PHYTOCHEMICAL EVALUATION OF MUSA ACUMINATA BRACT USING SCREENING, FTIR AND UV-VIS SPECTROSCOPIC ANALYSIS

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ABSTRACT

The use of plants as medicines goes back to early man. Musa plant is known as the largest herbaceous flowering plant in the world. All the parts of these plants are beneficial to mankind in the medicinal aspects and ornamental uses. Musa acuminata - one of the cultivar variety of the family Musaceae – has been used in traditional medicine since hundred years to alleviate various diseases and health problem. The objective of this study is to investigate the preliminary phytochemical constituents present in the bract of the inflorescence and further confirmed by FTIR and UV-Vis spectroscopic studies. The results reveal the presence of alkaloids, flavonoids, terpenoids, coumarins, phenols, tannins, glycosides, steroids and saponins in different solvents.

KEY WORDS: Musa Acuminata, Bract, Phytochemical Screening, FTIR, UV-Vis.

INTRODUCTION

The world of plants is virtually an untapped reservoir of novel bioactive agents. Plants can be evaluated for a wide range of biological activities and can be used for treatment of diseases. Novel drug entities continue to be developed through research into their constituents. One such plant family of medicinal importance is Musaceae. Musaceae family has 2 genera and 42 different species and within 42 species, 32 species belongs to musa species [Nuengchamnong et al, 2004] – one of the largest known herbaceous flowering plant in the world. It includes banana and plantains [Evans, 2002]. Banana, an antique fruit crop known as ‘Apple of the Paradises’ has played interesting and important roles in the history of human civilizations. One of the cultivar variety Musa acuminata ‘Nendran’ (AAB) is an important cooking banana in Kerala and Tamilnadu. It is an arboresent perennial herb. On maturing, a true stem or growing point emerges as banana blossom (inflorescence) from the centre of the tightly rolled bunch of leaves. The banana inflorescence is an elongated, oval shaped, dark purple bud. As it opens it is seen that the slim, nectar rich, tubular, toothed,
white flowers are clustered in whorled double rows along the floral stack. Bracts lift from the first hand in 3 to 10 days. The bracts are soon shed and fully grown fruits in each cluster become a hand of bananas (Figure 1) [Simmonds 1962; Simmonds and Stover 1987].

Figure 1. Musa acuminata ‘Nendran’ fruit bunch with inflorescence

All parts of the plant have medicinal applications. Banana flowers are treated in several cuisines as vegetables, while bracts are used as cattle feed. Banana flowers are considered to be good source of vitamin A and C and are used in treatment of bronchitis, constipation, ulcers and good for diabetics. It is traditionally believed to be beneficial as a lactating agent and helps to relieve painful menstruation [Sheng et al, 2010].

Plants are potent biochemist of photochemical having biological significance. Researchers today are involved in isolation and characterisation of various plants and plant constituents [Savithramma et al, 2011]. Bioactive plant constituents can be extracted from any part of the plant like bark, leaves, flowers, roots, fruits, seeds etc [Cragg and Dravid, 2001].

Previous research on preliminary phytochemical screening of dried leaves and fruit peels of Musa paradisiaca revealed the presence of some glycosides, anthocyanins, tannins, flavonoids and carbohydrates [Anhwange 2008; Archibald 1949; Alisi et al 2008]. No research has been reported on the phytochemical screening of banana flowers except a quantitative study on saponins and flavonoids [Sheng et al, 2010] and later phenolic content by Logananyaki et al [Kitdamrangsant, 2008]. The bract part of Musa paradisiaca was reported to contain anthocyanins such as delphinidin, pelaragonidin, peonidin and malvidin [Pazmino et al, 2001; Pothavorn, 2008]. Total phenolic content in bract was reported as the lowest compared to other parts such as rhizome, fruit peel, ovary, petiole, pseudo stem and leaves [Logananyaki et al, 2010]. In this present study an attempt has been made on systematic screening of bract extract of Musa acuminata ‘Nendran’ (AAB) with the purpose of discovering new bioactive compounds.
Phytochemical analysis of nearly related species reported may vary from one to another due to the morphology. Hence attempt has been made to study the phytochemical screening of Musa acuminata bract collected from the cultivated farm of Coimbatore district of Tamilnadu, India.

