Research Article

Flower Extract as an Organic Indicator in Acid Base Titration

Saloni Desai *, Dhruti Ganatra, Anjali Maisuriya, Ishali Patel
ROFEL Shri G M Bilakhia College of Pharmacy, Vapi, Gujarat, India.
***Corresponding author's E-mail:** salonidesai161296@gmail.com

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ABSTRACT

Indicators are very essential chemicals. Therefore, for all types of acid base titration, synthetic indicators have always been the preferred option. As they show a sharp color change at intervals of pH in all types of titrations. However, due to certain disadvantages like high cost, availability problems, long process and complex synthesis, hazardous waste product and environmental issues, an attempt is required to substitute synthetic indicator with natural indicator. From maceration of flowers, anthocyanin color pigment was produced. A UV-Visible spectrophotometer was used to measure the quantitative amount of color pigment.

Keywords: pH indicator, natural pigments, end point, titrations, Anthocyanin.

INTRODUCTION

Chemical substances to gauge the amounts of acid and base are referred to as acid-base indicators. Under certain pH levels, this indicator will change colour. This indicator is useful to indicate the acidic and base on hydrogen ion concentration due to titration process. To mark the equivalence point of acid base titration or to determine the current pH of medium, acid-base indicators are frequently used.[1] Indicators are pigments or colour that can be extracted from many sources such as plants, fungus and algae.[2]

The aqueous extract of *Dendrobium sp. & Hippeastrum puniceum* flower exhibits striking shift in colour with change in pH. Because of this feature, it can be used as green indicator instead of more traditional synthetic indicator. Like phenolphthalein which are chemical based and may be harmful to human health. Some scientist has successfully tried to invent the natural indicator such as methanolic extract of the flowers of Targets erecta, Dianthus plumaris and Antirrhinum majus, Morus alba, Rosa indica, Hibiscus rosa sinesis etc.[1] In present study we used *Dendrobium sp. & Hippeastrum puniceum* flower's aqueous extract as natural and effective indicator for acid-base titration.

Orchids are member of the Orchidaceae family, a large and diverse collection of blooming plants with blooms that are frequently vibrant and fragrant. The family Orchidaceae contains the genus Dendrobium, which is primarily composed of epiphytic and lithophytic orchids. The petals of the blooms typically bend backwards and are 3-5 cm broad. The labellum, which has three lobes, is the petal that extends forward. Orchids have adapted to a broad range of settings including the high altitude of Himalayan Mountains and the dry climate of the Australian desert. They are hailed from either Japan or China. [3]

Hippeastrum puniceum is a bulbous perennial that originated in South America's tropical regions but has since naturalized. It belongs to *Amaryllidaceae Family*. Every leaf on a plant is bright green, strap-shaped (lorate), 30-60 cm long by 2.5-3cm wide, and tapered at the end to and acute apex. Plants contain four to six leaves per plant. The petals are orange red-with paler bases.

A type of organic pigments called anthocyanin, which may change colour with pH, is found virtually every flower that is red, blue or purple.[4] Various natural colored substances, like grape juice, brown tea, and various floral pigments, change hue when the environment's acidity or alkalinity. These substances are called acid-base indicators. [5] Most indicators currently in use are synthetic. Natural indicators obtained from diverse plant parts like flowers, fruits, and leaves will be more useful because synthetic indicators have some drawbacks like high cost, availability, and chemical contamination.[6] Additionally, some of these synthetic indicators can cause environmental pollution, gastrointestinal pains, skin rash, eruptions, erythema, and epidermal necrosis in addition to harmful consequences on users such diarrhea, pulmonary edema, hypoglycemia, and pancreatitis.[7]

The search for alternatives has gained more traction as a result of these justifications for the harmful consequences of synthetic indicators. These alternatives from plant origin are probably cheaper, readily available, ways to extract, less toxic to users, and environmental friendly.[8] Shown in Figure 1.

Scope of Research

Orchids and lilies were selected for research because the flower extracts and prepared indicators were more stable as compared to other flowers (bougainvillea, rose, and marigold)

Figure 1: Purple Orchids, White Orchid, Lily Flower

MATERIALS AND METHODS

Plant materials

Barbados lily flowers were collected from ROFEL Shri G.M Bilakhia College of Pharmacy's medicinal garden in the month of April. White and Purple Orchids were collected from the temple trash. The authenticity of each flower is validated at the biology department of B.K.M Science College, Valsad.

