

ISSN 2181-8622

Manufacturing technology problems



Scientific and Technical Journal Namangan Institute of Engineering and Technology

INDEX  COPERNICUS
INTERNATIONAL

**Volume 10
Issue 4
2025**



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STUDY OF POLYPHENOLIC COMPOUNDS IN JUJUBE (ZIZIPHUS JUJUBA MILL.) GROWN IN UZBEKISTAN

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Abstract: This research provides information about the locally cultivated *Ziziphus jujuba Mill.* (Chinese jujube). In particular, the study presents its botanical description, chemical composition, biological characteristics, and an analysis of studies conducted by foreign scientists. In laboratory conditions, aqueous and alcoholic extracts of this fruit were obtained. After filtration, alcoholic extraction was carried out in the next stage. The contents of flavonoids and gallic acids in the extracts and concentrates were determined using high-performance liquid chromatography (HPLC). The obtained results are presented in tables.

Keywords: jujube, extract, amino acid, vitamin, organic acid, polyphenol, flavonoid, gallic acid, apigenin, catechin.

Introduction. *Ziziphus jujuba Mill.* — a species of the genus *Ziziphus* belonging to the family *Rhamnaceae* — is traditionally cultivated in the Mediterranean region, South and East Asia, and southern China [1]. As of today, the total global orchard area of jujube — a subtropical fruit — covers 410 thousand hectares, with an annual yield of 7.5 million tons.

Jujube is widely cultivated in countries such as China, India, South Korea, Afghanistan, Pakistan, the United States, and Russia. In China alone, intensive-type jujube orchards occupy more than 200 thousand hectares.

Currently, jujube trees are also widespread in Asia, Australia, and Europe, including Slovakia. Due to their drought resistance and potential applications in the food industry, jujube trees attract significant attention from researchers and food manufacturers [2]. Because of their biological properties, jujube fruits are increasingly consumed fresh or used in food products.

Chemically, jujube fruits contain 81–83% moisture; vitamins (A, B-group, and especially vitamin C); sugars (22%, including galactose, fructose, and glucose); organic acids (citric, malonic, and malic acids); minerals (Ca, K, Fe); fatty acids (oleic acid — 71.7%, linoleic acid — 15%); amino acids (0.8%, including Asn, Pro, Arg, Ala, Glu, Ser, Asp); fiber (1.3%); and polyphenols [3]. Jujube fruits are used in a variety of food products (especially in India and Africa), including compotes, alcoholic beverages, marinades, cakes, and bread [4]. Furthermore, jujube fruits are known for their health-promoting effects and are widely used in traditional medicine to detoxify the body, prevent anemia, boost energy, reduce pain, and strengthen the immune system. In folk medicine, the peel and pulp of the jujube fruit are used as expectorants and remedies for coughs and colds. The leaves possess anti-inflammatory, antidiarrheal, and

antiemetic effects; they are consumed as tea and used as cold compresses to reduce swelling and treat colds, as well as for managing diabetes. The seeds of jujube exhibit sedative and sleep-inducing properties [5].

The peel of the jujube fruit changes color from green to yellow, then to reddish-yellow, and finally to red as it ripens – representing distinct ripening stages commonly referred to as green, white, half-red, and fully red [6]. Fresh jujube fruits spoil quickly, and under ambient conditions, their composition deteriorates within ten days after harvest [7]. Therefore, dried jujube products have become the main commercial form due to their long shelf life, convenience, and high quality.

Despite its many beneficial qualities and wide use, the fruit of jujube remains insufficiently studied. Due to its high content of biologically active compounds, it can have a significant positive impact on human health [8]. Extracts from different parts of the fruit – peel, pulp, and seeds – have shown high antioxidant activity, primarily attributed to phenolic compounds. Jujube contains numerous phenolic compounds such as chlorogenic acid, gallic acid, protocatechuic acid, and caffeic acid.

The quality of commercial jujube products largely depends on their content of bioactive compounds such as polyphenols, flavonoids, and carotenoids. However, various factors – including geographic origin, cultivar, processing, and storage conditions – can influence their bioactivity.

Thus, while jujube is an excellent source of nutrients and phytochemicals, its full potential in commercial applications and contribution to healthy nutrition remains to be fully explored.

Phenolic Compounds in Jujube Fruits

Interest in studying the phenolic compounds of jujube fruits is linked to the role of these biologically active substances (BAS) in providing antioxidant, anti-inflammatory, membrane-stabilizing, and other pharmacotherapeutic effects.

The total phenolic content of jujube fruits varies depending on the region of cultivation, cultivar, and whether the raw material is fresh or dried. For instance, in jujubes grown in different regions of China, the total phenolic content ranged from 0.9 to 1.2 g per 100 g of dry raw material [9].

Other Chinese researchers reported that the phenolic content in various fresh jujube cultivars ranged between 359.38 and 1041.33 $\mu\text{g/g}$ [10]. Another study indicated that the total phenolic content in fresh jujube fruits is about 366 mg per 100 g [11].

