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PHARMACOGNOSY AND PHARMACOLOGICAL EFFECTS OF TRIBULUS TERRESTRIS: A REVIEW

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ABSTRACT

Tribulus terrestris (family Zygophyllaceae), commonly known as Gokshur or Gokharu or puncture vine, has been used for a long time in both the Indian and Chinese systems of medicine for treatment of various kinds of diseases. Its various parts contain a variety of chemical constituents which are medicinally important, such as flavonoids, flavonol glycosides, steroidal saponins, and alkaloids. It has diuretic, aphrodisiac, antiurolithic, immunomodulatory, antidiabetic, absorption enhancing, hypolipidemic, cardiotonic, central nervous system, hepatoprotective, anti‑inflammatory, analgesic, antispasmodic, anticancer, antibacterial, anthelmintic, larvicidal, and anticariogenic activities. In order to evaluate the therapeutic claims made for this plant in traditional medicine, the ethanol extract of T. ferrestris (fruit) was tested for activity against artificially induced urolithiasis in albino rats. Tribulus terrestris is used as a urinary anti-infective in folk medicine. To validate this use, the in vitro anti-bacterial activity of methanolic extracts of different parts (fruits, stems plus leaves and roots) of T. ter- restris L. Antifungal activity of natural products is being studied widely. Saponins are known to be antifungal and antibacterial. We used bioassay-guided fractionation to have isolated eight steroid saponins from Tribulus terrestris L.

KEYWORDS: Tribulus terrestris; Antiurolithiatic activity; Antibacterial activity; antifungal activity.

INTRODUCTION

The traditional medicinal methods, especially the use of medicinal plants, still play a vital role to cover the basic health needs in the developing countries and moreover the use of herbal remedies has risen in the developed countries in the last decades.[1] In this connec- tion, plants continue to be a rich source of therapeutic agents. TT is an annual plant of the family Zygophyllaceae, which is commonly known as Tribulus, Hard thorns, and goat head in China. It is mainly planted in the Mediter-ranean and in sub-tropical regions such as India, China, South America, Mexico, Spain, Bulgaria, and Pakistan. It is a small, prostrate, 10–60 cm high, hirsute or silky hairy shrub. The leaves are opposite, often unequal, paripinnate, pinnate from 5 to 8 pairs and elliptical or an oblong lanceolate. The fruits from the five mericarps are ax-shaped, 3–6 mm long, and arranged radially and have a diameter of 7–12 mm and a hard texture. The root is slender, fibrous, cylindrical and frequently branched, bears a number of small rootlets and is light brown in colour. The fruits and roots of TT, as a folk medicine, have been used for thousands of years in China. Over the last several years, it has been certified for its pharmaceu-tical activities for improving sexual function and cardiac protection and providing anti-urolithic, antidiabetic, anti-inflammatory, antitumour and antioxidants effects.[2]

The main constituents of T. terrestris are saponins, diosgenins, alkaloids and amides.[3]

TRIBULUS TERRESTRIS PLANT

- **Kannada – Negalu**
- **Hindi – Cholagokhru**
- Bengali – Gokhru
- Malayalam – Neringil
- Marati – Lahangokharu
- Sanskrit – Gokshura
- Tamil – Nerunji
- Telugu – Palleru

- Punjabi – Lotal[4]

Batanical description

It is small prostrate, 10-60 cm height, hirsute or silky hairy shrub. Leaves are opposite, often unequal, paripinnate; pinnae from five to eight pairs, elliptical or oblong lanceolate. Flowers are yellow in color. It carpel fruits are of characteristic, stellate shape, somewhat round-shaped, compressed, five cornered, and covered with princkles of very light yellow color. There are several seeds in each crocus with transverse partition between them. The seeds are oily in nature. When fresh, the root is slender, fibrous, cylindrical, frequently branched, bearing a number of small rootlets and its of light brown color. Fruits and roots are mainly used as a folk medicine for the treatment of various ailments. Root occurs in pieces, 7-18 cm long and 0.3-0.7 cm in diameter,

- Kingdom – Plantae
- Division – Phanerogram
- Subdivision – Angiospermae
- Class – Dicotyledonae
- Subclass – Polypetalae
- Series – Disciflorae
- Order – Giraniales
- Family – Zygophyllaceae[5]

Phytochemical investigations

Many different compounds with a variety of biological properties and chemical structures have been identified from TT, including steroidal saponins, flavonoids, glycosides, phytosterols, tannins, terpenoids, amide derivatives, amino acids, and proteins. Among the different types of constituents, steroidal saponins and flavonoids are considered to be the most important metabolites with various bioactivities.

