

PHARMACOGNOSTICAL AND PRELIMINARY PHYTOCHEMICAL STANDARDIZATION OF A SIDDHA DRUG "PULA"

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Many of the herbals mentioned in the ISM pharmacopoeia remain untapped for the therapeutic potentials. *Securinega virosa* (Roxb.ex Willd.)Baill. (Euphorbiaceae), which is known as 'Pula' in Siddha system of medicine, is one among them. Almost all parts of the plant have been claimed to be medicinally important. Literature review shows that this drug has not been given due attention in pharmacopoeia. The present study furnishes a comprehensive data for the anatomical features of the leaf, petiole, stem, root, wood and bark of *S. virosa*. Fluorescence characters, ash value, extractive values of root and leaves of *S. virosa* have been determined. Qualitative organic analysis of root and leaf powders in alcoholic extracts has been performed. The results of the investigation are to provide dependable diagnostic features of the vegetative organs for the proper standardization of the drug.

Keywords : Pharmacognosy; Pula; *Securinega virosa*.

Introduction

Securinega virosa (Roxb.ex Willd.)Baill. is a well branched shrub that grows on hills 200 m high and on base slopes. It is used by traditional healers to treat diarrhoea¹, epilepsy, oedema, syphilis². It is also used for the treatment of liver disease³, inflammation and pain². Its extracts are used for the expulsion of round worms². In addition some of these biological activities have been confirmed. For example, a 0.03% of methanol extract of the fruit pulp inhibited the growth of *Trichophyton mentagrophytes* and *Candida albicans*⁴. While extracts of leaves exhibited antibacterial activity against *Staphylococcus aureus* and *Pasteurella pestis*⁴. Similarly, a 1:1 aqueous ethanolic extract of the aerial parts was shown to have antispasmodic activity against both histamine and acetylcholine induced spasms⁵.

S. virosa is distinguished in folklore claims many human ailments. A lacuna in the pharmacopoeia for pharmacognostical parameters of the plant prompted the present investigation.

Materials and Methods

S. virosa was collected from Courtallam hills, Western Ghats of Tamil Nadu and identified with the help of flora of Tamil Nadu, India⁶. Anatomical studies of stem, root, leaves and bark carried out employing the customary techniques of microtomy and photomicrography. The materials were macerated using Jeffrey's solution to study the cellular components. Fluorescent analysis of the leaf and root powder and the extracts of the leaf in different solvents were carried out according to the methods of Chase and

Pratt⁷, physico-chemical characters were determined by standard methods.

