



UV/visible Spectroscopic Studies and Analytical Evaluation of *Dicliptera verticillate* Leaves Extracts as Eco-friendly Indicator for Acid-Base Titration

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Synthetic indicators usually employed in acid-base titration to determine the end point is expensive and non-biodegradable. Hence there is need to develop an eco-friendly and cost effective acid-base indicator from plant origin to serve as efficient alternative for the synthetic indicators. This research work focused on the development of easily available, cost effective and eco-friendly indicator from *Dicliptera verticillate* leaves extracts for acid-base titration. UV/visible spectrometer was used to analyzed the ethanol extract (EE), ethanol extract in basic medium (EEB), ethanol extract in acidic medium (EEA), hot water extract (HWE), Hot water extract in basic medium (HWEB) and hot water extract in acidic medium (HWEA) to determine the wavelength of maximum absorption (λ_{max}), and

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the absorbance of the extracts in different media in order to evaluate the chromophoric properties of the plant extracts phytochemicals before and after titrations. Titrimetric analysis was carried out in the reactions of strong acid/strong base (HCl/NaOH), strong acid/weak base (HCl/NH₄OH), weak acid/strong base (CH₃COOH /NaOH) and weak acid/weak base (CH₃COOH/NH₄OH). The results revealed that both extracts exhibit hypsochromic shift (shift to a shorter wavelength) with corresponding hypochromic effect (decreased in the absorption intensity) in both acidic and basic media. The titrimetric results revealed that the plant extracts give the end point comparable to that of the synthetic indicators such as methyl orange and phenolphthalein. Hence ethanol and hot water extracts of *Dicliptera Verticillate* can be utilized as suitable alternative indicators for acid - based titrations, since it is biodegradable and cost effective.

Keywords: *Uv/visible spectroscopy; natural indicator; Dicliptera verticillate; acid-base titration.*

1. INTRODUCTION

“Acid-base indicators are chemicals used to determine whether an aqueous solution is acidic, neutral, or alkaline. This is done by observing the change in colour of the solution when indicator is added. It is used to possibly determine the unknown concentration of acid or base through titrimetric analysis. When there is any change in the colour of analyte solution due to indicator, the titration is complete and the final volume of titrant is noted and further calculations are made to determine the concentration of the analyte” (Nwosu et al., 2014). Hence, indicators are used to determine the end point of any titrations. Examples of synthetic acid-base indicators include, phenolphthalein, methyl red and methyl orange etc.

“The search for natural compounds as acid-base indicators started increasing interest due to the non-biodegradability and high cost associated with synthetic indicator” [1]. “The advantages of using natural dye from plant extract as acid – base indicator is that it is cheap, easily available, easy to prepare, simple to extract, nontoxic and environmentally friendly since it can easily degrade when released to the environment” [1]. The colour change in the titrimetric process may be attributed to the type of phytochemicals present in the plant extract [2]. “The major phytochemical responsible for the colour change of the natural indicator extracted from the plant source is anthocyanin which is a subgroup of flavonoids. It is a secondary metabolite that contributes to the purple, blue or red colours in fruits, leaves or flowers. Anthocyanin can react or interact with acids or bases, resulting in changes at its molecular structure and hence the exhibition of colour changes” [3]. All the anthocyanin has been considered to be derivatives of 3,5,7-Trihydroxy flavylum chloride. Various anthocyanins and anthocyanidins differ

in the nature, number and position of hydroxyl groups, methoxy groups and sugar residue on their structural moiety [3].

The use of natural indicator from plant extract have been reported by many authors. Nwosu, [4] reported the potentials of natural indicator from Hibiscus (red species), Bougainvillea and rose flowers. Industrial and analytical potentials of the plant extracts from the fruit of *Telfrair occidentalis* have also been reported by Azundo, et al., [5]. Utilization of Hibiscus flower extract, mango seed extracts, ginger stem extract and kola nut seed extract as natural indicators for acid – base titration has been reported [6]. Akpakpan et al., [2], reported the acid-base indicator potentials of two varieties of kola nut extracts. Potentials of *Dissotis rotundifolia*, *Centrosema pubescens* and *Allium cepa* extracts as indicators for acid-base titration was reported by Odiogonyeni et al., [7].

“However, there is no report on the utilization of aqueous and ethanol extracts of *Dicliptera verticillata* leaves as acid base indicator. *Dicliptera verticillata* (Forssk). is a perennial herb, occurring in several parts of the world including Mauritania to Niger, Nigeria, India, Burma, Southern Africa, Zambia, Senegal, Sudan and Lake Chad” [8]. “*Dicliptera verticillata* is a medicinal plant used for the treatment of malaria and diarrhoea in Burkina Faso. In India, the leaves are used in preparation of various soups. It is also used as flavouring and spicing agent” [8]. “The phytochemical constituents of the ethanol and aqueous extracts of this plant leaves” have been reported by Akpakpan et al., [9]. The plant contains several phytochemicals which can enhance colour changes during acid - base titration.

However, this present study is aimed at assessing the potentials of this plant extracts as green indicators for acid-base titration in order to

ascertain their suitability or otherwise in replacing the synthetic indicators which are not environmentally friendly.

2. MATERIALS AND METHODS

2.1 Samples Collection and Preparation

Fresh samples of *Dicliptera verticillata* leaves were collected from a farm in Eket road, Mkpata Enin local Government, Akwa Ibom State, Nigeria. The samples were identified by a botanist in University of Uyo, Nigeria.

The samples were washed, squeezed and cut into pieces using knife. 20% extracts were prepared by soaking 20 g of the fresh sample separately in 100 ml of ethanol and hot water, after 24 hours, the extracts were filtered using Whatman filter paper and the filtrates obtained were characterized using UV/visible spectrometer, and was used as indicator for the titration to evaluate their indicator properties.



Fig. 1. *Dicliptera verticillata* plant

2.2 Experimental Procedure

2.2.1 Characterization of *Dicliptera verticillata* extract using UV-VIS Spectroscopic analysis

UV/visible Spectroscopic analysis was conducted on the *Dicliptera verticillata* extract at room temperature. The extracts were diluted to 1: 10 (v/v) using the same solvent. The raw extracts, and the extracts in acidic and basic medium were analyzed using Genesys spectrophotometer in the wavelength ranging from 200 - 800 nm, scan speed 5 nm/s and 1nm resolution in order to generate their characteristic absorption spectra.

2.2.2 Titrimetric analysis

The experimental work was done using the extracts of *Dicliptera verticillata* leaves. Another

experiment was also carried out using the standard indicator (phenolphthalein and methyl orange) [10]. Exactly 25 ml of each base (sodium hydroxide or ammonium hydroxide) was measured into a conical flask and 1.0 ml of the *Dicliptera verticillata* leaves extracts was added using a 1.0 ml dropper. A 50 ml burette was filled with either hydrochloric acid or ethanoic acid and titrated against each base (sodium hydroxide or ammonium hydroxide) until a sharp colour change was observed signifying the end of the reaction. Another titration was carried out using the standard indicator (phenolphthalein and methyl orange). Each titration sequence was done in triplicate using either the standard indicator or the plant extract and the end points recorded. The mean value was calculated for each set of titrations. The titrations were carried out for the strong acid vs strong base (HCl/NaOH), strong acid vs weak base (HCl/NH₄OH), weak acid vs strong base (CH₃COOH/ NaOH) and weak acid vs weak base (CH₃COOH/ NH₄OH). Titration was done using 0.5 M acids and 0.5 M bases, the end point values obtained are presented in Table 2.

2.3 Colour Changes

The colour of the extracts and their colours in acidic and basic medium are presented in Table 3, and Figs. 3 and 4.

3. RESULTS AND DISCUSSION

3.1 UV/visible Spectroscopic Analysis

UV/visible absorption spectra of ethanol and hot water extracts of *Decliptarea verticillata* leaves in different media is presented in Fig. 2, and their wavelength of maximum absorption with corresponding absorbance are presented in Table 1.

