

STRUCTURAL, MORPHOMETRIC AND GC–MS METABOLIC ANALYSIS OF SEVEN SAUDI ARABIAN DATE PALM CULTIVARS

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Keywords: *Phoenix dactylifera* L., Saudi date cultivars, GC-MS analysis, Phytochemical profiling

Abstract

This study presents a multidimensional structural–metabolic analysis of seven Saudi Arabian date palm (*Phoenix dactylifera* L.) cultivars integrating biomass allocation, morphometric traits, and GC–MS phytochemical profiling of embryo, seed, and fruit tissues. Significant variation was observed in biomass, dry matter distribution, and morphometric parameters, with a hierarchy (fruit flesh > seed > embryo). GC–MS analysis revealed tissue-specific metabolic specialization. Embryos and seeds were enriched in unsaturated fatty acids, oleic and linoleic acids, whereas fruit flesh contained higher proportions of oxygenated metabolites, including γ -lactones and carbonyl compounds. Hierarchical clustering based on phytochemical composition separated cultivars into distinct chemotypic groups, highlighting cultivar-dependent metabolic divergence. These findings indicate coordinated structural allocation and metabolite accumulation define cultivar identity and provide a framework for cultivar discrimination and targeted nutritional and industrial applications.

Introduction

The date palm (*Phoenix dactylifera* L.), belongs to Arecaceae, is among the earliest domesticated fruit crops and remains a key agricultural resource in arid and semi-arid regions. For centuries it has played a central cultural, economic, and ecological role in the Arabian Peninsula and the Middle East. In addition to its religious significance in Islamic tradition, it is characterized by remarkable ecological resilience, tolerating extreme temperatures, prolonged drought, and high soil salinity. These adaptive traits make it a fundamental component of sustainable agriculture in desert ecosystems. Worldwide cultivation exceeds 120 million trees, with a large proportion located in the Arabian Peninsula (El-Juhany 2010). Saudi Arabia represents a major center of date palm diversity, hosting nearly 450 cultivars among approximately 5,000 varieties reported globally (Bashah 1996). *Phoenix dactylifera* exhibits substantial genetic and morphological variability, reflected in differences in fruit size, shape, texture, color, and chemical composition (Barrow 1998, Mansour 2005).

Dates are widely recognized as nutrient-rich fruits containing high levels of natural sugars, dietary fiber, and essential minerals such as potassium, magnesium, calcium, and iron (Al-Shahib and Marshall 2002, 2003). They also contain numerous bioactive compounds, including phenolics, flavonoids, sterols, carotenoids, and anthocyanins, which contribute to antioxidant, antimicrobial, and anti-inflammatory activities (Al-Farsi *et al.* 2005, Boudries *et al.* 2007, Baliga *et al.* 2011). In addition, date seeds represent a valuable but underutilized by-product rich in oil, fiber, and fatty acids with potential nutritional and industrial applications (Hamada *et al.* 2002). Traditionally, cultivar identification has relied on morphological descriptors; however, these methods are often labor-intensive and influenced by environmental factors, particularly due to the long juvenile phase of date palms before fruiting (Sedra *et al.* 1998). Consequently, physicochemical characterization combined with advanced analytical techniques has become essential for reliable

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cultivar discrimination. Among these approaches, gas chromatography–mass spectrometry (GC–MS) provides detailed phytochemical profiling and enables accurate differentiation of metabolite composition among cultivars.

Given the economic and nutritional importance of Saudi Arabian date palm cultivars, comprehensive characterization of their structural and chemical attributes is necessary. Therefore, the present study evaluates selected Saudi date cultivars through integrated analysis of morphometric characteristics and phytochemical composition of fruits, seeds, and embryos using GC–MS profiling. This approach aims to enhance cultivar discrimination and provide insights into tissue-specific metabolic specialization and potential nutritional value.

Materials and Methods

Preserved fruits of seven Saudi Arabian date palm cultivars, stored at room temperature under dry conditions, were collected from three major date-producing regions in the Kingdom of Saudi Arabia: Ajwa, Anbar, Labanah, and Safawi from Al-Madīnah Al-Munawwarah (24°28'N, 39°36'E; 619 m), Sukkary and Nabtat Ali from Qassim (27°05'N, 43°28'E; 753 m), and Khalas from Wadi Addawasir (20°27'N, 44°47'E; 624 m). Approximately 0.5 kg of fruits from each cultivar were randomly collected and transported to the laboratory under controlled conditions to preserve their physicochemical integrity before analysis.