Observations

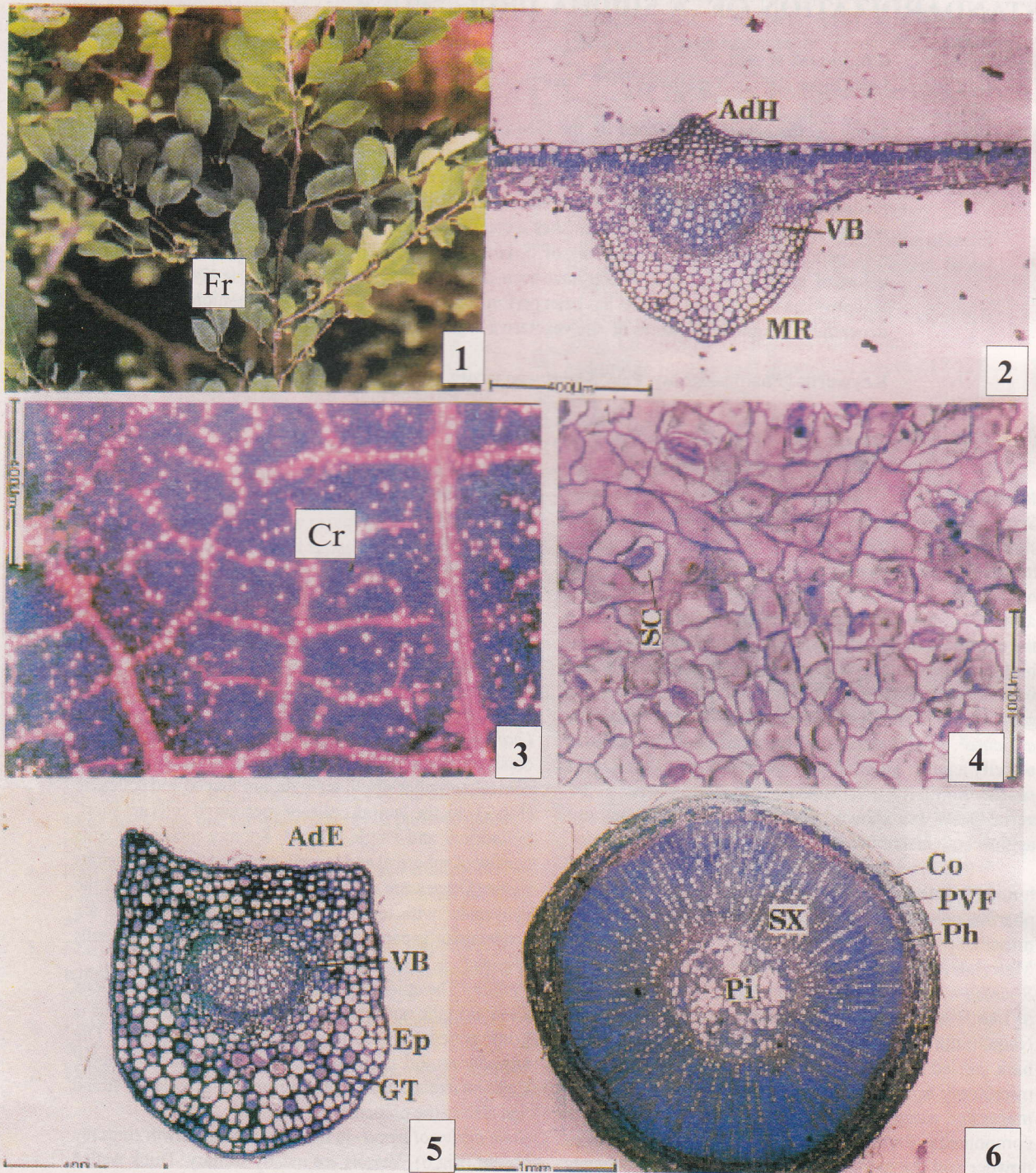
Exomorphology- *S. virosa* is characterized by unarmed branches. Leaves are thinly pubescent. Flowers are greenish yellow and occur in axillary clusters. Male flowers are numerous, female flowers are whitish - generally few. Fruits are globose with persistent calyx (Fig.1).

Microscopic features

Leaf - Leaf structure is mesomorphic, hypostomatic and dorsiventral; midrib is abaxially hemispherical and adaxially conical; vascular strand is single, arc shaped and collateral (Fig.2). In the lamina palisade mesophyll cells are short, cylindrical and single layered. Spongy mesophyll cells consist of spherical and loosely arranged parenchymatous tissue. Druses are abundant all along the veins and the vein ends with terminal sclereids (Fig.3). Stomata are predominantly paracytic with two unequal subsidiary cells (Fig.4).

Petiole - The epidermal layer is thin and less conspicuous all around the outline of the petiole. The ground tissue is homogeneous, parenchymatous and compact. The vascular strand is single, bowl - shaped and collateral. Calcium oxalate druses are seen around the vascular strand and also in the phloem cells (Fig.5).

Stem and root wood - In the stem wood, growth rings are absent (Fig.6). Vessels are mostly solitary. Thick walled lignified, non - septate libriform fibres are present. Xylem



Figs. 1-6. *Securinega virosa* (Roxb. ex. Wild.) Baill.

Fig.1. A shoot with fruits; **Fig. 2.** T.S. of leaf through midrib; **Fig. 3.** Cleared leaf under polarized light showing the occurrence of crystals along the veinlets; **Fig. 4.** Leaf epidermis showing paracytic stomata; **Fig. 5.** T.S. of old stem showing secondary growth.

(AdE- Adaxial epidermis; Cr- Crystals; Co-Cortex; AdH-Adaxial hump; Fr-Fruit; Ep-Epidermis; GT-Grund tissue; MR-Midrib; Ph-Phloem; Pi-Pith; PVF-Peri vascular fibres; Sc-Subsidiary cells; Sx- Secondary Xylem)

Table 1. Physico-chemical constants of *S. virosa*.