MATERIALS AND METHODS

Chemicals and Reagents

All the chemicals and reagents used for screening test were of analytical grade.

Collection of Plant material

Fresh inflorescence of Musa acuminata ‘Nendran’ (MAN) were collected from banana farm in Thirumalayampalayam, Coimbatore, India. The florets and bracts (Figure 2a) were separated. The bracts were washed with water, shade dried (Figure 2b) and ground to powder using an electronic blender, sieved and the fine powder was stored in air tight container.

![Figure 2(a). MAN inflorescence with floret and bract](image)

![Figure 2(b). Shade dried MAN bract](image)

Preparation of sample extract for phytochemical screening

Petroleum ether, chloroform, ethyl acetate, methanol and water extracts of bract of Musa acuminata was obtained by maceration. 20 g of the powdered bract was soaked in 200ml of each of the solvents for 48h at room temperature. It was then agitated using mechanical shaker to obtain successive extracts and filtered. The filtrates obtained were evaporated to dryness under vacuum using rotary evaporator [Syed Imran et al, 2012]. These extract were used for preliminary phytochemical screening.

Qualitative phytochemical analysis

Phytochemical screening of MANB was carried out using standard qualitative methods as described by various researchers Kotate [1999] and Harborne [1984]. Preliminary
qualitative phytochemical screening for bioactive compounds was carried out by the methods described below [Kotate et al, 2010; Harborne, 1998; Egwaikhide and Gimba, 2007; Savithramma et al, 2011]:

(i) Detection of carbohydrates
   To 2 ml of the extract, 2 ml of Molish’s reagent and 2 ml of concentrated sulphuric acid was added. Formation of a reddish ring indicated the presence of carbohydrate.

(ii) Detection of reducing sugar
    2 ml of Fehling’s solution was added to 2 ml of the extract and boiled for 5 minutes. Formation of a brick red precipitate indicated the presence of reducing sugar.

(iii) Detection of alkaloids
      A little of the extract was stirred with Mayer’s reagent (potassium mercuric iodide). Formation of cream coloured precipitate indicated the presence of alkaloids.

(iv) Detection of saponins
     About 2 ml of the extract was diluted with 20 ml of distilled water and shaken in a graduated cylinder for 15 minutes. Formation of 1 cm layer of foam indicated the presence of saponins.

(v) Detection of tannins
    To 2 ml of the extract, few drops of 1% lead acetate were added and the formation of yellowish precipitate indicated the presence of tannins.

(vi) Detection of flavonoids
     To a small quantity of the extract dilute sulphuric acid was added. The appearance of orange colour indicated the presence of flavonoids.

(vii) Detection of terpenoids
      To 2 ml of extract, 2 ml of acetic acid and sulphuric acid were added. Formation of bluish green ring indicated the presence of terpenoids.

(viii) Detection of phlobotannins
       2 ml of the extract was boiled with 1% hydrochloric acid. Deposition of red precipitate indicated the presence of phlobotannins.
(ix) Detection of coumarins
3 ml of 10% sodium hydroxide was added to 2 ml of extract. Formation of yellow colour indicated the presence of coumarin.

(x) Detection of cycloglycosides
To 5 ml of extract, 2 ml of acetic acid, 1 drop of 1% ferric chloride and 1 ml of sulphuric acid was added. Formation of greenish ring indicated the presence of cycloglycosides.

(xi) Detection of total phenol
Extract was treated with 3-4 drops of ferric chloride solution. Formation of deep blue colour indicated the presence of phenol.

(xii) Detection of quinones
The extract with 5 ml of hydrochloric acid resulted in yellow precipitate, indicating the presence of quinones.

(xiii) Detection of anthraquinones
To 2 ml of extract, 2 ml of 10% ammonium hydroxide was added. Formation of pink colour indicated the presence of anthraquinones.

