

Quantitative Evaluation of Ascorbic Acid in Iraqi *Vitis vinifera* Cultivars: A Biochemical and Organic Analysis

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ABSTRACT

Background: *Vitis vinifera* (grapes) represent one of Iraq's most significant fruit crops, widely cultivated in regions such as Babylon, Diyala, and Kurdistan. Beyond their economic value, grapes are a rich source of bioactive compounds, notably vitamin C (ascorbic acid), a key antioxidant essential for redox balance, collagen synthesis, immune function, and cellular protection against oxidative stress. **Objectives:** This study aimed to evaluate the concentration of vitamin C in selected *Vitis vinifera* cultivars grown in different regions of Iraq. **Methodology:** Twelve local *Vitis vinifera* cultivars were collected and analyzed during the 2021–2023 growing seasons from vineyards in central and northern Iraq. Ascorbic acid concentration in its berries was quantified using standardized biochemical assays. Comparative analysis across cultivars, regions, and seasons was performed. Climatic parameters including temperature were recorded to evaluate their influence on ascorbic acid accumulation. **Results:** Significant inter-cultivar differences in vitamin C concentration, ranging from 7.68 to 10.36 mg/100 g fresh weight. Environmental stress particularly high temperature and low hydrothermal coefficient was negatively correlated with ascorbic acid retention. Black Sadany, Shekh-Nuradeen, and Moon Drop exhibited the highest and most stable vitamin C content across seasons, indicating strong biochemical resilience under heat stress. Berry color and ripening period had minimal influence. **Conclusions:** Considerable genetic variation in vitamin C content exists among Iraqi *Vitis vinifera* cultivars suggesting that genotype and temperature are the primary determinants of ascorbic acid biosynthesis. The identification of high ascorbic acid genotypes provides a promising basis for enhancing the nutritional, preventive, and therapeutic potential of improved antioxidant capacity.

1. Introduction

Fruits and berries are vital in our everyday diets because they offer far more than just carbohydrates, proteins and fats they also deliver a rich array of bio-active compounds that help maintain our body's internal balance and protect against illness ¹. Among the most familiar fruit crops are apples (*Malus domestica*), pears (*Pyrus communis*), strawberries (*Fragaria × ananassa*) and grapes (*Vitis vinifera* L.) ². The grape (*Vitis vinifera*) in particular has a long history and remains one of the most economically important fruit crops globally, used both fresh and in products like juice and raisins ³. In Iraq, *Vitis vinifera* is especially significant: it is widely grown in regions such as Babylon, Diyala and Kurdistan, where the soil and climate help produce high-quality fruit ⁴. Beyond being an agricultural commodity, grapes are nutritionally dense they contain vitamins, phenolic compounds and flavonoids that all work together to support human health and well-being. One of the standout nutrients in grapes is ascorbic acid (vitamin C) ⁵. This compound plays multiple critical roles: it is involved in collagen production (important for skin, bones and blood vessels), catecholamine synthesis (which affects stress and mood regulation), and the metabolism of bile acids (helping in digestion and fat processing) ⁶. Vitamin C also helps regulate cholesterol levels and reinforces the immune system by shielding cells from oxidative damage ^{7,8}. As a powerful antioxidant, vitamin C neutralizes reactive oxygen species (ROS) and helps maintain redox balance within the body ⁹. When vitamin C is lacking, people risk weakened immunity, slower wound healing, scurvy in extreme cases, and a higher vulnerability to conditions tied to oxidative stress such as obesity, heart disease, cancer and certain respiratory illnesses ¹⁰. Importantly, humans cannot make vitamin C internally because we lost the enzyme L-gulonolactone oxidase during evolution therefore we must obtain it through our diet, especially from fruits and vegetables ¹¹. Recent work shows that more than half of the world's population intake falls below recommended levels of vitamin C which is particularly acute in low- and middle-income countries ¹². Although no