Steroidal saponins

Spirostanol and furostanol saponins are considered the most characteristic chemicals in TT. To date, 108 kinds of steroidal saponins have been isolated from TT. Among them, there are 58 kinds of spirostane saponins and 50 kinds of furostane saponins. The steroidal saponins, such as protodioscin and protogracil- lin, are thought to confer TT unique biological activities.

Flavonoids

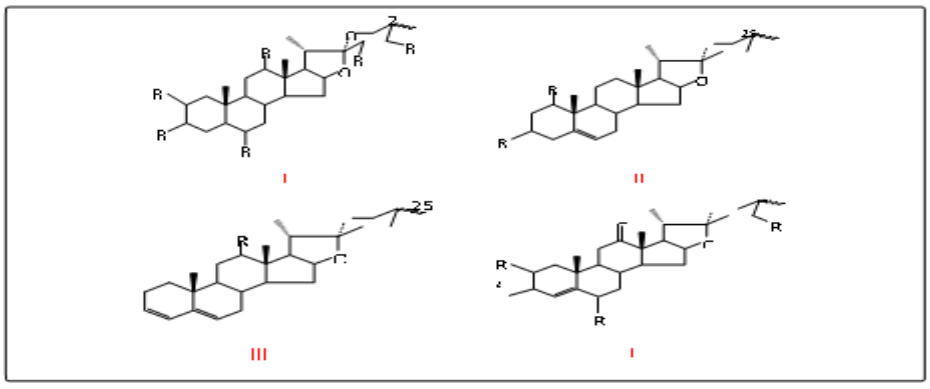
The flavonoids of TT are mainly derivatives of quercetin, kaempferol and isorhamnetin. Quercetin,isoquercitrin, rutin,quercetin-3-O-gent,quercetin-3-O-gentr, quercetin-3-O-rha-gent, quercetin-3-O-gent-7-O-glu are flavonoids with quercetin as the basic parent structure [34–36]. Isorhamnetin, isorhamnetin-3-O-glu, isorhamnetin-3-O-gent, isorhamnetin-3-O-rutinoside, isorhamnetin-3-O-gentr, isorhamnetin- 3,7-di-O-glu, isorhamnetin-3-O-p-coumarylglu, isorhamnetin-3-O-gent-7-O-glu, isorham- netin-3-O-gentr-7-O-glu are flavonoids with isorhamnetin as the basic parent structure. Kaempferol, kaempferol-3-O-glu, kaempferol-3-O-gent, kaempferol-3-O-rutinoside, kaempferol-3-O-gent-7-O-glu, tribuloside are flavonoids with kaempferol as the basic parent struc- ture.

Alkaloids

Tribulusamide C, tribulusterine, tribulusin A, harmine, harman, harmmol, tribulusimide C, terrestriamide, N-trans- coumaroyltyramine, N-trans-caffeoylyramine, terrestribisamide are the main alkaloids iso- lated from the stems, leaves, and fruits of TT. The nuclear mainly belong to β-carboline alkaloids and amide alkaloids. Others Other components of TT include organic acids, amino acids and other substances. Organic acids isolated from TT are benzoic acid, vanillic acid, 2-methyl benzoic acid, ferulic acid], succinic acid, palmitic acid mono- glyceride, succinic acid, docosanoic acid, Tribulus acid and others. The main amino acids are alanine and threonine. In addition, TT also contains 4-keto- pinoresinol, uracil nucleic acid, coumarin, emodin, and physcion.

Pharmacological Activities

- Diuretic activity
- Aphrodisiac activity
- Antiurolithic activity
- Immunomodulatory activity
- Antidiabetic activity
- Absorption enhancer
- Hypolipidemic activity
- Activity in cardiac disorders
- Central nervous system (CNS) activity
- Hepatoprotective activity
- Anti inflammatory activity[6]



Skeletal types of spirostane saponins in T. terrestris[3]

Anti-urolithiatic activity from plant drugs

The two siddha drugs aerva lanata and vediuppuchunnam tested for hyperoxaluria calculi induced in rats using ethylene glycol in drinking water. Increased the urine volume, and reduced calcium oxalate and other crystallizing salts.[7]

The Actinidiachinesis(plum) vacciniummacrocarpon (cranberry) and Ribesnigrum (blackcurrant) juice decreased the urinary pH whereas the excretion of oxalic acid and the relative super saturation for uric acid increased cranbery juice acidifies urine, and found useful in the treatment of urinary tract infection.[8]

The effects of seven plant drugs Verbena officinalis Lithospermummofficinale Taraxacumofficinale Equisetum arvense, Arctostaphylosura-ursi Arctiumlappa and Silenesaxifraga drugs are considered that it prevents and treat the kidney stone formation due to the kidney stone formation due to the presence of saponins.[9]

The leaf extract of coleus aromatics' has shown in reduction in deposition of urinary calculi induced by glycolic acid in experimental rats.[10]

ANTIBACTERIAL ACTIVITY

Chemicals. Methanol (99.5%) (Merck); Ampicillin sodium (Merck); Gentamicin hydrochloride (Merck).

