

CHEMICAL COMPOSITION AND BIOLOGICAL ACTIVITY OF TRAPA NATANS L.

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HEMIJSKI SASTAV I BIOLOŠKA AKTIVNOST TRAPA NATANS L.

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ABSTRACT

Trapa natans L., water chestnut (indian name singada, montenegrin name kasaronja) belonging to Trapaceae family is annual aquatic floating herb native to Asia but widely distributed in the Europe and in North America. The presence of two species, *Trapa natans* L. and *Trapa longicarpa* M.Jank. ssp. *scutarensis* M.Jank has been confirmed on the Skadar lake in Montenegro. *Trapa natans* is a plant demonstrating promising pharmacological activity mostly due to the presence of its constituents, phenols and flavonoids, showed hepatoprotective potential, antioxidant, anti-inflammatory, antidiabetic, antifungal and in vitro antimicrobial activity.

Keywords: *Trapa natans*, water chesnut, pharmacology effects, medical use

SAŽETAK

Trapa natans L., vodeni kesten (indijsko ime singada, crnogorsko ime kasaronja) koji pripada porodici Trapaceae jednogodišnja je vodena plutajuća biljka poreklom iz Azije, ali široko rasprostranjena u Evropi i Severnoj Americi. Prisustvo dve vrste, *Trapa natans* L. i *Trapa longicarpa* M.Jank. ssp. *scutarensis* M.Jank potvrđena je na Skadarskom jezeru u Crnoj Gori. *Trapa natans* je biljka koja pokazuje obećavajuću farmakološku aktivnost uglavnom zahvaljujući prisustvu njenih sastojaka, fenola i flavonoida, pokazala je hepatoprotektivni potencijal, antioksidativno, protivupalno, antidijabetičko, antimikotično i in vitro antimikrobno delovanje.

Ključne reči: *Trapa natans*, vodeni kesten, farmakološki efekti, medicinska upotreba



INTRODUCTION

Trapa natans L., water chesnut, belonging to *Trapaceae* family and it is annual aquatic floating herb native to Asia, where it is known for its local name Singhara (1). Nowadays plants from *Trapa* genus are widely distributed in the Europe and naturalized in North America (2). Number of *Trapa* species has not been precisely defined so far, however it is certainly more than 20 (3). A presence of two species, *Trapa natans* L. and *Trapa longicarpa* M.Jank. ssp. *scutarensis* M.Jank. has been confirmed on the Skadar lake in Montenegro (4). Local name for water chesnut in Montenegro is kasa-ronja and its parts such a fruit with seed were of a great importance in the nutrition of population during the years of poverty (5). In Asian continent, including India and China, water chesnut has been widely used for medical purposes since ancient times, (6) however this plant species has not been use in traditional medicine in Montenegro.

Trapa natans Is one of the most important medicinal plants of Indian Ayurveda due to its use in the treatment of stomach disorders, genitourinary system, liver, kidney and spleen disorders (6).

Based on pharmacological potential *Trapa natans* L. has been recognized as the most significant plant species belonging to *Trapa* genus. Therefore this review will summarize the current knowledge from the available literature referring to *Trapa natans*.

DESCRIPTION

Trapa natans L. grows in lakes and some rivers requiring conditions such as sunny places and slightly acidic water.

It has surfacing and submerged leaves. Characteristics of triangular surfacing leaves involve toothed edge and an inflated petiole which forms a rosette on the surface of the water. Submerged leaves are feather-like and they are circled around the stem of the plant. White flowers are formed on the axils and in July come out to the surface and bloom. Fruit is in form of nut, and has a hard „wooden“ surface with four sharp spines which are edible. Stems are rarely branching, long, rope-like and can attain lengths of up to 5 meters. The seeds germinate in the substratum, firmly attached to the lateral roots (7).

CHEMICAL COMPOUNDS

Literature data indicate the presence of essential minerals, proteins, lipids, carbohydrates as well as vitamins (B1, B2, B5, B6, E, A, C), dietary fibers and polyphenols such as phenolic acids and flavonoids as well as hydrolyzable tannins in water chesnut (8). Peels of water chestnuts have been shown to have a much higher content of phenolic compounds than

kernels (9). Ten phenolic compounds have been identified in *Trapa natans* leaf extract so far, while three phenolic acids, gallic, ellagic and ferulic acid, as well as quercetin 3-O-galactoside (hyperoside), are present in a large quantity comparing to other compounds (10).

