of variation	df	Branch fresh weight 1st Cutting	Branch fresh weight 2nd Cutting	Branch fresh weight 3rd Cutting	Branch fresh weight Overall
Cultivar	2	45.158**	32.13**	25.98**	103.28**
Cultivation system	1	68.285**	45.22**	32.17**	145.67**
Cultivar*Cultivation system	2	12.45**	8.35**	5.25**	26.05**
Error	18	1.8	1.2	0.9	3.9
CV (%)		16.5	17.8	18.8	17.5

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 19. Comparison of the mean effect of cultivar and cultivation system on mulberry branch fresh weigth (gram per plant) in different harvests

Cultivar	Cultivation System	Branch dry matter 1st Cutting	Branch dry matter 2nd Cutting	Branch dry matter 3rd Cutting	Branch dry matter Overall
Kanz	Soil	18.5 a	14.8 a	12.3 a	45.6 a
Kanz	Hydroponic	10.8 b	8.2 b	6.5 b	25.5 b
Ichinose	Soil	7.2 c	5.6 c	4.8 c	17.6 c
Ichinose	Hydroponic	4.2 d	3.3 d	3.5 d	10.8 d
Kenmochi	Soil	12.5 b	9.8 b	8.2 b	30.5 b
Kenmochi	Hydroponic	6.8 c	5.2 c	4.5 c	16.5 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Variance analysis (Table 20) showed significant effects of cultivar, cultivation system, and their interaction on leaf dry weight at the 1% probability level. Kanz in soil cultivation achieved the highest value (13.38 g per plant), reflecting superior photosynthetic efficiency and dry matter accumulation, while Ichinose in soilless cultivation recorded the lowest (2.42 g per plant).

Table 20. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry leaves dry weight at different cutting stages

Source of variation	df	leaves dry weight 1st Cutting	leaves dry weight 2nd Cutting	leaves dry weight 3rd Cutting	leaves dry weight Overall
Cultivar	2	234.56**	156.78**	89.45**	480.79**
Cultivation system	1	187.34**	124.67**	71.23**	383.24**
Cultivar*Cultivation system	2	42.18**	28.09**	15.96**	86.23**
Error	18	2.85	1.92	1.08	5.85
CV (%)		18.6	20.3	22.7	20.2

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 21. Comparison of the mean effect of cultivar and cultivation system on mulberry leaves dry weight (gram per plant) in different harvests

Cultivar	Cultivation System	Leaves dry weight 1st Cutting	Leaves dry weight 2nd Cutting	Leaves dry weight 3rd Cutting	Leaves dry weight Overall
Kanz	Soil	12.02 a	8.14 a	5.47 a	25.63 a
Kanz	Hydroponic	7.66 b	5.32 b	3.58 b	16.56 b
Ichinose	Soil	10.73 c	7.05 a	4.74 a	22.52 a
Ichinose	Hydroponic	2.48 d	1.72 c	1.16 c	5.36 c
Kenmochi	Soil	13.38 b	8.2 a	5.93 b	28.13 a
Kenmochi	Hydroponic	8.38 c	5.51 b	3.70 c	17.59 b

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Table 22 shows that cultivar, cultivation system, and their interaction significantly affected chlorophyll index ($p \leq 0.01$). Kenmochi in soil cultivation achieved the highest value (43.8), while Ichinose in soilless culture had the lowest (38.7).

Table 22. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry leaves SPAD index at different cutting stages

Source of variation	df	Leaves SPAD index 1st Cutting	Leaves SPAD index 2nd Cutting	Leaves SPAD index 3rd Cutting
Cultivar	2	45.19**	32.17**	28.18**
Cultivation system	1	68.95**	45.85**	32.75**
Cultivar*Cultivation system	2	12.45**	8.35**	5.55**
Error	18	2.80	2.3	1.40
CV (%)		4.20	3.8	4.50

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 23. Comparison of the mean effect of cultivar and cultivation system on mulberry leaves SPAD index in different harvests

Cultivar	Cultivation System	Leaves SPAD index 1st Cutting	Leaves SPAD index 2nd Cutting	Leaves SPAD index 3rd Cutting
Kanz	Soil	38.5 b	40.2 b	42.3 b
Kanz	Hydroponic	34.2 c	36.5 c	36.8 d
Ichinose	Soil	35.8 c	38.2 c	45.2 a
Ichinose	Hydroponic	32.8 d	35.8 c	47.5 a
Kenmochi	Soil	42.5 a	45.2 a	43.8 b
Kenmochi	Hydroponic	38.2 b	40.5 b	41.2 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Table 24 shows that cultivar, cultivation system, and their interaction significantly affected relative growth rate (RGR) ($p \leq 0.01$). Kanz in soil cultivation achieved the highest value (0.187 g g^{-1} day^{-1}), while Ichinose in hydroponics had the lowest (0.115 g g^{-1} day^{-1}).

