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Solubility and Dissolution Enhancement of Ramipril by Nanocrystallization

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Abstract: Drug nanocrystals are solid drug particles that have a mean diameter smaller than 1000 nm. The term "Nanocrystal" is also a trademark owned by Elan Pharma International Ltd, based in Ireland. Drug nanocrystals are a crucial approach for improving the capacity of poorly soluble medications to be absorbed when taken orally. Nanocrystallization shown to be an effective strategy for improving the solubility characteristics of Ramipril using the emulsion solvent diffusion method. Based on the solubility and *in vitro* dissolution experiments, it is evident that the nanocrystal formulations have the potential to enhance the bioavailability of Ramipril by enhancing its solubility and dissolving rate in comparison to the pure medication. Therefore, the utilization of a nanocrystal drug delivery system can be employed to augment the solubility and dissolving rate of a weakly soluble molecule such as Ramipril, thereby improving its bioavailability.

Keywords: Ramipril, Solubility, Dissolution, Nanocrystal

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Introduction

"Nano" means "dwarf" in Latin, which is where the phrase "nanotechnology" comes from. About the width of six carbon atoms or ten water molecules is one nanometer (nm), which is one-billionth of a meter. The width of a human hair is around 80,000 nanometers, while the width of a red blood cell is about 7,000 nanometers. Contrasted with atoms, which are smaller than 1 nm, many molecules, including proteins, are between 1 nanometer and bigger in size (Ahire *et al*., 2018; Ahirrao *et al*., 2022)

In his 1959 speech, "There's plenty of room at the bottom," physicist Richard Feynman initially laid out the conceptual foundations of nano-technologies. Feynman speculated on the prospect of controlling matter at the nanoscale and envisioned the entire Encyclopedia Britannica printed on the tip of a pin, among other things, as he investigated the potential of managing material at the scale of individual atoms and molecules (Keservani and Sharma 2018a; Keservani *et al*., 2018). The capacity to manipulate materials down to the nanoscale scale was first described by Norio Taniguchi of the University of Tokyo in 1974, who coined the term nanotechnology. Back then, miniaturization was mostly driven by the electronics industry's quest for better tools to make smaller, more complicated, and faster electronic devices on silicon chips (Ahirrao *et al*., 2023).

Nanotechnology has molecular-level applications in pharmaceutics and medicine, where it is employed to improve human health. Novel and potentially useful uses of nanotechnology in pharmaceutics include gene therapy, drug carrier system formulation, and diagnostic tool creation. The size of the particles is the main factor that makes nanotech medications superior to their traditional equivalents. Nano-sized drugs and therapeutic products have the potential to cause bioactivity to begin at a lower concentration and at an earlier stage. Some examples of nano drug delivery systems are solid lipid nanoparticles, nanovesicular systems, molecular systems, nanocrystals, nanospheres, nanosponges, and nanoemulsions (Ahire *et al*., 2020).

The rationale behind this study was that, in the field of pharmaceutical R&D, numerous cutting-edge technologies, including computer-aided drug design, combinatorial chemistry, and high throughput screening, have produced an abundance of promising new drug candidates within the past 20 years. There is a problem with the aqueous solubility of several of these potential drugs. There are methods that can improve the solubility of medications that are not very water-soluble, but they come with limitations including high dosage requirements and low drug loading. Nevertheless, nanonization has emerged as a viable alternative to previously unavailable methods of improving the solubility, bioavailability, and safety of medicines with low water solubility (Ahire and Kshirsagar, 2023).

The liver esterase enzyme converts the prodrug ramipril to the active metabolite ramipril. Hypertension and congestive heart failure are treated with this medication, which is an ACE inhibitor. By blocking the action of the angiotensin converting enzyme, ramipril reduces the synthesis of angiotensin II from angiotensin I. Consequently, the heart is able to pump blood more easily into big passageways by relaxing the muscles of the arteries and simultaneously widening them. When taken orally, Ramipril has an absolute bioavailability of 28-30%. Methyl alcohol and water both dissolve it to varying degrees (Keservani *et al*., 2023).