exact figures for Iraq were found in this text, the implication is clear: increasing the availability and use of vitamin C-rich foods is a public health priority. In Iraq, despite the rich variety of local *Vitis vinifera* cultivars and their adaptation to a range of agro-ecological zones, detailed information on how vitamin C levels vary among these local grape types is scarce. Understanding how environmental factors such as regional temperature swings and rainfall patterns affect vitamin C accumulation in the berries will help identify the best cultivars for nutritional purposes. These insights are useful not only for improving diet quality and health outcomes, but also for guiding future breeding programs aimed at stronger climate resilience and enhanced antioxidant capacity in grape varieties across the Middle East ¹³. This study was designed to quantitatively evaluate the concentration of ascorbic acid in selected *Vitis vinifera* cultivars grown in different regions of Iraq, compare how ascorbic acid content varies by cultivar, region, and over the 2021-2023 growing seasons, and identify cultivars that consistently produce high levels of ascorbic acid and may serve as valuable dietary antioxidant sources and breeding material for nutritionally enhanced grape production.

2. Materials and Methods

Study Design and Duration: This investigation was conducted from 2021–2023 at the Laboratory of Industrial Technologies, Baghdad, Iraq; targeting the quantification of ascorbic acid (vitamin C) in diverse *Vitis vinifera* L. cultivars grown under Iraqi conditions. Twelve cultivars differing by ecological–geographical origin, intended use, and berry color were included (Table 1). Hejazi was selected as the reference (control) based on its stable agronomic performance in local vineyards.

Ethical Statement: No human or animal subjects were involved in this study. The research complied with institutional and national ethical standards regarding the collection and use of plant materials and environmental data.

Plant Material and Sampling: Grape berries were collected at full physiological ripeness from experi-

Table 1. Characteristics of Studied *Vitis vinifera* Varieties

No.	Variety/Form	Origin	Berry Color	Purpose
1	Hejazi (control)	Central Iraq	Black	Universal
2	Tre Rash	Northern Iraq	Red	Table
3	Eayn al-Thawr	Babylon	Black	Table
4	Moon Drop	Imported (California)	Black	Table
5	Crimson	Imported (Chile)	Red	Table
6	Samarkandi	Kurdistan	Red-Purple	Universal
7	Nubari	Southern Iraq	White-Green	Table
8	Ari Uzumu	Turkey	White	Universal
9	Agh Gavra	Northern Iraq	White	Universal
10	Ashqarbasha	Local Iraqi variety	Pink	Universal
11	Shekh-Nuradeen	Kurdistan	Black	Universal
12	Black Sadany	Middle Euphrates region	Dark black	Table

mental plots maintained under uniform cultivation practices. Each sample was harvested during its optimal maturity stage to ensure biochemical consistency and minimize post-harvest variation. For each variety, duplicate samples (≈100 g each) were randomly taken from multiple vines, immediately cooled, and homogenized in an ice bath to prevent oxidative degradation of ascorbic acid. Analyses were carried out immediately after sample preparation to ensure precision and accuracy.

Determination of Vitamin C Content: The titrimetric method was employed to determine the concentration of ascorbic acid in the grape berries. This method is based on the redox reaction of vitamin C with a standardized iodine solution and potentiometric detection of the endpoint, providing high analytical reproducibility and sensitivity for fruit matrices¹⁴. 2,6-dichlorophenolindophenol titration using 0.01 N iodine with starch indicator. Measurements were performed in duplicate for all samples, and results were expressed as milligrams of ascorbic acid per 100 grams of fresh weight (mg/100 g).

Statistical Analysis: Data processing was performed using GraphPad Prism 5.0 (GraphPad Software Inc., USA). Descriptive statistics (mean ± standard deviation) were calculated for each cultivar. Significant differences among means were assessed by one-way ANOVA followed by Tukey’s post-hoc test, with a significance threshold of $p \leq 0.05$ ¹⁵.

