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Bioprocessing of Rose Hip Seed By-Products and Its Effects on Phenolic Composition and Antioxidant Activity

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Abstract

Rose hip processing generates seed-rich by-products that remain underexplored beyond oil extraction, despite their potential as a source of phenolic compounds and antioxidant activity. This study investigates the effect of bioprocessing (short-term fermentation) on the phenolic composition and antioxidant activity of rose hip (*Rosa* spp.) seed by-products, with relevance to cosmetic-oriented applications related to oxidative stress modulation. Rose hip seeds were obtained after juice production and subjected to short-term fermentation (14 days at 21 °C) using *Saccharomyces cerevisiae*, followed by mechanical separation and drying. Non-fermented and bioprocessed seeds were analyzed for individual phenolic compounds and antioxidant activity (DPPH, ABTS, FRAP), and correlation and multivariate analyses were conducted. Bioprocessing reduced total identified phenolics from 15.79 to 10.72 mg/g DW (≈32%), primarily due to a decrease in epigallocatechin (10.89 to 6.50 mg/g DW). In parallel, the relative contribution of phenolic acids increased, including gallic acid (0.50 to 0.60 mg/g DW) and salicylic acid (0.98 to 1.20 mg/g DW), indicating a selective compositional redistribution accompanied by partial degradation. Antioxidant activity decreased after bioprocessing (DPPH ~340 to ~250 µmol TE/g DW) but remained substantial. Correlation analysis identified epigallocatechin as the main contributor to antioxidant capacity. These findings show that rose hip seeds behave as a process-sensitive phenolic matrix in which bioprocessing alters the balance of individual compounds without complete loss of antioxidant activity. The results indicate that seed-derived by-products retain functional potential for further valorization in cosmetic-oriented applications.

Keywords: rose hip seeds; bioprocessing; phenolic redistribution; epigallocatechin; antioxidant activity; oxidative stress; cosmeceuticals; by-product valorization

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1. Introduction

Rose hips (*Rosa* spp.) are recognized as a complex plant matrix combining hydrophilic antioxidants [1], lipophilic constituents [2], and structurally diverse phenolics [3–5]. Rose species are widely distributed in Europe, Asia, and other temperate regions, and the seed fraction represents a substantial proportion of fruit mass, which supports the relevance of rose hip seed by-products as a raw material for valorization. Industrial processing, however, remains uneven. Pulp is commonly utilized for food products, while seeds and pomace are treated as secondary residues despite accounting for a substantial fraction of the fruit

mass and retaining bioactive compounds. The distribution of these compounds is strongly tissue-dependent; therefore, rose hip side streams should be considered differentiated raw materials whose functional value depends on both tissue origin and processing history.

Research on rose hip seeds has been largely focused on oil recovery. Comparative studies of solvent and ultrasound-, microwave-, subcritical-, and supercritical-fluid extraction have shown that extraction technology influences both yield and composition of seed oil. These oils are rich in linoleic and α-linolenic acids and contain tocopherols, carotenoids, and minor phenolics, supporting their relevance for high-value applications. At the same time, variability related to species, genotype, and growing conditions has been consistently reported [6–10]. This body of work confirms the technological and compositional importance of rose hip seeds, but it also shows that most available evidence remains centered on the lipid fraction. This oil-centered approach leaves the non-lipid fraction of seeds insufficiently explored.

Recent work has demonstrated that defatted rose hip seed residues retain extractable phenolic compounds, including flavan-3-ols and phenolic acids, indicating that seed valorization should extend beyond lipid recovery [11,12]. In parallel, biological studies have suggested potential cosmetic relevance of seed-derived materials, including antioxidant and enzyme-inhibitory activities, as well as effects on skin-related processes in experimental systems. However, these reports are mainly based on seed oils or isolated extracts and do not provide direct evidence of anti-aging efficacy in skin-relevant biological models. Despite this, most studies have focused on seed oils or isolated extracts, while the behavior of seed matrices subjected to prior processing remains less well defined.

Rose hip pomace and pulp-derived fractions are well described as phenolic-rich matrices with a complex composition and high antioxidant potential. Demir et al. [13] identified catechin, gallic acid, and other phenolic compounds as dominant constituents in *Rosa* spp., directly associated with strong radical-scavenging activity. Nađpal et al. [14] reported total phenolic content exceeding 500 mg GAE/100 g FW in rose hip products, together with high antioxidant capacity across different forms, indicating that both fresh and processed materials retain substantial bioactivity. The stability of these compounds depends strongly on processing conditions. Paunović et al. [15] showed that drying leads to measurable changes in phenolic content and antioxidant activity. Pirone et al. [16] demonstrated that dehydration reduces ascorbic acid concentration, with losses dependent on temperature and processing time. These results indicate that rose hip by-products do not represent compositionally stable systems. Processing introduces systematic variation in both phenolic concentration and extractability. Direct comparison between studies is therefore limited by differences in extraction methods, drying regimes, and raw material state.