Pure solid medication particles having an average diameter less than 1000 nm are known as drug nanocrystals (Keservani and Sharma, 2018b). The Noyes-Whitey equation states that the drug dissolution rate is proportional to the square of the particle size because a smaller particle has a larger effective surface area in the diffusion layer. Another Irish company, Elan Pharma International Ltd., owns the trademark Nanocrystal. To increase

the oral bioavailability of scarce water-soluble medicines, drug nanocrystals are a crucial strategy. Nanocrystals of pharmaceuticals can be utilized to stabilize drugs that are sensitive to chemical changes. The surfactant shield effect and the drug protection monolayer composed of degraded drug molecules explain the enhanced stability by reducing the accessibility for destructive agents (Ahirrao *et al*., 2023).

By reducing the drug's particle size, this study aimed to build nanocrystal of Ramipril, which will increase the drug's solubility and bioavailability. The emulsion solvent diffusion process is used to manufacture drug nanocrystals. The resulting nanocrystals' solubility and dissolution characteristics were evaluated in comparison to that of the pure medicine.

Materials and Methods

Standard curves for Ramipril:

To prepare a primary stock solution, dissolved a known weight of medication in an adequate quantity of methanol in a 100 ml volumetric flask. Top off the flask with 100 ml of phosphate buffer pH 6.8. A phosphate buffer solution with a pH of 6.8 was used to further dilute the stock solution until it reached a concentration of 10 µg/ml. The absorption maximum was obtained by scanning the resulting solution in the UV Spectrophotometer's range.

Preparation of standard curves:

Concentration solutions are made from the stock solution using the phosphate buffer pH (6.8) solution, which is manufactured as described above. The UV- spectrophotometer was used to measure the absorbance of these solutions at λ max. By plotting the absorbance on the Y-axis and the concentration on the X-axis, a standard curve was prepared (Chander *et al*., 2011).

Preformulation studies:

To assess the drug-stabilizer interaction, infrared spectrophotometer experiments were conducted prior to formulation. The IR spectrophotometer was used to acquire the infrared spectra of the medication, stabilizers, and physical mixes of these substances. The KBr-press was used to manufacture the pellets under a hydraulic pressure of 150 kg/cm^2. At room temperature, the spectra are scanned over the wave number range of 4000 to 400 cm^{-1} (Sinco *et al*., 2011).

Formulation of Ramipril nanocrystals:

The nanocrystals are synthesized using the emulsion solvent diffusion process. The preparation procedure entails the following steps (Phanchaxari *et al*., 2011).

Formulation of Ramipril nanosuspensions:

Preparation of drug solution:

The drug solution was prepared by adding a precisely weighed sample (100 mg) of the medication to 10 ml of methanol. After the medication solution has been created, added 10 ml of water containing stabilizers and mixed continuously on a homogenizer set to 1000 rpm for 2 h. Stirring continuously at 500 rpm for 3-4 h removed the organic solvent (Phanchaxari *et al*., 2011).

Lyophilization of nanosuspensions to obtain the nanocrystals:

To improve the chemical stability of nanocrystals, ramipril nanosuspensions were lyophilized using a freeze dryer. The lyophilization process involves adding a cytoprotective agent to the freshly made nanosuspensions. To summarize, the Ramipril nanosuspensions undergo a two-hour rapid cooling process to -500 °C, followed by primary and secondary drying at 1.03 mbar and 0.001 mbar, respectively (Rainer Muller *et al*., 2008).

Characterization of Ramipril nanocrystals:

In vitro drug release investigations, drug content, zeta potential, particle size, and all of the formulations undergo evaluation.

Determination of drug content:

Weighed 10 mg of Ramipril nanocrystals equivalent, dissolved them in methanol, and then added 100 ml of phosphate buffer pH 6.8 to get the final volume. 10 ml of the aforementioned solution

was diluted with 100 ml of phosphate buffer (pH 6.8). The drug content was assessed by determining the absorbance of the resultant solution at λ max (277 nm) using a UV Spectrophotometer (Phanchaxari *et al*., 2011).

In vitro dissolution studies:

For the purpose of conducting *in vitro* drug dissolving tests on all nanocrystal formulations, the USP dissolution device Type II was rotated at a speed of 100 rpm. With each batch, 900 ml of phosphate buffer pH 6.8 was used as the dissolution media for 10 mg equivalent Ramipril nanocrystals in capsules. Throughout the investigation, the bath temperature was kept at 37±0.5°C. Five milliliters of the solution was taken out of the dissolving device at 5,10, 20, 30, 40, 60, 90, and 120 min intervals. New dissolving media was added to the samples. The UV Spectrophotometer measured the sample solutions' absorbance values at λ max, which was 277 nm. It determined the cumulative proportion of medication release**.**

Determination of Particle size and Zeta potential:

The zeta size analyzer and dynamic light scattering technique were used to determine the mean particle size and zeta potential of Ramipril nanocrystal formulations. To get the right scattering intensity before measuring, the freeze-dried powders were redissolved in water (Dianrui Zhang *et al*., 2012).

