

**EVALUATION OF BEETROOT AS NATURAL INDICATOR****Dipali Dhumal, \* Savita D. Sonawane Sanjay K. Bais,***Fabtech College of Pharmacy, Sangola**Tal-Sangola, Dist.-Solapur**Maharashtra -413307***ABSTRACT**

*Beetroot (Beta vulgaris) is fleshy, large root growing in the plant of the same name. Beetroot Ranks in top 10 vegetables with respect to its antioxidant capacity. They possess antimicrobial and antiviral Effects. Beetroot exhibits dark crimson red color pulp which has a sweet taste. They impart this color due to Presence of specific chromophore group which is used in food and cosmetic industries. The present manuscript Focuses on the extraction of Beta vulgaris pigment and it's use as an indicator and preparation of pH indicator Strips. In this research endeavour, beetroots were procured, dried, powdered and used as a natural indicator. This indicator can be a substitute for synthetic indicators. Synthetic indicators have teratogenic and carcinogenic Effects, allergic effect, hence they possess a great health hazard. In comparison to synthetic indicator, natural indicators are environment friendly, bio-degradable and have less health hazards.*

*A study has been done to investigate the indicator activity of aqueous extract of root pigments and compared with that of already existing synthetic indicators. Pigments were extracted using hot water and a definite volume was added which gave accurate and reliable results for all the four different types of neutralization titrations - strong acid against strong base, strong acid against a weak base, weak acid against strong base and weak acid against weak base. The work proved to be acceptable in introducing root pigments as a substitute to the synthetic acid-base indicators.*

**Keywords:** *Indicator, BetaVulgaris, Titration, Endpoint, betaxanthins, Betanin.*

\*Corresponding Author Email: - [dipalidhumal2912@gmail.com](mailto:dipalidhumal2912@gmail.com)

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## INTRODUCTION

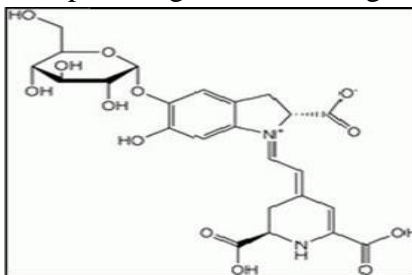
Beta vulgaris or beetroot, also known as the table beet, garden beet, red or golden beet. Beta vulgaris of Family chenopodiaceae, contains bioavailable compounds and micronutrients such as phenolic compounds, Carotenoids, betalains, vitamins and minerals.<sup>1</sup> Beetroot is a potential source of valuable water -soluble nitrogenous pigments, called betalains, which Are composed of two main groups, the red betacyanin sand the yellow betaxanthins. The colour of Betanin depends on pH increases. Once the pH reaches alkaline levels Betanin degrades by hydrolysis, Resulting in a yellow-brown colour.<sup>2</sup> Natural pigment in plants are highly colored substance and may show colour changes with variations of pH. But due to environmental pollution, availability and cost, the search for natural compounds as an acid –base indicator was started.<sup>3</sup>



**Figure No. 1: Beta Vulgaris Root**

Titrimetric analysis involves the determination of the volume of a solution of known concentration (standard), required to react quantitatively with a solution of the substance to be analysed. An Indicator's Colour change occurs over a range of hydrogen ion concentrations. This range is termed the colour change Interval.<sup>4</sup>

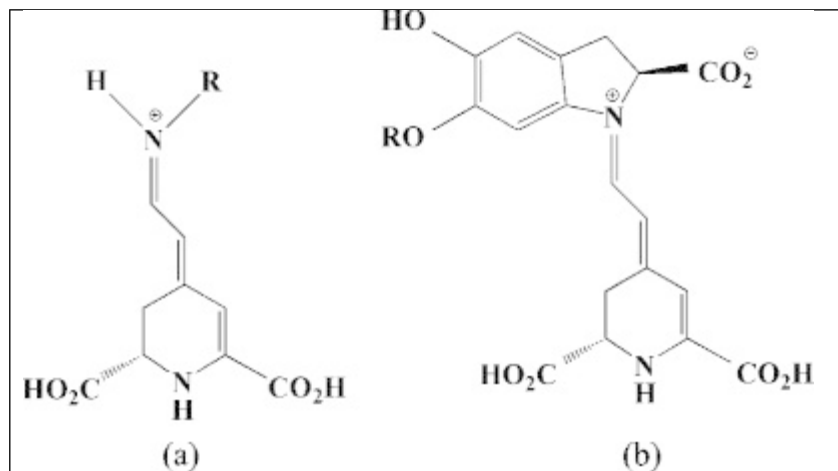
The beet is derived from the wild beet or sea beet (*Beta maritima*) which grows on the coasts of Eurasia.2 Ancient Greeks called the beet teutlion and used it for its leaves, both as a culinary herb and medicinally. The Indians also used the beet medicinally, but were the first to cultivate the plant for its root.<sup>5</sup> They referred to the beet as beta.3 Common names for the beet include: beetroot, chard, European sugar beet, red garden beet, Harvard beet, blood turnip, maangelwurz, mangel, and spinach beet.



