Boerhaavia diffusa – A Wild Herb with Potent Biological and Antimicrobial Properties

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Abstract

Boerhaavia diffusa, *commonly known as* punarnava *in Sanskrit, is a herbaceous plant of the family Nyctaginaceae. The whole plant or its specific parts (leaves, stem, and roots) are known to have medicinal properties and have a long history of use by indigenous and tribal people in India. The medicinal value of this plant in the treatment of a large number of human ailments is mentioned in Ayurveda, Charaka Samhita, and Sushrita Samhita. It has many ethnobotanical uses (the leaves are used as vegetable; the root juice is used to cure asthma, urinary disorders, leukorrhea, rheumatism, and encephalitis), and is medicinally used in the traditional, Ayurvedic system. Besides, the* B. diffusa *plant is reported to posses many pharmacological, clinical, and antimicrobial properties. Recently, the authors observed potent antiviral efficacy of this plant against phytopathogenic viruses. The antiviral agent isolated from this plant was found to be a glycoprotein with a molecular weight of 16–20 kDa. Administered by foliar spraying in the field, this antiviral agent could protect some economically important crops against natural infection by plant viruses.*

A program for the screening of *Boerhaavia diffusa* extracts (from different plant parts) against a wide range of phytopathogenic viruses was started by the authors at the Department of Botany, Lucknow University, Lucknow, Uttar Pradesh, India some three decades ago. So far, over one thousand plants of different taxonomic families have been screened for their antiviral activity (Chessin *et al.*, 1995). The aqueous extracts of plant material were prepared and tested in vitro as well as in vivo against phytopathogenic viruses on their hypersensitive and systemic hosts. The botanical identity of each plant was established before the extracts were prepared. Of the large number of plants screened, *B. diffusa* root extracts were found to have a broad spectrum and very high antiviral activity (Verma and Awasthi, 1979), and hence selected for further studies.

In view of the high antiviral activity exhibited by this plant, this paper presents a review of the other uses of *B. diffusa*, such as its therapeutical and pharmacological uses and general domestic utility.

Boerhaavia diffusa is a herbaceous member of the family Nyctaginaceae. It is widely distributed in the tropics and subtropics (CSIR, 1988). It has a long history of uses by indigenous and tribal people and in Ayurvedic or natural herbal medicines (Dhar *et al*., 1968). The major active principle present in the roots is alkaloidal and is known as punarnavine.

In the traditional system of medicine, *B. diffusa* roots have been widely used for the treatment of dyspepsia, jaundice, enlargement of spleen, and abdominal pain (Kirtikar and Basu, 1956), and as an antistress agent. The worldwide use of *B. diffusa* roots to treat liver disorders was validated when researchers demonstrated, in 1980 and 1991, that its root extract had antihepatotoxic properties (Chandan *et al*., 1991; Rawat *et al*., 1997).

Pharmacological studies have demonstrated that *B. diffusa* possesses diuretic (Gaitonde *et al.*, 1974), anti-inflammatory (Bhalla *et al.*, 1968), antifibrinolytic (Jain and Khanna, 1989), anticonvulsant (Adesina, 1979), and antibacterial properties (Olukoya *et al.*, 1993), which makes it a very useful medicinal plant. The aqueous extracts of roots are also a rich source of a basic protein, known as

systemic resistance inducing protein (BD-SRIP). The aqueous solution of this protein, when applied before virus infection/inoculation, induces strong systemic resistance in several susceptible plants against commonly occurring viruses (Verma and Awasthi, 1979). All these properties have made this plant very interesting, and the plant has played an important role in the treatment of human and plant diseases.

Nomenclature and systematic position

Confusion exists in the literature on the correct identity of punarnava (*Boerhaavia diffusa*). Several species of *Boerhaavia* are being used under the name *punarnava* in India. It is generally agreed that the red variety is *B. diffusa*, whereas the white variety has been identified as belonging to the *Trianthema* species. The roots of *Trianthema* are often used as an adulterant for *punarnava*. The two genera belong to different families, and *Trianthema* species neither contain the active constituent (punarnavine), nor have red flowers. The whole *Boerhaavia diffusa* plant (fresh as well as dried) is the genuine source of the drug *punarnava*, and is considered official in Indian Pharmacopoeia.