Solubility studies:

Ramipril nanocrystal formulations' solubility was investigated in various solvents, including phosphate buffer pH (6.8) and distilled water. 10 ml of the appropriate solvents were mixed with an excess of nanocrystal formulation. For 24 h, the ingredients were shaken vigorously in a mechanical shaker. Careful visual inspection was carried out to confirm that the mixture has an excess of Ramipril solids, which indicated that saturation has been achieved. To find out how much Ramipril dissolved in each solvent, the combinations were filtered and the filtrates are diluted appropriately (Abdul Hasan and Gopinath, 2013).

Selection and evaluation of best formulation:

Particle size, *in vitro* drug release, and solubility tests all contributed to the final formulation decision.

Infrared spectroscopic studies:

The chosen nanocrystal formulation was subjected to infrared spectroscopy in order to determine the drug-excipient interactions. The KBr-press was used to manufacture the pellets under a hydraulic pressure of 150 kg/cm^2. At room temperature, the spectra were scanned over the wave number range of 4000 to 400 cm^{-1} (Sinco *et al*., 2011).

Morphological studies of nanocrystals by using Scanning Electron Microscopy:

The scanning electron microscope was used to evaluate the morphology of the chosen Ramipril nanocrystal formulation. On a brass stub, using carbon double-sided tape, all samples were checked. Metal sample plates were used to bond and mount powder samples. An electrical potential of 2.0 kV at 25 mA for 10 min was used to sputter coat the samples with gold. In the trials, a voltage of 20 kV was employed for excitation (Yuan Gao *et al*., 2011).

X-ray Powder Diffraction analysis:

The medication and freeze-dried powder samples were examined using an X-ray diffractometer to determine their crystalline state. With a wavelength of 1.5405A˚ and a copper line as the radiation source, XRPD was performed in symmetrical reflection mode. This procedure was conducted according to standard employing a 40 kV and 30 mA. Scan rates of 0.1000◦/min were used for the samples, and the 2θ scanning range was from an initial angle of 4˚ to a final angle of 90˚ (Dianrui Zhang *et al*., 2012).

Results and Discussion

Standard Curves for Ramipril:

The pH of the calibration medium was 6.8 and it was made according to the I.P. process using phosphate buffer. The scanning of a 10 µg/ml drug

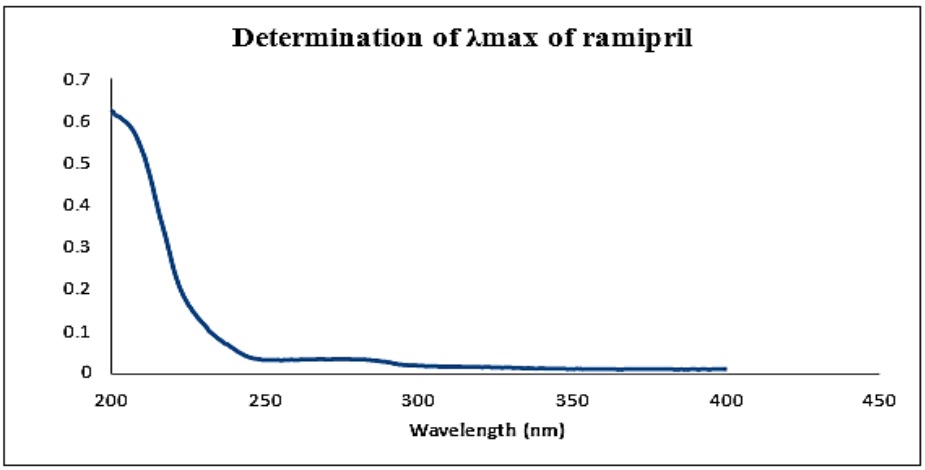

Fig. 1: λ max of ramipril using phosphate buffer pH (6.8).