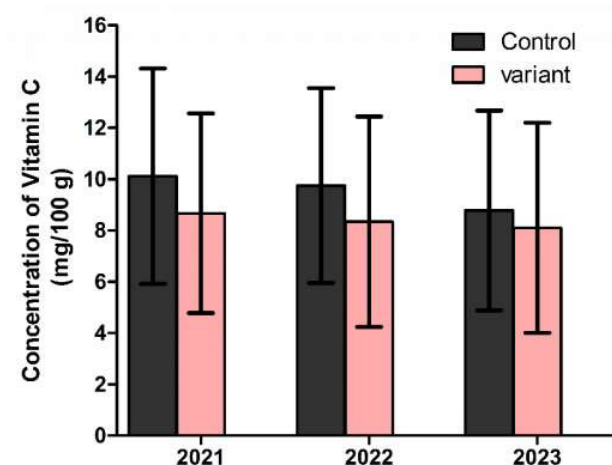
3. Results

Meteorological Factors: During 2021–2023, weather conditions varied, 2021 was warm and humid with a hydrothermal coefficient (HTC) = 1.4 and sum of active temperatures = 2245 °C, favoring the highest vitamin C accumulation 10.36 mg/100g. The lowest was 7.68 mg/100g, the vitamin C concentrations decreased with high temperature climate. RSD% ranged from 1.5% to 3.8% across all sample configurations, demonstrating excellent assay reproducibility alongside a highly reliable calibration curve with correlation coefficient $R^2 > 0.99$ (Figure 1).

Ripening Period: Average vitamin C levels among

Table 2. Climatic conditions during *Vitis vinifera* growing season (May–September) in Iraq, 2021–2023

Year	Sum of active temperatures (> +10 °C)	Hydrothermal Coefficient (HTC)	Climatic description
2021	2245 °C	1.4	Moderately warm and relatively humid season with adequate rainfall supporting stable vegetative growth.
2022	2529 °C	0.8	Hot and dry season characterized by limited precipitation and increased evapotranspiration stress.
2023	2658 °C	1.2	Hot season with semi-dry conditions; moderate moisture availability allowed acceptable berry formation.

**Figure 1:** Vitamin C levels in *Vitis vinifera* between control group and means of other genotypes (2021–2023). Differences not statistically significant.

introduced varieties exceeded the control by 5.3 % (0.49 mg/100 g) and were not dependent on ripening time (Figure 2)

Varietal Differences: Among genotypes, Hejazi (Control) started highest mean (9.86 mg/100 g) then little dropping indicates poor environmental resilience and high sensitivity to yearly variation. Tre Rash has moderate Vitamin C level (8.1 mg/100 g), but strong reduction after that suggesting limited biochemical stability. Eayn al-Thawr versus to He-

jazi, has high early ascorbate but significant loss it later that could be influenced by temperature fluctuations. Moon Drop is among the highest initial levels (9.8 mg/100 g) but slightly dropped suggesting good antioxidant potential but still declining under stress. Crimson has lower initial content (8.31 mg/100 g) with similar decline and has moderate variability; typical mid-range cultivar. Samarkandi has pattern mirrors Crimson with modest content with steady but low retention. Nubari has decent average and maintains moderate ascorbate concentration with minor inter-year deviation that suggesting stable genotype under Iraqi field conditions. Ari Uzumu has declining pattern; significant dropping in concentration that indicate higher sensitivity to temperature or storage conditions. Agh Gavra has fairly balanced, no extreme highs or lows with average Vitamin C stability among tested forms. Ashqarbasha has moderate baseline, but notable reduction and slight recovery later. Shekh-Nuradeen has distinct behavior then small rise before mild decline that implying better mid-season adaptability. Finally, Black Sadany has the most consistent performer overall, with high Vitamin C (9.7 mg/100 g), remaining relatively rich across all years with strong candidate for high-antioxidant breeding selection (Figure 3)

Color of berries: Vitamic C did not vary statistically significantly according to color: Red berries (8.10 mg/100 g), Pink berries (8.32 mg/100 g),

