Study on the determination of pharmaceutical products containing amoxicillin and clavulanic acid by HPLC/DAD method

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ABSTRACT

Amoxicillin, a β-lactam antibiotic, is commonly combined with clavulanic acid to extend its antibacterial spectrum. Dissolution testing of pharmaceutical formulations containing these two substances has seldom been reported, largely due to the chemical instability of clavulanic acid and the analytical challenges of simultaneously quantifying both components. This lack of standardized methodology represents a significant research gap in the quality control of Amoxicillin/Clavulanate products. The objective of this study was to develop and validate a robust HPLC/DAD method for dissolution testing of the commercial formulation Vigentin. Dissolution was performed in ultrapure RO water (resistivity 18.2 MΩ·cm at 25 °C) under tightly controlled temperature and stirring speed. Chromatographic analysis employed a C18 column, a sodium dihydrogen phosphate/methanol (95:5, v/v) mobile phase, and UV detection at 220 nm. The validated procedure met all requirements for specificity, repeatability, and system suitability, with excellent linearity ($R^2 > 0.9998$). Dissolution quantification of six samples showed amoxicillin release between 93.9% and 98.9%, and potassium clavulanate between 103% and 107%, all above the minimum acceptance criterion of 75%. These results demonstrate that the proposed method effectively fills the methodological gap and provides a feasible, reliable tool for routine pharmaceutical quality evaluation of this clinically important drug combination.

Keywords: Amoxicillin; Clavulanate potassium; Solubility; Liquid chromatography (HPLC); Drug quality control.

1. INTRODUCTION

Amoxicillin, a β-lactam antibiotic, is often formulated in combination with other compounds to broaden its antibacterial spectrum and overcome resistance. Clavulanic acid, a β-lactamase inhibitor, is the most widely used partner, and together they form a fixed-dose combination that is highly effective against resistant bacterial strains. Despite its clinical importance, the simultaneous dissolution testing of Amoxicillin and Clavulanic Acid has not been standardized in major pharmacopoeias, highlighting a significant analytical gap. Several studies have previously addressed the simultaneous determination of amoxicillin with other co-formulated agents. For instance, Becze et al. (2022) [1] developed and validated a multichannel HPLC-DAD method for the simultaneous determination of amoxicillin and doxycycline in pharmaceutical formulations and wastewater samples, achieving high reproducibility. Razuc et al. (2021) [2] established a simple and reliable HPLC method for quantifying amoxicillin and sulbactam pivoxil in both assay and dissolution samples, demonstrating its potential for routine quality control. De Marco et al. (2017) [3] provided a comprehensive review on the properties and analytical approaches for amoxicillin, emphasizing green chemistry trends. Tsou et al. (1997) [8] reported a rapid HPLC method using a β-cyclodextrin column for simultaneous assay of amoxicillin and clavulanic acid, but their study focused on assay recovery rather than dissolution profiling. Similarly, Foulstone and Reading (1982) [7] quantified these two compounds in biological fluids for pharmacokinetic assessment, without evaluating dissolution or impurity detection. More recently, Dhull et al. (2025) [4] and others [5, 6] have highlighted advances in

liquid chromatography for antibiotics, but none addressed standardized dissolution testing for the amoxicillin–clavulanic acid combination. In Vietnam, some efforts have been reported, such as Nguyen and Tao (2023) [9], who developed a UV-Vis spectrophotometric method for the simultaneous quantification of amoxicillin and clavulanic acid. Other groups have used HPLC or spectrophotometric approaches for assay or dissolution of these drugs [10, 11]. However, these methods often lack full validation or do not meet pharmacopoeial requirements for dissolution testing, limiting their applicability in regulatory quality control. Taken together, the literature demonstrates that while simultaneous quantification of amoxicillin with other β-lactam inhibitors (such as sulbactam) has been explored, there is still no robust, validated, and pharmacopoeia-compliant HPLC-DAD method for evaluating the dissolution of amoxicillin–clavulanic acid tablets. This is of particular importance because clavulanic acid is chemically less stable than other inhibitors, making its quantification in dissolution tests more challenging yet more clinically relevant.

The novelty of this study lies in uniting comprehensive method validation, pharmacopoeia-compliant dissolution testing, and impurity profiling into a single workflow. By applying this approach to commercial Vigentin tablets, we aim to establish a reliable and practical procedure for routine quality control of amoxicillin–clavulanic acid formulations, thereby addressing an urgent analytical need in both global and local contexts.

2. RESEARCH OBJECTIVES AND MATERIALS

2.1. Research objective

Establishing a procedure for determining the dissolution of Amoxicillin/Clavulanic Acid (figure 1a and figure 1b) combination drugs using HPLC/DAD is essential in pharmaceutical research and production. Developing such a method is crucial for evaluating drug release in simulated biological environments, which contributes to assessing bioavailability, therapeutic efficacy, and product quality assurance.

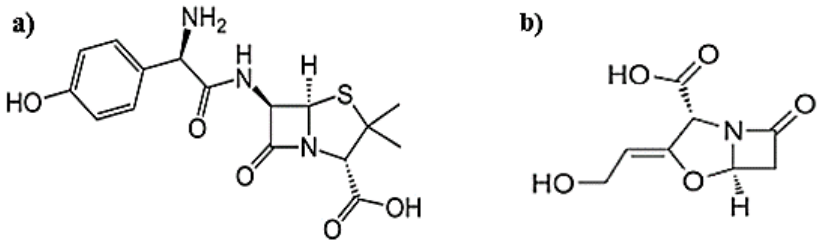

Figure 1. *Chemical structure of a) Amoxicillin, b) Acid Clavulanic.*

2.2. Equipment, reagents, and standards

a) Dissolution conditions

The dissolution medium consisted of 900 mL of ultrapure RO water (Resistivity 18.2 MΩ·cm at 25 °C) [12]. The dissolution temperature was maintained at 37 ± 0.5 °C. The apparatus used was a paddle-type dissolution tester operating at a rotation speed of 75 rpm for a total duration of 30 minutes. All parameters were programmed and controlled via the Veego dissolution testing system.

The dissolution medium consisted of 900 mL of ultrapure RO water (resistivity 18.2 MΩ·cm at 25 °C) [12]. While the United States Pharmacopeia (USP) recommends phosphate buffer media for dissolution testing of amoxicillin/clavulanate tablets [13], ultrapure RO water was selected in this study to minimize matrix interference and enhance the stability of the analytes. The dissolution temperature was maintained at 37 ± 0.5 °C, using a paddle-type dissolution tester operated at 75 rpm for 30 minutes. All parameters were programmed and controlled via the Veego dissolution testing system.

Table 1. *Equipment and supplies.*

No	Equipment and Supplies	Brand/ Model	Origin/Specifications
1	Analytical balance	Aczet / CY 224	India; Capacity 220 g, Readability 0.1 mg
2	Weighing paper, filter paper	Whatman (GE Healthcare)	UK/ Grade 1 (11 μm)
3	Membrane filter, vial	Agilent Technologies/ Nylon/PVDF	USA/0.45 μm/2 mL
4	Volumetric flask	ISOLAB/ Class A, Borosilicate glass	Germany/50 mL, 100 mL
5	Bulb pipette	ISOLAB/ Class A, Glass	Germany/ 10 mL
6	High Performance Liquid Chromatography system	Waters Corporation/ Acquity Arc Sample Manager FTN-R	USA
7	Dissolution tester	Veego/ VDA 14D, 6 vessels	India
8	Ultrapure water purification system	Merck Millipore/ Milli-Q Direct 8	Germany; Resistivity 18.2 MΩ·cm
9	pH meter (Accuracy ±0.002 pH)	Metrohm/ 913 pH Meter	Switzerland
10	Ultrasonic machine	Elma/ Elmasonic S	Germany; F: 37 kHz
11	Amoxicillin standard	National Institute of Drug Quality Control (NIDQC)	Vietnam/(86.9%)
12	Clavulanic acid standard	National Institute of Drug Quality Control (NIDQC)	Vietnam/(42.07%)
13	Finished product: Vigentin	Pharbao/Tablets (AMX + CLA)	Vietnam/(VD-22223-15)
14	Mobile phase solution: Methanol	Merck/ HPLC grade	Germany
15	Mobile phase solution: Sodium dihydrogen phosphate dihydrate	Supelco (Merck)	Germany

b) Chromatographic conditions

The analysis was performed using a High-Performance Liquid Chromatography system equipped with a Diode Array Detector (HPLC/DAD). Detection was carried out at a wavelength of 220 nm under isocratic elution conditions. The mobile phase consisted of sodium dihydrogen phosphate dihydrate/methanol in a 95:5 (v/v) ratio, with a flow rate of 1.0 mL/min. The injection volume was 20 μL, and the chromatographic column used was a reversed-phase C18 column (250 mm × 4.6 mm, 5 μm).

c) Sample preparation method

Amoxicillin reference standard (purity 86.9%), used for standard solution preparation. Potassium Clavulanate Reference Standard (purity 42.07%), used for combination standard with Amoxicillin. Vigentin tablets containing 500 mg Amoxicillin and 62.5 mg Potassium Clavulanate per tablet; used for test solution preparation; RO Water (Reverse Osmosis Water): High-purity water used as solvent for solution preparation and dilution. Sodium Dihydrogen Phosphate Dihydrate ($NaH_2PO_4 \cdot 2H_2O$): Used to prepare phosphate buffer (pH adjusted to 4.4) for mobile phase composition.

