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### Phytochemical studies of *Svensonia hyderbadensis* (walp.) Mold: A rare medicinal plant

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#### ABSTRACT

*Svensonia hyderbadensis* is a medicinal plant and included under the list of rare plant species. Phytochemical studies of stem and leaves of *Svensonia hyderbadensis* (Verbenaceae) revealed that the presence of secondary metabolites of flavonoids, phenols, glycosides, lignin, tannins, terpenoids, saponins, steroids and coumarins; and absence of alkaloids. The plant parts consist of secondary metabolites and the result helpful for the isolation of active principle to treat the hepatotoxic diseases.

**Keyword:** Secondary metabolites, Phytochemical studies, *Svensonia hyderbadensis*, Methanol, Ethyl acetate.

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#### INTRODUCTION

*Svensonia hyderbadnesis* is a rare medicinal plant belongs to the family Verbenaceae used to cure hepatotoxic diseases ([antihepatotoxicity.blogspot.com](http://antihepatotoxicity.blogspot.com)). Since ancient times, people have been exploring the nature particularly plants in search of new drugs. This has resulted in the use of large number of medicinal plants with curative properties to treat various diseases [1]. Nearly 80% of the world's population relies on traditional medicines for primary health care, most of which involve the use of plant extracts [2]. In India, almost 95% of the prescriptions were plant based in the traditional systems of Unani, Ayurveda, Homeopathy and Siddha [3]. The study of plants continues principally for the discovery of novel secondary metabolites. Around 80% of products were of plant origin and their sales exceeded US \$65 billion in 2003 [4]. The active principles found in plants are used to treat different ailments. Most of these active principles are derived from secondary metabolites. Hence the present study has been carried out to screen the *Svensonia hyderbadensis* for secondary metabolites.

#### MATERIALS AND METHODS

##### Screening of Phytochemicals

Stem and leaves of *Svensonia hyderbadenesis* were collected from the forest of Mamandur nearly 20 KM to Tirupati, A.P. India, in the year 2010. The stem and leaves were air dried for

10 days and kept in the hot air oven at 60°C for 24-48 hs and ground to fine powder. This powder extracted with different solvents by using soxhlet apparatus. The preliminary tests, for the detection of secondary metabolites were carried out with hexane, ethyl acetate and methanolic extracts. 500 mg of each extract was dissolved in 100 ml of the respective solvent and filtered through Whatman No.1 filter paper. Thus, the filtrates obtained were used as test solutions for the following preliminary phytochemical screening tests.

### 1. Test for Flavonoids [5]

The test solution of the extract was dissolved in one ml of alcohol and then subjected to the following tests:

**a. Ferric chloride test:** A few drops of neutral ferric chloride solution was added to one ml each of above alcoholic solution. Formation of blackish red color indicates the presence of flavonoids.

**b. Shinoda's test:** To one ml of alcoholic extract, a small piece of magnesium ribbon or magnesium foil was added and a few drops of conc. HCl were added. Change in colour (from red to pink) shows the presence of flavonoids.

**c. Zinc-HCl reduction test:** A pinch of zinc dust and a few drops of conc. HCl were added to alcoholic extract. Magenta colour indicates the presence of flavonoids.

**d. Lead-acetate test:** To one ml of alcoholic extract, a few drops of aqueous basic lead acetate solution were added. Reddish brown bulky precipitate indicates the presence of flavonoids.

### 2. Test for Steroids [6]

The test solution of the extract was dissolved in 5 ml of chloroform separately and was subjected to the following tests:

**a. Salkowski test:** One ml of conc. Sulphuric acid was added to the above solution and allowed to stand for 5 minutes after shaking. Lower layer turning into red in colour indicates the presence of steroids.

**b. Liebermann-Burchard test:** To one ml of the extract, treated with chloroform, a few drops of acetic anhydride, one ml of conc. H<sub>2</sub>SO<sub>4</sub> was added from the walls of the test tube and allowed to stand for 5 minutes. Formation of reddish brown ring was formed at the junction of the two layers and the upper layer turns into green indicates the presence of steroids.

