Advanced UV Spectrophotometry-Classical Least Squares Determination of Paracetamol, Phenylpropanolamine HCl, and Chlorpheniramine Maleate in Tablet Dosage Form

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Abstract.

This study aims to develop and validate an advanced UV spectrophotometry-Classical Least Squares (CLS) for the simultaneous determination of Paracetamol (PA), Phenylpropanolamine HCl (PH), and Chlorpheniramine Maleate (CH) in tablet dosage form. The method involves the preparation of standard solutions of the three compounds across their respective concentration ranges 3-9 µg/mL for PA, 170-512 µg/mL for PH, and 7-19 µg/mL for CH. The CLS spectrophotometry UV method was applied to these spectra to construct calibration, which were then used to predict the concentrations of PA, PH, and CH in tablet dosage form. Validation of the method demonstrated excellent linearity, precision, and accuracy, with recovery rates of 99.26% for PA, 100.06% for PH, and 99.41% for CH. Sensitivity analysis revealed Limits of Detection (LOD) of 0.10 µg/mL for PA, 1.20 µg/mL for PH, and 0.30 µg/mL for CH, and Limits of Quantification (LOQ) of 0,30 µg/mL for PA, 4.00 µg/mL for PH, and 1.50 µg/mL for CH. The method's robustness and reliability make it an excellent choice for routine quality control of these compounds in pharmaceutical tablet formulations.

Keywords: *Paracetamol,Phenylpropanolamine HCl, Chlorpheniramine Maleate, Classical Least Squares and UV Spectrophotometry.*

I. INTRODUCTION

The accurate and precise determination of pharmaceutical compounds within tablet dosage form is critical for ensuring the efficacy and safety of medications [1, 2]. Paracetamol (PA), Phenylpropanolamine HCl (PH), and Chlorpheniramine Maleate (CH) are commonly combined in tablets for their synergistic effects in treating cold and flu symptoms. Each of these compounds plays a distinct therapeutic role, PA serves as an analgesic and antipyretic, PH acts as a nasal decongestant, and CH functions as an antihistamine. Given their widespread use, the development of robust analytical methods for their simultaneous quantification is of paramount importance [3, 4]. Several studies have been published to determine the levels of PA, PH, and CH both in combination and with other compounds, namely High Performance Liquid Chromatography (HPLC) [5, 6], High Performance Thin Layer Chromatography (HPTLC) [7], Thin Layer Chromatography-Densitometry [8], Fourier Transform Infrared Spectroscopy (FTIR) [9], Liquid Chromatography Tandem-Mass Spectrometry [10], spectrophotometry [4, 11].Ultraviolet (UV) spectrophotometry is a widely used analytical technique due to its simplicity, cost-effectiveness, and rapid analysis capabilities [12-14].

However, the overlapping absorption spectra of these compounds pose a significant challenge in their simultaneous determination. To address this issue, advanced chemometric methods such as Classical Least Squares (CLS) can be employed. The CLS method allows for the deconvolution of overlapping spectra, enabling the accurate quantification of each component in a complex mixture [15, 16]. The integration of UV spectrophotometry with the CLS method not only enhances the accuracy and precision of the measurements but also streamlines the analytical process. This approach provides a robust tool for the pharmaceutical industry, ensuring that the therapeutic efficacy of multi-component tablets is maintained and compliance with regulatory standards is achieved [16, 17]. To the best of our knowledge, no previous

research has employed the UV spectrophotometric method with CLS for the determination of PA, PH, and CH in tablet dosage form. This study aims to develop and validate an advanced UV spectrophotometry-CLS for the simultaneous determination of PA, PH, and CH in tablet dosage form.

II. METHODS

The instrument used was UV spectrophotometer (Shimadzu 1800), MATLAB software version 7, mortar and pestle, analytical balance (Baeco Germany), and glassware (Oberoi).All chemicals were of analytical grade, Ethanol (Merck), PA, PH, and CH raw materials, Molexflu® tablets (each tablet contains PA 500 mg, PH12.5 mg, CH 2 mg), Molex Ayus Pharmaceutical, Indonesia.To prepare standard stock solutions of PA, PH, and CH, each compound was weighed accurately at 50 mg using an analytical balance. The weighed compounds were then transferred into separate 50 ml volumetric flasks. Ethanol was gradually added to each flask while swirling to ensure complete dissolution of the compounds. Once dissolved, the volume in each flask was made up to 50 ml with ethanol, resulting in standard stock solutions with a concentration of 1000 µg/mL for each of PA, PH, and CH. Calibration curves were constructed using the CLS method with UV spectrophotometry; concentrations ranging from 3–9 g/mL for PA, 170-512 g/mL for PH, and 7–19 g/mL for CH. Absorbance was measured using a UV spectrophotometer (Shimadzu 1800). Determination of PA, PH, and CH in tablet dosage form, twenty tablets were weighed and finely pulverized. A specific quantity of the resulting powder, equivalent to 500 mg of PA, 12.5 mg of PH, and 2 mg of CH, was accurately measured and transferred to a 50 mL volumetric flask. Ethanol was added to achieve the volume mark, and the solution was then filtered through filter paper.