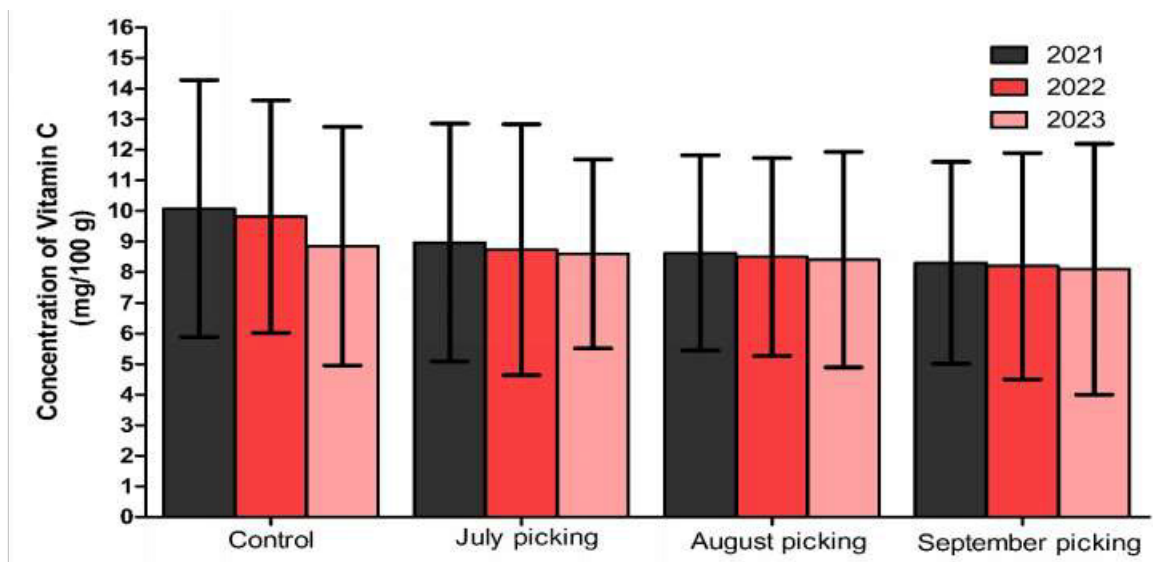


Figure 2: Vitamin C levels in *Vitis vinifera* depending on ripening periods: July, August, and September, over (2021–2023) versus control group.

White berries (8.84 mg/100 g) and Black berries (9.31 mg/100g) (Figure 4)

4. Discussion

The present study provides one of the most comprehensive biochemical evaluations of vitamin C variability among *Vitis vinifera* cultivars grown under Iraqi climatic conditions. The results demonstrate considerable genotypic differences in ascorbic acid accumulation, with concentrations ranging between 7.68 and 10.36 mg/100 g over the 2021–2023 growing seasons. These values fall within the expected global range reported for table and universal grape varieties, confirming that Iraq's agro-climatic environment supports the production of grapes with nutritionally relevant antioxidant potential¹⁶. Genotypic differences were the most influential factor determining vitamin C concentration across the studied cultivars. Black Sadany, Shekh-Nuradeen, and Moon Drop consistently recorded the highest ascorbate concentrations, averaging above 9 mg/100 g in comparison with Hejazi (control) concentration, whereas Tre Rash, and Eayn al-

Thawr showed lower and less stable values across the three years. These findings align with the established understanding that genetic background governs the biosynthetic efficiency of the L-galactose pathway, the principal metabolic route for ascorbic acid synthesis in plants^{17,18}. The superior performance of Black Sadany is notable, as it demonstrated the highest mean ascorbic acid stability across seasons, suggesting a genotype-specific resilience to temperature fluctuations. Comparable studies in Asian *Vitis* selections have also documented consistent ascorbate retention among dark-berried genotypes, attributing this to higher antioxidant enzyme activities and pigment-associated stabilization^{19,20}. Such metabolic robustness is particularly valuable for breeding programs aimed at developing cultivars suitable for arid regions where oxidative stress is frequent. Climatic variation, particularly temperature and hydrothermal coefficient (HTC), exerted a measurable influence on ascorbic acid accumulation. The warm-humid 2021 season (HTC = 1.4) favored the highest vitamin C accumulation (10.36 mg/100 g), whereas the hot, dry 2022 season (HTC = 0.8) coincided with a marked reduction in mean

