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Cold Storage Effects on Quality of Date Fruits (Phoenix dactylifera L.) from The Northern Saharan Oases System: Physicochemical, Biochemical, Microbiological Implication and Prospects for Human Health

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ABSTRACT: Date palm (*Phoenix dactylifera* L.) fruits are a dietary staple in arid regions and a rich source of bioactive compounds. Bent Qbala, a traditional cultivar from the M'zab valley, has been little studied, and how it responds to storage temperature is unknown. We assessed how refrigeration (+4 °C) and freezing (−18 °C) over three months affect its quality, bioactive compounds and microbial load, focusing on phenolic stability.

Tamr-stage fruits harvested in November 2025 were analyzed at harvest and after one and three months (physicochemical, sugar, phenolic, DPPH, LC-MS/MS and microbial parameters) using ANOVA, Tukey's test and PCA.

The fruits (8.20 g; pulp/fruit ratio 87.4 %) met quality criteria for Saharan cultivars. Moisture rose only in frozen fruit (22 to 27 %), shifting texture from dry to semi-soft. After three months, total phenolics fell by 25.6 % at +4 °C but only 4.7 % at −18 °C, and flavonoids and DPPH IC50 followed suit (+47 % vs +11.5 %). Yeast and mould counts exceeded 10^3 CFU g^{-1} in every quantified sample, even fresh fruit (1.08×10^6), and were lower under refrigeration than freezing at three months (1.3×10^5 vs 1.05×10^6). PCA (82.9 % of variance) set the +4 °C/3-month samples apart from all others. This study provides the first integrated assessment of storage options for the minor Saharan date cultivar 'Bent Qbala'. Our findings demonstrate that storage selection for this variety entails a fundamental trade-off between quality retention and microbiological safety a challenge that cannot be resolved solely at the product level. From a biochemical perspective, storage at −18 °C is the most effective approach to minimize the degradation of phenolic compounds and sugars. Preserving these bioactive constituents is essential for maintaining the fruit's nutritional value and potential health benefits, both of which warrant further investigation through *in vivo* and human clinical studies.

1. INTRODUCTION

Date palm (*Phoenix dactylifera* L.) is a keystone crop of arid and semi-arid regions, where it underpins food security, household income and the functioning of oasis agro-ecosystems. Dates are energy-dense fruits rich in carbohydrates, dietary fibre,

vitamins (B, C), minerals (potassium, magnesium, iron) and antioxidant phenolic compounds [1] and in dry regions they often replace fresh fruit during periods of scarcity [2].

Algeria ranks among the world's leading producers, with about 1.32 million tonnes reported [3] and production is concentrated in the Saharan provinces. In the M'zab valley (Ghardaïa province), traditional palm groves in Ghardaïa, Berriane, Atteuf and Melika maintain locally adapted cultivars, among them Bent Qbala, a variety valued for its aromatic flavour and soft flesh but far less studied than Deglet Nour or Ghars.

Dates are energy-dense fruits whose sugar content commonly reaches 70–80 % of dry matter and which also contain phenolic acids, flavonoids and other compounds with antioxidant activity [4,5]. Fruit composition depends on cultivar, maturity stage, climate and cultural practices [6]., and these attributes are modified after harvest by temperature, humidity and packaging.

Because the harvest is concentrated in autumn while demand extends over the year, storage is a central postharvest operation. Fruits at the Tamr stage still hold 20–27 % moisture, which makes them prone to moisture exchange with the environment, sugar and colour changes, insect infestation and spoilage by yeasts and moulds [7,8]. Low-temperature storage is widely recommended to slow these processes [9. 10], but its effects differ between cultivars and storage conditions, and no data exist for Bent Qbala cultivar.

Quality dynamics are, however, only one side of the storage problem. Whether a storage option can be adopted depends on who holds the fruit, who has access to cold facilities, and where value is captured along the chain. Date chains in the Algerian Sahara are characterised by informal collectors, household-level conditioning and the absence of an organized marketing circuit [11]. Studies that link product-level quality dynamics to the structure of the value chain, which is the type of cross-level interaction central to agricultural systems research, remain scarce for minor Saharan cultivars.

This study therefore aimed to (i) describe the value chain of Bent Qbala dates in Ghardaïa; (ii) characterize the fruit morphologically, physicochemically, biochemically and microbiologically at the Tamr stage; (iii) quantify the effect of refrigerated (+4 °C) and frozen (−18 °C) storage over three months; and outlining its implications for human health.

2. MATERIALS AND METHODS

2.1. Study area

Ghardaïa province lies in the centre of the northern Algerian Sahara, 600 km south of Algiers (32°30' N, 3°45' E; altitude 480 m; [12]. and covers 86 560 km² (Fig. 1). It is bordered by Laghouat (200 km) to the north, Djelfa (300 km) to the north-east, Ouargla (190 km) to the east, El Meniaa (240 km) to the south and El Bayadh (350 km) to the west. The province comprises seven dairas and ten communes and is best known for the M'zab valley, a UNESCO World Heritage site with five fortified settlements (ksour) [13].

The climate is typically Saharan, with a hot, dry season (April–September), a temperate season (October–March) and a large difference between summer and winter temperatures [14].According to Emberger's climagram, the province belongs to the Saharan bioclimatic zone with mild winters (Q2 = 3.60) with mean temperature of 25.23 °C, and annual rainfall 41.02 mm [14]. The landscape comprises hamadas, regs, ergs and dayas [15].

Agriculture has expanded rapidly: cultivated land grew by 64 % between 2015 and 2021, from 44,155 to 72,500 ha, an average of 4,724 ha per year [16]. The oases create a specific microclimate and an intensified, irrigated, multi-layered production system [17]. Date palms form the upper storey; fruit trees (citrus, fig, apricot, pomegranate, olive, peach, almond, apple) form the middle storey; and vegetables (squash, watermelon, melon, tomato, eggplant, pepper) and fodder crops (barley, alfalfa, clover) form the lower storey [18, 17, 19].

The dates samples studied in this research originate from the El Atteuf region, located 8 km north of the Ghardaïa province and 606 km south of Algiers. The region lies at a latitude of 32°27' N, a longitude of 3°45' E, and an altitude of 458 m, covering a total area of 750 km² [20].

