International Journal of Applied Pharmaceutics

ISSN- 0975-7058 Vol 17, Special Issue 1, 2025

Original Article

ENHANCING PIPERINE SOLUBILITY THROUGH MULTICOMPONENT CRYSTALS WITH CINNAMIC ACID: CHARACTERIZATION AND EVALUATION

USWATUL HASANAH[1], DHIANDRA ARYA WARDHANA[2], ERIZAL ZAINI[3]*

[1,3]Department of Pharmaceutics, Faculty of Pharmacy, Universitas Andalas, Padang, West Sumatra-25163, Indonesia. [2]Undergraduate Programme, Faculty of Pharmacy, Universitas Andalas, Padang, West Sumatra-25163, Indonesia
*Corresponding author: Erizal Zaini; *Email: erizal@phar.unand.ac.id

Received: 17 Sep 2024, Revised and Accepted: 20 Nov 2024

ABSTRACT

Objective: Piperine, an active alkaloid in *Piper nigrum* and *Piper retrofractum*, is known for its anti-inflammatory, antioxidant, and bioavailability-enhancing properties. However, its low water solubility limits its therapeutic potential.

Methods: This study addresses that limitation by enhancing piperine's solubility through forming multicomponent crystals using cinnamic acid as a coformer in a molar ratio of 4.5:5.5 via Liquid-Assisted Grinding (LAG). Various techniques, including X-ray Diffraction (XRD), Differential Scanning Calorimetry (DSC), Fourier Transform Infrared Spectroscopy (FT-IR), and Scanning Electron Microscopy (SEM), were used to characterize these crystals. Solubility tests were conducted in CO_2-free distilled water using sonication and analyzed with high-performance liquid chromatography (HPLC).

Results: The DSC results revealed an endothermic peak at 79.91 °C, and XRD analysis showed no new peaks, indicating that the crystal structure of piperine remained unchanged. FT-IR analysis revealed hydrogen bond formation, and SEM showed smaller particle sizes in the modified crystals. Most importantly, the solubility of the multicomponent crystals increased 1.785-fold compared to intact piperine.

Conclusion: These findings suggest that piperine-cinnamic acid multicomponent crystals, forming a eutectic mixture, significantly improve piperine's solubility, with statistical analysis confirming the results ($p<0.05$).

Keywords: Piperine, Cinnamic acid, Multicomponent crystal

DOI: https://dx.doi.org/10.22159/ijap.2025.v17s1.22 Journal homepage: https://innovareacademics.in/journals/index.php/ijap

INTRODUCTION

Piperine is a secondary metabolite from the alkaloid group, commonly isolated from the fruit and roots of the *Piper nigrum* and *Piper retrofractum* species of the Piperaceae family. Piperine is known to possess numerous pharmacological activities, including antioxidant, anti-inflammatory, antipyretic, antifungal, antidiarrheal, antidepressant, antithyroid, antimutagenic, antitumor, analgesic, hepatoprotective, antihypertensive, and other pharmacological activities. Additionally, piperine can act as a bioenhancer, improving drug absorption by slowing down drug metabolism, thereby increasing the bioavailability of drugs with low bioavailability [1, 2].

The clinical use of piperine is still limited due to its low solubility in water, which is 40 mg/l at 18 °C. This low solubility results in poor bioavailability of piperine in plasma, necessitating high doses to achieve therapeutic effects. However, if absorption is enhanced, the resulting increase in bioavailability could lead to toxicity risks, particularly to the reproductive and nervous systems. To address this issue, it is necessary to modify the drug compound both physically and chemically to enhance the solubility of piperine. One method that can be employed is the multicomponent crystal method. Previous research has been conducted to improve the solubility of piperine in water, such as the formation of solid dispersions and inclusion complexes that have proven to increase the solubility of piperine by 1.389 and 7.88 fold, respectively [3, 4].

Multicomponent crystals can alter the physicochemical properties of drugs, including enhancing solubility and dissolution rate, which can potentially improve bioavailability [5, 6]. Multicomponent crystals consist of two components: the Active Pharmaceutical Ingredient (API) and a coformer, which interact with each other to form non-covalent bonds such as hydrogen bonds, ionic bonds, and other interactions like polymorphism and π-π interactions [7]. A significant advantage of multicomponent crystals is that they do not affect the pharmacological effects of the active ingredient but can modify its physical properties, such as increasing dissolution rate, solubility, and compressibility. Several active ingredients that have been successfully enhanced in solubility using the multicomponent crystal method include ketoprofen [8], trimethoprim [9], fenofibric acid [10], mefenamic acid [11], and simvastatin [12].

The coformer to be used in this study is cinnamic acid. Cinnamic acid is generally regarded as a safe (GRAS) substance. The selection of a coformer must not negatively affect the API being used. The GRAS status of cinnamic acid makes it a potential candidate for use as a coformer in forming multicomponent crystals. Additionally, the selection of the coformer is based on the synthon and pKa approaches. The synthon approach suggests that the coformer should have functional groups capable of interacting with the active drug's functional groups. Cinnamic acid, an aromatic carboxylic acid, contains benzene and carboxylic acid groups, which are expected to form hydrogen bonds with the ketone group in piperine. Cinnamic acid has also been widely used as a coformer in the formation of multicomponent crystals with compounds such as carbamazepine, itraconazole, AMG 517, ibuprofen, moxifloxacin, isoniazid, and lovastatin [13].