### 3. Test for Terpenoids

Fresh plant material was treated with 5 ml of 1% aqueous hydrochloric acid. After 3-6 hours, the extract was treated with 1 ml of Trim-Hill reagent (10 ml of acetic acid, 1 ml of 0.2% copper sulphate in water and 0.5 ml of concentrated hydrochloric acid) and heated on a water bath. The appearance of blue colour indicates the presence of diterpenoids while green colour indicates the presence of monoterpenoids.

### 4. Tests for Tannins [7]

The test solution of the extract was dissolved in minimum amount of water separately, filtered and filtrates were then subjected to the following test:

**a. Ferric chloride test:** To the filtrate, a few drops of ferric chloride solution was added. A blackish precipitate indicates the presence of tannins.

**b. Gelatin test:** To the filtrate, gelatin (Gelatin dissolves in warm water immediately) solution was added. Formation of white precipitate indicates the presence of tannins.

**c. Lead acetate test:** To the filtrate, a few drops of aqueous basic lead acetate solution were added. Reddish brown bulky precipitate indicates the presence of tannins.

#### **5. Tests for Glycosides [8]**

**a. Kellar Kiliani test:** The test solution of the extract was dissolved in glacial acetic acid and after cooling, 2 drops of ferric chlorides solution was added to it. These contents were transferred to a test tube containing 2 ml of concentrated sulphuric acid. A reddish brown colour ring observed at the junction of two layers.

**b. Conc. H<sub>2</sub>SO<sub>4</sub> test:** 1 ml of conc. H<sub>2</sub>SO<sub>4</sub> was added to 1ml of test solution and was allowed to stand for 2 minutes. The formation of reddish colour indicates the presence of glycosides.

**c. Moisch's test:** A mixture of Molisch's reagent and conc. H<sub>2</sub>SO<sub>4</sub> (1:1) was added to the test solution. Formation of reddish-violet coloured ring at the junction of two liquids indicates the presence of glycosides.

#### **6. Tests for Saponins**

The test solution of the extract was separately mixed with 20 ml of distilled water and then agitated in a graduated cylinder for 15 minutes. Foam formation indicates the presence of saponins.

#### **7. Test for Alkaloids [6]**

The test solution of the extracts was dissolved in chloroform and the solution was extracted with dil. HCl or dil. H<sub>2</sub>SO<sub>4</sub> and acid layer was taken and tested for presence of alkaloids.

**a. Mayer's test:** To the acidic solution, Mayer's reagent (Potassium mercuric iodide solution) was added. Cream coloured precipitate indicates the presence of alkaloids.

**b. Wagner's test:** To the acidic solution, Wagner's reagent (Iodine in potassium iodide) was added. The formation of reddish brown precipitate indicates the presence of alkaloids.

**c. Dragendorff's reagent test:** 2 ml of Dragendorff's reagent and 2 ml of dilute HCl were added to the test solution. An orange-red coloured precipitate indicates the presence of alkaloids.

#### **8. Test for Phenols [6]**

**a. Phenol test:** When 0.5 ml of FeCl<sub>3</sub> (w/v) solution was added to 2 ml of test solution, formation of an intense colour indicates the presence of Phenols.

**b. Ellagic acid test:** The test solution was treated with few drops of 5% (w/v) glacial acetic acid and 5% (w/v) NaNO<sub>2</sub> solution. The solution turns muddy or niger brown precipitate occurs in the extract indicates the presence of phenols.

### 9. Test for Quinones

The test solution of the extract was treated separately with alcoholic potassium hydroxide solution. Quinones give coloration ranging from red to blue.

### 10. Tests for Lignin [6]

**a. Labat test:** The test solution was mixed with gallic acid, it develops olive green colour indicating the positive reaction for lignins.

**b. Lignin test:** Formation of red colour, when 2% (w/v) furfuraldehyde was added to the test solution indicates the presence of lignin.

### 11. Coumarins

1g of plant powder is placed in a tube in the presence of a few drops of distilled water. The tube is covered with paper soaked in NaOH is diluted and boiled. Yellow fluorescence indicates the presence of coumarins after examination under ultra-violet [9].