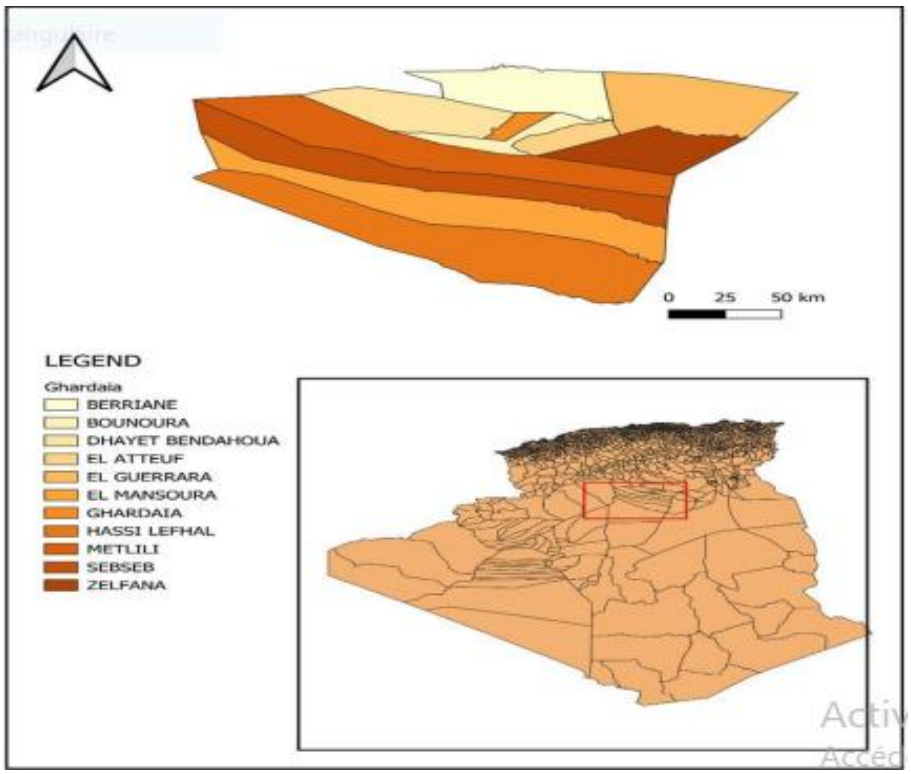


Fig. 1. Location of the study area: Ghardaïa province, north-central Algerian Sahara [20]

2.2. Value chain characterization

The date palm value chain in the Ghardaïa region (particularly within the M'zab Valley and oases like El Atteuf) serves as a vital socio-economic and ecological pillar, characterized by high varietal diversity ranging from Deglet Nour for commercial markets to resilient local varieties like Bent Qbala.

The value chain was described by segment (input supply, production, collection, conditioning/processing and marketing), identifying the actors and the degree of formalization of each segment.

2.3. Plant material and sampling

Fruits of the Bent Qbala cultivar were harvested at the Tamr stage in November 2025 from at least three different palms Fruits were sampled at random to ensure representativeness, cleaned of foreign bodies, placed in labelled polyethylene bags and kept until analysis.

2.4. Storage design

Fruits were packed in transparent polyethylene terephthalate (PET) boxes and stored for three months at +4 °C and −18 °C. Samples were analysed before storage (T0, fruit at ambient temperature, ≈ 25 °C) and after 1 and 3 months of storage (Fig. 2).

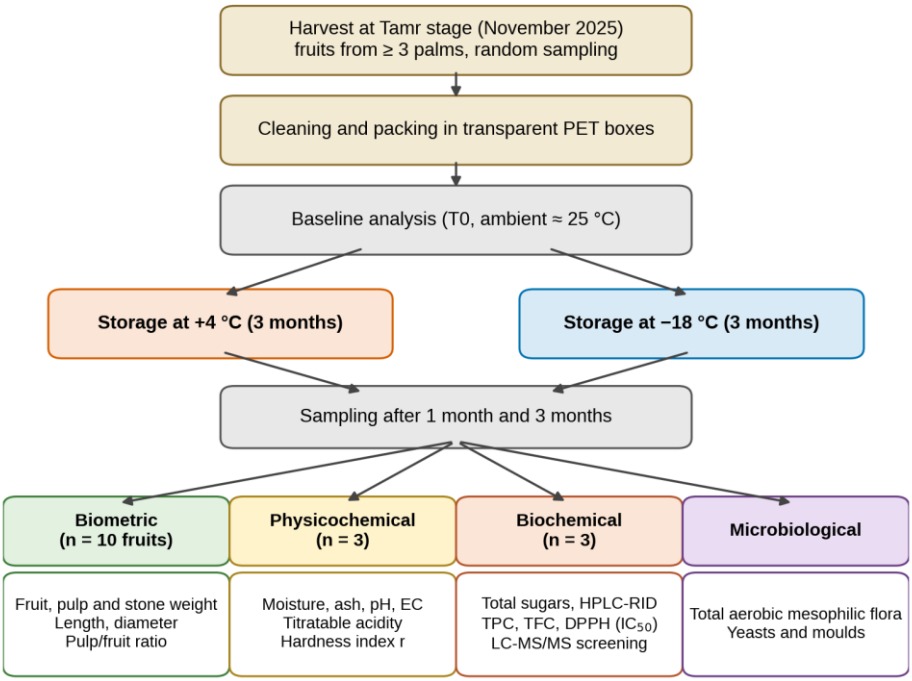


Fig. 2. Experimental design and analyses performed. TPC: total phenolic content; TFC: total flavonoid content; EC: electrical conductivity.

2.5. Biometric measurements

Ten fruits were randomly selected. Each fruit was weighed on a precision balance, and its length and diameter were measured with a dial micrometer. After stoning, the weight, length and diameter of the stone were measured in the same way, and pulp weight was obtained by difference. The pulp/fruit ratio was calculated as pulp weight divided by whole-fruit weight. Results are reported as mean ± standard deviation (SD).

2.6. Physicochemical analyses

All determinations were carried out in triplicate. pH and electrical conductivity (EC) were measured in an aqueous date solution according [21]. Moisture was determined by oven drying at 105 °C for 24 h. Ash was obtained by incineration of the dried sample in a muffle furnace at 550 °C for 5 h, and organic matter as the complement (% ash = 100 − % organic matter) [22]. Titratable acidity was determined using AOAC method 942.15 by extracting organic acids from 2 g of date sample in 100 mL of boiling distilled water. The solution was titrated with 0.1 N NaOH using phenolphthalein, with results expressed as grams of citric acid per 100 g of dates [23]. Total sugars were determined calorimetrically by reading absorbance at 490 nm against a glucose calibration curve (0–0.25 mg mL^{-1}; $y = 4.6286x - 0.0192$; $R^2 = 0.9925$) and are reported as g per 100 g; results were expressed in percentage of fresh weight [24]. The hardness index r was calculated as the ratio of total sugars to moisture content and the fruits classified following [25]: soft dates $r < 2$, semi-soft $2 < r < 3.5$, dry $r > 3.5$.

2.7. Sugar profile by HPLC-RID

Fructose, glucose and sucrose were analyzed by high-performance liquid chromatography with refractive index detection (HPLC-RID) on an amino column The analysis was performed on a SHIMADZU HPLC system equipped with an RID-20A refractive index detector. Chromatographic separation was achieved on a Shim-pack GIST NH2 column (250 x 4.6 mm, 5µm) maintained at 35°C using a mobile phase of acetonitrile and ultra-pure water (80/20, v/v) at a flow rate of 1.0mL/min. An injection volume of 10µL was used, with the sample stored in the autosampler cooler at 15°C. Compounds were identified by comparison of retention times with authentic standards and quantified by external calibration (fructose, 3 points, 24–51; glucose, 4 points, 15–60; sucrose, 4 points, 15–42; concentration units mg mL−1]).