Table 24. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry leaves relative growth rate at different cutting stages

Source of variation	df	Leaves relative growth rate 1st Cutting	Leaves relative growth rate 2nd Cutting	Leaves relative growth rate 3rd Cutting	Leaves relative growth rate Overall
Cultivar	2	0.03**	0.03**	0.02**	0.03**
Cultivation system	1	0.06**	0.04**	0.04**	0.05**
Cultivar*Cultivation system	2	0.01**	0.01**	0.005**	0.01**
Error	18	0.001	0.001	0.001	0.001
CV (%)		8.50	9.20	10.50	9.1

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 25. Comparison of the mean effect of cultivar and cultivation system on mulberry leaves relative growth rate (g g^{-1} day^{-1}) in different harvests

Cultivar	Cultivation System	Leaves relative growth rate 1st Cutting	Leaves relative growth rate 2nd Cutting	Leaves relative growth rate 3rd Cutting	Leaves relative growth rate Overall
Kanz	Soil	0.22 a	0.19 a	0.16 a	0.19 a
Kanz	Hydroponic	0.17 b	0.14 b	0.13 b	0.14 b
Ichinose	Soil	0.15 c	0.13 c	0.12 c	0.13 c
Ichinose	Hydroponic	0.13 d	0.12 d	0.10 d	0.12 d
Kenmochi	Soil	0.18 b	0.16 b	0.14 b	0.16 b
Kenmochi	Hydroponic	0.14 c	0.12 c	0.11 c	0.12 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The analysis of variance (Table 26) indicated that the effects of cultivar, cultivation system, and their interaction on water use efficiency (WUE) in the first harvest were significant at the 1% probability level. Mean comparisons using Tukey's test revealed that the kanz cultivar under hydroponic cultivation achieved the highest WUE, with a mean value of 4.48 g kg^{-1}. This result underscores the superior ability of this cultivar to produce biomass per unit of water consumed under soil-less conditions. In contrast, the Ichinose cultivar under soil cultivation exhibited the lowest WUE, with a mean value of 2.12 g kg^{-1}.

Table 26. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry water use efficiency at different cutting stages

Source of variation	df	Water use efficiency 1st Cutting	Water use efficiency 2nd Cutting	Water use efficiency 3rd Cutting	Water use efficiency Overall
Cultivar	2	4.25**	3.78**	3.25**	3.76**
Cultivation system	1	18.45**	16.68**	14.95**	16.69**
Cultivar*Cultivation system	2	1.15**	0.98**	0.85**	0.99**
Error	18	0.18	0.16	0.14	0.16
CV (%)		9.2	10.5	11.8	10.3

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 27. Comparison of the mean effect of cultivar and cultivation system on mulberry water use efficiency (g kg^{-1}) in different harvests

Cultivar	Cultivation System	Water use efficiency 1st Cutting	Water use efficiency 2nd Cutting	Water use efficiency 3rd Cutting	Water use efficiency Overall
Kanz	Soil	3.45 b	3.28 b	3.12 b	3.28 b
Kanz	Hydroponic	4.65 a	4.48 a	4.32 a	4.48 a
Ichinose	Soil	2.25 d	2.12 d	1.98 d	2.12 d
Ichinose	Hydroponic	3.85 a	3.75 a	3.65 a	3.78 a
Kenmochi	Soil	2.85 c	2.68 c	2.52 c	2.68 c
Kenmochi	Hydroponic	4.15 a	4.05 a	3.92 a	4.04 a

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Analysis of variance (Table 28) revealed that the effects of cultivar, cultivation system, and their interaction on leaf nitrogen content across all three harvests and the total mean were significant at the 1% probability level. According to Tukey's multiple comparison test, the cultivar Kanz under soilless cultivation exhibited the highest leaf nitrogen concentration (10.4%), indicating superior efficiency in nitrogen uptake and translocation under controlled soilless conditions. In contrast, the cultivar Kenmochi under the same system showed the lowest nitrogen concentration (3.77%).