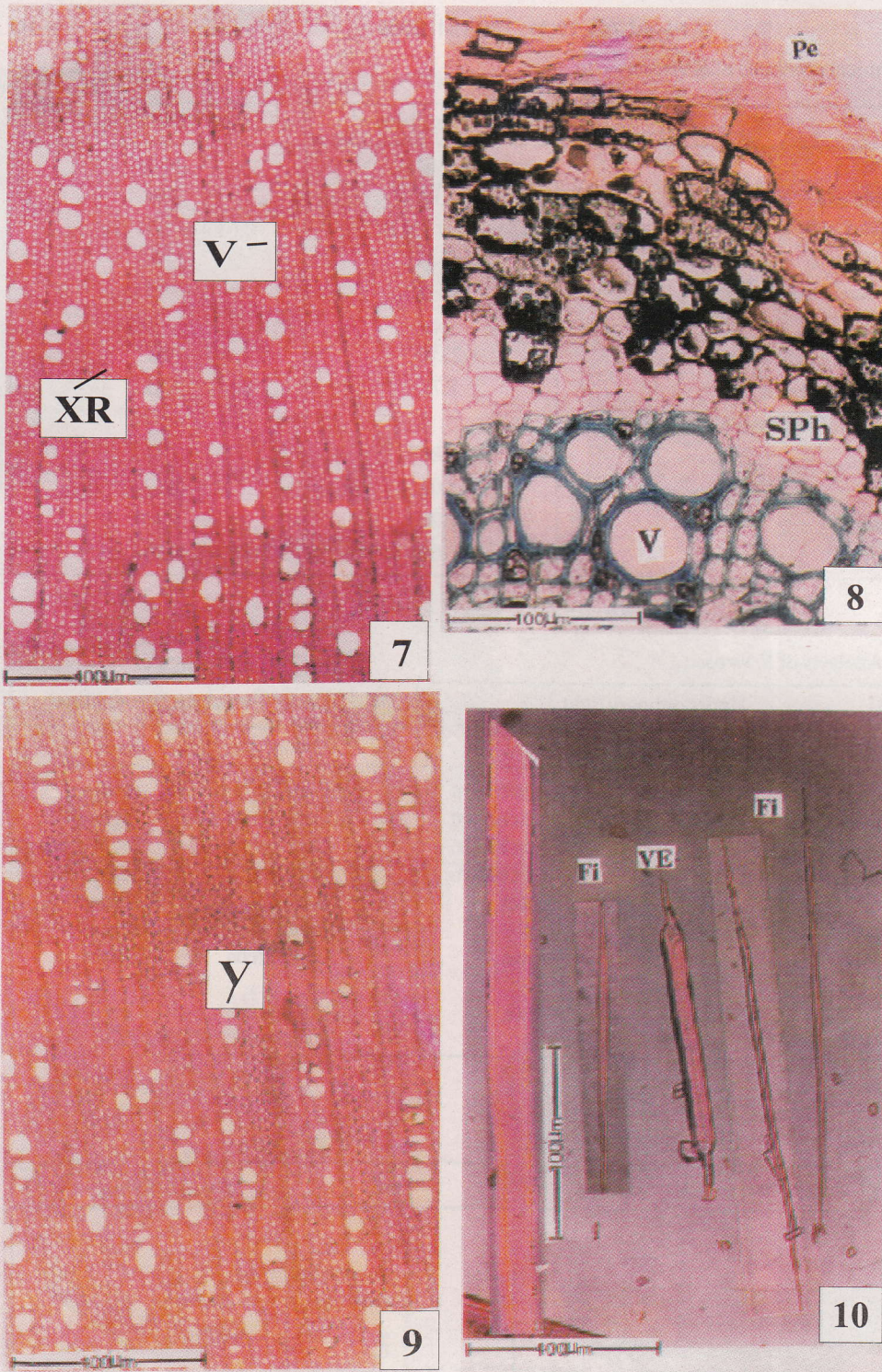
S.No	Parameter	Leaf Value (% w/w)	Root Value (%w/w)
1.	Ash Value	9.6	10.78
2.	Water soluble ash	5.16	4.94
3.	Alkalinity of water soluble ash	0.5 ml of 0.1 N HCl/g	0.6 ml of 0.1 N HCl/g
4.	Acid insoluble ash	0.3	1.5
5.	Moisture content	11.13	10.3
6.	Solubility		
	a. Water soluble extractive	23.96	9.8
	b. Alcohol soluble extractive	8.76	6.85
7.	Extractive Value (successive extraction)		
	a. Hexane	2.84	1.14
	b. Benzene	1.09	0.19
	c. Chloroform	0.96	0.44
	d. Alcohol	0.64	0.36

Table 2. Fluorescent Analysis of *S. virosa*.