Numerous studies have been devoted to identifying and isolating individual biologically active compounds in jujube fruits. Extracts from fruits collected in various regions of China contained gallic and caffeic acids, (-)-epicatechin, phloridzin, and cyanidanol. The concentration of (-)-epicatechin was shown to vary significantly depending on the geographic origin of the fruit [12].

According to Xiaofang Xue and co-authors, phenolic compounds identified at different ripening stages of jujube fruits include (+)-catechin, epicatechin, rutin, quercetin,

kaempferol, spinosin, phloridzin, luteolin, gallic and chlorogenic acids, and at the final stage — isorhamnetin.

General data on phenolic-type biologically active substances in jujube fruits indicate that their phenolic acids are represented by gallic, chlorogenic, caffeic, ferulic, and other acids. Flavonoids include flavan-3-ols (procyanidins B1, B2, B3, (+)-catechin, (-)-epicatechin), flavonols (quercetin, rutin, quercetin and kaempferol derivatives), as well as flavanones (eriodictyol derivatives) and the dihydrochalcone phloretin-3',5'-diglucoside [13,14].

Studies have shown that among the phenolic compounds identified in jujube fruits, catechin (2.46–3.74 mg/100 g of fresh raw material) and rutin (0.88–3.60 mg/100 g of fresh raw material) are predominant in quantity [15, 16, 17]. In addition to known flavonoids, a new compound of this group — zivulgarin (4-beta-D-glucopyranosylswertisin) — was also identified in jujube fruits [18, 19, 20].

This research focuses on obtaining extracts from jujube grown in Uzbekistan and determining the flavonoid content in these extracts.

Research Methods.

Determination of flavonoid content. Flavonoid analysis was carried out using HPLC with gradient elution and a diode-array detector (DAD). Acetonitrile and a buffer solution were used as the mobile phase. Spectral data were analyzed in the wavelength range of 200–400 nm.

Chromatography conditions: Chromatograph — *Agilent Technologies 1260*; Mobile phase (gradient mode): acetonitrile – buffer solution, pH = 2.92 (4% : 96%) 0–6 min, (10% : 90%) 6–9 min, (20% : 80%) 9–15 min, (4% : 96%) 15–20 min; Injection volume — 10 μ L; Flow rate — 0.75 mL/min; Column — *Eclipse XDB-C18*, 5.0 μ m, 4.6 \times 250 mm; Detector — diode-array detector, wavelengths 254 and 320 nm.

Main Part.

Dried fruits of the *Ziziphus jujuba Mill.* species grown in the Tashkent region were used for the study. Aqueous and alcoholic extracts and concentrates were prepared from the fruits.

First, the fruits were crushed into small pieces using knives (due to their fibrous consistency). Then, 50 g of the material was weighed and placed into a flask. Distilled water was added (in a 1:3 ratio), and the mixture was extracted in a water bath at 50°C for 2 hours.

The extract was filtered through a cloth filter, separating the solid residue and the liquid phase. The remaining solid residue was placed back into the flask, and 70% ethanol was added in a 1:2 ratio. Extraction was carried out for 3 hours at 80°C using a reflux condenser.

After completion, the mixture was filtered again. Samples from the aqueous and alcoholic extracts were taken for analysis, and the remaining portions were evaporated using a rotary evaporator at 45°C to remove excess water and alcohol. The evaporation continued until the volume was reduced to one-quarter of the original.

The content of polyphenolic compounds in the obtained extracts and concentrates was determined using high-performance liquid chromatography. The obtained results are presented in the following tables.

Table 1. Polyphenolic Compounds in the Samples.

№	Name of Polyphenols	Flavonoid Content, mg/100 g			
		Apigenin	Catechin	Gallic acid	Total
1	Aqueous extract	-	0.177	9.682	11.217
2	Alcoholic extract	1.358	5.385	3.389	10.132
3	Concentrate (aqueous)	2.365	1.363	35.562	39.348
4	Concentrate (alcoholic)	2.469	5.456	21.786	29.711

From the Table.

As can be seen from the table, the amounts of apigenin and catechin in the aqueous extract and concentrate are very small, whereas their contents are somewhat higher in the alcoholic extract and concentrate. In contrast, the amount of gallic acids is higher in the aqueous extract and concentrate than in the alcoholic ones. Thus, it has once again been confirmed that gallic acids dissolve better in water than in alcohol.

Conclusion. In this study, the flavonoids and gallic acids contained in aqueous and alcoholic solutions of the local jujube (*Ziziphus jujuba Mill.*) species were analyzed using high-performance liquid chromatography (HPLC).

According to the results obtained, only the flavonoids apigenin and catechin were detected in the solutions, while no other flavonoid types were identified. The concentrations of these flavonoids were also relatively low. Therefore, it is necessary to conduct additional experiments to achieve maximum extraction of flavonoids into the solvent.

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