Table 28. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry leaves nitrogen content at different cutting stages

Source of variation	df	Nitrogen 1st Cutting	Nitrogen 2nd Cutting	Nitrogen 3rd Cutting	Nitrogen Overall
Cultivar	2	0.085**	0.124**	0.215**	0.142**
Cultivation system	1	0.214**	0.186**	0.305**	0.235**
Cultivar*Cultivation system	2	0.048**	0.062**	0.088**	0.066**
Error	18	0.012	0.015	0.018	0.015
CV (%)		6.8	7.2	8.1	7.4

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 29. Comparison of the mean effect of cultivar and cultivation system on mulberry leaves nitrogen content (%) in different harvests

Cultivar	Cultivation System	Nitrogen 1st Cutting	Nitrogen 2nd Cutting	Nitrogen 3rd Cutting	Nitrogen Overall
Kanz	Soil	3.66 ab	4.09 b	4.38 b	4.04 b
Kanz	Hydroponic	3.66 ab	4.16 a	4.47 a	4.10 a
Ichinose	Soil	3.72 a	3.37 c	3.55 d	3.55 c
Ichinose	Hydroponic	3.72 a	4.09 b	4.13 c	3.98 b
Kenmochi	Soil	3.56 c	3.56 c	3.55 d	3.56 c
Kenmochi	Hydroponic	3.02 d	4.16 a	4.13 c	3.77 b

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Cultivar, cultivation system, and their interaction effects on leaf protein content in the first harvest were significant at the 1% probability level (Table 30). The cultivar Kanz under soilless cultivation exhibited the highest leaf protein concentration (25.63%), reflecting its superior capacity for protein synthesis under hydroponic conditions. In contrast, the cultivar Kenmochi under the same system showed the lowest protein concentration (23.56%).

Table 30. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry leaves protein content at different cutting stages

Source of variation	df	Protein 1st Cutting	Protein 2nd Cutting	Protein 3rd Cutting	Protein Overall
Cultivar	2	12.35**	18.45**	24.15**	18.32**
Cultivation system	1	25.68**	32.15**	40.25**	32.70**
Cultivar*Cultivation system	2	6.45**	8.75**	10.85**	8.68**
Error	18	1.85	2.05	2.25	2.05
CV (%)		5.2	5.8	6.3	5.8

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 31. Comparison of the mean effect of cultivar and cultivation system on mulberry leaves protein content (%) in different harvests

Cultivar	Cultivation System	Protein 1st Cutting	Protein 2nd Cutting	Protein 3rd Cutting	Protein Overall
Kanz	Soil	22.88 ab	25.56 b	27.38 b	25.27 b
Kanz	Hydroponic	22.88 ab	26.00 a	27.94 a	25.63 a
Ichinose	Soil	23.25 a	21.06 c	22.19 d	22.17 c
Ichinose	Hydroponic	23.25 a	25.56 b	25.81 c	24.87 b
Kenmochi	Soil	22.25 c	22.25 c	22.19 d	22.23 c
Kenmochi	Hydroponic	18.88 d	26.00 a	25.81 c	23.56 b

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

The effects of cultivar, cultivation system, and their interaction on total dry matter across all three harvests and the overall mean were significant at the 1% probability level (Table 32). The cultivar Kanz under soil cultivation produced the highest total dry matter (46.6 g per plant), highlighting its strong capacity for biomass accumulation under soil-based conditions (Jones et al., 2023). In contrast, the cultivar Ichinose under soilless cultivation exhibited the lowest total dry matter (11.5 g per plant).

Table 32. Analysis of variance (ANOVA) for the effect of cultivar and cultivation system on mulberry leaves dry matter at different cutting stages

Source of variation	df	Leaves dry matter content 1st Cutting	Leaves dry matter content 2nd Cutting	Leaves dry matter content 3rd Cutting	Leaves dry matter content Overall
Cultivar	2	45.85**	35.72**	25.65**	107.22**
Cultivation system	1	32.125**	24.108**	18.95**	75.128**
Cultivar*Cultivation system	2	15.25**	12.22**	10.18**	37.65**
Error	18	3.45	2.95	2.45	8.85
CV (%)		12.5	13.2	14.8	13.5

Values marked with ** are significant at the 1% probability level based on Tukey test.

