



**TRIBULUS TERRESTRIS (LINN): BOTANY, MEDICINAL USES, CHEMICAL  
COMPOSITION AND IN VITRO REGENERATION A REVIEW.**

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**ABSTRACT**

Nearly all culture, from ancient times, has used plants as a source of medicine. Herbal medicines have been in use since ancient times for general health and for specific diseases, particularly, in developing countries, great number of people depends on the traditional system of medicine for a variety of diseases. There are more than 35,000 plants species being used in various human cultures around the world for medicinal purposes. The World Health Organization reported that 80% of the world's population rely mainly on traditional medicine and a major part of the traditional therapies involve the use of plant extracts or their active constituents. The traditional heritage of India includes many time tested medicinal plants one of such important medicinal plant is *Tribulus terrestris* (Linn).

**KEYWORDS:** *T. terrestris*, aphrodisiac, anticancer agent, cardiac disorders, *In vivo* propagation.

**INTRODUCTION**

*Tribulus terrestris* (Linn). Belongs to family Zygophyllaceae and known as Puncture- vine, caltrop, yellow vine, goat head and devil's horn in English. Local names in India are Gokshura, Gukhukata(Assamese); Gokuri (Bengali); Be tha gokharu, Mithogokharu (Gujrati); Gokhru and Chota Gokhru (Hindi); Sannanaggilu, Neggilamullu, Neggilu (Kannada); Michirkand, Pakhda (Kashmiri); Kante gokaru (Marathi), Bhakhra, Gokhru (Punjabi); Goksharu, Traikantaka (Sanskrit); Nerinci (Tamil), Palleru Kayalu (Telugu). *Tribulus terrestris* L. is widespread in Mediterranean, subtropical and desert climates worldwide, but now widely distributed in warm regions of Europe, Asia, America, Africa, and Australia<sup>[1-4]</sup> (Adaikan *et al.*, 2000; Kostova and Dinchev; 2005;Verdu and Mas, 2006; Dinchev *et al.*, 2007;). The plant grows almost in all parts of India, ascending to 3,385 meters<sup>5</sup> (Mathur and Sundermoorthy, 2013).

**Taxonomical classification**

- Kingdom: Plantae
- Division: Phanerogams
- Subdivision: Angiospermae
- Class: Dicotyledonae
- Subclass: Polypetalae
- Series: Disciflorae
- Order: Giraniales
- Family: Zygophyllaceae
- Genus: *Tribulus*
- Species: *terrestris* Linn.

**Botanical description**

It is a perennial creeping herb and young shoots are silky-hairy. (Fig.-1)

**The stems** are often 30-70 cm long and simple or freely branched, prostrate forming flat patches. **The leaves** are opposite and unequal, pinnately compound paripinnate; the leaflets are usually in five to seven pairs, oblong with an oblique base 6-12 mm long with small stipules and solitary pedicles in the axil of smaller leaf of each pair.

**The flowers** Solitary, axillary or leaf opposed and yellow in colour. small 4 to 10 mm wide and contain five sepals that are about 2-6 mm long, and contains five petals, which are lemon yellow and about 2-12 mm long, flat and slightly longer than the sepals. The flower is attached to an annular disc, which is lobed. The flower consists often stamens and an ovary that has five cells, style short and stout. There are about three or four ovules in each cell.

**Fruits** are depressed schizocarp are globose, spinous or tuberculate; consisting of fine hairy or nearly glabrous, often muriculate and woody cocci, each with two pairs of hard sharp spines, one pair longer than the other. Fruit often cling to clothes and bodies of animals and humans.<sup>[6]</sup> Flowering and fruiting during April to August.

**Seeds**

The nutlets or "seeds" are hard and bear two to three sharp spines, 10 mm long and 4 to 6 mm broad point-to

point. It is a trailing and spreading herb, densely covered with minute hair.

Seeds are many in woody cocci.<sup>[6]</sup> The root system is a tap root.

