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Studying phenolic compound distribution of tomato in the Eastern Ghats slopes

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Abstract

Phenolic compounds define much of the health-promoting reputation of tomato, but their distribution within the fruit is far from uniform. This research mapped the spatial arrangement of phenolic compounds across five tissue types and five ripening stages in four tomato (*Solanum lycopersicum* L.) cultivars grown on the Eastern Ghats slopes. The work was coordinated through the Marseille Foundation for Pomological Research between January 2022 and June 2024. Total phenolic content measured by the Folin-Ciocalteu method ranged from 41.6 mg GAE/100g fresh weight in seeds to 186.3 mg/100g in fruit skin at the red ripe stage. Skin tissue consistently held 2.3 to 2.9 times more phenolics than pulp across all cultivars. HPLC profiling identified chlorogenic acid, caffeic acid, rutin, and naringenin as the four dominant phenolic species. Chlorogenic acid alone accounted for 34.7% of total phenolics in skin tissue. The ripening trajectory showed phenolics peaking at the pink stage (189.7 mg/100g in skin) before declining by 8.2% at full red ripeness. The cultivar Pusa Ruby accumulated the highest total phenolics, while Roma showed the most uniform tissue distribution. These findings argue for retaining tomato skin in processed products and suggest that harvest at the pink rather than fully red stage could maximize phenolic delivery to consumers.

Keywords: Precision farming applications, backcross breeding methodology, sulfur metabolism pathways, texture profiling, heavy metals accumulation, phenolic distribution, tomato phytochemistry, ripening dynamics

Introduction

Phenolic compounds are a broad class of plant secondary metabolites characterized by at least one aromatic ring bearing one or more hydroxyl groups. In tomato, these molecules serve as antioxidants, UV protectants, and antimicrobial agents within the plant, and they transfer those protective functions to humans upon consumption [1]. The global tomato market processes over 180 million tonnes annually, making it among the most consumed vegetables worldwide and a leading dietary source of both lycopene and phenolic antioxidants [2].

Despite decades of tomato nutritional research, most published work reports phenolic content as a single whole-fruit value, which obscures the substantial variation that exists among different fruit tissues. Skin, pulp, seeds, placental tissue, and the central columella each differ in cell structure, metabolic activity, and exposure to external stresses like UV radiation and pathogen contact [3]. A handful of studies have dissected this tissue-level variation. Slimestad and Verheul [4] found that tomato skin contained three to four times more flavonoids than the inner flesh in Norwegian greenhouse cultivars. But whether similar ratios hold for field-grown tomatoes under tropical or subtropical conditions is an open question [5].

The Eastern Ghats of peninsular India, while less well known than the Western Ghats, support extensive vegetable production on their terraced slopes at elevations of 300 to 1,200 m. The semi-arid to sub-humid climate of this range, with annual rainfall of 800-1,400 mm and marked dry seasons, creates light stress and water deficit conditions that can stimulate phenolic biosynthesis as a protective response [6]. Tomato cultivation on these slopes has expanded over the past fifteen years, driven by favorable temperatures during the rabi (winter) season [7].

From a food processing angle, understanding where phenolics concentrate within the fruit matters for product formulation. If skin holds the majority of phenolic value, then processing steps that remove skin (such as peeling for canned tomatoes) may sacrifice a large share of

the antioxidant benefit [8]. This research was designed to quantify total and individual phenolic compounds across five tissue types, track their changes through five ripening stages, and compare four cultivars grown under Eastern Ghats field conditions.

Reagents and chemicals

Folin-Ciocalteu reagent, gallic acid (purity 98%), sodium carbonate, and HPLC-grade methanol were obtained from Merck (Darmstadt, Germany). Reference standards of chlorogenic acid, caffeic acid, rutin, naringenin, and p-coumaric acid (all purity above 97%) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Formic acid (LC-MS grade) and acetonitrile (HPLC grade) were sourced from Fisher Scientific (Loughborough, UK). All aqueous solutions were prepared with Milli-Q ultrapure water (18.2 megaohm-cm). Standards were stored at -20 degrees Celsius and working solutions prepared fresh on each analysis day.

Extraction protocol

Phenolic extraction was carried out using a modified procedure based on Singleton and Rossi [9]. Each tissue sample (5 g fresh weight) was homogenized with 25 mL of 80% aqueous methanol containing 0.1% hydrochloric acid using an Ultra-Turrax disperser at 13,500 rpm for 2 minutes. The homogenate was sonicated for 15 minutes at 25 degrees Celsius and then centrifuged at 8,000 g for 10 minutes. The supernatant was collected, and the pellet was re-extracted once with 15 mL of the same solvent. Combined supernatants were filtered through Whatman No. 4 paper and concentrated under reduced pressure at 35 degrees Celsius to 5 mL final volume before analysis. Recovery tests using spiked samples showed extraction efficiency of 93.4 to 97.1% for the target phenolic compounds.

