RASĀYAN J. Chem.
Vol. 11 | No. 2 |773 - 779 | April - June | 2018
ISSN: 0974-1496 | e-ISSN: 0976-0083 | CODEN: RJCABP
http://www.rasayanjournal.com
http://www.rasayanjournal.co.in

Thunbergia erecta L. FLOWER AS AN ALTERNATIVE ACID-BASE NATURAL INDICATORS

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ABSTRACT
As known that synthetic acid-base indicators can cause some environmental problems such as chemical pollution, availability problems and high cost. Therefore, many researchers are now trying to find an alternative indicator to substitute the synthetic indicators by optimizing the natural pigment of the plant. Related to this, an attempt to investigate the indicator activity of *Thunbergia erecta* L. flowers extract as natural indicators had been conducted. Flower pigment of the plant was extracted by maceration using ethanol and 1% HCl. The flower extract showed visible color change at pH from 1 to 14 and its absorbance was measured using a UV-Vis spectrophotometer. The indicator activities of flower extract had been applied in titration using a strong acid-strong base with given concentration. The extract showed an absorption band at 315 and 269 nm. The result showed pH range of indicators presented in at pH from 10 to 11 with 0.382 error percentage. Thus, *Thunbergia erecta* L. flower can be used as an alternative indicator to be commercial indicators that are more environmentally friendly.
Keywords: *Acid-Base, Alternative indicator, Flower, Natural indicator, Thunbergia erecta* L.

INTRODUCTION

Titration is a basic chemical technique for quantitative analysis of an unknown substance concentration using standard solutions of known concentration[1]. In the acid-base titration, the equivalent point is very difficult to observe because the reaction between an acid and base will yield a salt and water that are colorless. At this moment, the function of the indicator can be used in acid-base titrations[2].

Indicators are substances that change color according to the concentration of hydrogen ions (H^+) of the liquid or solution that is added. Measurement of the concentration of hydrogen ions in solution is required to determine the value of the acid dissociation constant. A good indicator is a weak acid or weak base that is slightly soluble in water[3]. The indicators used in the acid-base titrations generally is commercial indicators. Commercial indicators are relatively expensive and have a toxic effect on the user and can also cause environmental pollution[4]. In addition, the commercial indicator has weaknesses such as lack of availability problems[5]. For this reason, it has been a lot of research to find an alternative to substitute the commercial indicators with natural indicators derived from the natural products. These alternatives will be cheaper, more available, easier to be extracted, less toxic to the user and environmental friendly[4].

Natural products that can be used as acid-base indicators including fruits, flowers and plants have been studied for volumetric acid-base titration at room temperature, 60 °C, 92 °C, and 98 °C[6]. The followings are natural products that had been developed as acid-base indicators. *Jacaranda acutifolia* flower extracts can be used as acid-base indicators which have the same equivalent point with standard indicators. This indicator is suitable for weak acid-weak base titration[7]. Extracts of *Nerium indicum* had been effectively used as acid-base indicators that can replace phenolphthalein indicator because its availability is abundant, easily prepared, precise and accurate results[8]. Then, extracts of *Aspilia africana* with different solvents had also been applied to be acid-base indicators and can replace phenolphthalein indicator[9]. Natural indicators of roses (*Rosa setigera*), Alamanda (*Allamanda cathartica*), and Hibiscus (*Hibiscus rosa-sinensis*) can be used as a substitute for commercial indicators[10]. In addition, the water extract of *Hibiscus rosa-sinensis* and *Euclea natalensis* can be used as an alternative to acid-base indicators because they contain anthocyanin

http://dx.doi.org/10.31788/RJC.2018.1121844

and naphthoquinones. They contributed to change color depending on pH range[11]. Then, *Ipomoe cairica* and *Caesalpinia pulcherrima* extracts can also be used as a natural acid-base indicator[12]. The methanol and water extracts of *Euphorbia milli* and *Erythrina varigata* can be used to substitute acid-base synthetic indicators because they are more advantageous, economical, easy to prepare, simplicity, easy availability, environmental friendly, inert and accurate results[13]. The other natural pigments that can be effectively used as a substitute for commercial indicators are *Citrullus lanatus*[3], *Lawsonia inermis*[14], *Mirabilis jalapa* and *Punica granatum*[15], *Acalypha wilkesiana*[16], *Ipomoea biloba*[5], *Phyllanthus reticulatus*[17], *Syzygium cumini*[18], and *Argyreia cuneata*[19].