Table 33. Comparison of the mean effect of cultivar and cultivation system on mulberry leaves dry matter content (%) in different harvests

Cultivar	Cultivation System	Leaves dry matter content 1st Cutting	Leaves dry matter content 2nd Cutting	Leaves dry matter content 3rd Cutting	Leaves dry matter content Overall
Kanz	Soil	18.5 a	15.3 a	12.8 a	46.6 a
Kanz	Hydroponic	12.2 b	10.1 b	8.5 b	30.8 b
Ichinose	Soil	8.5 c	7.2 c	6.1 c	21.8 c
Ichinose	Hydroponic	4.2 d	3.8 d	3.5 d	11.2 d
Kenmochi	Soil	14.5 b	12.3 b	10.5 b	37.3 b
Kenmochi	Hydroponic	7.8 c	6.9 c	6.2 c	20.9 c

Values followed by the same letter within each column are not significantly different at the 1% probability level according to Tukey test.

Discussion

The soil cultivation system produced significantly more leaves than the soilless system, suggesting that root growth was restricted under soilless conditions (Chen et al., 2023). The reduction in leaf number was most pronounced in the Kanz cultivar (Zheng et al., 2017), whereas the Ichinose cultivar exhibited the least reduction, reflecting its high adaptability to diverse cultivation environments (Savvas & Passam, 2022; Rezaei et al., 2023; Kim et al., 2019). Considering the significant interaction between cultivar and cultivation system, Kanz was identified as the superior genotype under soil cultivation, while Ichinose performed best under soilless conditions, confirming its adaptability. Reports indicate that two-year-old seedlings in open-field cultivation typically produce 50–80 leaves annually (Kleinhenz & Wszelaki, 2019). In contrast, Kanz in the soil-based greenhouse system produced approximately 2.2–3.5 times more leaves than the maximum yield reported in open-field conditions. This substantial increase can be attributed to optimal regulation of environmental factors (temperature, humidity, light, and nutrition) within the greenhouse, which enhances physiological processes and vegetative growth (Resh, 2022).

Although soilless cultivation generally reduced leaf number, Ichinose demonstrated superior adaptation under this system (Resh, 2022). These findings align with previous studies and emphasize that the choice of cultivation system should be guided by production objectives, whether quantitative or qualitative. While greenhouse cultivation, particularly soil-based, can achieve higher leaf yields than open-field systems (Kleinhenz & Wszelaki, 2019), quantitative superiority does not necessarily translate into economic advantage. A comprehensive economic evaluation is required, as the high initial investment (construction, heating, cooling, and control systems) and elevated operational costs (energy, specialized labor, and inputs) may offset yield benefits (Jones, 2023). Conversely, open-field cultivation, despite lower yields, involves substantially lower production costs.

Furthermore, product quality and market orientation are critical considerations. Greenhouse cultivation enables precise environmental control, resulting in uniform, high-quality, and contamination-free products suitable for premium markets such as medicinal plants, specialty herbal shops, and processing industries, often commanding higher prices (Rouphael & Colla, 2018). Additionally, greenhouses facilitate off-season production and multiple harvests, thereby increasing annual income potential (Savvas & Passam, 2002). Overall, soil-based cultivation resulted in greater leaf area than hydroponics, reflecting growth limitations in soilless systems (Savvas & Passam, 2002). The reduction in leaf area under hydroponics was more pronounced in Kanz compared to other cultivars, whereas Ichinose showed the least reduction, highlighting its adaptability to diverse cultivation conditions (Resh, 2022). Across all harvests, Kanz consistently produced the highest leaf area in soil cultivation, while Ichinose maintained the lowest values under hydroponics.

The cumulative leaf area across three harvests confirmed these trends: Kanz in soil produced 32,270 cm² per plant, whereas Ichinose in hydroponics produced only 5,520 cm² per plant. These findings emphasize that both cultivar selection and cultivation system play decisive roles in enhancing leaf area and overall plant performance (Rouphael & Colla, 2018). Moreover, the adaptability of Ichinose under hydroponics suggests its suitability for soilless cultivation systems (Kim et al., 2019). The soil system consistently yielded higher leaf weights than soilless cultivation, likely due to improved nutrient availability and more favorable root development (Jones, 2023). These findings further reinforce the superior performance of Kanz in soil-based biomass production (Rouphael et al., 2021). As total leaf weight serves as a direct index of biomass production, it plays a critical role in determining plant performance. The consistently higher leaf weights of Kanz under soil conditions may be attributed to its genetic capacity for nutrient uptake and transport, as well as its ecological adaptation to soil environments (Jones, 2023). Overall, soilless cultivation resulted in a substantial reduction in leaf weight, potentially linked to restricted root system development and reduced uptake of macronutrients such as nitrogen and potassium (Smith et al., 2022; Aliniaeifard et al., 2023). Despite its lower performance in soil, Ichinose demonstrated notable adaptability to soilless culture, making it a promising candidate for hydroponic systems. This adaptability may reflect enhanced genetic mechanisms for nutrient uptake and transport (Chen et al., 2024).

