



Effect of methanolic extract of *Tribulus terrestris* leaves on microbial growth

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Abstract

Tribulus terrestris, 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 alkaloids¹. In the present study we tried to evaluate the antimicrobial potency of methanolic extract of leaves of *Tribulus terrestris* using bacteria *E. coli* and *S. aureus*. The results are summarized at the end in the form of table.

Keywords: *Tribulus terrestris*, antimicrobial, alkaloids

Introduction

Medicinal plants are used at the household even by women taking care of their families, at the village level by medicine men or tribal shamans, and by the practitioners of classical traditional systems of medicine such as Ayurveda, Chinese medicine, or the Japanese Kampo system. According to the World Health Organization, over 80% of the world's population, or 4.3 billion people, rely upon such traditional plant-based systems of medicine to provide them with primary healthcare.

Allopathic medicine too owes a tremendous debt to medicinal plants: one in four prescriptions filled in a country like the United States is either a synthesized form of or derived from plant materials.

Even from the earliest trade data available, it is clear that the global market for medicinal plants has always been very large. According to the International trade Centre, as far back as 1967, the total value of imports of starting materials of plant origin for the pharmaceutical and cosmetics industry was of the order of USD 52.9 million. From this amount, the total values grew to USD 71.2 million in 1971, and then showed a steady annual growth rate of approximately 5-7% through to the mid-1980s

To give an example of the extent of trade volumes even at that time, according to one report commissioned by the World Wide Fund for Nature, the total import in 1980 of vegetable materials used in pharmacy" by the European Economic Community was 80,738 tons. India was the largest supplier by far, with 10,055 tons of plants and 14 tons of vegetable alkaloids and their derivatives.

However, it is only during the last decade that the real significance of the medicinal plants sector has begun to be realized. Interest in natural materials by the dominant economic powers had waned from the late 1960s to the early 1980s as new possibilities in biotechnology and the synthesis of drugs beckoned. But by the mid- 1980s, there was a renewed interest in natural materials and approaches to health care, coupled with recognition that

technology alone could not solve the pressing health care needs of the world's population.

This new drive for natural and plant-based medicines made itself felt in the market from the mid-in1980s onwards. Growth in the market in various regions is now on average 3 to 4 times the average growth rates of the national economies in the same regions. Some of these phenomenal rates, in some cases nearly 20%, imply that the market is now doubling in size every 4-5 years.

The participation of various companies in the market also a *Tribulus terrestris* tests to its new strength and importance by 1990, some 223 major companies worldwide (of which about half were in the United States) were reportedly screening plants for new leads; the figure had been zero in 1980.

Botany is a branch of biology studying plant life, including: structure, growth, taxonomy, systematic, reproduction, metabolism, physiology, biochemistry, development, diseases, ecology, and evolution of plants. Ethnobotany is the study of the relationship between plants and people and their culture.

William Harshberger (1895-1896), botanist in USA, termed "Ethnobotany" Leopold Glueck, 19th century German physician, ethno botanist Richard Evans Schulte, called father of modern ethnobotany.

Present plant: *tribulus terrestris* (Puncture vine)

- Kingdom: Plantae
- Division: Phanerogams
- Subdivision: Angiosperm
- Class: Dicotyledonae
- Subclass: Polypetalous
- Series: Disciflorae
- Order: Giraniales
- Family: Zygophyllaceae
- Genus: *Tribulus*
- Species: *Tribulus terrestris*

Tribulus terrestris is commonly known as *Gokshur* (Sanskrit); puncture vine, land English); *Gokharu* (Hindi); *Bethagokharu*

or *Nanagokharu* (Gujarathi); *Nerinjil* (Tamil); and *Khar-e-khusak khurd* (Urdu). It is distributed along a wide geographic perimeter. It is found all over India up to 11,000 ft in Kashmir, Ceylon, and all warm regions of both hemispheres. It is a common weed of the pasture lands, road sides, and other waste places, chiefly in hot, dry, and sandy regions including West Rajasthan and Gujarat in India.