Methanol (HPLC grade) is mixed with phosphate buffer in a 95:5 (v/v) ratio to prepare the mobile phase. Membrane Filter (PTFE or Nylon) 0.45 µm is used to filt both standard and test solutions before HPLC injection to separate particulates.

d) Expression for dissolution quantification

The dissolution content was calculated using the following equation [14]:

$$m_t = \frac{S_t \,.m_c \,.HL_c \,.V_t \,.D_c}{S_c \,.V_c \,.D_t}$$

Where: *mt* is the amount of Amoxicillin dissolved in the test solution after dissolution (mg); *mc* is the mass of the standard substance weighed (mg); *St* and *Sc* are the peak areas of the test and standard solutions, respectively; *HLc* is the labeled content of Amoxicillin in the standard (%); *Vt* and *Vc* are the dissolution media volumes of the test and standard solutions (mL); *Dt* and *Dc* are the dilution factors of the test and standard solutions, respectively.

This formula was applied to quantify the in vitro dissolution of Amoxicillin and Potassium Clavulanate, accounting for purity, dilution, and volume factors in both standard and test solutions.

e) Experimental procedure diagram

The quality control testing of Vigentin tablets (Batch No. 250210) was performed using high-performance ~~High-Performance Liquid Chromatography~~ (HPLC), following detailed steps (1) through (9), as illustrated in figure 2. Dissolution testing was conducted using six tablets in 900 mL of ultrapure water maintained at 37 ± 0.5 °C and stirred at 75 rpm for 30 minutes. The samples were analyzed using a U-HPLC system equipped with a UV detector set at 220 nm. Chromatographic separation was achieved under isocratic conditions with a flow rate of 2 mL/min, using a C18 column (25 cm × 4.6 mm, 5 µm) and an injection volume of 20 µL. The standard solution was injected in triplicate, while the test solutions were analyzed in sextuplicate.

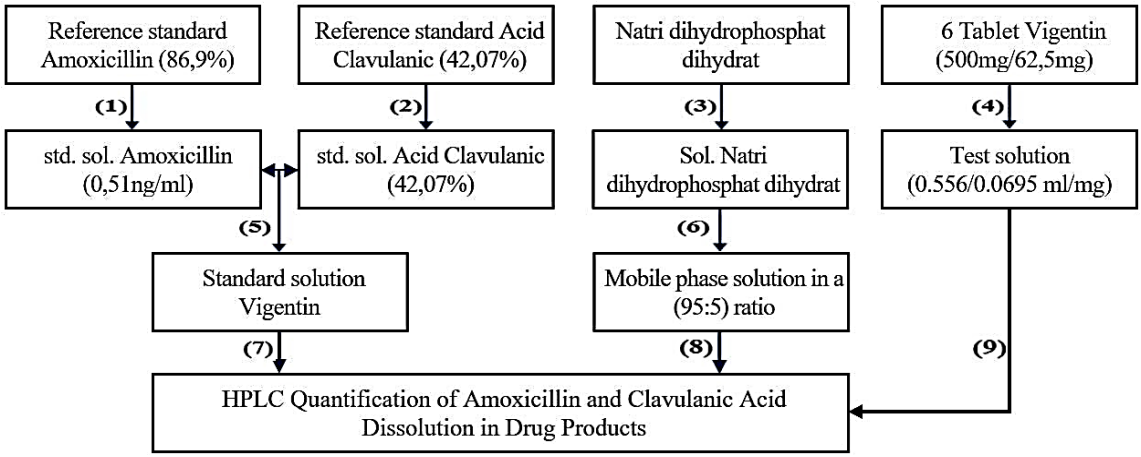

Figure 2. *Diagram of the dissolution test procedure for Vigentin.*

- Sample and Standard Solution Preparation for HPLC Analysis:

(1) Preparation of Amoxicillin standard solution: Accurately weigh 58.6 mg of Amoxicillin (86.9% assay) into a 100 mL volumetric flask. Dilute to volume with ultrapure water to obtain a 0.51 mg/mL solution.