2.8. Phenolic compounds and antioxidant activity

Extracts were prepared following [26] with modifications: 30 g of pulp was macerated in 100 mL of methanol–water (80:20, v/v) on an orbital shaker for 3 h at laboratory temperature. The mixture was filtered and evaporated to dryness in a rotary evaporator at 40 °C to obtain the crude extract, which was kept refrigerated until analysis. All assays were run in triplicate. Total phenolic content (TPC) was determined by the Folin–Ciocalteu assay [27] and expressed as mg gallic acid equivalents per 100 g dry weight (mg GAE 100 g^{-1} DW). Total flavonoid content (TFC) was determined by the aluminium complex method, with maximum absorption at 430 nm [28], and expressed as mg quercetin equivalents per 100 g DW (mg QE 100 g^{-1} DW). Radical-scavenging activity was assessed with the stable DPPH radical [29] by monitoring the decrease of absorbance at 517 nm. Inhibition (%) was calculated as $[(A_{control} - A_{extract}) / A_{control}] \times 100$, where $A_{control}$ is the absorbance of the DPPH solution without extract and $A_{extract}$ that of the extract–DPPH mixture. Activity is expressed as IC_{50}, the extract concentration (mg mL^{-1}) that inhibits 50 % of the DPPH radical; a higher IC_{50} therefore indicates lower activity.

2.9. Targeted LC-MS/MS screening

A targeted screening of 40 phenolic, terpenoid, vitamin and amino-acid compounds was performed by liquid chromatography–tandem mass spectrometry in multiple reaction monitoring (MRM) mode using a Shimadzu 8040 UPLC-ESI-MS/MS system equipped with a Nexera XR LC-20AD binary pump. Separation was achieved on a Restek Ultra C18 column (150×4.6 mm, 3 µm) using a mobile phase consisting of water with 0.1% formic acid (Solvent A) and methanol (Solvent B) at a flow rate of 0.2 mL/min and an injection volume of 5 µL. Electrospray ionization (ESI) was operated with CID gas at 230 kPa, conversion dynode at −6.00 kV, DL temperature at 250 ∘ C, heat block at 400 ∘ C, nebulizing gas flow at 3.00 L/min, and drying gas flow at 10 L/min. Compounds were considered detected when a chromatographic peak was found at the expected retention time; peak areas are reported as semi-quantitative indicators because no calibration was performed.

2.10. Microbiological analyses

Total aerobic mesophilic flora (TAMF) and yeasts and moulds were enumerated and expressed as colony-forming units per gram (CFU g^{-1}). The total mesophilic aerobic flora was enumerated according to standard NF ISO 4833-2008, while yeasts and molds were enumerated following standard NF ISO 7218. Results were compared with the reference limits of 10^6 CFU g^{-1} for TAMF and 10^3 CFU g^{-1} for yeasts and moulds.

2.11. Statistical analysis

Statistical analyses were conducted using XLSTAT 2014. Data were analyzed using analysis of variance (ANOVA) followed by Tukey's post-hoc test for multiple comparisons at a 95% confidence level ($p < 0.05$). Mean parameter values were subsequently subjected to principal component analysis (PCA) and Pearson correlation analysis to evaluate relationships among variables.

3. RESULTS

3.1. Structure of the Bent Qbala value chain

The chain comprises five segments (Fig. 3): input supply, production, collection, conditioning/processing and marketing, involving growers, input suppliers, collectors, storers, packers, traders and exporters. A single actor often holds several roles; a grower may also act as collector, storer, trader or exporter.

Input supply covers pollen and manure, provided by informal actors, together with fertilisers, plastics, packaging and equipment, supplied mostly through formal channels, and financing structures. Production is carried out by date growers, who maintain the palm groves and harvest the fruit once ripe, two steps that largely determine the quality of the final product. Collection is the least formalised and least documented link: it does not appear in the official list of activities of the Ministry of Commerce, and its actors prefer to operate informally to avoid taxes and state control. Conditioning and processing are generally carried out by producer households, where Bent Qbala is made into syrup or packed in wool sacks or plastic buckets, in bulk or in individual packs, for the local market. Marketing is marked by the absence of an organised commercial circuit, which creates problems for the disposal of a high-value product.

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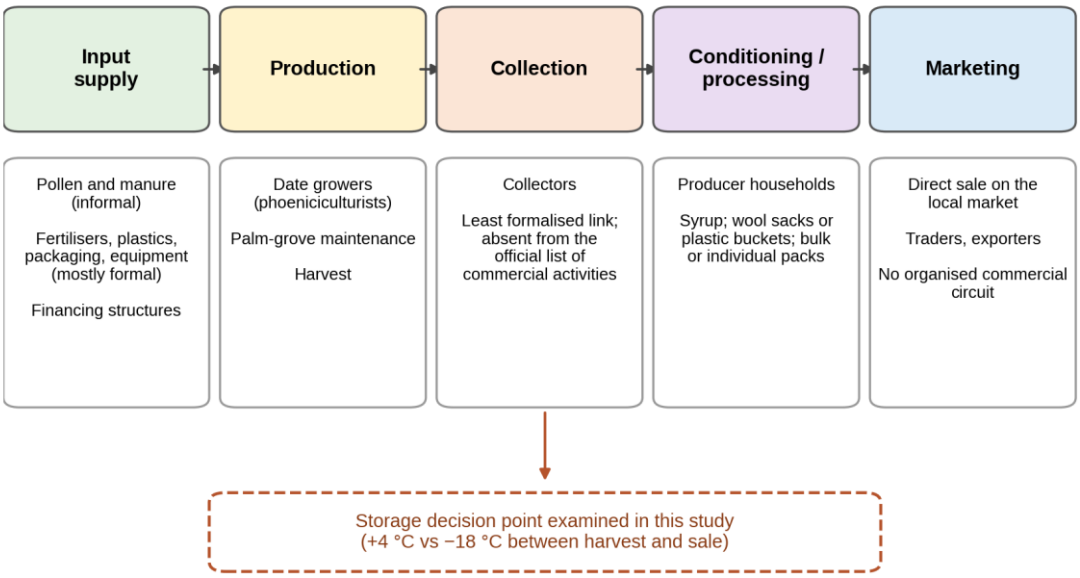


Fig. 3. Schematic of the Bent Qbala value chain in Ghardaïa. The dashed box indicates where the storage options examined in this study would intervene.

This sector relies on a dual model of traditional, small-scale oasis agriculture and modern agricultural extensions equipped with advanced technologies like drip irrigation and solar pumping. Managed by smallholders, cooperatives, and intermediaries, the value chain is increasingly driven by food processing innovations, organic certification, and commercial investments, yet it continues to face critical challenges, including water scarcity, soil salinity, and a shortage of skilled labor needed to maintain aging palm groves.

3.2. Morphology and biometry

Bent Qbala fruits are ovoid, amber-brown, soft, tender and slightly fibrous, with an aromatic flavour (Table 1). Mean fruit weight was 8.20 g, with 7.17 g of pulp and 1.03 g of stone, giving a pulp/fruit ratio of 87.43 % (Table 2). Mean length and width were 3.81 and 2.25 cm.

Table 1. Morphological descriptors of Bent Qbala dates.

	Shape	**Colour**	**Consistency**	**Plasticity**	**Texture**	**Taste**
Bent Qbala	Ovoid	Amber brown	Soft	Tender	Fibrous	Aromatic

Table 2. Biometric characteristics of Bent Qbala dates (mean ± SD, n = 10).

