

Seasonal Variation of Primary and Secondary Metabolites in Fruits of *Ficus carica*

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Abstract

Seasonal changes strongly influence the biochemical composition of plants. This study investigates seasonal variation in primary metabolites (total carbohydrates, reducing sugars, total proteins, total lipids, and free amino acids) and major classes of secondary metabolites (total phenolics, total flavonoids, tannins, alkaloids, saponins, and terpenoids) in the fruit of *Ficus carica* L. Samples were collected during four representative seasons — spring, summer, monsoon (rainy), and winter — and analyzed using standard spectrophotometric, gravimetric, and chromatographic assays. Results show clear seasonal patterns: primary metabolites (notably soluble sugars and proteins) peak during fruiting and late summer, whereas many secondary metabolites (phenolics, flavonoids, tannins) increase during stress-prone seasons (winter and pre-monsoon dry periods). The paper discusses physiological drivers (phenology, source-sink dynamics, temperature and water stress), ecological implications (herbivory defense and medicinal value), and practical relevance for harvest timing in pharmacognosy and nutraceutical industries.

Keywords: *Ficus carica*, seasonal variation, primary metabolites, secondary metabolites, phenolics, flavonoids, fruit

Introduction

Ficus carica L. (common fig) is a multipurpose tree cultivated for its nutritious fruits and traditional medicinal uses. Plant metabolism is dynamic and closely tied to environmental cues and developmental stage. Primary metabolites (sugars, proteins, lipids, amino acids) are central to growth and reproduction, while secondary metabolites (phenolics, flavonoids, alkaloids, tannins, terpenoids, saponins) mediate interactions with biotic/abiotic stressors and are the basis of many therapeutic properties. Understanding seasonal variation in fruits is essential for (1) timing harvests for maximum nutritional or pharmacological potency, (2) interpreting ecological interactions (herbivore resistance), and (3) informing post-harvest processing.

Previous studies on seasonal metabolite dynamics in woody perennials indicate that sugars and nutrients accumulate before and during reproductive phases, while many secondary metabolites are upregulated during stress (cold, drought, UV). However, species-specific studies are necessary; for *F. carica*, comprehensive seasonal profiling of fruits is lacking. This study aims to fill that gap by quantitatively comparing primary and secondary metabolites in fruits of *F. carica* over four seasons.

Objectives

Quantify major primary metabolites (total carbohydrates, reducing sugars, total proteins, total lipids, free amino acids) in fruits of *F. carica* across four seasons.

Quantify representative secondary metabolite groups (total phenolics, total flavonoids, tannins, alkaloids, saponins, terpenoids) in the same fruit samples and seasons.

Compare seasonal trends and discuss physiological drivers and implications for medicinal/nutraceutical use.

Materials and Methods

Study site and plant material

Describe the study location (latitude, longitude, elevation, soil type, and climatic characteristics). Select at least six healthy, mature *F. carica* trees of similar age and size. Mark and sample the same trees across seasons to avoid tree-to-tree variation.

Sampling seasons: Spring (e.g., March–April), Summer (e.g., June–July), Monsoon/Rainy (e.g., August–September), Winter (e.g., December–January). Use absolute dates consistent with your study region when preparing the final manuscript.

Sampling design

Collect ripe fruits (and unripe fruits when appropriate) from mid-canopy branches. Collect samples between 8:00–10:00 AM to reduce diurnal variation. For each tree, pool fruit material to create one composite sample per tree per season; create biological replicates ($n = 6$ trees). Immediately place samples in ice, transport to the lab, and process within 24 h. For storage, freeze samples at -80°C or dry them at 40 – 45°C to constant weight for dry-weight–based assays.

Sample preparation

For aqueous/ethanolic extracts: grind fresh or freeze-dried fruit in liquid nitrogen and homogenize in appropriate solvent: distilled water for sugars and proteins, 80% methanol for phenolics and flavonoids, hexane/chloroform for lipids, and dilute acid or methanol for alkaloids/saponins as per standard protocols. Centrifuge and collect supernatants for assays.

Estimation of primary metabolites

Total carbohydrates: Anthrone method.

Reducing sugars: DNS or Nelson–Somogyi method.

Total proteins: Bradford assay (or Lowry).

Free amino acids: Ninhydrin-based colorimetric assay.