(2) Preparation of Clavulanic acid standard solution: Transfer 10 mL of a 42.07% Clavulanic Acid solution into the flask of step (1). Filter through a 0.45 µm membrane to yield a 0.065 mg/mL Clavulanic Acid concentration.

(3) Preparation of buffer solution: Dissolve 19.86 g of sodium dihydrogen phosphate dihydrate in 2250 mL of ultrapure water. Adjust pH to 4.4 to obtain phosphate buffer.

(4) Preparation of test solution: Dissolve six Vigentin tablets (each containing 500 mg Amoxicillin and 62.5 mg Clavulanic acid) in 250 mL of mobile phase (buffer + methanol). Filter through a 0.45 µm membrane. Final concentrations: 0.556 mg/mL (Amoxicillin) and 0.0695 mg/mL (Clavulanic acid).

(5) Combination of standards: Use the mixed standard solution from steps (1) and (2) as the reference standard.

(6) Preparation of mobile phase: Mix the phosphate buffer (step 3) with methanol in a 95:5 ratio.

(7) Finalization of standard solution: The mixture in step (5) serves as the finalized reference standard.

(8) Finalization of mobile phase: Use the mobile phase from step (6) for analysis.

(9) HPLC Injection: Inject both standard (step 7) and test solution (step 4) into the HPLC system for dissolution quantification. All procedures followed pharmacopeial standards [14].

3. CALCULATION AND DISCUSSION

3.1. Method validation

3.1.1. Specificity of the method

The chromatograms of individual components yielded retention times of 7.147 minutes for Amoxicillin and 4.277 minutes for Potassium Clavulanate. Analysis of the chromatogram from the placebo sample (figure 3a) confirmed the presence of Amoxicillin by comparison with the standard chromatograms (figure 3b and figure 3c). Additionally, several minor peaks appeared between 0.5 and 5.5 minutes, indicating the presence of impurities, excipients, or matrix components in the real sample. The appearance of these minor peaks suggests possible degradation products or excipient interferences. Nevertheless, the Amoxicillin peak remained well-separated and unoverlapped, thereby ensuring accurate quantification and confirming the method's specificity. In the chromatogram shown in figure 3b, the standard mixture spectrum displays two primary peaks: Potassium Clavulanate with a retention time of approximately 4.298 minutes and Amoxicillin at 7.166 minutes. These peaks are symmetrical, sharp, and well-resolved, indicating that the chromatographic conditions employed are appropriate for the simultaneous analysis of both active compounds. This clear separation is essential to ensure the method's selectivity and quantification accuracy.

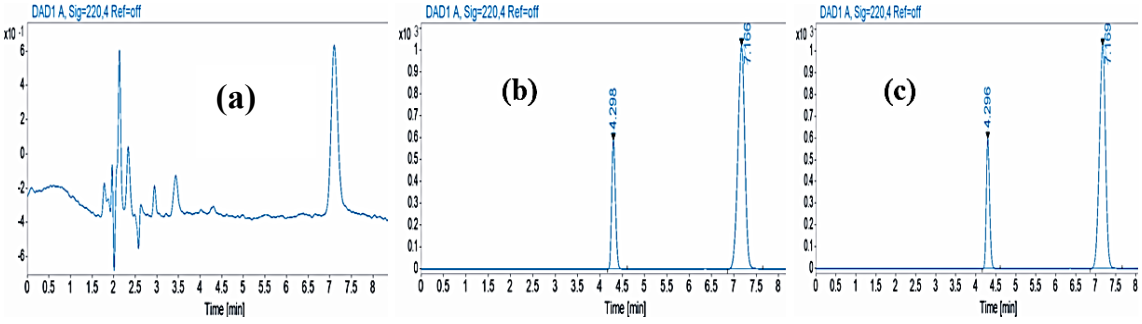

Figure 3. *(a) Chromatogram of the test sample containing Amoxicillin, (b) chromatogram of the standard mixture of Potassium Clavulanate and Amoxicillin, and (c) chromatogram of the individual standard solutions of Amoxicillin and Potassium Clavulanate.*

Figure 3c - The chromatogram of the individual standards of Amoxicillin and Potassium Clavulanate - further confirms the specific retention times for each compound: 4.296 minutes for Potassium Clavulanate and 7.169 minutes for Amoxicillin. Compared to the mixed standard, the peak shapes and positions remain consistent, demonstrating the high stability and reproducibility of the analytical method (see table 2).

