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ESTIMATION OF PIPERINE IN MARKET SAMPLES OF BLACK PEPPER AND POLY- HERBAL FORMULATIONS

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ABSTRACT

Black pepper, used as a spice and seasoning contains piperine which is medicinally important. *Piper nigrum* and *Piper longum* containing Piperine are used In many ayurvedic preparations. The safety and efficacy of allopathic drugs can be proved on basis of standardization data. But, there are very few Poly Herbal Formulations for which standardization data is available. The objective of the present study was to standardize pepper and pepper containing Poly Herbal Formulations. Two samples of black pepper and two samples of Poly Herbal Formulation (Trikatu Churna- Ritesh Pharmaceutical and Yasthama tablet- Shri Yash Remedies) containing *Piper nigrum* as one of the main ingredient were collected from different parts of Gujarat. All four samples were subjected to qualitative estimation (TLC) of Piperine by method mentioned in Indian Herbal Pharmacopoeia and quantitative estimation (UV Spectroscopy) by method mentioned in manual published by Ministry of Health and Family Welfare , Govt. of India. The Piperine content in the two market samples was found to be 1.25 % (w/w) and 1.89 % (w/w) respectively. The Poly Herbal Formulations gave 1.05 % (w/w) and 1.024 % (w/w) of piperine content.

INTRODUCTION:

Black pepper is used globally as a spice and seasoning. *Piper nigrum* and *Piper longum* containing Piperine are used in many ayurvedic preparations. Piperine is basically used in preparations intended for stomach and digestive disorders, colds, bronchitis, neuralgia, scabies, piles, various skin diseases, etc. Standardization of the Poly-Herbal Formulations was carried out by estimation of piperine content.

EXPERIMENTAL METHODS:**Plant Material Collection:**

- ❖ The fruits of plant *Piper nigrum* (Piperaceae) were collected from markets of Baroda and Ahmedabad.
- ❖ Trikatu Churna- Ritesh Pharmaceutical and Yasthama Tablet – Shri Yash Remedies were chosen as the poly-herbal

formulations to be standardized.

1) Qualitative Estimation : As per method mentioned in Indian Herbal Pharmacopoeia.

Test Solution- The powdered drug (1 g) was refluxed with methanol (10 ml) for 30 min, filtered and the filtrate was evaporated to dryness. The residue was dissolved in methanol (3ml).

Reference Solution- Piperine (1 mg) dissolved in methanol (1 ml).

Solvent System- Toluene: Diethyl ether: Dioxane (9.4: 3.2: 2.4)

Procedure- About 5 microlitres of each of test solution and reference solution was applied on 2 different tracks on a precoated silica gel GF₂₅₄ plate (5 * 15 cm) of uniform thickness (0.2 mm). Plate was developed in solvent system up to 10 cm.

Visualization of Spots- Under UV 254 light.

Evaluation – All spots appeared violet in UV. The major spot at Rf 0.48 corresponds to piperine. The other spots visible are at Rf 0.27, 0.53, 0.59, 0.64.

2) Quantitative Estimation: As per method mentioned in the manual printed by Ministry of Health and Family Welfare, Govt. of India.

Preparation of Piperine Standard solution:

- 0.100 g Piperine was weighed and placed in 100 ml volumetric flask.
- 70 ml Ethylene dichloride was added to it to dissolve the Piperine and volume was made up to the mark.
- 10 ml from it was pipetted into 100 ml volumetric flask and diluted up to volume.
- 1, 2, 3, 4, 5, 6 ml aliquots were pipetted into six 100 ml volumetric flasks and

diluted to volume with Ethylene dichloride.

- UV spectrophotometer was adjusted to zero absorbance with Ethylene dichloride and absorbance of each final solution was read at maximum 342- 345 nm using UV light source and Ethylene dichloride in reference cell.
- A graph of concentration against observed absorbance was plotted.

Procedure:

- Sample was ground to pass 60 mesh sieves and blended uniformly.
- 0.5 g test sample was weighed accurately and transferred to 125 Erlenmeyer flask, Protected from light.
- 70 ml Ethylene dichloride was added and refluxed and stirred for 1 hour.

- It was cooled to room temperature and filtered quantitatively through paper into 100 ml volumetric flask.
- Rest of the extracted residue was transferred to filter, washed thoroughly and diluted to volume.
- 2 ml of this solution was pipetted into 100 ml volumetric flask and diluted to volume.
- Absorbance was recorded at maximum 342-345 nm . Percent Piperine content was obtained from the calibration curve.

RESULTS AND DISCUSSION:

Sample A- Black pepper (Baroda)

Sample B- Black pepper (Ahmedabad)

Sample C- Trikatu Churna- Ritesh Pharmaceutical

Sample D- Yasthama Tablet – Shri Yash Remedies

1) Qualitative estimation (Thin Layer Chromatography)

| Sample | | | | | |
|------------------|--------------|--------------|-------------|--------------|--------------------------|
| Rf Values | A | B | C | D | Standard Piperine |
| | 0.478 | 0.473 | 0.48 | 0.476 | 0.48 |
| | 0.69 | 0.74 | 0.70 | 0.72 | -- |

2) Quantitative estimation (UV Spectroscopic Method)

| Sample | Absorbance | Concentration ($\mu\text{g/ml}$) | Percentage of Piperine |
|---------------|-------------------|--|-------------------------------|
| A | 0.2444 | 1.2427 | 1.25 |
| B | 0.3218 | 1.8921 | 1.89 |
| C | 0.1382 | 0.3515 | 1.05 |
| D | 0.1510 | 0.3840 | 1.024 |

CONCLUSION:

- The Piperine content in the two market samples of pepper was found to be 1.25% (w/w) and 1.89 % (w/w) respectively.
- The Poly Herbal Formulations gave 1.05 % (w/w) and 1.024 % (w/w) of Piperine content.

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