

Physicochemical and metabolite profiling of three *Capsicum* species from West Sumatra, Indonesia

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Abstract. Yenrina R, Weliardi, Putra AR, Humaira SS, Anggraini T. 2026. Physicochemical and metabolite profiling of three *Capsicum* species from West Sumatra, Indonesia. *Biodiversitas* 27 (4): d270405. <https://doi.org/10.13057/biodiv/d270405>. Fresh chili quality and bioactivity vary by species and growing conditions, but integrated data for West Sumatra remain limited. This study provides the first integrated physicochemical-metabolite characterization of three *Capsicum* species commonly cultivated and marketed in the region: Red Curly chili (*Capsicum annum*; RC), Bird's Eye chili (*Capsicum frutescens*; BE), and Ghost Pepper (*Capsicum chinense*; GP). Fruit morphometrics, surface color (CIELAB), proximate composition, and antioxidant activity (DPPH assay at 1000 ppm) were evaluated using a descriptive analytical approach, and metabolite profiling was performed using untargeted LC-MS/MS QTOF with putative identification based on MS/MS spectral library matching (MSI level 2). These three species exhibit distinct morphological and color parameters, with RC exhibiting the highest red intensity (a^* and chroma) and BE the lowest lightness (L^*). All samples exhibited high moisture content (78.70-82.46%) and high carbohydrate content, with low fat and protein levels. Strong antioxidant activity was detected across species (83.38-86.67%). Untargeted LC-MS/MS analysis of single composite samples per species identified 147 putative metabolites (MSI level 2) (73 in RC, 78 in BE, and 70 in GP), and qualitative differences in metabolite detection patterns were observed among species. Glycosylated terpenoid-related compounds were detected in BE, abietic-type diterpenoids were detected in GP, and RC showed the presence of apocarotenoid-related compounds such as β -ionone together with diverse diterpenoid annotations. As analyses were based on single composite samples and putative metabolite annotation (MSI level 2), results should be interpreted as qualitative trends rather than statistically validated differences. Overall, this study establishes a region-specific integrated physicochemical-metabolite profile of *Capsicum* species from West Sumatra, providing a structured reference for future quantitative metabolomics, functional evaluation, and cultivar differentiation.

Keywords: *Capsicum*, DPPH radical scavenging, proximate composition, untargeted LC-MS/MS, West Sumatra chilies

Abbreviations: BE: Bird's Eye chili (*Capsicum frutescens*), RC: Red Curly chili (*Capsicum annum*), GP: Ghost Pepper (*Capsicum chinense*)

INTRODUCTION

Chili (*Capsicum* spp.), domesticated in Mesoamerica, is now one of the most widely cultivated vegetable and spice crops worldwide (Saleh et al. 2018; Duranova et al. 2022). Its adaptability to diverse agroecological conditions has made *Capsicum* an important component of global food systems (Finger and Pereira 2016; Antonio et al. 2018). In Indonesia, chili is a major horticultural commodity and an essential ingredient in daily cuisine (Kurniawan 2023). High national production highlights the importance of understanding regional variation in chili quality and composition (Duranova et al. 2022).

Despite its economic importance, chili quality is not uniform across growing regions. Environmental factors such as climate, altitude, soil properties, and cultivation practices influence plant metabolism and fruit development, resulting in region-specific physicochemical and biochemical traits. Therefore, chili grown in different locations cannot be assumed to have identical quality characteristics, even

within the same species (Butt et al. 2021; Duranova et al. 2022).

West Sumatra is an important chili-producing and marketing region in Indonesia, supplying fresh chili for local consumers and nearby areas (Hamidah et al. 2020). Its distinct agroecological conditions may affect plant growth, fruit development, and metabolite accumulation, thereby contributing to variation in chili quality and chemical composition (Aryani et al. 2022). However, comprehensive data on the physicochemical and biochemical composition of fresh chili cultivated in this region remain limited. From a biodiversity perspective, variation among *Capsicum* species grown in local agroecosystems reflects interactions between genetic background and environmental conditions, which may produce distinct biochemical expressions related to local adaptation.

Chili peppers contain diverse bioactive compounds that influence their sensory properties and nutritional value. Vitamins A, C, and E, together with carotenoids and phenolic compounds, are major contributors to antioxidant activity in chili fruits (Martínez-Ispizua et al. 2021; Sadef

et al. 2022). Ascorbic acid and capsaicinoids, including capsaicin and related compounds, are important metabolites, with capsaicinoids responsible for pungency and associated bioactivities (Orobiyi et al. 2015; Clark and Lee 2016). Their accumulation is influenced by both genetic and environmental factors, resulting in substantial phytochemical variation within the genus *Capsicum* (Hayano-Kanashiro et al. 2016; de Jesus Luna-Ruiz et al. 2018). Therefore, integrated physicochemical characterization combined with LC-MS/MS-based metabolite profiling is needed to clarify possible species- and environment-associated biochemical variation.

Previous studies on *Capsicum* diversity have mainly emphasized morphological characterization. In Indonesia, including West Sumatra, local chili has largely been evaluated using qualitative and quantitative morphological descriptors to assess diversity and phylogenetic relationships (Ferniah and Pujiyanto 2017; Suliansyah et al. 2023; Zulkarnain et al. 2023). In contrast, metabolomics-based studies from outside Indonesia have shown substantial variation among species, cultivars, and domestication groups in phenolic compounds, capsaicinoids, flavonoids, and organic acids using LC-MS/MS or HPLC/MS approaches (Zamljen et al. 2021; Cervantes-Hernández et al. 2022; Lozada et al. 2023).

However, integrated studies combining physicochemical traits, antioxidant activity, and untargeted metabolite profiling within a specific agroecological setting remain limited, particularly for Indonesian *Capsicum* germplasm. Such data are essential to establish a regional biochemical baseline, enable species comparison under similar conditions, and provide reference metabolite profiles for local chili diversity. No previous study has simultaneously analyzed proximate composition, antioxidant activity, and untargeted LC-MS/MS metabolite profiles of fresh chilies in West Sumatra, limiting understanding of species-specific quality traits in this region.

This study aimed to: (i) characterize the proximate and physicochemical properties of fresh chili fruits cultivated in West Sumatra, (ii) evaluate their antioxidant activity, and (iii) identify metabolite composition using untargeted LC-MS/MS analysis. Red Curly chili (*Capsicum annuum* L.; RC), Bird's Eye chili (*Capsicum frutescens* L.; BE), and Ghost Pepper (*Capsicum chinense* Jacq.; GP) were selected because they represent major cultivated species in Indonesia with contrasting morphology, pungency, and aroma characteristics. Accordingly, this study addresses the following research question: Do different *Capsicum* species cultivated under the same agroecological conditions in West Sumatra exhibit detectable differences in physicochemical characteristics and metabolite profiles? We hypothesize that species identity may be associated with detectable differences in fruit physicochemical characteristics and metabolite profiles under shared agroecological conditions, acknowledging that replication is limited. Therefore, the results are interpreted as preliminary evidence of species-associated trends rather than statistically confirmed differences. This hypothesis is tested through integrated analysis of fruit morphology, proximate composition,

antioxidant activity, and untargeted LC-MS/MS metabolite profiling.

MATERIALS AND METHODS

Plant material and sampling

Three *Capsicum* species used in this study were Red Curly chili (*C. annuum*), Bird's Eye chili (*C. frutescens*), and Ghost Pepper chili (*C. chinense*), hereafter referred to as RC, BE, and GP, respectively. The materials represent locally cultivated farmer-propagated populations rather than registered commercial cultivars and correspond to commonly traded market species in West Sumatra. Seeds and plant materials were obtained from smallholder farmers in Belimbing, Padang City, West Sumatra, Indonesia (0.895904°S, 100.424185°E). The area is characterized by tropical humid climate and lowland elevation (approximately 80-100 m above sea level), where these chili species are traditionally cultivated.

Fruits were collected in June 2025 during the typical harvest season. For each species, mature fruits were harvested from multiple plants across several smallholder plots within the same production area (approximately 10 plants from 3 plots; fruits were collected proportionally across plants) to reduce plant-level bias. For each species, three independent biological replicates ($n = 3$) were prepared, each consisting of a composite sample of approximately 20-30 mature fruits pooled from different plants. Each composite replicate was constructed from a distinct set of plants to maintain biological independence among replicates. Pooling within each replicate was performed to minimize plant-to-plant variability and to obtain a representative chemical profile of commonly traded *Capsicum* species. Sampling within a common agroecological zone was intended to minimize environmental variability and enable comparison of species-associated trends under shared growing conditions.

Freshly harvested fruits were transported to the laboratory on the same day without preservatives. Samples were wrapped with clean polyethylene bags and kept chilled at 10°C during transport. Only fruits at commercial maturity, defined as fully ripe fruits exhibiting uniform red coloration across the entire pericarp surface, were selected for analysis.

Research procedures

Sample preparation

Chili fruits were washed with distilled water to remove surface contaminants and then air-dried at room temperature. Physical characterization was performed on fresh samples. For subsequent analyses, all samples were processed on the same day of harvest (within approximately 1 hour) to minimize post-harvest metabolic alterations. The edible portions were homogenized using a stainless-steel blender. Each biological replicate (composite sample) was homogenized independently. The homogenate was immediately divided into separate aliquots designated for color measurement, antioxidant activity determination, proximate composition analysis, and LC-MS/MS profiling.

For physicochemical and antioxidant analyses, three independent biological replicates per species ($n = 3$) were used, and each measurement was conducted in analytical triplicate to ensure measurement reliability. Aliquots intended for LC-MS/MS profiling were placed in airtight containers and stored at -20°C until analysis. Each aliquot was thawed only once, and all measurements were conducted after a single freeze-thaw cycle to prevent metabolite degradation or compositional changes. Metabolite profiling was performed on a single composite homogenate per species, with replicate injections used as technical (instrumental) replicates.

Color measurement

Color characteristics were determined by measuring L^* , a^* , and b^* values using a colorimeter (Hunterlab ColorFlex L2, USA). The L^* parameter indicates lightness, a^* represents the green-red axis, and b^* represents the blue-yellow axis. Color was measured on homogenized samples, representing overall pigment extraction rather than intact fruit surface color. Measurements were conducted using three independent biological replicates per species ($n = 3$), each analyzed in triplicate. Color intensity and hue were further described using chroma and hue angle ($^{\circ}\text{Hue}$) (Anggraini et al. 2025).

Physical characteristics

Physical characteristics of the chili fruits were measured according to procedures commonly applied in chili characterization studies (Hawa et al. 2021). Fruit length and diameter were measured using a digital caliper with a precision of 0.01 mm, while individual fruit weight was determined using a digital analytical balance with a precision of 0.001 g. Measurements were conducted on 20 randomly selected fruits from the pooled composite samples representing each species, which were collected from multiple plants within the sampled plots. The selected fruits represented multiple plants across the sampled plots to reduce the likelihood of pseudo-replication at the plant level. Each fruit was measured individually, and the results were expressed as mean \pm SD.

Proximate analysis

Proximate composition was determined following the methods of (AOAC 2019; Syukri 2021). Analyses were conducted using homogenized chili samples prepared as described in the sample preparation section. Approximately 5 g of homogenate was used for each proximate analysis. Moisture was determined by drying samples in a hot-air oven at 105°C until constant mass was achieved. Ash content was quantified by incineration in a muffle furnace at 600°C for 6 h, followed by gravimetric measurement of the mineral residue. Lipid content was extracted using a Soxhlet apparatus with n-hexane as solvent and determined gravimetrically after solvent removal. Crude protein was analyzed using the Kjeldahl method, and nitrogen values were converted to protein using a factor of 6.25. Carbohydrates were estimated by difference. Analyses were conducted using three independent biological replicates per species ($n = 3$), each derived from separately prepared

composite samples, with analytical triplicate measurements for each replicate. Results are presented as mean \pm SD.

Antioxidant activity DPPH

Antioxidant activity was determined using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay methods of (Nanda et al. 2023). Chili extracts were prepared by mixing 100 mg of homogenized chili sample with 10 mL of methanol and extracting for 15 min at room temperature with an ultrasonic bath. The extract was diluted with methanol to obtain a final concentration of 1000 ppm (1 mg/mL). For the assay, 1 mL of the extract was mixed with 2 mL of 25 μM DPPH solution in methanol and incubated in the dark at room temperature for 30 min. The decrease in absorbance was measured at 517 nm using a UV-Vis spectrophotometer. A blank consisting of 1 mL of methanol and 2 mL of DPPH solution was prepared under the same conditions.

Antioxidant activity was evaluated at a single extract concentration (1000 ppm) and expressed as percentage radical scavenging activity. Therefore, the results represent relative antioxidant activity at this concentration only and do not reflect overall antioxidant capacity or IC_{50} values. Measurements were conducted using three independent biological replicates per species ($n = 3$), each analyzed in analytical triplicate. Antioxidant activity was expressed as percentage radical scavenging activity according to the following equation:

$$\% \text{ inhibition} = \frac{\text{Abs. blank} - \text{Abs. sample}}{\text{Abs. blank}} \times 100$$

LC-MS/MS metabolite profiling

LC-MS/MS analysis was performed using a Waters Xevo G2-XS QToF mass spectrometer (Waters, USA) coupled to a liquid chromatography system. Before analysis, system suitability was verified using standard solutions of biotin and chloramphenicol.