	Length (cm)	**Width (cm)**	**Fruit weight (g)**	**Pulp weight (g)**	**Stone weight (g)**	**Pulp/fruit (%)**
Mean ± SD	3.81 ± 1.93	2.25 ± 0.92	8.20 ± 0.48	7.17 ± 0.52	1.03 ± 0.62	87.43

All the criteria for acceptable physical quality of dates (whole fruit ≥ 6 g, pulp ≥ 5 g, length ≥ 3.5 cm, diameter ≥ 1.5 cm; [30; 31;8] were met. Under the grading scheme used here, weight (> 8 g) falls in the "good" class and length (3.5–4 cm) in the "acceptable" class. Fruit weight was lower than the values reported for Bent Qbala by ([32] ;13.41 g) and ([33]; 10.75 g),

whereas width (2.25 cm) was close to the 2.26 cm reported by the former, whose fruits were longer (4.36 cm). Typical weights for this cultivar in Algeria range from 8 to 13 g [34].

3.3. Physicochemical changes during storage

Moisture, EC and total sugars differed significantly among the five conditions (one-way ANOVA, $P < 0.001$), pH differed weakly ($P = 0.039$), and ash and titratable acidity did not change (Table 3, Fig. 4).

Table 3. Physicochemical characteristics of Bent Qbala dates before and during storage (mean ± SD, n = 3). Within a column, means followed by different letters differ significantly (Tukey's HSD, $P < 0.05$).

Condition	Moisture (%)	pH	EC (ms)	Ash (%)	Titratable acidity (%)	Total sugars (g 100 g^{-1})	Index r
Initial (T0)	22.0 ± 0.19 c	6.30 ± 0.01 b	2.30 ± 0.03 c	1.79 ± 0.03 a	0.4 ± 0.06	80 ± 0.23 a	3.64
+4 °C, 1 month	22.0 ± 1.70 c	6.49 ± 0.18 ab	2.10 ± 0.06 d	1.79 ± 0.05 a	0.4 ± 0.06	76 ± 0.09 c	3.45
−18 °C, 1 month	25.0 ± 0.55 ab	6.69 ± 0.23 a	2.44 ± 0.02 b	1.79 ± 0.04 a	0.4 ± 0.06	77 ± 0.21 b	3.08
+4 °C, 3 months	24.0 ± 0.37 bc	6.34 ± 0.05 ab	2.79 ± 0.015 a	1.79 ± 0.09 a	0.4 ± 0.06	74 ± 0.12 e	3.08
−18 °C, 3 months	27.0 ± 0.89 a	6.42 ± 0.07 ab	2.30 ± 0.047 c	1.79 ± 0.02 a	0.4 ± 0.06	75 ± 0.33 d	2.78
F (4, 10)	16.2***	3.83*	134.3***	0.00 ns	–	348.1***	–

*** $P < 0.001$; * $P < 0.05$; ns, not significant. Index r = total sugars / moisture, calculated from the means.

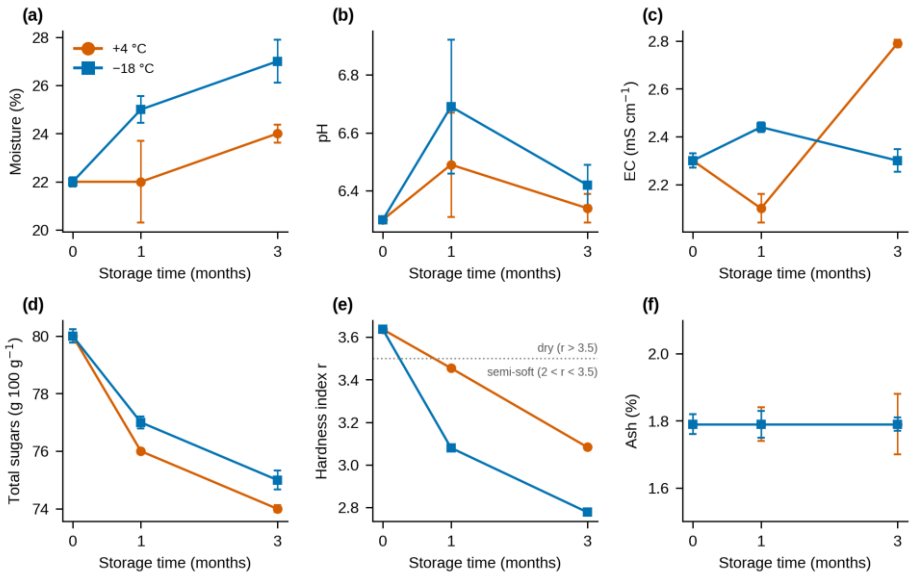


Fig. 4. Changes in (a) moisture, (b) pH, (c) electrical conductivity, (d) total sugars, (e) hardness index r and (f) ash of Bent Qbala dates stored at +4 °C and −18 °C. Error bars are SD (n = 3). Dotted line in (e): limit between dry and semi-soft dates [25].

Moisture was 22 % at harvest and stayed at 22 % after one month at +4 °C, but rose to 25 % after one month and 27 % after three months at −18 °C; at +4 °C it reached 24 % at three months. Moisture was thus affected by both temperature ($F_{1,8}$ = 26.2, P < 0.001) and duration ($F_{1,8}$ = 11.6, P = 0.009) without interaction (Table 4). The lower the storage temperature, the higher the moisture, and the −18 °C/3-month samples had the highest moisture of the experiment (P < 0.001 against T0).

Table 4. Two-way ANOVA (storage temperature × duration; four storage conditions, n = 3; F with 1 and 8 degrees of freedom).

Variable	Temperature	Duration	Temperature × duration
Moisture	26.2 (P < 0.001)	11.6 (P = 0.009)	0.0 (P = 1.00)
pH	2.5 (P = 0.15)	5.7 (P = 0.044)	0.5 (P = 0.51)
EC	10.5 (P = 0.012)	141.0 (P < 0.001)	321.2 (P < 0.001)
Total sugars	68.4 (P < 0.001)	273.5 (P < 0.001)	0.0 (P = 1.00)
Ash	0.0 (P = 1.00)	0.0 (P = 1.00)	0.0 (P = 1.00)

pH ranged from 6.30 to 6.69 and was only marginally affected: the sole significant difference was between T0 (6.30) and −18 °C/1 month (6.69). Values remained close to neutrality throughout. EC ranged from 2.10 to 2.79 mS cm^{-1}; it first decreased at +4 °C and increased at −18 °C after one month, then reached its maximum at +4 °C after three months, producing a strong temperature × duration interaction. Ash content was identical in all conditions (1.79 %), and titratable acidity remained at 0.4 ± 0.06 %.

3.4. Total sugars and hardness index

Total sugars decreased from 80 % at harvest to 76 % (+4 °C) and 77 % (−18 °C) after one month and to 74 % and 75 % after three months; all five conditions differed significantly (Table 3). Both temperature (P < 0.001) and duration (P < 0.001) were significant, without interaction. Relative to T0 this corresponds to losses of 5.0 and 3.8 % after one month and 7.5 and 6.3 % after three months, i.e. slightly lower at −18 °C. The hardness index r was 3.64 at harvest (dry dates, r > 3.5) and fell to 3.45 and 3.08 after one month and to 3.08 and 2.78 after three months, which places all stored samples in the semi-soft class (2 < r < 3.5); the lowest value was at −18 °C after three months, the condition with the highest moisture.

