# PHARMACOPEIA ANALYSIS OF CAFFEINE ISOLATION OF CAFFEINE QUANTIFICATION OF CAFFEINE USING COLORIMETRIC METHOD

PHARMACOGNOSY-III PRACTICE (2020)

- Pharmacopoeia: A book published usually under the jurisdiction of the government and containing a list of drugs, their formulas, and methods for making medicinal preparations, requirements and tests for their strength and purity, and other related information. (European Pharmacopoeia, Turkish Pharmacopoeia-Adaptadion of European Pharmacopoeia etc)
- Pharmacopoeia Analysis: It is an analysis to determine whether the active substances or excipients used in pharmaceutical preparations comply with the standards reported in the pharmacopoeia.

# **Experiments**

Description

Solubility

Identification reactions

Foreign alkaloids

1,3,7-trimethylxanthantine

COFFEINUM (TF 1974)

# **DESCRIPTION**

- >White
- ➤ Crystal structure
- > Bitter

# **SOLUBILITY**

- Water
- Ethanol
- Ether
- Chloroform

# **DENTIFICATION REACTIONS**

#### Identification Reaction A

#### **Murexide Test**

The capsules are exposed to NH<sub>3</sub> vapour.

This color disappears with the addition of alkali

**Purple** color is formed.

#### identification Reaction B

The solution in water forms a white precipitate with 10% solution of tannic acid in water.

#### Identification Reaction C

The solution in water does not precipitate with iodine TS. Caffeine is converted to enol form by addition of dilute HCl, reacts with iodine and brown precipitate occurs. With the addition of NaOH, caffeine turns into keto form and the precipitate disappears.

#### Reaction

When heated with 2 parts of water, it gives a solution which is neutral to the turnusole and becomes clear when cooled.

#### Foreign alkaloids

The saturated solution in water does not form precipitate with Mayer's reagent (K-Mercury iodide).

# ISOLATION OF CAFFEINE

The extract becomes alkali and the tannins are separated

Folia Theae (~ 10gr) Extracted with 20 ml 10% CaO.

Extracted with 20 ml CHCl<sub>3</sub> (3 times).

CHCl<sub>3</sub> solutions are transferred to the flask and distilled

CRYSTAL CAFFEIN

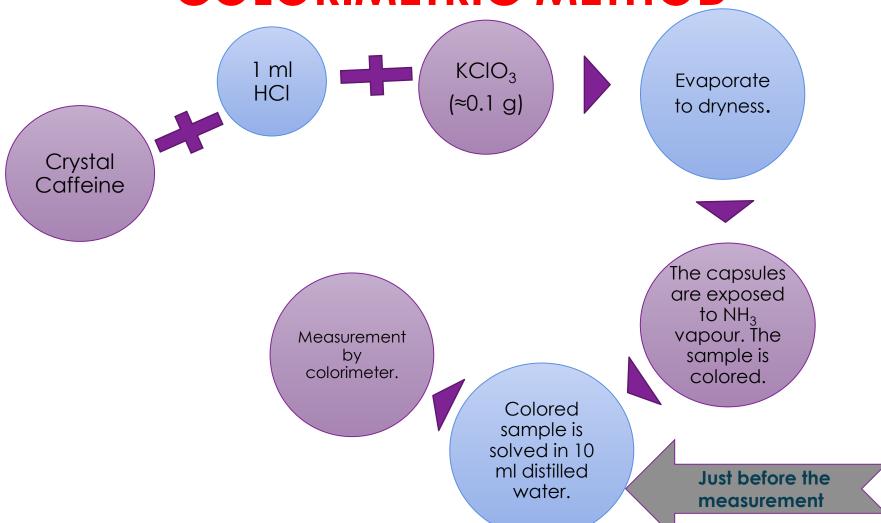
Filter into capsule (pleated filter paper) and water is evaporated

Cleaning with activated charcoal.

Raw caffeine is solved using 10 ml boiling water.

To separate pigment and foreign materials

# QUANTIFICATION OF CAFFEINE USING COLORIMETRIC METHOD

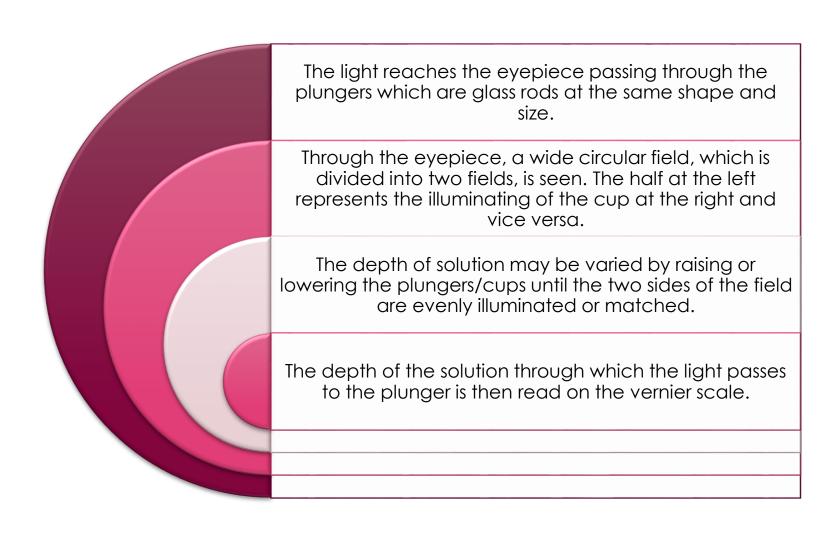


# COLORIMETRIC METHOD

To determine the concentration of a colored substance by comparison to a standard solution at a specific concentration is called colorimetry. The device used for this purpose is called colorimeter.



### **MEASUREMENT**



Lambert-Beer law is used for the calculation. Matching the illuminating of two fields means matching the absorbances.

$$A_1 = A_2$$

$$E \times I_n \times C_n = E \times I_s \times C_s$$

E=Absorption coefficient (cm<sup>-1</sup> . g<sup>-1</sup> . L)

I = Depth of the solution (cm)

c = Concentration (g/L)

$$C_{n} = \frac{I_{s} \times c_{s}}{I_{n}}$$

# CALCULATION

The color intensities of the sample and the standard solutions are compared to calculate the percentage of alkaloids.

 $c_s \!\!: 0.1 \text{ g/L}$   $l_n$  ve  $l_s \!\!: \text{the value read on the vernier scale}$ 

 $\begin{array}{ccc} \textbf{10,...} \ \textbf{g} \ \textbf{drug} & \textbf{c}_{\textbf{n}} \ \ \textbf{g} \ \textbf{caffeine} \\ \textbf{100} & ? \end{array}$ 

# THE ISSUES TO BE CONSIDERED FOR USING COLORIMETRIC METHOD

- > Solutions should be clear and freshly prepared.
- > The solution should be colourfull or coloured before the measurement.
- The color tone of the solutions should not be too light or too dark.
- > Cups and plungers should be clean.