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MORPHOLOGICAL EXPRESSIONS IN SEEDLINGS OF *OROXYLUM INDICUM* (L.) VENT. – A WELL KNOWN MEDICINAL TREE OF TROPICS AND SUBTROPICS

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Abstract. The main aim of the present study was to record morphological expressions and taxonomic significance in seedlings of *Oroxylum indicum*. Seedlings have been observed up to 5th leaf stage for 84 days from morpho-taxonomic view point. Seedling germination was *Macaranga* type. The morphological attributes found taxonomically significant are collet (with distinct ring), paracotyledon blade (anisocotylar, reniform, bilobed and deeply emarginated), glabrous hypocotyl and epicotyl and leaves simple up to 4th leaf stage, but compound afterwards.

Key words: *Oroxylum indicum*, seedling, morphology

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Introduction

Oroxylum indicum (L.) Vent. (family Bignoniaceae Juss.) also known as Broken bones tree, Indian trumpet flower, Midnight horror, Shivnak, Sonapatha and Shyonaka. The tree is a night-bloomer and flowers are adapted to natural pollination by bats. *O. indicum* is being used as medicinal herb for thousands of years without any known adverse effects (Ahad *et al.* 2012). Destructive and non-sustainable collection methods coupled with low regeneration and habitat destruction have posed serious threats to the survival and availability of this highly useful tree. The existence of *O. indicum* in natural population is highly threatened and it has been categorized as endangered and vulnerable by the Government of India (Ravikumar & Ved 2000; Saraf *et al.* 2013). There have been number of scientific studies conducted to evaluate the toxic effects of the plant. Almost all the studies have shown that *O. indicum* is not toxic to humans and experimental animals even up to high doses. Every part of this tree possesses medicinal value. It is an important herb in Ayurvedic medicine and indigenous medical system. In Indian system of medicine it has been used as a single drug or as a component of certain poly-herbal drug preparations. It is an active ingredient of well known Ayurvedic formulations like *Amartarista, Awalwha, Brahma rasayana, Bruhat pancha, Chyawanaprasha, Dantyadarishta, Dasamoola, Dhanawantara ghrita, Mulayadi kwath, Narayana taila* and *Shyonaka patpak*. Different parts of the plant are found to have anti-allergic, anti-anorexic, anti-arthritic, anti-bacterial, anti-bronchitic, anti-cancer, ant-carcinogenic, anti-diabetic, anti-fungal, anti-helminthic, anti-hepatotoxic, anti-inflammatory, anti-leucodermatic, anti-microbial, anti-oxidant, anti-proliferative, anti-rheumatic, anti-tumor, anti-tussive, anti-ulcer, anti-viral, immuno-stimulating and gastro-protective activity. The root and stem bark are also used in the treatments of diarrhoea, dysentery, erythema, gastralgia, hoarseness, infantile, measles, sore throat, urticaria, snake-bite and scorpion-sting. The seeds are found effective against gastralgia and also used as purgative. The fruits are found good in diseases of the heart, throat and piles. The tree is often grown as an ornamental for its strange appearance. Materials used include the wood, tannins and dyestuffs. It is also a plant with edible leaves and stems. Young shoots and unripe fruits are eaten as vegetable. The tender fruits are carminative and used in the treatment of stomach trouble in North East India. The tree is also frequently lopped for fodder. Wood of the

tree is used to make match boxes. Stem bark and fruits of the tree are used as mordant and yield tannin and dye. Sathe *et al.* (2013) reported true vivipary in *O. indicum*.

