

## **Original Research Article**

### **Efficacy of four cultivars of banana bract extract as PH indicator**

**Gina M. Zamora**

Cagayan State University, Tuguegarao, Cagayan, Philippines

#### **\*Corresponding author**

Gina M. Zamora

Email: [gmz0530@yahoo.com](mailto:gmz0530@yahoo.com)

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**Abstract:** This study aimed to prepare paper strips from four common cultivars of banana (*Musa* spp. namely: lakatan, latundan, sabapahaba and sabapabilog) bract extract as pH indicator using three methods of extraction namely; Manual, Soxhlet and Rotary Evaporator to act as potential alternative pH indicator. Specifically, it sought to answer the following questions: (1) Which among the banana bract cultivar extract paper strips and methods of extraction show the best color reaction as pH indicator; (2) What is the stability of the four banana bract cultivar extracts and methods of extraction as pH indicator in terms of: a. Shelf-life at room temperature and, b. Shelf-life at refrigeration temperature; (3) How does the banana bract cultivar extract paper strips compare with pH meter and commercial pH strips (Multistix) in terms of determining the pH of urine samples? And; (4) is there a significant difference between the banana bract cultivar extract paper strips and pH meter or commercial pH strips (Multistix) in terms of determining the pH of urine samples? The study consisted of three phases: Phase 1. Preparation and Determination of Banana Bract Extract Paper Strip as pH Indicator; Phase 2. Stability of the Banana Bract Extract Paper Strip pH Indicator at Room and Refrigeration Temperature. And Phase 3. Comparison of the Banana Bract Extract Paper Strip as Urine pH Indicator with pH Meter and Commercial Urine Strips (Multistix). Result of the study showed that Lacatan (Barangan) bract extract paper strips processed by Rotary Evaporator method and stored at room temperature gave the best color reaction as pH indicator. Furthermore, it was comparable with pH meter and commercial urine pH strips (Multistix) in determining the pH of urine samples.

**Keywords:** Acidity, Banana bract, Basicity, Cultivars, Efficacy, pH indicator, Stability, Anthocyanin.

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#### **INTRODUCTION**

The demand for measuring pH has grown dramatically in recent years. This includes environmental, agricultural, waste water, pharmaceutical and educational applications, to name just a few. In the laboratory, monitoring the pH level of urine, blood, sweat, and other body fluids are clinically important in the diagnosis of acidosis in the body metabolism. The urine pH will reflect a number of metabolic, digestive, and other processes. Routine chemical examination of the urine is very important and has changed dramatically since the early days of urine testing, owing to the development of the reagent strip method for chemical analysis [1].

However, considering the significant benefits of commercialized pH indicators in measuring all acids and bases level of solution, particularly that of urine, disadvantages arise due to high value it imposed in the market. The devastating impact imposed on health and environment brought by the use of chemical pH indicators and limited known sources of raw materials, gave the idea that alternative pH indicators naturally present in some plants particularly banana bract (*Musa* spp.) should be utilized. This will eliminate the

hazardous effects and minimize the cost of chemical pH indicators for urine analysis.

According to scientific study conducted by Castillo *et al.*; [2], the extracts coming from banana bract show different colors when added to solutions of different pH concentration. The feasibility of banana bract to act as pH indicator is due to the anthocyanin pigment which is responsible for the blue, red, and purple color of the plant particularly the first covering of the bract. This plant is found richly growing in tropics and easily propagated that is why accessibility and availability is not a problem.

This study therefore focused on the formulation of pH paper strips using the active components of banana bract extract as urine pH indicator. The effectiveness of the pH paper strips with its accuracy as an indicator had been evaluated.

#### **MATERIALS AND METHODS**

Four Banana bract cultivars: Lacatan (Barangan), Latundan (Manila), Saba (Damilig-Lolloy) and Saba (Damilig-Bato) were gathered and bought from the banana plantation in Rizal, Cagayan. The identification of the different banana cultivars were authenticated at

the Department of Agriculture Regional Field Office Region 2. The reagents utilized such as 95 % ethanol, Sodium phosphate, Phosphoric acid and distilled water for the extraction procedure and preparation of solutions with known pH were purchased. The Soxhlet apparatus and Rotary evaporator were used for the extraction procedures.

The varying solutions of known pH were prepared and the pH level was validated using pH meter. The pH level of the urine samples collected were also measured using the pH meter and commercial urine pH strips (Multistix). Other laboratory apparatuses such as Analytical balance, beakers, stirring rod, filter paper, buchner funnel, graduated cylinder, vials, reagent bottles, thermometer and petri dishes were used in the study. Fifteen (15) randomly collected urine samples were collected and analyzed in the study.

The study consisted of three phases: Phase 1. Preparation and Determination of Banana Bract Extract Paper Strip as pH Indicator. Phase 2. Stability of the Banana Bract Extract Paper Strip pH Indicator at Room and Refrigeration Temperature. Phase 3. Comparison of the Banana Bract Extract Paper Strip as Urine pH Indicator with pH Meter and Commercial Urine Strips (Multistix).

### Research Design

The research utilized a qualitative experimental method of research in the conduct of phase 1 and phase 2 of the study. The color reaction of the pH paper strips processed from the four cultivar extracts were established using solutions with known pH ranging from pH 1 to pH 14. A color chart served as the major output of each color reaction. The stability of the four banana bract cultivar extracts paper strips were tested in terms of its shelf-life at room and refrigeration temperature.

Phase 3 of the study on the comparison of banana bract extract paper strip with commercial urine pH strip (Multistix) and pH meter utilized t-test comparison of means to compare the significant differences between the treatments.

### Banana Bract Extraction

The first three (3) layers of the four Banana bract cultivars were washed and air dried. Three different extraction methods were utilized as follows:

#### 1. Manual Method

Each banana bract cultivar was cut into very fine pieces using scissors. The finely cut banana bract were placed in sterile gauze and the extract was collected by squeezing the flesh by hand. The sample extracted was filtered to remove the residue of banana bract layers and was collected in a petri dish. The same procedure was followed for all banana bract cultivars.

