

Simultaneous Determination of Hydroquinone, Tretinoin, and Resorcinol in Cream Preparations Using the Spectrophotometry UV-Vis Development and Validation Method

Aji Putra Raharjo¹, Sukrasno Sukrasno², Winni Nur Auli^{1*}, Akmal Hammami¹

¹Department of Pharmacy, Institut Teknologi Sumatera, Terusan Ryacudu Street, Way Hui, Jati Agung, South Lampung, Lampung, 35365, Indonesia

²School of Pharmacy, Institut Teknologi Bandung, Ganesa Street No. 10, Coblong, Bandung City, West Java, 40132, Indonesia

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Correspondence:

Winni Nur Auli

winni.auli@fa.itera.ac.id



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Abstract

Background: Skin-whitening creams may contain undeclared or prohibited active ingredients, including hydroquinone, tretinoin, and resorcinol, which can pose health risks when used excessively or without medical supervision. A rapid simultaneous screening method is therefore important for routine quality control and market surveillance.

Objective: This study aimed to develop and validate simultaneous analytical methods for determining the levels of hydroquinone, tretinoin, and resorcinol in cream preparations using a UV-Vis spectrophotometer.

Methods: The wavelengths used in the UV-Vis spectrophotometric method for hydroquinone, tretinoin, and resorcinol were 294 nm, 339 nm, and 276 nm, respectively. The concentration ranges of the method were 5.0–40.0 µg/mL for hydroquinone and resorcinol, and 0.75–6.00 µg/mL for tretinoin. Validation included selectivity, linearity, accuracy, precision, limit of detection (LOD), and limit of quantification (LOQ). Three pharmaceutical creams and three cosmetic creams obtained from an e-market were analyzed.

Results: The method showed excellent linearity ($R^2 = 0.9998–0.9999$), recoveries of 99.15%–100.15%, and precision with %RSD below 2%. LOD values for hydroquinone, tretinoin, and resorcinol were 0.698, 0.078, and 0.740 µg/mL, respectively; corresponding LOQ values were 2.328, 0.261, and 2.466 µg/mL. Pharmaceutical cream results were consistent with label claims. Cosmetic cream A contained hydroquinone and tretinoin, whereas cosmetic creams B and C contained all three analytes. The measured concentrations reached 4.891% hydroquinone, 0.412% tretinoin, and 3.512% resorcinol.

Conclusions: Sample A contains hydroquinone and resorcinol, while samples B and C contain hydroquinone, tretinoin, and resorcinol, respectively, with very high levels in all types of cosmetic cream samples.

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INTRODUCTION

The rapid growth of public interest in cosmetics has led to the emergence of various types of cosmetic products. Cosmetics are formulations intended for external application to the human body, including the epidermis, hair, nails, lips, external genital organs, as well as teeth and the oral mucosa. Their primary purposes are to cleanse, provide fragrance, enhance appearance, and to protect, maintain, and preserve specific body parts in good condition.¹ In Indonesia, cosmetic products must comply with the requirements set by the Indonesian Food and Drug Authority (BPOM RI), which are verified through product registration numbers. However, cases of misuse of hazardous substances in cosmetic products are still frequently reported, such as facial whitening creams containing high concentrations of active ingredients like hydroquinone, tretinoin, and resorcinol.

Several skin-whitening products have been reported to contain active ingredients such as hydroquinone, tretinoin, and corticosteroids, which may pose serious toxic effects, particularly when used over prolonged periods.² Previous studies have investigated the presence of hazardous substances in cosmetic creams, and earlier investigations successfully quantified these active components in various cream formulations, demonstrating that both hydroquinone and tretinoin could be effectively detected through validated spectroscopic and chromatographic techniques. However, only a limited number of studies have addressed the detection of resorcinol in cosmetic or pharmaceutical creams using

UV-Vis spectrophotometric methods. Earlier research demonstrated that the simultaneous determination of hydroquinone and tretinoin along with mometasone by UV-Vis spectrophotometry could be considered relatively efficient.³

Compared to complex methods like HPLC, UV-Visible spectrophotometry is a faster, cheaper, and more eco-friendly option for routine quality testing. It requires simpler sample preparation and uses far fewer hazardous chemical solvents.⁴ This method is also highly accurate and reliable, with a strong correlation score ($r = 0.9999$) and a very low error rate.⁵ It is sensitive enough to detect harmful ingredients in complex cosmetic creams, making it ideal for checking illegal products. These results match findings from other market studies. For example, recent online marketplace tests using UV-Vis at 541 nm detected hydroquinone in 9 samples (ranging from 0.22 to 3.62 µg/ml), with an LOD of 0.637 µg/ml and an LOQ of 2.124 µg/ml.⁶