Reagents and Glass wares:

The study was effectively carried out utilizing analytical grade reagents that were made accessible by ROFEL Shri G.M. Bilakhia College of Pharmacy. The entire experimental activity was carried out using a clean and consistent set of glassware. According to the Indian Pharmacopoeia, the reagents and volumetric solutions were prepared.

Preparation of flower extract:

Flower petal were crushed in a mortar and then transferred into conical flask with 100ml water, to yield concentrated extract, followed by the extraction technique known as maceration. Each extract was then kept in tightly sealed container and kept out of direct sunlight.

Methods:

pH determination on the basis of colour changes:

By Test-tubes: In 2 test tubes add few drops of acidic and basic solution respectively, and add few drops of prepared extract in both the test tubes and after that Observe the color change. Results shown in **Table no. 1 and Figure 2.**

By pH paper: Rub the flower petals on paper strips and dry, dry them at room temperature for few minutes and then Put few drops of acid and base on the prepared strips. Results shown in **Table 1 and Figure 2.**

Table 1: pH Determination

Flower	Color change	
	pH PAPER	TEST TUBE
White Orchid	Acidic: -Colorless Basic: - Green	HCl-Colorless NaOH-Green
Lily	Acidic- Orange Basic- Yellow	HCl- Pink NaOH-Yellow
Purple orchid	Acidic- Pink Basic- Yellow	HCl-pink NaOH-green

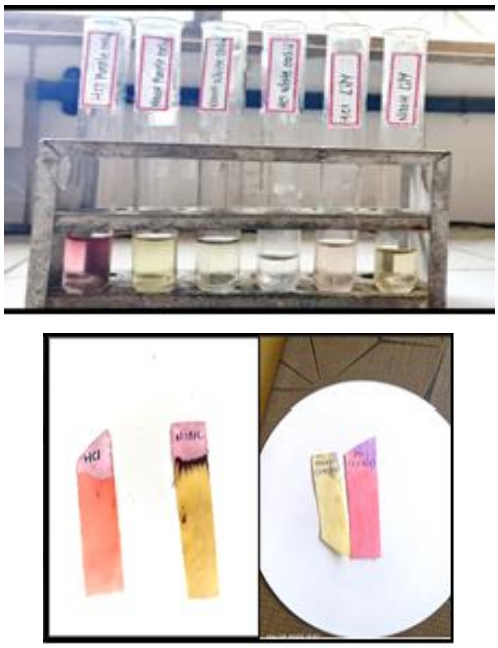

Figure 2: Test sample in acid base solution and pH Paper

ACID – BASE TITRATION:

Strong Acid Vs Strong Base: Burette: NaOH, Conical flask: 10 ml HCl, Standard indicator: phenolphthalein, Organic indicator: extract of orchids and lily (1ml).

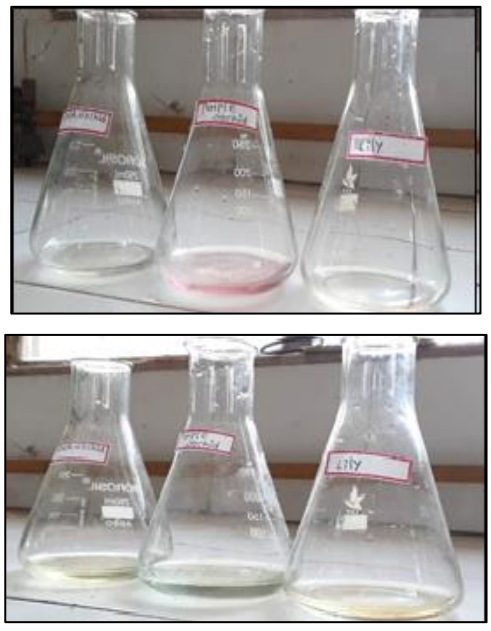

Figure 3: i) Colour Before titration, **ii)** Colour After titration

Stability Study:

Extract was stored in cool temperature. Stability study was carried out by using titration method. The stability study is observed up to 15 days. Observation shown in **Table 2**.

