



Physicochemical Standardization and Pharmacognostical investigation of *Zyziphus oenoplia* (L.) Mill. Root.

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Abstract

The experiments of Pharmacognostical standardization in fresh, powdered plant material and anatomical sections of the root of *Zyziphus oenoplia* (L.) Mill was carried out to determine its morphoanatomic and microscopical characters and also some of its quantitative physiochemical standards. Roots were cylindrical, blackish brown surface, longitudinal striations with irregular transverse ridges with splintery- fibrous fracture and yellowish brown in colour. Internally roots show the presence of cork, epidermis, periderm, vessels, cortex, phloem, xylem, endodermis and starch grains. The powder microscopy revealed the presences of Fibres and other cell structures. Physicochemical investigation includes ash values, extractive values and moisture content, swelling index, foaming index was evaluated. These present investigation will be useful towards establishing pharmacognostic standards on which helps batter in understanding their identification, purity, quality and medicinal importance in depth which can provide the reliable information for further pharmaceutical applications and research.

Keywords: *Zyziphus oenoplia* (L.) Mill, Pharmacognostical standardization, Physico-chemical parameters.

Introduction

Zyziphus oenoplia, belongs to the family "Rhamnaceae". It is commonly known as Jackal Jujube or Small fruited jujube in English, Bahukantaka, Karkandhau in Sanskrit, Shiakol in Bengali and pargi in kannada. *Zyziphus oenoplia* mostly found in India, Pakistan, Bangladesh, Sri lanka, Malaysia and Australia. It grows throughout India in dry forests and open bushy places along the roadside forests and thickets. The plant *Zyziphus oenoplia* is an erect, straggling or climbing shrub up to 3 m tall; branches fasciculate or not, often densely rusty tomentose; nodes slightly enlarged around the leaf scars. Leaves 1-8 × 2-3 cm, alternate, obliquely ovate or elliptic, crenate or sub-entire, oblique at the base, subrounded, apex acute or acuminate, 3-4 nerved, softly pubescent above, softly pilose beneath; petioles 2- 5 mm long, pubescent; stipular spines solitary, recurved. Inflorescence axillary shortly pedunculate cymes. Pedicels about 2 mm long, pilose. Calyx lobes 1.5-2 mm long, ovate-triangular, apex acute, glabrous inside, brownish, apparently hairy outside. Petals 0.8-1.0 mm long, spatulate, clawed, shorter than calyx. Stamens 0.7-0.9 mm long, Disc glabrous 10-lobed; lobes opposite each calyx lobe, emarginate. Ovary globose, glabrous, 2-celled, immersed in disk; styles 2, united to above the middle; stigma obtuse. Drupe 5-7 × 5-6 mm, globose or ovoid-globose, small, base with persistent calyx tube, apex mucronulate, black and shining when ripe; fruiting

pedicel 3-4 mm long, pilose. Seeds 1-2, 1 cm long, shiny, globose. The flower & fruiting of the plant occurs in the month of August-January^{1,2}. It can be propagated by seeds. Traditionally fruits and roots of this plant were used for different diseases³. The different parts of plant, contain different types of phytoconstituents. The root bark contains two new cyclopeptide alkaloids, zizyphine-A and zizyphine-B. betulinic acid, d-glucose, d-fructose, sucrose and unidentified polysaccharides, Betulinic acid. Stem bark also contains cyclopeptide alkaloids Zizyphine (A-G) and abyssinine A and B. The leaves of *Zyziphus oenoplia* contains methylated flavonol 3' 4' – diOMe quercetin, betulinic acid, a pentacyclic triterpine which is the common secondary metabolite of *Zyziphus oenoplia* and also produced some selective cytotoxicity against human melanoma cells^{4,5}. According to Ayurveda, the plant root is very useful in the treatment of fresh cuts and wounds, ulcer, ascaris infection, stomachalgia etc.² One of tribe called 'Chakmas' in Bangladesh were use the plant traditionally for gastrointestinal disorder⁶ and also among the 'Munda' tribe, the fruit is used as an ingredient in the preparation of stomach ache pill.² The roots are astringent bitter, anthelmintic, digestive and antiseptic. They are useful in hyperacidity, ascaris infection, stomachalgia and healing of wounds. Fruits are found to have medicinal properties like blood purifier, febrifuge, abdominal pain. In Siddha the fruit

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and the seed is used in fever, retention of urine, poisoning, aphrodisiac, tonic etc². Pharmacognostical profiles on root of this plant till not established. So the prime target of this present investigation is to study the macro anatomical, microscopical, pharmacognostical features and physicochemical standards of root of *Zyziphus oenoplia* (L.) Mill,. This may help not only to produce a standard monograph for the proper identification of the plant also gives an immense help for its proper use.