3.5. Sugar profile by HPLC-RID

Fructose (retention time 6.04 min) and glucose (6.49 min) were identified, whereas sucrose was not identified (Table 5). Fructose accounted for 68 % and glucose for 32 % of the two monosaccharides, a fructose: glucose ratio of 2.15. The fructose calibration was excellent (R^2 = 0.9993) but based on three points; the glucose calibration was acceptable (R^2 = 0.9937, maximum back-calculation error ≈ 9 % at the lowest levels), and the sample glucose concentration was at the bottom of its range, so the glucose value and the ratio are the least reliable. The sucrose calibration (R^2 = 0.8906) was not suitable for quantification, so the absence of sucrose is not conclusive.

Table 5. HPLC-RID sugar profile of Bent Qbala dates fruit.

Sugar	RT (min)	Peak area	Calibration	Concentration	Share of F + G (%)
Fructose	6.037	736,322	y = 19432.8x + 100950; R^2 = 0.9993 (3 points; 24–51)	32.7	68
Glucose	6.494	467,565	y = 21533.1x + 140857; R^2 = 0.9937 (4 points; 15–60)	15.2	32

Sugar	RT (min)	Peak area	Calibration	Concentration	Share of F + G (%)
Sucrose	not identified	–	y = 34074.9x + 58456.6; R^2 = 0.8906 (4 points; 15–42)	not quantifiable	–

3.6. Phenolic compounds and antioxidant activity

TPC was 430 mg GAE 100 g^{-1} DW at harvest (Table 6, Fig. 5). After one month it was 390 (−9.3 %) at +4 °C and 420 (−2.3 %) at −18 °C; after three months 320 (−25.6 %) and 410 (−4.7 %), respectively. TFC followed the same pattern: 25.03 mg QE 100 g^{-1} DW at harvest, 23.19 and 24.07 after one month, and 19.42 (−22.4 %) and 23.56 (−5.9 %) after three months. Antioxidant activity decreased accordingly: IC_{50} rose from 9.28 mg mL^{-1} to 10.89 and 9.74 after one month and to 13.64 (+47.0 %) and 10.35 (+11.5 %) after three months. Losses at +4 °C were thus 4–5 times those at −18 °C after three months for TPC (25.6 vs 4.7 %), and the divergence between temperatures widened with time for all three indicators.

Table 6. Total phenolic content (TPC), total flavonoid content (TFC) and DPPH IC_{50} of Bent Qbala extracts during storage .

Variable	Initial	+4 °C, 1 mo	−18 °C, 1 mo	+4 °C, 3 mo	−18 °C, 3 mo
TPC (mg GAE 100 g^{-1} DW)	430	390 (−9.3 %)	420 (−2.3 %)	320 (−25.6 %)	410 (−4.7 %)
TFC (mg QE 100 g^{-1} DW)	25.03	23.19 (−7.4 %)	24.07 (−3.8 %)	19.42 (−22.4 %)	23.56 (−5.9 %)
DPPH IC_{50} (mg mL^{-1})	9.28	10.89 (+17.3 %)	9.74 (+5.0 %)	13.64 (+47.0 %)	10.35 (+11.5 %)

Higher IC_{50} = lower antioxidant activity.

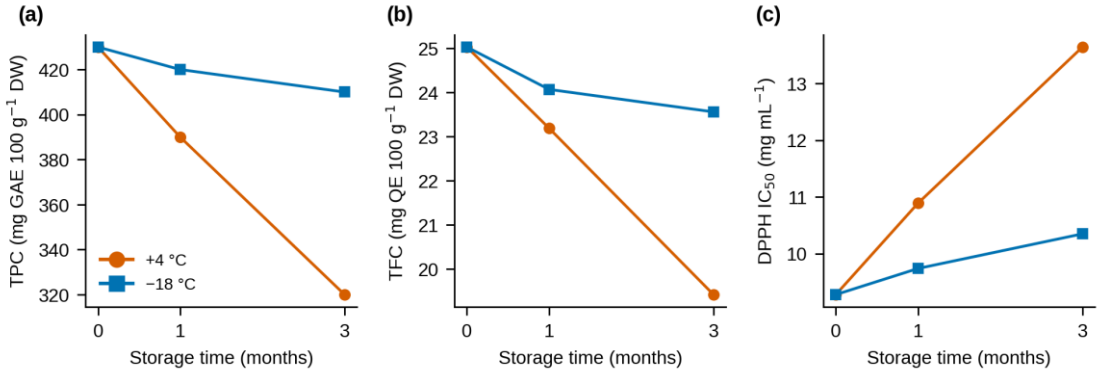


Fig. 5. Changes in (a) total phenolic content, (b) total flavonoid content and (c) DPPH IC_{50} of Bent Qbala extracts during storage at +4 °C and −18 °C.

3.7. Targeted LC-MS/MS screening

Twenty-two of the 40 target compounds produced a peak (Table 7, Fig. 6). Three of them, AAPH (a radical initiator), 4-aminoantipyrine and Sudan III (a dye), are laboratory reagents or dyes rather than date constituents and are not considered further, leaving 19 candidate natural compounds. L-Proline (peak area 5.57 × 10^6) and catechin (5.37 × 10^6) gave the strongest signals, followed by salicylic (1.98 × 10^6) and ferulic acids (1.04 × 10^6) and resveratrol (8.5 × 10^5). Flavonoid glycosides and aglycones (quercetin-3-glucoside, myricetin, naringenin, chrysin-6-C-glucoside, rutin, oroxylin A) were detected at lower levels. Eighteen compounds were not detected, including quercetin, luteolin, apigenin, chrysin, epicatechin, caffeic,

vanillic and sinapic acids, vanillin and curcumin. Oleuropein, oleanolic acid, thymol and β-carotene are unusual in dates and should be regarded as tentative until confirmed with a second transition and an authentic standard.

Table 7. Compounds detected by targeted LC-MS/MS (MRM) in Bent Qbala dates. Peak areas in arbitrary units.

Family	Compound (peak area)
Flavan-3-ol	Catechin (5.37×10^6)
Phenolic acids	Salicylic (1.98×10^6), ferulic (1.04×10^6), gallic (7.0×10^4), p-coumaric (6.9×10^4)
Stilbene	Resveratrol (8.48×10^5)
Flavonoids	Quercetin-3-glucoside (3.61×10^5), myricetin (1.56×10^5), naringenin (7.9×10^4), chrysin-6-C-glucoside (2.7×10^4), rutin (1.55×10^4), oroxylin A (1.6×10^3)
Terpenoids and others*	Oleuropein (1.59×10^5), oleanolic acid (1.43×10^5), thymol (1.22×10^5), β-carotene (9.6×10^4)
Vitamins and amino acids	L-Proline (5.57×10^6), riboflavin (6.45×10^5), nicotinic acid (2.47×10^5)

* Tentative identification. Reagents and dyes (AAPH, 4-aminoantipyrine, Sudan III) detected in the run are excluded.