Given the increasing reports of cosmetic products containing hazardous compounds such as hydroquinone, tretinoin, and resorcinol, there is a need for an analytical method that is practical, rapid, cost-effective, and efficient for determining the concentrations of these substances. Therefore, the validation of a method for the simultaneous quantification of hydroquinone, tretinoin, and resorcinol using UV-Vis spectrophotometry is crucial, as it may serve as a reference and potential solution to this issue.

METHODS

Tools and Materials

Tools. The equipment used in this study included a UV-Visible spectrophotometer Genesys 150 (Thermo Scientific®), cuvettes, an analytical balance (Ohaus®), a sonicator, micropipettes (DRAGONLAB®), drop pipettes, spatulas, and standard glassware.

Materials. Hydroquinone (EASTMANTM®), tretinoin (TONKING®), resorcinol (SIGMA-ALDRICH®), ethanol p.a (SIGMA-ALDRICH®), pharmaceutical cream samples (Bioacne, Melanox, Reticor), cosmetic cream samples (Cosmetic A, Cosmetic B, Cosmetic C), and filter paper were used in this study.

Research Procedure

Preparation of Standard Solutions: Standard solutions of hydroquinone, tretinoin, and resorcinol were prepared at concentrations of 1000 µg/mL, 150 µg/mL, and 1000 µg/mL, respectively. Each stock solution was subsequently diluted tenfold.

Sample Preparation: A total of 50 mg of Melanox, 150 mg of Reticor, and 2 g of Bioacne were homogenized in ethanol p.a. and sonicated for 25 minutes, followed by a tenfold dilution of each sample. Similarly, 250 mg of each cosmetic sample (A, B, and C) was homogenized in ethanol p.a., subjected to sonication for 25 minutes, and then diluted tenfold.

Determination of Maximum Wavelength: Standard solutions of hydroquinone, resorcinol, and tretinoin were diluted to final concentrations of 10 µg/mL, 10 µg/mL, and 1.5 µg/mL, respectively, using ethanol p.a. The absorbance measurements were performed with a UV-Vis spectrophotometer within the wavelength range of 200–380 nm.

Selectivity: Method validation for the selectivity parameter was carried out using placebo cream solution, sample solution, individual standard solutions of each compound, and a mixed standard solution. Absorbance analysis and spectrum identification were conducted for all test materials using a UV-Vis spectrophotometer at a wavelength range of 200–380 nm.

Linearity: A series of hydroquinone and resorcinol standard concentrations of 5, 10, 20, 30, and 40 µg/mL, and tretinoin standards of 0.75, 1.5, 3.0, 4.5, and 6.0 µg/mL were prepared for linearity testing. Calibration curves were constructed from the absorbance values of each standard, which were subsequently used to determine the limits of detection (LOD) and quantification (LOQ).

Accuracy: Solutions corresponding to 80%, 100%, and 120% of the target concentrations for hydroquinone, tretinoin, and resorcinol were analyzed in triplicate using a UV-Vis spectrophotometer. The absorbance values obtained were substituted into the linear regression equation to calculate the recovery percentage.

Precision: Method validation for precision was performed to assess repeatability and intermediate precision. Repeatability was evaluated using 100% standard solutions of hydroquinone, tretinoin, and resorcinol, with six replicates each, and the relative standard deviation (%RSD) was calculated. Intermediate precision was assessed over three consecutive days, with six replicates per day, and %RSD values were determined from the daily averages.

Sample Content Determination: The prepared samples were analyzed using a UV-Vis spectrophotometric method, with each sample measured in triplicate. The absorbance values obtained were substituted into the linear regression

equations to calculate the concentrations of each compound. These concentration values were then used to determine the percentage content, which was subsequently compared with reference values.

RESULT AND DISCUSSION

Determination of Maximum Wavelength

The maximum wavelength in UV-Vis spectrophotometry is an important parameter for identifying chemical compounds. Each compound possesses a characteristic maximum wavelength, influenced by the presence of chromophore or auxochrome groups.⁷ In this study, UV-Vis absorbance measurements were performed on three different compounds, namely hydroquinone, tretinoin, and resorcinol, within the wavelength range of 200–380 nm. The results showed that these compounds exhibited maximum wavelengths of 276 nm, 294 nm, and 339 nm, respectively.^{5,6,7} The obtained values were considered valid since they corresponded with the theoretical maxima for each active compound.

