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THE CONTENT OF WATER-SOLUBLE VITAMINS IN SUNFLOWER HUSKS AND THEIR APPLICATION POSSIBILITIES IN THE COMPLEX BIOCONVERSION FOR FOOD AND FEED PREPARATIONS

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Abstract
This study investigates the enrichment of sunflower husks with water-soluble vitamins and assesses their potential applications in the production of functional food and feed supplements. Using High-Performance Liquid Chromatography (HPLC) analysis, sunflower husks were identified as a valuable source of B-group vitamins and ascorbic acid. The findings highlight the high biological activity of these compounds and their suitability for bioconversion processes aimed at enhancing the nutritional value of various food and feed products. Moreover, the research emphasizes the economic and ecological advantages of utilizing sunflower husk-derived bioactive compounds, promoting sustainable resource use and reducing agricultural waste. This work provides a scientific basis for further biotechnological development of plant-derived vitamin complexes for industrial applications.
Keywords: sunflower husks, water-soluble vitamins, High-Performance Liquid Chromatography (HPLC), feed supplements, bioconversion, bioactive compounds, functional food, sustainable utilization.

Introduction

In recent years, there has been a growing focus in the food industry on the recovery of high-value bioactive compounds from agricultural by-products, aiming to promote sustainability and economic efficiency. Among these by-products, sunflower husks — a residue from sunflower oil production — represent a promising raw material for the extraction of valuable bioactive substances, particularly water-soluble vitamins. Studies have shown that plant-based residues are often rich in vitamins, minerals, and antioxidants, making them suitable candidates for biotechnological valorization [1].

The chemical composition of sunflower husks includes a significant amount of cellulose, hemicellulose, lignin, and various phytochemicals such as B-group vitamins and ascorbic acid. This composition makes them highly suitable for complex bioconversion processes, which can convert these residues into functional food ingredients and feed supplements [2]. The development and application of such bioconversion technologies not only contribute to waste minimization but also promote a circular economy approach, where agricultural waste streams are efficiently utilized [3].

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Furthermore, utilizing sunflower husk-derived bioactive compounds aligns with global strategies aimed at sustainable development and reducing the environmental footprint of agro-industrial production. Research indicates that valorizing agricultural residues helps decrease greenhouse gas emissions, conserve natural resources, and generate added economic value from what would otherwise be waste [4]. Given the growing need for sustainable and functional food sources, advancing the use of sunflower husks through innovative biotechnological approaches will be critical in supporting future food security and environmental sustainability.

Materials and methods

Reagents and Equipment. Vitamin standards were sourced from reputable suppliers: Vitamin B12 from *Rhydburg Pharmaceuticals* (Germany), Vitamin C from *Carl Roth GmbH* (Germany), Vitamin B9 from *DSM Nutritional Products GmbH* (Germany), and Vitamins B1, B2, B3, B6, and PP from *BLDPharm* (China). For chromatographic analysis, HPLC-grade reagents including water, acetonitrile, glacial acetic acid, and sodium hydroxide were used.

The quantification of water-soluble vitamins in the sunflower husk samples was carried out using a high-performance liquid chromatography (HPLC) system — *LC-40 Nexera Lite* — manufactured by Shimadzu Corporation (Japan) [5].

Sample Preparation and Analysis. Sunflower husk samples were first ground to a uniform particle size. A known mass of each sample was extracted with an aqueous solvent system under optimized conditions to maximize the recovery of water-soluble vitamins. The extracts were then filtered through a 0.45 µm membrane filter before injection into the HPLC system. Chromatographic separation was performed on a reverse-phase column under gradient elution conditions, and vitamin quantification was achieved by comparing retention times and peak areas with those of authenticated standards.

All experiments were conducted in triplicate to ensure the reproducibility and reliability of the data. Calibration curves for each vitamin were generated using external standards, and the concentrations in the samples were calculated based on these calibration curves.

Preparation of Standard Solutions. Standard solutions of vitamins C (CAS 50-81-7), B1 (CAS 59-43-8), B6 (CAS 58-56-0), B3 (CAS 59-67-6), B12 (CAS 68-19-9), and PP (CAS 98-92-0) were prepared by dissolving 5 mg of each vitamin in 50 mL of 0.1 N hydrochloric acid (HCl) solution to achieve a concentration of 100 mg/L. For vitamins B2 (CAS 83-88-5) and B9 (CAS 59-30-3), standard solutions were prepared by dissolving 5 mg of each vitamin in 50 mL of 0.025% sodium hydroxide (NaOH) solution [6].