In the laboratory, fruits were manually separated into fleshy pericarp, seeds, and embryos. Each component was carefully cleaned, weighed, and prepared for subsequent physical and phytochemical analyses. Fruit color parameters were determined using a Minolta CR-10 chromameter according to the Hunter L*, a*, b* color space system. The color index (CI) was calculated using the equation: $CI = (1000 \times a) / (L \times b)$. Moisture and dry matter contents of fleshy tissues, seeds, and embryos were determined by oven drying at 80 °C for 24 h and calculated from the difference between fresh and dry weights. Morphometric characteristics, including perimeter, projected area, and volume, were estimated from measured dimensions using standard geometric equations. For phytochemical profiling, methanolic extracts of fleshy tissues, seeds, and embryos were analyzed using gas chromatography–mass spectrometry (GC–MS) with a PerkinElmer Clarus 500 system equipped with an Elite-1 capillary column. Helium was used as the carrier gas, and chromatographic separation was achieved under a programmed temperature gradient. Mass spectra were recorded at an ionization energy of 70 eV across a scan range of 40–550 m/z. Compounds were identified by comparison of the obtained spectra with reference spectra from the NIST mass spectral library. To evaluate chemical relationships among cultivars, hierarchical cluster analysis was performed based on the phytochemical profiles obtained from GC–MS analysis. Clustering was conducted using Ward's agglomerative method with Euclidean distance as the similarity metric. This method minimizes within-cluster variance and groups samples according to their overall chemical similarity. Separate dendrograms were constructed for embryo, seed, and fruit flesh extracts, as well as for the combined phytochemical dataset, to visualize patterns of similarity and differentiation among the seven date palm cultivars. All measurements were performed in triplicate, and results are expressed as mean $\pm$ standard error (SE) calculated from three independent replicates.

Results and Discussion

A multidimensional structural–metabolic assessment of seven Saudi Arabian *Phoenix dactylifera* cultivars revealed pronounced cultivar-level differentiation across physicochemical, morphometric, and phytochemical traits. Fruit color parameters measured using the Hunter L*, a*, b* system showed clear variation among the cultivars (Table 1). Labanah and Sukkary exhibited the highest lightness values ($L^* = 50.14$ and 43.57), followed by Nabtat Ali (41.00), whereas

Ajwa and Safawi displayed the darkest fruits (20.57 and 20.29). Redness (a^*) was highest in Khalas and Anbar (83 and 80), while Labanah and Sukkary showed lower values. Yellowness (b^*) varied less, although Labanah, Nabtati Ali, and Sukkary exhibited relatively higher values. The calculated color index classified the cultivars as Ajwa (81.88, Dim gray), Anbar (64.91, Sienna), Khalas (68.12, Maroon), Labanah (10.69, Burly wood), Nabtati Ali (23.81, Saddle brown), Safawi (136.40, Dim gray), and Sukkary (17.82, Golden rod).

Table 1. Fruit colour variation, colour index, and CIELAB colour coordinates of seven Saudi Arabian date palm cultivars.

Colour variation	Colour index	L*	a*	b*	Cultivar
Dim gray	81.88	20.57	64.57	41.57	Ajwa
Sienna	64.91	27.57	80	46.86	Anbar
Maroon	68.12	29.71	83	43.71	Khalas
Burly wood	10.69	50.14	34	64.86	Labanah
Saddle brown	23.81	41	51.14	61.57	Nabtati Ali
Dim gray	136.4	20.29	62.57	30	Safawi
Golden rod	17.82	43.57	39.71	54.29	Sukkary

Table 2. Biomass allocation, moisture content, and morphometric traits of embryo, seed, and fruit flesh among Saudi Arabian date palm cultivars.