For sample preparation, an accurately weighed portion of the homogenized chili sample was transferred into a 10 mL volumetric flask, extracted with methanol under sonication, and diluted to volume with the same solvent. The extract was thoroughly mixed and filtered through a 0.22 μm GHP/PTFE membrane filter before injection. Chromatographic separation was carried out on an HSS T3 reversed-phase column (100 mm \times 2.1 mm internal diameter, 1.8 μm particle size) using a gradient elution system. Mobile phase A consisted of 0.1% formic acid in acetonitrile, and mobile phase B consisted of 0.1% formic acid in ultrapure water. The flow rate was 0.6 mL/min, and the injection volume was 10 μL .

Mass spectrometric detection was performed using Electro Spray Ionization (ESI) in both positive and negative ion modes, with TOF MSe acquisition over an m/z range of 50-1200. LC-MS/MS measurements were conducted in technical replicate injections of the composite extract for each species. A single composite extract per species was analyzed for LC-MS/MS, without biological replication. Therefore, metabolite profiling results are considered qualitative and exploratory, and no quantitative comparisons among species are intended.

Data processing and compound screening were performed using UNIFI software (Waters), which incorporates a mass spectral library for putative identification of metabolites (MSI level 2).

Data analysis

Data are presented as mean±Standard Deviation (SD). For color, proximate composition, and antioxidant activity, values were calculated from three independent biological replicates per species (n = 3). Physical traits were measured from 20 individual fruits per species (n = 20). This study aimed to provide baseline physicochemical characterization and metabolite profiling of representative composite samples rather than to compare populations statistically; inferential statistical tests (e.g., ANOVA) were not performed. Consequently, the results are interpreted as descriptive summaries rather than formal statistical comparisons among species.

RESULTS AND DISCUSSION

Color characteristics of fresh chili fruits

Color attributes of the three *Capsicum* species were assessed using the CIELAB system, in which L* indicates lightness, a* the red-green axis, and b* the yellow-blue axis. Chroma reflects color saturation (0 = gray; 100 = highly vivid), while the hue angle (0°-360°) represents the dominant color tone (Nawaz et al. 2019). Descriptive values for color parameters of Red Curly chili (RC), Bird's Eye chili (BE), and Ghost Pepper (GP) are presented in Table 1.

All samples showed low L* values, indicating dark fruits. BE had the lowest L* (27.00), suggesting a darker surface than RC (30.98) and GP (31.46). Lower L* values generally reflect reduced lightness due to lower reflectance, often linked to higher pigment density in plant tissues (Rodríguez-Pulido et al. 2017; Dutta and Nath 2023).

RC displayed the highest a* (39.43) and chroma (44.05), indicating the strongest red coloration and greatest color saturation. In contrast, BE and GP exhibited lower a* values (28.15 and 28.02) and chroma (31.29 and 32.89), consistent with less intense red appearance. Although carotenoids were not quantitatively measured, the exclusive detection of apocarotenoid-related compounds in RC, including β-ionone and blumenol C glucoside, may be qualitatively consistent with carotenoid turnover associated with pigmentation (Table 4). However, as metabolite identification was putative and relative abundance was not assessed, this observation should be interpreted as a qualitative association rather than direct evidence of pigment concentration differences.

According to previous reports, elevated a* values in *Capsicum* are commonly associated with red carotenoids such as capsanthin and capsorubin (Morales-Soriano et al. 2019; Berry et al. 2021). Higher chroma likewise indicates greater color vividness and pigment contribution, which may reduce perceived lightness through increased light absorption (Rodríguez-Pulido et al. 2017).

All samples had positive b* values (13.63-19.60), indicating yellow color components. GP showed a relatively higher b* (17.22) and the highest hue angle (31.57°), suggesting a shift toward orange tones compared with RC (26.43°) and BE (25.84°). Increases in b* and hue angle are commonly linked to greater contributions of yellow-orange carotenoid fractions, producing a more orange fruit appearance (Morales-Soriano et al. 2019; Hendrawan et al. 2021).

Physical characteristics of chili fruits

Morphological traits were analyzed to compare fruit size and shape among three *Capsicum* species: *C. annuum* (RC), *C. frutescens* (BE), and *C. chinense* (GP). Fruit length, diameter, and weight were used as key morphometric descriptors for varietal differentiation. Results are summarized in Table 2, and representative fruit morphology is shown in Figure 1.

The results of physical analysis (length, diameter, and weight) of the three *Capsicum* species are presented in Table 2. Fruit dimensions are reported as mean±standard deviation (n = 20 fruits per species) and reflect within-sample variation of the collected material.

Table 1. Color characteristics of fresh chili fruits from different species

Sample	CIE L* a* b* values			Hue (°)	Chroma	Color
	L*	a*	b*			
RC	30.98±2.13	39.43±1.32	19.60±1.34	26.43	44.05	Red
BE	27.00±1.22	28.15±1.70	13.63±0.86	25.84	31.29	Red
GP	31.46±2.14	28.02±1.59	17.22±1.60	31.57	32.89	Red

Note: Values are presented as mean±standard deviation (n = 3)

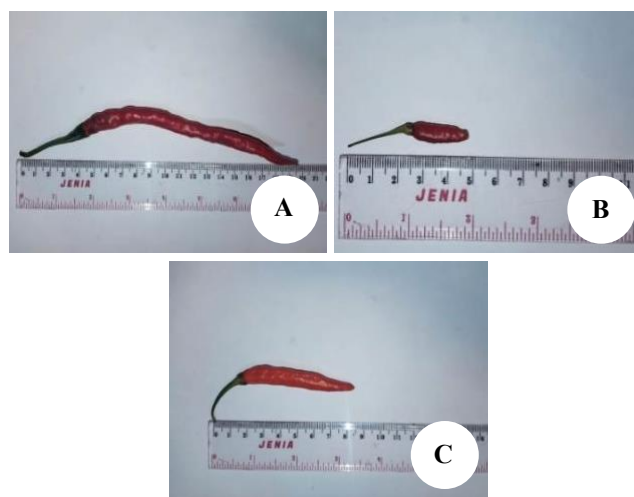


Figure 1. Physical measurements of three species of chili: A. Red Curly Chili (*Capsicum annuum*), B. Bird's Eye Chili (*Capsicum frutescens*), and C. Ghost Pepper Chili (*Capsicum chinense*)

Descriptive values indicate that RC exhibited an elongated fruit form (11.88±2.10 cm length; 0.73±0.11 cm diameter), yielding an elongation ratio (L/D ≈ 16.3). BE showed a shorter but wider fruit form (5.90±0.89 cm; 0.96±0.20 cm; L/D ≈ 6.1), while GP presented smaller fruits (3.14±0.44 cm; 0.62±0.11 cm; L/D ≈ 5.1). These descriptive measurements were also reflected in fruit mass, with BE showing the highest unit weight (3.42±1.00 g), followed by RC (2.59±0.61 g) and GP (0.77±0.17 g). These observations describe morphological characteristics of the sampled fruits and should not be interpreted as statistically validated species-level differentiation.