Selectivity Assay

Selectivity of the method was assessed through several comparisons: between the spectrum of the mixed standard solution and the placebo, the mixed standard and each individual standard compound, the mixed standard and pharmaceutical cream samples, as well as the mixed standard and cosmetic cream samples. The purpose of comparing the mixed standard with the placebo cream was to evaluate whether the placebo produced spectra similar to the active compounds.

The placebo samples were prepared using the same procedure as cosmetic sample preparation. The diluted filtrate of the placebo was then analyzed using a UV-Vis spectrophotometer. The results indicated that the spectra of the mixed active compounds showed no overlap with the placebo cream filtrate. Thus, the mixed standard solution was confirmed to be selective for resorcinol, hydroquinone, and tretinoin at their respective maximum wavelengths of 276 nm, 294 nm, and 339 nm.^{5,6,7}

Linearity Assay

Linearity was evaluated by measuring the absorbance of a series of mixed standard solutions containing hydroquinone, tretinoin, and resorcinol. The test was conducted by measuring absorbance values of three replicates of hc (5, 10, 20, 30, and 40 µg/mL), tretinoin (0.75, 1.5, 3.0, 4.5, and 6.0 µg/mL), and resorcinol (5, 10, 20, 30, and 40 µg/mL). Measurements were performed in triplicate at 294 nm for hydroquinone, 276 nm for resorcinol, and 339 nm for tretinoin. The obtained spectra (**Table 1**) demonstrated a clear compliance with Beer–Lambert's law, which states that absorbance increases proportionally with analyte concentration.⁸

Table 1. Linearity Test Data for Calibration Curve Construction by Comparing Analyte Concentration with Absorbance

Resorcinol				Hydroquinone				Tretinoin			
Conc. (µg/mL)	Abs.	Average	% RSD	Conc. (µg/mL)	Abs.	Average	% RSD	Conc. (µg/mL)	Abs.	Average	% RSD
5	0.219	0.2163	1.163	5	0.199	0.1957	1.561	0.75	0.131	0.1287	1.956
	0.216				0.195				0.129		
	0.214				0.193				0.126		
10	0.376	0.3777	0.404	10	0.356	0.3543	0.431	1.5	0.228	0.2273	0.916
	0.378				0.354				0.225		
	0.379				0.353				0.229		
20	0.721	0.7203	0.289	20	0.688	0.6900	0.383	3	0.438	0.4380	0.457
	0.718				0.693				0.436		
	0.722				0.689				0.440		
30	1.064	1.0633	0.109	30	1.019	1.0177	0.113	4.5	0.646	0.6463	0.389
	1.062				1.017				0.644		
	1.064				1.017				0.649		
40	1.426	1.4233	0.177	40	1.368	1.3687	0.152	6	0.863	0.8620	0.201
	1.421				1.371				0.863		
	1.423				1.367				0.860		

Abbreviation: Conc = concentration; Abs = absorbance.

The calibration curves, constructed by plotting analyte concentrations against mean absorbance values, yielded highly linear regression models of $y = 0.0345x + 0.0360$ ($R^2 = 0.9998$) for resorcinol, $y = 0.0335x + 0.0224$ ($R^2 = 0.9998$)

for hydroquinone, and $y = 0.1398x + 0.0200$ ($R^2 = 0.9999$) for tretinoin. These correlation coefficients ($R^2 \geq 0.9998$) confirm an excellent linear relationship across the specified concentration ranges for all three compounds.

According to the Indonesian Pharmacopoeia, 6th Edition, the acceptance criterion for linearity requires an R^2 value greater than 0.98.⁹ Based on the linearity test results for the three active compounds resorcinol, hydroquinone, and tretinoin the R^2 values obtained were 0.9998, 0.9998, and 0.9999, respectively. These results indicate that the applied method fully complies with the linearity validation criteria. These correlation coefficients ($R^2 \geq 0.9998$) signify an exceptionally robust proportional relationship between analyte concentration and instrument response. This strong linear relationship shows that the target molecules absorb light predictably, without getting disrupted by other substances. These regressions can be trusted to accurately measure the exact amount of each substance.¹³ The absorbance values measured for each analyte were inserted into the respective regression equations of each compound.