Literature survey revealed that comprehensive information on seedling morphology of *O. indicum* is not available (Dey *et al.* 1978; Vasanth *et al.* 1990; Ali *et al.* 1998; Bhuyan *et al.* 2000; Mao 2002; Gokhale & Bansal 2006; Kamkaen *et al.* 2006; Dinda *et al.* 2007; Yin *et al.* 2007; Zaveri & Jain 2007; Tenpe *et al.* 2009; Vikas *et al.* 2009; Babu *et al.* 2010; Islam *et al.* 2010; Lawania *et al.* 2010; Mishra *et al.* 2010; Zaveri & Jain 2010; Bisht *et al.* 2011; Harminder *et al.* 2011; Kumar *et al.* 2011; Pande & Gupta 2011; Radhika *et al.* 2011; Ahad *et al.* 2012; Dwivedi & Boro 2012; Najar & Agnihotri 2012; Deka *et al.* 2013; Jasrai *et al.* 2013; Karnati *et al.* 2013; Raghu *et al.* 2013; Sathe *et al.* 2013; Bharali *et al.* 2014; Joshi *et al.* 2014; Ramaswamy *et al.* 2014; Saboo *et al.* 2014). With this background the present study is aimed. This study would help in conservation and quality management of an endangered and vulnerable medicinal plant – *O. indicum*.

Material and methods

The mature capsules of *O. indicum* were collected in the month of April and May 2014 before dehiscence from Ayurveda Garden, Banaras Hindu University, Varanasi, Uttar Pradesh, India (25°10'-25°37' N, 82°11'-83°01' E, 80.19 m a.s.l.). The capsules were furled and seeds were taken out. Seeds grown in garden soil during June to September 2014 in the green house of the Experimental Botanic Garden of the Department of Botany, Udai Pratap College (Autonomous), Varanasi, India and morphological expressions of the seedlings have been recorded. The average minimum and maximum temperature during observation period were 26.4±1 °C and 34.0±1 °C. Different developmental stages were considered for preparing a complete morpho-taxonomic description out of ten individuals. For seedling observation binocular microscope (MSZ-B1) was used. All the specimens at different leaf stages were documented in the form of herbarium sheets which have been deposited in the Herbarium (15.09.2014, AKS 014), Department of Botany, Udai Pratap College (Autonomous), Varanasi, Uttar Pradesh, India. The gross morphological features of seedlings were described following the terminology proposed by Vogel (1980). For seedling description Singh (2012) and Singh *et al.* (2014) were followed. Days and dates of appearance of paracotyledons and leaves upto 5th leaf stage (Tab. 1), and length of root, hypocotyl and epicotyl from paracotyledon stage upto fifth leaf stage and root/shoot length values (R/S values) (Tab. 2) were also recorded.

Results and discussion

Taxonomic description

Oroxylum indicum (L.) Vent. Decas. Gen. Nov. 8. 1808; Clarke in Hook. f. Fl. Brit. India 4: 378. 1884. *Bignonia indica* L. Sp. Pl. 625. 1753.

Deciduous soft wooded tree, 4-7 m tall. Bark thick, yellowish-grey. Leaflets 2-4 pairs, 7.0×3.5-8.0 cm, ovate-elliptic, acuminate, base obliquely rounded-obtuse. Petiolules up to 4.5 cm long. Flowers foetid, in large erect racemes. Calyx 2.0-2.5 cm long, campanulate, fleshy; lobes 5, subequal, rounded, obtusely toothed, dull purple. Capsules 40-60 cm long, flat, sword-shaped. Seeds many, 5-7 cm long, margins winged all round except base.

Flowering: August-November; Fruiting: December-June.

Geographical distribution: Native to the Indian subcontinent, Indonesia, Malayasia, Philippines, South China and Sri Lanka.

Seedling morphology

Seedlings of *Macaranga* type (Vogel 1980). Primary root non fibrous, tap root, much branched, more developed than shoot, glabrous with secondary root scars, light yellow, terete in cross section, much thick later, 1.5 cm long at paracotyledon stage, 3.1, 4.3, 5.8, 17.0 and 28.0 cm long at 1-5th leaf stages respectively; secondary roots same as primary root. Collet distinct with distinct ring, milky white, glabrous and slightly swollen (Fig. 1 C; Fig. 2 C).

Tab. 1. Days and dates of appearance of paracotyledons and leaves of the seedling of *Oroxylum indicum* after sowing on 17.06.2014.