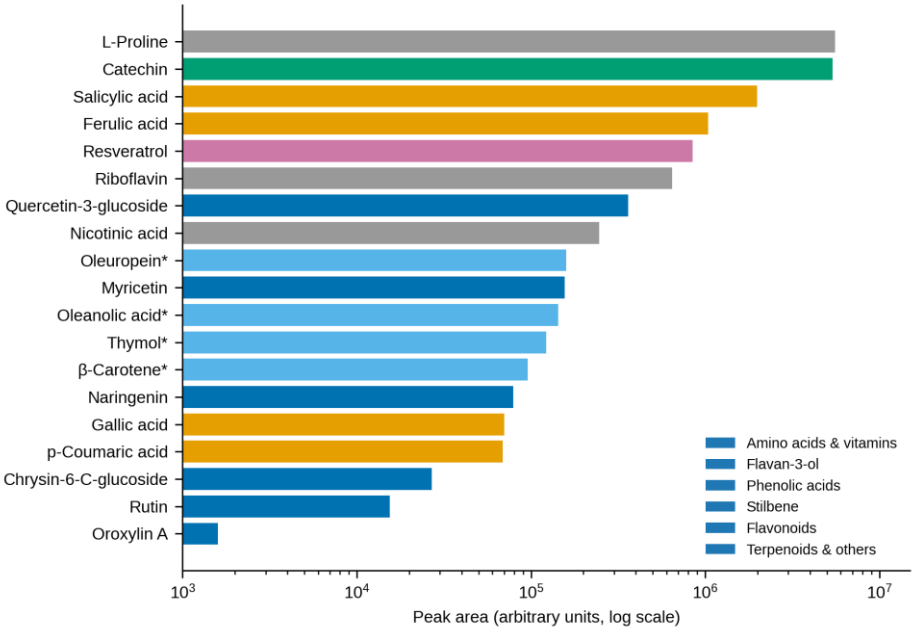


Fig. 6. Peak areas of the 19 natural compounds detected by LC-MS/MS (log scale). * Tentative identification.

3.8. Microbiological quality

TAMF counts were 2.35×10^3 CFU g^{-1} at harvest and remained between 1.25 and 2.45×10^3 in all stored samples, well below the 10^6 limit (Table 8, Fig. 7). The differences, at most 0.3 log units, are within the usual variability of plate counts and indicate stability rather than a treatment effect. Yeasts and moulds were far more variable: the fresh fruit already carried 1.08×10^6 CFU g^{-1}; counts fell to 7×10^3 after one month at +4 °C (−2.2 log) and were reported as not significant after one month at −18 °C, but after three months they were 1.3×10^5 at +4 °C and 1.05×10^6 at −18 °C, the latter equal to the initial value. All quantified yeast and mould counts exceeded the 10^3 CFU g^{-1} threshold.

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Table 8. Microbiological counts (CFU g^{-1}) of Bent Qbala dates before and during storage.

Group	Initial	+4 °C, 1 mo	−18 °C, 1 mo	+4 °C, 3 mo	−18 °C, 3 mo	Limit
Total aerobic mesophilic flora	2.35×10^3	1.55×10^3	2.45×10^3	1.25×10^3	1.35×10^3	$< 10^6$
Yeasts and moulds	1.08×10^6	7×10^3	8.5×10^4	1.3×10^5	1.05×10^6	$< 10^3$

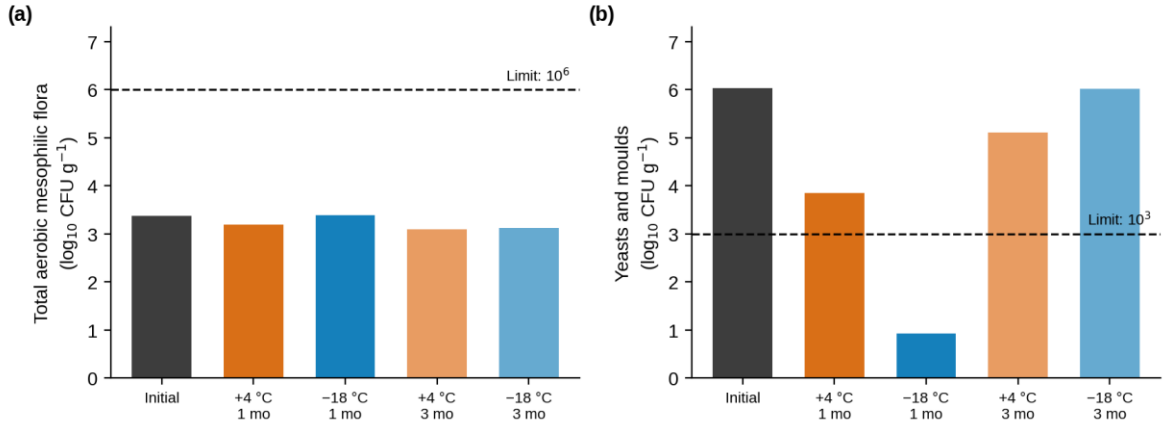


Fig. 7. Microbial counts of Bent Qbala dates during storage: (a) total aerobic mesophilic flora and (b) yeasts and moulds. Dashed lines: reference limits.

3.9. Multivariate structure: correlations and PCA

Across the five conditions, TPC, TFC and IC_{50} were almost perfectly correlated (TPC–TFC r = 0.99; TPC–IC_{50} and TFC–IC_{50} r = −1.00), the only significant relationships besides the arithmetic coupling between moisture and index r (r = −0.96; critical |r| = 0.878 for n = 5, P < 0.05; Fig. 8). Total sugars were positively associated with TPC (0.75) and TFC (0.79) and negatively with IC_{50} (−0.78), and EC was negatively related to TPC (−0.71) and TFC (−0.76), but these did not reach significance with five observations.

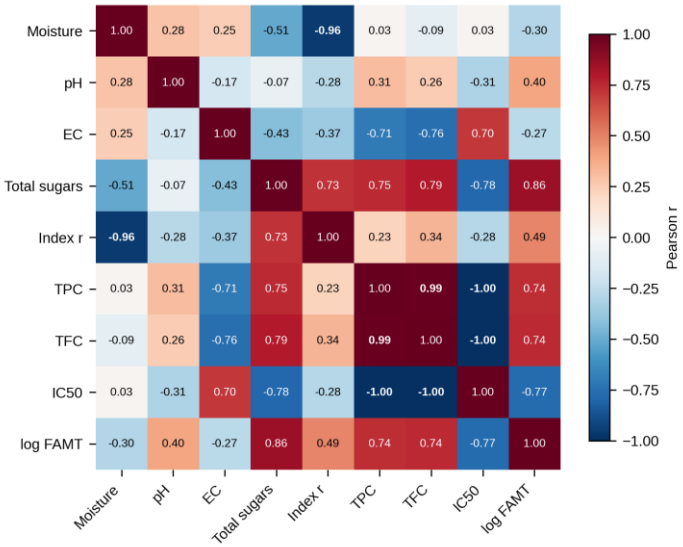


Fig. 8. Pearson correlation matrix among the quality, bioactive and microbial variables across the five storage conditions. Values in bold are significant at P < 0.05 (|r| ≥ 0.878).