Table 2: Stability Study

Days	White Orchid	Purple Orchid	Barbados Lily
Day-1	9.8ml	9.3ml	10.6ml
Day-3	9.3ml	9.8ml	10.1ml
Day-5	9.5ml	9.9ml	10.0ml
Day-7	9.5ml	9.7ml	9.9ml
Day-9	10.0ml	9.9ml	10.0ml
Day-11	10.5ml	9.6ml	10.0ml
Day-13	10.2ml	9.4ml	9.8ml
Day-15	10.1ml	9.5ml	9.7ml

Detection of Anthocyanin:

For testing of anthocyanin level using a differential pH, for 1ml of flower extract, added 9ml of KCl solution pH 1, put into 10ml volumetric flask and then homogenized. The same was done for the $CH_3COONa.3H_2O$ solution pH of 4.5. The absorbance of anthocyanin extract was measured at a maximum wavelength of 500 nm and 700 nm then recorded at that wavelength range.[9] The test was carried out two replicates where the anthocyanin level using differential pH method using equation 1 and shown in **Figure 4**.

Figure 4: Detection of anthocyanin

$$A = (A_{520} - A_{700})\ pH_1 - (A_{520} - A_{700})\ pH_{4.5}$$

$$\textbf{Anthocyanin level} = \frac{AxMWxDFx1000}{\varepsilon x1}$$

Where,

A= Absorbance

MW= the molecular weight of cyanidine-3-glucoside (499.2g/mol)

ε = molar absorptivity of cyanidine-3-glucoside

DF = dilution factor

1= cuvette wide (cm)

Calculation:

1) Calculation for Purple orchid

A = (A520nm–A700nm) pH 1.0 – (A520nm–A700nm) pH 4.5

= (0.1170-0.0027) pH 1.0 – (0.0640-0.0037) pH 4.5

= (0.1143)- (0.0603)

= 0.0603

$$\text{Anthocyanin pigment (g/l)} = \frac{AxMWxDFx1000}{\varepsilon x1}$$

$$= \frac{0.0603x449.2x10x1000}{26900x1}$$

$$= \frac{270,867.6}{26,900x1}$$

= 10,069.427 gm/l → 1.0069427 mg/100ml

2) Calculation for White orchid

A = (0.0096-0.0054) pH 1.0 – (0.0116-0.0076) pH 4.5

= (0.0042) - (0.00008816)

= 0.00411

$$\text{Anthocyanin pigment (g/l)} = \frac{AxMWxDFx1000}{\varepsilon x1}$$

$$= \frac{0.00411x449.2x10x1000}{26,900x1}$$

$$= \frac{18462.12}{26900x1}$$

=0.6863mg/l

3) Calculation for White orchid

A = (0.0133-0.0002) pH 1.0 – (0.0143-0.0057) pH 4.5

= (0.0131) - (0.0086)

= 0.0045

$$\text{Anthocyanin pigment (g/l)} = \frac{AxMWxDFx1000}{\varepsilon x1}$$

$$= \frac{0.0045x449.2x10x1000}{26900x1}$$

$$= \frac{20241}{26900x1}$$

=0.7514mg/l

RESULTS

Table 3: UV Absorbance Data of Purple Orchid, White Orchid, Barbados Lily

Sr. No.	λ max	Absorbance	
Purple Orchid			
		KCl	**Sodium Acetate**
1.	520 nm	0.0096	0.0116
2.	700nm	0.0054	0.0076
White Orchid			
1.	520 nm	0.0133	0.0143
2.	700nm	0.0002	0.0057
Barbados Lily			
1.	520 nm	0.1170	0.0640
2.	700nm	0.0027	0.0037

Table 4: Observation table of Titration (Barbados lily, Purple Orchid, White Orchid)