The variations in the absorbance and wavelength of maximum absorption of the extracts in different media is due to the presence of different forms of phytochemicals especially anthocyanin content in the plant extract. Absorption range of 230-270 nm indicate the transition electrons from lone pair and pi-electrons to antibonding sigma and pi bond in the phytochemicals present in the extracts. This absorption is consistent with the dominant group of hydroxyls, methoxy and benzene conjugated group in anthocyanin structures.

Table 1. wavelength of maximum absorption and the corresponding absorbance of each extract

Samples	λ_{max} (nm)	Absorbance
Ethanol extract (EE)	648 - 674	0.500
Ethanol extract in acidic medium (EEA)	650	0.202
Ethanol extract in basic medium (EEB)	648	0.215
Hot water extract (HWE)	558 - 600	0.500
Hot water extract in acidic medium (HWEA)	576	0.027
Hot water extract in basic medium (HWEB)	592	0.147

Comparing the wavelength of maximum absorption (λ_{max}) of ethanol extract and hot water extract, the absorption maxima of ethanol extracts is higher than that of hot water extract, this may be due to the fact that ethanol extract contains both the polar and some non-polar phytochemicals from the plant. The λ_{max} of ethanol extract ranges from 648 – 674 nm, at the absorbance of 0.5. The λ_{max} of both ethanol and water extracts in acidic and basic medium were within the ranges of that of the raw extracts, with corresponding decrease in absorbance. This is due to the dilution that takes place when these extracts were introduced into these media. This hypochromic effect was higher in acidic medium than basic medium.

“*Declipitarea verticilata* leaves extract showed multiple absorption peak in visible region, 500 nm – 700 nm and similar absorption peaks in UV region, 225-270 nm. In the visible region, absorption band corresponding to the complementary colours were observed” [11].

“Whereas multiple peaks in UV region was attributed to the multiple transitions from the ground state to the excited state of the phytochemical functional groups present in the extracts” [12].

3.2 Titrimetric Analysis

The results of the titrimetric analysis using *Dicliptera verticillata* leaves extracts and commercial indicators are presented in Table 2.

In strong acid/ strong base titration, the average titre value of *Dicliptera verticillata* leaves extracts was 28.00 cm³ and 28.08 cm³ for ethanol and hot water extracts respectively, whereas methyl orange gives a titre value of 28.00 cm³ and phenolphthalein 28.03 cm³. These results indicate that *Dicliptera verticillata* leaves extracts has end point comparable with standard indicators, hence it can serve as a suitable replacement for commercial indicator.

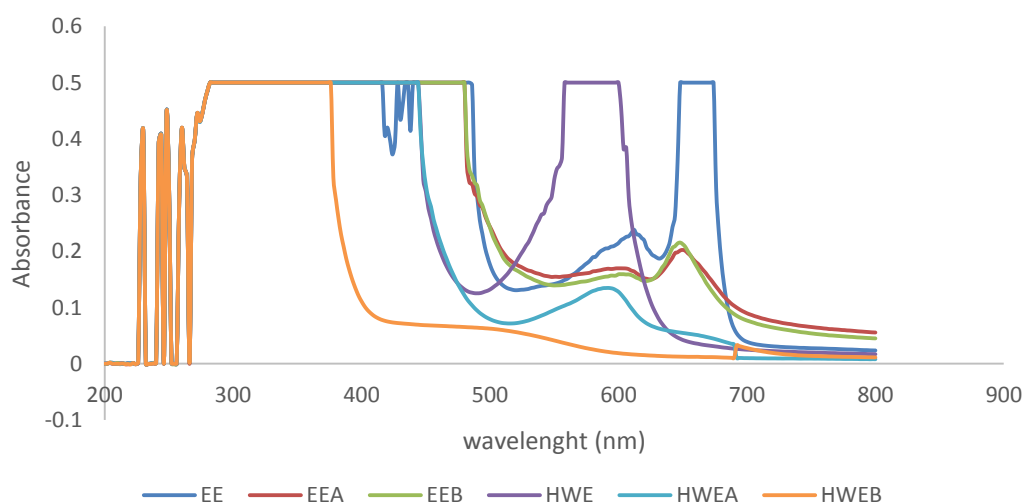


Fig. 2. UV/visible absorption spectra of ethanol and hot water extracts of *Declipitarea verticilata* leaves in different media