The plant was named in honor of Hermann Boerhaave, a famous Dutch physician of the 18th century (Chopra, 1969). *Boerhaavia*, a herbaceous plant, belongs to the Nyctaginaceae (four o'clock) family, order Thymilae, group Dicotyledons and phylum Angiosperms (Rendle, 1925). *Boerhaavia diffusa* is a perennial herb and it is ascribed the name *punarnava*, a drug known since long in the indigenous system of medicine in India. The name *punarnava* (*Punah punarnava bhawati iti*, in Sanskrit, translates as "that which becomes fresh again and again . . .") is probably derived from the perennial habit of the plant, which remains dry and dormant during summer and regenerates from the same old root stock in the rainy season, or from the Sanskrit phrase that denotes such therapeutic property (*Karoti shariram punarnavam*, in Sanskrit, translates as "that which rejuvenates the body") (Wahi *et al.*, 1997). Apart from *punarnava*, the plant has a host of other names in Indian languages: *biskhafra* in Hindi, *thazhuthama* in Malayalam, *mukaratte* in Tamil, *gadhapurna* in Bengali, *satodi* in Gujarati, *itsit* in Punjabi, etc. It is known as hogweed or pigweed in USA, and Erva Tostâo in Brazil.

Geographical distribution, origin, and habitat

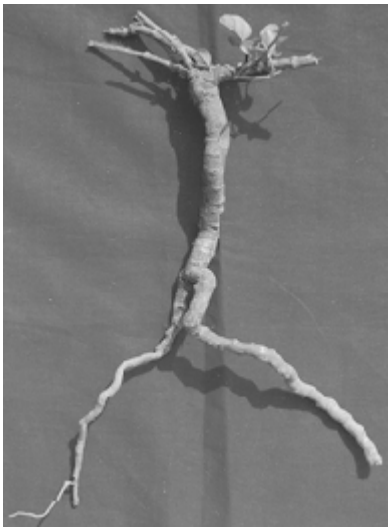
The genus *Boerhaavia* has several species, and is distributed in the tropical, subtropical, and temperate regions of the world (Heywood, 1978). It is found in Australia, China, Egypt, Pakistan, Sudan, Sri Lanka, South Africa, USA and in several countries of the Middle East. Out of the 40 species of this genus, 6 species are found in India – *B. diffusa, B. chinensis, B. erecta, B. repens, B. rependa*, and *B. rubicunda* (Chopra, 1969; CSIR, 1988).

Boerhaavia diffusa is also indigenous to India; it is found throughout the warmer parts of the country up to an altitude of 2000 m in the Himalayan region. It grows well on wastelands and in fields after the rainy season (Chopra, 1969). The plant is also cultivated to some extent in West Bengal (CSIR, 1988).

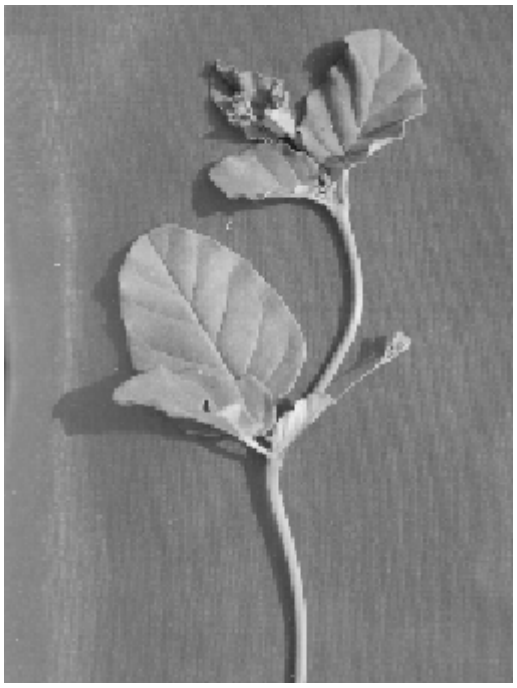
Generic description of *Boerhaavia diffusa*

Boerhaavia diffusa is a perennial creeping weed, prostrate or ascending herb, up to 1 m long or more, having spreading branches (Fig. 1). The roots are stout and fusiform with a woody root stock. The stem is prostrate, woody or succulent, cylindrical, often purplish, hairy, and thickened at the nodes. Leaves are simple, thick, fleshy, and hairy, arranged in unequal pairs, green and glabrous above and usually white underneath. The shape of the leaves varies considerably – ovate-oblong, round, or subcordate at the base and smooth above. Margins of the leaves are smooth, wavy, or undulate. The upper surface of the leaves is green, smooth, and glabrous, whereas it is pinkish white and hairy beneath. Leaves are up to 5.5 × 3.3 cm^2 in area. Flowers are minute, subcapitate, present 4–10 together in small bracteolate umbels, forming axillary and terminal panicles. These are hermaphrodite, pedicellate, and white, pink, or pinkish-red in color. Bracts are deciduous and involucrate. A perianth is present in the place of a calyx and corolla, which is tubular in shape, the tube being short and