Thunbergia erecta L. is a species belonging to Acanthaceae family which is commonly grown as a houseplant that can grow in the tropics and subtropics climates. This plant has a beautiful purple color flower and no smell. In *Thunbergia erecta* L. flower (as seen in Figure 1) contain anthocyanin compounds because of its purple color. As reported that anthocyanins are naturally occurring pigments that can produce blue, purple, violet, magenta, and yellow. These water-soluble pigments were found in flowers, fruit and leaves of plants[20].

Fig.-1: *Thunbergia erecta* L. Flower[21]

EXPERIMENTAL

Material

The materials (chemicals) used in this study are *Thunbergia erecta* L. fresh flowers, ethanol, hydrochloric acid, sodium hydroxide, and aquadest.

Apparatus

The followings are some apparatus that are used in the study including vacuum rotary evaporator, UV-Vis spectrophotometer (Shimadzu UV-1800), pH meter, pipette, mortal and pestle, burette, analytical balance, beaker glass, test tube, erlenmeyer flask, buchner funnel, and vacuum pump.

Sample extraction

25 grams of *Thunbergia erecta* L. fresh flowers were crushed then macerated with a mixture of 50 mL ethanol 95% and HCl 1% for 60 minutes. Furthermore, the extract was filtered with a buchner funnel and filter paper, and evaporated through a vacuum rotary evaporator at 35 ^oC until the volume stayed 1/3 from the initial volume to yield *Thunbergia erecta* L. flower extract.

Determination of the range pH value for indicator obtained from *Thunbergia erecta* L. flower extract

As much as 5 mL of solution with different pH value (from 1 to 14) was poured into each test tube and then added with 3 drops of *Thunbergia erecta* L. flower extract. After that, it was observed the color change and continued to measure its absorbance using UV-Vis spectrophotometer at wavelength of 200-600 nm. The results are the presence of color change for *Thunbergia erecta* L. flower extract and the shift of absorption band in UV-Vis spectrum.

Determination of the error percentage for indicator obtained from *Thunbergia erecta* L. flower extract

As much as 10 mL of 0.1 N HCl was poured into erlenmeyer flask and then added with 3 drops of *Thunbergia erecta* L. flowers extract. Then, the mixture was titrated with NaOH 0.1 N. Next, it was observed the color change during titration and changes in pH of the solution. In order to know the error percentage of indicator (phenolphthalein) and indicator of the plant flower can be stated as follows:

$$\%(X-1) = \frac{[OH^-][H^+]}{C_A} x\ 100$$

Where:

(X-1) = error percentage
[OH] = concentration of hydroxide ion at the end point of the titration
$[H^+]$ = concentration of hydrogen ion at the end point of the titration
C_A = $\frac{C_A^o V_A}{V_A+V_B}$ in which C_A^o= concentration of HCl (titrate)
V_A = volume of HCl (titrate)
V_B = volume of NaOH (titrant)

RESULTS AND DISCUSSION

Determination the absorbance of *Thunbergia erecta* L. flower extract using UV-Vis spectrophotometer

The result can be seen in Fig.-1 and explained as follows:

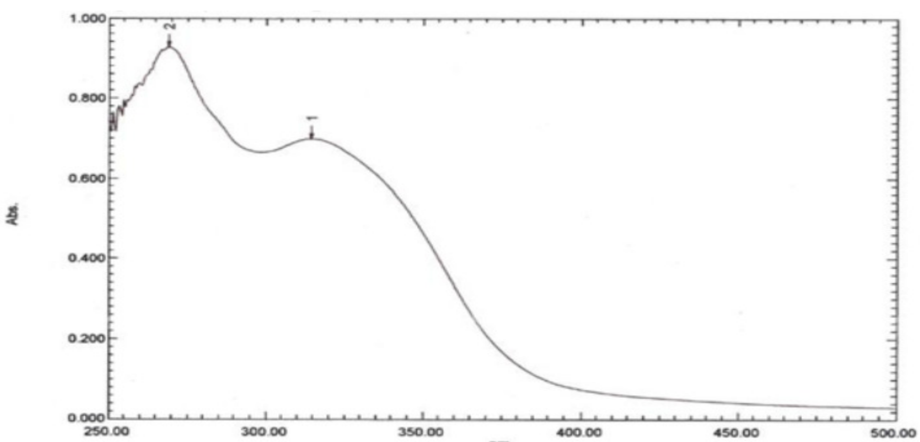

Fig.-1: UV-Vis spectrum of *Thunbergia erecta* L. flower extract

Figure-1 showed the UV-Vis spectrum of *Thunbergia erecta* L. flowers extract when added by HCl 1% that is a slightly acidic solution and has an absorption band at 269 and 315 nm. The function of the addition of this acidic solution in the extraction process is due to anthocyanins can be stable under this acidic conditions[20-22]. It meant that in acidic conditions, anthocyanins from *Thunbergia erecta* L. flowers will be more easily extracted.

The color change of indicator of *Thunbergia erecta* L. flower extract

The following is a color change from an indicator of *Thunbergia erecta* L. flower extract when mixed in solution with various pH value (from 1 to 14) as shown in Fig.-2.

Vol. 11 | No. 2 |773 - 779 | April - June | 2018

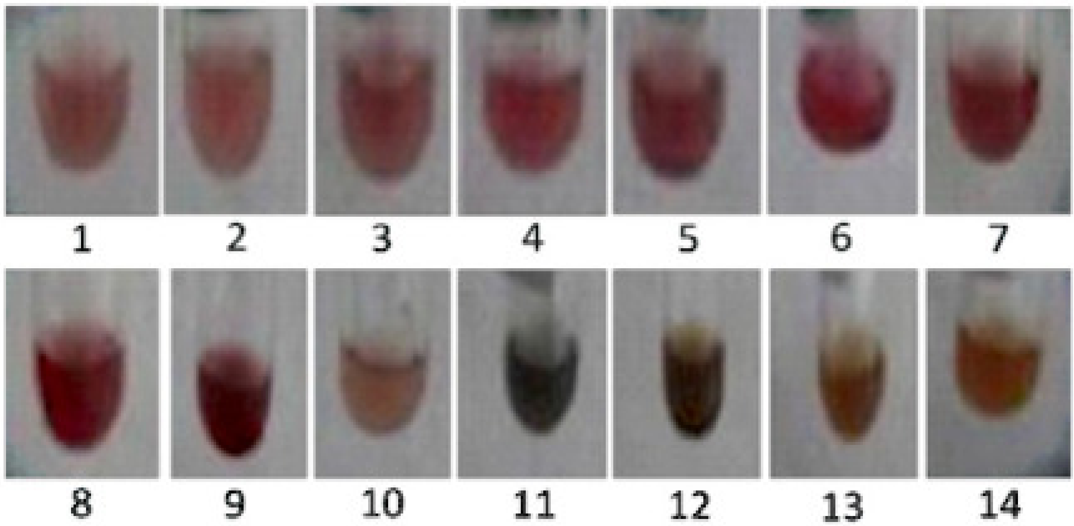

Fig.-2: The Color change of indicator of *Thunbergia erecta* L. flower extract in various pH value from 1 to 14

Figure-2 showed that the indicator at pH value from 1 to 9 has relatively stable color. In the acidic pH, anthocyanins are in the form of flavium cations which have little electrons, so the color is more stable[20,22]. According to Harborne (1987) declared that anthocyanins will be more stable in acidic solution as compared with neutral or alkaline solution[23]. On the other hand, it can be informed that color change of the indicator at pH value of 11 is reddish orange to green. Anthocyanins are the result of glycosylation of polyhydroxy and/or polymethoxy derivatives of 2-benzopyrolium salts or known as flavilium structures. Due to electro-deficiency, the flavilium core becomes highly reactive and only stable in acidic conditions[23]. The anthocyanin color changes with pH change and this is due to structural changes of the anthocyanin.

Determination the absorbance of *Thunbergia erecta* L. flower extract at various pH value (from 1 to 7 and from 8 to 14)

The respect absorbances (UV-Vis spectra) of the indicator resulted when it was mixed in solution with various pH value (from 1 to 14) can be shown by the different color as seen in Fig.-3 and Fig.-4.