#### 2. Soxhlet Method

The banana bract cultivars were cut into small pieces. Two hundred (200) grams of cut banana bract was submerged in 400 ml 95% ethanol. The material was soaked in the solvent (95% ethanol) for 48 hours and was filtered through Buchner funnel to separate the solid material (cut pieces of banana bract) from the liquid containing the dissolved chemical. The filtrate was poured into the Soxhlet separator and the procedure was allowed to come to completion until the desired compound was concentrated in the distillation flask. The concentrated compound was transferred to a petri dish. The same procedure was followed for all banana cultivars.

#### 3. Rotary Evaporator Method

The banana bract cultivars were cut into small pieces. Two hundred (200) grams of cut banana bract was submerged in 400 ml 95% ethanol. The material was soaked in the solvent (95% ethanol) for 48 hours and was filtered through Buchner funnel to separate the solid material (cut pieces of banana bract) from the liquid containing the dissolved chemical. The filtrate was poured into the Rotary Evaporator and the procedure was allowed to come to completion until the desired compound was concentrated in the distillation flask. The concentrated compound was transferred to a petri dish. The same procedure was followed for all banana cultivars.

### Preparation of Banana Bract Extract Paper Strip

The filter paper was cut into litmus paper strip size and the paper strips were soaked into the different banana bract cultivar extracts from the three extraction methods for 24 hours to allow absorption of the extract. Then, the filter paper strips were air dried for 6 hours. They were kept in two separate clean bottles, each labeled with the name of the banana bract cultivar and the method of extraction. One bottle was stored at room temperature and the other bottle at refrigeration temperature.

### Preparation of Buffer Solutions

Buffer solutions were prepared to establish the color reactions of the different banana bract cultivar paper strips to solutions of varying pH. Buffer solutions with pH ranging from pH 1 to pH 14 using Sodium phosphate/ Phosphoric acid buffer were prepared following this equation:  $pH = pka + \log \frac{[A^-]}{[HA]}$ .

For Buffer solutions with pH 1 to pH 3 the pka value used is 2.12; For Buffer solutions with pH 4 to pH 8 the pka value used are 7.12. For Buffer solutions with pH 9 to pH 14 the pka value used is 12.82.

To prepare 50 ml of the buffer solutions with different pH levels the following measurements were followed: pH 1 = 0.524 gm Sodium phosphate + 50ml

1M Phosphoric acid; pH 2 = 5.24 gm Sodium phosphate + 50 ml 1M Phosphoric acid; pH 3 = 53.34 gm Sodium phosphate + 50ml 1M Phosphoric acid; pH 4 = 0.004 gm Sodium phosphate + 50ml 1M Phosphoric acid; pH 5 = 0.054 gm Sodium phosphate + 50ml 1M Phosphoric acid. pH 6 = 0.540 gm Sodium phosphate + 50ml 1M Phosphoric acid; pH 7 = 5.389 gm Sodium phosphate + 50 ml 1M Phosphoric acid; pH 8 = 53.8619 gm Sodium phosphate + 50ml 1M Phosphoric acid; pH9 = 0.001 gm Sodium phosphate + 50ml 1M Phosphoric acid; pH 10 = 0.012 gm Sodium phosphate + 50ml 1M Phosphoric acid. pH 11 = 0.124 gm Sodium phosphate + 50ml 1M Phosphoric acid; pH 12 = 1.238 gm Sodium phosphate + 50ml 1M Phosphoric acid; pH 13 = 12.415 gm Sodium phosphate + 50ml 1M Phosphoric acid. pH 14 = 124.115 gm Sodium phosphate + 50ml 1M Phosphoric acid. Validate pH level using pH meter.

#### **Establishment of the Color Chart for Each Cultivar and Method of Extraction**

This step determines the change in color of the different banana bract extract paper strips in the different buffer solutions of known pH and establishing the color chart for each cultivar and method of extraction.

Banana bract extract paper strips from each banana bract cultivar were dipped into the different buffer solutions of known pH to determine the change in color for every pH level in order to establish a color chart for the four (4) banana bract cultivars and three methods of extraction.

The prepared banana bract extract paper strips were dipped into the buffer solutions of known pH for five (5) seconds. The color reaction was observed and recorded using the reference color chart (Appendix A) in the different pH levels after one (1) minute; disregarding the color change after one hour reaction time.

#### **Phase II. Determination of the Stability of the Banana Bract Extract Paper Strip as pH indicator**

Determination of the Shelf-Life of the Banana Bract Extract Paper Strips from the four cultivars and different methods of extraction at Room temperature and Refrigeration temperature

##### **A. Room Temperature**

The banana bract extract paper strips were kept in clean bottles and stored at room temperature (25°C - 30°C). It was tested for its color reaction in three (3) replicates with the different buffer solutions of known pH once (1) a week using the established color chart as a reference.

##### **B. Refrigeration Temperature**

The banana bract extract paper strips were kept in clean bottles and stored at refrigeration temperature (0°C - 8°C). It was tested for its color reaction in three

(3) replicates with the different buffer solutions of known pH once (1) a week using the established color chart as a reference.

#### **Comparison of the Banana Bract Extract Paper Strip as Urine pH Indicator with pH Meter and Commercial Urine Strips**

Fifteen (15) randomly collected midstream catch urine samples were tested and measured for pH level using the pH meter, commercial urine strips (Multistix) and banana bract extract paper strips.

#### **Data Analysis**

The t-test was computed using Microsoft excel on data analysis ToolPak was utilized to determine if the prepared banana bract paper strips were comparable with pH Meter and Commercial Urine pH Strips (Multistix) in determining pH level of urine samples.