Processing history is a key factor determining the functional behavior of rose hip by-products. Technological treatments such as oil extraction, mechanical fractionation, and thermal processing modify not only the composition but also the structure of the plant matrix. These changes affect compound accessibility and chemical stability. Concha et al. [17] demonstrated that oil extraction alters the physicochemical properties of rose hip residues, including defatted meal, indicating that the resulting material differs structurally from the original matrix. Studies on dehydration and processing show that compositional changes involve selective degradation and redistribution of compounds rather than uniform losses. Bioprocessing should therefore be interpreted as a transformation step that modifies the internal phenolic balance. Its effect cannot be reduced to a simple decrease in total phenolics. Short-term fermentation was selected in the present study as a mild bioprocessing strategy because fermentation can modify plant phenolics through enzymatic and microbial activity without the intensity of thermal treatment. In particular, esterases, tannin-acyl hydrolases, and related hydrolytic activities may contribute to the release or transformation of bound

phenolic compounds, while oxidative reactions and pH-related instability may simultaneously reduce more labile flavan-3-ols. Evidence for such transformations is available for pulp- and pomace-derived fractions. Information on seed-containing materials remains limited. In particular, data remain scarce for rose hip seed by-products obtained after juice processing that are subsequently subjected to short-term fermentation prior to drying and chemical analysis. The influence of matrix modification on the relative contribution of individual phenolic groups to antioxidant activity is still insufficiently defined. This gap is particularly relevant for evaluating the functional potential of seed-derived fractions in cosmeceutical applications.

The aim of this study was therefore to evaluate rose hip seed by-products before and after bioprocessing, with particular focus on phenolic composition and antioxidant activity, in order to determine whether processing induces compositional redistribution and to assess the potential of the resulting material for cosmeceutical applications related to oxidative stress modulation.

2. Materials and Methods

2.1. Sample Preparation

Rose hip samples were processed according to a previously published methodology, adapted for the present study. Rose hip fruits (*Rosa* spp.) were obtained from Farmer in Northern Lithuania. The fruits were harvested in summer 2025 and stored at 4 °C prior to processing. The procedure included juice production, short-term primary fermentation, mechanical separation of seeds, and subsequent drying. The main steps were maintained as described in [18], with minor adjustments related to sample type.

Fermentation was carried out in food-grade fermentation vessels at 21 °C for 14 days. A commercial active dry yeast (*Saccharomyces cerevisiae* strain) Oenoferm® Universal (Erbslöh, Geisenheim, Germany) was used as the starter culture to inoculate the mash. Yeast was added according to the manufacturer's instructions (approximately 20–30 g per 100 L of mash). After inoculation, the mixture was stirred and sealed with an airlock, and every two days the fermenting mash was thoroughly mixed to ensure uniform fermentation and prevent sedimentation. Fermentation progress was monitored daily by observing CO_2 release and measuring density changes. After 14 days, the process was considered complete, indicated by cessation of CO_2 evolution and stabilization of the specific gravity.

The seeds were dried in an infrared dryer (Ukrsushka, Dnipro, Ukraine) at 35 °C until the moisture content reached 9–10%. The dried material was then stored under appropriate conditions and used for subsequent analyses.

2.2. Reagents

Analytical and HPLC-grade solvents and reagents were used for chemical analyses. Acetonitrile (99.9%), methanol (99.9%), potassium persulphate (99%), 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (98%) (ABTS), and the reference compounds were obtained from Sigma-Aldrich (Steinheim, Germany); trifluoroacetic acid (≥99%), Trolox (≥98%) and apigenin were supplied from Fluka Chemika (Buchs, Switzerland). The purified deionized water (18.2 MΩ·cm) was produced using the Millipore Simpak1 Synergy 185 ultra-pure (Bedford, MA, USA) water system.

2.3. Determination of Physical and Functional Properties

2.3.1. Moisture Content

The moisture content of rose hip seed samples was determined using a gravimetric method. Approximately 1–2 g of sample was analyzed using a moisture analyzer (Shimadzu MOC63u, Shimadzu Corporation, Tokyo, Japan) at a constant temperature until a

stable mass was reached. The moisture content was expressed as a percentage of the initial sample weight.

2.3.2. Thousand-Seed Weight

The thousand-seed weight (TSW) was determined according to the International Rules for Seed Testing. Eight replications of 100 seeds from the pure seed fraction were manually counted and weighed using an analytical balance with a precision of 0.001 g. The TSW was calculated by extrapolating the average weight of 100 seeds to one thousand seeds (TSW = mean weight of 100 seeds × 10) using Microsoft Excel 365.

2.3.3. Bulk and Tapped Density

Bulk density was determined by gently filling a graduated cylinder with a known mass of sample and recording the occupied volume. Tapped density was measured after mechanically tapping the cylinder until a constant volume was achieved. Results were expressed as g/cm^3. The Carr index (*CI*) and Hausner ratio (*HR*) were calculated to evaluate flow properties:

$$CI = \frac{\rho_t - \rho_b}{\rho_t} \times 100$$

$$HR = \frac{\rho_t}{\rho_b}$$

where ρ_b is bulk density and ρ_t is tapped density.

2.3.4. Water Holding, Oil Absorption and Swelling Capacity

Water holding capacity (WHC) was determined by mixing a known mass of sample (approximately 1 g) with distilled water and allowing it to hydrate under controlled conditions. After centrifugation, the supernatant was removed, and the retained water was quantified. Results were expressed as grams of water per gram of dry sample (g/g). Oil absorption capacity (OAC) was determined by mixing the sample with a known volume of vegetable oil, followed by centrifugation to remove unbound oil. The amount of retained oil was calculated gravimetrically and expressed as grams of oil per gram of dry sample (g/g). Swelling capacity was evaluated by measuring the increase in volume of a known quantity of sample after hydration in excess water for a fixed period. Results were expressed as mL/g of dry sample and used to assess structural changes in the seed matrix induced by processing.

The sample-to-liquid ratio was 1:10 (w/v) for all determinations. For WHC and OAC, samples were allowed to hydrate or interact with liquid for 30 min at room temperature and were then centrifuged at 3000× g for 15 min. Refined sunflower oil was used for OAC determination. For swelling capacity, samples were hydrated in excess distilled water for 18 h at room temperature before volume measurement.

