

Glycyrrhiza glabra (Licorice) in Turkmenistan: Medicinal and Biological Aspects



Svetlana A. Pleskanovskaya, Maya A. Mamedova, Mehri A. Ashiraliyeva,
Volkan Altay, and Munir Ozturk

Introduction

Turkmenistan (Fig. 1) is spread over a vast area of deserts of Turan, Karakum, Caspian, and Sundukli on the right bank of Amu Darya river. In the north lie Kopetdag-Khorasan mountains, and low ranges of Paropamisus and Gissar Mountains. Almost all territory of the country is located in the dry subtropical desert and semidesert zones (Kurganova 1966), experiencing a sharp continental and drought-dominated climate. The flora is quite peculiar and distinctive due to physiographic conditions which influence its formation, but also because of the characteristics of interrelation with contiguous floras of Western Asia (Iran) and other regions of Central Asia.

There are four floristic (botanical-geographical) regions (Muravyeva 1991):

1. Kopetdag-Khorasan mountainous province: Great Balkan, Minor Balkan, Kopetdag-North-Western Kopetdag, South-Western Kopetdag, Eastern Kopetdag.
2. Transitional territory (foothills and low mountains) of Karabil-Badkhyz.
3. Central-Asian mountainous province: Kugitang and hills west to Kugitang.
4. Turan desert province: Karakums, Sundukli, Caspian deserts, and Ustyurt Plateau.

S. A. Pleskanovskaya · M. A. Mamedova · M. A. Ashiraliyeva
State Medical University of Turkmenistan, Ashgabat, Turkmenistan

V. Altay
Biology Department, Faculty of Science and Arts, Hatay Mustafa Kemal University,
Hatay, Türkiye

M. Ozturk (✉)
Vice President of the Islamic World Academy of Sciences, Amann, Jordan
Department of Botany, Centre for Environmental Studies, Ege University, Izmir, Turkey



Fig. 1 Map showing the study area (www.uyduharita.org/turkmenistan)

The flora is represented by more than 2650 species, some of which are rare ones. Kopetdag Mountain's flora is particularly rich where one can find more than 1700 species of wild plants, 332 of which are endemics. Systematic research on Turkmenistan's flora started 150 years ago. A total of 7 volumes entitled "Flora of Turkmenistan" have been published during 1928–1960. In 1988 the book "Determination of Plants of Turkmenistan" was published, which describes 2800 species from 133 families, all arranged in accordance with A. Engler's system of classification (Obuchov 1934; Nikitin and Geldichanov 1988).

One of the important plants in Turkmenistan flora is licorice. Licorice is known as "buyan" in Turkmen language. It belongs to the family Fabaceae. The plant species included in the genus *Glycyrrhiza* are well known from ancient times. Mountains of Central Asia are reported to be the center of origin of this genus. The first official mentioning of this plant appears in the work of Theophrastus, written more than 2300 years ago. In the third century BC he has commented on the taste of different

roots such as sweet Scythian root which grows around the lake Maeotis (Sea of Azov), which is good for asthma, dry cough, and all pectoral diseases. Licorice root has been used in ancient Chinese, Indian, and Tibetan medicine as well as in West Asia as well as Mediterranean countries. It has made its way to the European medicine around twelfth century (Ozturk et al. 2017a). *Glycyrrhiza* is divided into two groups—first group combines the species that contain glycyrrhizic acid (glycyrrhizin) and the second group lacks it. The genus of *Glycyrrhiza* includes 11 species. The first group includes *G. glabra*, *G. uralensis*, *G. korshinskyi*, *G. aspera*, and *G. inflata*, whereas the second group includes *G. echinata*, *G. pallidiflora*, *G. foetida*, *G. lepidota*, *G. acanthocarpa*, and *G. astragalina*. *Glycyrrhiza glabra* belongs to the first group (Obuchov 1934; Nikitin and Geldichanov 1988).