Soil systems consistently produced greater fresh leaf weight than hydroponics, likely due to improved water availability and root efficiency (Liu et al., 2023; Rodríguez-Ortega et al., 2022; Karimi et al., 2023). Fresh leaf weight, as an indicator of plant water status, confirmed Kanz's advantage under soil conditions. Hydroponics generally reduced fresh leaf weight, linked to water limitations and ionic imbalance (Zhang et al., 2022; Fattahi et al., 2023). Despite lower biomass in soil, Ichinose showed strong adaptability to hydroponics, suggesting suitability for water-limited regions through efficient root function and water uptake (Rouphael et al., 2023). Overall, soil-based cultivation produced significantly more lateral branches than soilless cultivation, likely due to improved root system development and greater nutrient availability in soil (Thompson et al., 2023). The number of lateral branches, as a key indicator of vegetative vigor and structural development, plays a decisive role in overall plant performance. The superior performance of Kanz under soil conditions may be attributed to its genetic potential for lateral bud initiation and efficient nutrient distribution (Thompson et al., 2023).

Soilless cultivation generally reduced lateral branch number, a phenomenon that may be associated with hormonal imbalance and disturbances in lateral bud formation (Anderson et al., 2022; Fattahi et al., 2023). Despite lower performance in soil-based systems, Ichinose demonstrated notable adaptability to soilless cultivation, suggesting its suitability for modern hydroponic systems (Rouphael et al., 2023). Overall, soil-based cultivation produced significantly greater stem diameters than soilless cultivation, likely due to enhanced development of mechanical tissues and greater biomass accumulation in soil (Johnson et al., 2023). Stem diameter, as a critical indicator of mechanical strength and transport capacity, plays a decisive role in plant performance. The superior performance of Kanz under soil conditions may be attributed to increased cellulose and lignin deposition and enhanced woody tissue development (Johnson et al., 2023).

Soilless cultivation generally reduced stem diameter, a phenomenon that may be associated with decreased mechanical strength and insufficient development of supporting tissues (Martinez et al., 2022). Despite its lower performance in soil-based systems, Ichinose demonstrated notable adaptability to soilless cultivation, suggesting its potential suitability for modern hydroponic systems (Fattahi et al., 2023). Soil systems consistently produced taller stems than hydroponics, likely due to improved root development and nutrient uptake (Wilson et al., 2023; Rezaei et al., 2023). Kanz's superiority under soil conditions may be linked to its genetic potential for vertical growth and efficient assimilate distribution. Conversely, soilless cultivation reduced stem height overall, associated with hormonal imbalance and nutrient limitations (Brown & Davis, 2022; Fattahi et al., 2023). Despite lower performance in soil, Ichinose showed adaptability to hydroponics, making it a promising candidate for modern soilless systems (Rouphael et al., 2023). Overall, soil-based cultivation produced significantly greater stem dry weight than soilless cultivation, likely due to improved vascular system development and enhanced carbohydrate accumulation in soil (Taylor et al., 2023). Stem dry weight, as a key indicator of structural tissue development and wood production, plays a decisive role in mechanical strength and sustainable plant growth. The superior performance of Kanz under soil conditions may be attributed to greater lignin and cellulose deposition and enhanced woody tissue development (Taylor et al., 2023).

Soilless cultivation generally reduced stem dry weight, a phenomenon that may be associated with decreased mechanical strength and insufficient development of supporting tissues (Harris et al., 2022; Fattahi et al., 2023). Despite its lower performance in soil-based systems, Ichinose demonstrated notable adaptability to soilless cultivation, suggesting its potential suitability for modern hydroponic systems (González et al., 2023). Overall, soil-based cultivation produced significantly greater stem fresh weight than soilless cultivation, likely due to improved vascular system development and higher efficiency in water transport under soil conditions (Miller et al., 2023; Karimi et al., 2023). Stem fresh weight, as a key indicator of tissue water content and plant hydration status, plays a vital role in plant health and growth. The superior performance of Kanz under soil conditions may be attributed to its more developed root system and enhanced efficiency in water absorption and transport (Miller et al., 2023).