2.4. Antioxidant Activity

ABTS + radical cation decolorization assay was adjusted according to the methodology described by Re and colleagues, with some modifications. A volume of 3 mL of ABTS + (2,2′-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid)) solution (absorbance 0.800 ± 0.02) was mixed with 20 µL of sample. Each sample was measured at 734 nm in a Cintra 202 (GBC Scientific Equipment, Knox, Braeside, VIC, Australia) spectrophotometer after 30 min. The ABTS working solution was prepared by mixing 7.0 mM ABTS stock solution with 2.45 mM potassium persulphate and allowing the mixture to react for 12–16 h in the dark at room temperature before use. Prior to analysis, the radical solution was diluted with ethanol to obtain an absorbance of 0.800 ± 0.02 at 734 nm.

The DPPH• free radical scavenging activity was established using the method suggested by Brand Williams, Cuvelie, and Berset [19], with some modifications: 2 mL of DPPH• (2,2-diphenyl-1-picrylhydrazyl) solution in 96.0% v/v ethanol was mixed with 20 µL of sample. A decrease in absorbance was determined at 515 nm in a Cintra 202 (GBC Scientific Equipment, Knox, Australia) spectrophotometer after 30 min. The concentration of the DPPH• solution was 0.1 mM.

The ferric-reducing antioxidant power (FRAP) assay was performed as described by Benzie and Strain, with some modifications. The FRAP solution consisted of TPTZ (0.01 M dissolved in 0.04 M HCl), $FeCl_3 \times 6H_2O$ (0.02 M in water), and acetate buffer (0.3 M, pH 3.6) at the ratio of 1:1:10. A volume of 3 mL of a recently prepared FRAP reagent was mixed with 2 µL of sample. The absorbance increase was established at 593 nm in a Cintra 202 (GBC Scientific Equipment, Knox, Australia) spectrophotometer after 30 min. Calculations for all antioxidant activity assays were carried out using Trolox calibration curves, and results were expressed as µmol of the Trolox equivalent (TE) per one gram of dry weight (µmol TE/g DW) [20]. The Trolox calibration curves were prepared in the range of 0.02–0.20 mM and showed good linearity, with $R^2 \geq 0.99$.

2.5. Identification and Quantification of Phenolic Compounds

Before HPLC analysis, phenolic compounds were extracted from the seed material using methanol/water (80:20, v/v) acidified with 0.1% trifluoroacetic acid. Ground seed sample (1.0 g) was mixed with 10 mL of extraction solvent and sonicated for 30 min at room temperature. The mixtures were then centrifuged at 5000× g for 10 min, and the supernatants were collected. The extraction was repeated twice under the same conditions, and the combined extracts were adjusted to a final volume of 20 mL. Prior to HPLC analysis, the extracts were filtered through 0.22 µm PVDF membrane filters.

Phenolic compounds were identified and quantified using high-performance liquid chromatography (HPLC) (Shimadzu, Kyoto, Japan). Chromatographic separation was carried out on a reversed-phase C18 column (250 × 4.6 mm, 5 µm particle size).

The mobile phase consisted of acidified water (0.1% formic acid, solvent A) and acetonitrile (solvent B), delivered under a gradient elution program optimized for the separation of phenolic acids, flavonoids, and flavan-3-ols. The flow rate was maintained at 1.0 mL/min, and the total run time was 60 min per sample. Detection was performed using a UV–Vis diode-array detector (DAD; Shimadzu Corporation, Kyoto, Japan), continuously recording absorbance spectra over a wavelength range suitable for phenolic compounds. Quantification was based on characteristic wavelengths: 280 nm for flavan-3-ols, 320 nm for phenolic acids, and 360 nm for flavonols. Identification of individual compounds was achieved by comparing retention times and UV spectral characteristics with those of authenticated reference standards. Quantification was carried out using external calibration curves prepared from serial dilutions of standard compounds. Results were expressed as mg per g of dry weight (mg/g DW). Representative chromatograms are provided in the Supplementary Materials.

2.6. Statistical Analysis

All experiments were performed in triplicate (n = 3), and results are expressed as mean ± standard deviation. Statistical analysis was carried out using Microsoft Excel 365 (Microsoft Corporation, Redmond, WA, USA) and IBM SPSS Statistics 26 (IBM Corp., Armonk, NY, USA). Differences between samples were evaluated using one-way analysis of variance (ANOVA), followed by Tukey's HSD post hoc test. Differences were considered statistically significant at $p < 0.05$. Normality and homogeneity of

variance were assessed using the Shapiro–Wilk test and Levene's test, respectively, prior to performing one-way ANOVA.

Pearson correlation analysis was performed using mean values of individual phenolic compounds (mg/g DW) and antioxidant assays (DPPH, ABTS, FRAP; µmol TE/g DW). For multivariate analysis, data were standardized using Z-score normalization prior to analysis. Principal component analysis (PCA) was applied to the normalized dataset to assess sample clustering and identify the main variables contributing to variation among non-fermented and bioprocessed seed and skin fractions. The first two principal components (PC1 and PC2) were used for visualization of sample distribution (score plot), while variable loadings were used to interpret the contribution of phenolic compounds and antioxidant activity to sample differentiation. All multivariate analyses and graphical representations were performed using Python 3.11 (Python Software Foundation, Wilmington, DE, USA) with the libraries scikit-learn 1.4, pandas 2.2, and matplotlib 3.8.