Table 2. *Summary of retention times for the compounds.*

Solution	$T_{Kali\ clavulanat}$ (min)	$T_{Amoxicillin}$ (min)
Reference sample	4.296	7.196
Sample under analysis	4.298	7.166
Placebo	-	7.012

Observation: The chromatographic method successfully achieved clear separation between Amoxicillin and Potassium Clavulanate. The retention time of Amoxicillin in the test sample matches that of the reference standard, confirming the presence of the active ingredient. Signal interferences in the test sample did not affect the quantification of Amoxicillin. The consistency and stability of retention times across different chromatograms validate the accuracy and repeatability of the method.

3.1.2. System suitability

System suitability of the HPLC-DAD system was evaluated by performing six replicate injections of the previously prepared standard solution and recording the chromatograms. The system suitability results are summarized in table 3.

Resolution (R_s) is a critical parameter in HPLC that quantifies the degree of separation between two adjacent chromatographic peaks. It is defined based on the retention times and peak widths of the analytes, and is mathematically expressed as:

$$R_s = \frac{2(t_{R1} - t_{R2})}{w_1 + w_2}$$

where t_{R1} and t_{R2} are the retention times and w_1, w_2 are the baseline peak widths of the first and second eluting compounds, respectively. A resolution value greater than 1.5 typically indicates baseline separation, essential for accurate qualitative and quantitative analysis. Resolution is influenced by multiple factors, including column efficiency, selectivity, and retention, and optimizing these parameters is crucial for method development and validation in analytical HPLC.

Table 3. *System suitability results.*

№	$T_{Kali\ clavulanat}$ (min)	$S_{Kali\ clavulanat}$ (mAU.s)	$T_{Amoxicillin}$ (min)	$S_{Amoxicillin}$ (mAU.s)	Resolution
1	4.280	3502.52	7.179	11092.05	13.20
2	4.280	3501.79	7.178	11088.60	13.23
3	4.281	3500.28	7.177	11089.27	13.22
4	4.281	3500.13	7.176	11054.51	13.22
5	4.282	3500.33	7.176	11063.77	13.22
6	4.283	3500.32	7.176	11110.93	13.21
Mean	4.281	3500.90	7.177	11083.19	-
RSD (%)	0.03	0.03	0.02	0.19	-
Requirement	RSD ≤ 2.0%	RSD ≤ 2.0%	RSD ≤ 2.0%	RSD ≤ 2.0%	≥ 3.5
Conclusion	Passed	Passed	Passed	Passed	Passed

The results in table 3 indicate that retention time, peak area, and resolution of Amoxicillin and Potassium Clavulanate in the standard solution met the acceptance criteria. Therefore, the method fulfills system suitability requirements.

3.1.3. Linearity

A series of diluted standard solutions of Amoxicillin and Potassium Clavulanate were prepared, analyzed by HPLC, and corresponding peak areas were recorded (table 4). From these data, linear regression equations were established, and the correlation coefficients (R^2) and y-intercepts were determined. The calibration curves are shown in figure 4a and figure 4b.

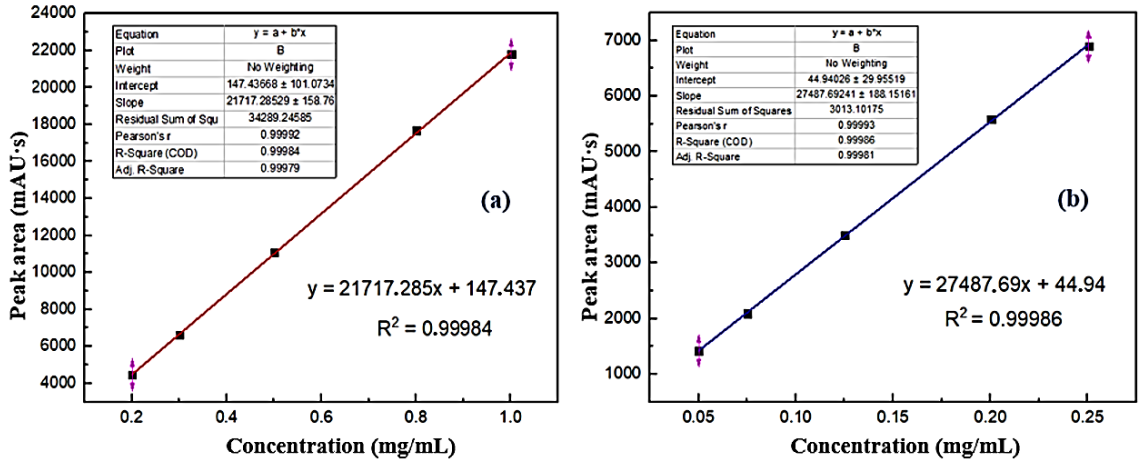

Figure 4. *Linear regression equations for a) Amoxicillin and b)Potassium Clavulanate.*