(xiv) Detection of steroids
2 ml of the extract was dissolved in 2 ml of chloroform. To this equal volume of acetic acid and concentrated sulphuric acid was added by the sides of test tube the formation of bluish green indicated the presence of steroids.

PHYTOCHEMICAL ANALYSIS BY SPECTROPHOTOMETER

Nature always stands as a golden mark to exemplify the outstanding phenomenon of symbiosis. The plants are indispensable to men for his life. Spectroscopic techniques are employed for qualitative and quantitative analysis of plant extract. The spectral studies like UV and FTIR are the preliminary work for the detection of phytochemical contents in the plants [Kotate, 2010; Harborne, 1998].

For UV-Vis and FTIR spectrophotometer analysis, the 5% stock solution of ethanol extract of MANB was used as such. The FTIR spectrum was taken on SHIMADZU FTIR - 8400F, at PSG College of Arts and Science College, Coimbatore, India, to detect the characteristic peaks and their functional groups.

To detect the UV-Vis spectrum profile, the extracts of Musa acuminata ‘Nendran’ bract were scanned in the wavelength ranging from 210–800 nm on SHIMADZU UV-Vis
spectrophotometer UV-1700 Pharmuspec model. The characteristic peaks were detected to confirm the different bioactive nutrients present in the sample extracts. The peak values of the FTIR and UV-Vis were recorded.

RESULTS AND DISCUSSION

Qualitative phytochemical analysis

Many compounds that occur in plant tissues are quite labile and almost inevitably may undergo change during extraction. Polarity of the solvent used for extraction may influence the group of bioactive compounds obtained from the plant material. The dry weight of plant material obtained after the extraction using different solvents are given in Table 1.

Table 1. Dry weight of plant materials obtained in different solvents

<table>
<thead>
<tr>
<th>Solvents used (Each 200 ml)</th>
<th>MAN Bract (mgs)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Petroleum ether</td>
<td>0.160</td>
</tr>
<tr>
<td>Chloroform</td>
<td>0.177</td>
</tr>
<tr>
<td>Ethyl acetate</td>
<td>0.100</td>
</tr>
<tr>
<td>Methanol</td>
<td>0.810</td>
</tr>
<tr>
<td>Water</td>
<td>0.910</td>
</tr>
</tbody>
</table>

The phytochemicals present in bract of Musa acuminata are summarized in Table 2.

Table 2. Phytochemical constituents present in MANB extract

<table>
<thead>
<tr>
<th>Phyto Compound</th>
<th>Petroleum ether</th>
<th>Chloroform</th>
<th>Ethyl acetate</th>
<th>Methanol</th>
<th>Water</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carbohydrates</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Reducing sugar</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Saponins</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Tannins</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Phlobotannins</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Coumarins</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Cycloglycosides</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Total phenols</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Quinones</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Anthraquinones</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Steroids</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
</tbody>
</table>

**Key:** “++” active compound copiously present, “+” active compound present, “-“active compound absent
The preliminary qualitative analysis (Table 2) showed the presence of alkaloids, saponins, tannins, flavonoids, terpenoids, coumarins, cycloglycosides, total phenols and steroids. All these phytonutrients were not extractable in one solvent. The petroleum ether extract of Musa acuminata ‘Nendran’ bract contains alkaloids and glycosides, whereas chloroform extract revealed the absence of phytochemical constituents. Extracts with ethyl acetate showed the presence of flavonoids. The presence of alkaloids, saponins, flavonoids, terpenoids, coumarins, glycosides, phenols and steroids was confirmed by methanol extract. The exceptional factor was tannin content which was found to be high in both methanolic and aqueous extract. Aqueous extract indicated the presence of coumarins and phenol. It is evident from the tabulation that the other phytoconstituents like quinones, steroids and phlobotannins were absent in all the five extracts.

The results allow summing up that Musa acuminata ‘Nendran’ bract is a good natural source of phytonutreints of biological significance.

**PHYTOCHEMICAL ANALYSIS BY SPECTROPHOTOMETER**

**FTIR spectra**

The results of FTIR peak values and functional groups are represented in Table 3. The FTIR spectrum profile is illustrated in the Figure 3. The FTIR analysis suggest the presence of different functional groups like of alkanes, aldehydes/ketones, alkene, ethers, amines and aromatics in ethanol extract of MANB.

