

BLOOD LEAF (*Iresine herbstii*) EXTRACT USED AS AN INDICATOR OF SOIL pH

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Abstract- This study had investigated on the possibility of blood leaf (*Iresine herbstii*) as a soil pH indicator. An aqueous solution was prepared and settled to remove impurities. The pH is 4.6. The solution was tested in a 0-14 pH solutions to check on the color changes in the scale. The soil pH was tested using pH paper and pH meter. The soil pH was also tested using the blood leaf extract and the tests showed the same result. In conclusion, the aqueous extract of blood leaf can be used as soil pH indicator. This can benefit gardeners and flower growers because they can use it as fresh extract or can be kept even for many weeks in closed container.

Index Terms- plant pigment, anthocyanin, pigment, blood leaf (*Iresine herbstii*), pH, soil

I. INTRODUCTION

Plants show characteristic colors due to the presence of strong pigments. Pigment may include include quercetin, from yellow onion skins, and anthocyanins, from red cabbage and blueberries. Anthocyanin comes from Latin; the Latin *anthos* is flower and *cyan* is blue. This family of molecules is responsible for the color found in radishes, eggplant, many different flowers, and even the varied colors we see in autumn leaves. Until the 19th century, people relied on various plants, animals, and minerals as sources of dyes for fabrics. Many natural plant pigments also have acid-base properties, and will change colors when exposed to different pH solutions. Anthocyanin pigments generally show this behavior. In several attempts, the red cabbage, sugar beets and other vegetables had been used as pH indicator in class activities and showed positive results. The blood leaf (*Iresine herbstii*) is usually used as a landscape ornamental plant which can easily be grown in the tropics. Its availability all year round makes it a very accessible plant for testing pH. The color looks like the red cabbage so anthocyanin seem to be present in this plant.

Anthocyanins are members of a class of nearly universal, water-soluble, terrestrial plant pigments that can be classified chemically as both flavonoid and phenolic. They are found in most land plants, with the exception of the cacti and the group containing the beet. They contribute colors to flowers and other plant parts ranging from shades of red through crimson and blue to purple, including yellow and colorless. These pigment apparently play a major role in two very different plant processes: for one, attracting insects for the purpose of pollination. Advantage is made of the fact that the pigments absorb strongly in the UV (ultraviolet), visually attracting insects but with light wavelengths that are invisible to humans. These

pigments play a major role in plant pollination - and in predation in carnivorous plants, attracting insects into the trap apparatus. Anthocyanins play a very versatile role in pollination, especially in the *Bromeliaceae*. Certain bromeliads turn a vivid red just before and during pollination but soon revert to the original green color characteristic of the photosynthesis pigment, chlorophyll.

Anthocyanins are not a biochemical dead end but rather a dynamic signalling device that can be switched on when needed by the plant to assist in pollination. They are then degraded by plant enzymes when no longer needed to attract pollinators to flowers.

In their second major role, anthocyanin-related pigments serve as a UV screen and are produced in response to exposure of the plant to UV radiation, protecting the plant's DNA from damage by sunlight. (UV causes the paired strands of genetic material in the DNA double helix to become cross-linked, preventing cell division and other vital cellular processes like protein production.

And in a third, and no less significant role, anthocyanins serve as anti-feedents, their disagreeable taste serving to deter predatory animals. In a related defense mechanism, anthocyanin production can be induced by ionizing radiation, which can damage DNA as readily as UV can. Chemical messengers apparently signal the damage to DNA and induce anthocyanin production in these plants.

The biosynthesis of this class of pigment is accomplished by a series of enzymes that are bound to cell membranes and that help convert two central biochemical building blocks derived from photosynthesis (acetic acid and the amino acid phenylalanine) found in the cell's cytoplasm through a series of discrete chemical steps into the final pigments, which are then excreted on the other side of the membrane into vacuoles in the epidermal cell layer. Significant genetic change in the DNA coding for the production of these enzymes results in loss of pigment production.

Anthocyanin pigments can be produced by growing plant cells in tissue culture. Plants having no pigmentation themselves in cultivation were subsequently demonstrated to produce anthocyanin in tissue culture.