3. Results and Analysis

It is important to characterize the physical and functional behavior of the raw material, since these parameters determine its suitability for downstream processing, extraction efficiency, storage stability, and potential incorporation into cosmetic-oriented formulations. In the context of bioprocessing, the assessment of seed morphology, density-related parameters, hydration behavior, and color provides useful information on whether short-term processing causes major structural deterioration or only mild matrix modification. Table 1 below summarizes the physical and functional properties of non-fermented and bioprocessed rose hip seeds.

Table 1. Physical and functional properties of non-fermented and bioprocessed rose hip seeds.

Parameter	Seeds (NF)	Seeds (F)	Unit
Thousand-seed weight (TSW)	11.73 [a]	11.60 [a]	g
Length	3.2 ± 0.4 [a]	3.2 ± 0.4 [a]	mm
Width	1.8 ± 0.2 [a]	1.8 ± 0.2 [a]	mm
Thickness	1.2 ± 0.2 [a]	1.2 ± 0.2 [a]	mm
Shape index (L/W)	1.78 [a]	1.78 [a]	–
Moisture content	7.5 ± 0.3 [a]	7.8 ± 0.3 [a]	%
Bulk density	0.42 ± 0.02 [a]	0.41 ± 0.02 [a]	g/cm^3
Tapped density	0.51 ± 0.02 [a]	0.50 ± 0.02 [a]	g/cm^3
Carr index	17.6 [a]	18.0 [a]	%
Hausner ratio	1.21 [a]	1.22 [a]	–
Water holding capacity (WHC)	2.1 ± 0.1 [b]	2.3 ± 0.1 [a]	g/g
Oil absorption capacity (OAC)	1.6 ± 0.1 [a]	1.5 ± 0.1 [a]	g/g
Swelling capacity	2.8 ± 0.2 [b]	3.0 ± 0.2 [a]	mL/g
Color (L*)	68.5 ± 1.2 [a]	64.2 ± 1.5 [b]	–
Color (a*)	5.4 ± 0.3 [b]	6.2 ± 0.3 [a]	–
Color (b*)	18.2 ± 0.6 [a]	17.0 ± 0.5 [b]	–

Note: Values are presented as mean ± standard deviation (n = 3), except for thousand-seed weight, shape index, Carr index, and Hausner ratio, which are presented as mean values. Different lowercase letters within the same row indicate statistically significant differences between non-fermented (NF) and bioprocessed (F) seeds according to one-way ANOVA followed by Tukey's HSD test ($p < 0.05$). NF, non-fermented seeds; F, bioprocessed seeds. L*, a*, and b* are CIELAB color parameters: L* = lightness, a* = green-red coordinate, b* = blue-yellow coordinate.

The data show that short-term bioprocessing had little effect on seed morphology and bulk behavior. Thousand-seed weight decreased only from 11.73 to 11.60 g, while length, width, thickness, and shape index remained unchanged. This indicates that the treatment did not induce measurable structural damage at the whole-seed level. Such stability is consistent with the general technological behavior of rose hip seeds reported

in processing studies, where the seed fraction is usually described as sufficiently robust to withstand drying, milling, and extraction operations without loss of physical identity; for example, Szentmihályi et al. processed air-dried rose hip seeds as an industrial by-product for comparative oil extraction, implicitly confirming their structural stability during technological handling.

The most meaningful changes were observed not in size descriptors but in hydration- and surface-related properties. Water holding capacity increased from 2.1 to 2.3 g/g and swelling capacity from 2.8 to 3.0 mL/g, whereas oil absorption capacity decreased slightly from 1.6 to 1.5 g/g. This pattern suggests a mild increase in matrix openness or hydrophilic accessibility after bioprocessing rather than extensive degradation. A useful comparison can be made with rosehip waste powder incorporated into waffle cones: some researchers reported water-holding values of about 2.64–3.08 g/g and swelling values of 4.23–4.48 mL/g for rosehip-enriched systems [21–24], while oil-holding capacity ranged from 0.96 to 1.19 g/g. Although those were food matrices rather than isolated seeds, the comparison is informative: our seeds showed comparable but slightly lower hydration capacity and higher oil absorption, which is reasonable for a denser, less fragmented seed-based material.

Color was the parameter most visibly affected by bioprocessing. The decrease in L* from 68.5 to 64.2, together with the increase in a^* and the decrease in b^*, indicates slight darkening and a shift toward a less yellow, more brown-red appearance. This is in line with observations from rosehip powder studies, where processing and formulation changes led to darker yellow-brown products and were associated with matrix composition and processing history. Michalska-Ciechanowska et al. showed that rosehip juice- and pomace-derived powders differed significantly in color, and that drying technology influenced moisture and water-related functionality at the same time [22]. Similarly, rosehip-enriched extrudates became more red as the proportion of rose hip material increased, confirming that color parameters in rose hip systems are sensitive markers of technological modification even when the matrix remains physically intact. In this context, the color shift observed here is best interpreted as evidence of surface-level biochemical change, not as proof of major seed deterioration.

A further point relevant to valorization is that the measured moisture content remained below 8% in both groups. This places the material in a range compatible with stable storage and further processing, and the slight increase observed after bioprocessing does not appear technologically critical. Instead, when considered together with the hydration-related parameters, the results suggest that the treatment acted as a mild conditioning step, preserving the processability of the seeds while slightly modifying their interaction with water. If statistical significance was confirmed, this should be indicated directly in the corresponding table using letter superscripts or another clearly defined notation.