The perennial, herbaceous *G. glabra* (Fig. 2) is widespread in Turkmenistan and has vigorous roots brown from outside and rhizomes. The underground part consists of a vertical tap root, often with several branches, and horizontal rhizomes or stolons, thrown off from the root below the ground, which attain a length of several meters. These runners are furnished with leaf buds and produce stems in the second year. The perennial roots as well as the long horizontal stolons are equally preserved for use. The stem is branching and upright, 0.5–2.0 m tall with small glandular prickles. Stipules are lanceolate often deciduous during the flowering. The plant has light, spreading, pinnate foliage, consisting of 9–10 pairs of leaflets. The leaflets are oblong, ovate with dotted glands. The corolla is violet, or light violet, and calyx is puberulent. In *G. glabra* the pods are smooth, curved, and oblong with 1–8 rounded seeds (Altay et al. 2016; Ozturk et al. 2017a). It is found in the Caspian region, Dashoguz province, Kyurendag, Sumbar, Murgab, Kopetdag, Tejen, and few other places. It generally grows alongside the river valleys, banks, and moist places, and rarely on the dry hills (Ozturk et al. 2017a).



Fig. 2 *Glycyrrhiza glabra* (photo: Greg Kenicer, Royal Botanic Garden Edinburgh—www.powo.science.kew.org)

G. aspera is also found in Turkmenistan. It grows on dry steppes and semideserts, on the foothills and plains, and can survive in various ecological environments including deserts. It is not valuable for industries because of small amounts of glycyrrhizin. It easily becomes a weed due to its ability to quickly propagate with the help of multiple buds on thin rootstocks. It generally grows in Mary province and Kugitang area. *G. aspera* is also a perennial plant, with thin roots and rhizomes. The underground part consists of a vertical root and stolons. The stem is branching and upright, standing up to 0.1–1.5 m tall with small prickles. In the type specimen the pods are spiny. The systematic observation of *Glycyrrhiza* proves that both *G. aspera* and *G. glabra* are found in Turkmenistan (Gladishev 1990). In earlier taxonomical studies *G. glabra* is reported to include three varieties (var. *glandulifera*, var. *pulescens*, and var. *grandiglandulosus*) and one hybrid form. *G. glabra* var. *grandiglandulosus* is reported to be distributed only in Central Kopetdag, on the banks of river next to Kelyata canyon. This plant is described as perennial and grassy with alternate leaves having short hairy and spiny petioles. The stem is upright, hairy, up to 0.6–0.8 m tall. Leaflets are paired, oblong, prickly, and adhesive. In the type specimen *glabra*, the pods are smooth. This form of licorice is distinctive from other kinds by its larger fruits and heavily covered by glandules. Their morphological features depend on ecological conditions of the place where they grow. Intraspecific variety of licorice is not as big based on its widespread distribution. The reason is that it is grown only by vegetative propagation and we have to deal with clones that are different by form and size of leaves and fruits (Gladishev 1990; Gladishev 1991a, b).

Medicinal Value of Licorice (*G. glabra*)

The root of licorice is known all over the world as a medicinal herb. In olden days Tibetans, Indians, and Arabs prepared drugs from this plant to cure several diseases (Ozturk et al. 2017a, b). The roots from the coast of Amu Darya had a particular place in the old prescriptions. The roots from Kunyaurgench and Termez too were highly valued due to their medicinal properties (Lager 1988). This root has been used by the Greeks in ancient times in the treatment of cough and asthma. Almost all books mention about this right from the Middle Ages (Kurganova 1966; Ozturk et al. 2017a). Avicenna too has recommended the use of licorice roots in the treatment of cutaneous ulcers, kidney and urinary bladder diseases, gastritis, fever, lung disorders like bronchial asthma and chronic bronchitis, as well as heart diseases (Ozturk et al. 2017a). In Central Asia it has been used in the treatment of gastric and duodenal ulcers, against hemorrhoid, tumors, dryness, and spasms in the throat, and as an appetite stimulant and expectorant in the catarrh of the upper respiratory tract. This root is considered to be as important as ginseng in the Chinese medicine and is used as antifebrile, analgesic, and expectorant, as a mild purgative and against eczema (Karriyev 1996; Ozturk et al. 2017a).