Plant material. Tribulus terrestris used in this study was collected at the end of November from lands around the Qom–Arak highway 10 km away from Arak in the Markazi province in Iran. Herbarium specimen of T. terrestris (voucher number 1237) is preserved in the herbarium of department of botany of Arak University, Arak, Iran.

Extraction procedure. Dried and powdered fruits, stems plus leaves and roots (100 g) were extracted with 85% methanol in a Soxhlet apparatus and the extracts were dried in vacuo by a Heidolph model Laborta 4001 rotary evaporator**.**

Media. The media used in broth dilution assays were brain-heart infusion broth (BHI broth) (Merck) and Mueller-Hinton broth (MH broth) (Merck) and the medium used in agar diffusion assay was Mueller- Hinton agar (MH agar) (Merck).

Test bacteria. Staphylococcus aureus ATCC 29213, Enterococcus faecalis ATCC 29212, Escherichia coli ATCC 25922, Pseudomonas aeruginosa ATCC 27853.

ANTIBACTERIAL ACTIVITY TESTS

Broth dilution assay. Broth dilution assay was car- ried out according to Murray et al [. A loopful of the bacterial culture from the slant was inoculated in the nutrient broth (BHI broth as well as MH broth) and in- cubated at 37±1ºC for 24 hours. The fresh broth (20 mL) was seeded with 0.25 mL of the 24-hour broth cul- tures and a two-fold serial dilution method was followed as below. The dried plant extracts were dissolved in 85% methanol to obtain an 80 mg/mL solution and ster- ilized by filtration through a 0.45 µm membrane filter. A 0.2 mL solution of the material was added to 1.8 mL of the seeded broth and this formed the first dilution. 1 mL of this dilution was diluted further with 1 mL of the seeded broth to produce the second dilution, and the process was repeated until six dilutions were obtained. A set of tubes containing only seeded broth was kept as control and 85% methanol controls were also main- tained. After incubation for 24 h at 37±1ºC the last tube with no visible growth of the bacteria was taken to rep- resent the minimum inhibitory concentration (MIC) of the test sample which is expressed in mg/mL. Moreover, the broth dilution assay was carried out with ampicillin and gentamicin in BHI broth as well as MH broth in the same way as the extracts and the MIC values of ampicil- lin and gentamicin were determined.

Agar diffusion assay. The dried plant extracts were dissolved in 85% methanol to a final concentration of 40 mg/mL and sterilized by filtration through a 0.45 µm membrane filter. Agar disc diffusion assay was then carried out according to Murray et al[11] using an in- oculum containing 106 bacterial cells on MH agar plates (1 mL inoculum/plate). The discs (diameter, 6 mm) were each impregnated with 50 µl of extract (2 mg/disc) at a concentration of 40 mg/mL and placed on the in- oculated agar and incubated at 37ºC for 24 h. Each test was carried out in triplicate with controls. Moreover, filter paper discs containing the antibiotics ampicillin and gentamicin were used as positive controls.[1]

ANTIFUNGAL ACTIVITYY

MEDIA

All strains used in this study were grown in two complete media consisting of a YEPD liquid medium (1% Bacto Pep- tone [Difco, USA], 0.5% yeast extract [Difco], 2% glucose [Sangon]), and a solid medium prepared by adding 2% agar (Sangon).

Antifungal susceptibility test

The in vitro minimal inhibitory concentrations (MICs) of the compounds were determined by the micro-broth dilution method according to the methods defined by the National Committee for Clinical Laboratory Standards (NCCLS, 2002). Candida krusei (ATCC6258) and Candida parapsilo- sis (ATCC22019) were quality controlled strains, and tested in each assay. Fluconazole (FLC), itraconazole (ICZ) and amphotericin B (AMB) obtained from their respective man- ufacturers served as the positive control. The drug MIC80 was defined as the first well with an approximate 80% reduc- tion in growth compared to the growth of the drug-free well. The eight compounds to be tested were dissolved in dimethyl sulfoxide (DMSO), and the stock solutions of the serial two-fold dilutions were prepared in RPMI 1640 medium (Gibco, USA) with the final concentrations between 128.0 and 0.25 g/mL (111.30–0.220 mol/L), and the final

concentrations of FLC, ICZ and AMB were 64.0–0.125 g/mL (209.15–0.410 mol/L), 2.0–0.004 g/ mL (2.83–0.006 mol/L) and 2.0–0.004 g/mL (2.16– 0.004 mol/L), respectively, depending on the MIC results from our preliminary study.[11]

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