



## PHYTOCHEMICALS SCREENING AND ACID- BASE INDICATOR PROPERTY OF ETHANOLIC EXTRACT OF *ALTHEA ROSEA* FLOWER

Ahmad Bala Ahmad\*, Naziru Alhassan Muhammad, Mustapha Balarabe Idris, Khalid Da'u Khalid

<sup>1</sup>Department of chemistry, Faculty of Applied Science, Jodhpur National University, Narnadi Jhanwar Road Boranada, Rajasthan, India

\*Corresponding author: [ahmadbala280@gmail.com](mailto:ahmadbala280@gmail.com)

### ABSTRACT

Secondary metabolites are unwanted compounds released by plants during excretion process but some of them are also used by the plants as protective agents. The presences of these compounds were reported to have medicinal properties. In the present study phytochemicals screening of the ethanolic extract of *Althea rosea* flower (garden hollyhock) revealed the presence of alkaloids, flavonoid, triterpenoids, saponins, tannins, cardiac glycoside, and phenolic compounds. Indicators currently in-used cause environmental pollution and they are hazardous to the living organism. The ethanolic extract of *Althea rosea* flower (garden Hollyhock) shows a perfect colour change at equivalent point during acid base titration as compared with synthetic indicators. Therefore the extract from this flower could be used as replacement for both methyl orange and phenolphthalein in neutralization reaction.

**Keywords:** *Althea rosea*, Ethanolic extract, Phytochemicals, Titration

### 1. INTRODUCTION

The medicinal value of Herbal medicines are also in great demand in the developed world and according to world health organization (WHO), more than 80% of the world's population relies on them for their primary health care needs. Phytochemical molecules such as vitamins, terpenoids, phenolic acids, lignins, tannins, flavanoids, alkaloids, amines e.t.c are bioactive chemicals naturally synthesized in all parts of the plant body which are rich in antioxidant activity are responsible for the medicinal properties of the plant. They are also regarded as secondary metabolites because the plants that manufacture them may have little need for them. Studies have shown that many of these antioxidant compounds possess anti-inflammatory, antiatherosclerotic, antitumor, antimutagenic, anticarcinogenic, antibacterial, and importantly, these studies will be helpful to isolate, characterize and studies their pharmacological activity. The previous studies showed that *Althaea rosea* possessed many pharmacological effects including antimicrobial, cardiovascular, prevention of urolithiasis, antiestrogenic, cytotoxic and immunomodulating effects. [1] Acid base titration is the determination of the concentration of an acid or base by exactly neutralizing the acid or base with an acid or base of known concentration using an indicator. Indicators are useful substances not only in acid base titration but in all volumetric analysis in which end point is to be determined such as redox reaction, gravimetric analysis and even in the determination of the amount of some elements present in solution like the amount of chloride present in water sample. The end point in traditional titrimetry is usually indicated by some substances added into the analyte solution, which change colour immediately after the

equivalence point has been attained [2]. The substances that change colour when the acidity of the solution changes are known as acid-base indicators [2].

Indicators work because they are weak acids which, when in solution, exist in equilibrium with their conjugate base. The acid and its conjugate base each have different colours, and as the equilibrium shifts from one direction to the other, the color of the indicator solution changes. Some indicators exhibit only two colours and some exhibit a wide range. Each indicator must be individually studied to determine its behavior as a function of pH. [3] In spite of the numerous instrumental techniques currently available for the chemical analyses of various samples, conventional methods of analysis such as gravimetry and titrimetry are still relevant [2]. Synthetic indicators have certain disadvantages like high cost, availability and chemical pollution hence natural indicators obtained from varies plant parts like flowers, fruits, leaves etc. will be more advantageous [3].

Since the use of synthetic indicators is found to be harmful to our environment which its effect is directly dangerous to the living organism, it is a task on the shoulder of chemist to find a way of substituting them by the more environmentally friendly substances; hence extraction from plant can serve the purpose. Some of these were *Dianthus plumarius* and *Antirrhium majus* [4], *Nerium odoratum* [5], *Ipomoea biloba* [6], *Jacaranda acutifolia* flower [7], *pride of barbados* [8], *Morus alba linn* fruit [9], *Nerium indicum* [2], *Moriga oleiflora* leaf [10] and *caesalpinia* [12].

*Althaea rosea* (L.) is a popular garden plant. It is native to China, southern Europe, the Middle East and decoction of flowers is used as anti-inflammatory, febrifuge, demulcent and astringent agent [1]. The roots are used in the treatment of

ulcers and flowers as well as roots are used in the treatment of inflammation of the kidneys and the uterus. Seeds are thought to be diuretic and febrifuge [12, 13, and 14].