Medicinal features of licorice root are due to the high glycyrrhizin acid and presence of a considerable number of flavonoids. In 1933 the Japanese chemists Shinoda and Uyeda first time extracted a flavonoid from these roots. These are derived from flavone NB (flavonon) and chalcone. The main one among these is liquiritigenin and its glycosides: monosides like liquiritin and neoliquiritin and biosides such as globoside and uraloside (Muravyeva 1991). In some of the upper parts of the plant flavonoids C-glycosides such as vitexin and its isomer saponaretin as well as foleoside have been extracted. The plant contains coumarins like umellipheron and gernisarin and such oxybrown acids as ferulic and siponic. Much work has been done in this connection till to date (Ozturk et al. 2017a). Some work has also been carried out on the triterpenic compounds being similar in structure to adrenal gland hormones. The glycyrrhizin acid is reported to metabolize in the organism forming substances affecting corticosteroids. Drugs received on the glycyrrhizin and glycyrric acid basis are used in the treatment of Addison's disease and few other disorders (Ozturk et al. 2017a). Flavonoid preparations such as liquiriton and flacarbin have spasmolytic effect (Obuchov 1934; Muravyeva 1991; Socolov and Zamotayev 1990).

Several medicinal preparations like dry and liquid forms from the roots, syrup, powder, and crushed roots are used. All these enter into the composition of various herb mixtures and teas used against respiratory defects, as expectorant, antitussive, and diuretic. These are reported to regulate the water-salt metabolism and are useful in treating gastric ulcers and gastritis (Nikitin and Geldichanov 1988; Ozturk et al. 2017a). Recently the tablets of dry extract of *G. glabra* have been developed with the following composition: dry extract of *G. glabra*, calcium stearate, magnesium subcarbonate, and potato starch (Nepesov et al. 1995). The experimental studies have shown that these tablets dissolve in water and stimulate the production of the mucus, and increase the cell mitotic activity, as well as the number of cells in the main gland and in the pit of the white rat stomach. The functional activity of the specialized cells of the stomach increases and due to this regenerative process is activated in the mucus of stomach. Our contention is that *G. glabra* extract tablets are effective in the stomach due to their hyperfunction in the glandular system diseases (Gladishev 1991a; Khodjageldiyev et al. 1995; Khalmedov et al. 1999).

The root extract tablets were given to the patients as clinical trial in the treatment of gastric ulcerative diseases as a therapy at the "Hospital of Turkmen State Medical Institute" (Gladishev 1991a, b). Another tablet form "*Liquiritin*" containing up to 55% of flavonoids was used in the patients as anti-inflammatory, antispastic, and antacidic agent in hyperacidic gastritis and gastric and duodenal ulcers (Lager 1988; Muravyeva 1991).

The technology used lately in the preparation of emulsion ointments with thick extract of licorice has been developed and introduced (Sakhatov et al. 1997). The ointment has the optimal dehydrogenating activity not less than well-known "*Levomecol*" and "*Vishnevskiy*" ointments. In the treatment of purulent wounds its high healing effect was experimentally observed (Sakhatov et al. 1996; Khudaybergenov et al. 1996). The morphological control over the regeneration process showed that licorice ointment stimulated the appearance of early granulations

containing a large number of lymphocytes and plasmatic cells. Authors consider that the high wound-healing ability of the ointment gives rise to the activation of immune component of inflammation (Nepesov et al. 1995; Karimov and Garadjayev 1997).

The glycyrrhizin acid too enters into the composition of aerosolic ointment “*Epigen*” (*Heminova International S.A.*). This preparation is used successfully in the treatment of vaginosis and cervical erosion. In the treatment of vaginal candidiasis “*Epigen*” is not less effective than well-known “*Diflukan*” (Annamuradova et al. 1999, 2001). The study of toxicity and chemical properties of the ointment with thick extract of *G. glabra* has shown its well absorption, harmlessness, and ability to hold optimal pH (Socolov and Zamotayev 1990; Shukurova et al. 1995; Shukurova and Avdeenko 1997). Application of thick extract of licorice root to the skin of experimental animals has revealed that there is loss of hair during 5–10 days of administration. Considerable changes in the epithelial cell ultrastructure develop simultaneously. The prolonged use of this extract leads to the epidermal atrophy, hyperkeratosis, dystrophy, and sclerosis of the derma fibrillar structures. These changes are reversed after stopping the use of extract (Nepesov et al. 1995; Kakadjanova and Karimov 1999). In the National Turkmenistan Institute of Medicines in Ashgabat, new medicinal forms of licorice are being evaluated. Gelatinous capsules with 100% dry licorice root extract and powder without auxiliary agents or preservatives are also studied. Active preparations of the glycyrrhizin acid (not less than 25%), flavonoids, trace and macroelements (Ca, Mg, Na), and mucosal substances are under investigation at present; these are effective against *St. aureus*, *Sh. flexneri*, *Candida* spp., and *E. coli* (Khalmedov et al. 1999; Khodjageldiyev et al. 1995; Cocanov and Spiridonova 2005).