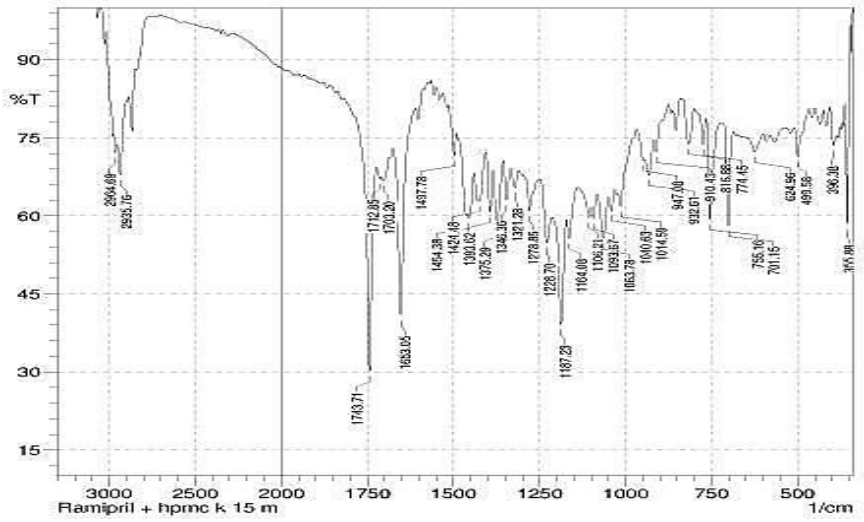

Fig. 2: IR spectrum of HPMC K15M and ramipril.

solution in a buffer solution with a pH of 6.8 was used to estimate the λ max of ramipril. The λ max was observed at 277 nm in a buffer solution with a pH of 6.8 (Fig. 1) (Chary *et al*., 2012). Phosphate buffer pH (6.8) was used to prepare the standard curves of ramipril. It was discovered that for pH 6.8, the linear correlation coefficient was 0.9995. Within a concentration range of 5 to 25 µg/ml, Ramipril abides under Beer's law.

Preformulation studies:

The calibration medium, which was prepared using phosphate buffer in accordance with the I.P. procedure, had a pH of 6.8. To determine the maximum concentration of ramipril, a buffer solution with a pH of 6.8 was used to scan a drug solution with a concentration of 10 µg/ml. In a buffer solution with a pH of 6.8, the maximum optical density (λmax) was recorded at 277 nm (Fig. 1) (Chary *et al*., 2012). Ramipril standard curves were prepared using a phosphate buffer with a pH of 6.8. The results showed a linear correlation coefficient of 0.9995 at a pH of 6.8. Ramipril follows Beer's rule when its concentration is between 5 and 25 µg/ml. Further drug authentication and interaction study was performed and results are depicted in Table 1 and Figures 2 - 6.

Formulation of Ramipril nanocrystals:

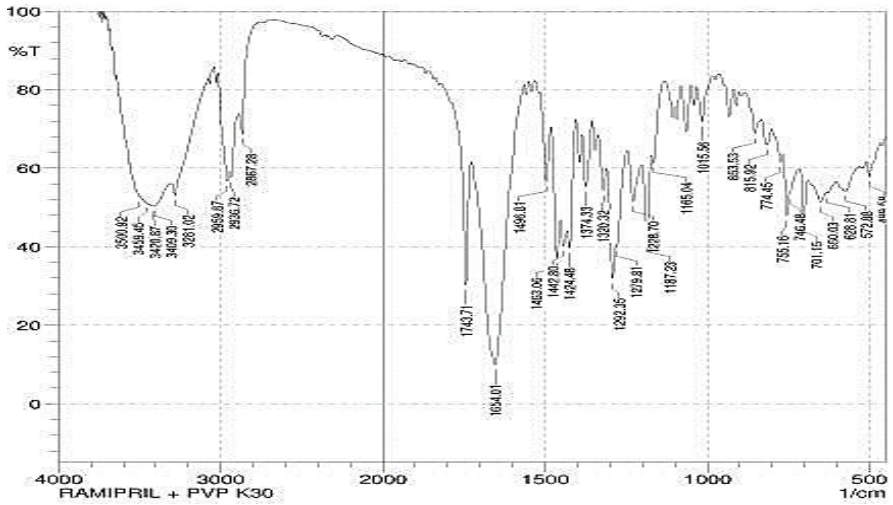

Fig. 3: IR spectrum of PVP K30 and ramipril.