LOD and LOQ Assay

The Limit of Detection (LOD) test is intended to determine the lowest concentration of an analyte that can still be detected, though not necessarily quantified, under the given experimental conditions.¹³ In contrast, the Limit of Quantitation (LOQ) refers to the lowest concentration of an analyte that can be quantified reliably within specific experimental conditions, based on accuracy and precision parameters.¹⁴ The calculated LOD and LOQ values obtained in this study demonstrate the exceptional sensitivity and regulatory utility of the developed multi-component method. Consequently, the concentration used for linearity, accuracy, and precision tests must not fall below the established LOQ values. Based on the experimental data, the calculated LOD and LOQ values were found to be below the lowest concentrations used in the linearity tests. The LOD values obtained for resorcinol, hydroquinone, and tretinoin were 0.740 $\mu\text{g/mL}$, 0.698 $\mu\text{g/mL}$, and 0.078 $\mu\text{g/mL}$, respectively. Meanwhile, the LOQ values were 2.466 $\mu\text{g/mL}$, 2.328 $\mu\text{g/mL}$, and 0.261 $\mu\text{g/mL}$, respectively. This method is so sensitive it can spot even low traces of tretinoin. Because of this, it easily finds hidden contamination, even in unevenly mixed creams.

Accuracy Assay

Accuracy testing was carried out as part of the method validation process in accordance with the Indonesian Pharmacopoeia, 6th Edition, with the objective of assessing how closely the analytical results approach the true or reference values.¹⁵ Accuracy is expressed as the percentage of recovery (% recovery); the smaller the deviation, the closer the recovery value is to the true result, ideally approaching 100%.¹⁶ The AOAC Guidelines state that the lower the analyte concentration in a sample, the wider the acceptable recovery range becomes.¹⁷

In addition, the accuracy study also considers the %RSD values from replicate measurements to ensure that repeatability precision remains within the acceptable limit ($<2\%$).¹⁸ According to the literature, the acceptable range for recovery values that meet the criteria is 95% to 105%.¹⁹ The results of this study showed recovery values ranging from 99.15% to 100.15%. These findings demonstrate that the accuracy validation complied with the acceptance criteria stated in the Indonesian Pharmacopoeia, 6th Edition. Recovery rates near 100% show the test safely collects all three ingredients from the cream, proving the method is accurate and error-free. The results of the accuracy test are presented in **Table 2**.

Table 2. The accuracy data were used to ensure that the precision of replication complied with the established requirements.

Concentration	Compound	R	Absorbance	C ($\mu\text{g/mL}$)	Concentration (%)	Average C ($\mu\text{g/mL}$)	% Recovery	% RSD
80%	Rs (8.0 $\mu\text{g/mL}$)	1	0.312	8.000	80.00	7.9420	99.28	0.632
		2	0.309	7.913	79.13			
		3	0.309	7.913	79.13			
	Hq (8.0 $\mu\text{g/mL}$)	1	0.292	8.048	80.48	7.9881	99.85	0.647
		2	0.289	7.958	79.58			
		3	0.289	7.958	79.58			
	Tn (1.2 $\mu\text{g/mL}$)	1	0.187	1.195	79.64	1.1898	99.15	0.694
		2	0.185	1.180	78.68			
		3	0.187	1.195	79.64			
100%	Rs (10.0 $\mu\text{g/mL}$)	1	0.380	9.971	99.71	9.9807	99.81	0.731
		2	0.378	9.913	99.13			
		3	0.383	10.058	100.58			

Concentration	Compound	R	Absorbance	C (µg/ml)	Concentration (%)	Average C (µg/ml)	% Recovery	% RSD
120%	Hq (10.0 µg/ml)	1	0.356	9.958	99.58	9.9682	99.68	0.754
		2	0.354	9.899	98.99			
		3	0.359	10.048	100.48			
	Tn (1.5 µg/ml)	1	0.229	1.495	99.67	1.4974	99.83	1.678
		2	0.226	1.474	98.24			
		3	0.233	1.524	101.57			
	Rs (12.0 µg/ml)	1	0.449	11.971	119.71	11.9710	99.76	0.484
		2	0.451	12.029	120.29			
		3	0.447	11.913	119.13			
	Hq (12.0 µg/ml)	1	0.424	11.988	119.88	12.0179	100.15	1.138
		2	0.430	12.167	121.67			
		3	0.421	11.899	118.99			
	Tn (1.8 µg/ml)	1	0.272	1.803	120.17	1.7954	99.75	0.690
		2	0.272	1.803	120.17			
		3	0.269	1.781	118.74			

Abbreviation: Rs = resorcinol; Hq = hydroquinone; Tn = tretinoin; R = replication; C = content.