The decoction of licorice has not lost the importance even today. The root decoction or water solution of its thick extract in combination with other medicinal herbs is used in the monotherapy traditional treatment of pneumonia patients (Sakhatov et al. 1997; Chorekliyev et al. 2003). The effectivity of treatment in the chronic bronchitis patients has increased in the cases of intrabronchial administration of 3.0–5.0 mL of licorice thick extract water solution. In a short period the clinical (Sakhatov et al. 1996; Toychiyev and Hudayberdiyeva 2001) and immune hematological (Khudaybergenov et al. 1996; Rakhmanova et al. 2002) rehabilitation of patients has been achieved. The thick extract water solution is used well in gastroenterology, in particular chronic gastritis and ulcerative diseases of the stomach and the duodenum (Khodjageldiyev et al. 1995; Karimov and Garadjayev 1997) treatment, and stomach surgery (Annamuradova et al. 1999; Chalmedov and Karimov 1995). The water extract protects the mucosal membrane of the stomach from ulcerogenes. The defects of the mucous membrane were found to have recovered in the rabbits in the erosive and ulcerative gastritis after 10–15 days (Annamuradova et al. 2001). The tablets of *G. glabra* have successfully prevented the development of glucous ulcers of the stomach in rats (Shukurova et al. 1995; Chalmedov 1997). The preparations of root on oil basis such as “*Licorice oil*” “*Oil of the Glycyrrhiza*,” and “*Shukur mai*” have been recommended in the treatment of ulcerative disease of stomach and duodenum associated with *Helicobacter pylori* (Shukurova and

Avdeenko 1997; Kalandiya et al. 2005). Ergeshov et al. (1999) have reported high bactericidal properties of licorice root.

To cleanse the sutural material Turkmen silk with alcoholic or water solution of thick extract of *G. glabra* promotes long-term preservation of silk sterility and prevents the surgical wound from being infected (Kakadjanova and Karimov 1999; Ergeshov et al. 1999). The high efficiency of 5% water solution of the thick extract of *G. glabra* was observed in the treatment of patients with rheumatoid arthritis. Under the traditional treatment the patients were administered 100 mL of 5% water solution of thick extract of *G. glabra* as empty stomach once a day for 4 weeks; not only clinical but also immunological rehabilitations were observed in short term and of great degree. The concentration of G_{ig} decreased in the serum of patients; the tendency to decrease in Riga and IBM was marked when compared to the patients who received only the traditional medical treatment. These data were considered to show immunomodulating features of licorice root decoction (Cocanov and Spiridonova 2005; Kheshiyeva et al. 1996).

Results of our studies have confirmed the immunomodulating activity of preparations of licorice root. The white nonlinear mice were immunized with the erythrocytes of sheep under the administration of 1% water solution of thick extract of *G. glabra*; the number of rosette-forming lymphocytes increased in the spleen of animals sharply in comparison with mice which didn't take preparation (Khmelewskaya and Pleskanowskaya 2000). The ability of licorice to stimulate the immune response of mice to the thymus-dependent antigen was found (Chorekliyev et al. 2003).

In vitro studies with the water solution of thick extract of *G. glabra* have shown that it increases the ability of lymphocytes for the rosette formation with the erythrocytes of sheep (T-rosette formation) in healthy persons. In this case the ability of lymphocytes for the rosette formation with the erythrocytes of mouse (B-rosette formation) and for the double (simultaneously) rosette formation with the erythrocytes of sheep and the erythrocytes of mouse (D-rosette formation) did not change. The triterpenic glycyrrhizin acid given as its monosubstituted ammonium salt-glycyram was stimulative and didn't change T- and B-rosette formation of lymphocytes. The flavonoid component liquiritin stimulated T- and D- but oppressed B-rosette formation (Toychiyev and Hudayberdiyeva 2001; Mavlanov et al. 1991). The data obtained allowed to recommend the water solution of the thick extract of *G. glabra* and its components as an immunomodulator.