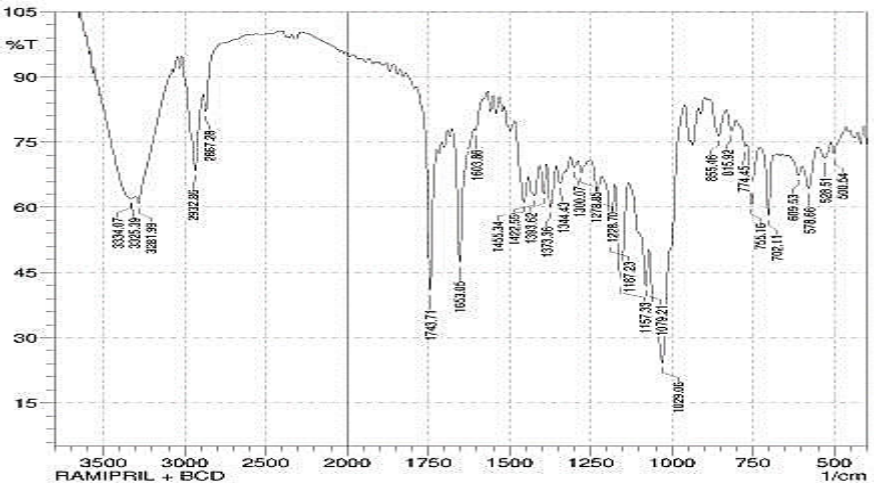

Fig. 4: IR spectrum of β-cyclodextrin and ramipril.

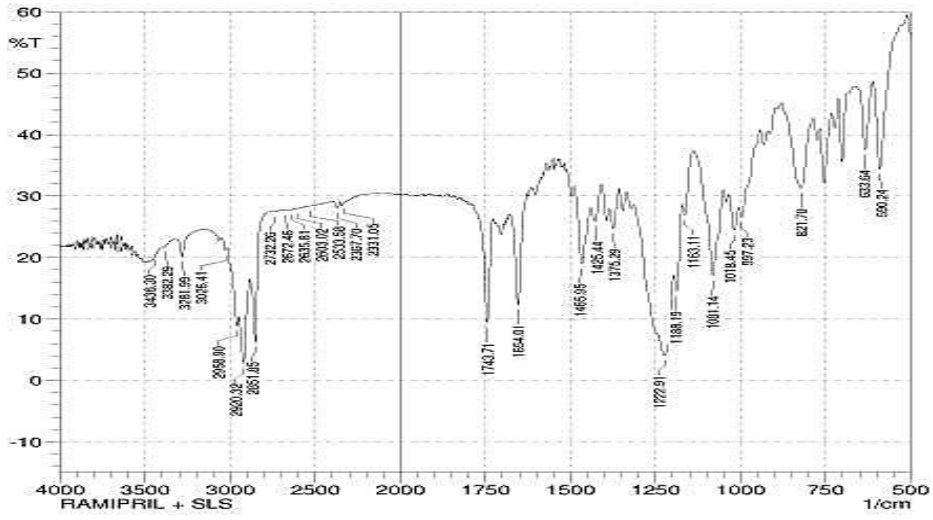

Fig. 5: IR spectrum of SLS and ramipril.

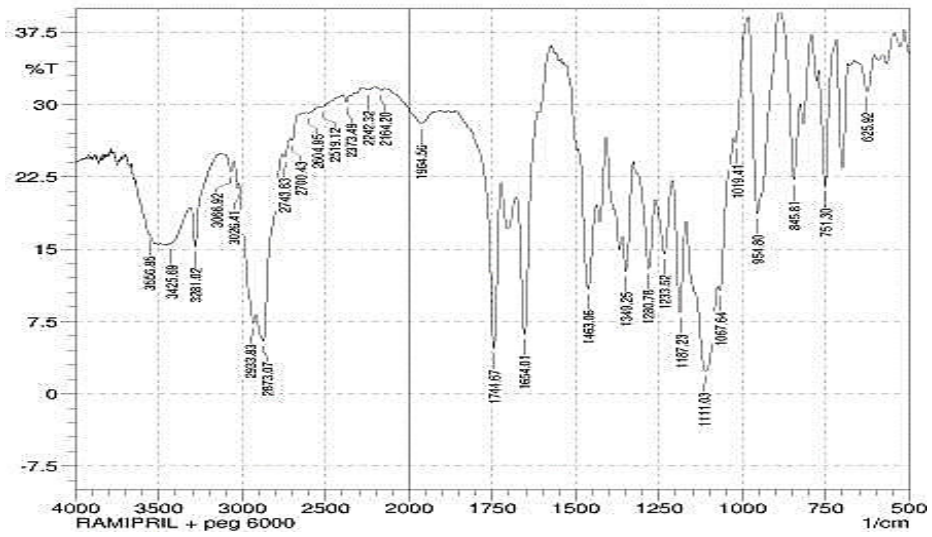

Fig. 6: IR spectrum of PEG 6000 and ramipril.

Table 1: IR peaks of a drug, stabilizers, physical mixture of Drug with stabilizers and formulations

S. No	Description	Characteristic peaks (cm^{-1}) obtained
1.	Ramipril	1373.36, 1228.70, 1063.78, 815.92, 753.23, 701.15
2.	HPMC K15M	3642.69, 2934.79, 1650.16, 1459.20, 945.15
3.	PVP K30	3748.78, 3398.69, 2956.01, 1667.52,1502.60
4.	β- cyclodextrin	1490.06, 1457.27, 1363.72, 948.04, 651.00, 431.10
5.	SLS	1654.01, 1470.77, 1248.95, 834.24, 634.60, 591.20
6.	PEG 6000	2165.17, 1650.16, 1469.81, 1240.27, 961.55
7.	Ramipril +HPMC K15M	2935.76, 1743.71, 1187.23, 755.16, 355.88
8.	Ramipril + PVPK30	2867.28, 1654.01, 1442.80, 1187.23, 701.15
9.	Ramipril+ β-CD	2932.86, 1653.05, 1157.33, 1029.06, 702.11
10.	Ramipril+ SLS	2920.32, 1743.71, 1654.01, 1465.95, 1081.44
11.	Ramipril + PEG 6000	3281.02, 1349.25, 1280.78,845.81, 751.30

According to Phanchaxari *et al*. (2011), the ramipril nanocrystals were created using the emulsion solvent diffusion process. The active medicinal ingredient was first dissolved in an organic solvent before being introduced to a non-solvent. This was the basic idea behind the procedure. Nanocrystals were precipitated after being exposed. The precipitation technique's main selling point was how inexpensive and easy it was to implement. According to Phanchaxari *et al*. (2011), this strategy also made scaling up easy (Baig *et al*., 2018, Gujarathi *et al*., 2023). The following procedure was used to manufacture ramipril nanocrystals: Different stabilizers such as HPMC K15M, PVP K30, β Cyclodextrin, SLS, and PEG 6000 were used to generate different formulations of ramipril nanocrystals (B1 - B20) at varied concentrations, as indicated in Table 1.

Characterization of Ramipril nanocrystals:

The drug content, particle size, polydispersity index, zeta potential, *in vitro* drug release, and solubility of each formulation were assessed.

Determination of drug content:

Table 2: Composition of ramipril nanocrystals

S. No.	Formulation Code	Drug Concentration (mg/10 ml)	Stabilizers Concentration (%)
1	B1A	100 mg in 10 ml	HPMC K15M (0.1%)
2	B1B	100 mg in 10 ml	HPMC K15M (0.2%)
3	B1C	100 mg in 10 ml	HPMC K15M (0.3%)
4	B1D	100 mg in 10 ml	HPMC K15M (0.4%)
5	B1E	100 mg in 10 ml	HPMC K15M (0.5%)
6	B2A	100 mg in 10 ml	PVP K30 (0.1%)
7	B2B	100 mg in 10 ml	PVP K30 (0.2%)
8	B2C	100 mg in 10 ml	PVP K30 (0.3%)
9	B2D	100 mg in 10 ml	PVP K30 (0.4%)
10	B2E	100mg in 10ml	PVP K30 (0.5%)
11	B3A	100 mg in 10 ml	BCD (0.1%)
12	B3B	100 mg in 10 ml	BCD (0.2%)
13	B3C	100 mg in 10 ml	BCD (0.3%)
14	B3D	100 mg in 10 ml	BCD (0.4%)
15	B3E	100 mg in 10 ml	BCD (0.5%)
16	B4A	100 mg in 10 ml	SLS (0.1%)
17	B4B	100 mg in 10 ml	SLS (0.2%)
18	B4C	100 mg in 10 ml	SLS (0.3%)
19	B4D	100 mg in 10 ml	SLS (0.4%)
20	B4E	100 mg in 10 ml	SLS (0.5%)

From B1 to B20, the drug content varied between 88.00% and 96.00% (Table 2). According to the findings, the method used to create the nanocrystals ensured that the medication was evenly distributed.