Aqueous extract of *Trapa natans* L. fruits

The most significant compounds in the aqueous *Trapa natans* L. fruit extract are phenolics which have gained increased scientific attention due to their prominent free radical scavenging activity. There is certain evidence that the aqueous extract of *Trapa natans* L. fruit exert great *in vitro* antioxidant activity against free radicals due to its polyphenolic content (9). (Table 1.)

Table 1. The total content of phenolic, flavonoid and tannin compounds in the aqueous *Trapa natans* L. fruit extract

gallic acid equivalents/g of dry material	63.81 mg
rutin equivalents/g of dry material	21.34 mg
tannin equivalent /g of dry material	17.11 mg

(source reference: Malviya, N. et all. 2010)

In addition, extract of *Trapa natans* L. in mentioned study showed a crucial property of protein oxidation inhibition. So far it has been known that only a few plant extract constituents exert that kind of activity which can inhibit the progression of a number of diseases implicating by oxidative stress (9).

Trapa natans methanol leaf extracts

Phytochemical analysis of methanol extract, as the least toxic when compared to acetone and ethyl acetate leaf extracts in the zebrafish (*Danio rerio*) model system, identified phenolic compounds such as p-hydroxybenzoic acid, phenolic acids and derivatives, flavonoids and flavonoid glycosides (10). (Table 2.)

Flavonoids are chemical compounds that protect cells against the damaging effects of reactive oxygen species, such as superoxide, singlet oxygen, hydroxyl radicals, peroxy radicals and peroxynitrite. There are many epidemiological studies that have shown flavonoid intake is inversely related to mortality due to cardiovascular disease (15).

Phenolics in plant species form one of the major groups of compounds which primary act as antioxidants. Antioxidant activity of phenolics is based on absorption and neutralization of free radicals, quenching singlet and triplet oxygen or decomposing peroxides (16). There are suggestions that polyphenolic compounds have inhibitory effects on mutagenesis and carcinogenesis in humans, by daily ingesting approximately 1.0 g from a diet rich in vegetables and fruits (17).



Table 2. Content of phenolic compounds of methanolic *Trapa natans* L. leaf extract

Phenolic acids and their derivatives	Flavonoids	Flavonoid Glycosides
<ul style="list-style-type: none"> ➤ Gallic acid hexoside isomer 1 and isomer 2 ➤ Gallic acid ➤ Protocatechuic acid ➤ p-Hydroxybenzoic acid ➤ p-Coumaric acid hexoside ➤ Caffeic acid ➤ p-Coumaric acid ➤ Ellagic acid ➤ p-Coumaroyl-digalloyl-O-glucose ➤ Ferulic acid 	<ul style="list-style-type: none"> ➤ Quercetin ➤ Pinobanksin ➤ Naringenin ➤ Rhamnetin 	<ul style="list-style-type: none"> ➤ Naringenin-7-O-hexoside ➤ Kaempferol di-O-hexoside isomer 1 ➤ Rutin ➤ Kaempferol-3-O-glucoside (Astragalin) ➤ Quercetin 3-O-rhamnoside ➤ Quercetin 3-O-galactoside

(source reference: Aleksic, I. et al. 2018)

Inorganic constituents of *Trapa natans*

Trapa natans contains many inorganic constituents such as minerals, phosphorus, calcium, iron, copper, magnesium, manganese, sodium and potassium (11). Mineral contents of 100g of the seeds were 32 mg (12).

PHARMACOLOGICAL PROPERTIES OF EXTRACTS OF TRAPA NATANS PARTS

Several studies demonstrated promising pharmacological activity of *Trapa natans* extract mostly dedicated to the presence of phenols and flavonoids.

Hepatoprotective activity

Trapa natans fruit peel ethanolic extract showed hepatoprotective potential against antitubercular drugs induced hepatotoxicity. Hepatotoxicity was induced in rats by intraperitoneal injection of antitubercular drugs and the level of marker enzymes confirming induced hepatotoxicity was measured. *T. natans* peel extract application (200 and 400 mg/kg) resulted in significant reduction in liver marker enzymes and antioxidant parameters (13). Study also showed that *T. natans* ethanolic root extract and its derived fractions have significant protective effect against hepatotoxicity and lipid peroxidation. Identification of ferulic acid and caffeic acid in *Trapa natans* extract might explain the mentioned activity due to antioxidative nature of both constituents (1).