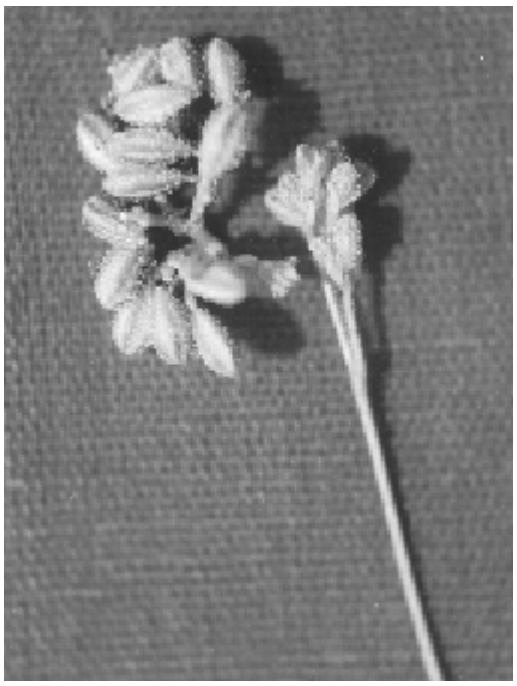
Naturally growing plants



Root



Stem and leaves



Flowers and fruits

Figure 1. ***Boerhaavia diffusa.***

narrow at the base and funnel-shaped at the top and constricted above the ovary. There are five lobes, which are small and acute. Two or three stamens are present and are slightly exserted. The stigma is peltate. The achene fruit is detachable, ovate, oblong, pubescent, five-ribbed and glandular, anthocarpous, and viscid on the ribs (Thakur *et al*., 1989). The seeds germinate before the onset of the monsoon. The plant grows profusely in the rainy season, and mature seeds are formed in October–November. Due to its sticky nature, the plant gets stuck on the clothes of human beings and on the legs of animals, which helps in its dispersal from one place to another. It has a large root system bearing rootlets. The tap root is tuberous, cylindrical to narrowly fusiform to conical or tapering, light yellow, brown or brownish gray. It is thick, fleshy and very bitter in taste. Some workers have studied the regeneration of this plant through tissue culture. Bhansali *et al.* (1978) reported induction of adventitious shoots using stem explants of *B. diffusa.* Roots were also regenerated from the leaf segments of *B. diffusa* when cultured in vitro. These roots contained 0.15% alkaloid punarnavine. Increase in levels of indole- acetic acid (IAA) in MS medium reduced the number of roots regenerated from the leaf segment, their length and alkaloid content (Shrivastava and Padhya, 1995).

Ethnobotanical uses

In Purulia (West Bengal), tribals eat this plant as vegetable. *Boerhaavia* leaves are cooked and eaten in Assam, where it is commonly found in the markets. Its roots are used in the treatment of piles by the inhabitants of the Garhwal Himalaya (Uttaranchal). The root paste is used to cure bloody dysentery by the Bhils of the Jhabua district in Madhya Pradesh. The decoction of the plant is given in the treatment of nodules in the body. The root juice is used in treating asthma, scanty urine, and internal inflammation disorders. *Boerhaavia diffusa* is used for curing ailments such as leukorrhea, rheumatism, and stomach ache by the Sahariya tribe in the Lalitpur district of Uttar Pradesh. This plant is also used by the tribes of Ambikapur district (Madhya Pradesh) for the treatment of elephantiasis. In the Indo-Nepal Himalayan terai region, the tribals harvest this plant for medicinal purposes, mainly for flushing out the renal system, and to treat seminal weakness and blood pressure (Mitra and Gupta, 1997).

