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Some phytochemical analyses of different extracts of the cogon grass *Imperata cylindrica* from Mizoram, India

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Cogon grass, *Imperata cylindrica* (L.) Räuschel, was studied for its flavonoid and phenol contents. A series of extracts of the underground (rhizome-root) parts was prepared by hot extraction using solvents of different polarities such as chloroform, methanol, and petroleum ether. The total flavonoid content of the plant extracts was determined based on the reaction with aluminum chloride, sodium nitrite, and sodium hydroxide. Quercetin was used as a standard reference. Chloroform extract showed highest content of flavonoids with 30.88 mg QE/g dry wt., followed by petroleum ether extract which was 22.05 mg mg QE/g dry wt., and methanol extract has the least value with 7.35 mg QE/g dry wt. The total phenolic content of the plant was estimated based on the reaction of Folin-Ciocalteu reagent using gallic acid as the standard reference. Again, the chloroform extract had the highest content with 7.54 mg GAE/g dry wt., while methanol extract contained 5.03 mg GAE/g dry wt., and petroleum ether extract contained 3.63 mg mg GAE/g dry wt. Our study shows that *I. cylindrica* is a good source of antioxidants.

Keywords: *Imperata cylindrica*, medicinal plant, total phenol, total flavonoid.

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INTRODUCTION

Cogon grass, *Imperata cylindrica* (L.) Räuschel, is a rhizomatous grass native to the Indomalayan and Australasian realms. The rhizome-root part serves as traditional medicine in different cultures of Southeast Asia for the treatment of a wide range of infections. The aerial part is a resilient fibre for which it serves as the most useful construction material for thatched roofs in traditional houses. The flowers and roots are used for important medicinal properties such as antibacterial, diuretic, skin softening (emollient), antipyretic (febrifuge), salivating (sialagogue), anticoagulant (styptic) and soothing (tonic) activities.[1] The roots are especially effective in the treatment of blood urine (haematuria), vomiting of blood (haematemesis), nosebleed (epistaxis), dropsy (oedema), and jaundice.[2] The leaves have been experimentally shown to have neuro-protective,[3] and vasodilative effects.[4]

The plant is abundant throughout the year in Mizoram and its propagation increases during summer after burning for slash-and-burn (*jhum*) cultivation. To the Mizo people, the plant is commonly used as antimicrobial in dermal wound, ringworm, cholera, dysentery and diarrhoea. It is traditionally unique to the Mizo people as the roots are known to be effective anthelmintic agents that can

eliminate both tapeworm and roundworms from the intestine. The roots maybe crushed and juiced, or directly consumed for intestinal helminthiasis.[5] The pharmacognosical and pharmacological properties remain unknown. It is therefore imperative to investigate its basic chemical and biological properties.

MATERIALS AND METHODS

Plant material

Mature *I. cylindra* were collected from Ngopa village, Champhai district, Mizoram, India, in January 2018. Ngopa is located between 23.8861° latitude north and 93.2119° longitude east. A voucher specimen was identified at the Botanical Survey of India (BSI), Shillong, Meghalaya, and is maintained at the herbarium section of University College, Aizawl, Mizoram (accession number PUC-I-2018-01). The roots were washed with distilled water and dried in shade at room temperature at 21–27°C.

Chemicals and reagents

All chemicals and reagents used were standard analytical grades procured either from Merck India, Mumbai, or HiMedia Laboratories Pvt. Ltd., Mumbai, India.

Extraction

The dried roots were crushed to fine powder in an electric blender. Continuous extraction was performed in a 5 L-capacity Soxhlet apparatus using three solvents of different polarities, *viz*. chloroform, methanol, and petroleum ether. The extracts were purified and concentrated in a vacuum rotary evaporator (Buchi Rotavapor® R-215). The crude extracts were obtained as semi-solid mass precipitate, which were stored at 4°C until further use.

Total flavonoid content

The total flavonoid content of the plant was determined based on the reaction with aluminum chloride following the method of Park *et al.*[6] Briefly, 1 ml of the plant extract (50 µg/ml) was mixed with 2 ml of distilled water in a test tube. After 5 minutes, 3 ml of 5% sodium nitrite ($NaNO_2$) and 0.3 ml of 10% aluminum chloride ($AlCl_3$) were added. 2 ml of sodium hydroxide (NaOH, 1 M) was added after 6 minutes and the total volume was made up to 10 ml with distilled water. After 1 hour, absorbance was measured at 510 nm in an ultraviolet-visible spectrophotometer (Labtronics LT-2700). A standard calibration curve was prepared using quercetin at different concentrations, *viz*. 10, 20, 40, 60, 80, and 100 µg/ml. The total flavonoid content of the plant extract was extrapolated from the standard curve. The final value was expressed as milligrams of quercetin equivalent per gram (QE/g) of the dried extract.

Total phenolic content

The total phenolic content of the plant was estimated based on the reaction of Folin-Ciocalteu reagent following the method of Kim *et al.*[7] with slight modifications. In brief, 1 ml of methanol extract of the plant (1 mg/ml of distilled water) was mixed with 5 ml of Folin-Ciocalteu reagent (diluted tenfold). After 3 minutes, 4 ml of sodium carbonate (Na_2CO_3) solution (0.7 M) was added and mixed completely. The mixture was left still for 1 hour at room temperature. Then, the absorbance was measured at 765 nm using UV-Vis spectrophotometer. Standard calibration curve was prepared using gallic acid at different concentrations, *viz*. 10, 20, 40, 60, 80, and 100 µg/ml. From the calibration curve, the amount of phenolic content of the plant extract was extrapolated. The total phenolic content was expressed as milligrams of gallic acid equivalent per gram (GAE/g) of the dry sample.