Precision Assay

The precision test was performed to assess the consistency of repeated experiments, both on the same day (repeatability) and across different days (intermediate precision).²⁰ This parameter reflects the degree of accuracy with which an experiment can reproducibly generate sample data, and the acceptance criterion is expressed as the coefficient of variation or relative standard deviation (%RSD). In general, for precision testing, the %RSD value between two sets of data should be $\leq 2.0\%$. However, acceptance limits for impurities may vary depending on their concentration levels and the sensitivity of the analytical method employed.^{21, 22}

Table 3. The repeatability precision data were obtained by comparing the measurement and calculation results performed on the same day.

Standard	Replication	Absorbance	Content (µg/ml)	Concentration (%)	%RSD
Resorcinol (10 µg/ml)	1	0.381	10.00	100.0	0.552
	2	0.380	9.97	99.7	
	3	0.383	10.06	100.6	
	4	0.380	9.97	99.7	
	5	0.378	9.91	99.1	
	6	0.378	9.91	99.1	
Hydroquinone (10 µg/ml)	1	0.358	10.02	100.2	0.480
	2	0.355	9.93	99.3	
	3	0.358	10.02	100.2	
	4	0.356	9.96	99.6	
	5	0.356	9.96	99.6	
	6	0.354	9.90	99.0	
Tretinoin (1.5 µg/ml)	1	0.229	1.49	99.7	0.718
	2	0.229	1.49	99.7	
	3	0.231	1.51	100.6	
	4	0.229	1.49	99.7	
	5	0.232	1.52	101.1	
	6	0.228	1.49	99.2	

In this study, measurements and calculations for repeatability and intermediate precision were performed over three consecutive days. Repeatability was assessed by conducting six replicate measurements on the same day, and the results are presented in **Table 3**. Based on the data in **Table 3**, all %RSD values obtained were below 2%, with the highest value recorded at 0.718%. These findings indicate that the repeatability test satisfies the validation criteria.

Intermediate precision was evaluated by performing six replicate measurements across three different days, with the results summarized in **Table 4**. The data showed that all %RSD values were also below 2%, with the highest value being 0.532%. Therefore, the intermediate precision test was confirmed to meet the validation requirements. RSD values below 0.75% show that the equipment, structure, and process remain extremely stable over time. This low variation proves that small daily changes, like slight room temperature shifts or different analysts handling the test won't affect the accuracy of the results. As a result, this method is highly reliable for routine quality control testing.

Table 4. The data for intermediate precision were obtained by comparing the measurement and calculation results performed on different days.

Standard	Day	Average Absorbance	Content (µg/ml)	Concentration (%)	%RSD
Resorcinol (10 µg/ml)	1	0.380	9.971	99.71	0.201
	2	0.380	9.957	99.57	
	3	0.379	9.928	99.28	
Hydroquinone (10 µg/ml)	1	0.356	9.963	99.63	0.221
	2	0.358	10.008	100.08	
	3	0.357	9.998	99.98	
Tretinoin (1.5 µg/ml)	1	0.230	1.500	99.98	0.532
	2	0.232	1.516	101.09	
	3	0.232	1.513	100.86	

Determination of Sample Content

The concentration values were determined by measuring the absorbance of each sample using a UV-Vis spectrophotometer.^{23, 24} The samples were classified into two categories: pharmaceutical cream samples containing a single active compound (hydroquinone, resorcinol, or tretinoin) and cosmetic cream samples with unknown active ingredients.⁸

For the pharmaceutical cream formulations, the active compound content was stated on the product labels. The samples analyzed included Bioacne cream, Melanox cream, and Reticor cream. According to the product label, Bioacne cream contains 5 mg of resorcinol in 10 g of cream preparation, equivalent to 0.05%. Melanox cream is labeled to contain 2% hydroquinone, whereas Reticor cream contains 0.1% tretinoin. After sample preparation, UV-Vis absorbance measurements were performed, and the results are presented in **Table 5**.