This pattern aligns with previously reported morphological variation in *Capsicum*, where fruit size and shape differ widely among species and cultivars (Peñuela et al. 2021). However, the present results represent locally cultivated and marketed materials rather than the full phenotypic range of each species. The high L/D ratio of RC quantitatively supports the elongated fruit form characteristic of *C. annuum*, while the lower L/D ratio of GP is consistent with the more globose tendency described for *C. chinense*. Previous studies have reported substantial within-species variation in fruit size and morphology across diverse *Capsicum* cultivars (Nimmakayala et al. 2021; Li et al. 2022). The morphometric values observed here fall within reported ranges, while providing region-specific descriptive data under shared agroecological conditions.

Similarly, Li et al. (2022) reported that *C. chinense* often exhibits the greatest fruit weight, whereas *C. frutescens* shows the lowest, and that fruit diameter tends to be larger in *C. chinense* and *C. frutescens* than in *C. annuum*. Although the absolute measurements in this study were lower than previously reported, the relative ranking among species was consistent, likely due to differences in genotype, growing environment, cultivation practices, and harvest maturity. Although absolute values differed from previous reports, the relative patterns observed here are presented as descriptive comparisons within the sampled material.

Notably, fruit weight did not increase proportionally with fruit length. Despite RC having the highest elongation ratio, BE had the greatest unit weight. This observation suggests that fruit mass may be influenced by multiple morphological components beyond length, including diameter, pericarp thickness, and tissue density, consistent with standardized *Capsicum* descriptors (IPGRI 1995).

Proximate analysis

Proximate analysis was conducted to characterize the basic nutritional and physicochemical composition of chili fruits from three *Capsicum* species. The proximate profiles are shown in Table 3.

Moisture was the dominant component in all samples (78.70%-82.46%), consistent with the high water content typical of fresh chili fruits. GP showed slightly lower moisture content compared with RC and BE. Elevated moisture in fresh produce is associated with parenchymatous tissues and high vacuolar water content (Gidado et al. 2024), but may also reduce storage stability due to increased susceptibility to microbial growth (Ajuru et al. 2017; Khan et al. 2019). The values observed fall within the expected range reported for fresh *Capsicum* fruits (83.26% to 92.20%) across species and cultivars (Malakar et al. 2018; Anaya-Esparza et al. 2021).

Ash content ranged from 0.85% to 1.10%, indicating differences in total mineral fraction among the species. Ash is widely used as a proxy for total inorganic constituents and reflects nutritionally important minerals (Tura et al. 2023). The ash values observed here fall within the broad range reported for fresh *Capsicum* fruits (0.47%-5.53%), supporting consistency with established compositional data (Anaya-Esparza et al. 2021; Gyamfi 2024). Among the samples, BE exhibited the highest ash content, indicating a relatively greater total mineral fraction compared with RC and GP.

Fat levels were low across all samples (0.32%-0.55%), consistent with fresh horticultural commodities (Don et al. 2019). In chilies, lipids are primarily linked to lipid-soluble bioactives, particularly carotenoids (e.g., capsanthin, capsorubin, β-carotene, violaxanthin, neoxanthin, lutein) and vitamin A-related compounds (Sunarmani et al. 2024). The measured values align with commonly reported ranges for fresh *Capsicum* fruits (0.30%-0.83%), with RC showing slightly higher lipid content than other species (Dahiru et al. 2018; Anaya-Esparza et al. 2021).

Protein levels were relatively similar, ranging from 1.15% to 1.58%, consistent with typical values for fresh chili fruits, which also indicates that species differences had a limited influence on the protein fraction of the fresh fruits. While plant-based foods contribute substantially to global protein intake and provide essential amino acids with health benefits (Hertzler et al. 2020; Pandey et al. 2023). Chilies contain relatively little protein and are not a major protein source, and the values obtained fall within the reported range for fresh *Capsicum* fruits (0.12%-2.03%) (Dahiru et al. 2018; Anaya-Esparza et al. 2021).

Table 2. Physical analysis (length, diameter, and weight of fresh chili)

Sample	Length (cm)±SD	Diameter (cm)±SD	Weight (g)±SD
RC	11.88±2.10	0.73±0.11	2.59±0.61
BE	5.90±0.89	0.96±0.20	3.42±1.00
GP	3.14±0.44	0.62±0.11	0.77±0.17

Note: Values are presented as mean±standard deviation (n = 20)

Table 3. Proximate composition of three chili fruits

Sample	Moisture content (%)	Ash content (%)	Fat content (%)	Protein content (%)	Carbohydrate content (%)
RC	82.46±0.25	0.85±0.05	0.32±0.02	1.15±0.07	15.22±0.22
BE	80.90±0.18	0.95±0.06	0.45±0.03	1.35±0.09	16.35±0.28
GP	78.70±0.20	1.10±0.05	0.55±0.02	1.58±0.08	18.07±0.26

Note: Values are presented as mean±standard deviation (n = 3)

Carbohydrates (by difference) accounted for 15.22%-18.07% of the sample composition, representing a substantial fraction of dry matter. Carbohydrates in plant foods include sugars, starches, and dietary fiber that support normal physiological functions (Xue et al. 2017). In chilies, carbohydrate levels vary by species and cultivar and reflect carbon allocation during fruit development, including cell wall polysaccharides and soluble solids (Khan et al. 2019). The carbohydrate values observed here exceed those reported in some studies on fresh *Capsicum* fruits (6.03%-8.19%) (Dahiru et al. 2018; Malakar et al. 2018; Anaya-Esparza et al. 2021), likely because the samples in this study had comparatively lower moisture content. Notably, GP showed relatively higher carbohydrate values compared with RC and BE, suggesting greater accumulation of soluble solids or structural carbohydrates in this species. Such variation may reflect differences in carbon partitioning during fruit development among *Capsicum* species.

Overall, proximate composition among the three species was broadly comparable, with only minor variations observed across components. However, some species-level differences were evident, including slightly lower moisture and higher carbohydrate levels in GP, and higher ash content in BE, indicating modest variation in water balance and mineral fraction among the species. Although the three species exhibited broadly comparable nutritional profiles, these compositional differences may influence their textural properties, flavor intensity, and processing suitability. Such variability is consistent with known influences of genotype, growing conditions, agronomic practices, harvest maturity, and analytical methods (Don et al. 2019; Makhziah et al. 2021).

Antioxidant activity

Antioxidant activity of fresh chili samples, evaluated using the DPPH radical scavenging assay, reflects the combined capacity of non-enzymatic bioactive compounds in *Capsicum* fruits to neutralize free radicals (Alam et al. 2016). All samples exhibited high radical scavenging capacity, with DPPH radical scavenging activity (mean \pm SD) of 86.67 \pm 6.94% for GP, 83.38 \pm 10.00% for BE, and 83.59 \pm 6.22% for RC (Figure 2).

DPPH antioxidant activity in chili fruits arises from the combined and potentially synergistic effects of multiple phytochemical groups rather than a single compound class (Alonso-Villegas et al. 2023). Phenolic compounds are major contributors due to their hydroxyl groups, which facilitate radical stabilization; predominant phenolics reported in *Capsicum* include catechin, quercetin, protocatechuic acid, and rutin (Lemos et al. 2019; Rodrigues et al. 2019).