The aim of the present research is to investigate the presence of some phytochemicals and acid- base property of the ethanolic extract of *Althea rosea* flower.

## 2. MATERIAL AND METHODS

All reagents and chemicals used in this study were of analytical grade.

### 2.1. Collection and Identification of the *Althea rosea* flower

The *Althea rosea* flower (Garden hollyhock) was collected from Jodhpur city, India and identified by; Dr. N. L. Vyas (a botanist). The flower was then cleaned with distilled water to remove soluble impurities and kept for further use.

### 2.2. Extraction of *Althea rosea* flower

10g was weighed and then soaked into 200 mL ethanol and allowed to stay for 48hours for complete extraction. The pH of the extracted solution was determined using pH meter and found to be 6.3 which is slightly acid.

### 2.3. Phytochemical analysis

Extracts were subjected to phytochemical screening as described by method in [15]. Some portion of an extract was taken and distributed in ten different test tubes and tested with specific reagents for the presence of the following natural organic compounds like alkaloids flavonoid, saponins, triterpens, tannins, steroid, phenolic compounds, glycosides and phlobatanins.

#### 2.3.1. Test for alkaloids

200mg plant extract is dissolved in 10ml methanol and then filtered. In 1ml filtrate 6 drops of Dragondroff's reagent is added. Appearance of orange precipitate indicates presence of alkaloids.

#### 2.3.2. Test for flavonoides

5ml of dilute ammonia solution was added to the filtrate followed by concentrated sulphuric acid. A yellow color observed indicates the presence of flavonoids.

#### 2.3.3. Test for Triterpenes

300 mg of extract mixed with 5 ml chloroform and warmed for 30 minutes. The chloroform solution is then treated with a small volume of concentrated sulphuric acid and mixed properly. The appearance of red color indicates the presence of triterpenes.

#### 2.3.4. Test for saponins

Boiled 300 mg of extract with 5 ml water for two minutes. Mixture was cooled and mixed vigorously and left it for three minutes. The formation of frothing indicates the presence of saponins.

#### 2.3.5. Test for tannins

To an aliquot of the extract added sodium chloride to make to 2% strength. Filtered and mixed with 1% gelatin solution. Precipitation indicates the presence of tannins.

#### 2.3.6. Test for steroids

200mg plant material was taken in 10 ml chloroform and then filtered. In 2ml filtrate, 2ml acetic anhydride and small amount of H<sub>2</sub>SO<sub>4</sub> was added, lack of appearance of blue green ring indicates absence of steroids.

#### 2.3.7. Test for phenolic compounds

Formation of intense green, purple, blue or black colours with addition of 1% ferric chloride solution to the extract this indicate the presence of phenolic compounds.

#### 2.3.8. Test for cardiac glycoside

*Keller-Kiliani test*: 5ml of extract was treated with 1ml of glacial acetic acid containing 1 drop of ferric chloride solution. This was underplayed with 1ml of concentrated sulphuric acid. No brown ring of the interface indicates the presence of deoxysugar characteristic of cardenolides.

#### 2.3.9. Test for Phlobatanins

Deposition of red precipitate when extract was boiled with 1% aqueous hydrochloric acid indicates absence of phlobatanin.

## 2.4. Titrations

Few drops of the extract was added as an indicator for the following four different type of titrations: strong acid against strong base (sodium hydroxide VS hydrochloric acid), strong acid against a weak base(hydrochloric acid VS ammonium hydroxide), weak acid against strong base (ethanoic acid VS sodium hydroxide) and weak acid against weak base(ammonium hydroxide VS ethanoic acid). Standard indicators (methyl orange and phenolthelein) were also used and in the same manner and results obtained were compared. At end point of the titration the color of the flower extract was changed from yellow to pink.

## 3. RESULTS AND DISCUSSION

Diverse natural compounds are produced by plants and many of these are involved in plant defense and some for medication to the living organism. The result of the phytochemical screening was shown in table 1. The result revealed that *Althea rosea* containn many phytochemicals. The previous studies indicated that *Althaea rosea* possessed antimicrobial, cardiovascular, prevention of urolithiasis, antiestrogenic, cytotoxic and many pharmacological effects. It have been reported that presence of phytochemicals were responsible for the pharmacological activities of many plant including *Althaea rosea flower* as confirmed in the present study.

