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CHARACTERIZATION OF FLAVONOID COMPOUNDS

CIPLUKAN STEM (Physalis angulata L.)

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ABSTRAK---*The use of plants in Indonesia is widely used as herbal medicines in an effort to maintain public health. One type of plants used as herbal medicines is the Ciplukan plant, Physalis angulata L. The activity of the P.angulata L. plant is as antihyperglycemic, antibacterial, antiviral, immunostimulant and immunosuppressant (immunomodulatory), anti-inflammatory, antioxidant, and cytotoxic properties. One of the secondary metabolite compounds which has this activity is the Flavonoid group. Therefore, the purpose of this study was to identify the flavonoid compounds contained in the Ciplukan plant, P.angulata L. The methods used were Phytochomia Test, Thin Layer Chromatography, Column Chromatography, and UV-Vis Spectrophotometry. Phytochemical test results show that the Ciplukan stem contains quinone, flavonoid, saponin, and monosesquiterpeoid compounds, and polyphenols. TLC test with n-hexane-ethyl acetate (7: 3) eluent showed a blue chromatogram with Rf 0.8 cm. Several fractions of the results of the chromatography column later in UV-Vis Spectrophotometry showed the compound was a flavonoid type of flavanone.*

Keywords---*Flavonoids, P.angulata L. Phytochemistry*

I. Introduction

Ciplukan, Physalis *angulata* L. is a Native American plant that is now widely distributed in tropical regions of the world. Ciplukan contains active compounds which include saponins (in buds), flavonoids (leaves and buds), polyphenols, and fisalin (fruits), Withangulatin A (fruits), palmitic acid and stearic (seeds), alkaloids (roots), Chlorogenic acid (stems and leaves), tannin (fruit), cryptoxanthine (fruit), vitamin C and sugar (fruit) (Ramesh & Mahalakshmi, 2014). From the research, one of the plants studied as an anticancer is *Ciplukan* (P.*angulata L*).

Based on the phytochemical results of some references that the chemical content of the leaves of the Ciplukan plant contains Phisalin and Withanolid compounds which were allegedly from various reports containing anticancer activity (Widodo et al., 2010). In addition, from various reports of this plant has many other properties because it contains chemical compounds whose function is to treat a disease, one of the chemical compounds contained in this Ciplukan plant is a flavonoid compound. Flavonoids are one of the secondary metabolites found in plants. This compound can be used as an anti-microbial, infection drug in wounds, anti-fungal, anti-virus, anti-cancer, and anti-tumor. Besides flavonoids can also

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be used as an anti-bacterial, anti-allergic, cytotoxic, and anti-hypertension (Sriningsih, 2008). However, it has not been reported the types of flavonoid compounds found on the stem of the Ciplukan plant. Therefore, it is necessary to know the types of flavonoid compounds found on the stem of the Ciplukan plant.

II. Materials and methods

Equipment used to identify and characterize secondary metabolite compounds is a set of Maserator, rotary evaporator (Heidolph Laborota 4000), oven, beaker glass, test tube, stirring rod, spatula, brown bottle, chromatographic column, analytical balance, chamber, capillary tube, TLC plates, and UV lamps (λ 254 and 366 nm) as stain removers. For the characterization process with the ultraviolet spectrum ultraviolet visible spectrophotometer (Shimadzu PharmaSpec UV-1700) was used.

The materials used for the isolation of secondary metabolite compounds are ciplukan plant stems obtained from the East Karawang area. The chemicals used are 96% ethanol as a solvent when maceration, ethyl acetate, and n-hexane as eluents in thin layer chromatography and as a solvent for column chromatography. The absorbent used in column chromatography is silica gel 60 F254 (0.063-0.200 mm), for thin layer chromatography (TLC) analysis, the finished plate is DC-Alufolien Kieselgel 60 F254 Merck (20x20 cm), filter paper and aluminum foil. Materials used for phytochemical tests are Mayer and Dragendorf reagents used for identification of alkaloids, Lieberman Burchad reagents (acetic anhydride and concentrated sulfuric acid) for identification of terpenoids and steroids, (concentrated hydrochloric acid and Magnesium powder) for identification of flavonoids and iron (III) fluoride (FeCl3) for phenolic identification.

III. Results and discussion

The extraction results obtained a thick extract of 13.06 grams which is not typical and blackish green. The percentage of extract yield obtained was 6.5%, this is not in accordance with the literature which suggests that the yield is not less than 9.65% (FHI, 2010). The factor that influences the amount of yield produced is the extraction method used, which is maceration, because maceration does not use heat assistance (lack of assistance from other forces), this maceration method only uses immersion so that the osmosis pressure of the solvent into the solid takes place static even though it has been remastered with solvents (Nurasiah, 2009).

As for the test results the water content is 4.8%, the results are in accordance with the literature that the water content in the extract must not exceed 10% (MOH RI, 2008), and may not exceed 11.7% (FHI, 2010). It aims to avoid the rapid growth of fungi in extracts (Soetarno and Soediro, 1997).