Stages	Days	Dates
Paracotyledon	7	24.06.2014
First leaf	11	28.06.2014
Second leaf	19	06.07.2014
Third leaf	34	21.07.2014
Fourth leaf	52	08.08.2014
Fifth leaf	84	06.09.2014

Tab. 2. Root, hypocotyl and epicotyl length and R/S values of the seedling of *Oroxylum indicum* from paracotyledon stage to fifth leaf stage. Values represent means of ten replicates.

Stages	Root (cm)	Hypocotyl (cm)	Epicotyl (cm)	R/S values
Paracotyledon	1.5	1.5	-	1.00
First leaf	3.1	2.7	2.0	0.65
Second leaf	4.3	2.7	3.1	0.74
Third leaf	5.8	2.7	4.2	0.84
Fourth leaf	17.0	2.7	10.0	1.34
Fifth leaf	28.0	2.7	12.5	1.84

Hypocotyl green, glabrous with brown lenticels; button shaped, 0.1 cm in diameter (Fig. 1 F); terete in cross section, 1.5 cm long at paracotyledon stage and 2.7 cm long at 1st to 5th leaf stages.

Paracotyledons two, opposite, phanerocotylar, slightly anisocotyler, foliaceous, persistant upto 5th leaf stage, exstipulate and petiolate. Petiole green, glabrous, terete and channeled in cross section, 0.8 cm long. Paracotyledon blade reniform, bilobed, blade lobes slightly unequal, 2.0×3.0 cm, mean length/width ratio (L/W) 0.7, base rounded, apex deeply emarginate, margins entire, adaxial surface dark green, abaxial surface light green, both surface glabrous (Fig. 1 A; Fig. 2 C-E). Venation acrodromous; multicostate reticulate; 5 primary veins distinct, central primary vein dichotomously divided, one branch enters into one lobe each, apically looped with each lateral vein on each side of central primary vein (Fig. 1 A).

Epicotyl green, glabrous, terete in cross section (Fig. 1 D), 2.0, 3.1, 4.2, 10.0 and 12.5 cm long at 1-5th leaf stages respectively. Internodes (at 5th leaf stage): first – 1.8 cm, second – 1.8 cm, and third – 1.0 cm long, later internodes elongating.

First leaves two, opposite, simple, exstipulate and petiolate (Fig. 2 D, E). Petiole green, glabrous, terete and channeled and winged in cross section (Fig. 1 E), 0.8 cm long. Leaf blade elliptical, 3.5×1.3 cm, mean L/W 2.7, base cuneate, apex acute, margins slightly wavy and undulate, adaxial surface dark green, abaxial surface light green, both surface glabrous. Venation brochidodromous; unicostate reticulate, single primary vein distinct, reaches to blade apex, 14 secondary veins distinct, 7 on each side of primary vein, alternate and looped at margins. Second leaf same as 1st leaf (Fig. 1 B). Third and fourth leaves same as 1st leaves but petiole base pulvinate and leaf blade base rounded, apex apiculate, margins wavy. Fifth leaf compound; trifoliate, leaflets unequal, lateral leaflets with oblique base and central leaflet base cuneate (Fig. 1 B; Fig. 2 E). Young leaves dark green but mature leaves with purple patches.