Linearity Evaluation of Amoxicillin and Potassium Clavulanate:

The developed HPLC method demonstrated excellent linearity for Amoxicillin across the concentration range of 0.2003–1.0016 mg/mL (figure 4a and table 4). The regression equation was y = 21717.285x + 147.437, with a high coefficient of determination (R^2 = 0.99984). The slope (21717.285 ± 158.76) confirmed strong sensitivity of the detector to change in analyte concentration, while the low intercept (147.437 ± 101.07) indicated minimal background interference (figure 4a). Peak areas increased proportionally from approximately 4468.51 to 21788.99 mAU·s, supporting a consistent and linear detector response.

For Potassium Clavulanate (figure 4b and table 4), linearity was observed in the range of 0.0501–0.2505 mg/mL, with a regression equation of y = 27487.69x + 44.94 and R^2 = 0.99986. Peak areas rose steadily from 1418.14 to 5591.85 mAU·s, indicating a uniform response within the tested range. The narrower range reflects the lower dosage level of this compound in combination products.

Table 4. *System suitability and linearity evaluation for Amoxicillin and Potassium Clavulanate.*

№	Dilution ratio (%)	Std. Sol Vol (mL)	Dil. Vol. (mL)	Linearity study of Amoxicillin		Linearity study of Kali clavulanate	
				Concentration (mg/mL)	Peak area (mAU·s)	Concentration (mg/mL)	Peak area (mAU·s)
1	40	2	20	0.2003	4468.51	0.0501	1418.14
2	60	3	20	0.3005	6628.13	0.0751	2092.44
3	100	5	25	0.5008	11083.19	0.1252	3500.90
4	160	10	20	0.8013	17674.49	0.2004	5591.85
5	200	10	10	1.0016	21788.99	0.2505	6898.49

At the 100% nominal concentration, y-intercept percentages were 1.33% for Amoxicillin and 1.28% for Potassium Clavulanate, both within the acceptable limit of ≤ 2%. These findings confirm that the method meets ICH Q2(R1) requirements for linearity and is suitable for quantitative analysis of both analytes.

3.2. Dissolution quantification results

The dissolution of six Vigentin tablets was quantified based on the formula provided in section 2.3d. The results for both active ingredients are summarized in table 5.

The results demonstrated that the dissolution of Potassium Clavulanate ranged from 103% to 107%, while that of Amoxicillin ranged from 93.9% to 98.9%. All six tested samples exceeded the

pharmacopoeial acceptance threshold of ≥75% [13, 14]. These findings are consistent with previous studies, in which commercial co-amoxiclav formulations also showed compliance with USP dissolution criteria [15, 16]. The higher values observed in this study highlight the robustness of the developed HPLC–DAD procedure for simultaneous quantification.

Table 5. *Dissolution results of Amoxicillin and Potassium Clavulanate components.*

Dissolution of the Amoxicillin component			Dissolution of the Kali Clavulanate component		
Tablet №	mt (mg)	Dissolution (% of mt compared to labeled mn: 500 mg)	Tablet №	mt (mg)	Dissolution (% of mt compared to labeled mn: 62,5 mg)
1	494.5	98.9%	1	64.5	103%
2	485.7	97.1%	2	66.6	107%
3	485.9	97.1%	3	65.0	104%
4	469.6	93.9%	4	65.2	104%
5	484.8	97.0%	5	64.3	103%
6	474.6	94.9%	6	64.6	103%
Requirement		≥ 75%	Requirement		≥ 75%
Conclusion		Complies	Conclusion		Complies

3.3. Quantification of amoxicillin-related impurities

The chromatogram in figure 5a shows a distinct peak for Impurity D with a retention time of 3.579 minutes. The peak is sharp, symmetrical, and well-separated from the baseline, indicating the method's high resolution and reliable detection capability for Impurity D. Figure 5b shows the peak for Impurity A at 4.573 minutes, which is also sharp with a stable baseline, confirming the specificity and separation efficiency of the chromatographic conditions. Figure 5c displays several well-resolved peaks at 1.153, 2.893, 3.551, and 4.193 minutes, without overlapping or baseline disturbance.