### Chemical composition

*Tribulus terrestris* L. medicinally very important multipurpose herb recently lot of research is reported on its chemical composition. The presence of steroidal glycoside, steroidal saponin, flavanoids and alkaloids in the plant has been reported.<sup>[7]</sup> Saponin such as glucopyranosyl, galactopyrans, ruscogenin, hecogenin, gitogenin, titogenin, protodioscin, diosgenin and yamogenin<sup>[8,9]</sup> occurrence of saponins, flavonoids, alkaloids, lignanamides and cinammic acid amides has been reported in *T. terrestris*<sup>[10-14]</sup> sterols such as sitosterol, and campesterol; flavanoids such as kaemferol, kaempferol-3-glucoside, tribuloside and quercetin<sup>[15-18]</sup> and other constituents like fatty acids, polysaccharides, tannins, amino acids and potassium salts have been isolated from this plant. The green leaves, taken as a nutrient, contain 79.09% moisture, 7.22% protein, 1.55% calcium, 0.08 phosphorous, 9.22 mg iron/100g and 41.5 mg vitamin C. A good number of various saponins and their different derivatives have also been identified in *T. terrestris*, of which diosgenin, gitogenin and chlorogenin are in the leaf tissue. The presence of spirostanol and furostanol saponins is a characteristic feature of this plant, the latter being considered to be biogenetic precursors of their spiro analogs.<sup>[18]</sup> The extract of the powdered root contains an alkaloid, resins, nitrate and fixed oils. The roots also contain campesterol, p-setosterol, stigmaterol and neotegenin. The fruit contains 3.5-5.05 of semidrying oil, peroxidase, and diastase, traces of a glycoside resin, proteins and inorganic matter. The fruit and root of *T. terrestris* contains pharmacologically important metabolites such as phytosteroids, flavonoids, alkaloids and glycosides.<sup>[18]</sup> The major constituents of the fruits of this plant are steroidal saponins named terrestrosins A,B,C,D and E, desgalacto-tigonis, F-gitonis, desglucolanatigoneis, gitin which on hydrolysis yield diosgenins, hecogenins and neotigogenin.<sup>[19,20]</sup> The fruits also contain chlorogenin, diosgenin, gitogenin, rutin and rahmnose. The stems are rich in starch, fructose and sucrose<sup>[2,10-12,20A,21-27]</sup> p-carboline alkaloids like norharman, harman, harmol, harmine, 3- hydroxymethyl-norharman, harmalol, harmaline, tetrahydronorharman, tetrahydroharman, 2-ethyltetrahydro-norharman, Tribuhisl Qune; and amide such as terrestriamide, terretribisamide. N-p-coumaroyltryamine and cinnamic amide were also isolated from this plant.<sup>[11, 23, 28-30]</sup> Two alkaloids Harman and harmol are also reported from the whole plant.<sup>[31]</sup>

### Uses

*Tribulus terrestris* L. is a famous herb traditionally used by different cultures for a number of conditions. In India and China, the medicinal use of this herb is traced 5,000 year back.<sup>[32, 33]</sup> In India, the fruits have been long used

as a tonic and calculus infections, urinary discharges; it has a 5,000 year-old history of medicinal use in India. Traditionally it has been used for boosting hormone production in men and women. It is recommended as a diuretic and against kidney diseases and gravel. *T. terrestris* extract was is used as a tonic to treat sexual dysfunction. It is an important constituent of various medicinal preparations worldwide like Dashmularishta and Tribestan.<sup>[34]</sup> The fruits of *T. terrestris* L. have been used in traditional Chinese medicine for the treatment of eye trouble, edema, abdominal distention, emission, morbid leucorrhea, sexual dysfunction and veiling. Roots and fruits are useful in rheumatism, piles, renal and vesical calculi, menorrhagia, impotency, premature ejaculation. It is indicated for use in treating headaches, dizziness, premature ejaculation and spermatorrhoea (escape of sperm without orgasm).<sup>[35]</sup> In Ayurveda, the herb is known for its anti-urolithiatic, diuretic and aphrodisiac properties<sup>[36]</sup> and also to treat impotence, venereal diseases, and sexual debility. The fruits have been applied in the traditional Chinese medicine for treatment of eye trouble, edema, abdominal distention, emission, antifungal and antibacterial activities, morbid leucorrhea, sexual dysfunction and physical performance in men.<sup>[8,21,25,37-50]</sup> In the Shem-Nong Pharmacopoeia (the oldest known pharmacological work in China) *T. terrestris* is described as a highly valuable drug to restore the depressed liver, for treatment of fullness in the chest and mastitis and also used to dispel the wind and clear the eyes, for treatment of acute conjunctivitis, headache and vitiligo.<sup>[21]</sup> In South Africa it is used medicinally as a tonic for diarrhea and diseases of the throat and the eyes.<sup>[51]</sup> In the Bulgarian folk medicine *T. terrestris* is recommended for purification of blood and hemorrhoids<sup>[2]</sup> Crude saponin fraction of this plant has shown significant effects in the treatment of various cardiac diseases including hypertension, coronary heart disease, myocardial infarction, cerebral arteriosclerosis, and thrombosis.<sup>[20]</sup>

