# SHORT REPORTS

# PISICIDAL TRITERPENOID SAPONINS OF THE PERICARPS OF SAPINDUS EMARGINATUS

WILAWAN MAHABUSARAKAM, G. H. N. TOWERS, PITTAYA TUNTIWACHWUTTIKUL, AND PICHAET WIRIYACHITRA

<sup>a</sup>Department of Chemistry, Faculty of Science, Prince of Songkla University, Hat Yai, Thailand.

<sup>c</sup>Department of Chemistry, Faculty of Science, Silpakorn University, Nakorn Pathom 73000, Thailand.

<sup>d</sup>Research Center for Natural Products, Faculty of Pharmacy, Chiang Mai University, Chiang Mai 50002, Thailand.

(Received 3 July 1990)

#### ABSTRACT

Two pisicidal triterpenoid saponins, 3-0-[ $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 3)- $\alpha$ -L-rhamnopyranosyl (1 $\rightarrow$ 2)- $\alpha$ -L-arabinopyranosyl] hederagenin and 3-0-[ $\alpha$ -L-arabinopyranosyl(1 $\rightarrow$ 2)- $\alpha$ -L-arabinopyranosyl] hederagenin, were isolated from the pericarps of Sapindus emarginatus (Sapindaceae).

Our previous studies indicated that the aqueous extract of the pericarps of Sapindus emarginatus (Sapindaceae) had good piscicidal activity. We now report the isolation and identification of the two piscicidal triterpenoid saponins,  $3-0-[\alpha-D-xylopyranosyl-(1\rightarrow 3)-\alpha-L-rhamnopyranosyl(1\rightarrow 2)-\alpha-L-arabinopyranosyl] hederagenin and <math>3-0-[\alpha-L-arabinopyranosyl(1\rightarrow 3)-\alpha-L-rhamnopyranosyl(1\rightarrow 2)-\alpha-L-arabinopyranosyl] heragenin.$ 

# General Experimental Procedures

Analyses were carried out by Scientific and Technological Research Equipment Center, Chulalongkorn University, Bangkok, Thailand. Melting points were measured with a micromelting point apparatus. Infrared spectra were obtained with a Jasco A-302 spectrophotometer. <sup>1</sup>H-NMR spectra of CDCl<sub>3</sub> solution were recorded with a Bruker WM-400 MHz spectrometer. <sup>13</sup> C-NMR spectra were obtained at 100.62 MHz. Adsorbents for tlc, prep.tlc and cc were from E. Merck.

<sup>&</sup>lt;sup>b</sup>Botany Department, University of British Columbia, Canada.

#### Plant Material

The whole fruits of *S. emarginatus* were purchased at the local market in Hat Yai, Thailand.

# Piscicidal Activity Test

Each saponin was dissolved in ethanol as the stock solution. Six concentrations of the test solution, viz. 2.0, 1.0, 0.5, 0.20, 0.10 and 0.05 ppm, 200 ml each, were prepared from the stock solution and put into a 250 ml beaker. There were six treatments of these five concentrations plus a control which had water and the same amount of ethanol as in the test solution. Ten fries of Tilapia (*Oreochronis niloticus* Linn.) about 1 cm in length were used in each test solution and control. Fish mortalities were observed after 24 hours.

#### Extraction and Isolation

Dry milled pericarps of *S. emarginatus* (600 g) were exhausively extracted with MeOH (3×2.5 l). The MeOH extract was concentrated in *vacuo* and the residue was partitioned between *n*-BuOH and water. The *n*-BuOH layer was evaporated to give a brown semisolid (237 g). Repeated cc of the *n*-BuOH fraction (155 g) on Si gel with CH<sub>2</sub>Cl<sub>2</sub>/MeOH/H<sub>2</sub>O gave a mixture of two saponins, 3-0-[ $\beta$ -D-xylopyranosyl-(1 $\rightarrow$ 3)- $\alpha$ -L-rhamnopyranosyl(1 $\rightarrow$ 2)- $\alpha$ -L-arabinopyranosyl] hederagenin and 3-0-[ $\alpha$ -L-arabinopyranosyl(1 $\rightarrow$ 3)- $\alpha$ -L-rhamnopyranosyl(1 $\rightarrow$ 2)- $\alpha$ -L-arabinopyranosyl] hederagenin (16.6 g.)

Separation of the mixture of the saponin acetates by prep.tlc, followed with the alkali hydrolysis of each of the saponin acetate gave 3-0-[ $\beta$ -D-xylopyranosyl( $1\rightarrow 3$ )- $\alpha$ -L-rhamnopyranosyl( $1\rightarrow 2$ )- $\alpha$ -L-arabinopyranosyl] hederagenin (1) and 3-0-[ $\alpha$ -L-arabinopyranosyl( $1\rightarrow 3$ )- $\alpha$ -L-rhamnopyranosyl( $1\rightarrow 2$ )- $\alpha$ -L-arabinopyranosyl]hederagenin (2).

The two saponins isolated were identified by spectral data and chemical studies. <sup>2-4</sup> <sup>13</sup>C-NMR data of the two saponins (1,2) are in agreement with the data previously reported.<sup>5</sup>

Lethal concentration for 50% death of Tilapia (*Oreochromis niloticus* Linn.) for both saponins (1 and 2) was between 0.10 to 0.20 ppm. Piscicidal effectiveness (LC<sub>50</sub>) of the saponins, 3-0-[ $\alpha$ -L-arabinopyranosyl] hederagenin and 3-0-[ $\alpha$ -L-arabinopyranosyl] hederagenin obtained from the partial hydrolyses of (1) and (2) were also found to be between 0.10 to 0.20 ppm.

Full details of the isolation and identification of the compounds are available on request to the authors.

### **ACKNOWLEDGMENTS**

The work was carried out with the aid of a grant from the International Development Research Centre, Ottawa, Canada. We are also grateful for assistance from the Chemistry of Biologically Important Natural Products, an activity of the International Development Program of Australian Universities and Colleges supported by the Australian Development Assistance Bureau.

## REFERENCES

- 1. Chiayvareesajja, S., Mahabusarakam, W., Maxwell, J.F., Wiriyachitra P., and Towers G.H.N. (1987). *J. Sci. Soc. Thailand* 13, 29.
- 2. Higuchi, R. and Kawasaki, T. (1976). Chem. Pharm. Bull. 24, 1021.
- 3. Encarnacion, R., Kenne, L., Samuelsson, G. and Sandberg, F. (1981). Phytochem. 20, 1939.
- 4. Shimizu, M., Arisawa, M., Morita, N., Kizu, H. and Tomimori, T. (1978). Chem. Pharm. Bull. 26, 655.
- 5. Kimata, H., Nakashima, T., Kokubun, S., Nakayama, K., Miyota, Y., Kitahara, T., Yata, N. and Tanaka, O. (1983). Chem. Pharm. Bull., 31, 1998.

## บทคัดย่อ

จากการแยกสารที่มีฤทธิ์ฆ่าปลาจากส่วนสกัดของ Sapindus emarginatus พบว่าเป็นสาร triterpenoid saponins 2 ตัวคือ 3-0-[ $\beta$ -D-xylopyranosyl(1  $\rightarrow$  3)- $\alpha$ -L-rhamnopyranosyl(1  $\rightarrow$  2)- $\alpha$ -L-arabinopyranosyl] hederagenin และ 3-0-[ $\alpha$ -L-arabinopyranosyl(1  $\rightarrow$  3)- $\alpha$ -L-rhamnopyranosyl(1  $\rightarrow$  2)- $\alpha$ -L-arabinopyranosyl] hederagenin