Subsequently, 200 µL aliquots of each B-group vitamin standard solution (B1, B6, B3, B12, PP) were mixed to create a composite solution with an individual vitamin concentration of 14.286 mg/L. From this stock solution, further dilutions were made to obtain standard solutions with concentrations of 7.143 mg/L, 3.571 mg/L, and 1.786 mg/L. Similarly, for Vitamin C, standard solutions with concentrations of 286 mg/L, 143 mg/L, 71.5 mg/L, and 57.2 mg/L were prepared. Distilled water was used as the blank (0 mg/L) for calibration curve generation [6].

Preparation of Sample Extracts. For the extraction of water-soluble vitamins, 1.0 g of the homogenized sunflower husk sample was accurately weighed into a 50 mL conical flask, and 25 mL of 0.1 N HCl solution was added. The mixture was subjected to ultrasonic extraction in a GT SONIC-D3 ultrasonic bath (China) at 60°C for 20 minutes [7]. After extraction, the mixture was cooled to room temperature, filtered, and the filtrate was brought to a final volume

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of 25 mL with distilled water in a volumetric flask. An aliquot of 1.5 mL of the extract was filtered through a 0.22 µm syringe filter into a HPLC vial and used for analysis.

Chromatographic Conditions. The determination of vitamins in both standard solutions and sample extracts was performed using an LC-40 Nexera Lite HPLC system, comprising the LC-40D pump, SIL-40 autosampler, and SPD-M40 photodiode array detector (PDA), with data acquisition via LabSolutions software version 6.92 (Shimadzu Corporation, Japan) [5]. Chromatographic separation was achieved on a reversed-phase Shim-pack GIST C18 column (150 × 4.6 mm, 5 µm particle size).

The mobile phase consisted of a gradient mixture of acetonitrile (solvent A) and 0.25% aqueous acetic acid solution (solvent B), detailed in Table 1. The injection volume was set to 10 µL, the flow rate to 0.6 mL/min, and the column oven temperature to 40°C.

Detection of the vitamins was carried out at three specific wavelengths: 265 nm, 291 nm, and 550 nm, according to their maximum absorbance profiles (Figures 1–3). For Vitamin C analysis, a 15-minute gradient program was applied (Table 2), with absorbance monitored at 265 nm [8].

Table 1. Mobile phase gradient program in the determination of vitamins.

Time, minute	Acetonitrile (A), %	0.5% acetic acid (B), %
0	0	100
3	0	100
14	20	80
17	50	50
18	0	100
25	Finish	

Table 2. Mobile phase gradient program for vitamin C quantification.

Time, minute	Acetonitrile (A), %	0.5% acetic acid (B), %
0	0	100
2	0	100
6	50	50
6,01	0	100
15	Finish	

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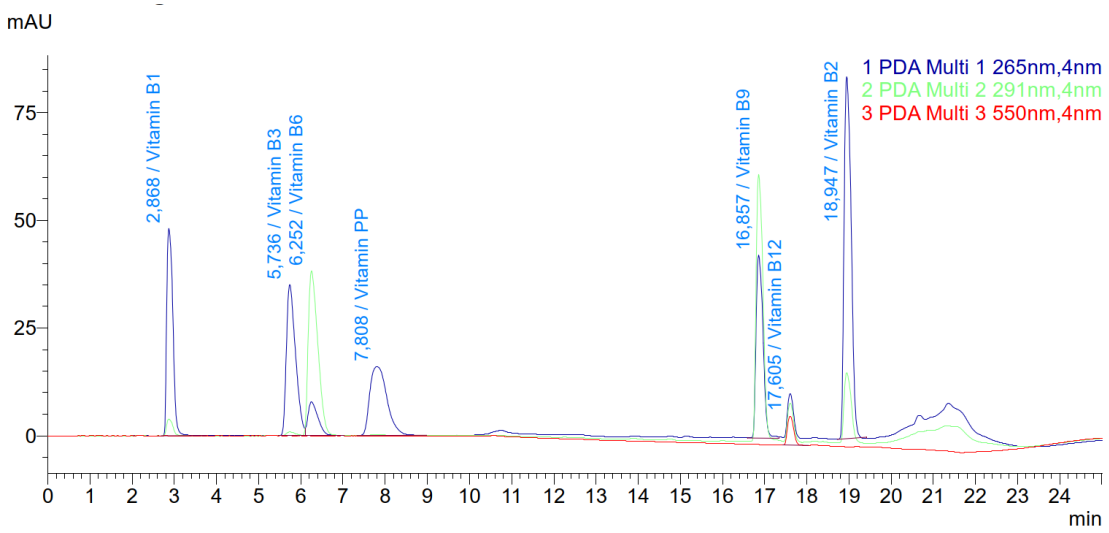

Figure 1. Chromatogram of a standard solution of vitamins.