Table 5. Data on Active Compound Determination in Cream Samples

Sample	Absorbance	Content (µg/ml)	Average Content (µg/ml)	Concentration (%)	Conclusion
Bioacne	0.379	9.942	9.932	99.32	Compatible to the label
	0.378	9.913			
	0.379	9.942			
Melanox	0.356	9.958	10.008	100.08	Compatible to the label
	0.359	10.048			
	0.358	10.018			
Reticor	0.228	1.488	1.490	99.33	Compatible to the label
	0.228	1.488			
	0.229	1.495			

Based on **Table 5**, the results indicate that the active ingredient content in the tested medicinal cream formulations, as determined by UV-Vis spectrophotometry, was consistent with the label claims, namely 99.32% for Bioacne cream, 100.08% for Melanox cream, and 99.33% for Reticor cream. According to the Indonesian Pharmacopoeia, Sixth Edition, hydroquinone cream preparations must contain 94–106% of the hydroquinone content stated on the label. The same reference also specifies that tretinoin cream preparations should contain 90–120% of the labeled tretinoin content.⁹ Meanwhile, the specification for resorcinol content is based on the United States Pharmacopeia (USP), 44th Edition, which allows a range of 90–110%.²⁴ Therefore, it can be concluded that the determination of active ingredient levels in the tested medicinal cream formulations complies with the requirements outlined in the Indonesian Pharmacopoeia, Sixth Edition.

The testing results of the three cosmetic cream samples are presented in **Table 6**. The analysis indicated that these creams contained combinations of active ingredients that contributed to the effectiveness of their formulations. In addition, the concentrations of the active compounds in the products were found to be considerably high. The identity of each analyte was verified via $\lambda_{\max}$ comparison with pure standards; and among the samples, Cosmetic Creams B, A, and C exhibited the highest concentrations of resorcinol, hydroquinone, and tretinoin, respectively.

According to the Indonesian National Agency of Drug and Food Control (BPOM), the distribution and unrestricted sale of cosmetic whitening creams containing hydroquinone are prohibited. The use of hydroquinone in cosmetic formulations is only allowed under the recommendation and supervision of dermatologists, with a maximum concentration of 2%.⁹ Similarly, tretinoin is permitted in cosmetic cream formulations at a concentration of 0.1%, and its application must also be under medical prescription and supervision. Hydroquinone concentrations in the range of 1.5–2% are generally recognized as safe and effective (GRASE) and have been proposed for use as an active ingredient

in over-the-counter (OTC) skin-lightening products.²⁵⁻²⁷ In contrast, BPOM regulations explicitly prohibit the use of resorcinol in cosmetic formulations.

Table 6. Determination of Active Ingredient Content in Cosmetic Cream Samples

Sample	Absorbance	Content (µg/ml)	Average Content (µg/ml)	Concentration (%)	Data Interpretation
Cosmetic A	λ 294 nm	1.242	36.406	4.891	Contains a prohibited substance at a high concentration
		1.255	36.794		
		1.257	36.854		
	λ 339 nm	0.262	1.731	0.233	Contains a prohibited substance at a high concentration
		0.264	1.745		
		0.266	1.760		
Cosmetic B	λ 276 nm	0.941	26.232	3.512	Contains a prohibited substance at a high concentration
		0.941	26.232		
		0.952	26.551		
	λ 294 nm	0.860	25.003	3.350	Contains a prohibited substance at a high concentration
		0.861	25.033		
		0.871	25.331		
	λ 339 nm	0.367	2.482	0.332	Contains a prohibited substance at a high concentration
		0.366	2.475		
		0.371	2.511		
Cosmetic C	λ 276 nm	0.794	21.971	2.920	Contains a prohibited substance at a high concentration
		0.791	21.884		
		0.790	21.855		
	λ 294 nm	0.716	20.704	2.753	Contains a prohibited substance at a high concentration
		0.714	20.645		
		0.712	20.585		
	λ 339 nm	0.453	3.097	0.412	Contains a prohibited substance at a high concentration
		0.453	3.097		
		0.449	3.069		

The quantitative determination of each cosmetic cream sample, as shown in **Table 6**, demonstrated that the concentrations of active compounds were extremely high, exceeding the limits recommended for medical supervision. Specifically, Cosmetic Cream A contained 4.8913% hydroquinone and 0.2327% tretinoin. Cosmetic Cream B contained 3.512% resorcinol, 3.350% hydroquinone, and 0.332% tretinoin. Meanwhile, Cosmetic Cream C contained 2.920% resorcinol, 2.753% hydroquinone, and 0.412% tretinoin. From a health and safety perspective, using products with high levels of hydroquinone (up to 4.89%) and tretinoin (up to 0.41%) seriously increases the risk of permanent skin discoloration, lasting redness, severe skin thinning, and internal toxicity. Additionally, adding resorcinol (up to 3.51%) is a major health hazard because it can disrupt thyroid function and trigger severe allergic skin reactions.¹