An additional practical aspect of utilizing rose hip seeds is their distinctive outer surface morphology. The seeds are covered with fine hair-like appendages that appear visually soft, yet direct handling revealed a pronounced and persistent prickling sensation on the skin. This observation is relevant for cosmetic applications, because it indicates that intact seeds are not suitable for direct incorporation into topical systems. From an application standpoint, only sufficiently milled and homogenized seed-derived material could be considered for further formulation studies. Whether fine grinding alone is sufficient to eliminate residual mechanical skin irritation remains unclear and should be verified experimentally in future formulation and skin compatibility studies. Antioxidant activity of non-fermented and bioprocessed rose hip seed and skin fractions is presented in the Figure 1 below.

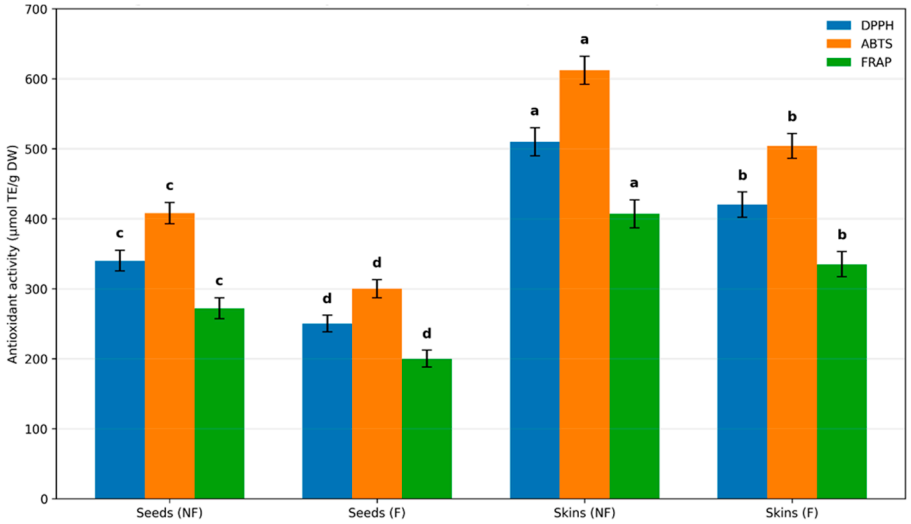


Figure 1. Antioxidant activity of non-fermented (NF) and bioprocessed (F) rose hip seed and skin fractions measured by DPPH, ABTS, and FRAP assays. Results are expressed as mean ± standard deviation (n = 3) in µmol Trolox equivalents per g dry weight (µmol TE/g DW). Different lowercase letters above bars indicate statistically significant differences among sample groups within each assay (one-way ANOVA, Tukey's HSD, $p < 0.05$).

The antioxidant data show two consistent effects. First, the skin fraction was more active than the seed fraction in all three assays, irrespective of processing. Second, bioprocessing reduced antioxidant activity, but the decrease was moderate and did not eliminate the functional advantage of the skin fraction. In the present study, non-fermented skins showed the highest activity, reaching approximately 510, 610, and 407 µmol TE/g DW in DPPH, ABTS, and FRAP, respectively, whereas non-fermented seeds reached about 340, 408, and 272 µmol TE/g DW. After bioprocessing, antioxidant activity declined in both fractions, but the reduction was proportionally stronger in seeds than in skins.

This pattern agrees well with published rosehip studies showing that the non-seed fraction generally contributes more strongly to antioxidant performance than seed-containing material. Some studies reported that tinctures prepared from rose hips without seeds had higher antioxidant activity and higher extractability of phenolics than seed-containing preparations, particularly in ABTS and FRAP assays [23–28]. In addition, pomace-based rose hip preparations have been reported to retain substantial ABTS scavenging capacity, confirming that solid by-product fractions can preserve relevant antioxidant potential after processing.

The assay ranking observed here, ABTS > DPPH > FRAP, is also plausible for rosehip-derived materials. In rosehip juice powders, ABTS and FRAP were both strongly associated with phenolic content, but ABTS tended to respond particularly well to hydrophilic antioxidant constituents. Rosehip pulp extracts have been described as rich in phenolics such as gallic acid, salicylic acid, catechin, and procyanidins, which are major contributors to radical-scavenging activity. This is consistent with the stronger activity of the skin fraction in the present study, since the present compositional data also indicate that skins retained a more concentrated phenolic profile than seeds.

Since oxidative stress is one of the main mechanisms involved in skin aging, the retention of antioxidant capacity after bioprocessing supports the feasibility of using rose hip seed-derived material as a source of bioactive ingredients for cosmetic or cosmeceutical formulations related to oxidative stress modulation. At the same time, the consistently higher activity of skins suggests that seed-focused valorization may be further strengthened by optimized extraction or by combining seed fractions with other rose hip side streams.

However, antioxidant assays alone do not provide direct evidence of anti-aging effects in skin models, and the results should therefore be interpreted as indicating functional potential rather than confirmed topical efficacy. Table 2 shows that bioprocessing reduced the phenolic content of rose hip by-products and selectively altered the composition of the seed fraction.

Table 2. Phenolic composition (mg/g DW) of non-fermented and bioprocessed rose hip seed and skin fractions.