Based on the considerations above, this study was conducted to form a piperine-cinnamic acid multicomponent crystal to improve the solubility of piperine. This multicomponent system was characterized using DSC, FTIR spectroscopy, XRD, and SEM as part of the evaluation.

MATERIALS AND METHODS

Materials

Piperine (Sigma-Aldrich, USA), cinnamic acid (TCI, Japan), ethanol pro analysis (Merck, Germany), methanol pro analysis (Merck, Germany), acetonitrile (Merck, Germany), glacial acetic acid (Merck, Germany), distilled water, and CO_2-free distilled water.

Preparation of piperine-cinnamic acid physical mixture

The physical mixture of piperine-cinnamic acid was prepared in various molar ratios from 1:9 to 9:1 of piperine and cinnamic acid for the binary phase diagram and the physical mixture for the characterization sample. The weighed powder was mixed in a flask

and vortexed for 30 seconds. The obtained powder was stored in a desiccator [14].

Formation of piperine-cinnamic acid multicomponent crystals using the solvent drop grinding method

Multicomponent crystals were prepared by weighing piperine and cinnamic acid in a molar ratio of 4.5:5.5 (1.28403 g: 0.8149 g), followed by grinding for 10 min while adding one drop of ethanol. The obtained powder was then stored in a desiccator [14].

Differential scanning calorimetry (DSC) analysis

Thermal analysis of the samples was performed using a DSC instrument (Shimadzu DSC 60plus, Japan) with calibrated temperature. A sample of 4 mg was placed in a sealed aluminum pan. The DSC was programmed with a temperature range of 30–200 °C at a heating rate of 10 °C per minute [14]. The study was conducted on piperine, cinnamic acid, the multicomponent crystal, and the physical mixture.

X-ray diffraction (XRD) analysis

X-ray diffraction analysis was conducted at room temperature using a Rigaku RINT-2500 diffractometer. The measurement conditions were as follows: Cu metal target, Kα filter, voltage 40 kV, current 40 mA, with analysis in the 2-theta range of 5°–50°. The sample was placed on a glass sample holder and leveled to prevent particle orientation during sample preparation. The analysis was performed on piperine, cinnamic acid, piperine-cinnamic acid physical mixture, and piperine-cinnamic acid multicomponent crystals [14].

Fourier transform infrared (FT-IR) analysis

Fourier transform infrared (FT-IR) (Shimadzu IRTracer-100AH, Japan) analysis was carried out by dispersing the sample on an Attenuated Total Reflectance (ATR) crystal until the entire surface was covered. The absorption spectra were recorded in the wavenumber range of 4000–600 cm^{-1}. The analysis was performed on piperine, cinnamic acid, piperine-cinnamic acid physical mixture, and piperine-cinnamic acid multicomponent crystals [14].

Scanning electron microscopy (SEM) analysis

The SEM analysis (Hitachi S-3400 N, Japan) was conducted on piperine, cinnamic acid, their physical mixture, and piperine-cinnamic acid multicomponent crystals by placing the sample on an aluminum sample holder and coating it with gold or palladium. The samples were observed using the SEM instrument at various magnifications, with the voltage set at 20 kV and the current at 12 mA [14].

Solubility test

The solubility test was conducted on piperine and the piperine-cinnamic acid multicomponent crystals by creating a saturated solution using CO_2-free distilled water. The CO_2-free distilled water was selected to maintain a stable and neutral environment, free from pH alterations caused by dissolved carbon dioxide, which could influence the solubility measurements of the active pharmaceutical ingredient. A sample equivalent to 40 mg of piperine was weighed and dissolved in 100 ml of CO_2-free distilled water. The sample was then sonicated for 30 min. After sonication, the sample was filtered using a 0.45 µm filter (Whatman filter paper). The obtained filtrate was then analyzed using High-Performance Liquid Chromatography (HPLC) (Agilent 1220, USA) with a mobile phase of acetonitrile: CO_2-free distilled water (containing 0.5% glacial acetic acid) (8:2) and a stationary phase of Phenomenex/ODS C18 column (4.6 x 150 mm). The flow rate was set at 1.5 ml/min, with an injection volume of 20 µl and a retention time of 1.4 min. The peak area was recorded, and the dissolved piperine concentration was calculated [14].

RESULTS AND DISCUSSION

DSC analysis is a screening tool for forming multicomponent crystal phases between piperine and cinnamic acid. The key observations from the DSC analysis include the melting point and the enthalpy of fusion for piperine, cinnamic acid, their physical mixture, and the resulting multicomponent crystal. DSC is commonly used to assess the thermal properties of materials and detect phase transitions. In this case, it helps to determine if a new multicomponent crystal phase has been formed between piperine and cinnamic acid. By comparing the melting points and enthalpies of the pure components, their physical mixture, and the multicomponent crystal, it is possible to identify shifts or changes in the thermal behavior that indicate new interactions or phase formation [15]. It is necessary to test different molar ratios of the mixture to determine the eutectic point of the piperine-cinnamic acid multicomponent crystal. DSC analysis of 11 different molar ratios indicated that the multicomponent crystal is an eutectic mixture. The eutectic point for the piperine-cinnamic acid multicomponent crystal occurs at a molar ratio of 4.5:5.5, as shown in table 1. The data indicates that the eutectic mixture is formed at this specific ratio, and the reduction in melting point suggests enhanced physical interaction between the piperine and cinnamic acid, creating a stable eutectic composition.