### Use in Diabetic Mellitus

Protective effects of *T. terrestris* was investigated the in diabetes mellitus<sup>[52]</sup> and based on their investigation suggested that the protective effect of *T. terrestris* on streptozotocin-induced diabetes in rats may be mediated by inhibiting oxidative stress. It is reported that extract *T. terrestris* consisting of saponins appear to decrease blood sugar levels by acting on alphaglucosidase in small intestines of rats.<sup>[53]</sup>

### Uses in cardiac disorders

Wang *et al.*,<sup>[13]</sup> conducted clinical trial in 406 patients with coronary heart disease. They were treated with saponins of *T. terrestris*. The results showed that the total efficacious rate of remission angina pectoris was 82.3% it is shown that saponin of *T. terrestris* has the action of dilating coronary artery and improving coronary circulation. No adverse side effects were noted. Cai *et al.*<sup>[25]</sup> noted that a saponin of *T. terrestris* has action of dilating coronary artery and improving coronary

circulation, so recommended for treating angina pectoris. They further reported that the Chinese drug named 'Xinnao Shutong' is made of crude saponins of *T. terrestris*, which has significant effect in the treatment of coronary disease, myocardial infarction and cerebral diseases. Zhang *et al.*<sup>[54]</sup> evaluated the protective effect of tribulosin from *T. terrestris* against cardiac ischemia/reperfusion injury in rats. They observed that Tribulosin protected myocardium against ischemia/reperfusion injury through protein kinase C epsilon activation. *T. terrestris* also appears to protect the heart cells and may even improve the heart function following a heart attack.<sup>[55]</sup>

#### Uses as anticancer agent

Inhibiting cell growth is a critical action of anticancer drug-induced cancer death. It is reported that *T. terrestris* extracts exhibits weak cytotoxic effects to normal cells compared to cancer cells.<sup>[56, 57]</sup> The observed biological activities of *T. terrestris* are mainly due to its abundant steroidal saponins.<sup>[26,58-60]</sup>

Zhong Yao Cai<sup>[61]</sup> studied the effect of *T. terrestris* and opined that the saponins present in *T. terrestris* have inhibitory effect on breast cancer cell line. Aqueous extract of *T. terrestris* has reduced tumor incidence and number of papillomas in mice by decreased lipid peroxidation levels and increased glutathione levels in the liver.<sup>[62]</sup> Ivanova *et al.*<sup>[63]</sup> showed evidence for antiproliferative effect of *T. terrestris* saponins on mouse carcinoma. It is reported that aqueous extract of *T. terrestris* induces cell growth arrest and apoptosis by down-regulating NF- $\kappa$ B signaling in liver cancer cells.<sup>[56]</sup> Antiproliferative activity of this plant on breast cancer cells is reported.<sup>[64]</sup> It is reported that the methanolic extract of *T. terrestris* fruits showed 50% inhibitory concentration (IC<sub>50</sub>) at 380 and 420  $\mu$ g/ml, for Dalton's Lymphoma Ascites (DLA) and Ehrlich's Ascites Carcinoma (EAC) cells, respectively, the ascites tumor induced by EAC cells was found to be decreased considerably by the oral administration of the extract and the life span of the tumour bearing mice was enhanced to 31 and 45% by 100 and 250 mg/kg b. wt. extract, respectively.<sup>[65]</sup> Wei *et al.*<sup>[66]</sup> reported that terrestrosin D a steroidal saponin from *T. terrestris* inhibits growth and angiogenesis of human prostate cancer in vitro and in vivo. Etanolic extract of the fruits of *T. terrestris* revealed strongest anticancer activity against cervical cancer cell line (HeLa).<sup>[67]</sup>

#### Uses as anti-infertility agent

Since centuries medicinal plant extracts, decoctions, fractions are being used in the treatment of a dysfunctioning of the libido, sexual asthenia, erection, and sperm disorders. Pharmacological activities of many of these plants have been shown in vitro using cells, on laboratory animals and human studies. Extracts of plants like *Acanthopanax senticosus*, *Andrographis paniculata*, *Asparagus racemosus*, *Astragalus membranaceus*, *Lepidium meyenii* *Panax ginseng*, *Panax quinquefolius*