Compound	Seeds (NF) (mg/g DW)	Seeds (F) (mg/g DW)	Skins (NF) (mg/g DW)
Gallic acid	0.50 ± 0.01 ^{c}	0.60 ± 0.02 ^{b}	14.43 ± 0.50 ^{a}
Epigallocatechin	10.89 ± 0.51 ^{b}	6.50 ± 0.30 ^{c}	20.35 ± 0.80 ^{a}
Catechin	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
p-Hydroxybenzoic acid	0.04 ± 0.00 ^{b}	0.06 ± 0.00 ^{a}	0.03 ± 0.00 ^{c}
Chlorogenic acid	0.00 ± 0.00 ^{b}	0.05 ± 0.00 ^{a}	0.00 ± 0.00 ^{b}
Caffeic acid	0.06 ± 0.00 ^{b}	0.07 ± 0.00 ^{a}	0.05 ± 0.00 ^{c}
p-Coumaric acid	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
Benzoic acid	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
Ferulic acid	0.51 ± 0.01 ^{a}	0.40 ± 0.01 ^{b}	0.32 ± 0.01 ^{c}
Quercetin-3-O-rutinoside	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
Naringin	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
Vanillic acid	0.05 ± 0.00 ^{b}	0.07 ± 0.00 ^{a}	0.04 ± 0.00 ^{c}
Salicylic acid	0.98 ± 0.03 ^{b}	1.20 ± 0.04 ^{a}	0.62 ± 0.02 ^{c}
Sinapic acid	0.19 ± 0.01 ^{a}	0.10 ± 0.00 ^{b}	0.08 ± 0.00 ^{c}
Ellagic acid	1.03 ± 0.02 ^{b}	0.80 ± 0.02 ^{c}	1.45 ± 0.05 ^{a}
Kaempferol-3-*O*-glucoside	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
Myricetin	0.06 ± 0.00 ^{a}	0.04 ± 0.00 ^{b}	0.05 ± 0.00 ^{ab}
Valeric acid	0.16 ± 0.00 ^{a}	0.12 ± 0.00 ^{b}	0.09 ± 0.00 ^{c}
trans-Cinnamic acid	0.24 ± 0.01 ^{a}	0.15 ± 0.01 ^{b}	0.11 ± 0.00 ^{c}
Apigenin	0.05 ± 0.00 ^{a}	0.03 ± 0.00 ^{b}	0.04 ± 0.00 ^{ab}
Quercetin	0.13 ± 0.01 ^{a}	0.08 ± 0.00 ^{b}	0.10 ± 0.00 ^{ab}
Kaempferol	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}	0.00 ± 0.00 ^{a}
Quercetin-3-*O*-glucoside	0.60 ± 0.05 ^{a}	0.45 ± 0.03 ^{b}	0.52 ± 0.04 ^{ab}

Note: Values are presented as mean ± standard deviation and expressed as mg/g DW. NF, non-fermented; F, bioprocessed. Different lowercase letters within the same row indicate statistically significant differences among sample types according to one-way ANOVA followed by Tukey's HSD test ($p < 0.05$).

The total amount of identified phenolics clearly separated the fractions. Non-fermented skins contained 37.68 mg/g DW, whereas non-fermented seeds contained 15.79 mg/g DW. Thus, the skin fraction contained about 2.4-fold more identified phenolics than the seed fraction. This agrees with the tissue-specific distribution reported by Kunc et al., who found substantially higher total phenolic levels in rose hip flesh with skin than in seeds across *Rosa* genotypes, confirming that the non-seed fraction is the principal phenolic reservoir of the fruit.

At the same time, the seed fraction was chemically distinct rather than merely diluted. In non-fermented seeds, epigallocatechin reached 10.89 mg/g DW, accounting for the dominant share of the identified phenolics, whereas ellagic acid, salicylic acid, quercetin-3-*O*-glucoside, ferulic acid, and gallic acid were present at 1.03, 0.98, 0.60, 0.51, and 0.50 mg/g DW, respectively. This profile indicates that the seed fraction was strongly flavan-3-ol-centered. By contrast, non-fermented skins combined 20.35 mg/g DW epigallocatechin with a markedly higher gallic acid concentration of 14.43 mg/g DW and 1.45 mg/g DW ellagic acid, showing that the skin fraction was not only richer in total phenolics, but also broader in compositional balance.

The strongest evidence of process-induced change was observed in seeds after bioprocessing. The total identified phenolics decreased from 15.79 to 10.72 mg/g DW, corresponding to a reduction of about 32%. However, the decrease was not uniform across compounds. Epigallocatechin declined from 10.89 to 6.50 mg/g DW, which represents a reduction of approximately 40% and explains most of the total loss. Ellagic acid decreased from 1.03 to 0.80 mg/g DW, sinapic acid from 0.19 to 0.10 mg/g DW, trans-cinnamic acid from 0.24 to 0.15 mg/g DW, and quercetin from 0.13 to 0.08 mg/g DW. In contrast, gallic acid increased from 0.50 to 0.60 mg/g DW, and salicylic acid from 0.98 to 1.20 mg/g DW. These data indicate that bioprocessing reduced the phenolic sum, but at the same time shifted the seed profile away from strong epigallocatechin dominance toward a relatively greater contribution of simpler phenolic acids.

This pattern is best interpreted as redistribution accompanied by partial degradation, rather than redistribution alone. The increase in gallic acid may reflect hydrolytic cleavage of ester-linked gallate or ellagitannin-related structures, whereas the decline in epigallocatechin and several other phenolics may be associated with oxidative conversion, polymerization, or structural instability during fermentation and subsequent drying. Microbial esterases, tannin-acyl hydrolases, and pH-dependent transformations are plausible contributors to this shift, although the present study was not designed to identify the exact reaction pathways. Thus, the observed compositional change likely reflects both selective release of low-molecular-weight phenolic acids and loss of more labile flavan-3-ol constituents. Drevelegka et al. reported that defatted rosehip seed waste still contained an extractable phenolic pool, with quinic acid and catechin identified among dominant constituents after deep eutectic solvent extraction. Our seed profile differs substantially, because epigallocatechin rather than catechin dominated the non-fermented seeds, while quinic acid was not among the quantified analytes. This difference most likely reflects a strong dependence of seed phenolic composition on pretreatment and extraction strategy. Therefore, the present data do not contradict previous studies; rather, they show that rose hip seed phenolics are highly process-sensitive and that bioprocessing produces a distinct compositional state that has not been sufficiently described in the literature.