Capsaicinoids such as capsaicin, dihydrocapsaicin, and nordihydrocapsaicin have also been reported to contribute to free-radical scavenging activity (Sarafi et al. 2018; Azlan et al. 2022). In addition, carotenoids (capsanthin, capsorubin, β -carotene, violaxanthin, neoxanthin, and lutein) and ascorbic acid are known to enhance antioxidant capacity through radical quenching and reducing mechanisms (Del Rocio Moreno-Ramírez et al. 2018; Azlan et al. 2022). These literature-based mechanisms provide a general

context for antioxidant activity in *Capsicum* but were not quantitatively assessed in the present study.

Phytochemical screening of fresh chili fruits has also confirmed the presence of flavonoids, tannins, alkaloids, saponins, anthraquinones, and polyphenols, which are widely associated with antioxidant potential (Dahiru et al. 2018). Accordingly, variation in DPPH scavenging activity among samples may reflect differences in overall phytochemical composition rather than the dominance of a single compound class; however, such relationships were not directly quantified in this study.

The antioxidant activity observed here is consistent with previous DPPH-based reports on fresh *Capsicum* fruits, which typically show inhibition values above 70% (Ranilla et al. 2010; Chávez-Mendoza et al. 2015), indicating that the measured values fall within the expected range.

In the present study, untargeted LC-MS/MS analysis detected several compounds commonly associated with antioxidant activity, including quercetin, luteolin, kaempferol derivatives, epicatechin, and cyanidin-related anthocyanins (Table 4), as well as apocarotenoid-related metabolites such as β -ionone and blumenol C glucoside. However, metabolite identification was qualitative (MSI level 2), and no quantitative correlation analysis was performed; therefore, these detections should be interpreted as general associations rather than direct evidence of contribution to antioxidant activity.

Furthermore, antioxidant activity was evaluated at a single concentration (1000 ppm); thus, the results represent relative activity at this concentration only and do not reflect overall antioxidant capacity (e.g., IC₅₀) or enable robust comparison among species. Accordingly, the observed values should be interpreted as descriptive trends rather than definitive differences in antioxidant capacity.

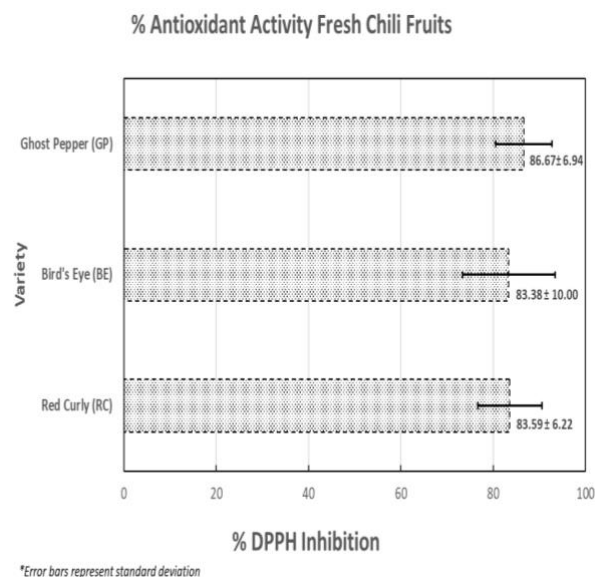


Figure 2. DPPH radical scavenging activity of fresh chili fruits from different *Capsicum* species (1000 ppm). Values are presented as mean \pm standard deviation (n = 3)

Table 4. Summary of representative metabolites identified by untargeted LC-MS/MS in three fresh *Capsicum* species

Compound name	Metabolite class	Sample		
		RC	BE	GP
Polyphenols and flavonoids				
p-Coumaric acid	Phenolic acids	✓	-	-
Luteolin	Flavones	✓	-	-
Apigenin derivatives	Flavones	-	-	✓
Quercetin derivatives	Flavonols	✓	✓	✓
Kaempferol derivatives	Flavonols	✓	✓	✓
Isorhamnetin derivatives	Flavonols	-	✓	✓
Rhamnetin derivatives	Flavonols	-	✓	✓
Epicatechin	Flavan-3-ols	✓	✓	✓
Afzelechin	Flavan-3-ols	-	✓	-
Cyanidin-3-rutinoside	Anthocyanins	✓	-	✓
Cyanidin-3,5-diglucoside	Anthocyanins	✓	✓	✓
Chalcone derivatives	Chalcones	✓	✓	✓
Terpenoids				
Julibroside C1	Terpenoids	-	✓	-
Neocomplanoside	Terpenoids	-	✓	-
Rindoside	Terpenoids	-	✓	-
Viscumneoside 3	Terpenoids	-	✓	-
Viscumneoside 5	Terpenoids	-	✓	-
Abietic acid	Diterpenoids	-	✓	✓
Dehydroabietic acid	Diterpenoids	-	✓	✓
β-Ionone	Apocarotenoids	✓	-	-
Blumenol C glucoside	Apocarotenoids	✓	-	-
Dihydroactinidiolide	Volatile compounds	✓	-	-
9-Hydroxylinalool glycoside	Monoterpene glycosides	✓	-	✓
Trans-carveol glycoside	Monoterpene glycosides	✓	-	✓
Menthoside	Monoterpene glycosides	✓	-	✓
Fatty acids				
4,8,12-Trimethyltridecanoic acid	Fatty acids	✓	✓	-

Note: ✓ / - : Compound putatively detected/not detected in the sample under the analytical conditions applied

LC-MS/MS-based metabolite profiling of fresh chili fruits

Untargeted LC-MS/MS profiling of three *Capsicum* species, *C. annum* (RC), *C. frutescens* (BE), and *C. chinense* (GP), resulted in the annotation of 147 metabolites across all samples. Based on qualitative annotation, 73 metabolites were detected in RC, 78 in BE, and 70 in GP. The annotated compounds were primarily distributed among flavonoids, fatty acids, other polyphenols, terpenoid-related metabolites, and several biosynthetic intermediates. Collectively, this dataset provides a comparative overview of secondary metabolite composition among three *Capsicum* species commonly traded in West Sumatra, providing an overview of metabolite distribution patterns (Table 4). Table 4 presents a comprehensive qualitative metabolite annotation; therefore, interpretation is based on overall detection patterns rather than quantitative comparison, serving as a synthesis of metabolite distribution across species.