Trait	Ajwa	Anbar	Khalas	Labanah	Nabtati Ali	Safawi	Sukkary
Embryo fresh wt. (g)	0.00125	0.0011	0.0008	0.0015	0.00145	0.001	0.00118
Seed fresh wt. (g)	1	1.1	0.63	1.08	1.02	0.82	1.3
Fruit fresh wt. (g)	6.2	12.7	6.3	8.9	12	5.4	9.8
Embryo dry wt. (g)	0.0009	0.00095	0.0007	0.001	0.00103	0.0008	0.00095
Seed dry wt. (g)	0.93	1	0.58	1.02	0.93	0.75	1.2
Fruit dry wt. (g)	5.2	10.3	5.4	8	9.9	4.5	8.4
Embryo dry matter (%)	75	86	87	67	78	80	83
Seed dry matter (%)	94	90	94	94	91	93	91
Fruit dry matter (%)	85	82	86	90	82	85	86
Embryo moisture (%)	25	14	12	33	22	19	17
Seed moisture (%)	6	10	6	6	9	7	9
Fruit moisture (%)	15	18	15	10	17	15	15
Embryo perimeter (mm)	7.8	7.2	5.9	6.8	7.1	6	7.3
Seed perimeter (mm)	58	82	58	75	69	63	65
Fruit perimeter (mm)	100	160	100	135	132	110	120
Embryo projected area (mm ²)	10.5	9.5	5.8	8.4	8.9	6	8.7
Seed projected area (mm ²)	112	145	92	135	138	112	145
Fruit projected area (mm ²)	480	950	430	680	800	480	650
Embryo volume (mm ³)	2.7	2.2	1	1.9	2	1.2	1.9
Seed volume (mm ³)	630	670	410	700	780	560	900
Fruit volume (mm ³)	6000	16000	5800	10000	14500	6200	10500

Integration of biomass allocation, morphometric traits, and GC–MS metabolite profiles indicates that cultivar identity reflects coordinated structural investment and metabolic specialization, consistent with previously reported germplasm heterogeneity (Mansour 2005, Sakr *et al.* 2010) (Table 2). Embryo fresh mass varied nearly two-folds (0.00080–0.00150 g), indicating differences in reserve deposition efficiency. Seed biomass showed stronger divergence, with Sukkary exhibiting the highest seed mass (1.30 g fresh; 1.20 g dry) and Khalas the lowest (0.63 g

fresh; 0.58 g dry). Fruit biomass further reinforced phenotypic differentiation, with Anbar and Nabtat Ali producing the largest fruits, consistent with classical morphological classifications (Khatchadourian *et al.* 1983, Mansour 2005). Seeds exhibited very high dry matter (90-94%) and low moisture (6-10%), conditions associated with enhanced storage stability and longevity (Ellis *et al.* 1990) and linked to seed vigor and reserve accumulation (Henning *et al.* 2010, Pereira *et al.* 2013). Embryos retained higher hydration (12-33%), reflecting their metabolically active status. Morphometric scaling followed a consistent hierarchy (fruit flesh > seed > embryo), confirming structural compartmentalization and cultivar-dependent fruit morphology previously reported in date palm germplasm (Sakr *et al.* 2010).