Anti-inflammatory activity

As mentioned earlier, phenolic compounds such as gallic acid were detected in water chesnut extracts. (10). It seems that gallic acid have useful pharmacological properties, including antioxidant, anti-inflammatory, anti-viral, and anti-cancer activities (18)(19). Consequently, gallic acid has a main anti-inflammatory activity in water chesnut extract and

there are evidences that its use may be a possible therapeutic agent against skin inflammatory disorders (20).

Antidiabetic activity

Trapa natans fruit peel methanol extract has shown antidiabetic activity against streptozotocin induced diabetic rats. One of the main advantages is that ethanolic extract of *Trapa natans* roots and its derived fractions can slow down the progression of hyperglycemia without inducing hypoglycemic effect. Antihyperglycemic activity of *Trapa natans* might be attributed to the presence of ferulic acid and caffeic acid (14). Ferulic acid is known to reduce oxidative stress and hyperglycemic response while caffeic acid has generative effect on Langerhans islets following reduction in expression of glucose transporter 2 in liver and consequently, reduce hyperglycemic effect (21)(22).

Antifungal activity

An isolated peptide from *Trapa natans* fruits possesses antifungal activity against *Candida tropicalis*. In one study, TnAFP1, one uncommon small antifungal peptide with molecular mass of 1230 Da was isolated and identified from the fruits of *T. Natans*. Antifungal peptide from *T. natans* fruit showed better antifungal activity against *C. tropicalis* with inhibitory effect on biofilm formation, compare to several previously known peptides (23).

Anthelmintic activity

Methanol extracts of fruit peel also have anthelmintic activity according to study. It showed dose-dependent activity on adult Indian earthworms *Pheritima posthuma* and aquarium worm *Tubifex tubifex*. Three concentrations viz., 5, 10 and 20 mg/ml of aqueous and methanolic extracts of fruit peels and root were studied to determine the time of paralysis and time of death and results showed that methanolic extracts possess significant anthelmintic activity at highest concentration of 20 mg/ml. Piperazine citrate in 10mg/ml



concentration was evaluated as standard reference and distilled water as control (24). Flavonoids, tannins and polyphenolic compounds show anthelmintic activity according to literature and it is expected that methanolic extracts of *Trapa natans* peel exert that kind of pharmacological effect (25)(26).

Antimicrobial activity

Several extracts of fruit peel showed *in vitro* antimicrobial activity. *Trapa natans* L. fruit peel extracted in different solvents with increasing polarity: 1,4-dioxan, chloroform, acetone, dimethylformamide, ethanol and water showed different intensity. Traditional use of water chestnut was mostly referring to its water extract, but founded studies pointed that the plant extracts, extracted in different organic solvents showed profoundly distinct antibacterial activity than aqueous extract. Maximum effect was with dimethylformamide while minimum was with petroleum ether extract. *Pseudomonas aeruginosa*, *P. vulgaris* and *P. pseudoalcaligenes* were completely resistant at all extracts, while best antibacterial activity was shown against *P. Putida* (best antibacterial effect shown by 1,4-dioxan extract) followed by *P. testosterone* and *P. morgani* respectively while *P. mirabilis* was inhibited by 4 of the solvents only (1,4-dioxan, acetone, dimethylformamide, ethanol). Amongst *Klebsiella* strains, *K. pneumoniae* showed good antibacterial activity with all the solvents. Study showed maximum antibacterial activity against Gram negative bacteria (27).

TRADITIONAL USE

Trapa natans in Montenegro has been predominantly used in nutrition, however its therapeutical potential in traditional medicine has not been clarified so far (5). On the other hand, in Asia continent including India and China, water chestnut was often used for medical purposes. Since ancient times, good pharmacological properties of this plant species as antipyretic, diuretic, hemostatic, constipating, antidiarrheal, have been recognized and applied in traditional Chinese and Indian medicine (6). It has been traditionally used in India in a form of liniments made from the fruits and as a cure for rheumatism, sore and sunburns (28).

CONCLUSIONS

There are scientific reports confirming pharmacological properties of *Trapa natans* in various diseases and conditions. Nevertheless the full therapeutic potential of this plant species has yet to be understood and discovered.

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