## **RESULTS AND DISCUSSION**

### **Establishment of Color Chart for the Four Banana Cultivars**

#### **LACATAN CULTIVAR**

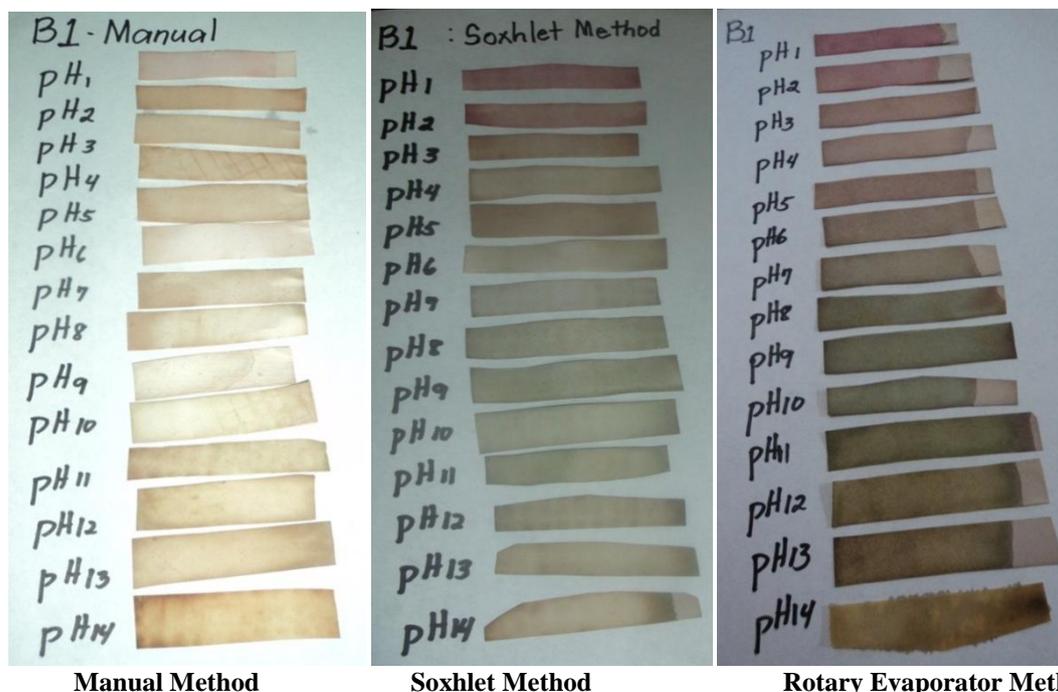
In the manual method of extraction, the original color of the prepared extract paper strip is Blanched almond. At pH 1 the paper strip turned to Light Pink. At pH 2 and pH 10 the banana bract paper strip did not change in color. pH 3, pH 6, pH 9, and pH 11 have the same color reaction of Navajo white; furthermore, pH 4, pH 5, pH 12 and pH 13 have the same color reaction of Mocassin; while pH 7 and pH 8 have the same color reaction of Bisque at pH 14 the color changed to Burly Wood. The result shows that Lacatan (Barangan) Bract Paper Strips extracted manually is not a good pH indicator because of many pH levels showing the same color reaction (Figure 1).

Lacatan Bract Extract Paper Strips Prepared by Soxhlet method is Antique White in color. It changed to Pale Violet Red at pH 1, Light Pink at pH 2, and Rosy Brown color at pH3. At pH 4 no color change was noted. At pH 5 it changed to Burly Wood and Bisque at pH 6. At pH 7 the color reaction is Dim Gray. The same color reaction is seen at pH 8 and pH 9 to Cadet blue; pH 10 and 11 to Olive Drab; pH12 and pH 13 to Dark Olive green; and Olive at pH 14. The different colors exhibited in acid pH (pH 1-6) indicate that Lacatan (Barangan) extracted by soxhlet method maybe a good acid pH indicator. This is supported by the study conducted by Castillo *et.al.*; [2] that anthocyanin is a better acid indicator. The change in color at extreme pH levels (acid and basic) is suggestive that it may be used as a qualitative pH indicator (Figure 1).

Lacatan Bract Extract Paper Strips Prepared by Rotary Evaporator Method is Antique White in color. It changed to Pale Violet Red, Light Pink, and Rosy Brown color at pH 1, pH 2, and pH 3 respectively. At pH 4 no color change was noted. At pH 5 it changed to Burly Wood and Bisque at pH 6. At pH 7 the color

reaction is Dim Gray the same color reaction is seen in pH 8 and pH 9 to Cadet blue; pH 10 to Medium Turquoise; pH 11 to Olive Drab; pH12 and pH 13 to Dark olive Green and pH 14 to Olive (Figure 1). The different colors exhibited in acidic pH (pH 1-6) indicates that Lacatan (Barangan) extracted by Rotary evaporator method may be a good acid pH indicator. This is supported by the study conducted by Castillo *et.al.*; [2] that anthocyanin is a better acid indicator. The

change in color at extreme pH is suggestive that it may be used as a qualitative pH indicator. Almost the same color reaction was observed with the color reaction extracted by soxhlet method. The only difference is the color reaction at pH 10 (Medium Turquoise) giving a different color reaction at pH 11 (Olive Drab) as compared with that extracted by Soxhlet method. This makes Lacatan (Barangan) Bract Paper Strips extracted by Rotary Evaporator a better pH indicator.



**Fig-1: Color reaction of Lacatan (Barangan) Bract Extract Paper Strips processed by the three (3) methods with solutions of varying pH.**

The best color chart for Lacatan cultivar was noted with the Lacatan Extract Paper Strips processed by Rotary Evaporator Method. This is evidenced by the variation of colors produced in the different pH solutions.

**LATUNDAN CULTIVAR**

The Latundan Bract Extract Paper Strip prepared by Manual method is Papaya Whip in color. At pH 1 the paper strip changed its color to peach puff. While at pH 2 the banana bract extract paper strip did not change in color. At pH 3, pH 12 and pH 13 it has the same color reaction of Mocassin; at pH 4 it turned to Light Golden Rod Yellow; at pH 5, pH 6, pH 7, pH 9 and pH 11 it has the same color reaction of Lemon Chiffon. At pH 8 it turned to Corn silk; Wheat at pH 10 and at pH 14 to Light Green (Figure 2). The color reaction shows that Latundan (Manila) Bract Paper strips prepared by manual method are not a good pH indicator because it gives the same color reaction at different pH levels.

The Latundan Extract paper strip prepared by Soxhlet method is Blanched Almond in color. At pH 1

the color changed to light pink, peach puff at pH 2, Mocassin at pH 3, Papaya whip at pH 4. There is no change in color at pH 5 and pH 6. At pH 7 the color changed to Corn silk, at pH 8 to Green yellow. The same color reaction was noted at pH 9 and 10 to Lime green, at pH 11 it changed color to Olive drab, at pH 12 and pH 13 to Dark olive green and at pH 14 to Olive (Figure 2). The color chart indicates that Latundan (Manila) Bract Extract paper Strips prepared by soxhlet method is a good pH indicator in lower pH as evidenced by the different colors exhibited in pH 1 to pH 4. This is supported by the study conducted by Castillo *et.al.*; [2] that anthocyanin is a better acid indicator.

The Latundan Bract Extract Paper Strips prepared by Rotary Evaporator Method is Blanched Almond in color. At pH 1 it changed color to light pink; pH 2 to peach puff; pH 3 to Mocassin and pH 4 to papaya whip. There is no change in color at pH 5 and pH 6. At pH 7 it changed to Corn silk in color and to Green yellow at pH 8. The same color reaction was noted at pH 9 and pH 10 to Lime green; at pH 11 it changed color to Olive Drab; pH 12 and pH 13 to Dark

olive green and Olive at pH 14. The color chart indicates that Latundan (Manila) Bract Extract Paper Strips extracted by Rotary evaporator method is a good pH indicator in lower pH as evidenced by the different

colors exhibited in pH 1 to pH 4. This is supported by the study conducted by Castillo *et.al.*; [2] that anthocyanin is a better acid indicator.

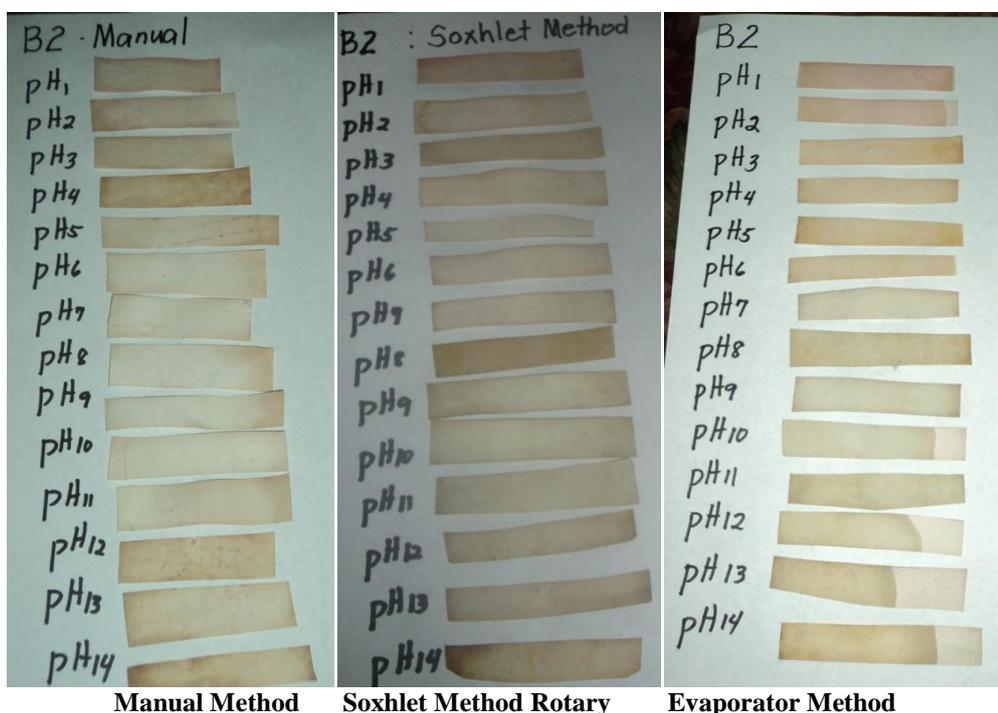


Fig-2: Color reaction of Latundan (Manila) Bract Extract Paper Strips processed by the three (3) methods with solutions of varying pH.

The best color chart for Latundancultivar was noted with the Latundan Extract Paper Strips processed by Soxhlet and Rotary Evaporator Methods.

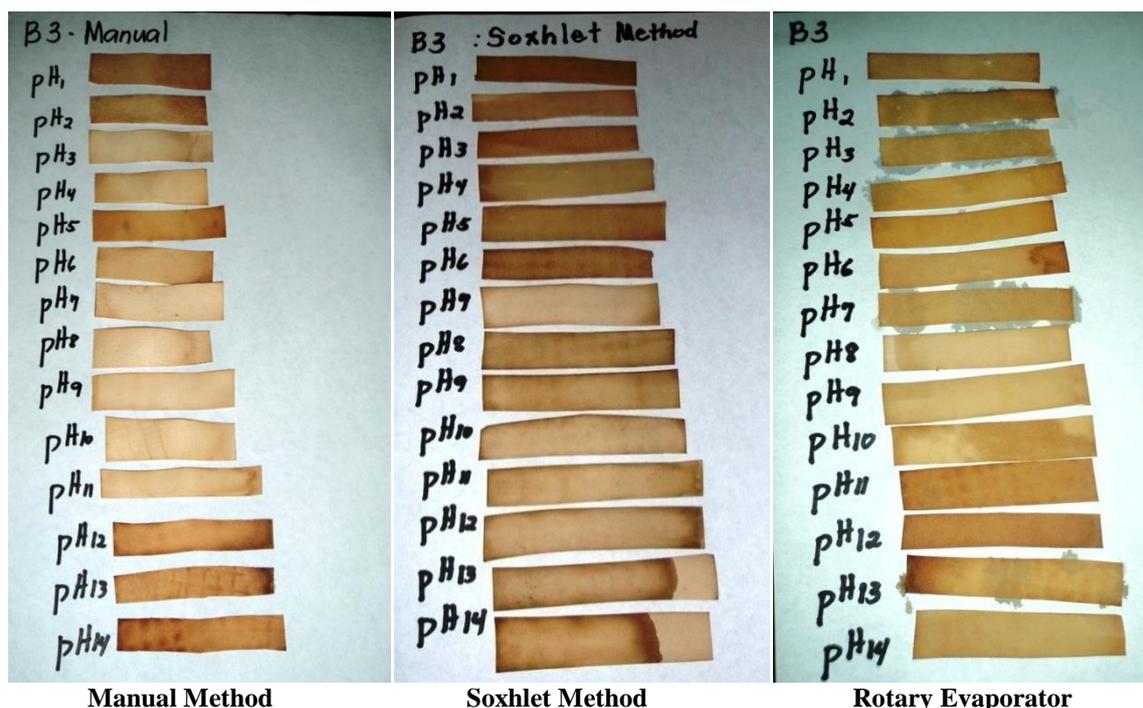
**SABA – LOLLOY CULTIVAR**

The Saba Lolloy Bract Extract Paper Strips prepared by Manual method is Navajo White in color. There is only one color reaction of the paper strips from the original color Navajo white to Burly Wood in this method of preparation. The pH at which the Saba Lolloy Bract Extract Paper Strips changed colors were pH 1, pH 2, pH 5, pH 6, pH 13 and pH 14 to Burly wood while pH 3, pH 4, pH 7, pH 8, pH 9, pH 10, pH 11, and pH 12 has no color change noted (Figure 3). This is a clear indication that Saba Lolloy Bract Extract Paper strips prepared by manual method are not a good pH indicator.

The Saba Lolloy Bract Extract Paper Strips prepared by Soxhlet method is Navajo White in color. At pH 1, pH 2, pH 3, pH 4, pH 5 and pH 6 the color

reaction is Peru. At pH 7 the color changed to Burly Wood. The same color reaction of Dark Golden Rod is seen at pH 8, pH 9, pH 10, pH 13, and pH 14 while at pH 11 and pH 12 the color changed to Saddle Brown (Figure 3). Saba Lolloy Bract Extract Paper Strips extracted by Soxhlet method is not a good pH indicator as evidenced by the same color reactions noted at different pH levels.

The Saba Lolloy Bract Extract Paper Strips prepared by Rotary Evaporator method is Navajo White in color. The color reaction at pH 1 to pH 6 is Peru. At pH 7 the color reaction is Burly Wood. Dark Golden Rod color reaction is seen at pH 8, pH 9, pH 10, pH 13 and pH 14. At pH 11 and pH 12 the color reaction is Saddle Brown (Figure 3). The color chart shows that Saba Lolloy Bract Extract Paper Strips prepared by Rotary Evaporator method is not a good pH indicator as evidenced by the same color reactions noted at different pH levels.



**Fig-3: Color reaction of Saba Lollo Bract Extract Paper Strips processed by the three (3) methods with solutions of varying pH.**

The best color chart for Saba- Lollo cultivar was noted with the Saba – Lollo Extract Paper Strips processed by Soxhlet and Rotary Evaporator Method.

**SABA – BATO CULTIVAR**

The Saba Bato Bract Extract Paper Strips prepared by Manual method is Navajo White in color. There is no change in color of the strips at pH 1 and pH 6. The same color reaction of Burly Wood is seen at pH 2, pH 3, pH 8, pH 10 and pH 11. At pH 4 the color changed to Wheat; Tan at pH 5 and pH 7; Saddle brown at pH 9, pH 12 and pH 13. At pH 14 the color changed to Peru (Figure 4). The color reaction is a clear indication that Saba Bato Extract Paper Strips prepared by manual method is not a good pH indicator as evidenced by the same color reactions at different pH levels.

The Saba Bato Bract Extract Paper Strips prepared by Soxhlet method is Navajo White in color.

Same color reaction were seen at pH 1 to pH 6 to Peru. At pH 7 the color changed to Burly Wood. Same color reaction were seen at pH 8, pH 9, pH 10, pH 11 and pH 14 to Dark Golden Rod.; pH 12 and pH 13 to Saddle brown (Figure 4). The color chart indicates that Saba Bato Extract Paper Strips prepared by Soxhlet method is not a good pH indicator as evidenced by the same color reaction noted at different pH levels.

The Saba Bato Bract Extract Paper Strips prepared by Rotary Evaporator method is Navajo White in color. Same color reaction was seen at pH 1 to pH 6 to Peru. At pH 7 the color changed to Burly Wood. Same color reaction were seen at pH 8, pH 9, pH 10, pH 11 and pH 14 to Dark Golden Rod.; pH 12 and pH 13 to Saddle brown (Figure 4). The color chart indicates that Saba Bato Extract Paper Strips prepared by Rotary Evaporator method is not a good pH indicator as evidenced by the same color reaction noted at different pH levels.

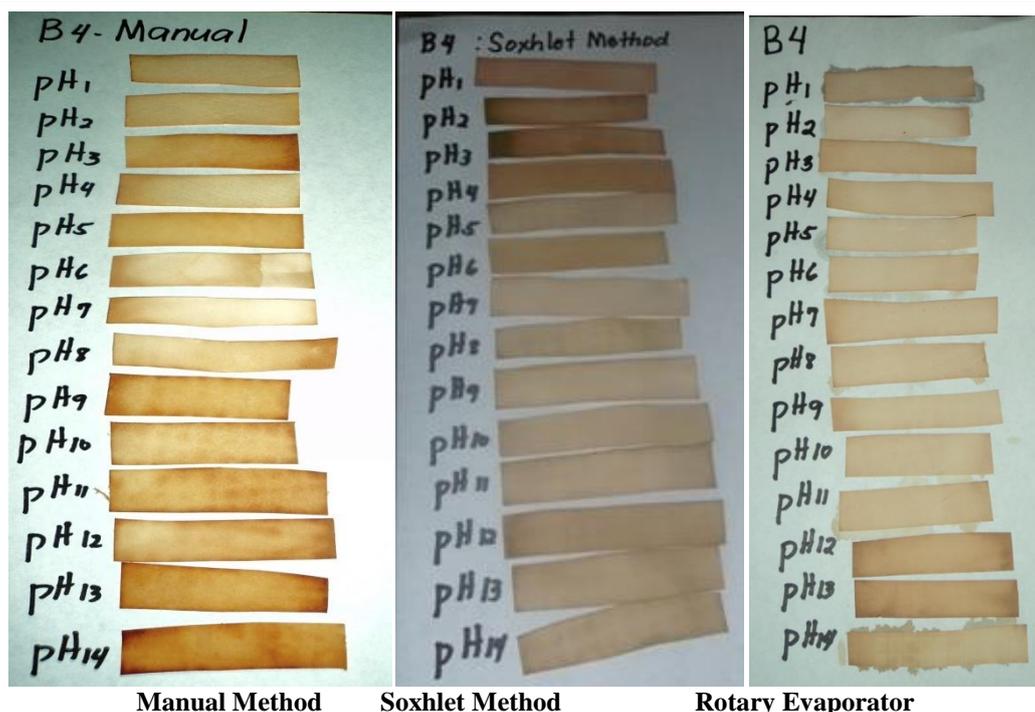


Fig-4: Color reaction of Saba Bato Bract Extract paper Strips processed by the three (3) methods with solutions of varying pH.

The best color chart for Saba- Batocultivar was noted with the Saba – Bato Extract Paper Strips processed by Soxhlet and Rotary Evaporator Methods.

The four cultivars of banana bract extract paper strips prepared by Manual method, Soxhlet method and Rotary Evaporator method showed color reactions when dipped with known pH solutions. However, Lacatan Banana Bract Extract Paper Strips showed the best color reaction as indicated by the variation of colors produced in the different pH

solutions on both Rotary Evaporator and Soxhlet Method. The two methods utilized ethanol as solvent. Pulmonol-Duran, *et.al.*; [3] and De Ross and Mercadante [4], proves that the use of ethanol or methanol in extracting anthocyanin on tropical fruits produces more amount of pigment. The amount of anthocyanin in each cultivar extract and the kind of anthocyanin present in each Banana cultivar may have contributed to the variation of color reactions in the different pH levels.

|                               |        |     |     |        |     |     |        |     |     |        |      |      |
|-------------------------------|--------|-----|-----|--------|-----|-----|--------|-----|-----|--------|------|------|
| B4<br>Saba Damilig-<br>Bato   | Red    | Red | Red | Red    | Red | Red | Blue   | Red | Red | Blue   | Blue | Blue |
| B3<br>Saba Damilig-<br>Lolloy | Red    | Red | Red | Red    | Red | Red | Blue   | Red | Red | Blue   | Blue | Blue |
| B2<br>Latundan                | Red    | Red | Red | Red    | Red | Red | Blue   | Red | Red | Blue   | Blue | Blue |
| B1<br>Lakatan                 | Red    | Red | Red | Red    | Red | Red | Blue   | Red | Red | Blue   | Blue | Blue |
|                               | M      | S   | R   | M      | S   | R   | M      | S   | R   | M      | S    | R    |
|                               | WEEK 1 |     |     | WEEK 2 |     |     | WEEK 3 |     |     | WEEK 4 |      |      |

Fig-5: Determination of the Stability of Banana Bract Paper Strip

As shown in Figure 5, Banana bract extract paper strip from Banana Bract 1 (Lakatan) extracted by manual method was stable until week 2 while those extracted by Soxhlet and Rotary evaporator method were stable until week 3. Banana bract extract paper strip from Banana bract 2 (Latundan) extracted by manual method was stable until week 2 while those extracted by soxhlet and rotary evaporator method were

stable until week 3. Banana bract extract paper strip from Banana bract 3 (Saba Damilig - Lolloy) extracted by manual method was stable until week 2 while those extracted by soxhlet and rotary evaporator method were stable until week 3. Banana bract extract paper strip from Banana bract 4 (Damilig -Bato) extracted by manual method was stable until week 2 while those extracted by soxhlet and rotary evaporator method were

stable until week 3. The graph indicates that the four banana bract cultivar extract paper strips has a long shelf-life at room temperature using Soxhlet and Rotary evaporator method of extraction.

Determination of the Stability of the Banana Bract Paper Strip stored at refrigeration temperature is shown in figure 6.