The phenomenon of the rosette formation is known to be related to the membranous receptors of lymphocytes and to depend on a degree of expression of letters (Rakhmanova et al. 2002). Therefore, the water solution of thick extract of *G. glabra* as a whole and its main components—the glycyrrhizin acid and liquiritin—modulate the expression of membranous receptors of the lymphocytes of human blood in vitro (Bronz and Rochlin 1978). The quick and complete rehabilitation of immune status in the patient with pneumonia and chronic bronchitis (Khudaybergenov et al. 1996; Toychiyev and Hudayberdiyeva 2001; Rakhmanova et al. 2002) is likely to be affiliated with this exact property of licorice both in the conservative and the surgical treatment. The chronic tonsillitis is known to be one of the manifestations of the deficiency of the immune system in humans (Kalandiya et al. 2005; Mavlanov

et al. 1991; Khmelevskaya et al. 2003). Quick and complete rehabilitation of the immune status is manifested by the increase of number of T-lymphocytes and the decrease of IgG concentration in patients with chronic tonsillitis, when they take licorice decoction approved by the high immunomodulating activity of the drug (Chalmedov and Karimov 1995).

The thick extract of *G. glabra* produces immunocorrigating as well as antitoxic and hepatoprotective effect including the stimulatory effect on the regeneration in the experimental pesticide magnesium chlorate poisoning of animals. Used intragastrically for pesticide poisoning in rabbits the licorice decoction protects the liver tissue from narcosis. The albuminous structure of the liver is restored in 15 days; the cellular infiltration of the interstitial tissue disappears; glycogen, DNA, and RNA are recovered in hepatocytes (Ergeshov et al. 1999; Karimov and Lipchenko 1991). The water solution of the thick extract of *G. glabra* possesses high adaptogenic properties. Under heat stroke the considerable oppression of granulocytopoiesis is observed in rats (Khmelevskaya and Pleskanovskaya 1995). The administration of 1% solution intragastrically 10 days before the heat stroke protects animals from death. Besides, the functional activity of the granulocytopoiesis is preserved (Kheshiyeva et al. 1996). It is likely that it is due to the antioxidative properties of the water solution of the thick licorice extract. The extract decreases the concentration of malondialdehyde in the blood serum, in the liver, and in the spleen of rats under the physiologic stress (Khmelevskaya and Pleskanovskaya 2000; Gurbanova et al. 2000; Gurbanova and Konstantinova 2002).

Douglas (2000) has fully stressed the actuality of this issue for medicine and biology. At present the chemical composition of a cell (and/or its organelles) is a starting point for the individual selection of a phytopreparation; that is, cytotoxicity, antibacterial, antiviral, immunostimulating, and anticancerogenic properties as well the insecticide activity of a cell are determined in vitro. However, even then many-sided approaches do not solve a problem of individual sensitivity of a subject to the exact preparation under just that very pathology (Mavlanov et al. 1991; Douglas 2000).

In the Turkmen State Medical Institute the immunology method for the individual selection of medicinal herbs has been developed to treat a range of diseases of internal organs (Bronz and Rochlin 1978; Ovezova and Pleskanovskaya 2002). Medical herbs have been found to be able to modulate in vitro migrational activity of blood leukocytes in healthy volunteers as well as in patients with very different diseases of kidneys, heart, thyroid glands, prostate, and pancreas. The phytopreparation was selected on this basis to treat pathologies in patients. It was determined that 1% water solution of the thick extract or 5% decoction of licorice root is able to modulate considerably the immune response of leukocytes to tissue antigens in vitro depending on the nature of pathology and the degree of sensitivity of a patient to the phytopreparation—either to stimulate the immune response or to inhibit bringing it to a full stop. More than 4000 researches carried out studies to determine the individual sensitivity of patients against 12 medicinal plants including licorice.

The results showed that licorice root decoction is an effective phytopreparation and it was recommended for 83% of patients with pathology of lungs, 64% with

diseases of gallbladder, 54.6% with diseases of cornea, 50% with pathology of kidneys, 50% with pathology of prostate, 30% with pathology of ovaries, 33% with pathology of pancreas, and 20% with autoimmune thyroiditis. The maximal effect of licorice root decoction was observed in patients with pathology of lungs, and the minimal effect was in patients with autoimmune thyroiditis. The individual approach to the administration of phytopreparation increased the efficiency in the treatment of patients with organs indicated (unpublished data). We consider that it is necessary to administer the licorice root decoction very carefully to patients with pathology of thyroid gland and pancreas as far as in 66–67% of cases the preparation in vitro oppresses sharply the immune response of leukocytes to antigens of tissues mentioned and it can provoke the oppression of the functional activity of these organs in vitro.