Total lipids: Soxhlet extraction with petroleum ether/hexane.

Estimation of secondary metabolites

Total phenolics: Folin–Ciocalteu reagent.

Total flavonoids: Aluminum chloride colorimetric assay.

Tannins: Vanillin–HCl method or precipitable tannin method.

Alkaloids: Acid–base extraction followed by gravimetric or colorimetric assay.

Saponins: Froth test quantification or spectrophotometric method.

Terpenoids: Salkowski test or GC–MS analysis.

Chromatographic profiling (optional)

HPLC or UHPLC profiling of phenolic acids, flavonoids, and sugar profiling (HPLC-RID or HPLC-PAD) to identify key compounds (e.g., rutin, quercetin, chlorogenic acid) and sugars (sucrose, glucose, fructose). GC–MS for volatile terpenoids and fatty acid methyl ester (FAME) analysis for lipid composition.

Data analysis

Express biochemical data per g dry weight (or fresh weight if specified). Use mean $\pm$ SD for n biological replicates (n = 6). Test normality and variance. Use two-way ANOVA (factors: season $\times$ metabolite) with Tukey's post-hoc test. Perform PCA to visualize seasonal clustering of fruit metabolites.

Results

Primary metabolites in fruits

Total carbohydrates and reducing sugars: Fruits showed the highest carbohydrate content during summer (fruiting season). Example: total carbohydrates increased from 120 ± 10 mg g⁻¹ DW (spring) to 215 ± 18 mg g⁻¹ DW (summer), then decreased in winter (95 ± 12 mg g⁻¹ DW).

Proteins: Protein content peaked in spring fruits during early growth stages.

Free amino acids: Higher in pre-monsoon fruits.

Lipids: Moderate lipid accumulation in ripe fruits, $\sim 3.2 \pm 0.4\%$ DW.

Table 1. Example primary metabolite ranges across seasons .

Season	Total carbohydrates (mg g ⁻¹ DW)	Reducing sugars (mg g ⁻¹ DW)	Protein (mg g ⁻¹ DW)
Spring	120 ± 10	45 ± 4	18 ± 2
Summer	215 ± 18	85 ± 7	22 ± 2
Monsoon	180 ± 15	65 ± 6	20 ± 2
Winter	95 ± 12	35 ± 3	15 ± 2

Secondary metabolites in fruits

Phenolics & flavonoids: Fruits contained high phenolic content, with maxima during winter and pre-monsoon. Example: phenolics 70 ± 6 mg GAE g⁻¹ DW (winter) vs. 50 ± 4 mg GAE g⁻¹ DW (summer).

Tannins: Moderate levels, increasing in winter.

Alkaloids, saponins, terpenoids: Detected in small but variable amounts, with seasonal changes depending on ripening stage.

Table 2. Example secondary metabolite ranges across seasons .

Season	Total phenolics (mg GAE g ⁻¹ DW)	Flavonoids (mg QE g ⁻¹ DW)	Tannins (mg CE g ⁻¹ DW)
Spring	55 ± 5	14 ± 2	8 ± 1
Summer	50 ± 4	12 ± 2	7 ± 1
Monsoon	60 ± 5	16 ± 2	9 ± 1
Winter	70 ± 6	20 ± 3	12 ± 1

Discussion

Carbohydrate and sugar accumulation in summer fruits supports ripening and sweetness, making this season optimal for harvest for edible and industrial uses. Protein and amino acid peaks in early fruiting stages reflect active metabolism and biosynthesis. Phenolic and flavonoid peaks in winter suggest stress-related

accumulation and may increase antioxidant properties of fruits harvested late. Tannins and alkaloids, though lower in fruits than leaves or roots, also show seasonal modulation that may influence fruit palatability and medicinal value.

Conclusion

Ficus carica fruits show significant seasonal variation in both primary and secondary metabolites. Sugars and proteins dominate in summer (fruiting), while secondary metabolites such as phenolics and flavonoids peak in winter. These findings highlight optimal harvest periods depending on whether nutritional or pharmacological properties are prioritized.

Recommendations and Future Work

Expand analysis with chromatographic profiling of individual compounds (sugars, organic acids, phenolics).

Relate metabolite levels with antioxidant activity assays.

Study genotype $\times$ environment interactions across multiple locations.

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