An aliquot of 0.4 mL of the filtrate was pipetted into a 10 mL volumetric flask, with additional solvent added to the mark. The absorbance of the solution was then determined using the CLS Spectrophotometry UV method. The analytical method was validated according to ICH guidelines, focusing on linearity, precision, accuracy, Limit of Detection (LOD), and Limit of Quantification (LOQ) [2, 15, 18]. Linearity was established by constructing calibration curves for PA, PH, and CH with concentration ranges of 3-9 g/mL, 170-512 g/mL, and 7-19 g/mL, respectively. Precision was assessed through repeatability, with Relative Standard Deviations (RSD) well within the acceptable limits of less than 2% [17, 18]. Accuracy was evaluated by recovery studies, yielding results between 80%, 100%, and 120%, demonstrating the method's reliability [18, 19]. The LOD and LOQ were determined based on the standard deviation of the response and the slope of the calibration curve. These validation parameters confirm the method's robustness and suitability for accurate quantification of the active components in tablet dosage form [20].

III. RESULT AND DISCUSSION

Absorbance spectra were obtained using a UV spectrophotometer (Shimadzu 1800) and analyzed through the CLS method. This approach effectively resolved the overlapping spectral bands of PA, PH, and CH (Figure-1), enabling precise quantification. The CLS technique utilized a matrix of absorbance data at specific wavelengths (210-280 nm) to solve a system of linear equations, allowing for the accurate determination of each component despite spectral interferences [15, 16].

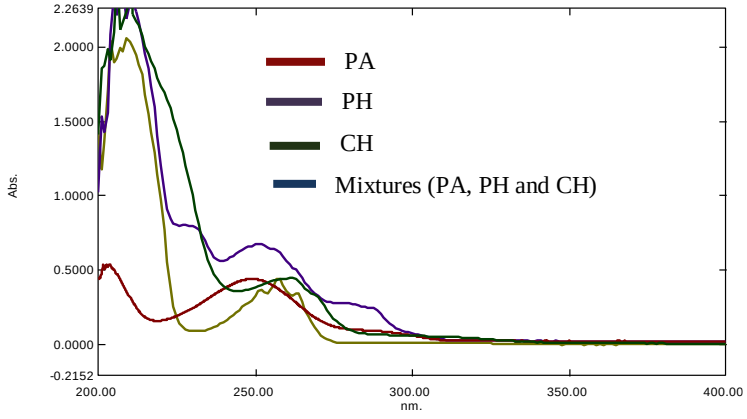

Fig 1. The absorption spectrum overlaps the spectrum of PA, PH, CH and mixtures (PA, PH and CH).

The advanced UV spectrophotometry- CLS method demonstrated high efficacy in the simultaneous determination of PA, PH, and CH in tablet dosage forms. Calibration curves for each analyte were meticulously constructed over the concentration ranges of 3-9 µg/mL for PA (Figure-2), 170-512 µg/mL for PH (Figure-3), and 7-19 µg/mL for CH (Figure-4). These curves exhibited excellent linearity, with correlation coefficients (R^2) consistently greater than 0.99 (Table-1), affirming the method's robustness in measuring analyte concentrations accurately [15, 17].