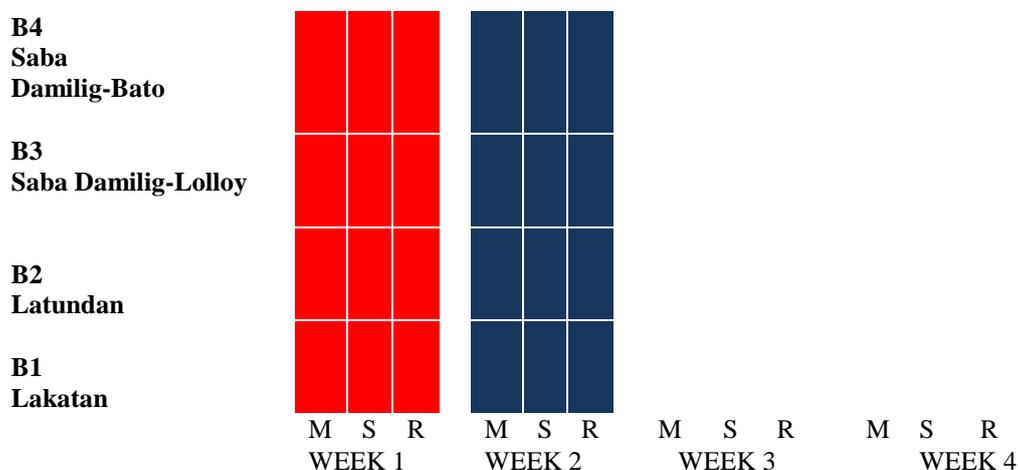


Fig-6: Stability of Banana Bract Extract Paper Strip Stored at Refrigeration Temperature

Legend:

- Color reaction stable
- Color reaction not stable
- M – Manual Method of extraction
- S – Soxhlet method of extraction
- R – Rotary Evaporator Method of extraction

Figure 6 shows that the Banana bract extract paper strips from the four cultivars: B1 (Lakatan); B2 (Latundan); B3 (Damilig- Lolloy) and B4 (Damilig – Bato) kept at refrigeration temperature were stable only until week 1. This short stability could be due to the moisture formed during storage of the paper strips at

lower temperature which could have destabilized the pigment anthocyanin as stated by Strasinger *et.al.*; [1].

In Table 1 the stability of the Four Banana Bract Cultivar Extract Paper Strips were compared at room temperature and refrigeration temperature.

Table 1: Stability of the Four Cultivars of Banana Bract Extract Paper Strips at Room and Refrigeration Temperature.

| Bract Type               | Method of Extraction | Week 1 |     | Week 2 |     | Week 3 |     | Week 4 |     |
|--------------------------|----------------------|--------|-----|--------|-----|--------|-----|--------|-----|
|                          |                      | Room   | Ref | Room   | Ref | Room   | Ref | Room   | Ref |
| B1<br>Lakatan            | Manual               | ✓      | ✓   | ✓      | ✗   | ✗      | ✗   | ✗      | ✗   |
|                          | Soxhlet              | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |
|                          | Rotary Evaporation   | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |
| B2<br>Manila             | Manual               | ✓      | ✓   | ✓      | ✗   | ✗      | ✗   | ✗      | ✗   |
|                          | Soxhlet              | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |
|                          | Rotary Evaporation   | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |
| B3<br>Damilig-<br>Lolloy | Manual               | ✓      | ✓   | ✓      | ✗   | ✗      | ✗   | ✗      | ✗   |
|                          | Soxhlet              | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |
|                          | Rotary Evaporation   | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |
| B4<br>Damilig-<br>Bato   | Manual               | ✓      | ✓   | ✓      | ✗   | ✗      | ✗   | ✗      | ✗   |
|                          | Soxhlet              | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |
|                          | Rotary Evaporation   | ✓      | ✓   | ✓      | ✗   | ✓      | ✗   | ✗      | ✗   |

LEGEND:

- ✓ - Color reaction stable
- ✗ - Color reaction not stable

As gleaned from table 1, banana bract extract paper strips from the four cultivars extracted by manual method and stored at refrigeration temperature were stable until week 1, when stored at room temperature were stable until week 2. Banana bract extract paper strips from the four cultivars extracted by Soxhlet method stored at refrigeration temperature were stable until week 1, when stored at room temperature were stable until week 3. Banana bract extract paper strips from the four cultivars when extracted by Rotary evaporator method stored at refrigeration temperature were stable until week 1, when stored at room temperature were stable until week 3. Probably, the reason why banana bract extract paper strips prepared by soxhlet and rotary evaporator methods had a longer stability was due to the use of a solvent solution (95% ethanol) in the extraction process which could have an effect on the amount of anthocyanin extracted and incorporated to the paper strip. This is supported by the studies conducted by De Rosso and Mercandante [4]

and Puzimo-Duran *et.al.*; [3]. More so, the banana bract extract paper strips prepared from the four cultivars and stored at refrigeration temperature had a shorter life span as compared to those stored at room temperature. This validates the result of the studies conducted by Rein [5]; Roobha *et.al.*; [6] that temperature affects the stability of anthocyanin pigment and other environmental conditions could destabilize the anthocyanin pigment. Refrigeration of the banana bract extract paper strips absorbed moisture during storage at lower temperature which could have destabilized the anthocyanin absorbed by the paper strips as cited by Strasinger *et.al.*; [1]

**Determination of Urine pH**

The pH concentration of Fifteen randomly collected samples were determined using pH Meter, Commercial Urine pH strips (Multistix) and the Banana Bract Cultivar Extract Paper strips.