**Figure No. 2: Betanin**

The beetroot, commonly called the beet, is a biennial plant that produces seeds the second year of growth and is usually grown as an annual for the fleshy root and young leaves.<sup>6</sup> The *Betavulgaris* has three basic varieties: chard, grown specifically for its leaves; beets, grown for its bulbous root, with edible leaves (with varieties in white, yellow and red roots); and sugar beets, grown for making sugar from the long, thick root. The beet is a root vegetable with purple-green variegated leaves. It has either a taproot with minimal secondary roots, or in the case of the sugar beet, a long carrot-like root with minimal secondary roots. The young plants, which are generally thinned by hand, are good to eat as young leaves (tasting somewhat like spinach).

The beet is propagated by seed (clusters holding from three to five seeds).<sup>7</sup>



**Figure No. 3: Betaxanthin and Betacyclin**

Commercial indicators are expensive and some of them have toxic effects on users and can also cause environmental pollution. For these reasons there has been an increasing interest in searching for alternative sources of indicators from natural origins.<sup>8</sup> These alternatives would be cheaper, more available, simple to extract, less toxic to users and environmentally friendly. Volumetric analysis is one of the key quantitative techniques used to analytically determine both inorganic and organic acid interaction with strong or weak acids and bases in raw materials, intermediates and finished products for quality assurance purposes.<sup>9</sup> This is accomplished via the use of appropriate weak organic dyes or acids pH indicators.

#### ADVANTAGES

To determine the pH of a solution using substances found in nature, such as fruits, flowers, instead of synthetic.

Natural indicators change in color in response to changes in the acidity or alkalinity of solution, providing a visual indication of its pH level.<sup>10</sup>

To utilize natural substances, such as plant extract.

These indicators offer an alternative to synthetic indicators, promoting sustainability and reducing environmental impact.

#### MATERIAL & METHODOLOGY

The entire Hydrochloric acid (HCL), Sodium Hydroxide (NaOH), Acetic acid (CH<sub>3</sub>COOH), Ammonia (NH<sub>3</sub>), methyl red, methyl orange was of analytical grade.

## Preparation of Indicator

100.0 gram of chopped Beetroot was added 100 ml of solution containing Ethanol- Hydrochloric acid (v/v ratio 99:1) and Ethanol- Water (v/v ratio 1:2) for 45 min, after allowing the beaker to cool for 15 minute, boiled Beetroot were squashed and the liquid was filtered .

The residues were squeezed once again and the liquid has evaporated to get a highly concentrated portion of the indicator. The extract was preserved in tightly closed container and stored away from the direct sun light.<sup>11</sup>

## Testing the Indicator

Beetroot extract

pH buffer solutions or household acids/bases (e.g., lemon juice, vinegar, baking soda, ammonia)

Test tubes or small containers

pH paper or pH meter (for comparison)

Add a few drops of the rose petal extract to each solution and observe the color change.<sup>12</sup> The Acidic solutions (pH < 7) typically turn the extract pink or red.

Neutral solutions (pH = 7) maintain the extract's original color.

Basic solutions (pH > 7) turn the extract greenish or bluish. Sure, here's a concise procedure for pH testing using a pH meter: extract should change color depending on the pH of the solution.<sup>[12]</sup>

## Procedure for Testing of pH

### Calibration

Prepare and calibrate the pH meter using standard buffer solutions (pH 4.01, pH 7.00, pH 10.01).

### Sample Preparation

Prepare the sample solution to be tested and ensure it's at room temperature.

### Testing

Rinse and blot the pH meter electrode, then immerse it in the sample solution.

### Stabilization

Wait for the pH reading on the meter to stabilize.<sup>13</sup>

### Recording

Record the stabilized pH reading displayed on the meter.

### Cleaning

Rinse the pH meter electrode with distilled water after use and store it properly.

### Storage

Store the rose petal extract in a sealed container, preferably in a cool, dark place to maintain its effectiveness.<sup>14</sup> For longer shelf life, refrigeration is recommended.

By following these steps, you can prepare a natural pH indicator using rose petals and test its effectiveness with various solutions.<sup>15</sup>

## Procedure

### Preparing the Beetroot Indicator

Cut the Beetroot

Peel and chop the beetroot into small pieces.

### Boil the Beetroot

Place the chopped beetroot into a saucepan and add enough water to cover the pieces.

Bring the water to a boil and let it simmer for about 10-15 minutes until the water turns a deep red color.<sup>16</sup>

### Strain the Solution

Remove the beetroot pieces by straining the solution through a sieve or strainer. Collect the deepred liquid in a container. This liquid is your beetroot indicator.

### Cool the Solution

Allow the beetroot indicator to cool to room temperature before using it.<sup>17</sup>

### Titration

Clean all glassware (burette, pipette, conical flask) thoroughly with distilled water.

Rinse the burette with the titrant solution and the pipette with the analyte solution to prevent contamination.<sup>18</sup>

### Filling the Burette

Clamp the burette vertically on a stand<sup>19</sup>

Fill the burette with the standard solution (titrant), ensuring no air bubbles are present in the nozzle.