The design of various molar ratios of piperine and cinnamic acid was developed to clarify the eutectic point of the two-component mixture, which was then analyzed using a binary phase diagram. A binary phase diagram is a graphical representation of the equilibrium conditions between different phases of a substance under varying temperatures and compositions [16].

Plotting these phase diagrams allows one to determine the specific eutectic composition, which corresponds to the lowest temperature at which the components can melt together, providing insight into the interactions between piperine and cinnamic acid at different ratios. This approach is essential for understanding the solid-liquid transition and the stability of the eutectic mixture, making it a crucial tool for verifying the formation of multicomponent crystals [17].

Table 1: Thermal properties of piperine–cinnamic acid binary phases, including melting point peaks and enthalpy of fusion (ΔH)

Binary phase	Peak 1		Peak 2	
	Melting point (C)	ΔH Fusion (J/g)	Melting point (C)	ΔH Fusion (J/g)
1:9	78.93	11.30	126.69	37.65
2:8	79.44	29.73	102.26	12.32
3:7	79.78	50.82	97.6	2.64
4:6	80.02	61.36	-	-
4.5:5.5	79.91	68.33	-	-
5:5	79.82	62.43	-	-
5.5:4.5	79.64	65.31	-	-
6:4	79.91	66.68	-	-
7:3	79.78	34.87	105.84	15.86
8:2	78.96	15.62	115.66	29.76
9:1	79.80	8.11	123.12	49.88

The binary phase diagram of piperine and cinnamic acid (fig. 2) shows the lowest melting point at the piperine–cinnamic acid molar ratio of 5.5:4.5, with a melting point of 79.64 °C. In the DSC thermogram (fig. 1), the molar ratio of 5.5:4.5 exhibits a single but slightly broader endothermic peak, which raises concerns that the two components may not have completely melted together at the same time. In contrast, the piperine–cinnamic acid molar ratio of 4.5:5.5 demonstrates a sharper endothermic peak in the thermogram. This suggests a more uniform melting process, indicating better interaction between the components and a clearer eutectic mixture. The sharpness of the peak may reflect a more well-defined eutectic composition where the active

compound and the coformer melt simultaneously, ensuring optimal performance of the multicomponent crystal. Therefore, the piperine–cinnamic acid multicomponent crystal formation was selected with a molar ratio of 4.5:5.5, displaying a sharper endothermic peak compared to other molar ratios. The multicomponent piperine–cinnamic acid crystal at this ratio had a melting point of 79.9 °C. The reduction in melting point suggests weaker crystal lattice energy, corresponding to weaker crystal-forming interactions. Solubility requires disruption of the crystal lattice, so the weaker the lattice, the better the solubility [18].

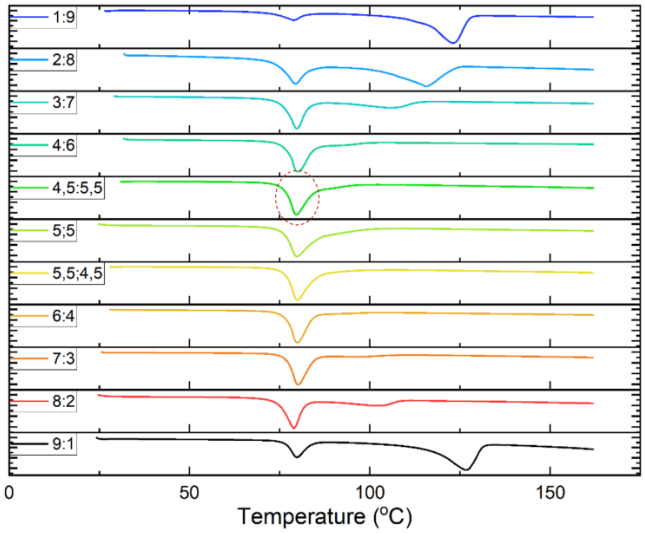

Fig. 1: DSC thermograms overlay of piperine–cinnamic acid binary mixtures at various molar ratios. The 4.5:5.5 ratio shows a single sharper peak

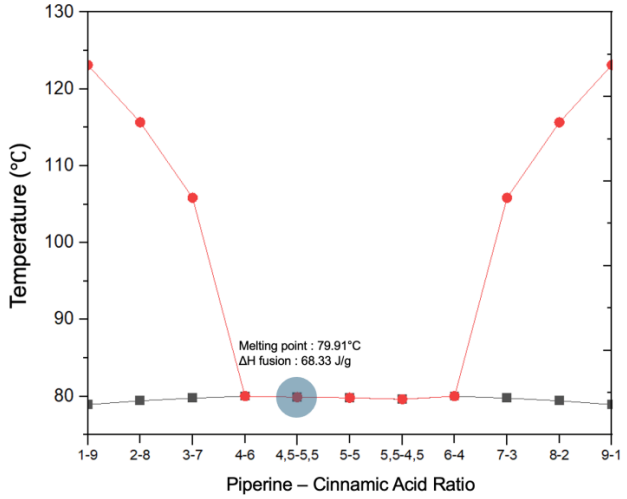

Fig. 2: Binary phase diagram of piperine–cinnamic acid, showing melting point variations across molar ratios and identifying the eutectic composition by evaluating the melting behavior

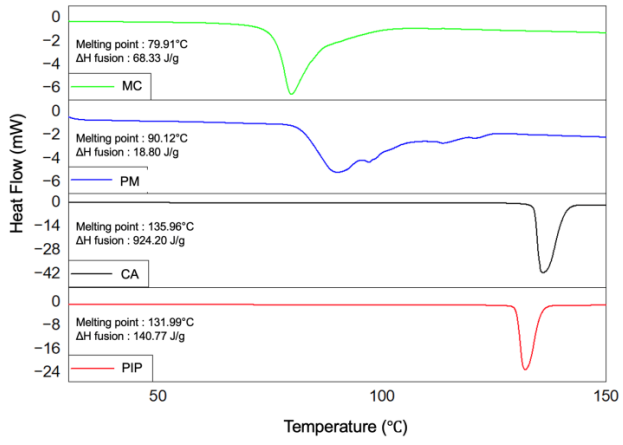

Fig. 3: DSC thermograms overlay of multicomponent crystal (MC), physical mixture (PM), cinnamic acid (CA), and piperine (PIP). Melting points and enthalpy of fusion (ΔH) values for each sample are labeled near their respective thermograms

In fig. 3, based on the DSC analysis, the melting points of piperine (131.99 °C) and cinnamic acid (135.96 °C) were observed. The melting points were reduced for the multicomponent crystal (79.91 °C) and the physical mixture (90.12 °C). This decrease in melting point is attributed to the physical interactions between cinnamic acid and piperine, which weaken the crystal lattice. As the lattice energy diminishes, the energy required to break the interactions within the crystal structure also decreases, leading to a lower melting point [19].

In fig. 3, the thermogram of the physical mixture shows more than one endothermic peak, indicating that the two constituent components have not melted simultaneously. In contrast, the thermogram of the multicomponent crystal displays a single endothermic peak with a melting point that is relatively lower than its components. This suggests that the active compound and the coformer have melted together, signifying their successful interaction.

The lower melting point of the multicomponent crystal compared to its components and the single endothermic peak in the thermogram align with the XRD results (fig. 4), suggesting that the formed multicomponent crystal is likely a eutectic mixture. A eutectic mixture is a specific composition of at least two solid components that transition into a liquid phase at a particular temperature, known as the eutectic point [20]. This temperature corresponds to the lowest melting point possible for various component compositions.

XRD analysis is a crucial technique for determining the properties of solid substances, including their crystalline phases, amorphous content, and solid-state reactions. In the study of multicomponent crystals, the diffraction patterns of the solid phase often differ from those of the pure APIs. These distinct diffraction patterns provide valuable information about the structural changes that occur when forming multicomponent crystals. The appearance of new peaks or the disappearance of existing ones can indicate the formation of a new crystalline phase, revealing insights into molecular interactions, structural modifications, and potential improvements in the physicochemical properties, such as solubility and dissolution rate, which are essential for drug formulation and development [21].

Based on fig. 4, the diffractogram of the multicomponent crystal and the physical mixture of piperine and cinnamic acid exhibit patterns that closely resemble those of their pure components. The diffractogram of multicomponent crystal results show no new diffraction peaks, indicating that no new crystalline phase has formed from the mixture [22]. This indicates that the eutectic mixture does not involve the formation of a new crystalline phase but represents a specific arrangement of the existing crystalline structures. However, a decrease in intensity is observed at the same diffraction peaks. In X-ray diffraction analysis, peak intensity changes can indicate crystalline structure alterations. For the piperine sample, the reduction in peak intensity at specific 2 theta positions (14.201, 22.3253, and 29.8413) suggests that the crystal lattice may have undergone some structural modification. Similarly, the cinnamic acid sample shows a decrease in peak intensity at 2 theta positions of 9.7795 and 22.3253. These reductions in peak intensity could be attributed to several factors, such as partial disordering within the crystal lattice or a rearrangement of molecules [23]. The XRD results demonstrate that the eutectic mixture is characterized by the coexistence of crystalline phases of the pure components. While this may appear similar to a physical mixture, the melting point reduction observed in the DSC analysis confirms the thermodynamic stability of the eutectic system. This behavior is consistent with eutectic formation, where two components form a stable phase with distinct melting characteristics at specific ratios.

Thermodynamic activity, including changes in temperature or pressure during the sample preparation or analysis, may lead to a loss of crystallinity or a decrease in the regularity of the crystal structure. This could result in a less defined or partially amorphous material, explaining the observed reduction in peak intensity. Understanding these changes is crucial for determining how the multicomponent crystal's properties, such as solubility or stability, are affected [24].

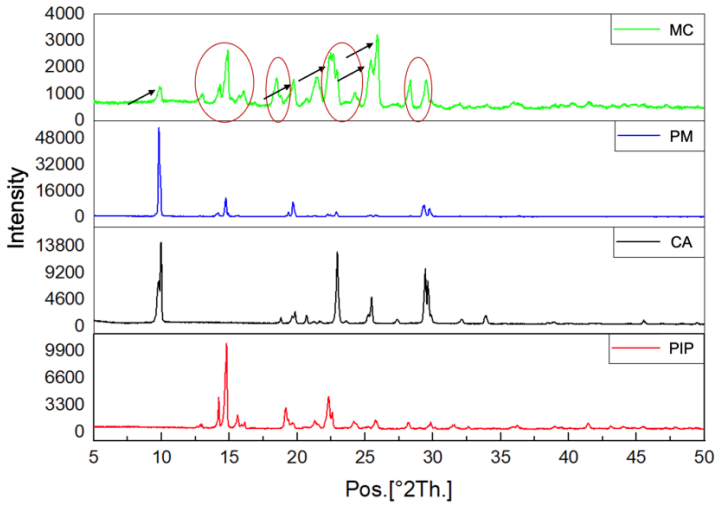

Fig. 4: XRD diffractograms overlay of multicomponent crystal (MC), physical mixture (PM), cinnamic acid (CA), and piperine (PIP). Peaks corresponding to cinnamic acid (black arrows) and piperine (red circles) are highlighted to illustrate retained crystalline features in the multicomponent crystal

FT-IR analysis is a technique that can be used to observe chemical interactions between solid molecules. The formation of multicomponent crystals can exhibit significant differences in the shift of transmission bands in the FT-IR spectrum. The results of the FT-IR spectra and the respective wavenumber data for each sample are shown in fig. 5. This method is crucial in identifying changes in functional groups and detecting the formation of hydrogen bonds or other non-covalent interactions, critical indicators of multicomponent crystal formation. Shifts in wavenumbers suggest alterations in the molecular environment, potentially confirming the interaction between piperine and cinnamic acid in the crystal structure [25].

The results of the FT-IR spectroscopy analysis show that the piperine spectrum has absorption peaks at wavenumbers 1610.82 cm^{-1}, 1250.89 cm^{-1}, 1112.99 cm^{-1}, and 830.34 cm^{-1}, indicating the presence of C=O, C-N, C-O, and C=N groups in piperine. Meanwhile, the cinnamic acid spectrum shows absorption at wavenumbers 2829.04 cm^{-1}, 1627.34 cm^{-1}, and 766.8 cm^{-1}, representing O-H,

C=O, and C=C groups in cinnamic acid. There were noticeable shifts in the spectrum after forming the multicomponent crystal and the physical mixture of piperine and cinnamic acid. The C=O group in piperine shifted from 1610.82 cm^{-1} to 1581.49 cm^{-1} in the multicomponent crystal. This shift is due to the C=O group in piperine donating electrons to the O-H group in cinnamic acid to formed a intermolecular interaction such as a hydrogen bond, observed at 2941.08 cm^{-1} peak in the multicomponent crystal spectrum. However, the evidence remains qualitative, and future studies employing molecular simulations or quantitative spectroscopic techniques could provide further validation of the extent and nature of these hydrogen bonds. Conversely, the shift in the C=O group remains within the expected wavenumber range, indicating that molecular interactions have occurred between the two components [26, 27]. This interaction confirms the formation of a multicomponent crystal, as the hydrogen bond formation is consistent with the observed wavenumber shifts, particularly in the C=O stretching region. The alteration in the molecular environment further corroborates the involvement of piperine and cinnamic acid in forming the multicomponent system.

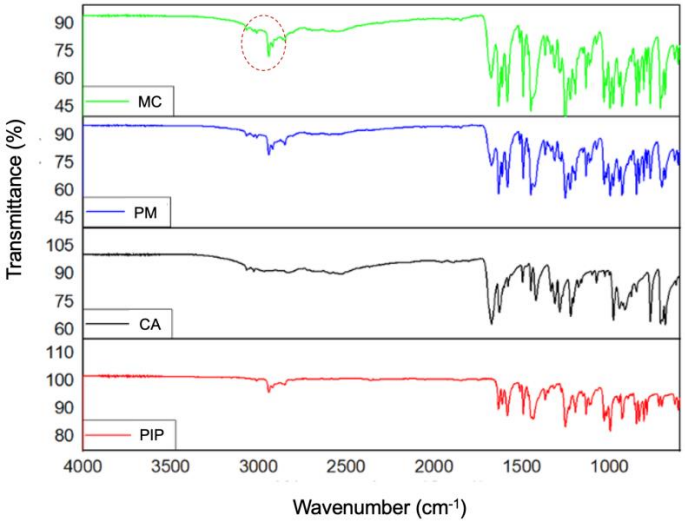

Fig. 1: FT-IR spectra overlay of multicomponent crystal (MC), physical mixture (PM), cinnamic acid (CA), and piperine (PIP). A possible-OH peak at 2941 cm-1, indicative of hydrogen bonding, is marked

Cinnamic acid possesses an aromatic ring structure and a carboxylic acid group, both of which can play a role in enhancing the solubility of piperine. The aromaticity of cinnamic acid allows for potential π-π stacking interactions with piperine's aromatic ring, which may stabilize the eutectic mixture and increase solubility by reducing the intermolecular forces between piperine molecules. Additionally, the carboxylic group of cinnamic acid can form hydrogen bonds with piperine, further improving its solubility by facilitating favorable molecular interactions that promote dissolution in the solvent. These structural properties likely contribute to the observed solubility enhancement of piperine when combined with cinnamic acid in the eutectic mixture.

