GLYCYRRHIZA GLABRA L. A MIRACLE MEDICINAL HERB

Yogesh Badkhane\textsuperscript{1*}, A.S. Yadav\textsuperscript{2}, A. Bajaj\textsuperscript{1}, Ajit K. Sharma\textsuperscript{3}, D. K. Raghuwanshi\textsuperscript{1}

\textsuperscript{1}Molecular Biology and Seed Technology Laboratory, Dept. of Botany, Govt. MotilalVigyanMahavidyalaya, Bhopal-08.
\textsuperscript{2}Higher Education Dept., Govt. of Madhya Pradesh, Bhopal, India.
\textsuperscript{3}The Environmental Planning & Coordination Organisation(EPCO) Bhopal, India.

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ABSTRACT

\textit{Glycyrrhiza glabra} Linn. commonly known as “Licorice”, “Mulethi” in Hindi, is an important medicinal plant. Its roots and rhizomes are important in drug development with various pharmacological activities. Licorice is used traditionally both as a flavouring agent in confectionery and medicine in tropical countries especially in India, China and hot temperate regions. Licorice has also been shown to have memory enhancing properties, antidiabetic, antioxidant, anti-inflammatory, antibacterial activities. Hence, since in traditional medicines as an antiallergic, demulcent, emollient, as a fungicide since ancient time. It is also prescribed for the treatment of many diseases, such as addison’s disease, coughs, asthma, arthritis, bronchitis, peptic ulcer and for allergic complaints. Glycyrrhizin and glycyrrhizic acid reported from Licorice, have remarkable biological activities. This article presents comprehensive analyzed information on the botanical, chemical, traditional use and medicinal aspects of \textit{Glycyrrhiza glabra}, additionaly tissue culture studies of \textit{G. glabra} have been focused upon in this paper.

Corresponding author
Yogesh Badkhane
Senior Research Fellow, Biotechnology,
Dept. of Botany, Govt. Motilal Vigyan Mahavidyalaya,
Bhopal, (M.P.) India.
badhkane.yogesh@rediffmail.com

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INTRODUCTION

*Glycyrrhiza glabra* Linn. belongs to the family Leguminosae, is a genus of perennial herbs and under shrubs distributed in the subtropical and warm temperate regions of the world. *Glycyrrhiza glabra* Linn. commonly known as liquorice and sweet wood in English, Jothi-madhu, Mulethi in Hindi, Yasht-madhu, Madhuka in Sanskrit, Jashtimadhu, Jaishbomodhu in Bengali, Atimaduramu, Yashtimadikam in Telugu, Jethimadhu in Gujarati and Atimaduran in Tamil (Chopra *et al.* 2002). Licorice extracts and its principle component, glycyrrhizin, have intensive use in foods, tobacco products and in traditional use and herbal medicine. It is cultivated for its rhizomes (underground stems) that contain the compound glycyrrhizin, which is 50 times more sweetener than sugar. It is cultivated in the Mediterranean basin of Africa, in southern Europe, and in India (*The Indian pharmaceutical codex*, 1953., African pharmacopoeia, 1985., Ghazanfar, 1994., Chin *et al.*, 1992) widely cultivated in Punjab and sub Himalaya tracts (Dhuke *et al.*, 2002), Baramulla, Srinagar, Jammu, Dehradun, Delhi and South India (Meena *et al.*, 2010).

Botanical description

Licorice is an herb or under shrub, perennial. Stem 50-150 cm tall, woody at base, densely scaly glandular punctate, white hairy. Leaves 5-14 cm, 11-17-foliate; stipules caducous, linear, 1-2 mm; petiole densely yellow-brown glandular hairy and villous; leaflets ovate-oblong, oblong-lanceolate, or elliptic, 1.7-4 × 0.8-2 cm, abaxially densely yellow scaly glandular punctate and pubescent on veins, adaxially glabrescent or pilose, base rounded, apex rounded or retuse and with mucro. Erect grows to about 2m height. Its oval leaflets, flat pods and purplish white flower clusters look very beautiful. Pods 1 cm long, flat pods oblong to linear, 1–3 cm long by 6mm wide, more or less densely echinate glandular, many seeded or abbreviated 2- or 3-seeded. (*The Indian pharmaceutical codex*, 1953. African pharmacopoeia, 1985.). Seeds 2-8, dark green, ca. 2 mm in diam., smooth. Flower. May-Jun, fruit. Jul-Sep. 2n = 16* (Flora of China, Flora of Pan Himalayas, 1964). It has extensive root system and its fibrous main taproot with bright yellow interior is used for medicinal purpose (*Wealth of India*, 1985) (Olukoga & Donaldson, 1998). The commercial name of the dried rhizome and root of the plant is liquorice (*Medicinal Plants*, 1998). Roots and Rhizome are the officinal part, which are nearly cylindrical pieces, up to 1m long and 5–20 mm in diameter.Externally, the bark is brownish grey to dark brown, longitudinally wrinkled, occasionally bearing small dark buds in rhizomes or small circular or transverse rootlet-scar in roots. The roots are long, cylindrical, thick and multibranched (African pharmacopoeia, 1985. European pharmacopoeia, 2nd edition, 1995., Akao *et al.* 1991). The dried, peeled or unpeeled underground stems and roots constitute the drug known in the trade as liquorice (Disabled World).

Taxonomic classification

- **Class:** Equisetopsida
- **Subclass:** Magnoliidae
- **Superorder:** Rosanae
- **Order:** Fabales
- **Family:** Leguminosae/ Fabaceae - Papilionoideae
- **Genus:** *Glycyrrhiza*
- **Species:** *glabra* Linn.

Chemical constituents:

A high number (More than 400) of chemical compounds have been isolated from *Glycyrrhiza* species, flavonoids (liquiritiside, isoliquiritiside) are believed to be responsible for the bioactivities of licorice. The contents of these saponins and flavonoids may vary significantly due to different plant species and geographic sources (Zhang and Ye, 2009). polysaccharides, pectins, Steroid hormones, sacharose, glucose, amino acids, mineral salts, (Obolentseva *et al.*, 1999) Triterpenoid saponins Glycyrrhizin (2-15%) also known as Glycyrrhizinic acid, present in the form of calcium and potassium salts (Bradley, 1992). Glycyrrhetinic acid (18-beta-glycyrrhetinic acid, GA), bitter principle (glycymarin), Steroid hormone, Resinous oil, starch, glucose, asparagin, manitol, atropine, choline, betaine, progesterone, steroids, tannins etc. (Snow, 1996; Fukai *et al.*, 1998; Arystanova *et al.*, 2001. Mills 1991, mills *et al* 2000, schulze *et al* 2001, weiss *et al* 2000). The major component in the roots and stolons of Glycyrrhiza plants that imparts a sweet flavor glycyrrhizin is an oleanane-type triterpene saponin. This compound represents a mixture of potassium-calcium-magnesium salts of glycyrrhizic acid that varies within a 2-25 % range. The natural saponins, glycyrrhizic acid is a molecule composed of a hydrophilic part, two molecules of glucuronic acid, and fragment, glycyrrhetic acid (Obolentseva GV. 1999).

Structure of major chemical constituents:

The structure of major chemical constituents of *Glycyrrhiza glabra* L. has been shown in (Fig. 1)

Traditional uses:

*Glycyrrhiza glabra* Linn. has been widely used in folk medicine and is today a highly commercially important target species. In the traditional system of medicine, the roots and rhizomes of *G. glabra* as an anti-inflammatory agent in the treatment of allergic reactions (Hikino, 1985), antimicrobial, antilucer, expectorant and anxiolytic activities (Wang & Han, 1993; Asl & Hosseinzadeh, 2008). It has also been known to relieve rheumatism, osteoarthritis and arthritis, regulate low blood sugar, and was used for Addison's disease. It finds applications in arthritis and mouth ulcers. (*Wealth of India*, 1985., Bradley 1992). It has also been used as antiallergic, demulcent, emollient, fungicide (Dhuke *et al.*, 2002), respiratory, gastrointestinal, cardiovascular, eye, and skin disorders and for their
antiviral effects (Zhang, Ye M. 2009). The root extract produces mild estrogenic effects, and it has proven to be useful for some in treating symptoms of menopause, relieving menstrual cramps and regulating menstruation. The roots and rhizomes of *Glycyrrhiza glabra* has been in clinical use for centuries to treat liver diseases and is a major component of polyherbal formulations for the cure of hepatotoxicity (Rajesh *et al*., 2000); antiallergic, demulcent, emollient, fungicide, peptic ulcer (Dhuke *et al*. 2002), to prevent liver for its toxicity and to treat tuberculosis and adrenocorticoid insufficiency. (Bradley 1992; Schambelan 1994)

