

IDENTIFICATION OF BELLADONNA ALKALOIDS

PHARMACOGNOSY
3RD CLASS, 2ND
SEMESTER

LAB.4



**IDENTIFICATION OF
BELLADONNA
ALKALOIDS**

Qualitative Analysis

Quantitative Analysis

**Chromatography
T.L.C**

**Chemical Test
1-Specific Test
2-General Test**

Titration Method

What is the difference between Titration and Back-Titration?

- **In titration**, amount of standard solution, which is chemically equivalent to the analyte amount, is added. **In back titration**, excess of the standard titrant is added to determine the analyte amount.
- Normally, **in a titration**, only one direct reaction is taking place, which is between the standard titrant and the analyte. **In a back-titration**, two chemical reactions are taking place. One is with the standard and analyte, and the other is with the excess-standard titrant and a standard solution.

1- Quantitative analysis

- This analysis is done by the back titration method, since the resultant extract is not crystal, but liquid containing more than one component.
- In this test we will add an excess amount of acid of known normality and titrate the resultant mixture with a base of known normality.
- The excess of acid is determined by the use of methyl red indicator, there by indicating the amount of our alkaloid in the extract indirectly.



Aim: - Identify the belladonna alkaloids quantitatively.

Equipments and reagents:-

- Burette
- Small flask
- Chloroform
- N/20 Sulphuric acid
- N/20 sodium hydroxide
- Methyl red solution

Procedure:-

- 1) Dissolve the residue in 2ml of chloroform.
- 2) Add 5ml of N/20 Sulphuric acid.
- 3) Warm to remove the chloroform, cool.
- 4) Titrate the excess of acid with sodium hydroxide.
- 5) Using methyl red solution as an indicator.
- 6) The end point is the change in the mixtures color from pink to yellow.
- 7) Each N/20 of Sulphuric acid is equivalent to 0.01447g of alkaloids calculated as hyoscyamine.

Results:-

By the use of the equation

Excess of acid (5ml) – volume of the NaOH used in the titration (from your exp.) = volume of the acid reacted
(this volume \times 0.01447 = weight of alkaloids calculated as hyoscyamine)

2- Qualitative analysis

A) The specific test for Tropane alkaloid.

1) Vitali morin test

Aim: - To identify the Tropane alkaloids from other alkaloids.

Equipments and reagents:-

- Small beaker
- Fuming nitric acid
- Alcoholic KOH

Procedure:-

- 1) Take few mls of the extract.
- 2) Add to it drops of fuming nitric acid and evaporate.
- 3) Then add 2ml of alcoholic KOH.

Results:-

Violet color will be resulted.

Discussion:-

- The nitric acid will nitrate the benzene ring, and the color will be developed after addition of KOH.

2) Gerhard's Test

Aim: - To identify the Tropane alkaloids from other alkaloids.

Equipments and reagents:-

- Small beaker
- 2% HgCl_2 in 50% aqueous ethanol.

Procedure:-

Add 2% HgCl_2 in 50% aqueous ethanol to 0.006g of atropine.

Results:-

A deep red color will be developed.

B) General Tests

1- Mayer's test

Aim:- to indicate in general the alkaloid as other alkaloids.

Equipment and reagents:-

- Petri dish.
- Ethanol.
- HCL.
- Mayer's reagent.

Procedure:-

- 1) Take few crystals of Tropane alkaloid.
- 2) Dissolve in few mls of ethanol in a Petri dish.
- 3) Add two drops of HCL.
- 4) Add two drops of Mayer's reagent.

Results:-

White precipitation will occur.

2- Wagner's reagent  red-brown precipitate

3- Dragendorff's reagent  orange precipitate

T.L.C

Aim: - Used to identify qualitatively the belladonna alkaloids

Equipments and reagents:-

- Glass jar with its cover.
- Silica gel plates.
- Standard reagent.
- Mobile phase (acetone: water: ammonia (90:7:3))
- Spray reagent (dragendorff's reagent)
- Capillary tube.

Procedure:-

- 1) Prepare the mobile phase and put in the glass jar, cover the jar and leave it for 45 min. for saturation.
- 2) Apply the sample and the standard by the use of capillary tube on the silica gel plate.
- 3) Leave the plate in the jar until the solvent reaches 3/4 of the plate. Remove the plate, dry, and then spray with the spraying reagent.

Result

Orange spots appear for both standard and sample.

Note: -

Other used mobile phase =

- Chloroform: Acetone: Diethyl amine (50: 40: 10)
- Methanol: Benzene (66%: 33%)
- Chloroform: Diethyl amine (90:10)



THANK YOU