All three cosmetic creams were found to contain active substances that are prohibited by BPOM for use and distribution in cosmetic whitening creams due to potential health risks to consumers. These products are nevertheless widely available through e-commerce platforms. In fact, they are distributed and sold freely without any labeling that discloses their active ingredients or concentrations. Similar products are abundant in the market, particularly on online trading platforms. The high consumer demand for such cosmetic creams is primarily driven by their easy accessibility without a prescription, relatively low prices, and the rapid whitening effect they provide. These quantitative findings strongly mirror recent marketplace literature, which reported a high prevalence of heavily adulterated whitening formulations across online e-commerce platforms containing hydroquinone concentrations from 0.22 to 3.62 µg/ml. Notably, the percentage levels identified in this study (up to 4.8913%, equivalent to 48,913 µg/ml) drastically surpass standard marketplace surveillance data, highlighting an aggressive trend in illicit cosmetic compounding. Because these unlabeled black-market creams are typically manufactured without standard quality control protocols, they are highly prone to poor homogenization. The high precision and low detection limits (LOD and LOQ) of this UV-Vis method make it a cost-effective tool for regulatory screening. It enables the rapid detection and quantification of hazardous multi-component mixtures, providing a reliable approach to safeguard public health against unsafe cosmetic exposure.

Limitation

This study presents several limitations that should be taken into consideration. The analytical method developed in this research was validated only using UV-Vis spectrophotometry, in which the identification and quantification of hydroquinone, tretinoin, and resorcinol were primarily based on absorbance values and maximum

wavelength characteristics. Therefore, the method has not yet been compared with more advanced confirmatory analytical techniques such as HPLC or LC-MS, which may provide higher specificity and sensitivity. In addition, the study employed a limited number of samples, consisting of only three medicinal cream samples and three cosmetic cream samples obtained from e-market, which may not fully represent the wide variety of cosmetic products circulating in the market. Furthermore, the validation parameters evaluated in this study were restricted to selectivity, linearity, accuracy, precision, limit of detection (LOD), and limit of quantification (LOQ), while other important validation aspects such as robustness and ruggedness were not investigated. Moreover, further studies involving broader sample variations, additional validation parameters, and comparative analyses using more advanced analytical instruments are necessary to strengthen the reliability and applicability of the developed method. Comparing our results to recent studies shows that our method performs just as well as existing ones. Our linearity matches the near-perfect scores ($r = 0.9999$) and low error rates (1-3%) reported by Kurniawan et al. (2022).⁴ Additionally, our method easily handles complex cream bases, and its detection limits compare well with recent findings that successfully found target ingredients in real marketplace samples at low concentration levels.⁶

CONCLUSION

The validation of the UV-Vis spectrophotometric method for the simultaneous determination of hydroquinone, tretinoin, and resorcinol in cream formulations demonstrated adequate selectivity. The quantification of concentrations in the three cream samples Bioacne, Melanox, and Reticor was consistent with the specifications stated on their respective labels. Nevertheless, this method requires further refinement by expanding the number of tested samples, particularly cosmetic whitening creams. Therefore, additional studies are recommended to comprehensively establish the overall validity and reliability of this analytical method.

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GENERATIVE AI DISCLOSURE STATEMENT

Generative AI assistance (GPT; OpenAI, 2026) was used to English paraphrasing of this paper. All AI-assisted drafts were critically evaluated and corrected by the authors. The researchers hold total responsibility for the integrity of the data and the conclusions presented herein.

AUTHOR CONTRIBUTION STATEMENT

Aji Putra Raharjo: Conceptualization Methodology, Investigation, Visualization, Software, Validation, Writing - Original Draft; **Sukrasno Sukrasno**: Supervision; **Winni Nur Auli**: Supervision, Conceptualization, Writing - Original Draft, Writing - Review & Editing; **Akmal Hammami**: Writing - Original Draft, Writing - Review & Editing.

CONFLICT OF INTEREST DECLARATION

The authors declare that there are no conflicts of interest among the authors or with any external parties in the preparation of this manuscript.

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