Sl.No	Treatment	Under ordinary light		Under ultra-violet light (254 nm)	
		Leaf	Root	Leaf	Root
1.	Hexane extract	Light brown	Light brown	Green	Brown
2.	Chloroform extract	Greenish yellow	Light brown	Green	Light green
3.	Alcohol extract	Green	Light brown	Dark green	Light green
4.	Benzene extract	Green	Light brown	Dark green	Light green
5.	Water extract	Brown	Brown	Green	Green
6.	Acetone extract	Brown	Red	Green	Green
7.	Powder + 1N NaOH in water	Green	Brown	Light green	Brown
8.	Powder + 1N HCl	Brown	Brown	Light green	Green
9.	Powder + H ₂ SO ₄ (1:1)	Dark green	Red	Dark green	Red

Table 3. Qualitative Organic Analysis of *S. virosa*.

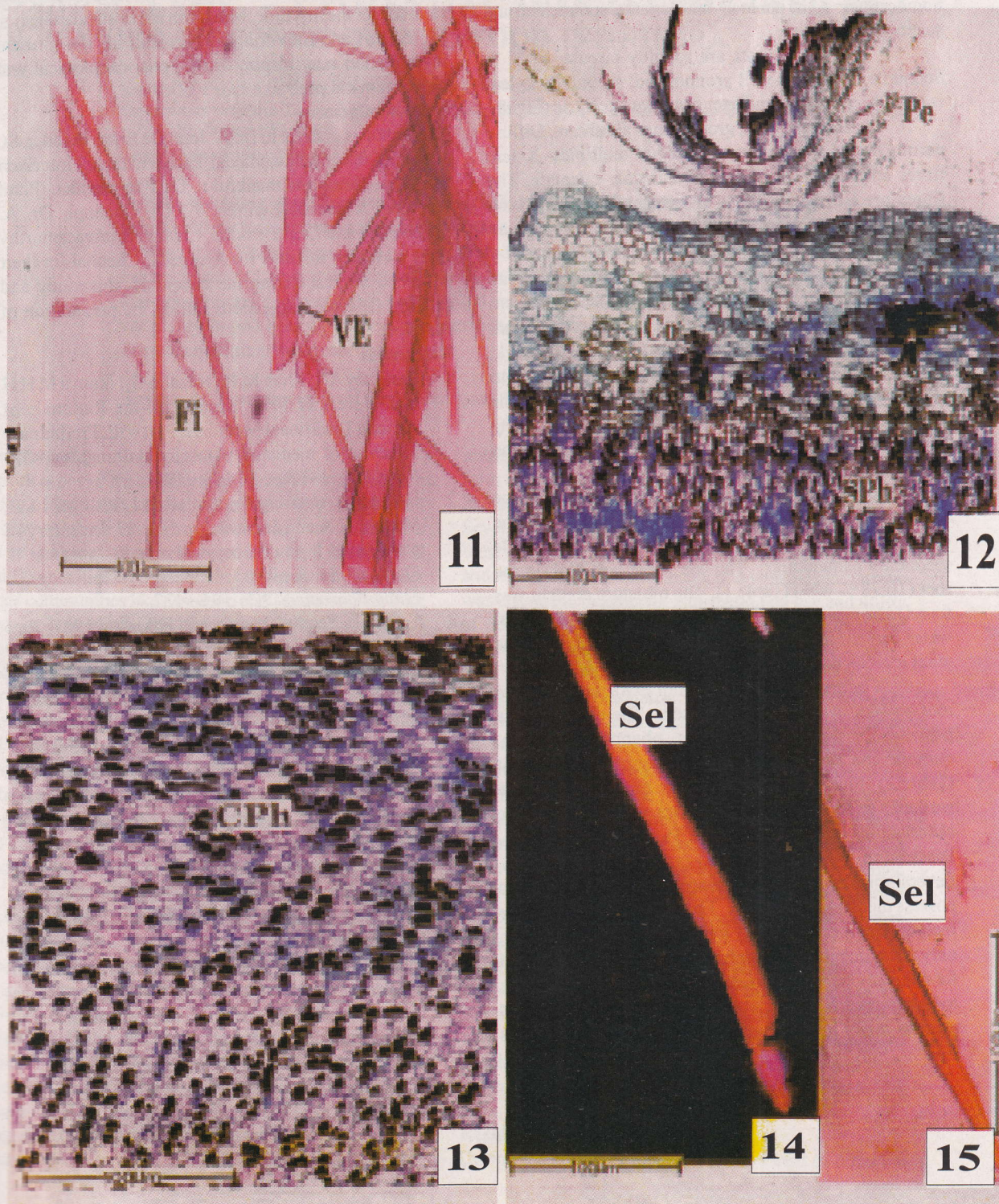
S.No	Contents of the alcoholic extract	Leaf	Root
1.	Steroid	+	+
2.	Triterpene	+	+
3.	Alkaloid	+	+
4.	Phenol	+	+
5.	Flavonoid	+	+
6.	Coumarin	+	+
7.	Quinone	+	+
8.	Furan	-	-
9.	Tannin	-	-
10.	Glucoside / Sugars	+	+