Polyphenols and flavonoids

Phenolic compounds, synthesized via the phenylpropanoid pathway, are key secondary metabolites involved in plant

structural integrity, development, and stress responses. In *Capsicum*, flavonoids and phenolic acids constitute the dominant phenolic classes (Echave et al. 2020), contributing substantially to the chemical diversity of pepper fruits. In this pathway, phenylalanine is converted through the sequential action of Phenylalanine Ammonia-Lyase (PAL), cinnamate-4-hydroxylase (C4H), and 4-coumarate-CoA ligase (4CL) into p-coumaroyl-CoA, a central precursor of flavonoid biosynthesis. Subsequent reactions mediated by Chalcone Synthase (CHS) and Chalcone Isomerase (CHI) generate naringenin, a key intermediate leading to multiple flavonoid subclasses (Yang et al. 2025).

Consistent with known biosynthetic pathways, the LC-MS dataset revealed multiple putatively annotated flavonoid subclasses across all three species. Consistent with these biosynthetic pathways, the LC-MS dataset revealed multiple putatively annotated flavonoid subclasses across all three species. Among early phenylpropanoid metabolites, *p*-coumaric acid was putatively detected in the RC dataset but was not annotated in BE and GP under the present analytical conditions. Phenolic acids derived from cinnamic, coumaric, and caffeic pathways are widely reported in *Capsicum* fruits (Xavier and Pérez-Gálvez 2016).

Flavonoids detected in this study comprised several subclasses commonly reported in pepper fruits. Flavone derivatives, including luteolin- and apigenin-related compounds, were detected across the datasets and are widely reported in *Capsicum* (Imran et al. 2019), while quercetin and luteolin have been reported to account for a substantial proportion of total flavonoids (Antonio et al. 2018; Ab Rahman et al. 2024). Several findings of quercetin- and luteolin-based glycosides, including both O- and C-glycosides, have also been demonstrated in pepper pericarp through MS-MS and UV analyses (Wahyuni et al. 2011). The consistent detection of flavone and flavanone derivatives across the three species supports the conservation of core flavonoid biosynthesis within the genus.

Flavonols constituted another prominent group, including quercetin, kaempferol, kaempferide, rhamnetin, and isorhamnetin derivatives. Kaempferol has been described as a common flavonol in *Capsicum* with antimicrobial activity (Mokhtar et al. 2017), while isorhamnetin has also been previously reported in *Capsicum* (Liu et al. 2025). Variation in their detection among datasets, including the absence of certain derivatives in RC, may reflect analytical detection limits or biological variation among samples.

Flavan-3-ols, including afzelechin and epicatechin, were detected across all species and are widely associated with antioxidant activity and plant defense (Grojja et al. 2023; Lu 2024). Flavan-3-ols are widely recognized for their antioxidant properties and their role as precursors of condensed tannins involved in plant defense. Anthocyanin-related metabolites, represented by cyanidin-3-rutinoside and cyanidin-3,5-diglucoside, were also putatively identified, indicating the presence of cyanidin-type anthocyanins associated with pigmentation and antioxidant activity (Devraj et al. 2025). In *Capsicum*, anthocyanin accumulation is regulated by environmental and transcriptional factors (He et al. 2025) and contributes to fruit coloration (Zhou et al. 2020).

Chalcone derivatives were also putatively detected. Although chalcones are typically present at low concentrations due to rapid enzymatic conversion, their detection is consistent with ongoing flavonoid biosynthesis in fruit tissues (Sugimoto et al. 2024; Yang et al. 2025).

Some metabolites were detected only in specific datasets. Such absence should be interpreted cautiously, as it may reflect analytical detection limits or annotation constraints rather than true biological absence. Overall, these observations indicate the presence of conserved flavonoid-related metabolites across the genus, with variation in qualitative detection patterns among samples.

Terpenoids

Terpenoid profiling revealed a diverse array of terpenoid glycosides and carotenoid-derived compounds across all species (Table 4). Glycosylated forms represented a large proportion of the annotated terpenoid-related metabolites, consistent with glycosylation as a major modification in plant secondary metabolism (Zhang et al. 2022).

Based on annotation counts from single composite samples, a greater number of terpenoid-related compounds were detected in BE (38) compared with RC (33) and GP (28). This observation is qualitative and may reflect analytical sensitivity, ionization efficiency, or annotation bias rather than true biological differences in metabolite abundance.

In BE, frequently detected compounds included were present as glycosides, including Julibroside C1, Rindoside, Neocomplanoside, Viscumneoside 3, and Viscumneoside 5. This pattern reflects a higher detection frequency of conjugated terpenoids in BE within the present dataset (Zhang et al. 2022). GP showed a partially overlapping detection pattern with BE, sharing compounds such as abietic acid, dehydroabietic acid, and phytolaccoside D. Abietic acid has previously been reported in *Capsicum* (Jang et al. 2024). These observations reflect similarities among the detected compound classes under the applied analytical conditions.

In contrast, RC showed a distinct detection pattern within this dataset. RC was only detected by the presence of dihydroactinidiolide, a volatile compound reported as common in *C. annuum* (Li et al. 2025). More notably, RC was the only species in which β -ionone was putatively detected. β -Ionone is an apocarotenoid formed via oxidative cleavage of carotenoids by Carotenoid Cleavage Dioxygenases (CCD) (Paparella et al. 2021), and its presence in *C. annuum* has previously been linked to carotenoid substrates (Mohd Hassan et al. 2019). The occurrence of β -ionone is consistent with apocarotenoid formation, potentially associated with aroma formation and ecological signaling functions (Paparella et al. 2021). Supporting this observation, glycosylated apocarotenoid-related metabolites such as blumenol C glucoside were also detected in RC, consistent with previous reports in *C. annuum* metabolomes (Guevara et al. 2021).

RC additionally showed detection of multiple diterpenoid-related compounds and their derivatives. The coexistence of oxidized aglycones and glycosylated forms is consistent with diterpenoid biosynthesis followed by downstream

modification through oxidation and sugar conjugation (Zhang et al. 2022).

Monoterpene glycosides (e.g., 9-hydroxylinalool-9-O- β -D-glucopyranoside, trans-carveol-6- β -D-glucopyranoside, and menthoside) were detected in RC and GP. In plants, volatile terpenoids are commonly stored as glycosylated forms that can be enzymatically hydrolyzed upon tissue disruption or stress (Yazaki et al. 2017). Differences in their detection among datasets may therefore reflect variation in metabolite stability, storage, or analytical sensitivity.

Overall, differences in terpenoid-related annotations among species should be interpreted as qualitative detection patterns rather than evidence of pathway dominance or differential biosynthetic regulation.

Other identified metabolites

In addition to flavonoids and terpenoids, other compounds were putatively annotated, including branched-chain fatty acids such as 4,8,12-trimethyltridecanoic acid. Similar compounds have been reported in *Capsicum* (Keum et al. 2012).

Beyond the key metabolites discussed, the untargeted LC-MS/MS dataset also captured additional flavonoid aglycones (e.g., alpinetin, galangin, genkwanin, herbacetin, tangeretin), flavonoid glycosides (e.g., baicalin, rhoifolin, eriocitrin/neoeriocitrin, prunin, paeonin), chalcone derivatives, and diverse terpenoid-related compounds, including annotations resembling andrographolide-, ginsenoside-, and ruscogenin-like structures. The complete putatively annotated metabolites are presented in Table 5. Because metabolite identification relied on spectral library matching, all assignments should be considered putative annotations rather than confirmed compound identifications.