Record the initial volume of the titrant in the burette.<sup>20</sup>

### Strong acid Vs Strong base (Hydrochloric acid and sodium hydroxide)

5 ml sodium hydroxide of was pipette out into a clean conical flask and 3 drops phenolphthalein indicator was added and titrated against Sodium hydroxide taken in the burette.<sup>21</sup> Appearance of permanent pale pink colour is the end point.<sup>22</sup>The titration must be repeated for concordant values. Same procedure can be followed with different indicators including methyl red, methyl orange and natural indicator obtained from beetroot.<sup>23</sup> The titration was carried in triplicate and standard deviation was calculated from the results. Same procedure was carried for strong acid – strong base (Hydrochloric acid- Sodium hydroxide), and Weak acid -weak base (Acetic acid – Ammonium hydroxide). Strong acid- weak base (Hydrochloric acid Ammonium hydroxide)<sup>24</sup>

### Measuring the Analyte

Use the pipette to measure a precise volume of the analyte solution.

Transfer the measured analyte into the conical flask<sup>25</sup>.

Add a few drops of the indicator to the analyte solution if an indicator is required for your titration.

Sr No	Indicators	Mean value	Colour change	pH range
1	PH	20.1333	Colorless to pink	6
2	MR	20.9666	Pink to yellow	7
3	MO	21.7666	Pink to yellow	7
4	Beetroot extract	24.1333	Pink to colorless	10

**Table No. 1: Titration 1 Strong Acid – Strong Base Titration**

### Weak acid Vs Strong base:(Acetic Acid & Sodium Hydroxide)

With reference to titration II, the volume of sodium hydroxide needed for the neutralization of acetic acid was recorded and calculated for standard deviation and standard error. Specific end point was found for each indicator used in the experiment.<sup>26</sup> The mixture of solution turns a colorless solution to pink when using phenolphthalein as indicator solution, pink color to yellow in color when using both methyl red, methyl orange turns orange to yellow, whereas pink color turns colorless solution when crude methanolic extract of beetroot was used as an acid base indicator. Each set of color change have its unique range of pH specific against the indicators used.<sup>27</sup>

The neutralization reaction found occurs in particular pH range like pH 6, 6, 6 and 10 against phenolphthalein, methyl red, methyl orange and beetroot extract respectively. The mean and standard deviation shows closeness to the accuracy of the results (table-2).

Sr No.	Indicators	Mean value	Colour change	pH range
1	PH	7.06667	Colorless to pink	6
2	MR	8.8	Pink to yellow	6
3	MO	4.3667	Pink to yellow	6
4	Beetroot extract	9.733	Pink to colorless	10

**Table No.2: Titration Weak Acid-Strong Base**

## RESULT

Beetroot can be used as a natural pH indicator because it contains pigments called betalains that change color depending on the acidity or alkalinity of a solution.

Sr No	Vol of acid (ml)	Burette reading		Volume of titrant(ml.) mean= $\frac{\sum x}{n}$
		Initial (ml)	Final(ml)	
1	10.0	0.0	9.5	9.5
2	10.0	0.0	9.4	
3	10.0	0.0	9.6	
4	10.0	0.0	9.5	
5	10.0	0.0	9.5	

**Table No.3: Titration of HCl against NH<sub>3</sub> using aqueous golden beet root extract indicator**

## DISCUSSION

The preparation of a pH indicator from rose petals effectively demonstrates the use of natural Pigments, particularly anthocyanins, as sensitive markers for pH changes. These pigments exhibit Clear and distinct color changes across different pH levels: red in acidic environments, purplish in Neutral conditions, and green or blue in alkaline settings. This confirms the viability of rose petals as a functional pH indicator. To enhance the efficacy of the rose petal indicator, several improvements could be made. Using a Mixture of water and alcohol for extraction could increase the yield of anthocyanins, resulting in More intense and reliable color changes. Standardizing the extraction protocol, including the Amount of petals, water, and alcohol used, could improve consistency. Exploring natural Preservatives could extend the shelf life of the extract while maintaining its eco-friendly Attributes. The preparation and use of an herbal rose indicator have significant educational value. It offers a Hands-on approach to learning about pH, natural product chemistry, and sustainable practices. This method can serve as an engaging introductory experiment in chemistry courses, helping Students grasp the concept of acid-base indicators and the role of natural compounds in science.

## CONCLUSION

In conclusion, natural beetroot indicators offer a compelling, eco-friendly alternative for pH measurement. While they may not completely replace synthetic indicators in all applications due to certain limitations in precision and stability, their benefits make them an excellent choice for many educational and practical uses. Future research could focus on enhancing the stability and range of beetroot indicators, further expanding their applicability. In acid-base titrations, indicator is used to show a sharp color changes at end point which are mostly organic dyes.

Due to environmental pollution, availability and cost, the search for natural acid-base indicator was started. In the present study the extract of *Beta vulgaris* was used to replace the synthetic indicators due to the disadvantage of less availability and high cost of synthetic dye. Extract of *Beta vulgaris* gives sharp and intense color changes as compared to synthetic indicator. The extracts were evaluated by using strong acid-strong base, strong acid-weak base, weak acid-strong base and weak acid-weak base.

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