**Table 2: pH Level of the Fifteen Urine Samples Using Different pH Measurements for Urine**

| Urine Sample | pH Meter |     |     |     | Commercial Urine pH Strips |     |     |     | Banana Bract extract paper Strips |     |     |     |
|--------------|----------|-----|-----|-----|----------------------------|-----|-----|-----|-----------------------------------|-----|-----|-----|
|              | R1       | R2  | R3  | AVE | R1                         | R2  | R3  | AVE | R1                                | R2  | R3  | AVE |
| 1            | 5.0      | 5.0 | 5.0 | 5.0 | 6.0                        | 5.0 | 5.0 | 5.3 | 6.0                               | 5.0 | 5.0 | 5.3 |
| 2            | 5.0      | 5.0 | 5.0 | 5.0 | 5.0                        | 5.0 | 5.0 | 5.0 | 5.0                               | 6.0 | 5.0 | 5.3 |
| 3            | 5.0      | 5.0 | 5.0 | 5.0 | 5.0                        | 5.0 | 6.0 | 5.3 | 5.0                               | 5.0 | 5.0 | 5.0 |
| 4            | 6.0      | 6.0 | 6.0 | 6.0 | 6.0                        | 6.0 | 6.0 | 6.0 | 6.0                               | 5.0 | 6.0 | 5.7 |
| 5            | 6.0      | 6.0 | 6.0 | 6.0 | 5.5                        | 6.0 | 6.0 | 5.5 | 5.0                               | 5.0 | 6.0 | 5.3 |
| 6            | 6.0      | 6.0 | 6.0 | 6.0 | 6.0                        | 6.0 | 6.0 | 6.0 | 5.0                               | 6.0 | 6.0 | 5.7 |
| 7            | 6.0      | 6.0 | 6.0 | 6.0 | 6.0                        | 6.0 | 6.0 | 6.0 | 6.0                               | 6.0 | 6.0 | 6.0 |
| 8            | 5.5      | 6.0 | 6.0 | 5.8 | 6.0                        | 5.0 | 6.0 | 5.7 | 6.0                               | 5.0 | 5.0 | 5.3 |
| 9            | 6.0      | 6.0 | 6.0 | 6.0 | 5.0                        | 6.0 | 6.0 | 5.7 | 6.0                               | 5.0 | 6.0 | 5.7 |
| 10           | 5.5      | 6.0 | 5.5 | 5.7 | 5.0                        | 6.0 | 6.0 | 5.7 | 6.0                               | 6.0 | 6.0 | 6.0 |
| 11           | 7.5      | 7.0 | 7.0 | 7.2 | 7.0                        | 7.5 | 7.0 | 7.2 | 7.0                               | 7.0 | 7.0 | 7.0 |
| 12           | 7.8      | 7.5 | 7.4 | 7.6 | 8.0                        | 7.5 | 8.0 | 7.8 | 7.0                               | 7.0 | 8.0 | 7.3 |
| 13           | 8.0      | 8.0 | 8.0 | 8.0 | 8.0                        | 8.0 | 7.5 | 7.8 | 8.0                               | 8.0 | 8.0 | 8.0 |
| 14           | 7.4      | 7.5 | 7.5 | 7.5 | 7.0                        | 8.0 | 7.0 | 7.8 | 7.0                               | 8.0 | 7.0 | 7.3 |
| 15           | 7.7      | 7.5 | 7.6 | 7.6 | 8.0                        | 8.0 | 8.0 | 8.0 | 8.0                               | 7.0 | 8.0 | 7.7 |

Using pH meter, Multistix and Banana bract extract paper strips, the fifteen (15) urine samples has a pH range from pH 5.0 to pH 8.0. This indicates that urine samples are acidic and alkaline in nature.

The study hypothesized that there is no significant differences between the banana bract cultivar extract paper strips and pH meter or Multistix as urine pH indicator. This is analyzed using t-test as shown in Table 3.

**Table 3: t-Test of Differences Between pH Meter and Lacatan Bract Extract Paper Strips and Between Multistix and Lacatan Bract Extract Paper Strips as Urine pH Indicator**

|                                    | Mean | Variance | N  | Df | t-computed | t tab  | Prob                 |
|------------------------------------|------|----------|----|----|------------|--------|----------------------|
| pH meter                           | 6.29 | 1.030    | 15 | 28 | 0.2989     | 2.0484 | 0.7672 <sup>ns</sup> |
| Lacatan bract extract paper strips | 6.78 | 0.998    | 15 |    |            |        |                      |
| Commercial pH strips (Multistix)   | 6.43 | 1.124    | 15 | 28 | 0.66817    | 2.0484 | 0.5095 <sup>ns</sup> |
| Lacatan bract extract paper strips | 6.18 | 0.998    | 15 |    |            |        |                      |

ns= not significant

Table 3 shows the test of differences between the pH meter and Lacatan bract extract paper strips and between multistix and lacatan bract extract paper strips as urine pH indicator. The t-test reveals that there is no significant differences between pH meter and Lacatan bract extract paper strips in measuring pH of urine samples since the probability value is 0.7672 which is greater than 0.05. This proves that Lacatan bract extract paper strips are a possible substitute in measuring pH of urine. Furthermore, this is shown in their variance. pH meter has a more scattered value with a variance of 1.030 than Lacatan bract extract paper strips with a variance of 0.998.

**Color Reference Chart**

The table also shows that there is no significant differences between Multistix and Lacatan bract extract paper strips in measuring pH of urine samples since the probability obtained is 0.5095 which is greater than 0.05. This proves that Lacatan bract extract paper strip is a possible substitute in measuring pH of urine. Furthermore, this is shown in their variance. Multistix has a more scattered value with a variance of 1.124 as compared with the variance of Lacatan bract extract paper strips with a value of 0.998. Based on the results of Table 3 the null hypothesis is accepted.



**CONCLUSIONS**

Based on the findings of the study, the following conclusions were drawn:

1. The different banana bract cultivars might contain different types of anthocyanin, the main active component of the banana bract on its potential as

pH indicator, which resulted in the difference in the color reactions of the four cultivars utilized in the study with solutions of varying pH. Furthermore the use of 95 % ethanol as a solvent in extracting the anthocyanin pigment from the four banana cultivars in both Soxhlet and Rotary

Evaporator methods may have increased the amount of anthocyanin extracted which was incorporated into the paper strips giving a more distinct color reaction of the pH paper strips.

2. The processed Banana bract cultivar paper strips are better kept at room temperature than at refrigeration temperature in order to have a longer shelf life.
3. The Lacatan (Barangan) Bract extract paper strips processed by Rotary Evaporator method are as effective as the pH meter and commercial urine pH strips (Multistix) in determining the pH of urine samples.
4. 4. Based on the findings of the study, Lacatan (Barangan) bract extract paper strips is a possible substitute in determining urine pH as it shows variance of colors at different pH levels.

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