Figs. 7-10. *Securinega virosa* (Roxb.ex. Wild.) Baill.

Fig.7. T.S. of stem wood; **Fig.8.** T.S. of old root showing secondary growth; **Fig.9.** T.S. of old root wood; **Fig. 10.** Fibres and vessel element of stem wood.

(Fi- Fibre; Pe-Periderms; SPh-Secondary phloem; V-Vessel; VE-Vessel element; XR- Xylem ray)



Figs. 11-15. *Securinega virosa* (Roxb.ex. Wild.) Baill.

Fig.11. Macerated root xylem showing vessels and fibres; **Fig.12.** T.S. of stem bark; **Fig.13.** T.S. of root bark; **Fig.14.** Fibre sclereid of stem bark; **Fig.15.** Fibre sclereid of root bark.

(Co-Cortex; Cph-Collapsed phloem; Fi-Fibre; Pe-Periderm; Sph-Secondary phloem; VE-Vessel element)

rays mostly narrow and short, uni or biseriata and homocellular. Axial parenchyma is sparse, apotracheal and scanty (Fig. 7, 10).

The lateral roots are roughly circular in T. S. Secondary growth is well pronounced; secondary xylem is initially concentric and later becomes excentric (Fig. 8).

In the root xylem, growth rings are absent. Diffuse porous. Pores are solitary or in radial multiples. Xylem fibres are narrow, thick walled and non-septate. The fibres are filled with large dense starch grains. Axial parenchyma is apotracheal and scanty. Xylem rays are uni or biseriata, very high and heterocellular (Fig 9, 11).

Stem and root bark - In the stem bark, periderm is broad with distinct phellem and phelloderm. Secondary phloem is broad and distinct (Fig 12). Spindle shaped sclereids are scattered among phloem element (Fig. 14). In the root bark, periderm is thin. Secondary phloem is distinguished into outer broad zone of tannin filled parenchyma cells and the inner zone consists of tannin free intact phloem cells. Long narrow fibre-like sclereids are seen in the phloem region (Fig 13, 15).

Preliminary Phytochemistry - The water solubility value was found high for the leaf of *S. virosa* (23.96 w/w) due to the presence of polar compounds and water soluble secondary metabolites. The ash value was high in the root (10.78 % w/w) (Table 1). The moisture content was found to be maximum (11.3 %) and was indicative of total water content and of the volatilizable matter from the plant ingredients (Table 1).

For qualitative test, alcohol extract of leaf and root powder of the plant was used. The extracts were positive for steroids, triterpenes, phenols, coumarin, quinone and sugars. Furan and tannin were absent (Table 2). Fluorescence characters of the plant were studied under day light and ultra violet light. The observations are tabulated (Table 3).

Discussion

Several members of Euphorbiaceae have been found to possess medicinal properties. The Tribe *Phyllanthoideae* includes many economically important species. However, only a few members of Euphorbiaceae have been tapped for clinically active compounds and a bulk of taxa remains to be explored. *S. virosa* is one such plant, which seems to possess many interesting therapeutic compounds. The

anatomy of *S. virosa* shows certain histological features, recorded for the first time, which will aid in standardization of the leaf and root drugs. The result of the present investigation may inspire further pharmacological and phytochemical probes.

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