A preliminary resolution calculation (Rs) between the peaks at 2.893 and 3.551 minutes yields:

$$R_S = \frac{2(t_{R2} - t_{R1})}{\omega_1 + \omega_2} = \frac{2\text{x } 0.658}{0.2 + 0.2} = 3.29 > 2$$

Where: ω_1 *and* ω_2 are the baseline widths of the peaks, $\omega_1 = \omega_2 \approx 0.2$

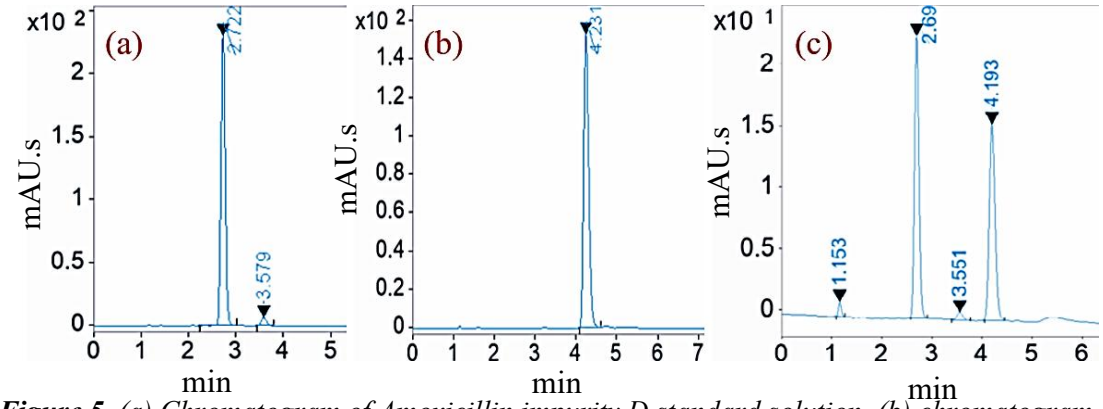

Figure 5. *(a) Chromatogram of Amoxicillin impurity D standard solution, (b) chromatogram of Amoxicillin impurity A standard, (c) system suitability test chromatogram.*

The resolution value of R_s = 3.29 meets pharmacopeial requirements, indicating that Impurities D and A are fully separated with stable retention times and flat baselines. The system suitability test

confirms the HPLC system's stability and reliability. Subsequently, six replicate injections of the Amoxicillin standard solution were performed to calculate the mean peak area for quantifying related impurities (table 6), while table 6 shows impurity peak areas detected in raw material batches.

Table 6. *Peak area values of the Amoxicillin standard solution across 6 samples.*

Replicate	Retention time (min)	Peak area (mAU·s)	Replicate	Retention time (min)	Peak area (mAU·s)
1	4.737	170.49	4	4.645	172.31
2	4.655	174.43	5	4.654	169.97
3	4.648	170.93	6	4.646	172.32
Average					171.74

The results presented in table 5 indicate that the retention time of the Amoxicillin peak exhibited a narrow range (4.645–4.737 minutes), demonstrating stable chromatographic conditions. The variation in peak area across replicates was minimal (169.97–174.43 mAU·s), with an average value of 171.74 mAU·s, indicating high repeatability and accuracy. These results meet the criteria of the System Suitability Test, confirming that the HPLC system is operating reliably and is suitable for constructing a calibration curve for the quantification of Amoxicillin in both raw materials and finished products.

Analysis of seven different raw material batches (table 7) revealed the common presence of two major impurities, Amoxicillin $D_{0.53}$ and Amoxicillin $D_{0.68}$, along with the maximum single impurity (max individual impurity) and several other sporadically detected impurities across certain batches.

Table 7. *Peak area values of detected impurities in the test solution from 7 product batches.*

Material Batch	Detected impurity	Sample 1 (mAU·s)	Sample 2 (mAU·s)	Material batch	Detected impurity	Sample 1 (mAU·s)	Sample 2 (mAU·s)
124120984	Amoxicillin D0.53	13.10	10.08	124121084	Amoxicillin D0.53	12.98	13.90
	Amoxicillin D0.68	34.62	24.02		Amoxicillin D0.68	26.98	29.25
	MI Impurity	12.92	12.63		MI Impurity	14.56	15.29
	Other impurities	–	6.06	124121086	Amoxicillin D0.53	13.68	16.04
124120986	Amoxicillin D0.53	10.27	11.64		Amoxicillin D0.68	28.13	30.14
	Amoxicillin D0.68	21.65	29.57		MI Impurity	14.16	15.21
	MI Impurity	–	11.84	624090681	Amoxicillin D0.53	14.07	14.00
124121082	Amoxicillin D0.53	13.29	14.22		Amoxicillin D0.68	28.31	25.88
	Amoxicillin D0.68	30.74	29.41		MI Impurity	14.21	13.50
	MI Impurity	14.96	49.44	624091385	Amoxicillin D0.53	13.69	13.78
	Other impurities	–	14.35		Amoxicillin D0.68	30.37	28.08