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. Its carpel fruits are of characteristic, stellate shape, somewhat round-shaped, compressed, five cornered, and covered with prickles of very light yellow color. There are several seeds in each crocus with transverse partitions 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 is 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, cylindrical, fibrous, frequently branched, bearing a number of small rootlets, tough, woody, yellow to light brown in color, surface rough due to the presence of small nodules; fracture fibrous; odor aromatic; taste sweetish astringent. The fruits of the herb are known as "Chih-hsing" in China or goat head in USA. The spiky fruit looks like the cloven hoof of a cow and, hence, is known as go-ksura (cow-hoof). Fruits are faint greenish yellow with spines. They are globose, consisting of five, nearly glabrous, muriculate, wedge-shaped, woody cocci, each with two pairs of hard sharp spines, one pair longer than the other. Tips of spines almost meet in pairs together forming pentagonal framework around the fruit. Outer surface of the schizocarp is rough. There are several seeds in each coccus, with transverse partitions between them. Odor of fruits is faintly aromatic and taste is slightly acid.

Material and Method

Soxhlet Extraction

Thoroughly washed dried leaves of of a *TRIBULUS TERRESTRIS* extract of, of plant material were dried in shade for four days. Plant parts were placed in an oven at 38 °C and then powdered with mortar and pestle. 100 grams of each respective plant parts and 300 ml of ethanol were place in around boTribulus Terrestrisom and it was kept for soxhlet extraction for thoroughly. The ethanol extract kept at room temperature for 24 hours. The extracts were filter using what *Tribulus terrestris* man filter paper (No. 1) and dried at temperature below 45°C for ethanol removal to obtain the dense extract and then they were kept in sterile boTribulus Terrestrisles under refrigerated conditions until use.

Phytochemical Screening

The phytoconstituents of crude extract obtained above is done using chemical methods and the results are summarized in the table. No.1

Biological screening

The bacteria *E. coli*, and *S. aureas* were procured from Department of Biotechnology, KSWU Bijapur. They were pre-cultured in nutrient broth overnight in a rotary shaker at

37°C, centrifuged at 10,000 rpm for 6 min, pellet was suspended in double distilled water and the cell density was standardized spectrophotometrically (A nm). Culture grown on Potato dextrose agar medium. The Petri dishes were flooded with 9 to 11 ml of distilled water and the conidia were scraped using sterile spatula.

Antibacterial activity was measured using agar dilution technique. Briefly, the ethanol extracts were dissolved in dimethyl sulfoxide (DMSO, Merck) and serially diluted in molten Mueller Hinton Agar (MHA, Sigma) in petridishes (100 mm×15 mm) to obtain final concentrations: 100, 50, 25 and 12.5 µg/ml. The solvent did not exceed 1% concentration and did not affect the growth of the organisms. All bacterial strains were grown in Mueller Hinton Broth (MHB, Sigma) for 4 h at 37°C. Bacterial suspensions with 0.5 McFarland standard turbidity, which is equivalent to 108 cfu/ml were prepared by dilution with Mueller Hinton broth. The diluted inoculum was added to a Steer's replicator calibrated and incubated for 24 h at 37 °C. After incubation, all dishes were observed for microbial inhibition by the disc diffusion method. The results are summarized in the table. No.2

Results and Discussion

Table 1: Results of phytochemical testing

Class of compounds	Methanol extract
Phenols	+
Flavanoids	+
Steroids	-
Glycosides	+
Lignans	+
Saponins	-
Alkaloids	+

+ = Presence of constituents; - = Absence of constituents

Table 2: Results of antibacterial activity.

Test Sample	Concentration (µg/ml)	Zone of inhibition (in mm)	
		<i>E.Coli</i>	<i>S.aureus</i>
Sample-1	1000	13	14
Sample-2	500	11	12
Sample-3	250	09	10
Sample-4	175	06	08
Streptomycin	100	16	15
D. Water	--	06	06

Cup diameter= 6 mm Quantity of drug solution and control fed into each cup = 5µl

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