Materials and Methods

Collection and Authentication

Roots of *Zyziphus oenoplia* (L.) Mill were collected from the road sides of Benavalli forest area, Sagar road, 8-10 km from Shimoga district of Karnataka, India, in the month of September 2009 in a quantity sufficient for all the experiments in a single batch. The plant material was authenticated by Dr. K. Krishna Swamy, Principal investigator (UGC major research project) Dept. of Botany, Sahyadri Science College, Shimoga. One voucher specimen was submitted and preserved in the Department of Pharmacognosy, National College of Pharmacy, Shimoga (Herbarium no.NCP/09-10/005).

Pharmacognostic Standardization

Dried roots of *Zyziphus oenoplia* (L.) Mill was placed in a test tube containing water and boiled over Bunsen flame for a few minutes to soften the hard root. Transverse section was taken by cutting with a razor blade at right angle to the longitudinal axis of the root material. A thin section of the plant material (root) was mounted in chloral hydrate solution and warmed gently for clearing of the section, stained with phloroglucinol - hydrochloric acid (1:1) and mounted in glycerin⁷⁻¹⁰. Cover slip was put with care to avoid air bubbles. The section was observed under using simple microscope and photograph was taken. The shape, size, colour, odour, taste, fracture roots were determined. Dried roots Powder was used for the observation of powder characteristics. The drug powder was separately treated with phloroglucinol-hydrochloric acid (1:1) solution⁹⁻¹⁰.

Physico-Chemical Evaluations

The quality control parameters studied include ash values, loss on drying, crude fibers content, foaming index, swelling index etc. were determined. Extractive values were also determined to find out the amount of

water, alcohol, chloroform and petroleum ether soluble components present on it¹¹⁻¹⁶. The data obtained from the above studies are shown in

Physicochemical constants	Values (%)
Ash values	
Total ash	5.03 ± 0.06
Water soluble ash	1.0 ± 0.06
Acid insoluble ash	1.43 ± 0.12
Extractive values	
Petroleum ether (60-80°C)	2.17 ± 0.29
Chloroform	9.03 ± 0.06
Ethanol (90%)	15.03 ± 0.15
Aqueous	18.07 ± 0.12
Loss on drying	
Dry matter content	88.63 ± 0.47
Moisture content	11.13 ± 0.15
Crude fiber content	
Foaming index	80.20 ± 0.20
Swelling index	294.15 ± 0.04
	0.81 ± 0.02

Result and Discussion

The roots of *Zyziphus oenoplia* (L.) Mill. were observed to be Cylindrical 50-140mm in diameter, blackish brown surface, longitudinal striations with

Table 1: Physicochemical Analysis of Root of *Zyziphus oenoplia* (L.) Mill.
N= Three experiments for each parameter. The values shown are mean ± S.E

irregular transverse ridges. Fractures were splintery-fibrous and yellowish brown in colour



Figure 1: Morphological Features of *Zyziphus oenoplia* (L.) Mill root.

The roots of *Zyziphus oenoplia* (L.) Mill. having Characteristic odour, Bland taste and hard texture, yellowish brown Fracture. In microscopic studies, the root shows all the typical characteristics of root. The TS of the root (**Fig.2 & 3**) was circular in outline. Outer cuticular layer or cork was followed by thin walled epidermal cells. Phellogen part was bilayered, immediately below the cork and cells tangentially elongated. Cortex portion of the root composed of 16-

18 layers of horizontally elongated parenchyma with small intercellular spaces. The parenchyma cells contain starch grains which were both simple and compound. A continuous ring consists of phloem parenchyma. Phloem was seen in several thin patches around the well developed xylem calls. Biseriate, reddish orange coloured Medullary rays observed. Xylem vassals occupied the entire central portion and were traversed regularly by rows of medullary rays whose cells were lignified. Each ray cell was radially elongated and contains starch. The root section was shown absence of pith at the centre of the section.

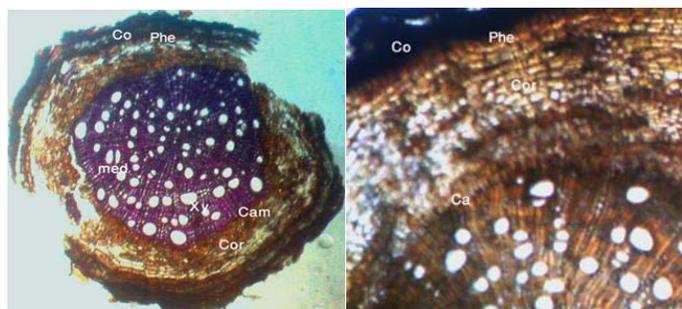


Figure 2: T.S. of Young Root (1.6mm Thick). (Phe- Phellogen, Co- Cork, Cor- Cortex, Xy- xylem, Med- Medullary rays, Cam- Cambium layer.)