Figure-3 and Figure-4 showed the UV-Vis spectra of an indicator of *Thunbergia erecta* L. flower resulted when it was mixed in solution with various pH value (from 1 to 14). The absorption bands shown by the indicator at pH value from 1 to 9 are not significant changes. For a while, the absorption bands of indicator began to shift at pH 10 and the absorption bands of it had exactly changed at pH 11 as shown in the UV-Vis spectra above. Based on these data, it can be determined that the indicator of *Thunbergia erecta* L. flower extract possess pH range from 10 to 11. The following are the change of absorption bands (I and II) of the indicator *Thunbergia erecta* L. flower extract at pH value from 1 to 14 as shown in Table-1.

Table-1 showed the shift of absorption bands for the indicator of *Thunbergia erecta* L. flower extract at pH value from 1 to 14. At pH value from 1 to 9, the indicator showed λ_{max} absorption band I (316.5 – 318.0 nm) and absorption band II (246.0 – 268.1 nm) that are not relatively change. While, at pH value from 10 to 11, the shift of absorption band I don't happen (no peaks). From this, it can be considered that the range of pH for indicator lied on pH value from 10 to 11. This was supported by changing color from orange to bluish green. In addition, at pH value from 12 to 14, the absorption band I shifted at λ_{max} from 360.5 nm to 364.0 nm whereas the absorption band II shifted slightly at λ_{max} from 276.0 nm to 278.0 nm. On the other hand, the indicator of *Thunbergia erecta* L. flower can be able to stabilize the color change at pH value from 1 to 9 with red color. Then, the indicator has pH range from 10 to 11 with a color change from orange to bluish green. For a while, the indicator can also be able to stabilize the color change from greenish yellow to yellow at pH value from 12 to 14.

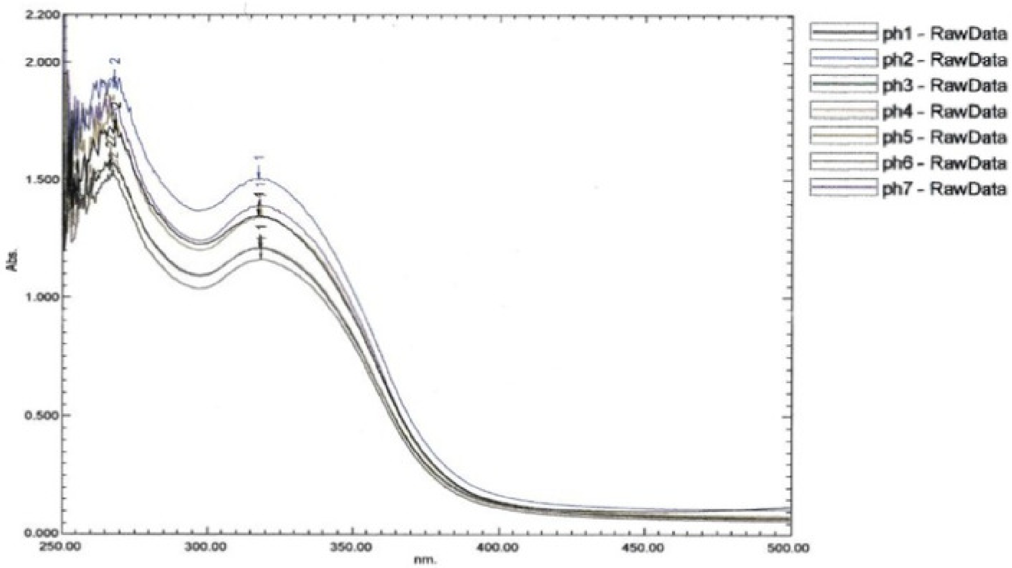

Fig.-3: UV-Vis Spectra of the indicator of *Thunbergia erecta* L. flower extract in solution with various pH value (from 1 to 7)