**Medicinal Uses**:

A medicinal use of licorice includes cough suppression, treatment of early Addison disease, treatment of liver disease and dyspepsia also in the prophylaxis and treatment of gastric and duodenal ulcers (African pharmacopoeia, 1985. The wealth of india, 1995, Hikino 1985., Bruneton 1995 ). Licorice is very sweet, moist and soothing herb is an anti-inflammatory, used in treatment of arthritis and mouth ulcers (Chevallier, 1996). Demulcent for sore throats, expectorant in the treatment of coughs and much used in cough medicines bronchial catarrh. Externally, the root is used for herpes, eczema and shingles and Addison’s disease, asthma, bronchitis, arthritis, and allergic complaints (Brown, 1995). Licorice extracts have been used for more than 60 years in Japan to treat chronic hepatitis, and therapeutic benefit against viruses, Glycyrrhizin and glycyrrhizic acid have been shown to inhibit growth and cytopathology of numerous RNA and DNA viruses (Hattori *et al*. 1989, Ito *et al*. 1988), a preliminary report which covers the isolation of licopyranocoumar and ant-HIV activity of licorice phenolics, inhibit the HIV-induced giant cell formation for molt-4 cells (Hatano *et al*., 1988), a major component of Glycyrrhiza, which exhibited higher anti-UV activity (SI=20.6) (Kato *et al*. 2012), including hepatitis A (Crance *et al*., 1990) and C (Van *et al*., 1988), Herpes simplex (Pompei *et al*., 1979. Partridge *et al*., 1984) and CMV (Numazaki *et al*., 1994). Licorice preparations have been used to sooth and heal skin eruptions, like skin diseases and herpetic lesions. The root is antisipasmotic, demulcent, diuretic, emollient, expectorant, laxative, pectoral, tonic (Grieve, 1984; Launert, 1981; Lust, 1983; Uphof, 1959; Mills, 1993; Stuart). The root has also been shown to have a hormonal effect similar to the ovarian hormone (Chiej, 1984).

### Food and Agriculture Organization of the United Nations (FAO)

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**Liquorice root in cough medicines:**

Most cough medicines are prepared to treat either dry coughs or "wet" coughs, usually known as productive coughs. Licorice root with glycyrrhizin treats the latter, working as an expectorant to loosen and thin mucus, thus making the productive cough more. The tea form of licorice root is recommended for coughs, but syrup is also available. Pure form of licorice is fifty times sweeter than sugar (Hikino, 1985). Its sweetness property that has resulted in its use around the world in cough syrups and lozenges as well as candy extensively used in cough drops and syrups(Leung *et al*., 1996).

Licorice is used to make a wonderfully soothing honey cough syrup, demulcent that will help break up mucus settled in your lungs, and it is an expectorant, it will help expel the mucus as well. Licorices have an expectorant effect, increase the secretion of the bronchial glands and make it an effective remedy for these ailments. Glycyrrhizin promotes the production of mucus in the respiratory tract. It may make the respiratory tract mucus less sticky and may also promote its removal from the body.

**Memory enhancing properties:**

Several medicinal plants used in ayurvedic polyherbal formulations for curing the dementia and so many medicinal plants showing the memory enhancing property under several researcher studies in the current trend one such plant is *G. glabra*. The roots and rhizomes of *Glycyrrhiza glabra* is an efficient brain tonic it increases the circulation into the CNS system and balance the sugar levels in the blood (Rathee *et al*., 2008). Liquorice shows significantly improved learning and memory on scopolamine induced dementia. Oxygen free radicals and other products of oxidative metabolism have been shown to neurotoxic (Sayre *et al*., 1997). The elevated plus-maze and passive avoidance paradigm were employed to evaluate learning and memory parameters. Aqueous extract of *G. glabra* were administered for 7 successive days in separate groups and observed in mice. (Parle 2004). The protective effect of liquorice extract may be attributed to its antioxidant property by virtue of which susceptible brain cells get exposed to less oxidative stress resulting in reduced brain damage and improved neuronal function thereby enhancing the memory (Dhingra *et al*., 2004).