The powder microscopy of the root powder reveals the presence of cork- about six layers, thin walled, tubular, polygonal cells. Moss and liverwort cells, wood elements- xylem vessels and fibres inter lock with each other to form a spindle shaped structure, starch grains- simple as well as compound (Fig.3).

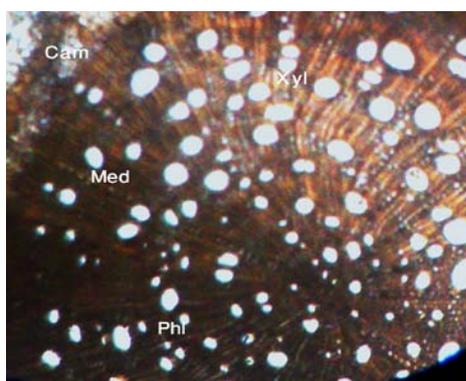


Fig.-3. TS of the root of *Zyziphus oenoplia* (L.) Mill.

(Co –cortex, Phe- Phellogen, Cor- Cortex, Ca- cambium., Med- Medullary rays, Xyl- Xylem, Phl- Phloem.)

The physical constant evaluation of the drugs is an important parameter in detecting adulteration. Ash values are helpful in determining the quality and purity of crude drugs, especially in the powdered form. The object of ashing vegetable drugs is to remove all traces of organic matter which may otherwise interfere in an analytical determination.^{11,12} Extractive values are useful for (1) evaluation a crude drug, (2) give an idea

about the nature of the chemical constituents present in a crude drug, and (3) useful for the estimation of specific constituents, soluble in that particular solvent used for extraction. The foaming ability of an aqueous decoction of plant materials and their extracts is measured in terms of a foaming index. The phytochemical investigation has shown that the ethanol and aqueous root extracts of the plant contained saponins. Many medicinal plant materials are

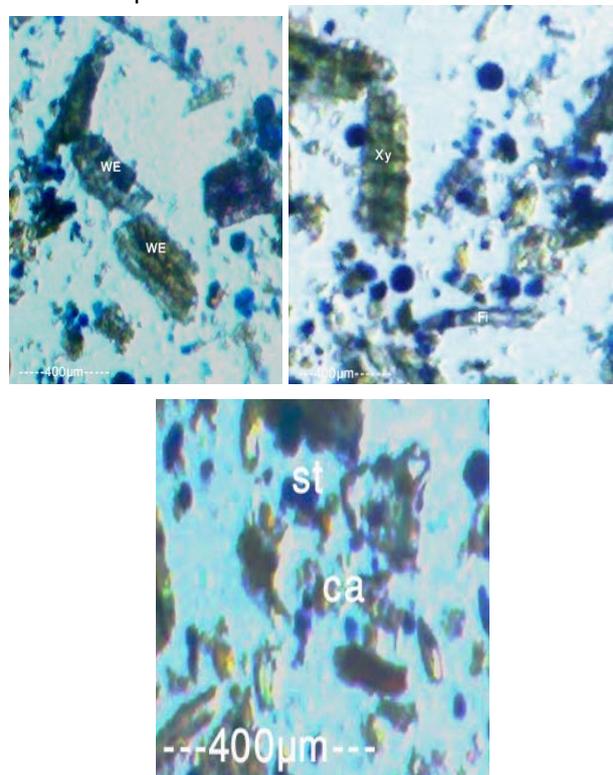


Fig.- 4. Powder characteristics. (WE- Wood elements, Xy- Xylem, Fi- Fibers, St- Starch, Ca- Calcium oxalate crystals.)

of specific therapeutic or pharmaceutical utility because of their swelling properties, especially gums and those containing an appreciable amount of mucilage, pectin or hemicellulose. The percentage of active chemical constituents in crude drugs is mentioned in air dried basis. The moisture content of the drugs should minimize in order to prevent decomposition of crude drugs either due to chemical change or microbial contamination. The ash values, extractive values and moisture content of root were determined. The results are depicted in Table 1. This study of Pharmacognostic including physico-chemical parameters is meant for proper identification of the plant, adulterant detection and also compilation of quality control standards of crude drugs. Thou this plant have been a long history of traditional medicine for various human ailments, it is important to standardize it for use as a drug.

Various numerical standards reported in this work along with the diagnostic microscopic features and the Pharmacognostical constants for the root of this plant

could be very helpful for the proper compilation of an appropriate monograph for its proper identification and also its right execution in human health service.

Conclusion

In this present studies which reported first time, suggest a methodical approach to macroscopic, microscopic characters and physiochemical screening of *Zyziphus oenoplia* (L.) Mill root in the field and powdered form can serve as a basis for proper identification, collection and investigation. These parameters ensures the quality of the drug and also use as reference material in preparation of herbal monograph of this plant for better commercial use in pharmaceutical industry.

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