Environmental factors affecting anthocyanin production included light intensity and wavelength, with blue and UV being most effective, temperature, water and carbohydrate levels, and the concentrations of the elements nitrogen, phosphorous and boron in the growth medium. Anthocyanin production can be induced by light, blue being the most effective color. Low light levels also induce the formation of different flavonoid pigments, which is another interesting adaptive response on the part of plants. (Tillandsias), for example, develop a bright red coloration due to induced anthocyanin production if grown in strong light.

For some additional observations on possible alternate roles for anthocyanin in *Tillandsia*, (see noted bromeliaexpert David Benzing's personal observations as quoted in Paul T. Isley III's excellent book *Tillandsia*.)

Since the anthocyanin pigments are regularly made by plants like the blood leaf, it is therefore very convenient to tap its potential as pH indicators.

Modern methods of determining pH are available nowadays but these are only methods are only available in sophisticated laboratories in universities and science centers. In the Philippines, most florists and subsistence gardeners do not have the access to these protocols. This research gives an accessible method to these gardeners so that they can test their soil pH using this indigenous method using an ornamental plant that is readily available instead of using chemicals which can be toxic. Hence this study.

II. MATERIALS AND METHOD

About a hundred gram of blood leaf was collected and put in a blender and around 10 mL distilled water were mixed and blended until the a thick extract was produced. The extract was filtered and kept in a tightly closed bottle. Using a pH meter the pH was tested and it was recorded a 4.6. The impurities settled and it was separated by decantation.

A set of solution from 0-14 pH was prepared and stored in closed container. The extract was tested to show the color changes. Distinct color changes appeared. The color changes of the extract is shown below in Fig 1. . Based on this color changes, four soil samples were tested using the blood leaf extract. The pH meter and pH paper records were used to verify the data observed using the extract as indicator.

The four soil samples were tested using pH meter and pH paper. The ash from burnt organic matter and lime was also tested to show the changes in the higher pH levels.

III. DATA ANALYSIS AND INTERPRETATION

A. Color change of the blood leaf extract along the pH scale

The blood leaf extract was subjected to the solutions pH 0-14. The color changes were very distinct which shows that it can be a pH indicator. The color changes were maintained even after an hour which is not true to some commercial indicators. Table 1 shows the color changes of the extract in the pH solutions 0-14. The real color changes that were observe is shown in Plate 2. The color changes had been observable even after one hour which is not true to some commercial pH indicator.

Table 1 shows the color changes of the extract in the pH solutions 0-14. The real color changes that were observed are shown in Plate 2.

Table 1. Color Changes when Blood leaf extract were subjected to the pH solutions.

pH	Color change
0	Dark green
1	Light green

2	Very light pink
3	Light pink
4	pink
5	Dark pink
6	Darker pink
7	Darkest pink
8	Darker pink
9	pink
10	Very light pink
11	indigo
12	green
13	Light green
14	Yellow green

B. pH test of the Soil Samples using blood leaf Extract

The pictures below showed a comparison of the blood leaf extract scale with the different soil samples.

Picture 1 is a comparison of the pH4 blood leaf extract and pH5. The clay soil showed a reading reading of of 4.5 which coincides with the result using the blood leaf extract indicator.

Picture 2 showed the comparison of color change in the soil from Balenben which is between pH 5 -6. The reading coincides which the pH meter reading which is 6.5.

The soil from the strawberry farm showed a darer pink color as shown in picture 3. When compared with the blood leaf extract indicator, it showed a pinkish color between pH 6-7 which coincides with the pH meter reading which is 6.3.