The skin data are also consistent with previous reports on rose hip flesh- and pomace-rich matrices. Teleszko et al. showed that rose hip tinctures prepared without seeds had higher phenolic extractability and stronger antioxidant performance than seed-containing preparations, especially in ABTS and FRAP systems. Some studies demonstrated that rosehip pomace-based materials retained substantial antioxidant potential after processing [29–31]. In this context, the high phenolic density observed in the skin fraction in the present study is expected. Compared with seeds, skins behaved as the quantitatively dominant fraction, whereas seeds provided the more informative model for evaluating process-related compositional shifts.

This redistribution is particularly relevant for the chosen manuscript focus. For cosmetic-oriented applications, the value of the seed fraction is not determined only by total phenolics, but by which compounds remain or become relatively enriched after processing. After bioprocessing, the seed fraction still retained 6.50 mg/g DW epigallocatechin, 1.20 mg/g DW salicylic acid, 0.80 mg/g DW ellagic acid, and 0.60 mg/g DW gallic acid-equivalent acidic enrichment, indicating that the material remained phenolically active despite compositional simplification. The seed fraction therefore should not be interpreted as degraded waste but as a chemically redirected by-product fraction whose phenolic balance was altered by bioprocessing in a potentially useful way. To identify which phenolic compounds contributed most strongly to antioxidant activity, correlation analysis was performed between individual phenolics and DPPH, ABTS, and FRAP values, and results are shown in Table 3 below.

Table 3. Pearson correlation coefficients between phenolic compounds and antioxidant activity.

Variables	DPPH	ABTS	FRAP	Sum Phenolics	Epigallocatechin	Ellagic Acid	Gallic Acid	Salicylic Acid
DPPH	1.00	0.91 **	0.88 **	0.82 **	0.88 **	0.74 **	0.65 *	0.61 *
ABTS	0.91 **	1.00	0.89 **	0.85 **	0.86 **	0.78 **	0.69 **	0.66 *
FRAP	0.88 **	0.89 **	1.00	0.80 **	0.84 **	0.72 **	0.67 *	0.63 *
Sum phenolics	0.82 **	0.85 **	0.80 **	1.00	0.79 **	0.85 **	0.70 **	0.66 *
Epigallocatechin	0.88 **	0.86 **	0.84 **	0.79 **	1.00	0.68 *	0.52	0.48
Ellagic acid	0.74 **	0.78 **	0.72 **	0.85 **	0.68 *	1.00	0.63 *	0.55
Gallic acid	0.65 *	0.69 **	0.67 *	0.70 **	0.52	0.63 *	1.00	0.77 **
Salicylic acid	0.61 *	0.66 *	0.63 *	0.66 *	0.48	0.55	0.77 **	1.00

Note: Pearson correlation coefficients were calculated using mean values of phenolic compounds (mg/g DW) and antioxidant activity assays (DPPH, ABTS, FRAP; µmol TE/g DW); * $p < 0.05$, ** $p < 0.01$.

Strong positive correlations were observed between epigallocatechin and antioxidant activity, particularly with DPPH and ABTS, confirming its dominant role in radical scavenging within the seed fraction. This is consistent with its high concentration in non-fermented seeds (10.89 mg/g DW) and its marked decrease after bioprocessing (6.50 mg/g DW), which coincided with the reduction in antioxidant activity. Gallic acid and salicylic acid showed weaker but consistent correlations with antioxidant assays, despite their increase after bioprocessing (gallic acid: 0.50 to 0.60 mg/g DW; salicylic acid: 0.98 to 1.20 mg/g DW). This indicates that the enrichment of these acids did not compensate for the loss of epigallocatechin in terms of total antioxidant capacity. A similar relationship between flavan-3-ols and antioxidant activity has been reported in rose hip extracts, where catechin-type compounds were strongly associated with radical scavenging capacity, and simpler phenolic acids contributed less to overall activity. Principal component analysis (PCA) score and loading biplot of non-fermented and bioprocessed rose hip seed and skin fractions based on standardized phenolic composition and antioxidant activity data are presented in the Figure 2 below.