To further clarify the morphological structure of the formed eutectic mixture, a SEM analysis was conducted. This method aims to microscopically examine the surface morphology of the samples, providing valuable information about their texture and surface shape. The SEM observations of piperine, cinnamic acid, the physical mixture, and the piperine-cinnamic acid multicomponent crystal can be seen in fig. 6. Through SEM analysis, surface characteristics such as roughness, particle size, and arrangement patterns can be discerned. Compared to the pure components, surface texture and particle size changes may be why piperine's solubility was increased in the form of an eutectic mixture with succinic acid.

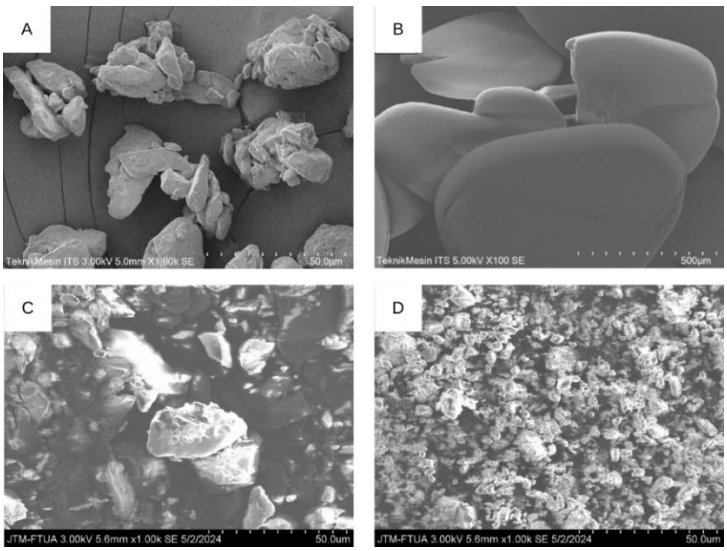

Fig. 2: SEM microphotographs of (A) piperine (5.00 mm × 1.00k magnification), (B) cinnamic acid (×100 magnification), (C) physical mixture (5.6 mm × 1.00k magnification), and (D) multicomponent crystal (5.6 mm × 1.00k magnification)

Based on observations from the image at 5.00mm×1.00k magnification in fig. 6, piperine exhibits a rod-like crystalline morphology. At the same time, cinnamic acid displays a flattened spherical shape with a relatively smooth surface and non-uniform size. In the physical mixture, the morphology appears as a combination of piperine crystals and cinnamic acid. However, in the multicomponent crystal, the particle size is noticeably smaller and more irregular. This change is attributed to using the LAG method during the multicomponent crystal formation [28]. The reduction in crystal particle size increases the surface area, which enhances the interaction with solvents and potentially improves solubility [22]. This morphological change supports the hypothesis that the multicomponent crystal formation improves dissolution properties by increasing the surface exposure to the solvent, thus facilitating better solubility than pure compounds or physical mixtures [27]. However, this conclusion is based on qualitative observations, as quantitative particle size distribution data, such as those obtained via laser diffraction or DLS, were not available for this study. Future work should incorporate these techniques to provide definitive evidence of particle size reduction and its correlation with solubility enhancement.

The solubility test was conducted by dissolving piperine and the piperine–cinnamic acid multicomponent crystal in CO_2-free distilled water. The samples were sonicated for 30 min to observe the impact of multicomponent crystal formation on piperine's solubility. The amount of dissolved piperine was then determined using HPLC analysis at the maximum absorbance wavelength of piperine in CO_2-free distilled water, which is 342 nm. This method allows for comparing the solubility enhancement provided by the multicomponent crystal system, as the sonication helps disperse the sample evenly and ensure accurate measurements of dissolved piperine. Analyzing the solubility via HPLC, any significant improvement in dissolution due to the crystal structure modification can be quantitatively assessed.

Table 2: Solubility data of piperine and its multicomponent crystal (MC)

Compound	Solubility* (mg/100 ml)	Increase in solubility
Piperine	0.033±0.002	-
Piperine – cinnamic acid eutectic mixture	0.059±0.003	1.785

*Data represent the mean±SD from three independent solubility tests conducted in CO_2-free distilled water

Table 2 presents the solubility test data for piperine and the piperine–cinnamic acid multicomponent crystal. The average concentration of dissolved piperine in CO_2-free distilled water was found to be 0.033±0.002 mg/l, while for the multicomponent crystal sample, the average solubility increased to 0.059±0.003 mg/l. This data reveals a 1.785-fold increase in the solubility of piperine within the multicomponent crystal system.

The increased solubility of piperine in the multicomponent crystal is related to the results of DSC characterization, which showed a reduction in the melting point and fusion energy of the multicomponent crystal. A lower fusion energy indicates less energy is required to melt the compound. Additionally, the improvement in solubility is associated with a decrease in crystallinity, as indicated by XRD analysis and the morphological changes observed through SEM characterization. The reduction in particle size of the multicomponent crystal, achieved through grinding the pure components, increases the contact surface area between the compound and the solvent, thereby enhancing solubility [22, 27]. While CO_2-free distilled water provides a controlled environment for solubility testing, it does not replicate the ionic strength or pH of physiological fluids. Future studies will consider solubility evaluations in phosphate buffer (pH 7.4) to align with *in vivo* relevance.