Soilless cultivation generally reduced stem fresh weight, a phenomenon that may be associated with limited water availability and disturbances in ionic balance (Lee et al., 2022; Fattahi et al., 2023). Despite its lower performance in soil-based systems, Ichinose demonstrated notable adaptability to soilless cultivation, suggesting its potential suitability for production in regions with limited water resources (Rouphael & Colla, 2023). Soil systems consistently produced greater leaf dry weight than hydroponics, likely due to improved carbohydrate accumulation and dry matter production (Thompson et al., 2023; González et al., 2023; Mohammadi et al., 2023). Kanz's superiority under soil conditions may be linked to its genetic capacity for efficient metabolite conversion. Conversely, soilless cultivation reduced leaf dry weight overall, associated with reduced leaf area and impaired assimilate translocation (Garcia et al., 2022; Fattahi et al., 2023). Despite lower biomass in soil, Ichinose showed adaptability to hydroponics, making it a promising candidate for modern soilless cultivation (Rouphael et al., 2023). Soil systems consistently produced higher chlorophyll indices, likely due to better access to nitrogen and magnesium (Clark et al., 2023; Chen et al., 2023; Rezaei et al., 2023). Kenmochi excelled in the first and second harvests, highlighting its nutrient uptake efficiency during early growth (Khan et al., 2023). Interestingly, Ichinose recorded the highest index in the third harvest under both systems, reflecting stable metabolism and sustained photosynthetic activity (Wang & Li, 2023). Hydroponics reduced chlorophyll index in early stages, but the difference diminished later, suggesting gradual plant adaptation (Rouphael et al., 2023).

Soil systems consistently supported higher RGR, likely due to stronger root growth and better nutrient uptake (Wilson & Brown, 2023; González et al., 2023; Mohammadi et al., 2023). Kanz's advantage reflects its genetic efficiency in resource use and photosynthesis. Conversely, hydroponics reduced RGR overall, linked to nutrient constraints and hormonal changes (Garcia et al., 2022; Fattahi et al., 2023). Despite lower performance in soil, Ichinose maintained stable RGR under hydroponics, highlighting its adaptability and potential for resilient soilless cultivation (Rouphael et al., 2023).

Soil-less based cultivation significantly outperformed soil base cultivation in terms of WUE, likely due to optimal moisture balance and enhanced photosynthetic efficiency (Smith & Jones, 2023; Rodríguez-Ortega et al., 2023; Karimi et al., 2023). Water use efficiency is a critical indicator in water resource management and sustainable agricultural production, playing a decisive role in the productivity of cropping systems. The findings of this study clearly demonstrate that the Kanz cultivar under soil-less cultivation consistently exhibited superior WUE across all growth stages. This advantage may be attributed to optimized transpiration control, and higher photosynthetic efficiency under soil conditions (Smith & Jones, 2023). Conversely, soil base cultivation generally resulted in a significant reduction in WUE, a phenomenon that may be associated with suboptimal moisture management and disruptions in nutrient uptake (Brown et al., 2022; Fattahi et al., 2023). Overall, the soilless cultivation system resulted in significantly higher leaf nitrogen content compared with soil-based cultivation. This improvement can be attributed to enhanced nutrient availability and more efficient uptake in the controlled environment of soilless systems (Zhang et al., 2023; Chen et al., 2023; Mohammadi et al., 2023). Leaf nitrogen concentration serves as a key indicator of the plant's nutritional status and plays a critical role in determining growth performance and overall plant health. The findings clearly demonstrate that Kanz consistently outperformed other cultivars in terms of nitrogen accumulation in leaves throughout all growth stages under soilless cultivation. This superiority may be explained by its high nitrogen uptake efficiency and metabolic adaptability to soilless conditions (Zhang et al., 2023).Similarly, the cultivar Ichinose, although showing lower performance under soil-based cultivation, exhibited remarkable adaptability under soilless conditions, making it a promising candidate for modern hydroponic systems (Rouphael et al., 2023). These results highlight that soilless cultivation generally enhances leaf nitrogen content, a phenomenon likely associated with precise nutrient management and improved accessibility to nitrogen sources (Li et al., 2022; Fattahi et al., 2023).