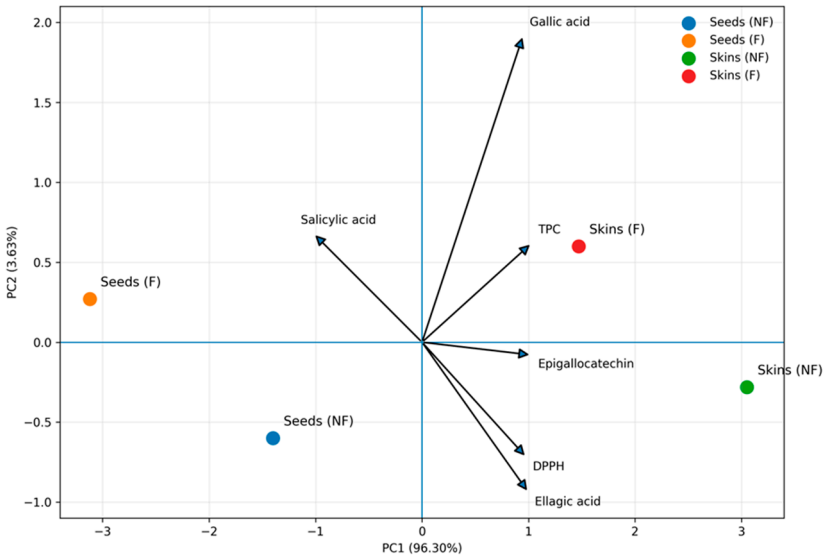


Figure 2. Principal component analysis (PCA) score and loading biplot of non-fermented and bioprocessed rose hip seed and skin fractions based on standardized phenolic composition and antioxidant activity data. PC1 explained 96.30% of the total variance, while PC2 explained 3.63%. Sample points represent non-fermented and bioprocessed seed and skin fractions, and arrows indicate the contribution of selected variables to sample differentiation. Data were standardized using Z-score normalization prior to PCA. NF, non-fermented; F, bioprocessed; TPC, total phenolic content.

The first two principal components explained the majority of variance, with PC1 primarily associated with antioxidant activity, total phenolics, and epigallocatechin and PC2 reflecting variation in phenolic acids, particularly gallic and salicylic acid. Samples were clearly separated along PC1, where skin fractions clustered at high PC1 values, reflecting their higher phenolic content (37.68 mg/g DW) and stronger antioxidant activity (DPPH ≈ 510 µmol TE/g DW), while seed fractions were located at lower PC1 values, consistent with their lower phenolic density (15.79 mg/g DW in NF seeds). Bioprocessing effects were captured along PC2. Bioprocessed seeds were shifted relative to non-fermented seeds, reflecting the decrease in epigallocatechin (10.89 to 6.50 mg/g DW) and the relative increase in gallic and salicylic acids (0.50 to 0.60 mg/g DW and 0.98 to 1.20 mg/g DW, respectively). In contrast, the position of skin samples changed less markedly, indicating that bioprocessing had a stronger compositional impact on seeds than on skins. This confirms that sample type (seed vs. skin) determines the overall level of bioactivity, while bioprocessing primarily modifies the internal phenolic balance of the seed fraction.

The experimental results can be interpreted in the context of oxidative stress-related mechanisms relevant to skin aging. Non-fermented seeds exhibited higher antioxidant activity, primarily associated with epigallocatechin (10.89 mg/g DW), indicating a stronger capacity to neutralize reactive oxygen species (ROS). Reduction in ROS is associated with decreased formation of oxidative stress markers such as malondialdehyde (MDA) and hydrogen peroxide (H_2O_2), which reflect lipid peroxidation and cellular oxidative damage [32,33].

Bioprocessing resulted in a decrease in antioxidant activity, corresponding to the reduction in epigallocatechin (6.50 mg/g DW), accompanied by an increase in gallic and salicylic acids. This change reflects a shift in phenolic composition rather than a complete loss of bioactivity. The lower antioxidant capacity indicates reduced efficiency in controlling ROS levels and, consequently, a weaker impact on oxidative stress markers such as MDA. Oxidative stress is a key factor in skin aging, contributing to damage of lipids, proteins, and nucleic acids, and is associated with collagen degradation and structural alterations in skin tissue [34,35]. Higher antioxidant activity in non-fermented seeds therefore indicates a greater potential to limit oxidative damage, whereas bioprocessed seeds represent a modified phenolic system with a retained but reduced capacity to influence these pathways.

From an application perspective, rose hip seed fractions can be considered as functional ingredients in cosmeceutical formulations such as creams, serums, or emulsions targeting oxidative stress-related skin aging. Non-fermented seeds, due to their higher epigallocatechin content and antioxidant activity, are more suitable for applications focused on reducing oxidative damage and protecting skin components. Bioprocessed seeds, characterized by a shifted phenolic profile, may provide complementary effects through phenolic acids and can be used in formulations requiring a milder but still bioactive antioxidant contribution.

4. Conclusions

Rose hip seeds were shown to be a structurally stable by-product fraction suitable for further processing. Short-term bioprocessing did not significantly affect seed morphology, density, or dimensional parameters, indicating that the material remains technically applicable for downstream use. Bioprocessing induced a selective change in phenolic composition of the seed fraction. The total identified phenolics decreased from 15.79 to 10.72 mg/g DW, mainly due to a reduction in epigallocatechin from 10.89 to 6.50 mg/g DW. At the same time, gallic acid increased from 0.50 to 0.60 mg/g DW and salicylic acid from 0.98 to 1.20 mg/g DW, indicating a shift toward a relatively higher contribution of low-molecular-weight phenolic acids.

Antioxidant activity decreased after bioprocessing but remained measurable across all assays. Correlation analysis showed that antioxidant capacity was primarily associated with epigallocatechin, while PCA demonstrated that sample type determined overall activity levels and that bioprocessing mainly affected the internal phenolic distribution of the seed fraction.

The results indicate that rose hip seeds function as a process-sensitive phenolic matrix, in which composition can be altered without loss of structural integrity. The retained antioxidant activity and the presence of phenolic acids indicate that this fraction has potential for further valorization in cosmetic-oriented applications related to oxidative stress modulation. However, the present study is limited to chemical and functional characterization, and antioxidant assays alone do not provide direct evidence of anti-aging effects or topical efficacy in skin systems. Further work should address extraction efficiency, formulation strategies, and evaluation of skin compatibility of milled seed-derived materials. Additional biological studies are also needed before any anti-aging or cosmeceutical efficacy claims can be made.

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