Picture 4 below showed a change to color green when tested with blood leaf extract. The pH meter and pH paper showed a pH 12 which is the same as the extract reading.



pH 5- (clay soil=pH 4.5) - pH4

Picture 1

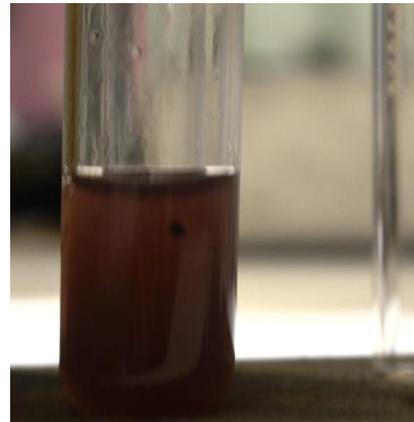


pH6-(b. soil pH5.5) – pH 5

Picture 2



**pH6 (St soil =6.5) pH 7
picture 3**



**pH 11 Ash tested with blood leaf extract
Picture 5**



**Lime water tested with blood leaf extract
Picture 4**

Soil pH Test

The table shows the result of the pH test. There are five (5) samples, 3 soil samples, lime and ash that were tested using the pH meter and pH paper and the blood leaf extract. The table shows that three tests showed similar results. Among the five soil samples, the clay soil is the most acidic which showed a pH of 4.3 in the pH meter reading between 4 to 5 pH in the pH paper reading and showed light pink color in the blood extract pH test. The soil from a vacant lot has a pH of 5.6 using the pH meter and similar result was observed using the blood leaf showing a pink color which is darker than the other samples which are acidic. The darker pink coloration has a higher pH.

This shows the soil samples were in the range of 4-7. This means that the soils were acidic. The ash showed a reading at pH 11 which showed a color change of indigo and the lime solution showed a greenish color. Thus the two substances were basic.

Table 2. pH Readings of the Soil samples using blood leaf extract As compared to pH paper and pH meter readings.

Indicators	Clay Soil La Trinidad , Benguet	Soil From Vacant Lot with plants Balenzen Baguio City	Soil From Strawberry Farm, la Trinidad, Benguet	ash	lime
pH METER	4.3	5.6	6.3	11	12
pH PAPER	4 to 5	5 to 6	6 to 7	11	12
plant extract from Blood leaf	4-5 pink color	5- 6 pink	6-7 pink	11 indigo	12 green

IV. CONCLUSION AND RECOMMENDATION

Based on the results of this study, it is therefore noted that the blood leaf extract can be a pH indicator for soil considering its easy preparation and acquisition of the plant material. Since it is an ornamental plant, the material is readily available. Small scale gardeners and florists can do it even extract it using mortar and pestle. The changes are easy to observe the changes in color do not disappear immediately. Further studies can be done by analyzing the type of anthocyanin present in blood leaf.

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REFERENCES

- [1] Abbas, S.K. (2012) Study of acid-base indicator property of flowers of Ipomoea biloba. International Current Pharmaceutical Journal 2012, 1(12): 420-42

- [2] ArifKhan, P.M. and M. Farooqi (2011) Analytical Applications of Plant Extracts as Natural Indicator.
- [3] Bondre,S. et. Al. (2012) Study on Isolation and Propagation of Anthocyanin and its application as pH indicator.
- [4] Journal of Advanced biotechnology and Research ISSN 0976-2612 Online ISSN 2078-5995X, Vol.3 Issue 3 2012
- [5] Chalker- Scott(1999). Environmental Significance of Anthocyanins in Plant Stress Responses. Phytochemistry and Photobiology 70(1):1-9
- [6] Flavonoids: Chemistry, Biochemistry and Applications (E book) edited by Oyvind M. Andersen, Kenneth R. Markham
- [7] Izonfou, W. A et al.(2006) Acid-base indicator properties of dyes from local plants I: Dyes from Basella alba(Indian spinach) and Hibiscus sabdariffa (Zobo) J. Appl. Sci. Environ. Mgt. Vol. 10 (1) 5 – 8
- [8] Pillai, M.M.(2012) Study on Isolation and Purification of Anthocyanins and its Application of pH Indicator

Internet Sources

- [1] <http://ph/anthocyanin.html> retrieved July 12, 2014.
- [2] <http://www.charlies-web.com/specialtopics/anthocyanin.html> (retrieved July 17, 2014
- [3] <http://chem.answers.com/compounds/natural-ph-indicators>(retrieved July 5, 2014)

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Appendix

Plate 1. Blood Leaf



Plate 2. pH readings of the soil samples using pH paper



Plate 3. color changes of the blood leaf extract in the pH solutions 0-14



| pH |
|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |