Isolation and Identification of Secondary Metabolite Compound Extract Etil Acetate from the Leaves of Durian *Durio zibethinus* L.

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Abstract: The aim of this study was to isolate and identify secondary metabolite in ethyl acetate extract of durian leaves (*Durio zibethinus* L.). Isolation and purification is carried out by maceration, partitioning (ethyl acetate and water), fractionation (column chromatography) and preparative Thin Latyer Chromatography (TLC). Interpretation of infrared (IR) spectra and phytochemical screening showed that the secondary metabolite is a steroid compound.

1 INTRODUCTION

Indonesia is known as a tropical area and even the largest for the world scale with a variety of plants that are of high value to the community, namely plants that have medicinal properties. One of them is the durian plant. This plant is very familiar, especially the fruit. Durian Leaves are traditionally stew used as an antipyretic for children. Phytochemicals of durian leaves (*Durio zibethinus* L.) are flavonoids, steroids/terpenoids, and glycosides in ethyl acetate extract (Aruan, 2019). Steroids are a group of secondary metabolites. This is very important in the medical field. Isolation in the process of separating chemical contained in a material includes four important stages of maceration, partitioning, purification, and identification. This research is based on the results of Insanu 2011 study on flavonoids in ethanol extracts from durian leaves. This study aim to examine the chemical content of durian leaves, to be developed further as a natural medicine.

2 MANUSCRIPT PREPARATION

2.1 Preparation Sample

Samples were collected from Tornaginjang Sibolga Village, Medan, North Sumatra, Indonesia.

Fresh durian leaves are cleaned of dirt, weighed, then dried in a drying cabinet that is not exposed to direct sunlight, then the dried durian leaves are crushed using a blender. Blending durian leaves were tested for steroid / terpenoid identification. 1 gram of dried durian leaf is soaked with ethyl acetate, then filtered using filter paper, the filtrate is collected in a vaporizer cup and then evaporated on a water bath and the Liebermann-Burchard reagent is added through the wall of the evaporating cup. If the color purple or red turns blue or blue or green blue indicates the presence of steroids / terpenoids (Harborne, 1987).

2.2 Pemurnian Sampel

Dried durian leaves, crushed using a blender, macerated with ethanol, then partitioned using ethyl acetate. Ethyl acetate extract was fractionated with

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n-hexane: ethyl acetate (5: 1) eluent. The spot is monitored under UV light at 265 and 365 nm and then sprayed with 1% cerium sulfate in order to see patches showing some amount of compound in the ethyl acetate fraction. The results of the first column chromatography obtained 8 fractions followed by antidiabetic testing. Obtained F3 fraction which has antidiabetic activity with 82.85% inhibition. F3 fraction was carried out by the second column chromatography with eluent n-hexane: ethyl acetate 5: 1 and obtained fraction 6, 7, 8 with the same Rf distance called F3-1 fraction. The F3-1 fraction was then carried out preparative chromatography with eluent n-hexane: ethyl acetate 5: 1 and obtained F3-1-1 fraction with Rf 0.7 with a weight of 81 mg with white powder. The purity fraction F3-1-1 was determined by TLC and eluent n-hexane: ethyl acetate 5: 1, n¬-hexane: acetone 5: 1, n-hexane-chloroform 5: 1; 2-D TLC n-hexane: ethyl acetate 5: 1. Pure isolate F3-1-1 fraction was characterized and identified using infrared spectrophotometry (IR).

3 RESULT AND DISCUSSION

The results of screening of phytochemical extract of durian leaf ethyl acetate with Liebermann Burchad reagent showed that the green solution of the compound was a steroid group (Harborne, 1987).
The results of the identification of the F3-1-1 fraction using infrared spectrophotometry (IR) can be seen in Figure 1 and the interpretation of the spectrum in Table 1.

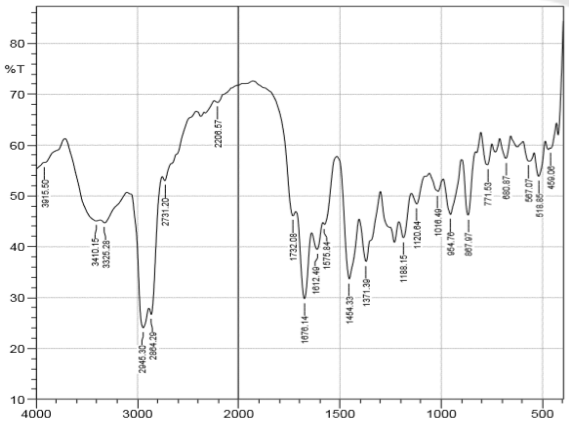

Figure 1: IR Spectrum F3-1-1.

In Table 1 shows the peak of the absorption spectrum at 3410.15 cm-1 in the presence of OH groups; at a wavelength of 2945.30 cm-1 there is a stretching group C-H; at a wavelength of 1676.14 cm-1 group C = C alkene absorption peak; at the other absorption peak 1454.33 cm-1 in the presence of CH_2 groups; peak absorption spectrum of 1371.39 cm-1 indicates the presence of CH_3 groups; and peak absorption spectrum at 1120.64 cm-1 indicates the presence of C-OH groups (Cole, 1963). IR spectrum information reinforces data that there are no aromatic functional groups but only alkene groups in isolation.

Table 1: Spektrum Interpretation of IR dari F3-1-1.

Wavenumber cm^{-1}	Functional Groups
3410.15	-OH
2945.30	C-H
1676.14	C=C
1454.33	CH_2
1371.39	CH_3
1120.64	C-O

4 CONCLUSIONS

The best effluent for purification of ethyl acetate extract from durian leaves (Durio zibethinus L.) by using thin layer chromatography (TLC) is n-hexane: ethyl acetate (5: 1) eluent. The type of secondary metabolites obtained from the separation of ethyl acetate extract from durian leaves by screening, TLC, and IR information are steroid groups.

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