Learning and memory enhancing activity of *Glycyrrhiza glabra* root extract affects was observed on in three months old male Wistar albino rats. The aqueous extract of root of *Glycyrrhiza glabra* was administered orally in three doses (75, 150 and 300 mg/kg)
for 4 weeks. Elevated plus-maze and Morris water maze tests were conducted to evaluate the learning and memory parameters and served as the exteroceptive behavioral models. (Chakravarthi et al. 2012). The roots and rhizomes of *Glycyrrhiza glabra* is an efficient brain tonic it increases the circulation into the CNS system and balance the sugar levels in the blood (Rathee et al., 2008) significant action an memory enhancing activity as dementia disorder (Dhingra et al., 2004).

**Glycyrrhiza glabra** Linn. *as an Antioxidant:*

Antioxidant may offer resistance against the oxidative stress by scavenging free radicals, inhibiting lipid peroxidation and by many other mechanisms and thus prevent disease, and today widely used as free radicals inhibitors in food for maintaining the freshness, flavor and odor for a longer period. (Panchawat et al., 2010). The roots of *Glycyrrhiza glabra* is a potential source of antioxidants and urease inhibitors. (Mehreen et al. 2012) Extract of *G.glabra* was tested by studying the inhibition of radiation induced lipid peroxidation in rat liver microsomes. Chemical constituents are glycyrrhizin, flavones, coumarins. It shows its activity through free radical scavenging property (Naik & Satav, 2003). The relative reducing activity in terms of antioxidant activity of extracts was determined by using individual extract (15mg) as well as its combination with equal amount of ascorbic acid (Ashawat et al. 2007). Ethanol extract of *G. glabra* possess considerable antioxidant activity and protective effect against the human lipoprotein oxidative system using *in vitro* models (Visavadiya et al. 2009).

### Economic Importance and Conservation status:

There are number of popular traded products of *Glycyrrhiza glabra* L. which are widely accepted throughout the global market are as follows: Abana (HeartCare), Diabecon (GlucoCare), Diakof (CoughCare Sfree), Geriforte (GeritCare / StressCare), Herbolax (Laxa Care), Himcocid, Koflet (Cough Care), Menosan, Septilin (ImmunoCare), Septilin syrup, Rumalaya forte, Anti-Wrinkle Cream, Baby Lotion, Geriforte Aqua, Geriforte Vet, Himpyrin, Himpyrin Vet, Regurin, Yashi-madhu (Himalaya Herbal Healthcare, India), Lioele Magic Lip treatment, Lioele Cosmetic Co., Ltd. [Korea], Mandarin O2 Foaming Cleanser, H&H Co., Ltd. [Korea], Licorice Flavonoids, Henan Bis-Biotech Co., Ltd [China], Satibo Capsule(Arabic), Shanghai Cynking Trading Co., Ltd. [China], Pure Intensive GEL AF5000 Serum, KB Cosmetics [Korea], AV Free Clean Foam Cleanser, CU SKIN Co., Ltd. [Korea], MX7 Return To Control Skin Essence, MBention Co., Ltd. [Korea], Licorice Root Extract, Copalyton Biotechnology Co., Ltd. [China], NanoVital VITANICS Skin Toner, Vitacos Corporation [Korea], Flodol 30 Patches, Alta Care Laboratories [Italy], ACIGARD Tablet, Bliss Ayurveda India Private Limited [India], VADO (Dietary Supplement, Herbal Medicine, Herbs), Promopharma S.R.L. [Italy], Foldex Cream. Purex Health Care Pharmaceuticals [Egypt], Ginseng tea, Covic International [Korea], Germall Free Preservative, Naturogin Inc. [Korea], Clopidogrel Bisulphate, Shreeji Pharma International [India].

A report of the task force, Planning Commission, New Delhi on conservation and sustainable use of medicinal plants listed *Glycyrrhiza glabra* L. under list of major plants required by Indian pharmaceutical industries. The demand of this plant was estimated 5000 tonnes per annum is being largely met through imports from Pakistan, Iran and Afghanistan, which need support for indigenous production. There is no concerted effort has yet been made for conservation, careful cultivation and harvesting of this plants so as to replenish the exhausting supply. Though the plant has immense medicinal value, high demand it is gradually declining from the nature due to over exploitation, environmental pollution and lack of indigenous production.

That is why there is an urgent need of replenishment of the short supply and conservation of this exotic plant genetic resource. Generally *Glycyrrhiza glabra* L. are multiplied vegetatively by means of cuttings, but this process is slow in response and need to maintain large number of mother plants stock. From seeds plants are rather slow to grow (Brown, 1995).

Thus, large scale propagation through tissue culture technology could be possible option to ensure the availability of quality planting material for large scale cultivation and to meet the ever growing pharmaceutical demand for this plant.

### Cultivation and Conservation measurements:

**Conventional:**

Licorice is a cultivated crop in many places that grows slowly from seed (generally from 2 to 6 feet in height), and difficult to eradicate when well established. Licorice is a now very important commercial crop that is a mainstay in herbal medicine, as well as a flavoring for confectionery, snuff, foods, cough syrups and tobacco products. Propagation of *Glycyrrhiza glabra* by traditional methods using old crowns of lifted roots of the old plants. They are cut into 15 cm long. The plants can be made from the runners which are cut into 5 inches pieces with 2 to 3 buds. They are kept in moist moss for about 10 to 12 days in the shade then its starts to root. The same are planted to the fields. The *Glycyrrhiza glabra* plants are uprooted in the month of September after the rains. The material is then cut into 15-20cm long pieces and dried. It is cultivated in Russia, UK, USA, Italy, France, Germany, Spain, China and Northern India (Punjab and Sub-Himalayan tracts). Large scale commercial cultivation is seen in other countries i.e. Spain, Sicily and England (Chopra and Chopra, 1958).

**Micropropagation studies:**

Micropropagation has a great commercial potential due to the speed of propagation, slashed production area demand and therefore the ability to multiply elite clones exhibiting superior growth and enhanced stress tolerance (Garton and Mosses, 1985; Kane et al., 1989). Micropropagation also can be used to establish and maintain disease free and virus free plant stock. This is done by culturing by the meristematic part of the plant, which typically is not virus infected. Once plants are developed from the apical meristem of the plant, they can be maintained and sold as virus free plants. Micropropagation can be achieved by using different parts...
of the plant as the primary ‘explant’ such as, the apical meristem, nodal bud, shoot buds, axillary buds, leaf segment or through production of somatic embryos, a process commonly known as somatic embryogenesis.

Shoot Multiplication:

Micropropagation of *Glycyrrhiza glabra* L. aims at two things: production of large number of plantlets and propagation of the selected genotypes without inducing any genetic variation. A preliminary report in vitro micropropagation of *Glycyrrhiza glabra* L. was published as early as 1980 by Shah and Dalal. A lot of research work has been done for its micropropagation by shoot tip and nodal culture. Thengane et al. (1998) described a stepwise protocol for the micropropagation of *Glycyrrhiza glabra* L. on a simple minimal medium using shoot tip and nodal explants where very high multiplication rates with healthy root system. They observed enhanced multiplication ratio 1:10 in murashige and skoogs medium fortified with N6 benzyladenine (BA, 0.88-8.88 um). In vitro regeneration was achieved in loricance using nodal segments as an explants and recorded cent per cent multiplication on MS medium supplemented with BAP (0.5 mg/l) and NAA (0.05 mg/l). Maximum number of shoots/explant was produced on MS medium containing higher BAP (2.0 mg/l) level combined with NAA (0.05 mg/l). In vitro rooting was found better on full strength MS medium supplemented with IBA (0.5 - 1.0 mg/l) (Patel et al. 2007).

Sharma et al. (2008) reported cost effective micropropagation protocol in selected plants for the conservation *Artemisia annua* L., *Glycyrrhiza glabra* L., *Bacopa monnieri* L. and Shoot tips, axillary buds and young leaf was used as explants. They studied the effect of cost effective alternates such as Nutrients, Carbon source, tap water, growth regulators and in vitro hardening under ordinary conditions like natural light and temperature.

Yadav and Singh (2012) reported one protocol on micropropagation by manipulating growth regulators, culture conditions and other external factors influencing in vitro multiplication of *G. glabra*. The highest bud-break (86.6%) with the longest shoot length (8.0 cm) and maximum number of shoots (3.0) was obtained when middle order nodes (3rd to 5th node from apex) collected between May to August were inoculated on MS medium supplemented with 6-Benzylaminopurine (2.0 mg/l) + α-naphthalene acetic acid (0.5 mg/l) under photoperiod of 16/8 h (light/dark cycle). Shrivastava et al. (2013) studied in conserving shoot apices of *Glycyrrhiza glabra* L. under slow growth conditions. Slow growth cultures were obtained by lowering incubation temperature and light intensity. A very low concentration of growth hormones (BAP and IAA) added to the conservation medium was beneficial for complete retrieval of the shoots. Very low concentration of BA (0.1 mgL-1) and IAA (0.05 mgL-1) was found beneficial for retrieval of the conserved shoots. Different combinations of osmotic agents (sucrose, sorbitol and mannitol), used for increasing the subculture period, 20 gm/l of mannitol suited best for slow growth conservation with only subculture in a year (Shrivastava et al., 2013).

Somatic Embryogenesis:

Somatic embryogenesis involves a process of development of embryos in callus cultures derived from the somatic explant in vitro and their subsequent development to full plants. Little information is available on the application of micropropagation technique for multiplication of *Glycyrrhiza glabra* L. An experiment was conducted by the base of auxins such as NAA and 2,4-D, 6-BA as cytokinins plant growing regulators in rang of 0.5 - 2.0 mg/l in a MS medium, well grown callus induced from explants were selected to transfer to MS medium with appropriate hormones for subculture. No callus were induced but When 2,4-D was used, all explants formed white callus and induction rates increased with the increase of concentration of 2,4-D. The maximum induction rate was recorded as 96 % in MS medium with 0.5 mg/l 2,4-D and 2 mg/l 6-BA with a light compact structure (Ali Parsaemehr and Badrossadat mousavi, 2009). Fu et al (2010) optimized embryogenic callus and embryogenesis and observed that the explants of hypocotyl give the highest calli formation frequency of 93.3% on Murashige and Skoog (MS) medium containing 2.0 mg/L 6-benzylaminopurine (6-BA) and 0.5 mg/L 2,4- dichlorophenoxyacetic acid (2,4-D). The maximum efficiency of embryo were obtained on MS medium with 0.5 mg/L 6-BA + 0.5 mg/L kinetin zeatin (KT) + 0.1 mg/L indole-3-butyric acid (IBA); the embryos could develop further on medium with 1000 mg/L malt extract (ME).

Molecular Marker Studies:

Molecular tools are more reliable than phenotypic observation for evaluating tissue culture induced variations (Leroy et al., 2000). In plants regenerated via somatic embryogenesis, the quality of somatic embryos determines the production of true-to-type plants. Many authors have reported that dedifferentiation of plant tissues leads to genetic modifications (Taylor et al., 1995; Hashmi et al., 1997; Rani et al., 2000), but on the contrary, several reports also confirmed genetic integrity of tissue culture derived plants (Dale et al., 1981; Haydu and Vasil, 1981; Jayanthi and Mandal, 2001; Gagliardi et al., 2004). ISSR and RAPD based evaluation of genetic stability is reported encapsulated micro shoot of *Glycyrrhiza glabra* L. after 6 month storage (Mehrotra et al. 2012). An experiment was conducted by Shirazi et al. (2012) for Glycyrrhizin and isoliquiritigenin production by hairy root culture of *Glycyrrhiza glabra* L. by hairy root culture. Hairy roots was induced by inoculation of the leaf and stem explants with *A. rhizogenes* strain AR15834 and reported hairy root cultures of *G. glabra* realized leading to the production of bioactive compound as glycyrrhizin and isoliquiritigenin.

CONCLUSION

The objective of this review was to indicate the recent advances within the exploration of licorice as an example its potential as a therapeutic agent. With the present information, it is evident that *Glycyrrhiza glabra* L. has pharmacological functions as well as antiviral, antibacterial drug, antifungal and inhibitor activities, among others. As the current information shows, it is also possible that Licorice might be useful in the development of new drugs for the treatment of various diseases. However, the present results suggest a possibility that Licorice can be further developed as a potential disease-curing remedy. This review emphasizes the potential
of Glycyrrhiza glabra to be employed in new therapeutic drugs and provide the basis for future research on the application of transitional medicinal plants. Furthermore, optimization of standard protocol for seed production of Glycyrrhiza glabra L. would enhance the quality and quantity of elite seeds material. This in turn would encourage farmers to undertake commercial cultivation of Glycyrrhiza glabra L. thus curbing the overexploitation of this plant in the wild and thereby complements the conservation process.

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Figure: Glycyrrhizin

Licochalcone C.

18-beta-glycyrrhetinic acid.

Fig. 1 Structure of major chemical constituents of Glycyrrhiza glabra L.
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