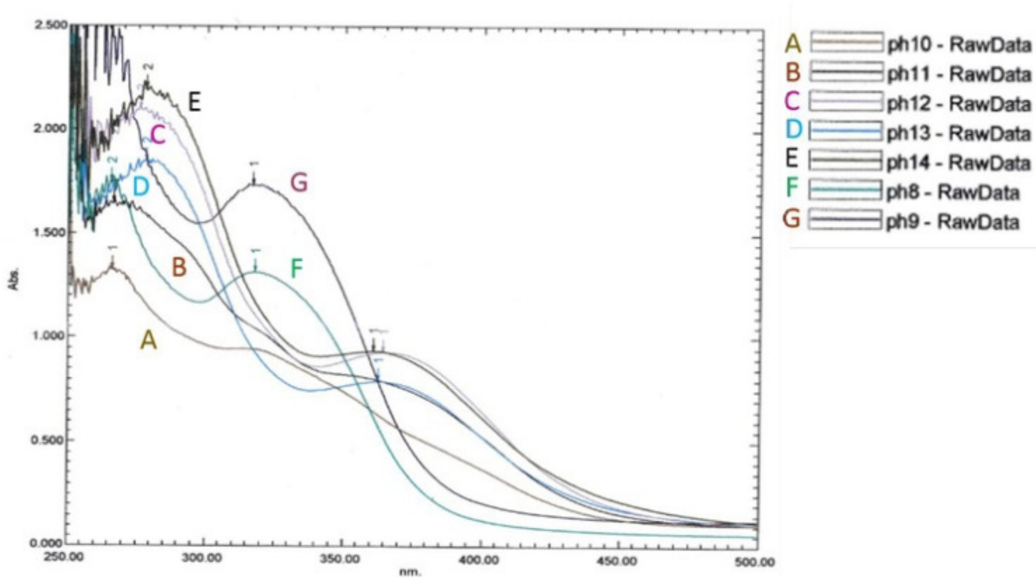

Fig.-4: UV-Vis spectra of the indicator of *Thunbergia erecta* L. flower extract in solution with various pH value (from 8 to 14)

Table-1: The change of absorption bands of the indicator *Thunbergia erecta* L. flower extract at pH value from 1 to 14

pH		1	2	3	4	5	6	7
Bands (λ_{max}, nm)	I	317.2	317.0	318.0	317.5	318.0	318.0	317.5
	II	268.1	267.5	266.5	266.5	266.0	267.0	264.5
pH		8	9	10	11	12	13	14
Bands (λ_{max}, nm)	I	317.5	316.5	No peak	No peak	364.0	362.5	360.5
	II	265.5	246.0	266.0	266.5	276.0	278.0	278.0

Determination of the error percentage for indicator obtained from *Thunbergia erecta* L. flower extract

A set of procedures to determine the error percentage of indicator of *Thunbergia erecta* L. flower extract had been conducted using the equation above and yielded a number of data as shown in Table-2.

Table-2: The Error percentage of the indicator *Thunbergia erecta* L. flower extract in the strong acid-strong base titration

HCl (mL)	An indicator of *Thunbergia erecta* L. flower extract			Phenolphthalein (PP)		
	NaOH (mL)	pH	% Error	NaOH (mL)	pH	% Error
10	20.00	10.07	0.356	10.10	8.13	2.696×10^{-5}
	20.10	10.13	0.409	10.10	8.11	2.586×10^{-5}
	20.00	10.10	0.382	10.20	8.15	2.839×10^{-5}
Average	20.03	10.10	0.382	10.13	8.13	2.707×10^{-5}

Table-2 displayed the error percentages through calculations results of indicator *Thunbergia erecta* L. flower extract. When applied in strong acid (HCl) and strong base (NaOH) titration, the indicator has the error percentage of 0.382%. For a while, indicator phenolphthalein as standard indicator possessed the error percentage of 2.707×10^{-5} %. In addition, *Thunbergia erecta* L. flower extract contains a natural anthocyanin pigment that changes its structure as a result of the color change of the solution under alkaline conditions.

CONCLUSION

An indicator of *Thunbergia erecta* L. flower extract showed λ_{max} absorption bands at 269 and 315 nm. The absorption bands of indicator began to shift at pH 10 and the absorption bands of it had changed at pH 11. It meant that indicator showed pH range from 10 to 11. Therefore, *Thunbergia erecta* L. flower can be an alternative acid-base natural indicators.

ACKNOWLEDGMENT

Our sincere thanks to Directorate of Learning and Student Affairs, the Ministry of Research, Technology and High Education, Indonesia for financial support for Student Creativity Program (SCP) with the SCP-RESEARCH Schema for the fiscal year 2015. Authors are also thankful to Department of Chemistry Department, Universitas Negeri Surabaya for providing facilities to complete this research.

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[RJC-1844/2018]