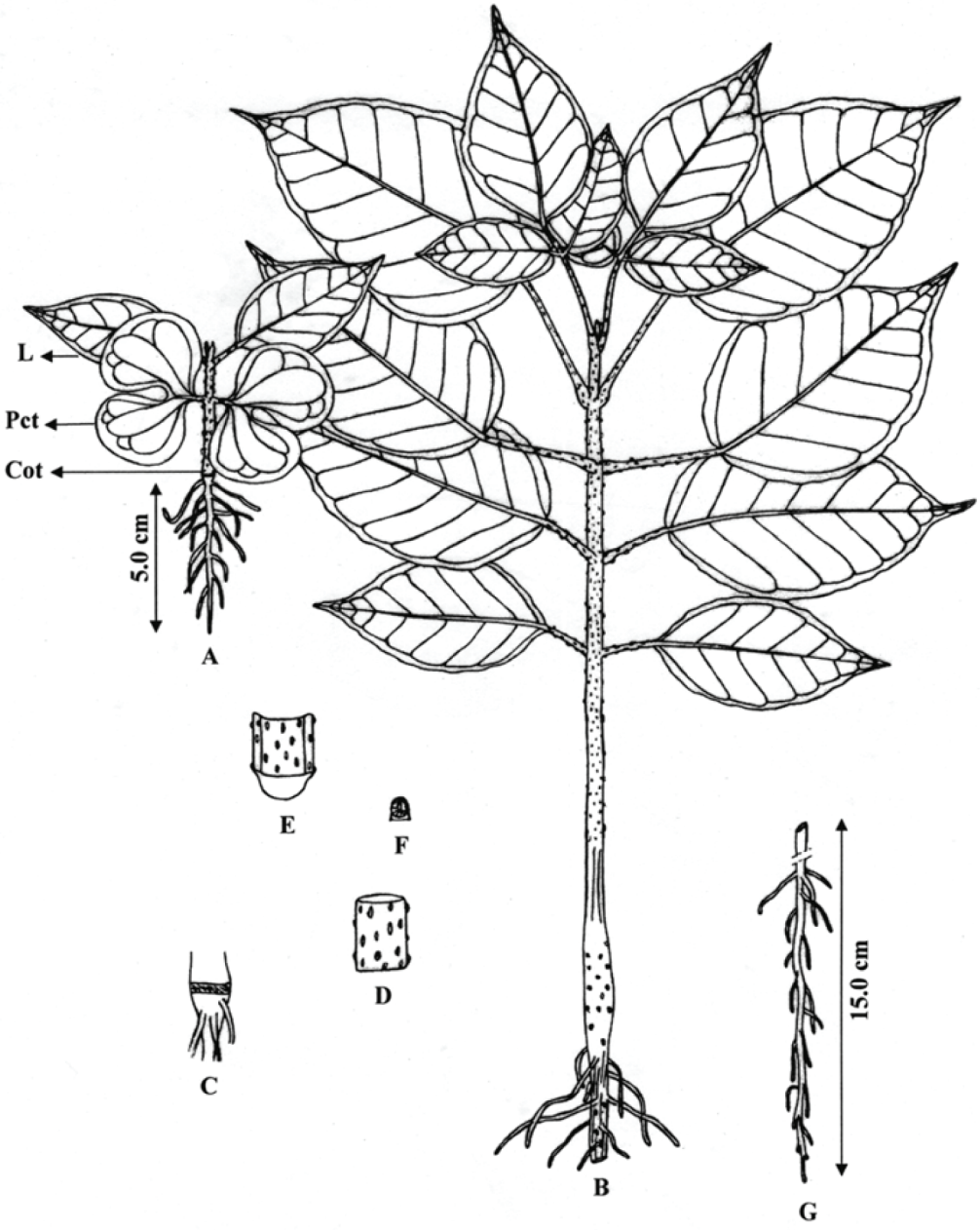

Fig. 1. Seedlings of *Oroxylem indicum* at different leaf stages and their parts: **A** – first leaf stage (**Cot** – collet; **L** – first leaf; **Pct** – paracotyledon); **B** – sixth leaf stage; **C** – collet; **D** – part of epicotyls; **E** – part of leaf petiole; **F** – single lenticel; **G** – main and lateral roots.

O. indicum is propagated naturally by seeds, which germinate in the beginning of the rainy season (June to September). Seedlings require moderate shade in the early stages. However, the seed set is poor and seed viability is low. Problems related with its natural propagation and indiscriminate exploitation for medicinal purpose has pushed *O. indicum* to the list of endangered plant species of India (AHAD *et al.* 2012).