Chemical composition of *Boerhaavia diffusa*

The *Boerhaavia diffusa* plant contains a large number of such compounds as flavonoids, alkaloids, steroids, triterpenoids, lipids, lignins, carbohydrates, proteins, and glycoproteins. Punarnavine $C_{17}H_{22}N_2O$ mp 236–237°C (Agarwal and Dutt, 1936; Basu *et al.*, 1947; Surange and Pendse, 1972), boeravinone A.F (Kadota *et al.*, 1989; Lami *et al.*, 1990; 1992), hypoxanthine 9-L-arabinofuranoside (Ahmad and Hossain, 1968), ursolic acid (Mishra and Tiwari, 1971), punarnavoside (Jain and Khanna, 1989), liirodendrin (Aftab *et al.*, 1996), and a glycoprotein having a molecular weight of 16–20 kDa (Verma *et al.*, 1979) have been isolated and studied in detail for their biological activity.

Chopra *et al.* (1923) reported that the plant contained large quantities of potassium nitrate, besides punarnavine. The herb and roots are rich in proteins and fats. The herb contains 15 amino acids, including 6 essential amino acids, while the root contains 14 amino acids, including 7 essential amino acids. Seth *et al.* (1986) isolated a new antifibrinolytic compound ‘punarnavoside’ from the roots of *B. diffusa*. Phytochemical screening of the roots from garden-grown in vivo plants of *B. diffusa* of different ages revealed that the maximum alkaloid content (2%) accumulated in the roots of 3-year-old mature plants.

Biological activity

As medicine in the traditional system

Different parts of the *B. diffusa* plant have been widely used by indigenous tribes in the traditional system of medicine. The roots have been widely used for the treatment of dyspepsia, jaundice, enlargement of spleen, abdominal pain, abdominal tumors, and cancers (Kirtikar and Basu, 1956).

The root powder, when mixed with *mamira* (*Thalictrum foliolosum*), is used to treat eye diseases. It cures corneal ulcers and night blindness (Gupta *et al.*, 1962), and helps restore virility in men. People in tribal areas use it to hasten childbirth (Shah *et al.*, 1983). The juice of *Boerhaavia* leaves serves as a lotion in ophthalmia. It is also administered orally as a blood purifier and to relieve muscular pain.

As medicine in the Ayurvedic system

In old Indian books of medicine such as the Charaka Samhita and Sushrita Samhita, it is mentioned that the Ayurvedic preparations made from *punarnava* – namely, *punarnavastaka kvath*, *punarnava kshar*, and *punarnava taila* – were used for the treatment of various ailments. The whole plant of *B. diffusa* is a very useful source of the drug *punarnava*, which is documented in Indian Pharmacopoeia as a diuretic (Chopra, 1969). The active principle contained in the herb is an alkaloid, known as punarnavine. The roots and leaves with flowers have been found to be highly potent (CSIR, 1988). In Ayurvedic medicine, different parts of this plant were reported to have various medicinal properties. It was used in renal ailments as diuretic (Anand, 1995); and to treat seminal weakness and blood pressure (Gaitonde *et al.*, 1974). It is also used in the treatment of stomach ache, anemia, cough, and cold, and as a diaphoretic, laxative, expectorant, and a potent antidote for snake and rat bites (Chopra *et al.*, 1956), in the treatment of nephrotic syndrome (Singh and Udupa, 1972), hepatitis, gall bladder abnormalities, and urinary disorders (Mudgal, 1975; Cruz, 1995). The flowers and seeds are used as contraceptive (Chopra *et al.*, 1956).

Pharmacological and clinical properties

Pharmacological studies have demonstrated that *punarnava* possesses punarnavoside, which exhibits a wide range of properties – diuretic (Gaitonde *et al.*, 1974); anti-inflammatory (Bhalla *et al.*, 1968); antifibrinolytic (Jain and Khanna, 1989); anticonvulsant (Adesina, 1979); antibacterial (Olukoya *et al.*, 1993); antistress agent; antihepatotoxic (Mishra, 1980; Chandan *et al.*, 1991; Rawat *et al.*, 1997); anthelmintic febrifuge, antileprosy, anti-asthmatic, antiscabies, and anti-urethritis (Nadkarni, 1976); and antinematodal activity (Vijayalakshmi *et al.*, 1979). An aqueous extract of thinner roots of *B. diffusa* at a dose of 2 ml kg^{-1} exhibited marked protection of various enzymes such as serum glutanic-oxaloacetic transaminase, serum glutanic-pyruvic transaminase, and bilirubin in serum against hepatic injury in rats (Rawat *et al.*, 1997). *Punarnava* possesses diuretic and anti-inflammatory activities and the maximum activity was observed in samples collected in the rainy season. Due to the combination of these two activities, *punarnava* is regarded therapeutically as highly efficacious for the treatment of inflammatory renal diseases and common clinical problems such as nephrotic syndrome, oedema, and ascites resulting from early cirrhosis of the liver and chronic peritonitis.

