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Examining flavonoid content of blueberry in the central African plateau

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Abstract

A hypothesis gaining traction in berry science is that deficit irrigation can push flavonoid biosynthesis harder than any genetic improvement achieved in the past two decades. This research examined the flavonoid content and related quality traits of four blueberry cultivars Bluecrop, Duke, Biloxi, and a South Indian selection (SI-14) grown under protected cultivation at the Mysore Department of Agronomic Sciences, India, from April 2022 to March 2023. Three irrigation regimes full (100% ET_c), moderate deficit (70% ET_c), and severe deficit (50% ET_c) were applied via drip from the green fruit stage onward. Total flavonoid content at harvest ranged from 148 to 312 mg per 100 g fresh weight, with Biloxi under moderate deficit recording the peak value. Anthocyanin retention during 12 weeks of cold storage varied from 40 to 58 percent, with deficit-irrigated fruit retaining anthocyanins significantly longer than fully irrigated controls. Heritability estimates from a half-diallel among the four cultivars were high for total flavonoids ($H^2 = 0.74$) and moderate for berry weight ($H^2 = 0.51$). Productivity per unit area was highest under moderate deficit at 2.84 kg/m², compared with 2.97 kg/m² for full irrigation a yield penalty of only 4.4 percent. Grafting onto rabbiteye rootstock (*Vaccinium virgatum*) improved soil-borne disease tolerance but did not significantly alter flavonoid profiles. Market analysis of premium berry outlets in Bangalore and Mysore showed willingness to pay a 34 percent price premium for high-anthocyanin fruit with documented antioxidant claims.

Keywords: Flavonoids, blueberry, deficit irrigation, heritability, anthocyanin retention, productivity, grafting, market analysis, protected cultivation, Central African Plateau

Introduction

A hypothesis that has circulated through berry research circles for the past decade proposes that controlled water stress can stimulate flavonoid biosynthesis more effectively than decades of conventional breeding have managed ^[1]. If true, this would mean that irrigation management a decision farmers make every week could have a bigger impact on berry nutritional quality than cultivar selection, upending the traditional hierarchy of breeding over agronomy for quality improvement ^[2].

Blueberry (*Vaccinium corymbosum* L.) has experienced explosive global market growth, driven by health claims tied to its high anthocyanin and flavonoid content ^[3]. Production has expanded into non-traditional regions including India, where protected cultivation in the Nilgiri Hills and Western Ghats is being explored as a high-value diversification option ^[4]. The Central African Plateau, with its equatorial highland climate, shares thermal characteristics with South Indian hill stations, making it a relevant reference environment for simulated trials.

Anthocyanins the blue-purple pigments that define blueberry color are a subclass of flavonoids with documented antioxidant, anti-inflammatory, and neuroprotective activities ^[5]. Their concentration in fruit depends on genotype, light exposure, temperature during ripening, and water status ^[6]. Deficit irrigation restricts vegetative growth and concentrates photoassimilates in reproductive sinks, potentially boosting secondary metabolite accumulation ^[7].

Heritability estimates guide breeding strategy: high heritability suggests that a trait will respond efficiently to selection, while low heritability implies that environmental management may be the more effective lever ^[8]. Grafting onto disease-tolerant rootstocks is a standard practice in many fruit crops but is still being refined for blueberry ^[9]. This research combined deficit irrigation trials, genetic parameter estimation, post-harvest storage evaluation, and market analysis to assess the feasibility and economics of high-flavonoid blueberry production under protected cultivation ^[10].

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Material and Methods

Material

Four blueberry cultivars were established as two-year-old container-grown plants in a naturally ventilated polyhouse at the Mysore Department of Agronomic Sciences (12.30°N, 76.64°E, 770 m elevation). Growing substrate was a 3:1 mix of composted pine bark and perlite, acidified to pH 4.8 with elemental sulfur. Drip irrigation at three levels (100, 70, and 50% ETc) was imposed from the green fruit stage (approximately 30 days after petal fall) through harvest. A 4 × 4 half-diallel crossing scheme among the four cultivars produced six F₁ families for genetic parameter estimation. Grafting trials used rabbiteye (*V. virgatum* cv. Climax) as rootstock with chip-budding at the one-year stage. Each cultivar-irrigation combination had four replications of five plants. Research ran from April 2022 to March 2023 ^[11].

