Borneo Journal of Resource Science and Technology (2023), 13(2): 186-202
DOI: https:doi.org/10.33736/bjrst.5646.2023

Characterisation of Chemical Compounds from the Root and Leaf Extract of *Abrus precatorius*

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Received: 18 April 2023 Accepted: 26 September 2023 Published: 31 December 2023

ABSTRACT

A total of five secondary metabolites were isolated and purified from dichloromethane root and leaf extracts of *Abrus precatorius* using different chromatographic methods. The structures were elucidated using different spectral data and were confirmed to be 5,7-Dimethoxyflavanone (**1**), Xanthoxylin (**2**), Hexadecanoic acid (**3**) successfully isolated from the root extract, and Beta-eudesmol (**4**) and Squalene (**5**) successfully isolated from the leaf extract. 5,7-Dimethoxyflavanone **(1)** and xanthoxylin **(2)** were reported for the first time from the root extract and this contributed to the pharmacological importance of *A. precatorius.*

Keywords: 5,7-Dimethoxyflavanone, *Abrus precatorius*, beta-eudesmol, hexadecanoic acid, squalene, xanthoxylin

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INTRODUCTION

Numerous efforts by researchers have been directed towards the provision of empirical proof to back the use of tropical plants in traditional medicinal practice (Wakawa *et al.*, 2021). Focus on medicinal plant research has increased worldwide and evidence abounds in the immense potentials of medicinal plants used in various traditional systems. Various medicinal plants have been studied using different scientific approaches and results from these studies have revealed the potentials of medicinal plants in pharmacology (Wakawa & Hauwa, 2013). These medicinal plants are of great importance to the health of the individuals and communities to larger extend, and nutritional benefits are derived from these plants since they are commonly used as vegetables. The use of these medicinal plants in most communities is commonly referred to as traditional medicine.

The use of traditional medicine at the primary health care level is widespread and plant-based treatments are being recommended for curing/managements of various diseases by traditional medical practitioners all over the world. The phytochemicals present in the fruits, vegetables and medicinal plants are gaining attention day-by-day for their active role in the treatment of various human diseases. *Abrus precatorius* (Linn) (Fabaceae) is a lean woody creeper and it is a widely distributed tropical medicinal plant with several therapeutic properties (Akinloye & Adalumo, 1981; Molgaard *et al.*, 2001). It is widely found in Africa, India and many other parts of the world. In West Tropical Africa, *A. precatorius* leaves have been employed to sweeten food, treat stomach complaints, treat fevers, cough and cold (as decoction). The leaves are casually chewed, and the vine is sometimes sold as a masticatory in Curacao (Morton, 1981). The plant is also traditionally used to treat tetanus, and to prevent rabies (Bhutia & Maiti, 2011). Glycyrrhizin (a natural sweetener), a compound obtain from the leaves and roots extract, is an important phytoconstituent of liquorice which is widely used in the pharmaceutical and food industries (Karwasara *et al.,* 2010), and it is reported to exhibit anti-HIV (Hirabayashi *et al*., 1991), immunomodulatory (Yoshida *et al*., 2006), anti-ulcerative and anti-inflammatory properties. This work is also in line with the view to discover more plants and plant materials with scientific proof that can be used in the treatment of diseases as well as material that can serve as potential sources of new product for food, and to

larger extend increase the value for the medicinal plant.

MATERIALS AND METHODS

Plant Material

Fresh plant materials were collected from uncultivated farm land in Garkida, Gombi Local Government Nigeria. They were authenticated in Modibbo Adama University Yola. The plant materials were given a voucher specimen number as HWP/AP/2014-11/001 (*Abrus precatorius* leaves) and HWP/AP/2014-11/002 (*A. precatorius* root). The fresh leaves were carefully plucked and washed under running tap water. They were then air dried and then spread in the laboratory to dry at room temperature until they were fully dried. The root was also washed under running tap water. It was then cut into smaller pieces, spread in the laboratory and allowed to dry at room temperature until they are fully dried.

Sample Preparation

The dried plant materials (leaves and root) were ground into fine powder form using laboratory mortar and pestle, and electric grinder. The powdered samples (mesh 30) were packed into clean, dry sample containers and were labelled appropriately and kept for further use. Extraction was carried out by the conventional solvent extraction method described by Fasihuddin *et al.* (2010) with slight modification. This was achieved by soaking the ground plant materials in solvents in the order of increasing polarity. A total of two kilograms of the dried and ground powdered samples were separately extracted using cold soaking method with hexane. The samples were soaked in the hexane with the ratio of 1:3 (sample: hexane) in five litres Erlenmeyer flasks at room temperature for five days. The resulting hexane solution was then filtered using Whatman filter paper No. 4 and the residue was then re-extracted with fresh hexane and filtered. Both extracts were combined and evaporated to dryness with a rotary evaporator (Heidolph Laborota 4000 efficient) under reduced pressure below 50 ^{o}C to obtain the hexane crude extract. The residues were re-extracted using similar procedure with dichloromethane, followed by ethyl acetate and methanol to obtain respective crude extracts. The dry weight and percentage yield of each crude extract was determined (simple percentage).

Isolation and Identification of Secondary Metabolites

Combination of the following methods were used for the isolation and identification of secondary metabolites.

Column chromatography

The column was prepared using methods as described by Firdous *et al.* (2013). Sample loading via wet-packing method and column elution with suitable solvent systems with increasing polarity was used as described by Fasihuddin *et al*. (2010) and Patra *et al*. (2012). The procedure was repeated using different solvent systems, based on increasing polarity. Samples from the column fractions were examined using TLC plates in few suitable solvent systems until fraction with single spot that appeared on TLC plate was treated as possible pure secondary metabolite as described by Patra *et al.* (2012).

Chromatotron

Chromatotron was also used for the separation of the substances and was observed by illuminating the disk with short-wave UV light, so that the desired fractions can be collected. The layer was covered with a fused silica disk, as normal glass is not transparent to short-wave UV light. The sample was then collected at time interval into vails and TLC analysis of the different fractions collected was performed (Varsha & Sonal, 2015)

Vacuum-liquid chromatography

Vacuum liquid chromatography was used for separation in step gradient elution of compounds in which the flow of compounds was activated by vacuum to enhance separation. Successive addition of eluting solvent in different ration combination was done, and band of fractions were collected into several conical flasks (John & Bruce, 1986)

At the end of the combined isolation methods used, 11.6 mg of compound **1** (5,7-dimethoxyflavanone) was obtained in combined solvent system Hexane: $CHCl_3$ (7:3) and Hexane: EA (4:1), 9.8 mg of compound **2**