The plant is reported to be efficacious in abdominal tumors and cancers. The drug proved useful as a hematinic and as a growth promoter in children fed with milk fortified with the drug. In the form of a powder or an aqueous decoction, the drug was found to be useful in the treatment of nephrotic syndrome and compared well with corticosteroids. The drug decreased the albumin urea; the serum protein was increased and serum cholesterol level was lowered.

Singh and Udupa (1972) reported that dried root powder showed curative efficiency when administered orally for one month to children or adults suffering from helminth infection. The subjects became worm-free within five days of treatment. The drug, singly or in combination with other drugs, was found to be effective in liver disorders, heart diseases (hypertension, angina, cardiac failure, etc.), respiratory tract infections, leukorrhea, spermatorrhea, etc. The purified glycoprotein from *B. diffusa* exhibited strong antimicrobial activity against RNA (ribonucleic acid) bacteriophages (Awasthi and Menzel, 1986). With much of the clinical research validating its long history of different uses in natural medicine, the commercial bulk of punarnava in India represents heterogeneous medicinal uses.

As feed

The entire plant including the roots is eaten as vegetable, in curries and soups. The roots and seeds are added to cereals, pancakes, and other foodstuffs. They are also served as bird feed or poultry feed. The plants are grazed by sheep, goats, and cows, and in West Bengal, it is believed that the plant enhances lactation period and also the amount of milk in cattle (CSIR, 1988).

Antiviral activity of *Boerhaavia diffusa*

In view of the pharmacological, clinical, and medicinal potential of this plant, the authors screened the root, leaf, stem, flower, and seed samples (collected at different stages of plant growth and from different locations, both fresh and dried) for their antiviral activity against a number of isometric as well as anisometric phytopathogenic viruses, in various host/virus combinations both in vitro and in vivo (Verma and Awasthi, 1979). Maximum antiviral activity, in each case, was recorded with the aqueous extract of dried root powder applied before virus inoculation. The active principle was purified and isolated (Verma *et al*., 1979). The roots of *B. diffusa* are a rich source of a basic protein, which is used for inducing systemic resistance in many susceptible crops against commonly occurring viruses (Verma and Awasthi, 1979; 1980; Verma *et al*., 1979; Awasthi *et al.* 1984; 1985; 1989). This protein or antiviral agent was active against tobacco mosaic virus in *Nicotiana glutinosa*, *Datura metel*, *Chenopodium amaranticolor*, and *Nicotiana tabacum* (Ky58 White Burley and NP31); sunnhemp rosette virus in *Cyamopsis tetragonoloba*, *Vigna unguiculata*, and *Crotalaria juncea*; and gomphrena mosaic virus in *Chenopodium amaranticolor*, *Vigna unguiculata*, and *Gomphrena globosa* when applied a few hours (2–24 h) before inoculation by the respective inocula of viruses (Verma and Awasthi, 1979; Awasthi *et al.*, 1984). The antiviral agent was a basic glycoprotein (70–80% protein and 8–13% carbohydrates) with a molecular weight of 16–20 kDa as determined by gel filtration chromatography (Verma *et al*., 1979). The resistance-inducing protein was found to be extremely thermostable (Verma and Awasthi, 1979). Following treatment with the systemic resistance inducing protein, the susceptible healthy host produced a virus inhibitory agent (VIA). The VIA showed the characteristics of the protein, and upon incubation with the virus, reduced infectivity of the viruses both in vitro and in vivo (Verma and Awasthi, 1980). Upon gel filtration on Sephadex G-75®, two active fractions, exhibiting protein characteristics, were recovered (Verma and Awasthi, 1980, Awasthi *et al*., 1987). The VIA was present both in treated as well as untreated leaves. The biophysical characteristics of induced VIA were also studied and it was found to be a basic protein (Awasthi *et al*., 1987). The glycoprotein occurring in *B. diffusa* roots functions as a signal molecule, and is of great interest as it has a role in stimulating the defence systems of plants against viruses.