RESULT

Total flavonoid content

The total flavonoid content of *I. cylindrica* root

extract was estimated by reacting with sodium nitrite ($NaNO_2$), aluminum chloride ($AlCl_3$), and sodium hydroxide (NaOH). Quercetin was used as standard antioxidant with concentration dependent activity as shown in Figure 1. The total flavonoid concentration was expressed as weight in mg of quercetin equivalent (QE) per gram of dry weight. The different extracts showed different amounts of phenolic content. The chloroform extract of the plant had the highest value of total flavonoids, while methanol extract had the least. From the standard curve, it was found that the total phenolic content of the chloroform extract was 30.88 mg QE/g dry wt., that of petroleum ether extract was 22.05 mg mg QE/g dry wt., and that of methanol extract was 7.35 mg QE/g dry wt.

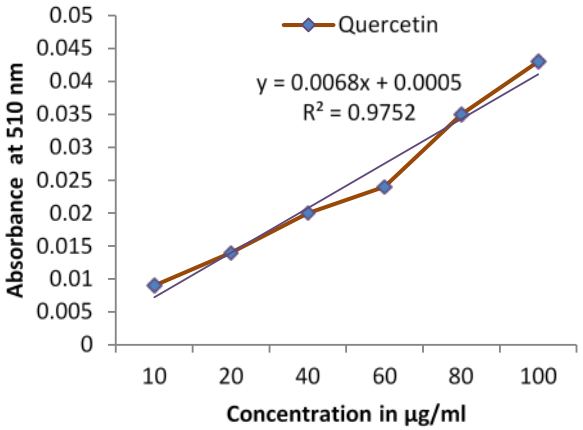

Figure 1 | Standard curve for quercetin.

Total phenolic content

The total phenolic content was determined from the reaction with Folin-Ciocalteu reagent. The standard antioxidant used was gallic which indicated concentration-dependent activity as shown in Figure 2. The total phenolic content was expressed as weight in mg of gallic acid equivalent (GAE) per gram of dry weight. The chloroform extract of the plant had the highest value of total flavonoids, while petroleum ether extract had the least. From the standard curve, it was found that the total phenolic content of the chloroform extract was 7.54 mg GAE/g dry wt., that of methanol extract was 5.03 mg GAE/g dry wt., and that of petroleum ether extract was 3.63 mg GAE/g dry wt.

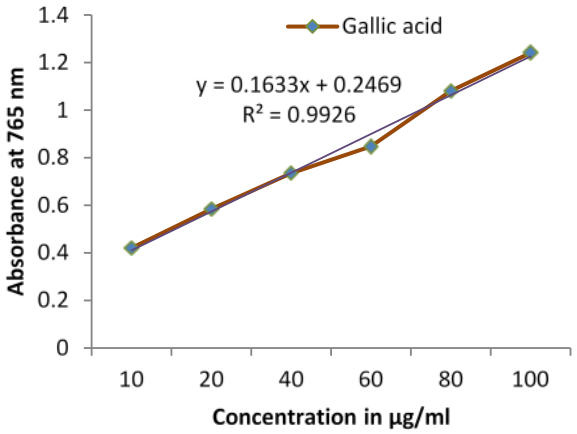

Figure 1 | Standard curve for gallic acid.

DISCUSSION

Free radicals inside the cells are harmful to the cellular and molecular activities. Reactive oxygen and nitrogen species are produced during normal metabolic reactions in the cells and they cause inherent molecular damage to DNA, lipids, proteins, and other vital biomolecules.[8] The cumulative effect called oxidative stress is therefore the major factor for the development of several genetic and immune problems including those of as cardiovascular, neurodegenerative, cancer and even aging.[9] Our body is endowed with special antioxidant defenses such as superoxide dismutases, hydrogen peroxide-removing enzymes, metal binding proteins, but they are inadequate mechanism to subdue all the harmful effects of such endogenous oxidants.

Thus, antioxidants are essential for maintaining

the oxidation equilibrium by effectively promoting the elimination of free radicals which are constantly produced as a result of cellular oxidative stress. They make the harmful free radicals to harmless molecules.[10] The protective function of antioxidants is not limited to scavenging the free radicals alone but extends to upregulation of enzymes involved in antioxidation and detoxification, modulation of redox cell signaling and gene expression. In this way the body balance of oxidation and free radical removal is constantly maintained in a normal physiological condition.[11]

Plants are a huge source of antioxidants. Medicinal plants are therefore among the best sources of these antioxidant compounds for their availability and ease of access.[12] Therefore, it is crucial for investigations to analyse acclaimed medicinal plants of their antioxidant potentials, in addition to their usual pharmacological activities. Thus, they can be useful in the treatment and management of complex diseases like autoimmune disease and cancer.[13,14] The present data also show that *I. cylindrica* is a good source of antioxidants and may serve as potential lead to pharmaceutical and nutritional supplementary development.

Phenolic derivatives and flavonoids natural compounds in plants and paly several roles in the plant's life such as general growth, reproduction, and defence against parasites and pests. Flavonoids themselves are a group of hydroxylated phenolic compounds having a benzo-γ-pyrone structure and are ubiquitously occurring in plants.[15] In fact, these phytocompounds are best known for their biological properties for their high antioxidant activities. They also play pivotal role in the modulation of the immune system at various molecular levels.[16] Hence, it is interesting to study the pharmacological properties of *I. cylindrica*.

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