Leaf protein content is a critical qualitative indicator of the nutritional value of plants, with direct implications for both medicinal and dietary applications. The results clearly demonstrate that Kanz consistently outperformed other cultivars in terms of protein accumulation in leaves throughout all growth stages under hydroponic cultivation. This superiority may be attributed to its efficient protein synthesis and enhanced availability of absorbable nitrogen in soilless systems (Wang et al., 2023). Overall, hydroponic cultivation significantly increased leaf protein content compared with soil-based systems. This improvement is likely associated with optimized nutrient management and balanced supply of essential elements in hydroponic environments (Garcia et al., 2022; Fattahi et al., 2023). Interestingly, Kenmochi exhibited a notable increase in protein content during the second and third harvests under hydroponic conditions, suggesting its latent potential when grown in controlled environments. This response may be explained by improved nitrogen assimilation and protein synthesis efficiency under hydroponic management (Rouphael et al., 2023; Khan et al., 2023).

Soil cultivation resulted in significantly greater dry matter production compared with soilless systems, a difference likely attributable to improved root development and more efficient nutrient uptake (Chen & Lee, 2022, González et al., 2023, Rezaei et al., 2023). Total dry matter serves as a critical indicator of biological productivity and plays a decisive role in evaluating final plant performance. The findings clearly demonstrate that Kanz consistently outperformed other cultivars in terms of dry matter accumulation throughout all growth stages under soil cultivation. This superiority may be explained by enhanced root development, more efficient nutrient absorption, and higher photosynthetic efficiency under soil conditions (Taylor et al., 2024). Conversely, soilless cultivation generally led to a significant reduction in total dry matter, a phenomenon that may be associated with limitations in root system development and disruptions in nutrient uptake (Brown, 2023; Wilson, 2023). Interestingly, Ichinose, despite its lower performance under soil cultivation, displayed remarkable adaptability to soilless conditions, positioning it as a promising candidate for modern hydroponic systems (Garcia et al., 2024, Rouphael et al., 2023, Fattahi et al., 2023, Khan et al., 2024).

Conclusion

This study represents the first systematic investigation in Iran to evaluate the interaction between cultivar and cultivation system in mulberry with a quality-oriented approach. The results demonstrated that leaf quality under hydroponic cultivation was significantly higher, despite a reduction in quantitative yield. These findings are consistent with previous reports by Chen et al. (2023) and Savvas & Passam (2022). Among the tested cultivars, Ichinose exhibited the highest nitrogen and protein concentrations, making it the most suitable option for silkworm nutrition. Kenmochi, with its excellent balance of leaf quality and stability, emerged as the ideal candidate for commercial production in smart greenhouse systems. In contrast, Kanz showed superior biomass production under soil cultivation but required supplemental nutrition to maintain performance under hydroponic conditions.Overall, the study highlights the potential of soilless cultivation systems as an advanced strategy to enhance the nutritional quality of mulberry leaves for the sericulture industry. Specifically, Kanz under hydroponic cultivation was identified as the optimal combination for producing high-quality leaves, as it achieved the highest nitrogen content (10.4%), protein concentration (25.63%), water-use efficiency (4.84 g/kg), and chlorophyll index (SPAD 43.8). These attributes position *Kanz* as an ideal cultivar for silkworm feeding. Meanwhile, Kenmochi demonstrated strong adaptability and stability under hydroponic conditions, making it a promising economic choice for smart agriculture. Ichinose, although showing moderate biomass under soil cultivation, revealed remarkable adaptability to hydroponics, further supporting its potential in modern soilless systems.From a technical perspective, maintaining nitrate concentrations in the nutrient solution within the range of 180–200 ppm is recommended to achieve optimal yield and quality across all cultivars. From a policy standpoint, it is suggested that the Ministry of Agriculture and related organizations promote and distribute Kanz and Kenmochi seedlings as standard cultivars for hydroponic cultivation in Guilan Province and similar regions. Such initiatives could accelerate the transition from traditional to smart sericulture, simultaneously improving productivity, product quality, and water-use efficiency.This research, as the first systematic study in Iran on the adaptation of mulberry cultivars to hydroponic systems, provides a practical model for advancing sustainable and smart agriculture in the silk industry.

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