In vitro dissolution studies:

The *in vitro* dissolution experiments of all formulations were compared to the pure medication. The drug release investigations conducted on the ramipril nanocrystals yielded data that are presented in Table 3 Figures 7-10. The *in vitro* release profile of all the formulations is much higher than that of the pure medication ramipril when compared. The formulations (B1A – B1E) that were created with varying

Table 3: Nanocrystals of the drug Ramipril

S. No.	Stabilizers Concentration	Formulation Code	Avg ± SD
1.	HPMC K15M (0.1%)	B1A	93.63±1.6226
2.	HPMC K15M (0.2%)	BIB	89.63±1.9561
3.	HPMC K15M (0.3%)	B1C	89.70±1.3856
4.	HPMC K15M (0.4%)	B1D	88.00±0.0000
5.	HPMC K15M (0.5%)	B1E	89.50±2.0621
6.	PVP K30 (0.1%)	B2A	92.86±1.8013
7.	PVP K30 (0.2%)	B2B	92.00±1.6055
8.	PVP K30 (0.3%)	B2C	90.33±1.0237
9.	PVP K30 (0.4%)	B2D	93.66±1.1547
10.	PVP K30 (0.5%)	B2E	93.00±1.4641
11.	BCD (0.1%)	B3A	95.86±0.5011
12.	BCD (0.2%)	B3B	95.33±1.5166
13.	BCD (0.3%)	B3C	91.66±1.1547
14.	BCD (0.4%)	B3D	95.33±1.5166
15.	BCD (0.5%)	B3E	93.00±1.0000
16.	SLS (0.1%)	B4A	90.66±1.5166
17.	SLS (0.2%)	B4B	91.66±1.1547
18.	SLS (0.3%)	B4C	94.33±1.1547
19.	SLS (0.4%)	B4D	92.33±1.1547
20.	SLS (0.5%)	B4E	96.00±1.7320

concentrations of stabilizer exhibited drug release percentages of 53.39%, 61.10%, 67.97%, 71.12%, and 74.81% at 120 min, respectively. The formulations (B2A - B2E) containing varying concentrations of stabilizer exhibited drug release percentages of 73.29%, 74.04%, 75.68%, 75.67%, and 78.78% at 120 min, respectively. The formulations (B3A - B3E) were created with varying concentrations of stabilizer (β-cyclodextrin) ranging from 0.1% to 0.5%. The percentage of medication released at 120 min for each formulation was 75.69%, 77.98%, 81.12%, 81.12%, and 83.43%, respectively (Singh and Keservani, 2017).

The drug's release rate from the nanocrystals was enhanced by increasing the concentration of stabilizers. The higher percentage of drug release observed in formulations B1A to B1E, which contained the stabilizer HPMC K15M, suggests that this water-soluble polymer was used to stabilize the suspension. HPMC K15M possesses surface active properties that are sufficient for this purpose and contribute to the increased drug release (Hecq *et al*., 2005). The higher percentage of drug release observed with the addition of stabilizer PVP K30 in formulations B2A to B2E suggests that PVP K30, a water-soluble chemical, was employed to enhance the pace at which the medication dissolves (Shahbaziniaz *et al*., 2013).

The higher drug release percentage observed in formulations B3A to B3E suggests that the stabilizer β-cyclodextrin forms a network through intermolecular interactions. This network provides protection and allows the stabilizer to self-associate in water, forming nano-scale aggregates with a small hydrodynamic radius. This, in turn, enhances the rate at which the drug dissolves (Phanchaxari *et al*., 2011).

According to Phanchaxari *et al*. (2011), the presence of SLS as a dispersion stabilizer in the formulation (B4A to B4E) increased the percentage of drug release. This is because SLS increases the activation energy, which prevents the agglomeration of precipitated nanocrystals,