Owing to the high antiviral efficacy of *B. diffusa* under laboratory conditions, it was tested under field conditions as well against a few viral diseases of economically important crops (Table 1). The purified glycoprotein from *B. diffusa* reduced infection and multiplication of tomato yellow leaf curl virus (Awasthi and Rizvi, 1999), papaya ring spot virus (Awasthi, 2000), and cucumber green mottle mosaic virus (Awasthi *et al*., 2003). The aqueous crude extract from the dried roots was also found significantly active against a number of viruses – mung bean yellow mosaic virus (Awasthi, 2000); bean common mosaic virus (Singh and Awasthi, 2002); water melon mosaic virus (Awasthi, 2002); bottle gourd mosaic virus in muskmelon (*Cucumis melo*), ridged gourd (*Luffa acutangula*), and bottle gourd (*Lagenaria siceraria*) (Awasthi and Kumar, 2003); cucumber mosaic virus in cucumber (*Cucumis sativus*) and muskmelon and water melon mosaic virus in watermelon (*Citrullus lanatus*) (P Kumar, personal communication); and mung bean yellow mosaic virus in mung bean (*Vigna radiata*) (S Singh, personal communication).

In treated plants, the antiviral agent from *B. diffusa* not only decreased disease symptom severity but also protected the plants against infection by viruses. A significant and many fold enhancement/increase in plant growth along with increase in yields was also observed (Table 1).

Table 1. Prevention and control of viral diseases of crops in fields by Boerhaavia diffusa, an antiviral agent.

Virus	Crop	Disease protection (%)	Increase in yield (%)	Reference
Potato virus X	Potato	68	22	Awasthi and Mukerjee (1980)
Tomato leaf curl virus	Tomato	71	24	Awasthi *et al*. (1984)
Complex infection of tomato mosaic and cucumber green mottle mosaic virus	Tomato Cucumber	75 62	16 19	Awasthi *et al*. (1985) Awasthi *et al*. (1985)
Tomato mosaic virus	Tomato	78	12	Awasthi *et al*. (1985)
Brinjal mosaic virus	Brinjal	64	9	Awasthi *et al*. (1985)
Pea mottle virus	Pea	47	26	Awasthi *et al*. (1985)
Oat sterile dwarf virus	Oats	42	NA[1]	Kempiak *et al.* (1991)
Tomato yellow mosaic virus	Tomato	68	29	Awasthi and Rizvi (1998)
Tomato leaf curl virus	Tomato	47	30	Awasthi and Rizvi (1999)
Mung bean yellow mosaic virus	Mung bean Black gram	46 NA	37 NA	Awasthi (2000) Awasthi (2000)
Bean common mosaic virus	Black gram Mung bean	42 NA	28 NA	Singh and Awasthi (2002) Singh and Awasthi (2002)
Water melon mosaic virus	Watermelon	49	18	Awasthi (2002)
Bottle gourd mosaic virus	Bottle gourd	68	42	Kumar and Awasthi (2003a)
Cucumber green mottle mosaic virus	Muskmelon	47	36	Awasthi *et al.* (2003)
Cucumber mosaic virus	Cucumber	59	27	Kumar and Awasthi (2003b)
Mung bean yellow mosaic virus	Mung bean	63	41	S Singh (personal communication)
Cucumber green mottle virus	Muskmelon	48	32	P Kumar (personal communication)
Cucumber mosaic virus	Cucumber	67	39	P Kumar (personal communication)
Mung bean yellow mosaic virus	Black gram	65	36	S Singh (personal communication)

1. NA = Data not available.

Conclusion

Boerhaavia diffusa is an important medicinal plant. The plant is widely used for the treatment of oedema, dropsical condition, and urinary troubles. A large number of publications on the chemistry, pharmacology, and several other aspects have been made, but no homogenous, pure, active principle of the plant in the form of a modern standardized drug has been introduced. A basic protein showing high systemic resistance inducing activity against plant viruses has been isolated, but it has not yet been purified to homogeneity and commercially made available. However, the plant is abundantly available in wild form over large tracts of land.

The commercial bulk of *B. diffusa* represents a heterogeneous population. Consequently, it quite often results in poor quality roots and biomass. The cardinal feature of modern cultivation of any plant with impressive uniformity and high productivity for end product is thus grossly lacking in *B. diffusa*. The need, therefore, is to genetically improve the available commercial bulk of *B. diffusa* in order to meet the natural requirement of the valuable products. Improved varieties with enhanced drug yields hold great promise. Mutation breeding may have an important role in this direction and may improve the yield and quality. Recently, Shukla (2002) has reported a substantial amount of genetic variability in *B. diffusa.* Of the seventy-one genotypes tested, only a few were elite lines and were found to have desirable material for commercial use. Therefore, commercial manufacture of active constituents from these improved elite lines would be useful and profitable.

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