**Table 1: Result of the phytochemical screening of *Althaea rosea* flower**

Phytochemicals	Ethanollic extract
Alkaloids	Present
Flavonoids	Present
Triterpenoids	Present
Saponins	Present
Tannins	Present
Steroids	Absent
Phenolic compounds	Present
Cardiac glycosides	Present
Phlobatanins	Absent

Some pigments of plants can be used as Indicators; this is as a result of their property of sharp change in color when the equivalent point is attained during titration, thus any substance or pigment possessed this property can be used as indicator. In the present study, the ethanolic extract of *Althaea rosea* flower was used as indicator was in the neutralization reaction between different Titrant/Titrant. The results obtained were compared with two synthetic indicators (methyl orange and phenolphthalein) and the end point of the titration were found to be precisely or nearly the same. The results were shown in table 2, 3 and 4 respectively.

**Table 2: Result of titration using *Althaea rosea* flower extract indicator**

Titrant / Titrant	Mean value of titration $\pm$ SD
0.5M HCl / NaOH	25.17 $\pm$ 0.047
0.1M HCl / NH <sub>4</sub> OH	25.10 $\pm$ 0.047
0.5M CH <sub>3</sub> COOH / NaOH	24.80 $\pm$ 0.082
0.1M CH <sub>3</sub> COOH / NH <sub>4</sub> OH	25.13 $\pm$ 0.082

**Table 3: Result of titration using methyl orange indicator**

Titrant / Titrant	Mean value $\pm$ SD
0.5M HCl / NaOH	24.90 $\pm$ 0.125
0.1M HCl / NH <sub>4</sub> OH	25.00 $\pm$ 0.125
0.5M CH <sub>3</sub> COOH / NaOH	24.90 $\pm$ 0.082
0.1M CH <sub>3</sub> COOH / NH <sub>4</sub> OH	25.13 $\pm$ 0.153

**Table 4: Result of titration using Phenolphthalein indicator**

Titrant / Titrant	Mean value of titration $\pm$ SD
0.5M HCl / NaOH	24.97 $\pm$ 0.094
0.1M HCl / NH <sub>4</sub> OH	24.93 $\pm$ 0.082
0.5M CH <sub>3</sub> COOH / NaOH	24.80 $\pm$ 0.082
0.1M CH <sub>3</sub> COOH / NH <sub>4</sub> OH	25.10 $\pm$ 0.082

#### 4. CONCLUSION

From the above results, it could be concluded that *Althaea rosea* flower contained alkaloids flavonoid, triterpenes, saponins, cardiac glycosides, tannins and phenolic compounds which might be responsible for the pharmacological activity of the flower. In addition, the ethanolic extract can be used in place of both methyl orange and phenolphthalein indicators during acid-base titration.

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#### 6. REFERENCES

1. Ali EA. The Pharmaceutical Importance of *Althaea officinalis* and *Althaea rosea* : A Review. Department of Pharmacology, College of Medicine, Thiqr University, Nasiriyah 2013, 9.
2. Khalid DK, Mustapha BI, Naziru AM, Ahmad BA. *British J. Pharm. Res.*, 2016; **9(1)**:1-4.
3. Sajni KA. *Int. Current Pharm. J.* 2012; **1(12)**:420-422.
4. Abugri DA, Apea OB, Pritchett G. *Green Sustainable Chem.* 2012; **2**:117-122.
5. Pimpodkar NV, Surve BS, Bhise SH. *J. Curr. Pharma. Res.*, 2014; **4(2)**:1124-1127.
6. Eze SO, Ogbuefi RA. *Asian J. Nat. Appl. Sci.*, 2014; **3(1)**:54-60.
7. Bhise SH, Shinde NG, Surve BS, Pimpodkar NV, Shikalgar SS. *Int. J. Nat. Prod. Res.*, 2014; **4(1)**:33-35.
8. Jaspreet S, Kanika A, Parminder N, Geeta D. *Int. J. Pharma.*, 2011; **2(4)**:177-179.
9. Pathade SK, Patil SB, Konda-war N, Magdum CS. *Int. J. Chem. Tech. Res.*, 2009; **3(1)**:549-551.
10. Khalid DK, Mustapha BI, Naziru AM, Ahmad BA. *Ame. Chem. Sci. J.*, 2016; **13(2)**.
11. Naziru AM, Ahmad BA, Khalid DK, Mustapha BI. *Euro. J Biom and Pharm Sci.*, 2016; **3(3)**:1-4.
12. Izonfuo WAL, Fekarurhobo GK, Obomanu FG, Daworiye LT. *J. Appl. Sci. Environ. Mgt.*, 2006; **10(1)**:5-8.
13. Pimpodkar NV, Shikalgar S, Shinde N, Bhise S, Surve B. *Asian. J. Pharm. Ana.*, 2014; **4(2)**:82-84.
14. Abugri1 DA, Apea OB, Pritchett G. *Green Sustainable Chem.*, 2012; **2**:117-122.
15. Harborne JB. *Phytochemical Methods: A Guide to Modern Technique of Plant Analysis.* Chapman and Hall, London. 1984.