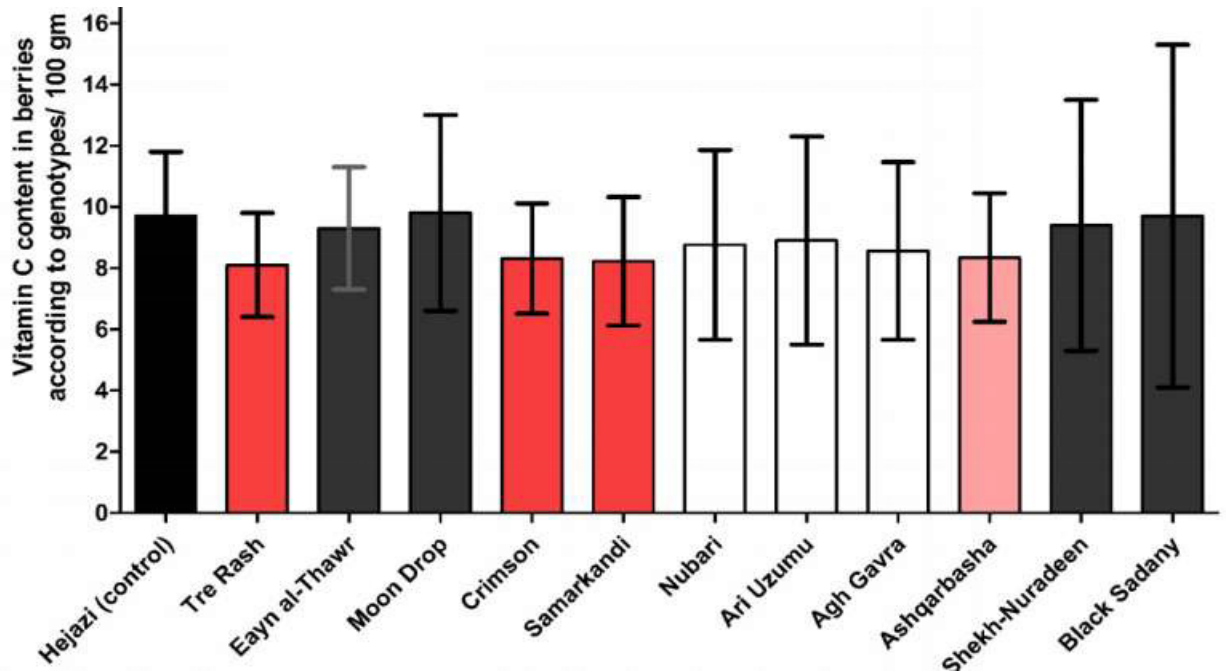


Figure 3: Vitamin C content in *Vitis vinifera* depending on genotypes (2021–2023).

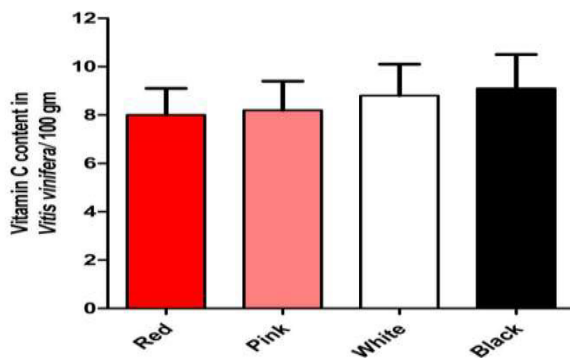


Figure 4: Vitamin C content in *Vitis vinifera* depending on color berries. Differences not statistically significant.

ascorbate levels. This inverse relationship between temperature stress and vitamin C concentration is consistent with the pattern observed a 62% reduction in Agh Gavra when temperatures exceeded 25 °C for extended periods. Elevated temperature in-

creases the rate of respiratory oxidation and ascorbate degradation, while limited water availability restricts substrate flow through the Smirnoff-Wheeler biosynthetic pathway²¹. Nevertheless, moderate recovery in 2023 (semi-dry, HTC = 1.2) indicates partial adaptation, suggesting that Iraqi grape varieties possess physiological plasticity to maintain a baseline antioxidant system even under suboptimal moisture conditions. These findings emphasize that while temperature stress negatively impacts ascorbic acid accumulation, irrigation management and canopy shading may mitigate its degradation. Contrary to some expectations, the present study found no statistically significant differences in vitamin C content across color groups, though a general trend persisted: black berries (9.31 mg/100 g) > white (8.84 mg/100 g) > pink (8.32 mg/100 g) > red (8.10 mg/100 g). This color-related gradient aligns partially with the known co-accumulation of anthocya-