Overall, the three *Capsicum* species share a conserved phenylpropanoid and terpenoid metabolite framework. Variation in metabolite detection patterns was observed among datasets, with BE showing a broader representation of glycosylated terpenoid-related annotations, GP showing partially overlapping patterns, and RC showing detection of apocarotenoid-related compounds and diverse diterpenoid annotations (Ab Rahman et al. 2024). These observations are qualitative and should be interpreted as exploratory trends rather than definitive species-specific differences.

Overall, the three *Capsicum* species shared broadly similar metabolite classes, while differences in qualitative detection patterns were observed among datasets. These observations should not be interpreted as definitive species-specific differences or quantitative variation. Metabolite identification was based on putative annotation (MSI level 2) and qualitative data; therefore, results are exploratory and do not support definitive interspecific differences. No quantitative or multivariate analyses were performed. Future studies should incorporate quantitative metabolomics and multivariate approaches for robust comparison.

The observed physicochemical and metabolite characteristics can be interpreted in a qualitative integrative framework. Variations in carbohydrate content among species may reflect differences in carbon allocation during fruit development.

Table 5. Complete list of putatively annotated metabolites detected by LC-MS/MS in three *Capsicum* species

Compound name	ESI mode	Sample		
		RC	BE	GP
Polyphenols and flavonoids				
Quercetin-3-O-robinobioside	+	-	-	✓
Quercetin-3-rhamnogentiobioside	+/-	-	-	✓
Quercetin 3-O-neohesperidoside	+	-	-	✓
Quercetin-7-O-rutinoside	+	✓	-	-
Luteolin	+	✓	-	-
Luteolin 7-O-β-D-(6"-acetyl)-glucopyranoside	-	✓	-	✓
Luteolin-7-β-D-glucopyranoside	-	-	✓	-
Luteolin 7-β-neohesperidoside	-	-	-	✓
Apigenin-7-O-acetyl-β-D-glucoside	-	-	-	✓
Isoschaftoside	+/-	✓	✓	-
4, 5, 7-Trihydroxyflavone-3-O-β-D-glucopyranoside	-	✓	-	-
5, 7, 2', 5'-Tetrahydroxyflavone	+/-	✓	✓	✓
8-(Δ ² -Isopentenyl)-5, 7, 3', 4'-tetrahydroxyflavone	-	-	✓	-
Naringenin	+	-	✓	✓
3-Hydroxynaringenin	-	✓	-	-
Naringenin-4-O-glucopyranoside	+/-	✓	✓	-
5, 7, 4-Trihydroxy-8-C-βD-flavanone glucoside	-	-	✓	-
Isorhamnetin	+	-	✓	-
Rhamnetin	+/-	-	✓	✓
8-C-prenylkaempferol	-	-	-	✓
Isorhamnetin-3-O-gentiobioside	+/-	-	✓	-
Isorhamnetin-3, 7-O-β-D-glucopyranoside	-	-	✓	-
Isorhamnetin-7-O-β-D-glucopyranoside	-	-	-	✓
Isorhamnetin-3-O-β-rutinoside	+	-	✓	-
Isorhamnetin-3-O-neohesperidoside	-	-	-	✓
Isorhamnetin-3-gentiobioside-7-glucoside	-	-	✓	✓
2"-O-acetyl-3'-O-methylrutin	+/-	-	✓	✓
2"-OAcetylrutin	+/-	✓	✓	✓
Kaempferol-3-gentiobioside	-	✓	-	✓
Kaempferol-3-α-L-di-rhamnosyl-(1à4)-β-D-glucopyranoside	-	✓	-	-
Kaempferol-3-O-(2G-α-L-rhamnosyl)-rutinoside	+	✓	-	-
Kaempferol-3-O-neohesperidoside	+/-	-	✓	-
Kaempferol-3-O-rhamnoside	+	✓	✓	-
Kaempferol-3-gentiobioside-7-glucoside	+	-	-	✓
Kaempferol-3-rutinoside-7-glucoside	+	-	-	✓
Kaempferol-3-O-β-D-glucopyranosyl-(1à2)-β-D-galactopyranosyl-(1à2)-β-D-glucopyranoside	-	-	✓	✓
Kaempferol-3-O-β-D-glucoside-7-O-α-L-arabinofuranoside	-	✓	✓	✓
3-O-[β-D-Glucopyranosyl-(1à2)-β-D-glucopyranosyl-7-O-α-L-glucopyranosyl]kaempferol	+	✓	-	-
7-α-L-Rhamnosyl kaempferol 3-O-β-D-glucopyranosyl (1à6)-β-D-glucopyranoside	+	-	-	✓
Kaempferide-3-O-α-L-(4-O-acetyl)-rhamnosyl-7-O-α-L-rhamnoside	+	✓	-	-
Kaempferide-4-methyl ether-3-glucoside	+/-	✓	-	✓
(2R, 3R)-Taxifolin-7-O-α-L-rhamnopyranosyl-(1à6)-β-D-glucopyranoside	-	-	-	✓
Cyanidin-3-rutinoside	-	✓	-	✓
Cyanidin 3, 5-diglucoside	-	✓	✓	✓
Afzelechin	+	-	✓	-
(-)-Epiafzelechin-3-O-β-D-allopyranoside	-	✓	✓	✓
Epicatechin 5-O-β-D-glucopyranoside	-	✓	✓	-
Alpinetin	+	✓	-	-
Galangin (Norisalpinin)	+	✓	-	✓
Gardenin C	+	✓	-	-
Genkwanin	+/-	✓	✓	-
Herbacetin	+	-	✓	-
Hydroxygenkwanin	+	-	-	✓
Isobavachin	+	✓	✓	-
Kushenol S	+	-	-	✓
Leachianone G	+	✓	-	-
Rubrofusarin	+	✓	-	-
Santin	+	-	✓	-
Sappanol	+	✓	✓	✓
Tangeritin	+	-	-	✓