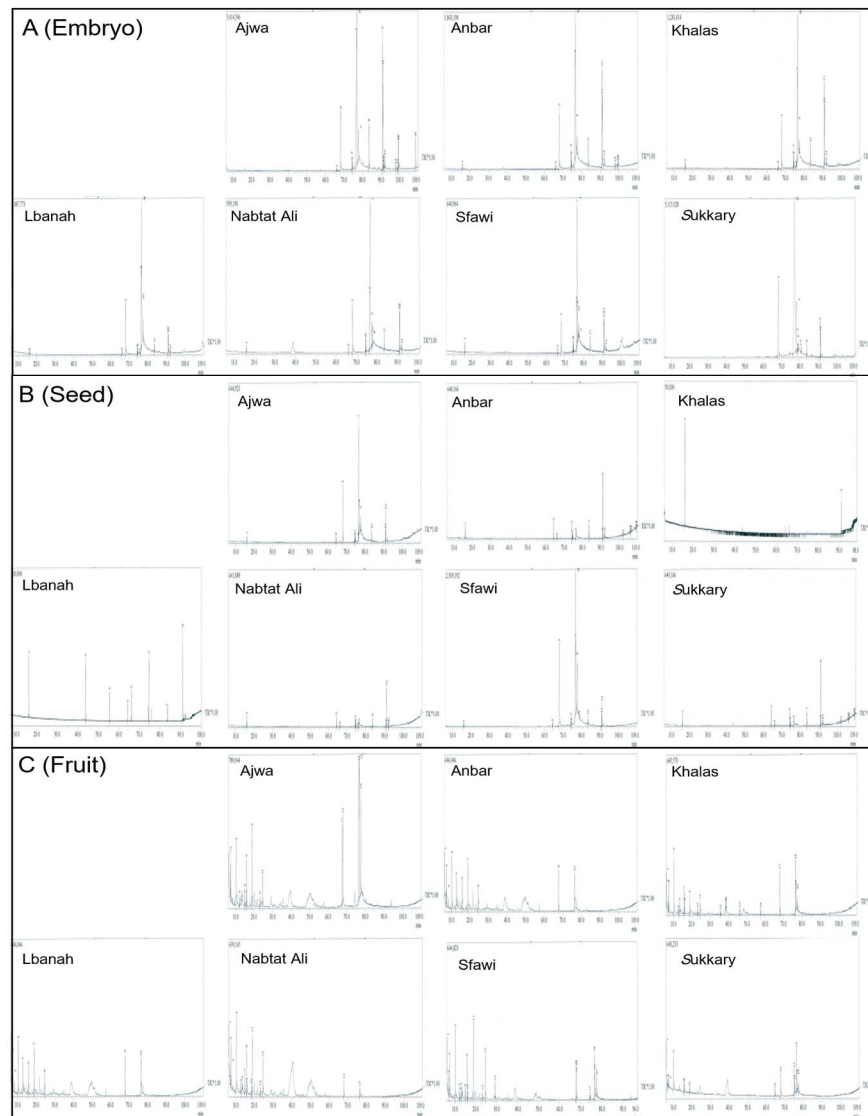


Fig. 1. Comparative GC-MS chromatographic profiles of embryo, seed, and fruit flesh tissues among seven Saudi Arabian date palm cultivars.

GC-MS analysis revealed strong organ-specific metabolic specialization. Embryos and seeds were dominated by long-chain fatty acids. In contrast, fruit flesh contained higher proportions of oxygenated metabolites (Fig. 1). Embryo extracts were particularly enriched in unsaturated C18–C22 fatty acids, with oleic acid and 9,12-octadecadienoic acid representing the principal metabolites. Khalas embryos showed the highest erucic acid level (34.961%), whereas Safawi exhibited the greatest oleic acid enrichment (54.969%). Across all cultivars, n-hexadecanoic acid formed a conserved saturated fatty acid backbone. Minor constituents included glycerol esters, methyl esters, and trace hydrocarbons, confirming embryo tissues as lipid-storage compartments. Seed chemistry showed greater cultivar variability. Oleic acid dominated most cultivars (33.636–47.532%), accompanied by n-hexadecanoic and cis-vaccenic acids, characteristic of lipid-rich storage tissues. Khalas seeds displayed a markedly distinct profile dominated by decane, 6-ethyl-2-methyl- (72.372%) and 1-hexyl-2-nitrocyclohexane (27.627%), indicating strong genotype-driven metabolic divergence. Labanah seeds were enriched in fatty acid methyl esters, including dodecanoic acid methyl ester (24.585%) and 15-tetracosenoic acid methyl ester (11.007%). Fruit flesh extracts exhibited the highest metabolic complexity and were enriched in oxygenated compounds. In most cultivars, 2-hydroxy- γ -butyrolactone dominated (17.296–30.596%), suggesting a potential role of lactone biosynthesis in fruit flesh metabolism. Sukkary fruit flesh showed a slightly different profile dominated by 3-hydroxycyclohexanone (26.499%). Additional oxygenated metabolites, including dl-glyceraldehyde dimer and related derivatives, indicate active carbohydrate-derived oxidative metabolism during fruit flesh maturation. Molecular analysis of zygotic embryos revealed substantial genetic diversity among the seven date palm cultivars. This genetic differentiation corresponds with the observed phytochemical and morphometric variation among cultivars, indicating coordinated genetic and metabolic divergence, consistent with previous molecular marker studies (Aladadi *et al.* 2018).