The piperine and the multicomponent crystal solubility test results were statistically analyzed using paired t-tests with the SPSS 26 software. The paired t-test for solubility analysis showed a significance value of $p<0.05$, confirming that the formation of the piperine–cinnamic acid multicomponent crystal significantly increased the solubility of piperine. The paired t-test demonstrates a significant improvement in solubility; however, the practical significance of this enhancement has not been quantified using effect size or confidence intervals. Additionally, particle size data and other physicochemical characteristics that could influence solubility were not collected. These limitations suggest the need for further studies to fully characterize the factors contributing to solubility enhancement and to evaluate the practical relevance of the observed results. In this study, the eutectic mixture of piperine and cinnamic acid enhanced solubility by 1.785-fold, demonstrating a moderate improvement compared to previous approaches like solid dispersions and inclusion complexes. Unlike inclusion complexes, which require specialized materials (e.g., cyclodextrins) and sophisticated processing techniques, the eutectic mixture relies on readily available components and simpler preparation methods. This makes it a cost-effective and scalable option for industrial applications, particularly in low-resource settings.

CONCLUSION

The results of this study demonstrate that the formation of a piperine–cinnamic acid multicomponent crystal significantly improves the solubility of piperine. The multicomponent crystal exhibited a 1.785-fold increase in solubility compared to intact piperine. This enhancement is closely linked to the reduction in melting point and fusion energy, as indicated by DSC analysis, and a decrease in crystallinity confirmed by XRD. The smaller particle size observed through SEM analysis also increased surface area and enhanced solubility. Statistical analysis using a paired t-test further validated the significant improvement in solubility. Therefore, the multicomponent crystal approach proves to be an effective method for enhancing the solubility of piperine.

ACKNOWLEDGMENT

The Authors would like to express a gratitude to the Research and Community Service Institute of Universitas Andalas for funding this research.

FUNDING

The Research and Community Service Institute of Universitas Andalas funded this research under the Undergraduate Thesis Research scheme in 2024 with contract number 143/UN16.19/PT.01.03/PSS/2024.

AUTHORS CONTRIBUTIONS

The study was conceptualized and methodologically designed by EZ and UH. EZ, UH, and DAW carried out the investigations, with DAW performing the formal analysis and contributing to data visualization alongside UH. The initial draft of the manuscript was prepared by UH and DAW, while EZ and UH provided critical revisions during the review and editing process. The project was supervised by EZ, with funding acquisition and project administration jointly managed by EZ and UH. Each author reviewed and approved the final version of the manuscript.

CONFLICT OF INTERESTS

The authors declare no conflict of interests related to this work.

REFERENCES

1. Tiwari A, Mahadik KR, Gabhe SY. Piperine: a comprehensive

review of methods of isolation, purification, and biological properties. Medicine in Drug Discovery. 2020 Sep;7:100027. doi: 10.1016/j.medidd.2020.100027.

