EXTRACTION AND QUANTIFICATION OF STEROLS FROM TRIBULUS TERRESTRIS L., SIDA ACUTA BURM F. AND TRIDAX PROCUMBENS L.

ALKA JINDAL, PADMA KUMAR

Laboratory of Plant Tissue Culture and Secondary Metabolites, Department of Botany, University of Rajasthan, Jaipur 302044, Rajasthan, India. Email: jindal4@gmail.com

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ABSTRACT

Extraction of secondary metabolites from medicinal plants has become important for the presence of bioactive compounds. The present study was aimed to detect and extract sterols in the different parts of Tribulus terrestris L. (root, stem, leaf and fruits), Sida acuta Burm f. (root, stem, leaf and buds) and Tridax procumbens L. (root, stem, leaf, bud, flower and pedicle). Preliminary detection and extracts of sterols was performed by well established methods. The extracts were separately dried and weighed. Result revealed that all the parts of the selected plants were rich in sterol content. Maximum sterol content recorded were 50.5, 34.7 and 18.6 mg/g dw for T. terrestris, S. acuta and T. procumbens, respectively.

Keywords: Sterols, Tribulus terrestris, Sida acuta, Tridax procumbens

INTRODUCTION

Medicinal plants are essential part of human medicine, since the dawn of civilization and are the backbone of traditional medicine system in India [1]. They represent rich source of antimicrobial agents [2]. Many of the plant materials used in traditional medicine are readily available in rural areas at relatively cheaper rate than the modern medicine [3]. Many plant families represent reservoir of effective chemotherapeutics and can provide valuable sources of natural antimicrobials [4,5].

T. terrestris (Family: Zygophyllaceae) is an annual plant distributed in warm regions of Asia, Africa, Europe, America and Australia [6-8]. It is an important medicinal plant and has been used extensively as tonic, aphrodisiac, analgesic, astringent, stomachic, anti-hypertensive, diuretic, lithotriptic and urinary anti-infective [9]. The main constituents of plant are saponins, diosgenins, alkaldoids and amides [10-12].

S. acuta (Family: Malvaceae) is an erect perennial shrub found throughout the hotter parts of India and Nepal. The plant has been used for eczema, kidney stone, headache, malaria, ulcer, fever, gonorrhea, abortion, breast cancer, poisoning, inflammation, feed for livestock, stops bleeding, treatment of sores, wounds and antipyretic [13-16].

T. procumbens (Family: Asteraceae) is a perennial herb. The plant has been used as feed for livestock and stops bleeding [17,18] and for treatment of diarrhoea, malaria, cough and asthma, boils, epilepsy, liquid purging, wounds, toothache and stomachache and paralysis [19-20].

MATERIAL AND METHODS

Plant Material

Different parts of Tribulus terrestris (root, stem, leaf and fruits), Sida acuta (root, stem, leaf and buds) and Tridax procumbens (root, stem, leaf, bud, flower and pedicle) were collected from different localities of Jaipur, in the month of June, 2008. Selected plants were identified at Herbarium, Department of Botany, University of Rajasthan, Jaipur. Voucher specimens (RUBL-20390, 20428 and 20389, respectively) were also submitted to the Herbarium, UOR. All the parts of selected plants were separately shade dried and were milled to a fine powder using a grinder.

Detection of Sterols

Two standard methods were used to determine the presence of sterols in each sample of selected plants [21,22]. 20 grams of finely powdered samples were Soxlet extracted with hot methanol (200 ml) on a water bath for 24 h and filtered by using Whatman filter paper No. 1. The extracts were concentrated on water bath for removal of excess methanol and were used for the detection of sterols.

Liberman-Burchard’s Test: 5 ml of chloroform and 2 ml of acetic acid were added to a portion of the methanol extract of each part of the selected plants. Appearance of green colour in each extract, indicate the presence of sterols.

Salkowski Test: Few drops of con. H₂SO₄ solution was added to each test extracts. A purple colour ring was observed at the upper surface, showed the presence of sterols.

Extraction and Quantification of Sterols

Different parts of selected plants viz., T. terrestris (root, stem, leaves and fruits), Sida acuta (root, stem, leaves and buds) and T. procumbens (root, stem, leaves, bud, flower and pedicle) were subjected to the sterols extraction [23]. Hundred grams of finely powdered plant part was Soxlet extracted with hot petroleum ether (500 ml) on a water bath for 24 h and filtered. Residual mass is taken and hydrolyzed in 30% HCl for 4 h on water bath. Resulting mixture was washed with distilled water till neutrality and dried in oven at 50°C. The dried material was dissolved in benzene for 1 day and filtered. Filtrates were dried in vacuo, weighed and stored in glass vials at 4°C till used.

Table 1: Detection of sterols of T. terrestris, S. acuta and T. procumbens

<table>
<thead>
<tr>
<th>Plant Part</th>
<th>T. terrestris Burchard’s</th>
<th>Salkowski</th>
<th>Sida acuta Burchard’s</th>
<th>Salkowski</th>
<th>T. procumbens Burchard’s</th>
<th>Salkowski</th>
</tr>
</thead>
<tbody>
<tr>
<td>Root</td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Stem</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Leaf</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>Fruit</td>
<td>-</td>
<td>++</td>
<td>Nd</td>
<td>Nd</td>
<td>Nd</td>
<td>Nd</td>
</tr>
<tr>
<td>Bud</td>
<td>Nd</td>
<td>Nd</td>
<td>+</td>
<td>+</td>
<td>++</td>
<td>-</td>
</tr>
<tr>
<td>Flower</td>
<td>Nd</td>
<td>Nd</td>
<td>dd</td>
<td>Nd</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Pedicle</td>
<td>Nd</td>
<td>Nd</td>
<td>dd</td>
<td>Nd</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

*: absent; +: trace amount; ++: moderate amount; +++: significant amount; Nd: not determined
RESULTS

In the present investigation preliminary detection of sterols has been done in the methanol extract of different parts of T. terrestris, S. acuta and T. procumbens and the results are presented in Table 1. All the methanol extract of the selected plants showed presence of sterols by showing positive response for Burchard’s and/or Salkowski test.

Sterol content estimated in each gram of dried plant material of T. terrestris, S. acuta and T. procumbens was recorded in Table 2. Content of sterols were obtained in leaf of T. terrestris (50.5 mg/g.d.w.), followed by bud of S. acuta (34.7 mg/g.d.w) and flower of T. procumbens (18.6 mg/g.d.w).

DISCUSSION

Sterols are subgroup of the steroids and occur naturally in plants, animals and fungi. They cannot be synthesized by humans and are thus consumed from the diet. They are incorporated in a variety of food products [24] due to their cholesterol-lowering effect, hence providing protection against cardiovascular disease [25]. They have shown inhibition of several cancer cell lines including colon [26], prostate [27] and breast [28]. The role of plant sterols as immune modulators [29] and anti-inflammatory agents [30] has also been described. Hence, it is need to screen medicinal plants for detection and extraction of sterols. Result revealed that the selected three plants (T. terrestris, S. acuta and T. procumbens) can be exploited as an important source of phytosterol for drug formulations.

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REFERENCES