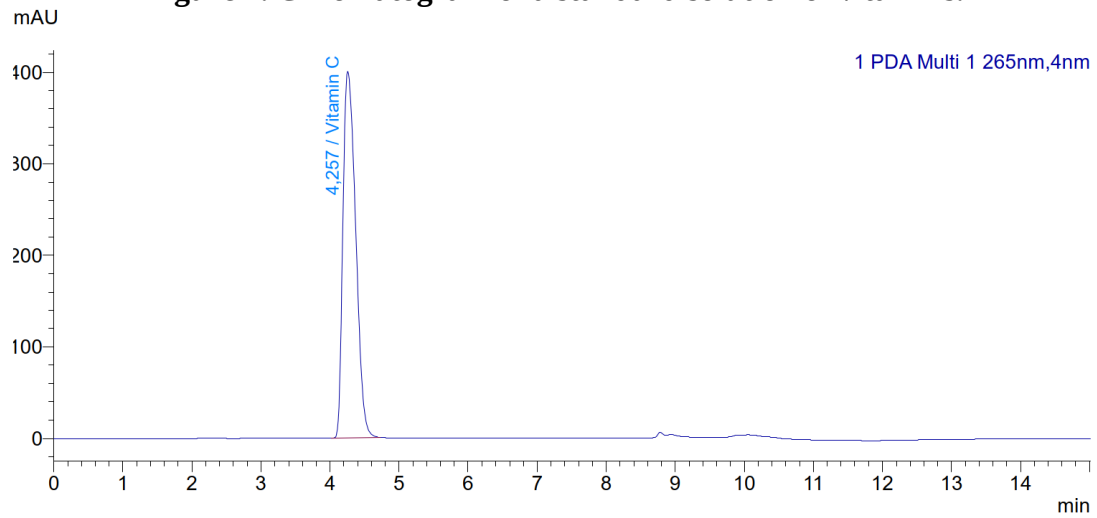

Figure 2. Chromatogram of a vitamin C standard solution.

Results

Determination of vitamins in the sample extract. A chromatogram of the sample extract (Figures 3-4) was obtained and based on the results, the amounts of vitamins in 100 g of the sample were calculated using the following formula and presented in Table 3.

$$X = \frac{C_{vit} \cdot V_{extract}}{m_{sample}} \cdot 100\ g$$

Here,

X – the amount of vitamins in 100 grams of the fruit sample, expressed in mg;

C_{vit} – the concentration of the vitamin in the extract determined by the HPLC method, expressed in mg/L;

$V_{extract}$ – the volume of the sample extract, expressed in liters (L);

$m_{samplem}$ – the mass of the sample taken for extraction, expressed in grams (g).

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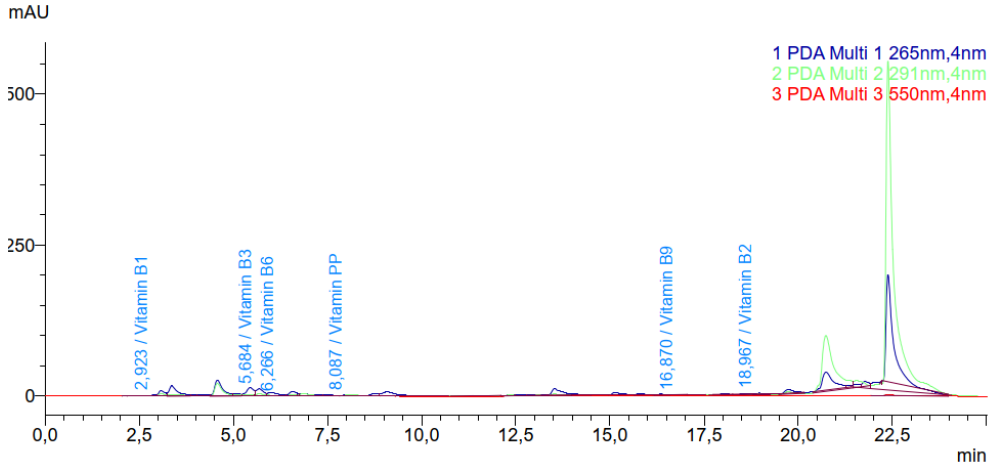

Figure 3. Chromatogram of the determination of vitamins in the sample extract.