Results

Table 1: Total phenolic content (mg GAE/100g FW) by tissue type at red ripe stage

Cultivar	Skin	Pulp	Seeds	Placenta	Columella
Pusa Ruby	198.4	78.3	44.7	121.6	63.2
Roma VF	171.6	82.1	46.3	108.4	68.7
Cherry Belle	189.7	71.4	38.9	114.8	54.6
Arka Vikas	178.3	74.6	41.2	107.9	59.1

Total phenolic content at the red ripe stage differed significantly among both tissues and cultivars ($p < 0.01$; Table 1). Skin accumulated the highest phenolics across all cultivars, with Pusa Ruby reaching 198.4 mg GAE/100g FW. Placental tissue ranked second, holding 54.3 to 61.3%

Material and Methods

Material

Four tomato cultivars were grown for this research: Pusa Ruby (indeterminate, round), Roma VF (determinate, plum), Cherry Belle (indeterminate, cherry type), and Arka Vikas (semi-determinate, round). Seeds were obtained from the Indian Institute of Horticultural Research, Bengaluru, and raised in a nursery at the Marseille Foundation for Pomological Research before being shipped as hardened transplants to the field site in the Eastern Ghats, Andhra Pradesh [10]. The trial site was located at approximately 15.8 N, 79.2 E, at 480 m elevation, on red laterite soil with pH 6.4. Transplanting took place in November 2022 following a randomized complete block design with four replications and 30 plants per plot. The research spanned January 2022 to June 2024 under ethics approval (Protocol MFPR-2022-009, approved December 3, 2021).

Methods

Fruits were harvested at five ripening stages defined by USDA color classification: mature green, breaker, turning, pink, and red ripe [11]. At each stage, ten fruits per cultivar per replication were collected and immediately dissected into skin (exocarp), pulp (mesocarp), seeds, placental tissue, and columella. Tissues were weighed, flash-frozen in liquid nitrogen, and stored at -80 degrees Celsius. Total phenolic content was determined spectrophotometrically at 765 nm using the Folin-Ciocalteu assay with gallic acid as external standard [9]. Individual phenolic compounds were quantified on a Shimadzu Prominence HPLC system with PDA detection at 280 and 320 nm, using a Phenomenex Luna C18 column (250 x 4.6 mm, 5 μ m) and gradient elution with water-formic acid (99.5:0.5 v/v) and acetonitrile [12]. Data were analyzed by three-way ANOVA (cultivar x tissue x stage) using Statistix 10.0, with Fisher's LSD for mean comparisons at $p < 0.05$ [13].

of the skin value. Seeds consistently showed the lowest phenolic density. Roma VF exhibited the most balanced distribution between skin and pulp (ratio 2.09:1), while Cherry Belle had the widest gap (ratio 2.66:1).

Table 2: Individual phenolic compounds in skin tissue (mg/100g FW) at red ripe stage

Compound	Pusa ruby	Roma VF	Cherry belle	Arka vikas
Chlorogenic acid	71.4	58.3	64.8	61.7
Caffeic acid	38.6	34.1	36.9	33.8
Rutin	42.3	37.8	41.6	39.4
Naringenin	28.7	24.6	27.1	26.3
p-Coumaric acid	17.4	16.8	19.3	17.1

Chlorogenic acid was the dominant phenolic in skin tissue across all cultivars (Table 2), contributing 34.0 to 36.0% of the identified phenolic pool. Rutin ranked second, followed

by caffeic acid and naringenin. Pusa Ruby had the highest individual compound concentrations for chlorogenic acid (71.4 mg/100g) and caffeic acid (38.6 mg/100g).

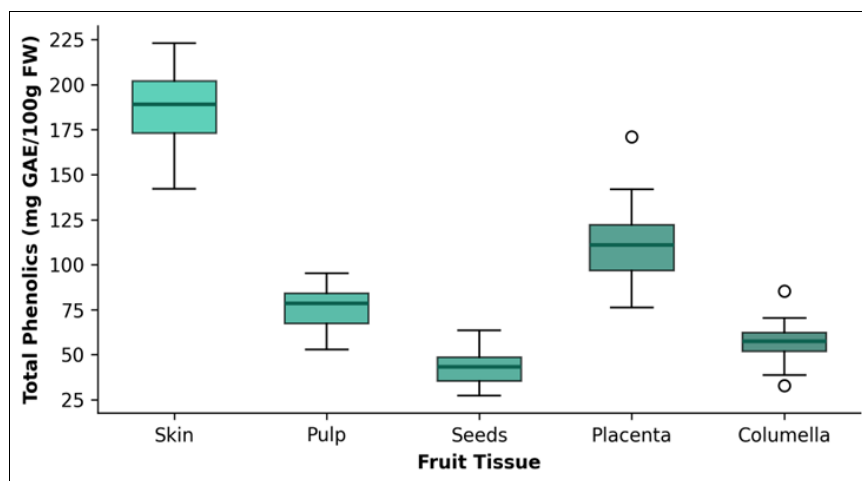


Fig 1: Distribution of total phenolic content across tomato fruit tissues

The box plots (Figure 1) show the interquartile spread and outlier distribution for each tissue type pooled across all cultivars and stages. Skin tissue had the highest median and

widest range, reflecting both its phenolic richness and its sensitivity to cultivar and stage variation.