The first two principal components explained 61.3 and 21.6 % of the variance (82.9 % cumulated; eigenvalues 4.29 and 1.51; Table 9, Fig. 9a). PC1 was defined by TPC (loading 0.97), TFC (0.99), total sugars (0.82), IC_{50} (−0.98) and EC (−0.78), i.e. an axis from preserved to degraded bioactive and sugar quality. PC2 was defined by moisture (0.85) and pH (0.74) and ordered the samples by moisture gain. The +4 °C/3-month sample was isolated at the negative end of PC1 (score −3.49; high EC and IC_{50}, low TPC, TFC and sugars), while the harvest sample lay at the positive end of PC1 and the negative end of PC2. The −18 °C samples had positive PC2 scores (moisture-rich) and moderate PC1 scores close to the +4 °C/1-month sample (Fig. 9b). The supplementary hardness index r was aligned with the negative direction of PC2 (correlation −0.83), and log TAMF with PC1 (0.77). PC1 is dominated by the +4 °C/3-month sample, which contributes about 71 % of its inertia.

Table 9. PCA results: correlations of variables with the first two principal components and sample scores.

Active variable	PC1 (61.3 %)	PC2 (21.6 %)	Sample score (PC1 ; PC2)
Moisture	−0.19	0.85	Initial: 1.95 ; −1.55
pH	0.27	0.74	+4 °C, 1 mo: 0.43 ; −0.47
EC	−0.78	0.03	−18 °C, 1 mo: 0.97 ; 1.35
Total sugars	0.82	−0.44	+4 °C, 3 mo: −3.49 ; −0.50
TPC	0.97	0.17	−18 °C, 3 mo: 0.13 ; 1.17
TFC	0.99	0.06	
DPPH IC_{50}	−0.98	−0.13	

Supplementary variables: hardness index r (PC1 0.42; PC2 −0.83); log TAMF (PC1 0.77; PC2 −0.05).

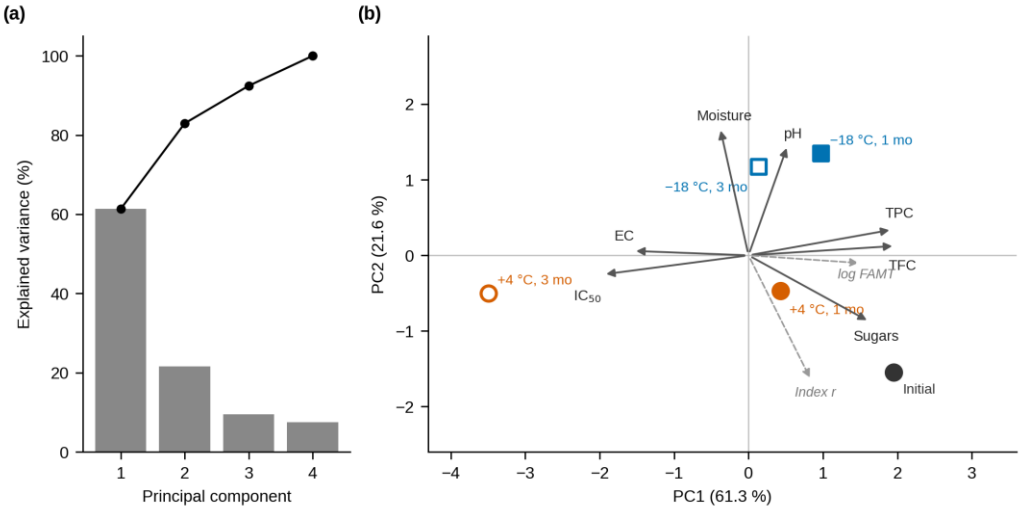


Fig. 9. PCA of storage conditions: (a) explained variance (bars) and cumulative variance (line); (b) biplot of the first two components. Solid arrows: active variables; dashed grey arrows: supplementary variables. Filled symbols: harvest and 1 month; open symbols: 3 months.

4. DISCUSSION

4.1. Fruit quality and its dependence on the production system

The fruits studied satisfied the physical quality criteria for dates, and their high pulp/fruit ratio (87.4 %) is typical of a good commercial quality [25]. Their weight was, however, below published values for the same cultivar, which, as the literature on

dates stresses, can arise from differences in climate, soil, irrigation, cultural practices and genetic material [34]. The chemical composition: moisture (22 %), pH (6.3), ash (1.79 %) and sugars (≈ 80 %) at harvest lie within the ranges reported for Algerian and other date cultivars [1; 8], but without comparable storage studies on Bent Qbala these values serve mainly as a baseline.

4.2. Moisture uptake and texture under cold storage

The most visible physical effect of storage was a gain in moisture, largest at −18 °C, which moved the hardness index from the dry to the semi-soft class. This result is consistent with fruit equilibrating with the humidity inside the box and the room, as discussed by [35] for stored dates, and with the fact that colder storage rooms and freezers hold air near saturation. this explanation remains a hypothesis. Its practical relevance is that moisture of 25–27 % is at the upper end of what grading schemes consider acceptable (25–30 %) and approaches the range in which yeasts and moulds can grow, which links the physical result to the microbiological one (section 4.4). It also means that the "soft" texture reported at harvest (Table 1) and the dry classification at T0 (r = 3.64) are not fully consistent, which may reflect the sugar basis issue noted in the Methods.

4.3. Sugars and bioactive compounds

Total sugars declined modestly (relative losses of 4–8 %), slightly less at −18 °C, so that cold storage did not compromise the sweet profile that characterizes the cultivar. By contrast, bioactive compounds were clearly temperature-sensitive: after three months at +4 °C, TPC and TFC fell by about a quarter and DPPH IC_{50} rose by nearly half, while at −18 °C the corresponding changes were 5–6 % and 11.5 %. This agrees in direction with reports that low-temperature storage helps retain phenolic compounds in dates [5;1]. The usual explanation, residual polyphenol oxidase activity and oxidation at refrigeration temperature that are largely arrested in the frozen state, is plausible.

The LC-MS/MS screening adds a qualitative picture. Catechin, ferulic and salicylic acids, resveratrol and flavonoid glycosides such as quercetin-3-glucoside are plausible contributors to the phenolic pool and to the antioxidant activity, and the dominance of L-proline matches the amino-acid richness of dates. The aglycones quercetin and luteolin were not detected. A larger fructose than glucose fraction (ratio 2.15) is higher than the near-equal proportions commonly reported for invert-sugar cultivars; given the weak calibration at the glucose level.

4.4. A quality–safety trade-off between +4 °C and −18 °C

The microbiological data qualify the chemical benefits of freezing. TAMF levels remained low across the board, but yeast and mold counts exceeded the reference value of 10^3 CFU/g in all samples analyzed, including fresh fruit. This suggests contamination or moisture accumulation prior to storage (during drying, handling, or harvesting), in addition to a break in the cold chain during storage. In this context, the temperature of +4 °C significantly reduced the levels after one month (−2.2 log) and maintained them at 0.9 log below the value observed at −18 °C after three months; however, the temperature of −18 °C better preserved the phenolic compounds and antioxidant activity, whereas the +4 °C temperature resulted in a lower microbial load (yeasts and molds) after three months, provided that the samples stored at −18 °C suffered a cold chain break, which skewed the values results that still need to be replicated.