Tetuin	+/-	✓	✓	-
5-O-Methylvisamminol	-	✓	✓	-
6-Aldehydo-7-methoxyisooopogonone B	-	-	✓	-
Apigenol	-	✓	-	✓
Chrysoeriol	-	-	-	✓
Scutellarein	-	-	✓	-
Sophoranodichromane B	-	-	✓	-
Suberectin	-	-	✓	-
Baicalin	+	✓	-	-
Isoengeletin	+	-	-	✓
Mirificin	+/-	-	-	✓
Oroxin B	+	✓	✓	✓
Rhoifolin	+	-	-	✓
Vaccarin	+/-	✓	✓	✓
Eriocitrin	-	-	-	✓
Neoeriocitrin	-	-	✓	-
Paeonin	-	-	✓	✓
Prunin	-	✓	-	-
E-p-Coumaric acid	+	✓	-	-
2, 4-Dihydroxy-6-methoxydihydrochalcone	+	✓	-	-
2, 6-Dihydroxy-4-methoxydihydrochalcone	+	-	-	✓
Psorachalcone A	+	-	✓	✓
1, 2, 3, 4-Tetrahydro-1, 6-dimethyl-4-(1-methylethyl) naphthalene	+	✓	-	-
Terpenoids				
Dihydroactinidiolide	+	✓	-	-
β-Ionone	+	✓	-	-
14-Deoxy-11-oxoandrographolide	+	✓	-	-
14-Deoxyandrographolide	+	✓	-	-
Abietic acid	+	-	✓	✓
Dehydroabietic acid	+	-	✓	✓
Oriediterpenol	+	✓	-	✓
ar-Abietatriene	+	✓	✓	✓
ent-16α, 17-Hydroxy-19-kauranoic acid	-	✓	-	✓
19β-Glucosyl-14-deoxy-11, 12-didehydroandrographoside	+	✓	✓	-
19β-Glucosyl-14-deoxyandrographoside	+/-	✓	-	-
25(R)-Ruscogenin-1-O-β-D-glucopyranosyl (1→2)-β-D-fucopyranoside	+	✓	✓	✓
Blumenol C glucoside	+	✓	-	-
Corymboside	+	✓	-	✓
Esculentoside L	+/-	✓	✓	✓
Hookeroside C	+	✓	✓	✓
Isoschaftoside	+/-	✓	✓	-
Julibroside C1	+	-	✓	-
Menthoside	+/-	✓	-	-
Nelumboside A	+	-	-	✓
Nelumboside B	+/-	-	✓	✓
Neocomplanoside	+	-	✓	-
Odoratin-7-O-β-D-glucoside	+	-	-	✓
Pinnatifinoside D	+	✓	-	-
Rhamnocitrin-3-O-β-D-glucoside	+	-	✓	-
Rindoside	+/-	-	✓	-
SecOglucosylhamaudol	+	-	✓	✓
Sophorabioside	+/-	✓	✓	✓
Undulatoside A	+	-	-	✓
prim-O-Glucosylcimifugin	+	-	✓	✓
trans-Carveol-6-β-glucopyranoside	+	-	-	✓
17-O-β-D-Glucopyranosyl-16β-H-entkauran-19-oic acid-19-O-β-D-glucopyranoside	-	✓	✓	✓
1 β,3β,6α-Trihydroxy-4α-(15)-dihydrocortic acid methyl ester-1-O-β-D-glucopyranoside	-	-	✓	-
20(R)-Ginsenoside Rh2	-	✓	-	-
25-Anhydrocimigenol-3-O-β-D-xylopyranoside	-	✓	-	-
Acacetin-7-O-(6"-O-acetyl)- β-D-glucopyranoside	-	-	✓	-
Aesculioside D	-	✓	-	-
Andrographatoside	-	✓	✓	-
Cichorioside B	-	-	✓	-
Gentiopicroside	-	✓	✓	✓
Khellol-β-D-glucoside	-	-	✓	✓

Limocitrin-3,7-O-β-D-glucopyranoside	-	✓	✓	-
Nevadensin-7-O-[α-L-rhamnosyl(1→6)]-β-D-glucoside	-	✓	-	-
Patuletin-7-O-(6"-isobutyryl)-glucoside	-	-	✓	✓
Patuletin-7-O-[6"-(2-methylbutyryl)]-glucoside	+/-	-	✓	✓
Peonidin 3-glucoside	-	-	✓	-
Pterodontoside E	-	✓	✓	-
Saponoside A	-	-	✓	✓
Taraxacolide-1-O-β-D-glucopyranoside	-	✓	✓	-
Turpinionosides E	-	✓	✓	-
Viscumneoside 3	-	-	✓	-
Viscumneoside 4	+/-	-	✓	✓
Viscumneoside 5	-	-	✓	-
Viscumneoside 7	+/-	-	✓	✓
Yadanzioside A	-	-	✓	✓
Yadanzioside L	-	-	-	✓
9-Hydroxylinalool-9-O-β-D-glucopyranoside	+	✓	-	-
Phytolaccoside D	+	-	✓	✓
17-O-β-D-Glucopyranosyl-16β-H-entkauran-19-oic acid	-	-	✓	-
Neoandrographolide	+	✓	-	-
19-O-[β-D-Apiofuranosyl-(1→2)-β-D-glucopyranoyl]-3,14-dideoxyandrographolide	-	✓	-	-
3-O-β-D-Glucopyranosyl-14,19-dideoxyandrographolide	-	✓	-	-
Fatty acid				
4, 8, 12-Trimethyltridecanoic acid	-	✓	✓	-

Note: ESI +/-: Compound detected in both positive and negative ESI ionization modes, ✓/-: Compound putatively detected/not detected in the sample under the analytical conditions applied

The detection of pigment-related compounds, including carotenoid-derived and apocarotenoid-related metabolites, is consistent with observed color parameters. In addition, the presence of flavonoids and terpenoid-related compounds is consistent with known contributors to antioxidant activity. However, because these relationships are based on qualitative observations and no correlation analysis was performed, they should be interpreted as general associations rather than direct functional relationships. Overall, the results provide a preliminary integrative overview rather than a mechanistic explanation of functional properties.

In conclusion, this study provides a qualitative and exploratory characterization of physicochemical properties and putative metabolite profiles of three widely marketed *Capsicum* species from West Sumatra: Red Curly chili (RC; *C. annuum*), Bird's Eye chili (BE; *C. frutescens*), and Ghost Pepper chili (GP; *C. chinense*) under a shared agroecological context. The species exhibited clear differences in color and composition, with RC showing the highest red intensity ($a^* = 39.43$; chroma = 44.05), BE the lowest lightness ($L^* = 27.00$), and GP the highest carbohydrate content (18.07%). All samples were characterized by high moisture (78.70-82.46%), low fat (0.32-0.55%), and low protein (1.15-1.58%). Antioxidant activity was consistently high across species (83.38-86.67% DPPH inhibition). Untargeted LC-MS/MS analysis annotated 147 metabolites, revealing species-associated detection patterns, including glycosylated terpenoid-related compounds in BE, abietic-type diterpenoids in GP, and apocarotenoid-related compounds (e.g., β-ionone) in RC. This exploratory study provides preliminary descriptive data on physicochemical traits and putative metabolite detection patterns in locally cultivated *Capsicum* species. These findings contribute baseline compositional data that may support future

nutritional evaluation, metabolite-focused studies, and cultivar selection strategies for *Capsicum* production in the region. Because metabolite annotations were putative and based on composite samples obtained from a single season and location, the results should be interpreted as indicative trends rather than definitive compositional differences. Future studies incorporating multiple biological replicates, seasons, and locations, together with quantitative metabolite analysis, are required to validate and extend these findings.

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