nins and ascorbate, which function synergistically in ROS neutralization^{22,23}. The relatively higher ascorbate content of dark genotypes such as Black Sadany and Shekh-Nuradeen likely reflects this synergistic antioxidant interaction. However, the absence of statistically significant differences suggests that in these Iraqi genotypes, vitamin C biosynthesis may be more strongly influenced by genotype-specific enzymatic regulation than by pigment concentration alone. Anthocyanin content and other phenolic compounds while correlated with color that also influence the redox equilibrium of berry tissues, indirectly affecting vitamin C stability during maturation and post-harvest storage. The results therefore support the observation of Gorokhova et al.¹⁹, who reported that darker berry pigments enhanced antioxidant efficiency but not necessarily the total ascorbate pool, as much of the redox buffering occurred through polyphenol oxidation pathways rather than direct vitamin C accumulation. Temporal sampling over July–September revealed minimal differences in ascorbic acid levels across ripening periods. Average variation did not exceed $\pm 5\%$ between early- and late-harvested berries, indicating that ascorbic acid reaches near-maximum concentration during veraison and remains relatively stable until full ripeness. European researches were showing that vitamin C accumulation peaks early in grape maturation and declines only slightly upon full ripening²⁴. This stability is advantageous for table-grape production, as it ensures consistent antioxidant quality across the harvest window. The observation that berry ripening time did not significantly affect ascorbate levels under Iraqi field conditions suggests that environmental stressors (temperature and humidity) exert a stronger regulatory effect than developmental stage during the final 4–6 weeks of maturation. However, the coefficient of variation across Iraqi cultivars (18–24 %) is comparable to that reported in temperate zones, confirming that genotype-linked variability remains the dominant factor independent of climate. Interestingly, white-berry cultivars Agh Gavra and Ari Uzumu showed average ascorbate levels (8.84 mg/100 g) statistically similar to the control variety Hejazi, despite originating from

distinct eco-geographical regions. This indicates that color phenotype does not strongly determine antioxidant concentration in the Iraqi context, reinforcing the idea that ascorbate biosynthesis is polygenically regulated rather than solely dependent on pigmentation¹³. Vitamin C serves multiple biochemical functions for collagen formation, immune defense, and prevention of lipid peroxidation which are crucial for populations in semi-arid regions where oxidative stress and heat exposure are prevalent²⁵. The identification of high-ascorbate cultivars such as Black Sadany (9.7 mg/100 g) and Shekh-Nuradeen (9.4 mg/100 g) thus has nutritional and therapeutic relevance for local diets. These varieties can contribute meaningfully to daily vitamin C intake, considering that a 100 g serving provides $\sim 10\%$ of the WHO-recommended daily allowance for adults. Furthermore, the demonstrated genotypic stability of these cultivars under high-temperature stress suggests potential for their use in functional food products, juice processing, or nutraceutical formulations. Such applications would enhance the economic value of local vineyards and strengthen Iraq's participation in the antioxidant-rich food market, which is expanding globally due to increasing consumer awareness of dietary antioxidants²⁶. The observed genetic variability in ascorbate concentration offers a valuable foundation for grape-breeding programs targeting enhanced nutritional quality and stress tolerance. Selection for high-vitamin C genotypes can be integrated with traits for heat resistance and water-use efficiency, essential under future climate scenarios projected for Iraq and neighboring regions¹⁷. Moreover, environmental modulation through improved irrigation and canopy management could further stabilize ascorbate levels without compromising yield. In this context, Black Sadany, Shekh-Nuradeen, and Moon Drop can serve as donor parents for introgression of antioxidant stability into other locally adapted lines, supporting the dual goal of nutritional enhancement and climate resilience. Limitations of this study include the lack of advanced chromatographic validation (e.g., HPLC) for vitamin C quantification. Future studies should

incorporate multi-year datasets and broader cultivar representation.

5. Conclusion

This study establishes clear genotypic differentiation in ascorbic acid accumulation among twelve *Vitis vinifera* cultivars cultivated in Iraq between 2021 and 2023. Environmental stress particularly high temperature and low humidity was found to significantly reduce ascorbic acid concentrations, confirming the sensitivity of this metabolite to climatic extremes. However, the minimal influence of berry color and ripening time suggests that genotype and environment interactions, rather than phenotypic traits, are the primary determinants of vitamin C biosynthesis under Iraqi field conditions. Identifying and cultivating high-ascorbate genotypes will contribute to public health by providing accessible sources of dietary antioxidants and support agricultural sustainability by promoting climate-resilient cultivars.

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Conflicts of Interest

The author declares no conflicts of interest.

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