The peak areas of the detected impurities were recorded in duplicate for each batch, showing relatively consistent values between measurements and thereby confirming good repeatability of the applied HPLC–DAD method. The maximum individual impurity (MI) was detected

inconsistently across batches, with certain elevated values (e.g., 49.44 mAU·s in batch 124121082), indicating the necessity for tighter control of by-product formation during manufacturing. Other minor impurities were observed only in two batches (124120984 and 124121082) at low levels (6.06–14.35 mAU·s). These findings may be attributed to purification steps or secondary reactions during processing and warrant further evaluation in line with previous reports highlighting impurity monitoring as an essential component of quality assurance for co-amoxiclav formulations [8, 15, 17].

4. CONCLUSIONS

This study successfully developed and validated an HPLC-DAD method for dissolution testing of Amoxicillin/Potassium Clavulanate tablets, addressing the lack of an official procedure for this clinically important drug combination. The method met all validation requirements for specificity, system suitability, linearity, and repeatability, with correlation coefficients above 0.9998 for both analytes. Application to seven product batches demonstrated consistent dissolution results, with Amoxicillin (93.9–98.9%) and Potassium Clavulanate (103–107%) both exceeding pharmacopoeial criteria (≥75%). Moreover, the method enabled simultaneous detection of related impurities (Amoxicillin $D_{0.53}$ and $D_{0.68}$) within specification limits, highlighting its robustness. These findings confirm that the proposed procedure is not only reliable for routine quality control but also provides added value by integrating dissolution profiling with impurity monitoring in a single workflow. The method can therefore contribute to strengthening regulatory compliance, enhancing product quality assurance, and supporting further pharmaceutical development of fixed-dose antibiotic combinations. Future research could expand this approach by assessing dissolution behavior under biorelevant media that mimic physiological conditions, as well as comparing the method's performance with established international pharmacopoeial standards. Additionally, long-term stability studies and evaluation across different dosage forms would further validate its applicability in comprehensive quality control.

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TÓM TẮT

Nghiên cứu xác định các chế phẩm dược phẩm chứa Amoxicillin và Acid Clavulanic bằng phương pháp HPLC/DAD

Amoxicillin - một kháng sinh β-lactam, thường được phối hợp với acid clavulanic nhằm mở rộng phổ tác dụng, song các thử nghiệm hòa tan đối với dạng chế phẩm phối hợp này ít được thực hiện do đặc tính không bền của acid clavulanic cũng như khó khăn trong định lượng đồng thời hai hoạt chất. Điều này tạo ra khoảng trống đáng kể trong nghiên cứu và kiểm soát chất lượng sản phẩm Amoxicillin/Clavulanate. Nghiên cứu này tập trung phát triển và thẩm định phương pháp HPLC/DAD bền vững để đánh giá độ hòa tan chế phẩm thương mại Vigentin. Quá trình hòa tan được tiến hành trong nước RO siêu tinh khiết (18.2 MΩ·cm, 25 °C) dưới điều kiện nhiệt độ và tốc độ khuấy kiểm soát chặt chẽ; phân tích sắc ký sử dụng cột C18 với pha động natri dihydrogen phosphate/methanol (95:5, v/v) và phát hiện UV tại bước sóng 220 nm. Phương pháp được thẩm định đáp ứng đầy đủ các tiêu chí về tính đặc hiệu, độ lặp lại và độ phù hợp hệ thống, với độ tuyến tính xuất sắc ($R^2 > 0,9998$). Kết quả định lượng độ hòa tan trên sáu mẫu cho thấy amoxicillin giải phóng 93,9% – 98,9% và kali clavulanate 103% – 107%, đều vượt ngưỡng chấp nhận tối thiểu 75%. Các kết quả này chứng minh phương pháp đề xuất đã khắc phục hạn chế phương pháp luận trước đây, đồng thời cung cấp công cụ khả thi, tin cậy cho đánh giá chất lượng thường quy của chế phẩm phối hợp Amoxicillin/Clavulanate có ý nghĩa lâm sàng quan trọng.

Từ khóa: Amoxicillin; Kali Clavulanat; Độ hòa tan; Sắc ký lỏng (HPLC); Kiểm nghiệm chất lượng thuốc.