After one month, storage at −18 °C proved to be equivalent to, or even superior to, storage at +4 °C in terms of sugars, total cell content (TCC), total fiber content (TFC), IC_{50}, and the number of yeasts and molds; it was inferior only in terms of moisture content and the resulting changes in texture. The results indicate that a temperature of −18 °C is the best choice for preserving chemical quality over both storage periods, provided that moisture absorption is limited by the packaging and that the initial microbial load is low, ensuring product hygiene and compliance with good storage practices.

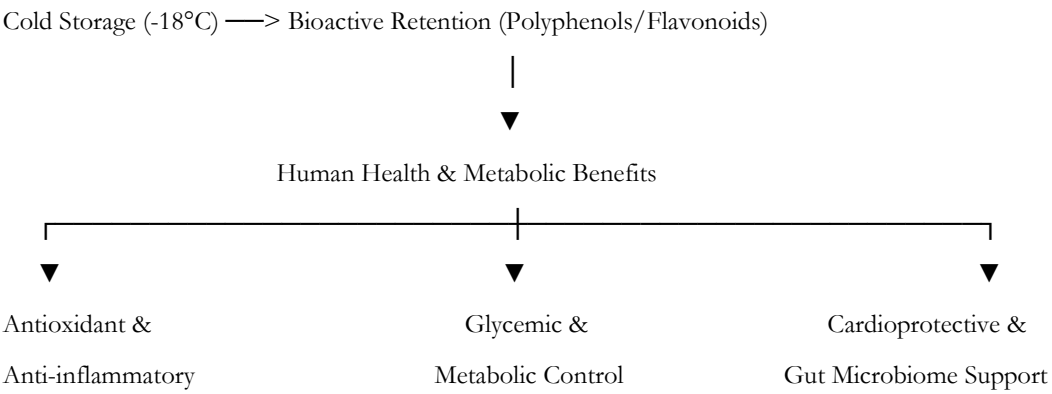
4.5. Implications for the value chain and for agricultural systems

Based on the results obtained, three implications for the supply chain emerge. First, the critical microbiological risk occurs prior to the storage stage. The high initial yeast and mold load stems from the stages (harvesting, sorting, collection, and home packaging) which, as shown in Section 3.1, are carried out by informal actors subject to limited oversight; consequently, investment in cold storage alone will not be sufficient to ensure compliance. Interventions at the interface between the producer and the collector such as adhering to hygiene rules during harvesting and drying, using clean containers, and checking moisture content before storage are likely to yield greater benefits than simply choosing between +4 and −18 °C. Second, the trade-off between quality and safety means that the best storage option depends on the product's intended use.

When a product's value lies in its nutritional and antioxidant properties (for example, for high-end or health-focused markets), freezing preserves these qualities more effectively, whereas for batches intended for rapid sale in local markets, refrigeration is sufficient and less costly. Third, the lack of an organized marketing system reduces the incentive to invest in storage, since the price premium associated with a better-preserved product cannot be easily captured.

4.6. Prospects for Human Health and Industrial Valorization

The retention of phenolic compounds and dietary fiber through optimized cold chain management directly impacts the therapeutic and nutraceutic potential of date fruits.



4.7. Antioxidant and Anti-Inflammatory Properties

Polyphenols preserved in cold-stored dates act as potent free radical scavengers, mitigating oxidative stress and downregulating pro-inflammatory pathways (such as NF-ϰB activation). This supports potential therapeutic applications in managing chronic inflammatory diseases [36].

4.8. Glycemic Management and Metabolic Health

Despite their high sugar content, dates exhibit a low-to-medium glycemic index (GI) due to their high soluble fiber and polyphenolic matrix, which inhibits α-glucosidase and α-amylase activities [37]. Preserving these enzyme-inhibiting polyphenols via low-temperature storage supports their inclusion in diabetic-friendly functional food formulations.

4.9. Pharmaceutical and Industrial Valorization

Cold-stored date matrices rich in bioactive compounds serve as valuable raw materials for:

- **Nutraceutical Formulations:** Standardized phenolic extracts utilized in dietary supplements.
- **Functional Food Ingredients:** Natural antioxidants replacing synthetic additives (e.g., BHT/BHA) in food systems.
- **Pharmaceutical Intermediates:** Prebiotic and anti-microbial fractions for gastrointestinal health formulations.

4.10. Limitations

Although our study provides a detailed characterization of several physicochemical variables, as well as the polyphenolic (LC-MS/MS), sugar (HPLC-RID), and microbiological profiles of the cultivar under study, certain limitations must be highlighted. On the one hand, the evaluation, which was limited to a single site, a single cultivar, and a single harvest season, does not account for interannual, edaphic, and genetic variability. Furthermore, the absence of measurements of water activity (aw), color, texture profile, and sensory evaluation limits the overall understanding of the product's technological and commercial stability during storage.

the bioavailability and in vivo therapeutic efficacy of the retained polyphenols were not evaluated in the present study. Further clinical investigations are therefore needed to determine their biological effects and validate their potential applications in functional foods and pharmaceutical products

5. CONCLUSION

Overall, our results provided insights into the biometric parameters, physicochemical composition, and bioactivity of Bent Qbala dates from Ghardaïa oases (Algeria). The studied date cultivar showed good physical quality, with a high pulp-to-fruit ratio, a high sugar content and a substantial phenolic content. Over three months of cold storage, sugars declined only moderately, while ash and titratable acidity remained stable. Storage temperature had the strongest effect on bioactive compounds: refrigeration at +4 °C caused marked losses in total phenolic and flavonoid contents and a pronounced decline in antioxidant activity (DPPH), whereas these changes were much smaller at −18 °C. Frozen storage, however, increased moisture content and shifted the texture from dry to semi-soft. Yeast and mould counts exceeded the stated limit under nearly all storage conditions. PCA summarized these results along two axes: a bioactive-preservation axis, which clearly separated the +4 °C/3-month samples from all other conditions, and a moisture axis, which ordered the frozen samples.

Within the value chain, these results show that the choice between refrigeration and freezing is a quality safety trade-off, and that the high initial microbial load, linked to upstream, largely informal handling, constrains what storage can achieve. Pre-storage sanitation, moisture control and moisture-barrier packaging should accompany any cold-storage option, and −18 °C appears preferable when preserving bioactive quality is the objective. Maintaining the integrity of these bioactive constituents unlocks significant prospects for incorporating dates into functional foods and pharmaceutical applications. Future research should prioritize in vivo clinical studies to evaluate the bioavailability and therapeutic efficacy of date polyphenols preserved under long term at low temperature storage.

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AUTHOR CONTRIBUTIONS

Ben Sayah Faiza: Conceptualization, Methodology, Investigation, Laboratory Analysis Writing original draft.

Boutouil Abderrahmane: Supervision, Writing, review & editing.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ETHICS STATEMENT

Ethical approval was not required for this study.

DATA AVAILABILITY STATEMENT

Data supporting the findings of this study are available from the corresponding author upon reasonable request.

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