**Table 1: Preliminary screening of secondary metabolites from stem and leaf extracts of *Svensonia hyderbadnesis***

| Primary Screening for secondary metabolites |                        | <i>Svensonia hyderbadnesis</i> |               |          |        |               |          |
|---|------------------------|--------------------------------|---------------|----------|--------|---------------|----------|
|   |                        | Stem                           |               |          | Leaf   |               |          |
|   |                        | Hexane                         | Ethyl acetate | Methanol | Hexane | Ethyl acetate | Methanol |
| Alkaloids                                   | Mayers test            | -                              | -             | -        | -      | -             | -        |
|   | Wagners test           | -                              | -             | -        | -      | -             | -        |
|   | Drgendorff's test      | -                              | -             | -        | -      | -             | -        |
| Flavonoids                                  | FeCl <sub>3</sub> test | -                              | -             | +        | -      | +             | +        |
|   | Shinoda's test         | -                              | -             | +        | -      | +             | +        |
|   | Zin-HCl reduction test | -                              | -             | +        | -      | +             | +        |
|   | Lead acetate           | -                              | -             | +        | -      | +             | +        |
| Phenols                                     | Phenol test            | +                              | +             | +        | -      | -             | +        |
|   | Ellagic test           | +                              | +             | +        | -      | -             | +        |
| Glycosides                                  | Kellar kilani test     | -                              | +             | -        | +      | +             | -        |
| Lignin                                      | Labat test             | -                              | -             | -        | -      | -             | +        |
|   | Lignin test            | -                              | -             | -        | -      | -             | +        |
| Tannins                                     | Gelatin test           | -                              | +             | +        | -      | -             | +        |
|   | FeCl <sub>3</sub> test | -                              | +             | +        | -      | -             | +        |
|   | Lead acetate           | -                              | +             | +        | -      | -             | +        |
| Terpenoids                                  |                        | +                              |               |          | +      |               |          |
| Saponins                                    |                        | -                              | +             | -        | -      | -             | +        |
| Steroids                                    | Slakowski              | -                              | -             | +        | -      | +             | +        |
|   | Liebermann Burchard    | -                              | -             | +        | -      | +             | +        |
| Coumarins                                   |                        | +                              |               |          | +      |               |          |

Note: '+' indicates presence and '-' absence

## RESULTS AND DISCUSSION

The constituents of the plant from the secondary metabolites provide excellent identification of the drugs [10]. Phytochemical analysis of the stem extracts of *Svensonia hyderbadnesis* revealed that the maximum number of the compounds were reported in methanolic extracts when compare with hexane and ethyl acetate (Table 1). Methanolic extracts showed rich in flavonoids, phenols, tannins, terpenoids and steroids. Whereas ethyl acetate extract was rich in phenols, glycosides, tannins and saponins. The same results were observed with *Boswellia ovalifoliolata* [11]. Ethanolic extract of *thymus* leaves consist of more secondary metabolites than the other

solvents [12]. Hexane extract showed positive results only for phenols. However the qualitative analysis of secondary metabolites in the leaf extracts of *Svensonia hyderabadensis* illustrates that the methanolic extract was rich in flavonoids, phenols, lignin, tannins, terpenoids, saponins and steroids. Ethyl acetate extract was rich in flavonoids, glycosides and steroids and hexane extract rich in glycosides. Compare to the methanolic extract of the stem and leaves showed similar phytochemical compounds like flavonoids, phenols, tannins, terpenoids and steroids but methanolic leaf extract shows additional compounds such as lignin and saponins. The reason could be that the methanol is high capacity to dissolve more secondary metabolites than the ethylacetate and hexane solvents. The results of the present study revealed that the methanol is the more efficient solvent to extract the phytochemical compounds from *Svensonia hyderabadensis* when compare to the selected solvents for the study.

### CONCLUSION

*Svensonia hyderabadensis* shows rich in secondary metabolites may have valuable pharmacological properties. Phenolic compounds identified during testing the phytochemicals could indicate that this species has an antioxidant activity at higher level. The saponins and flavonoids with great intensity in this case, could through their fungicidal and antiseptic properties. The plant could thus be considered as part of the antimicrobial, antioxidant and hepatotoxic activity.

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