Short communication **Bimonthly published scientific journal**

INTERNATIONAL JOURNAL of Indigenous Herbs and Drugs

ISSN-2456-7345

Analgesic activity of the aqueous extract of *Dioscorea hispida* Dennst (Kurot) rhizome

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Abstract

Objective: *Dioscorea hispida* Dennst is a wild yam that is locally known as 'Kurot' in Northern Samar, Philippines. The main objective of the study was to determine the analgesic activity of the kurot rhizome extract.

Methods: In this study, the rhizome of the plant extract was collected from Lavezares Northern Samar. An aqueous rhizome extract of the plant was prepared for analysis. The phytochemical characteristics like alkaloids, steroids and anthraquinones and the analgesic activity using acetic acid-induced writhing of the rhizome extract of kurot was determined.

Results: Results of the phytochemical screening and the analgesic activity showed that the rhizome extract of kurot may exhibit potential application in variety of analgesic products as shown in the decrease of the mean squirm of the experimental animals which is comparable to the commercially available drug.

Conclusion: In this work, it was concluded that the rhizome extract of kurot possesses analgesic activity.

Keywords: Analgesic activity, Kurot, Phytochemical screening, Acid-induced writhing

Plants, besides providing nutrition, have always formed an important source of chemical compounds, which can be used for medicinal purposes. Even in recent times, plants have been an important source of modern drugs. In recent years traditional medicine has become of interest to both scientists and the general population for a number of reasons, which include high price of allopathic drugs beyond the reach of the poorer segments of society in almost every country, lack of access to medical clinics and hospitals by the rural population of developing countries, the side-effects and toxicities of modern synthetic drugs [1].

Northern Samar has a rich assortment of plants distributed in different geographical and ecological conditions widespread in the province. Plants have been used since prehistoric times for treatment of various ailments [2]. The traditional systems of medicine together with folklore systems continue to serve a large portion of inhabitants, particularly in rural and tribal area regardless of the dawn of modern medicine [3]. Nowadays there are many drugs available as anti-inflammatory, analgesic, antioxidant and antimicrobial agents; however they have undesirable effects and adverse reactions [4]. Since, the modern medicines have some limited use the traditional plants are now becoming the promising approach for the treatment of several diseases [5]. Although, there were studies to use alternative sources of medicines but most of their findings were focused on plants. Only few are on wild yam. *Dioscorea hispida* Dennst (Kurot) belongs to the family Dioscoreaceae assigned to the order Dioscorales. The bioactive components of kurot extract have been paid more attention by researchers however there have been few investigations on the secondary metabolites and analgesic activity of kurot rhizome extract. Studies on the properties of *Dioscorea hispida* are very important due to their ready availability and their utilization in food and non-food applications. In this work, to make full use of the resources and to widen its application, the secondary metabolites and analgesic properties of kurot rhizome extract were determined.

In this work, fresh rhizome of *Dioscorea Hispida* Dennst (kurot) plant was collected from Lavezares, Northern Samar. The sample was brought to the College of Science Laboratory for extraction and taxonomic identification. The identity of the plant was confirmed by an expert from the College of Science and a voucher specimen (UEPCS 101) was submitted for the future reference. An aqueous rhizome extract of the plant was prepared for analysis. The extract was placed in airtight bottle and stored in refrigerator awaiting subsequent bioassays. 20 mL of rhizome extract of 100% concentration was prepared. A commercially available drug (mefenamic acid) serves as a positive control and distilled water as the negative control. The kurot rhizome extract was subjected for chemical analysis to identify and characterize some of the secondary metabolites. The standard procedures for phytochemical screening were adopted to screen the rhizome extracts of

Lee RG, Cui-Lim KMR. Analgesic activity of the aqueous extract of *Dioscorea hispida* Dennst (Kurot) rhizome. Int J Ind Herbs Drugs 2018; 3(1): 8-10.

Kurot for secondary metabolites such as alkaloids, steroids and anthraquinones [6]. Forty five albino mice, weighing about 20-35 g were used for acetic acid writhing test. The albino mice were acclimatized in the laboratory room in a few days and were given enough food and water under normal environmental condition before these were subjected to experimentation. Kurot rhizome extract, mefenamic acid solution and distilled water were administered orally to albino mice at a dosage 0.2 ml per 20 grams body weight (gbw). After 30 min, 0.2 ml of 0.7% acetic acid per 20 gbw was injected intraperitioneally to all sets of treatment. The pain experiment was done in conformity to the guidelines of the animal pain experimentation. Five min after acetic acid injection the number of squirms was counted for each mouse for 15 min using the equation below:

% reduction in squirms= (Number of squirms for treatment 1-No.of squirms for treatment 2) / (Number of squirms for treatment 1) ×100%

Where:

Treatment 1= No. of squirms for (acetic acid + solvent control)

Treatment 2= No. of squirms for (acetic acid + kurot extract)

The data were statistically treated to compare the difference between the analgesic potential of fresh rhizome extract of *Dioscorea Hispida* Dennst (kurot) and the analgesic potential of a commercial drug (mefenamic acid) using T-test and one way ANOVA.