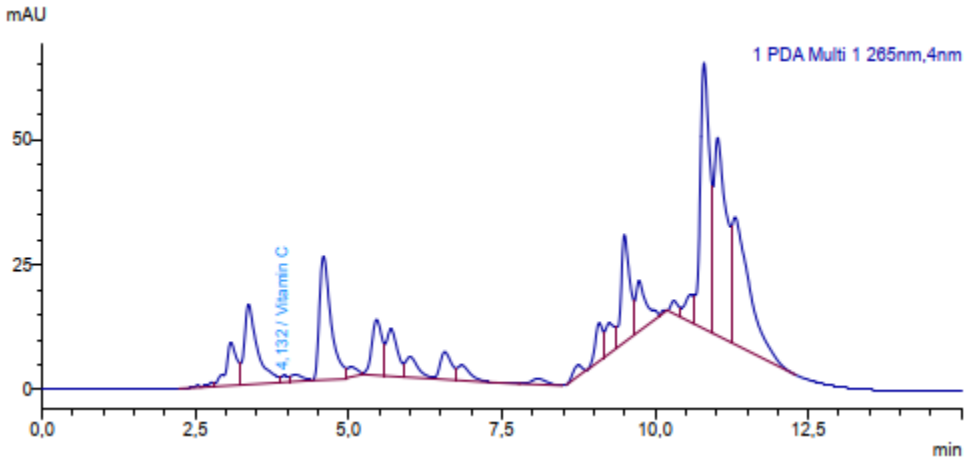

Figure 4. Chromatogram of vitamin C in the sample extract.

Table 3. Amount of vitamins in the extract and retention times.

Vitamin	Holding time, sec	Concentration, mg/l	Amount in 100 g of sample, mg
Vitamin B_1	2.923	0.656	1.640
Vitamin B_3	5.684	4.283	10.708
Vitamin PP	8.087	1.402	3.505
Vitamin B_9	16.87	0.273	0.683
Vitamin B_2	18.967	0.57	1.425
Vitamin B_6	6.266	0.412	1.030
Vitamin B_{12}	Not specified	0	0.000
Vitamin C	4.132	0.651	1.628

Discussion

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Bioconversion refers to the transformation of substances into other valuable forms through biological processes such as fermentation, microbial activity, enzymatic reactions, or other biochemical means [9]. The complex bioconversion of sunflower husks results in extracts that are enriched with biologically active compounds, particularly B-group vitamins and vitamin C. Incorporating such extracts into food products enhances their functional properties, offering significant health benefits to consumers [10].

The application potential of these vitamin-rich extracts in various product categories is outlined as follows:

- Bakery products: Enrichment with 3–5% extract can increase the levels of vitamins B3 and C by up to 30–40%.
- Dairy products: During fermentation, the addition of 2–3% extract can raise the concentrations of vitamins B2 and B6 by approximately 25–35%.
- Infant foods: Incorporating 1–2% extract can lead to a 20–30% increase in vitamin C content.
- Meat-based semi-finished products: Supplementation with 2–3% extract enhances the levels of vitamins B1, B3, and B6 by about 15–25%.
- Animal feeds: Adding 5–7% of the powdered extract can boost immune-enhancing vitamins by up to 40%.
- Sports supplements: The extract can contribute to covering 20–35% of the recommended daily intake of essential vitamins.

It should be noted that these values are based on laboratory-derived theoretical calculations. Further empirical studies and practical trials are required to validate the precise efficacy and stability of vitamin enrichment under industrial conditions [11-15].

Conclusions

The conducted research demonstrated that sunflower husks are rich in water-soluble vitamins, and that their complex bioconversion allows for potential application in both the food and feed industries. Vitamins identified through High-Performance Liquid Chromatography (HPLC), particularly B1, B2, B3, B6, B9, and C, significantly enhance the biological value of the sunflower husk extract.

These vitamins play vital roles in various metabolic and physiological processes essential for both human and animal health. Notably, the relatively high concentrations of vitamins B3 and C suggest that the extract could serve as an effective enrichment component in nutritional and functional products.

Moreover, the valorization of sunflower husks, traditionally considered waste, into high-value products through biotechnological processes supports sustainable production practices, waste-free technologies, ecological safety, and the development of export-oriented feed preparations.

It is recommended that future research should focus on evaluating the extract's resistance to technological processing, its bioavailability, and its functional impact on final product formulations. Additionally, it is necessary to develop practical mechanisms for the commercial integration of sunflower husk-based extracts into marketable products based on empirical data obtained through applied experiments.

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