2. Chopra B, Dhingra AK, Kapoor RP, Prasad DN. Piperine and its various physicochemical and biological aspects: a review. Chem. 2016;3(1):75-96. doi: 10.2174/1874842201603010075.
3. Jessica A, Naura R, Hasanah U, Zaini E, Fitriani L. Pembentukan multikomponen kristal piperin dan kuersetin. JOPS. 2021;4(2):1-11. doi: 10.36341/jops.v4i2.1881.
4. Fitriani L, Tirtania S, Umar S, Zaini E. Enhancing the solubility and dissolution rate of piperine via preparation of piperine-hydroxypropyl methylcellulose 2910 solid dispersion system using freeze-drying method. J Pharm Pharmacogn Res. 2024 Jan 1;12(1):175-83. doi: 10.56499/jppres23.1734_12.1.175.
5. DN CP, AS. A solubility enhancement of aceclofenac by new crystallization technique. Asian J Pharm Clin Res. 2022 Feb 7:113-8.
6. Munde MK, Shinde AM, Kulkarni NS, Tambe VS, Alhat HP. Comprehensive review on nanocrystal technology in pharmaceutical formulation. Int J Pharm Pharm Sci. 2023 Apr 1:1-7. doi: 10.22159/ijpps.2023v15i4.47317.
7. Qiao N, Li M, Schlindwein W, Malek N, Davies A, Trappit G. Pharmaceutical cocrystals: an overview. Indian J Pharm Sci. 2017;79(6):858-71.
8. Sonita H, Zaini E, Umar S. Preparation and characterization phisicochemical properties of multi-component ketoprofen with co-former glutamine. MS. 2023;8(2):849-60. doi: 10.37874/ms.v8i2.799.
9. Umar S, Farnandi R, Salsabila H, Zaini E. Multicomponent crystal of trimethoprim and citric acid: solid state characterization and dissolution rate studies. Open Access Maced J Med Sci. 2022;10(A):141-5. doi: 10.3889/oamjms.2022.7920.
10. Anggraini D, Salsabila H, Umar S, Aldi Y, Zaini E. Preparation and characterization of a eutectic mixture of fenofibric acid and nicotinic acid and evaluation of *in vivo* antihyperlipidemic activity. Sci Technol Indones. 2022 Oct 31;7(4):514-21. doi: 10.26554/sti.2022.7.4.514-521.
11. Izadihari R, Rosaini H, Zaini E, Yuliandra Y. Multicomponent crystals of mefenamic acid-tromethamine with improved dissolution rate. Sanat. 2019;23(6):988-96. doi: 10.35333/jrp.2019.63.
12. Zaini E, Evert A, Dona Octavia MD. Increase in dissolution rate of simvastatin by amorphous solid dispersion system with hydroxypropylmethylcellulose polymer. Orient J Chem. 2017;33(4):2080-4. doi: 10.13005/ojc/330457.
13. Putri D. Review: multi-component crystals: cinnamic acid as a co-former. Int J Pharm Sci Med. 2021 Jan 21;6(1):92-8. doi: 10.47760/ijpsm.2021.v06i01.008.
14. Octavia MD, Hasmiwati H, Revilla G, Zaini E. Multicomponent crystals of piperine-nicotinic acid: the physicochemical and dissolution rate properties. Trop J Nat Prod Res. 2023 Aug 31;7(8):3701-5.
15. Yamashita H, Hirakura Y, Yuda M, Teramura T, Terada K. Detection of cocrystal formation based on binary phase diagrams using thermal analysis. Pharm Res. 2013 Aug 21;30(1):70-80. doi: 10.1007/s11095-012-0850-1, PMID 22907418.
16. Sunil Dhoot A, Naha A, JuhiPriya J J, NehaXalxo N N. Phase diagrams for three-component mixtures in pharmaceuticals and its applications. J. 2018;10(2):132-7. doi: 10.5530/jyp.2018.10.31.
17. Hasanah U, Badriyya E, Safitri R, Yuliza S, Ihsan I, Saafrida RH. Ketoprofen-tromethamine: binary phase diagram of multicomponent crystal, dissolution rate, and analgesic activity evaluation. Sci Technol Indones. 2024 Jun 30;9(3):726-34. doi: 10.26554/sti.2024.9.3.726-734.
18. Dwichandra Putra O, Yonemochi E, Uekusa H. Isostructural multicomponent gliclazide crystals with improved solubility. Cryst Growth Des. 2016 Nov 2;16(11):6568-73. doi: 10.1021/acs.cgd.6b01279.
19. Sari YN, Zaini E, Ismed F. Peningkatan laju disolusi piperine dengan pembentukan multikomponen kristal menggunakan asam nikotinat. J Sains Farm Klin. 2019 Aug 28;6(2):180. doi: 10.25077/jsfk.6.2.180-185.2019.
20. Pena Pereira F, de la Calle I. Solvents and eutectic solvents. In: Worsfold P, Poole C, Townshend A, Miro M, editors. Encyclopedia of analytical science. 3rd ed. Cambridge: Academic Press; 2019. p. 184-90.
21. Zaini E, Afriyani FL, Fitriani L, Ismed F, Horikawa A, Uekusa H. Improved solubility and dissolution rates in novel multicomponent crystals of piperine with succinic acid. Sci Pharm. 2020;88(2). doi: 10.3390/scipharm88020021.
22. Hasanah U, Wahyuni L, Zaini E. Micronized eutectic mixture of fenofibric acid-saccharin formation for solubility and disoolution enhancement. Int J Appl Pharm. 2023 Feb 7;15 Special Issue 1:56-60.
23. Rabiej S. A comparison of two X-ray diffraction procedures for crystallinity determination. Eur Polym J. 1991 Jan 1;27(9):947-54. doi: 10.1016/0014-3057(91)90038-P.
24. Ainurofiq A, Mauludin R, Mudhakir D, Setianto AB, Soewandhi SN. The effect of compression force on alteration of desloratadine and its multicomponent crystal crystallinities using X-ray diffraction and ATR-FTIR techniques. KEM. 2018;787:43-51. doi: 10.4028/www.scientific.net/KEM.787.43.
25. Salsabila H, Fitriani L, Zaini E. Recent strategies for improving solubility and oral bioavailability of piperine. Int J Appl Pharm. 2021;13(4):31-9. doi: 10.22159/ijap.2021v13i4.41596.
26. Garrido B, Gonzalez S, Hermosilla J, Millao S, Quilaqueo M, Guineo J. Carbonate-β-cyclodextrin-based nanosponge as a nanoencapsulation system for piperine: physicochemical characterization. J Soil Sci Plant Nutr. 2019 Sep 1;19(3):620-30. doi: 10.1007/s42729-019-00062-7.
27. Fitriani L, Fitriandi AD, Hasanah U, Zaini E. Nano-cocrystals of piperine-succinic acid: physicochemical characterization and dissolution rate studies. Chemistry Select. 2022 Apr 12;7(14):e202104196. doi: 10.1002/slct.202104196.
28. Mannava MK, Gunnam A, Lodagekar A, Shastri NR, Nangia AK, Solomon KA. Enhanced solubility, permeability, and tabletability of nicorandil by salt and cocrystal formation. Cryst Eng Comm. 2021;23(1):227-37. doi: 10.1039/D0CE01316A.