The FTIR spectrum shows spectral bands or peaks due to the vibrations of individual bonds or functional groups. The fact that many functional groups can be identified by their characteristic vibration frequencies, makes the spectrum, the simplest and often the most reliable method of assigning a compound to its class. In spite of this, FTIR spectroscopy is most frequently used in phytochemical studies as a ‘fingerprint’ device, for comparing natural samples. The complexity of the spectrum lends itself particularly well to this purpose and such comparisons are very important in the complete identification of many types of plant constituent. The spectroscopy can also usefully contribute to structural elucidation, when new compounds are encountered in plants [Kokate et al, 2010].
Table 3. FTIR peak values of Musa acuminata ‘Nendran’ bract

<table>
<thead>
<tr>
<th>Possible Functional group</th>
<th>FTIR peak values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkane</td>
<td>2924, 1457</td>
</tr>
<tr>
<td>Aldehyde or ketone</td>
<td>1725, 1171</td>
</tr>
<tr>
<td>Alkene</td>
<td>1625</td>
</tr>
<tr>
<td>Ether</td>
<td>1251</td>
</tr>
<tr>
<td>Amine or amide</td>
<td>1551, 1051</td>
</tr>
<tr>
<td>Aromatic</td>
<td>979</td>
</tr>
</tbody>
</table>

Figure 3. FTIR spectrum of ethanol extract of MAN (B)

UV-Vis spectra

Spectral measurements are important in the identification of many plant constituents, purification of plant products and for screening crude plant extracts for the presence of particular classes of compounds.

The value of UV and visible spectra in identifying unknown constituents is obviously related to the relative complexity of the spectrum and to the general position of the wavelength. If a substance shows a single absorption between 250 and 260 nm, it could be any one of a considerable number of compounds (e.g. a simple phenol, a purine or pyrimidine, an aromatic amino acid and so on). Furthermore, spectral measurements in two or three other solvents and comparison with literature data might even indicate which particular compound it is. The above statements suggest that absorption spectra are of special value in plant studies [Harborne, 1998].

The UV-Vis spectral studies made on ethanol extract of Musa acuminata bract to understand its phytoconstituents are discussed below.
Table 4. UV-Vis absorption spectral data of Musa acuminata ‘Nendran’ bract

<table>
<thead>
<tr>
<th>Medium</th>
<th>Plant extract</th>
<th>Wavelength in nm</th>
<th>Absorption</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethanol</td>
<td>MANB</td>
<td>275</td>
<td>0.636</td>
</tr>
<tr>
<td></td>
<td></td>
<td>266</td>
<td>0.640</td>
</tr>
<tr>
<td></td>
<td></td>
<td>222</td>
<td>2.275</td>
</tr>
</tbody>
</table>

Figure 4. UV-Vis spectrum of ethanol extract MANB

Data in Table 4 and Figure 4 reveals that UV-Vis spectral peak values of Musa acuminata ‘Nendran’ bract extract in ethanol range between 222 to 275 nm. The MANB extract may contain carbonyl group (C=O) in conjugation with an olefinic group (C=C) and hence shows its UV absorption at longer wavelengths due to its lower energy gap between n and n* energy levels. It was also observed that the position of n → n* transition varies with the solvent used. [Dyer, 1987].

CONCLUSIONS

Curing of diseases and restoration of health have always been major objectives of humanity. The beneficial medicinal effects of plant materials result from the combinations of secondary metabolities present in the plants. Such discoveries led to an interest in the plants in the quest for new natural products.

The present findings and properties confirm that Musa acuminata ‘Nendran’ bract may have potential bioactive compounds. Further investigation can be done to exploit the pharmacological properties which in turn may help in the development of new bio products.
REFERENCES

13. Loganayaki, N, Rajendrakumaran, D, Manian S. 2010 Antioxidant capacity and phenolic content of different solvent extracts from banana (Musa paradisiaca) and mustai (Rivea hypocrateriformis). Food Sci Technol, 19,1251–1258.