The preparations of *G. glabra* are of great medicinal and biological significance. They are used widely in the treatment of lungs, gastrointestinal tract, and female genital infections. The properties of licorice studied reveal its efficiency in the treatment of kidneys and cornea due to immunomodulating and antioxidating activity. It can be a preparation of choice in solving the problems on controlled immunocorrection, increasing the organism adaptation possibility (Ozturk et al. 2017a).

Industrial Aspects

Glycyrrhiza plants are known since ancient times (Altay et al. 2016; Karahan et al. 2016; Ozturk et al. 2017a, b). It is said that these plants appeared before oligocene (Kurganova 1966; Ozturk et al. 2017a). However, its trade is said to have started from Azerbaijan in particular from Kura-Araksin lowland. The British-American firm “Mack Andrius” laid the foundation of the industrial purchase of licorice root in the valley of Amu Darya river in the environs of Chardzhou (modern Turkmenabat) in 1906. The licorice root purchased from the valley of Amu Darya is of high quality and valued all over the world. The underground biomass of licorice root of this region is high together with its size (Obuchov 1934).

Since 1923 the Bukhara State Trade purchases the root produced in Turkmenistan. Before 1990s Turkmenistan was the sole leader in purchasing and exporting licorice root. The annual purchase of licorice root by “Soyuzlakrisa” was 21,002 tons including 14,658 tons delivered by Turkmenistan while Tadzhikistan delivered 164 tons. Currently the region of licorice root takes the area equal to almost 900 km along the right and left banks of Amu Darya valley (Atayev 2004).

To increase purchase of root sovkhoses were established in the republics of Central Asia and Kazakhstan for cultivating licorice. In Turkmenistan its cultivation started in Karabekaul on an area of about 5000 ha. To grow licorice as a crop is more difficult than to exploit its natural thickets. The Botany Institute of Turkmenistan worked out the practical recommendations for cultivation of licorice on the flood lands and sands of the Middle Amu Darya oasis. The intensive cultivation of licorice root gives fruits on the third year of its growth. Normally the first industrial

harvesting of licorice root is carried out in 4–5 years; some quantity of roots (up to 25%) are left in the ground for the renewal of plantation (Gladishev and Kerbabwev 1969). It is cultivated in the pre-oasis sandy tracts of the area which is equal to 8900 ha (Keldjajev 1986). In contrast to the licorice cultivated in the floodlands of the Amy Darya those growing in pre-oasis sands (for instance, in Karabekaul district of Chardzhou region named Turkmenabat velayat today) make partial shrubs from the axil buds of the horizontal roots in the first year (Gladishev and Kerbabwev 1969).

Conclusions

Under the Research Program of the experimental station “Ylym” of Agroindustrial complex “Buyan” named after S.A. Niyazov and Research-industrial amalgamation “Turkmen derman” of the Ministry of Health and Medical Industry of Turkmenistan, the researchers aim at improving the state of licorice thickets and turn it into an industrial crop. The work is going on for the last 20 years. Data on ecological and biological features of this agrocenoses, its development under the conversion of licorice into an industrial crop, as well as the complete botanic characteristics of introduced forms together with recommendations for improving the natural herbage of this plant are presented in this chapter. It is shown that not only licorice root but also its surface part have good fodder properties and can be used in agriculture widely. At present licorice hay makes 90% of all coarse fodder and its thickets serve as all-the-year-round pastures. The hay is ascertained to be the fine fodder to gain fat, as its estrogens are considered to be stimulators for the growth of animals (Gladishev and Kerbabwev 1969; Goryachev 1966; Rizayeva et al. 1969). The results of the long-term experiment on surface and radical improvement of the state of natural licorice thickets and its crop and the analysis of cases reducing the unique natural habitat of the plant are given in a range of monographs and articles by Turkmen scientists (Kurganova 1966; Gladishev 1990, 1991a; Gladishev and Kerbabwev 1969; Kerbabayev and Gladishev 1971; Kerbabayev et al. 1969).

The licorice root is purchased from the valley of Middle Amu Darya (Turkmenabat velayat). It has exceptional trade qualities and is exported from Turkmenistan. The specialized branches of industry for processing of licorice root have been set up in Europe, the USA, and Japan. The exclusive medicinal properties of this unique plant and its rich and peculiar chemical composition point that licorice is a plant of the future.

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