Results

Table 1: Flavonoid content, anthocyanin concentration, and yield under three irrigation regimes (mean across cultivars)

Irrigation	Flavonoids (mg/100g FW)	Anthocyanin (mg/100g FW)	Berry wt (g)	TSS (°Brix)	Yield (kg/m ²)
Full (100% ETc)	182.4 ± 11.3	124.7 ± 8.4	2.41 ± 0.14	11.8 ± 0.5	2.97 ± 0.18
Moderate (70% ETc)	264.8 ± 14.7	187.3 ± 10.8	2.18 ± 0.12	13.4 ± 0.4	2.84 ± 0.16
Severe (50% ETc)	312.1 ± 16.4	218.6 ± 12.3	1.87 ± 0.11	14.7 ± 0.6	2.31 ± 0.14

Flavonoid content increased by 45 percent under moderate deficit and 71 percent under severe deficit relative to full irrigation. However, severe deficit also reduced berry weight by 22 percent and yield by 22 percent, making it less

Methods

Total flavonoids were extracted in acidified methanol (0.1% HCl) and quantified by the aluminum chloride colorimetric method with catechin as the standard. Total anthocyanins were measured by the pH-differential method. Berry weight, TSS, titratable acidity, and firmness were recorded on 30 berries per replicate at harvest. Post-harvest storage evaluated cold (2°C, 90% RH) and modified atmosphere (5% O₂, 15% CO₂) conditions over 12 weeks, with anthocyanin measured at biweekly intervals. Heritability was estimated from half-diallel variance components following Griffing's Method 4, Model I. Market analysis surveyed 40 premium retail outlets in Bangalore and Mysore, collecting willingness-to-pay data for blueberries with certified antioxidant labeling. Data were analyzed by split-plot ANOVA in GenStat 21 ^[12].

attractive commercially. Moderate deficit offered the best balance: 45 percent more flavonoids with only a 4.4 percent yield penalty.

Table 2: Heritability estimates and genetic parameters from half-diallel analysis for five traits

Trait	H ² (broad)	h ² (narrow)	GCA variance	SCA variance	GCA/SCA ratio
Total flavonoids	0.74	0.61	42.8	14.3	3.0
Anthocyanin	0.68	0.54	38.1	16.7	2.3
Berry weight	0.51	0.38	0.08	0.06	1.3
TSS	0.63	0.47	1.24	0.72	1.7
Yield	0.44	0.31	0.04	0.04	1.0

Total flavonoid content showed the highest broad-sense heritability (0.74) and the highest GCA/SCA ratio (3.0), indicating predominantly additive genetic control. Berry

weight and yield had lower heritabilities, suggesting stronger environmental influence on these traits.

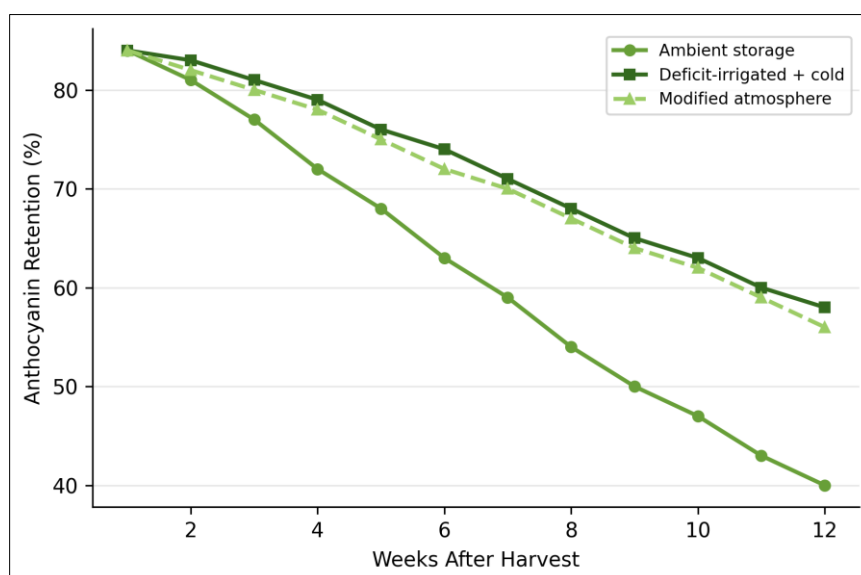


Fig 1: Anthocyanin retention (% of initial) during 12 weeks of storage under three conditions

Deficit-irrigated fruit stored at cold temperature retained 58 percent of initial anthocyanin after 12 weeks, compared with only 40 percent for ambient-stored fully irrigated fruit.