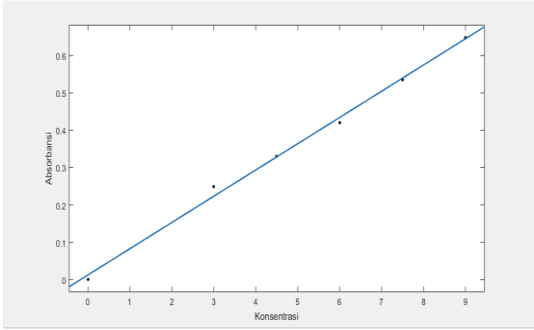

Fig 2. Calibration curve 3-9 µg/mL for PA

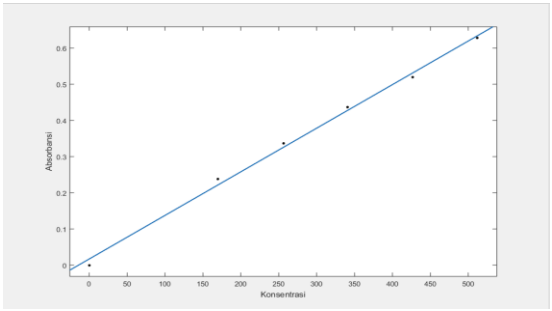

Fig 3. Calibration curve 170-512 µg/mL for PH

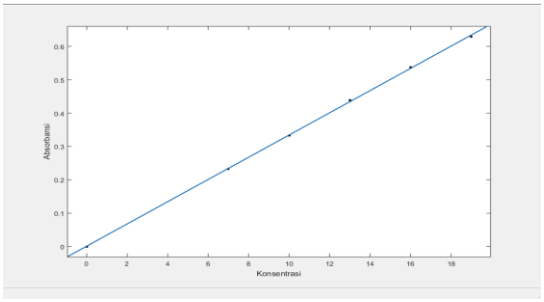

Fig 4. Calibration curve 7-19 µg/mL for CH

The analytical method was validated following ICH guidelines, focusing on linearity, precision, accuracy, Limit of Detection (LOD), and Limit of Quantification (LOQ). The results are summarized in the Table-1 below:

Table 1. Result Validation Method

No	Parameter	PA	PH	CH
1	Linearity	0.9997	0.9998	0.9997
2	Accuracy (%)	99.26	100.06	99.41
3	Precision (RSD)%	0.97	0. 33	0.54
4	LOD (µg/mL)	0.10	1.20	0.30
5	LOQ (µg/mL)	0,30	4.00	1.50

The validation results demonstrate that the analytical method adheres to ICH guidelines and is highly effective for the simultaneous quantification of PA, PH, and CH. The calibration curves exhibited exceptional linearity with correlation coefficients (R^2) exceeding 0.99 for all analytes, confirming a strong linear relationship between absorbance and concentration across the specified ranges. This indicates that the method is reliable for measuring concentrations within these ranges [17, 18]. Precision, resulting in Relative Standard Deviations (RSD) of less than 2% for all analytes. This low RSD underscores the method's high

reproducibility and consistency, ensuring reliable results in routine analytical settings [11, 17]. Accuracy was evaluated through recovery studies, which yielded recovery rates 80%, 100% and 120% for PA, PH, and CH. The recovery rates are within the acceptable range of 98% to 102% for PA and PH, indicating accurate quantification. These results affirm that the method provides accurate quantification of the analytes, effectively reflecting their true concentrations in the samples [21, 22]. Sensitivity was assessed by determining the LOD and LOQ. The low LOD and LOQ values highlight the method's ability to detect and quantify trace levels of the analytes, making it suitable for precise quality control in pharmaceutical formulations [22, 23].Overall, the method's validation confirms its robustness, precision, and sensitivity, making it an excellent choice for the accurate and reliable analysis of active pharmaceutical ingredients in tablet dosage forms [24]. The CLS spectrophotometry UV method was employed to quantitatively determine PA, PH, and CH in tablet dosage forms. The results of this study are presented in the following Table 2.

Table 2. Quantitatively Determine PA, PH, and CH in Tablet Dosage Form

No	Parameter	PA	PH	CH
1	Theoretical Content (mg/tablet)	500 mg	12.5 mg	2 mg
2	Observed Content (mg/tablet)	498.7 mg	12.4 mg	1.9 mg

The observed contents of PA, PH, and CH were 498.7 mg, 12.4 mg, and 1.9 mg per tablet, respectively, closely aligning with the theoretical contents of 500 mg, 12.5 mg, and 2 mg. This close agreement reflects the method's accuracy in quantifying the active ingredients [14-17].

IV. CONCLUSION

The Advanced UV Spectrophotometry-Classical Least Squares (CLS) method was successfully validated for the simultaneous quantification of PA, PH, and CH in tablet dosage forms. The method demonstrated excellent linearity, precision, accuracy, and sensitivity in accordance with ICH guidelines. The method's accuracy was confirmed by recovery rates within the acceptable range for PA and PH, and slightly above for CH. Precision studies showed low RSD, indicating high reproducibility. The LOD and LOQ confirmed the method's sensitivity. Overall, the Advanced UV Spectrophotometry-CLS method is a robust and reliable analytical tool, ideal for the quality control and routine analysis of pharmaceutical products containing PA, PH, and CH in tablet dosage forms.

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