Hierarchical cluster analysis further confirmed cultivar differentiation (Fig. 2). Embryo phytochemical profiles formed two clusters at linkage distance 8.83: Ajwa-Anbar-Khalas and Labanah-Nabtati Ali-Safawi-Sukkary, with the highest similarity between Labanah and Nabtat Ali (57.89%) and the lowest between Ajwa and Safawi (16%). Seed profiles produced a similar structure at linkage distance 9.5, while fruit flesh metabolites displayed greater dispersion (≈ 6.0 linkage distance). Combined phytochemical data generated three major chemotypic groups at a linkage distance of 22: Ajwa-Anbar-Khalas, Labanah-Nabtati Ali, and Safawi-Sukkary, with the

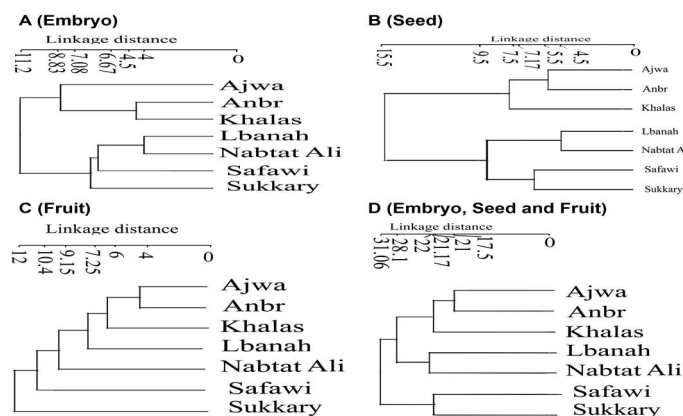


Fig. 2. Hierarchical cluster analysis dendrograms showing relationships among seven Saudi Arabian date palm cultivars based on phytochemical traits in different tissues: A: embryo, B: seed, C: fruit flesh, and D: combined dataset.

highest similarity observed between Ajwa and Anbar (37.5%). Hierarchical clustering provides a robust framework for similarity-based classification (Johnson and Wichern 2002), and the observed chemotypic divergence likely reflects underlying genetic differentiation influenced by environmental factors (Sezik and Basaran 1985). Comparable patterns of genetic diversity detected using RAPD and ISSR molecular markers have been reported in plants (Alyahya *et al.* 2025), where genetic variability is often associated with corresponding phytochemical and morphological differentiation.

Overall, integration of morphometric traits, biomass allocation, and metabolite profiling reveals a coherent model of metabolic compartmentalization in *Phoenix dactylifera*. Embryos represent the principal axis of lipid variability, seeds maintain a conserved storage architecture, and fruits flesh exhibit specialized oxygenated metabolism associated with flavor-related traits. These findings reinforce previously documented germplasm variability (Khatchdourian *et al.* 1983, Ellis *et al.* 1990, Mansour 2005, Henning *et al.* 2010, Sakr *et al.* 2010, Pereira *et al.* 2013) and provide a robust biochemical framework for cultivar discrimination and targeted utilization of Saudi Arabian date palm germplasm. In conclusion, the integrated structural and metabolomic assessment clearly demonstrates pronounced cultivar-level differentiation among the seven Saudi Arabian *P. dactylifera* cultivars. Variations in fruit color parameters, biomass allocation, morphometric scaling, and GC–MS metabolite profiles collectively reveal coordinated structural organization and tissue-specific metabolic specialization across embryo, seed, and fruit compartments. Hierarchical clustering further resolved distinct chemotypic groupings, confirming the robustness of biochemical differentiation among cultivars. These findings provide a comprehensive framework for cultivar discrimination and highlight the substantial biochemical diversity within Saudi Arabian date palm germplasm, supporting its targeted exploitation in breeding, quality optimization, and value-added applications.

Acknowledgements

This work was funded by the Deanship of Research and Graduate Studies at King Khalid University through the Large Research Project under grant number RGP2/113/47.

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(Manuscript received on 07 March, 2026; revised on 30 August, 2026)