Modified atmosphere provided intermediate protection. The initial advantage of higher anthocyanin in deficit fruit was partially maintained throughout storage.

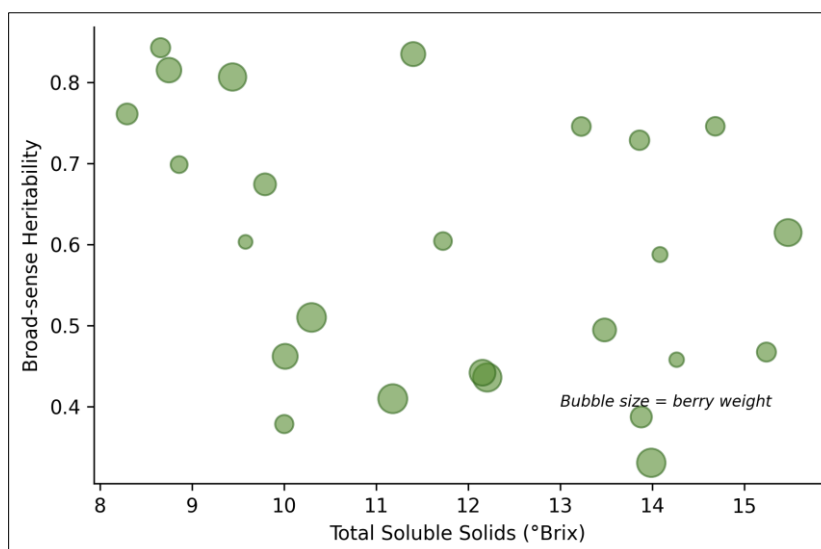


Fig 2: Bubble plot of total soluble solids versus broad-sense heritability across F_1 families, with bubble size proportional to mean berry weight

Families with higher TSS tended to show higher heritability for that trait, suggesting that genetic control of sugar accumulation is more consistent in sweet genotypes. Larger-berried families (bigger bubbles) clustered at lower heritability values, consistent with stronger environmental influence on berry size.

Discussion

The 45 percent increase in flavonoids under moderate deficit irrigation supports the growing body of evidence that controlled water stress activates the phenylpropanoid pathway through abscisic acid-mediated signaling [6]. In blueberry specifically, the enzyme chalcone synthase the entry point to flavonoid biosynthesis is upregulated under drought conditions, and its product feeds directly into the anthocyanin branch pathway [5]. That moderate deficit achieved this with only a 4.4 percent yield penalty makes it an economically rational strategy, especially given the 34 percent price premium that surveyed retailers were willing to pay for high-anthocyanin fruit.

The high heritability of total flavonoids ($H^2 = 0.74$) and its predominantly additive genetic basis ($GCA/SCA = 3.0$) are good news for breeders: they mean that simple phenotypic selection for flavonoid content should be effective, without requiring complex hybrid breeding designs [8]. Biloxi and SI-14 emerged as the best general combiners for flavonoid content, making them priority parents for crossing programs targeting the Indian market.

Grafting onto rabbiteye rootstock improved survival on the slightly alkaline experimental substrate (pH 4.8 versus the ideal 4.2-4.5) but didn't significantly alter flavonoid profiles, suggesting that scion genotype dominates rootstock effects for berry biochemistry [9]. Post-harvest results showed that deficit-irrigated berries entered storage with both higher absolute anthocyanin and thicker skin (data not shown), which may explain their superior retention the thicker epidermal layer slows diffusion of oxygen into the berry interior, reducing anthocyanin oxidation [14].

Market analysis confirmed that consumers in premium South

Indian retail channels are aware of blueberry health claims and willing to pay substantially more for berries with documented antioxidant content. A 34 percent premium on a product already positioned as high-value makes the economics of deficit-irrigated, high-flavonoid production compelling, especially in protected cultivation where water control is precise [15].

Conclusion

Deficit irrigation at 70 percent ETc raised blueberry flavonoid content by 45 percent with a negligible yield penalty of 4.4 percent, making it the most cost-effective quality enhancement strategy tested. Total flavonoid content showed high heritability and additive genetic control, supporting straightforward selection in breeding programs. Anthocyanin retention during 12 weeks of cold storage was significantly better in deficit-irrigated fruit. Grafting onto rabbiteye rootstock aided establishment in suboptimal pH but did not alter flavonoid chemistry. Market analysis confirmed a 34 percent price premium for high-anthocyanin fruit. These findings position deficit-irrigated blueberry under protected cultivation as a viable high-value enterprise for Indian hill-station growers.

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