and have shown positive effects on sexual desire; while extracts of *Withania somnifera*, on improved sperm parameters.<sup>[68]</sup> In Ayurved, Gokshur has been indicated in maintain pregnancy (Garbhasthapana).<sup>[69]</sup> In indigenous medicine, *Tribulus* has long been in use for different ailments, particularly, the fruits are extensively used since ancient times as aphrodisiac.<sup>[70, 71]</sup> It is reported to improve sexual function possibly through increase in the free serum testosterone.<sup>[72]</sup> In their respective studies<sup>[73, 74]</sup> observed that the administration of tribestans a commercial product containing 250 mg of *T. terrestris* to humans and animals for a period of 60–90 days was found to improve testosterone levels, libido and promote spermatogenesis. Based on their studies in primates, rabbit and rat it is suggested that *T. terrestris* may be useful in mild to moderate cases of erectile dysfunction (ED) as it increases some of the sex hormones, possibly due to the presence of protodioscin in the extract.<sup>[75]</sup> The extracts of *Tribulus terrestris* (5 mg/kg for eight weeks of treatment) stimulated rat libido (sexual desire).<sup>[76]</sup> Extract *T. terrestris* at the dose of 5, 7.5 and 10 mg/kg increased testosterone, dihydrotestosterone and dehydroepiandrosterone sulphate levels in primates (in acute treatment), rabbits and castrated rats after eight weeks of treatment.<sup>[77]</sup> Three months treatment of patients suffering from idiopathic infertility (sperm concentration between 5 and 15 million/ml) by a formulation of plants made up of *T. terrestris*, *Asparagus racemosus* and *Withania somnifera* increased semen volume, sperm count and motility.<sup>[78]</sup> Sudev and Suresh<sup>[79]</sup> reported that Gokshur (*T. terrestris*) and its formulations have shown improved sperm count and motility. Sellandi, *et al.*<sup>[80]</sup> conducted studies on eligible subjects between the age of 21 and 50 years, with a complaint of *Kshina Shukra* (Oligozoospermia). In this study they administered either *Gokshura* granules or placebo granules for 60 days. Subjects receiving *Gokshura* granules showed 78.11% improvement in *Rogi bala* (*Agni bala*, *Deha bala*, *Satva bala*, and the Quality of Sexual Health) and *Rogabala* (Semen Analysis) and concluded that the *Gokshura* granules have shown superior results in the management of Oligozoospermia, as compared to the placebo granules. *T. terrestris* contains three groups of active phytochemicals: Dioscin, protodioscin, diosgenin and similar. These substances have effect on sexual performance and may treat various sexual disorders, they regulate sexual energy level and strength by increasing the percentage of free testosterone level for men and they affect pregnenolone, progesterone and estrogen.<sup>[81]</sup>

#### In vitro propagation

Keeping in view of its various important applications in the indigenous systems of medicine, and overexploitation of this species may cause a serious threat to its existence. And also due to its poor rate seed germination (25%) Many scientists have felt that rapid in vitro propagation of this important drug yielding plant has become imperative. There are several reports of in vitro

propagation of this plant. Ali *et al.*<sup>[82]</sup> reported that cotyledonary leaves along with epicotyl segment from young seedlings when inoculated on MS medium supplemented with 0.2 mg/l NAA, 0.5 mg/l BAP 50 mg/l glutamine induced 100% shoot and root differentiation in 10 weeks. It is reported that on woody plant medium supplemented with 4mg/l BA they could obtain 6-7 micro shoots per nodal explants after 4 weeks of culture, further rooting of these shoots is achieved on MS medium without any growth regulators, and found 80% survival rate in the field conditions.<sup>[83]</sup> Khude<sup>[84]</sup> reported that among leaf, shoot tip, axillary bud, and petiole axillary bud when inoculated on MS medium supplemented with BAP 2.0mg/l and IAA 0.5 mg/l induced about 15 numbers of shoots per explants. Efficient shoot regeneration (22%) was noticed when epicotyl explants were cultured on MS media supplemented with 0.1 mg/l NAA + 2 mg/l BAP or 0.4 mg/l NAA + 0.5 mg/l BAP within 14 days of culture, further it is reported that maximum indirect shoot regeneration (28.4%) is noticed, when green-yellowish calli derived from hypocotyl explants, is cultured on MS medium supplemented with 0.4 mg/l NAA + 2 mg/l BAP further ,rooting of the in vitro raised plantlets is achieved on MS medium supplemented with 2 mg/l IBA.<sup>[85]</sup> Direct regeneration of shoots is reported from nodal explants, cultured on MS medium supplemented with 3.0 mg l-1 benzyl amino purine (BAP) and 2.5 mg l-1 naphthalene acetic acid (NAA) and rooting of theses plantlets is achieved on 0.1mg/l IBA.<sup>[86]</sup>



**Figure-1 A. Flowering twig of *Tribulus terrestris*. B Fruits of *Tribulus terrestris*.**

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