Standard (Phenolphthalein)		Extract	
Initial	**Final**	**Initial**	**Final**
Barbados Lily			
0.0ml	9.8ml	0.0ml	10.0ml
0.0ml	9.7ml	0.0ml	10.1ml
0.0ml	9.6ml	0.0ml	10.1ml
Mean	9.7ml	**Mean**	10.0ml
Purple Orchid			
0.0ml	9.3 ml	0.0ml	9.3ml
0.0ml	9.4ml	0.0ml	9.0ml
0.0ml	9.3 ml	0.0ml	9.1ml
Mean	9.3ml	**Mean**	9.1 ml
White Orchid			
0.0 ml	10.1 ml	0.0ml	9.8 ml
0.0 ml	10.0 ml	0.0ml	9.9 ml
0.0 ml	10.1 ml	0.0ml	9.7 ml
Mean	10.0 ml	**Mean**	9.8 ml

The results of the study demonstrated that the equivalence points of acid-base titrations using each different flower extract either coincide with or were nearly as close to those of using standard phenolphthalein indicator, with each flower extract indicator giving a sharp colour change at the equivalence point during titration, shown in Figure 5,6,7 and Table 3 and 4.

CONCLUSION

The research carried out found that the aqueous extract of each flower from *Dendrobium Sp. & Hippeastrum puniceum* can be used as a replacement for synthetic indicator because of its benefits, including ease preparation, efficiency, and the capacity to produce accurate and precise results in accordance with green chemistry.

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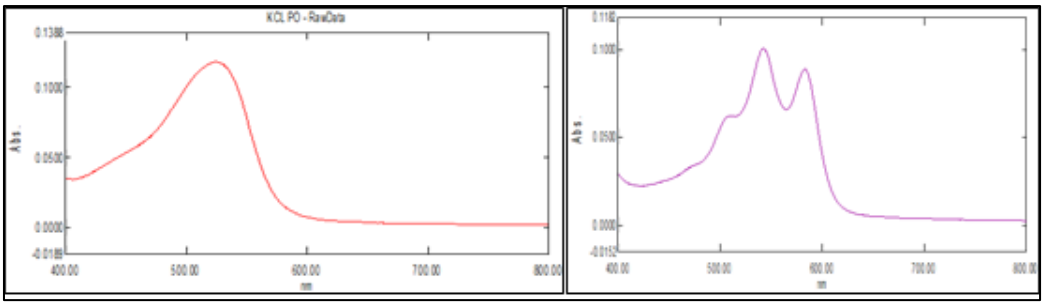

Figure 5: Spectra of Purple Orchid in KCl and In Sodium Acetate

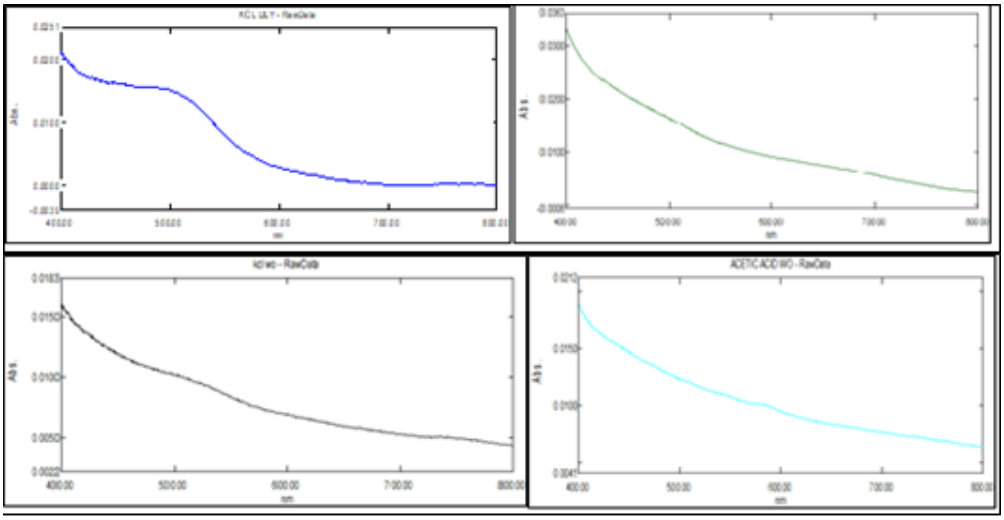

Figure 6: Spectra of Lily and White Orchid in KCl and In Sodium Acetate

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