21% of acetic acid was used to induce pain in the experimental animals. The pain experiments were performed in conformity to the guidelines of the ethics board. A commercially available drug was used as positive control and the distilled water as negative control. The determination of analgesic activity of *Dioscorea hispida* Dennst in this study was based on the number of squirms of the mouse. The squirm is the sign that the animal is in pain and the reduction in the number of squirms represent reduction in pain. The measurement of the analgesic activity was interpreted by the reduction of squirms of the experimental mice.

The secondary metabolites that were screened in the rhizome of *Dioscorea hispida* Dennst (Kurot) extract are alkaloid, steroids and anthraquinone. Screening test for alkaloids were performed using Dragendorff's reagent and Mayer's reagent. A positive result was indicated by the formation of orange precipitate. There was an orange precipitate that was formed when it was treated with Dragendorff's reagent. There was white precipitate formed when it was treated with Mayer's reagent. This clearly indicates that the alkaloid was present in the rhizome of Kurot extract. The Libermann-Burchard test was used to determine the presence of steroids specially the unsaturated ones. The positive results for this secondary metabolite gives color range of blue to green, red, pink or violet, because of their skeletons. The modified Borntrager's test was used to detect the presence of anthraquinone, a pink color indicated a positive result of anthraquinone. Table 1 shows the summary of the different secondary metabolites determined in the rhizome extract of kurot. Table 2 shows the mean number of squirms of the experimental mice for the 3 treatments. The dosage given corresponded to the body weight of each mouse and their squirms exhibited.

Table 1: The summary of the different secondary metabolites determined in the rhizome of kurot extract

Test for secondary metabolites	Result
Alkaloids	Positive
Steroids	Negative
Anthraquinone	Negative

Table 2: The average mean squirms of the experimental animals

Treatment (T)	Average gbw of mouse	Average dosage (0.2/20 gbw)	Mean (number of squirms) and standard error of mean
Acetic acid + distilled water (T1)	29.20	0.29	30.93+ 0.0034
Dioscorea hispida extract (T2)	29.13	0.29	13.93+ 0.0125
Commercially available drug (T3)	29.23	0.29	7.530+ 0.0012

gbw=Gram body weight; n=15; Mean±S.E.M

It was observed that after administration of acetic acid, the pulse rate of the experimental mice increased and became rapid and irregular. This implied that the animal was in pain. When tested, the number of squirms ranged from 24 to 38. After establishing that the animals were in pain, the test substances were administered to determine its analgesic activity. Results on the number of squirms experienced by the experimental animals are presented in Tables 2. The study shows the mean number of squirms of each of the experimental mice. After administration of the test substance, it was observed that the pulse rate of the animals returned to normal. This implied at the time of the experiment that the kurot extract administered to the experimental animals relieved the pain, which is shown in the reduced number of squirms in all of the mice in T2. The reduction in squirms manifested the analgesic activity of 100% of the rhizome of

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kurot extract. After the treatment of the test substance, the pulse rate returned to normal, the same reaction in T2. However, the number of squirms was lower than those in T2. This means that the commercially available drug is more effective as an analgesic compared to the 100% kurot extract. This study focused on the extraction of the kurot rhizome extract and the determination of the different secondary metabolites like alkaloids, steroids and anthraquinones. The effects of these different secondary metabolites on the analgesic activity were also tested in this work. The kurot rhizome extract was subjected to analgesic activity. Kurot rhizome extract exhibited analgesic activity. Statistical evaluation of the 3 treatments using t-test and one-way analysis of variance shoes that the kurot rhizome extract is comparable to the commercially available drug in terms of its analgesic activity. The results presented in this work bring new dimensions that could contribute to a better characterization and utilization of kurot rhizome extract.

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Acknowledgement

The authors are grateful to the University Research Office, University of Eastern Philippines for the support. We also thank BS Major in Chemistry of the College of Science, UEP for the help in the analysis and the College of Science for the laboratory facilities.

Conflict of Interest: None declared

Received: 24 January 2018, Revised: 12 February 2018, Accepted: 22 February 2018

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