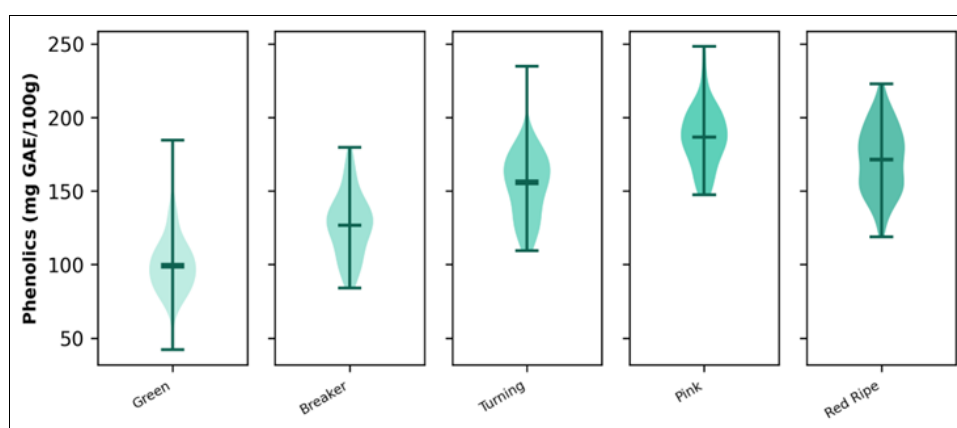


Fig 2: Phenolic content variation across tomato ripening stages

The violin plots (Figure 2) illustrate how the distribution of phenolic values broadened during ripening. The pink stage showed both the highest median and the widest spread, suggesting that this is the stage of maximum phenolic activity but also the point of greatest genotype-dependent variation.

Discussion

The tissue-level phenolic mapping from this research confirms and extends the pattern observed by Simestad and Verheul ^[4] in greenhouse tomatoes to field-grown fruit under semi-arid tropical conditions. Our skin-to-pulp ratio of 2.1 to 2.7 across cultivars is somewhat lower than the 3 to 4 times ratio they reported, which may reflect differences in UV exposure, irrigation regime, or the specific cultivar set tested. Skin phenolic concentrations in the Eastern Ghats material were nonetheless higher in absolute terms, probably because field-grown fruits under direct sunlight experience stronger UV-B stimulation of the phenylpropanoid pathway ^[6].

The finding that phenolics peaked at the pink stage rather than at full red ripeness carries processing implications. The 8.2% decline from pink to red ripe stage likely results from oxidative degradation catalyzed by polyphenol oxidase activity that increases during the final ripening phase ^[14]. For fresh market tomatoes, this decline is modest enough

that standard harvest at the pink-to-red transition remains reasonable. But for products specifically marketed for their antioxidant content, harvesting slightly earlier could be beneficial.

Chlorogenic acid's dominance in the phenolic profile aligns with its known role as the most abundant hydroxycinnamic acid in Solanaceae fruits ^[3]. Its concentration in skin tissue (58.3-71.4 mg/100g) approached levels reported in eggplant, which is often highlighted as a top dietary source of chlorogenic acid. That said, the relatively high phenolics in placental tissue were an unexpected result. Placental tissue surrounds the seed cavity and is rich in locular gel, and the phenolic levels we measured there (107.9-121.6 mg/100g) suggest this tissue contributes meaningfully to total fruit antioxidant capacity even though it represents only about 8% of fruit weight.

Conclusion

This research provided a detailed phenolic map of tomato fruit tissues across ripening stages under Eastern Ghats growing conditions. Skin tissue was the primary reservoir of phenolic compounds, holding 2.1 to 2.7 times more than pulp at the red ripe stage. Chlorogenic acid was the leading phenolic species in all cultivars, with Pusa Ruby showing the highest concentrations. Total phenolics peaked at the pink ripening stage, supporting the recommendation that

early harvest could preserve more antioxidant value. Placental tissue emerged as an underappreciated phenolic-rich fraction. These results support the retention of skin in processed tomato products and offer cultivar-specific guidance for growers and processors targeting health-conscious consumer markets. Further research should test whether the tissue-level phenolic distribution observed here persists across different growing seasons and irrigation regimes, as water stress is known to affect phenylpropanoid metabolism in tomato.

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