Table 2. Titrimetric analysis using ethanol and hot water extracts of *Dicliptera verticillata* leaves and commercial indicators

Acid/base	End point (ethanol extract)	End point (hot water extract)	End point (Methyl orange)	End point (Phenolphthalein)
HCl/NH ₄ OH	No visible colour change	No visible colour change	37.02 ± 0.01	34.06 ± 0.00
HCl/NaOH	28.00 ± 0.01	28.08 ± 0.01	28.00 ± 0.03	28.03 ± 0.03
CH ₃ COOH/NH ₄ OH	No colour change	33.02 ± 0.01	No visible colour change	No visible colour change
CH ₃ COOH/NaOH	30.00 ± 0.01	26.04 ± 0.01	32.40 ± 0.02	30.32 ± 0.01

However hot water extract gave a remarkable colour changes for all titrations while ethanol extracts gave colour changes for the titration of strong acid with strong base and weak acid with strong base. Hence ethanol extract is not recommended for the titration of strong acid with weak base, and weak acid and weak base.

Comparatively, *Dicliptera verticillata* leaves extracts gave titre values less than or comparable to that of the commercial indicators for all titrations, hence titration using this plant

extracts as indicator will consumes small volume of acids to reach the end point.

The titrimetric equations of reactions which occur during the titration processes, in the presence of these indicators are presented in equation 1- 4.

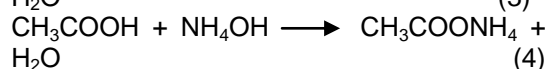
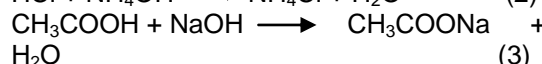
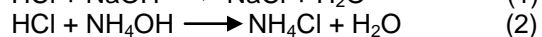
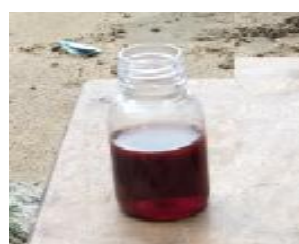


Table 3. Colour of the extracts in acidic and basic medium

Extracts	Colour of the extracts	Colour in Acid	Colour in Base
Hot water extract	Red	Light Red	Dirty green
Ethanol extract	Redish brown	Redish brown	green



Hot water extract (Red)



Hot water extract in base (Green)



Hot water extract in acid (Light Red)

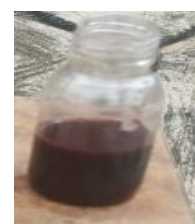
Fig. 3. Colour of the hot water extract in different medium



Ethanol extract (Redish brown)



Ethanol extract in base (dirty green)



Ethanol extract (Redish brown)

Fig. 4. Colour of ethanol extract in different media

“Plant extract showed different colours in different media, this is because of the changes in the chemical structure and functional groups (Chromophore) of its phytochemicals in different media” [13]. The plant extracts gave remarkable colour changes as shown in Figs. 3 and 4 [14].

4. CONCLUSION

This study was carried out to evaluate the potentials of ethanol and hot water extracts of *Dicliptera verticillata* leaves as eco-friendly acid-based indicator to serve as an effective replacement for synthetic indicators. The UV/visible spectroscopic studies revealed that the absorbance and λ_{max} of the extract changed in different media due to the changes in the structural features of phytochemicals/ chromophore present in the leaves extract. The results obtained for the titration using *Dicliptera verticillata* leaves extracts as indicator were comparable with that of synthetic commercial indicators. Hot water extract gives sharp colour changes than ethanol extract. Hence it is highly recommended that these extracts of *Dicliptera verticillata* leaves should be utilized as green indicator for acid-base titration especially at the primary and secondary school levels of education since they are easily available, cheap, biodegradable and eco-friendly.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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