Phytochemical Screening

Phytochemical screening test results using simplicia and ethanol extract of Ciplukan stem as listed in the table above shows that the Ciplukan stem contains Kuinon, Saponin, Flavonoid, Monosesquiterpenoid, and Polyphenol compounds. This is evidenced by the results of simplicia ciplukan stem containing Kuinon, Saponin, Flavonoid, Monosesquiterpenoid compounds, while the ethanol extract of Ciplukan stem also contained Kuinon, Saponin, Flavonoid, Monosesquiterpenoid, and Polyphenol compounds. The difference in the results obtained in the Phytochemical Screening test for simplicia and extracts can be influenced by the solvent used when extracting, namely 96% ethanol solvent, so it is suspected that the solvent can dissolve polyphenol compounds, which can be proven in Phytochemical Screening tests using positive extracts containing compounds containing polyphenols.

Purification by column chromatography

Isolation is the process of extracting or separating natural compounds using an appropriate solvent (Djamal, 2008). The isolates in the 1-11 vials tested by TLC in the eluent hexane-ethyl acetate ratio (7: 3) produced a single blue spot, this spot was seen in UV lamps at 366 nm wavelength so that the compound was thought to be a compound pure flavonoid. This is consistent with the opinion (Achmad, S.A, 1986) which states that if the compound tested by TLC gives a single stain, then the compound from the isolation is pure. Chromatogram profile identification of groups of flavonoid compounds showed blue spots when read at 366 nm wavelength (Marliana et al., 2005). Chromatograms produced from the fraction of chromatography results are shown in Figure 4.2.

Figure 1. Mixture of hexane-ethyl acetate (7: 3), seen in UV lamps with a wavelength of 366 nm.

The value of Rf can be defined as the distance traveled by the component divided by the distance traveled by the solvent. Compounds with low Rf values are more distributed in the stationary phase, while high Rf values will be distributed in the mobile phase. Stains with low Rf values are more polar compared to high Rf values. Therefore the Rf number is always smaller than 1 cm. To maximize separation the range of Rf values lies between 0.2 to 0.8 cm.

Characterization by UV-Vis Spectrometry

The isolates obtained from TLC results were identified qualitatively by UV-Vis spectrophotometer. UV-Vis spectrophotometry is used to determine the absorbance value of compounds at the maximum wavelength. Spectrum measurements are carried out at wavelengths of 200-400 nm. The typical spectrum of flavonoid compounds consists of two maximums in the range 240-285 nm (band II) and 300-550 nm (band I). Flavonoid compounds of flavanone group according to Markham (1988) provide an absorption range at wavelengths 275-295 nm in band II and 300-330 nm (shoulder) in band I, the results of analysis using UV-Vis Spectrophotometry conducted by Markham can be seen in Figure 4.3.

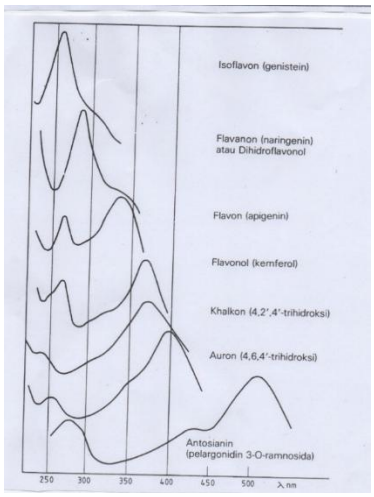

Figure 2. UV-Vis Spectrum Flavonoids with MeOH Solvents (Markham, 1988).

The results of the analysis using UV-Vis Spectrophotometry, isolates in the n-hexane fraction showed 2 bands at a maximum absorption of 229,263 nm in band II and shoulder shape at 301,902 nm which was band I. Compound of the isolation results showed that the Ciplukan stem was thought to contain flavonoid compounds of flavanon group , it is shown by the shape of the spectrum which is almost the same as the spectrum of flavanone contained in the library. The results of the analysis using UV-Vis Spectrophotometry can be seen in Figure 4.4.

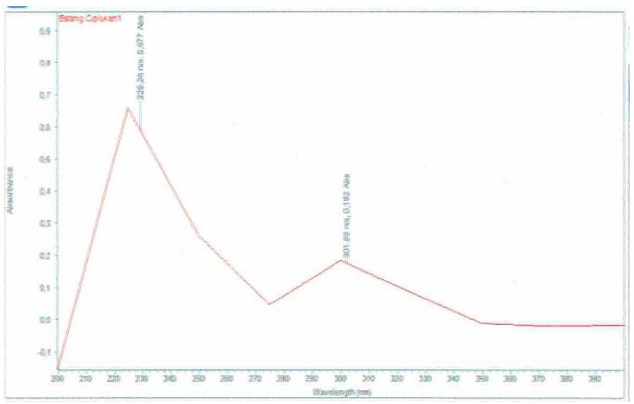

Figure 3. Isolate Spectrum using UV-Vis Spectrophotometry Information: Isolate Spectrum of Column Chromatography Fraction, Standard Solution: n.hexane, Interval: 25 cm.

IV. Conclusion

Flavonoid compounds are found on the Ciplukan stem. This is evidenced by the Phytochemical Test in both simplicia and Ciplukan stem extracts. The types of flavonoid compounds found on the Ciplukan stem are Flavanon compounds. This is evidenced by the results of Thin Layer Chromatography Test (TLC) and UV-Vis Spectrophotometry Test.

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