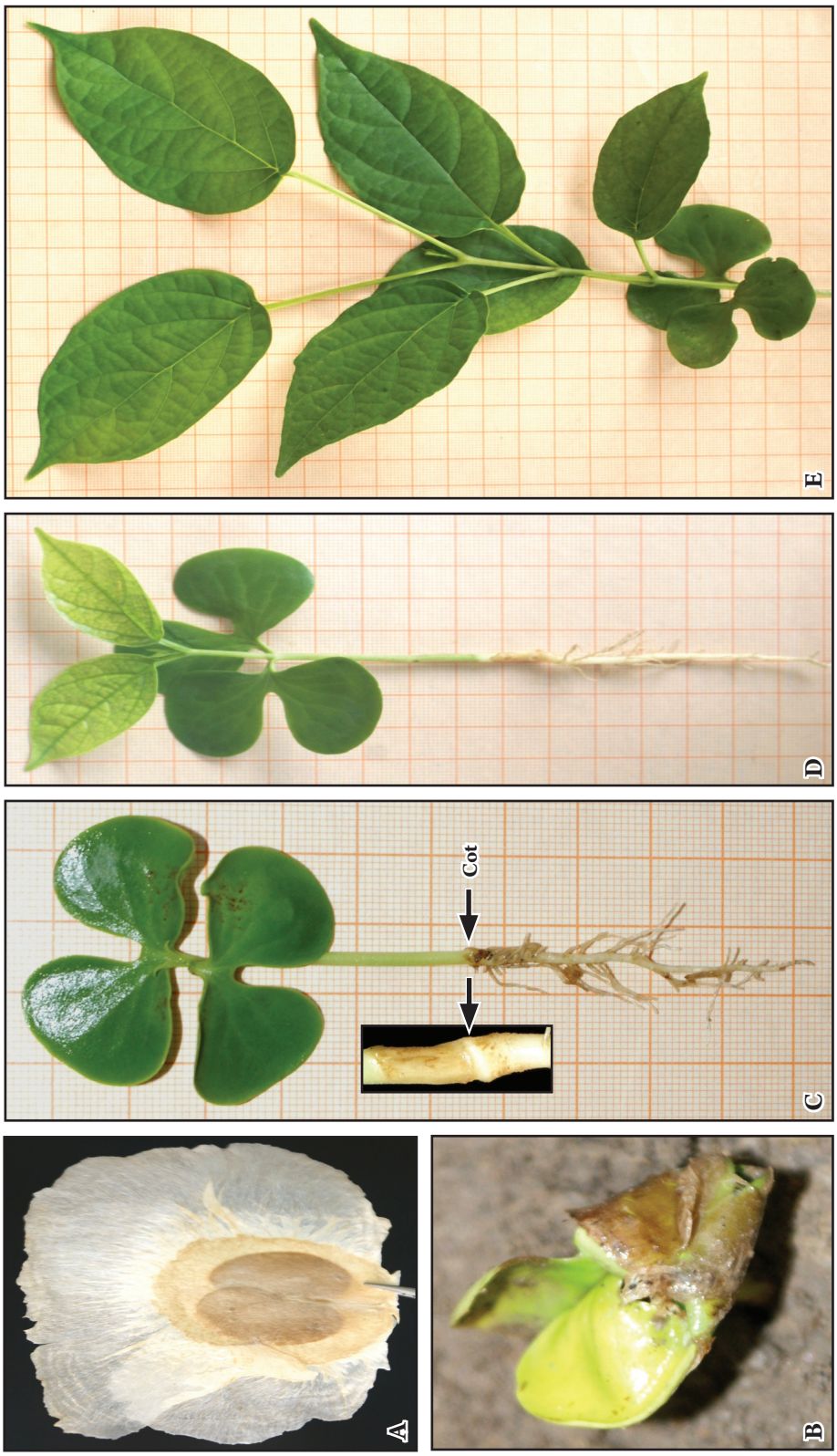

Fig. 2. Seed and stages of seedling development of *Oroxylum indicum*: **A** – seed; **B** – germinating seed; **C** – seedling at paracotyledon stage (**Cot** – collet); **D** – seedling at first leaf stage; **E** – seedling at third leaf stage.

Interest in medicinal plants as a re-emerging health aid has been fuelled by the rising costs of prescription drugs in the maintenance of personal health and well-being. Herbal-based traditional medicine has become popular in developed countries in recent years, and its use is likely to increase in the coming years. This system has advantages over the modern medicine as it is prophylactic. Therefore, it would be a wise step if most of medicinal plants, particularly those which are in high demand and rare should be cultivated. The manufacturers and the practitioners of traditional medicines depend solely on the procurement of medicinal plants from the wild, as cultivation of most of the medicinal plants has not gained momentum (SINGH *et al.* 2008). Domestication and cultivation of medicinal plants is one of the viable options to meet the growing demand from the industries and to reduce the extraction pressure in the natural habitats of medicinal plants. Knowledge of seedling ecology, morphology and taxonomy of medicinal plants could be beneficial for their cultivation and management.

The herbal drug industries are very fast growing sectors in the international market. Conservation and cultivation of medicinal and aromatic plants has many aspects and is a subject which needs constant probing and experimentation, to meet the stringent standards of world market in pharmaceutical products (SRIVASTAVA 2005; SINGH *et al.* 2014).

The commercial exploitation of the wild plant may leads to its extinction. Only little information is available on seedling morphology of most the medicinal plants. Further, seedlings also represent the final stage in the process of regeneration from seeds. Thus, understanding the relationship between the morphology and function of seedlings can also provide an insight into the regenerative strategies of species in different vegetation types and assist in constructing appropriate models for biodiversity management (SINGH *et al.* 2014).

The main aim of the present study is to record morphological expressions in seedlings of *O. indicum*. Seedlings attain 5th leaf stage in 82 days. During present observation some key morpho-taxonomic attributes have been recorded, which can be exploited for differentiation and identification of *O. indicum* at seedling stage on spot in nature and in the management of seedling bank. These key attributes are-seedling germination *Macaranga* type (cotyledons long persistent, leaf-like, free and spread in the air and have photosynthetic functions.) in the sense of VOGEL (1980), collet slightly swollen with distinct ring, hypocotyl green; glabrous with brown lenticels, paracotyledon blade; anisocotylar, reniform, bilobed, deeply emarginate; venation acrodromous, epicotyl green and glabrous, leaves simple upto 4th leaf stage, 5th and subsequent leaves are compound. First and second leaf petioles are winged and their blades with brochidodromous venation. Third and fourth leaves are the same as 1st and 2nd leavs but petiole bases are pulvinate, blade bases are rounded, apexes are apiculate and margins are wavy. Fifth and subsequent leaves compound with unequal leaflets and oblique base but central leaflet base cuneate. Paracotyledon blade and leaf blade very much distinct in shape and venation up to 4th leaf stage.

In addition to general morphological events, days and dates of appearance from paracotyledon stage upto 5th leaf stage and length of root, hypocotyl and epicotyl at paracotyledon stage and at different leaf stages up to 5th leaf stage along with R/S values have also been recorded (Tabs. 1 & 2). From Tab. 1, it is evident that paracotyledons appear after 7th days of sowing. Leaves appear at the intervals of 4, 8, 15, 18 and 32 days from 1st to 5th leaf stage respectively. Seedling reaches to 5th leaf stage after 84 days. The root length was minimum (1.5 cm) at paracotyledon stage and maximum (28.0 cm) at 5th leaf stage. The hypocotyl length was almost same from 1st leaf stage to 5th leaf stage (2.7 cm), whereas the length of epicotyl was minimum (2.0 cm) at 1st leaf stage and maximum (12.5 cm) at 5th leaf stage. At paracotyledon stage R/S value was 1.0, but at 1st, 2nd and 3rd leaf stages it was <1, whereas at 4th and 5th leaf stages it was >1.

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