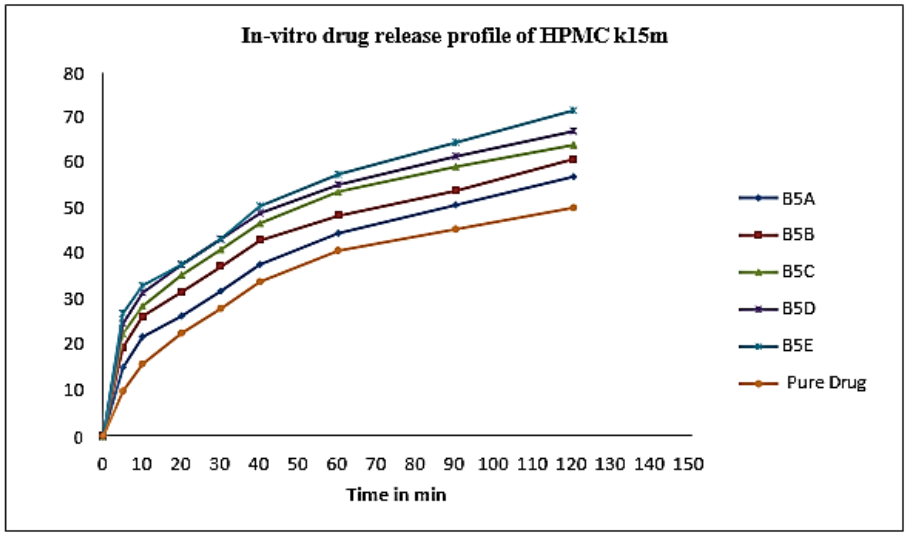

Fig. 7: *In vitro* dissolution release profile of ramipril nanocrystals stabilized by HPMC k15m.

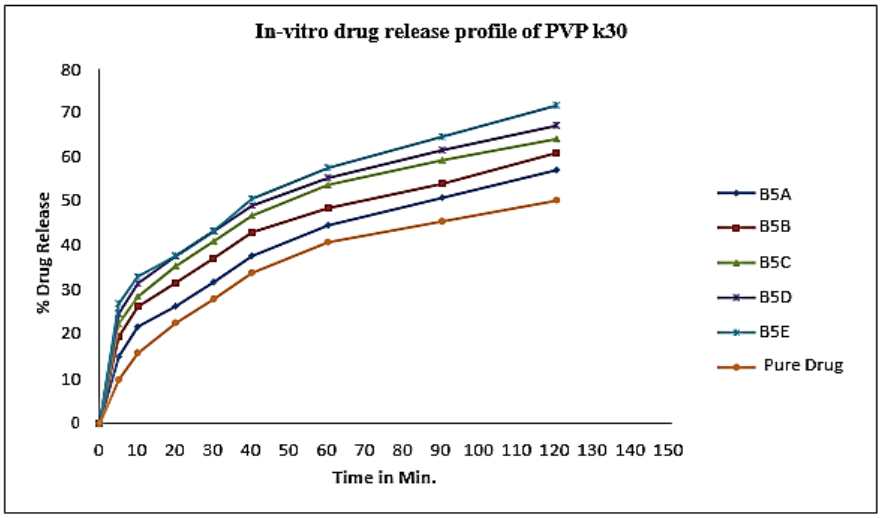

Fig. 8: *In vitro* ramipril nanocrystal dissolution release profile comparison Contains stabilizer PVP k30.

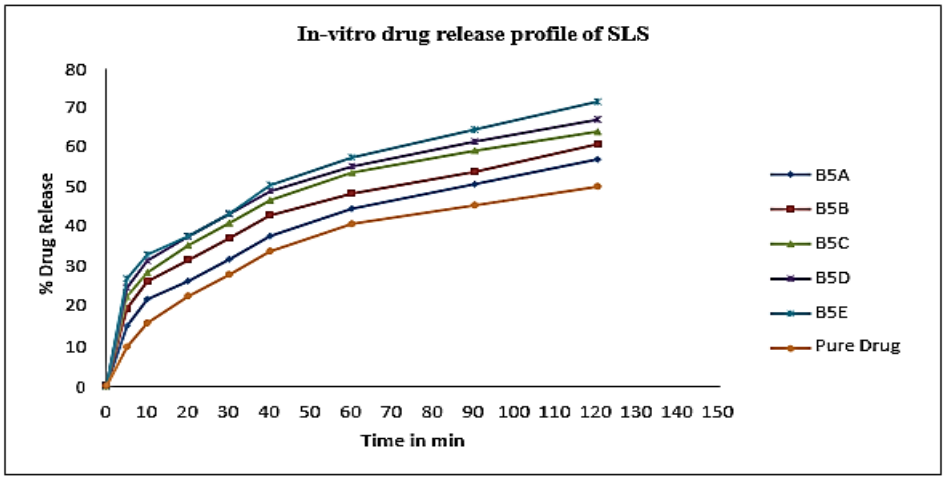

Fig. 9: Compare *in vitro* dissolution release profile of ramipril nanocrystals with SLS stabilizer.

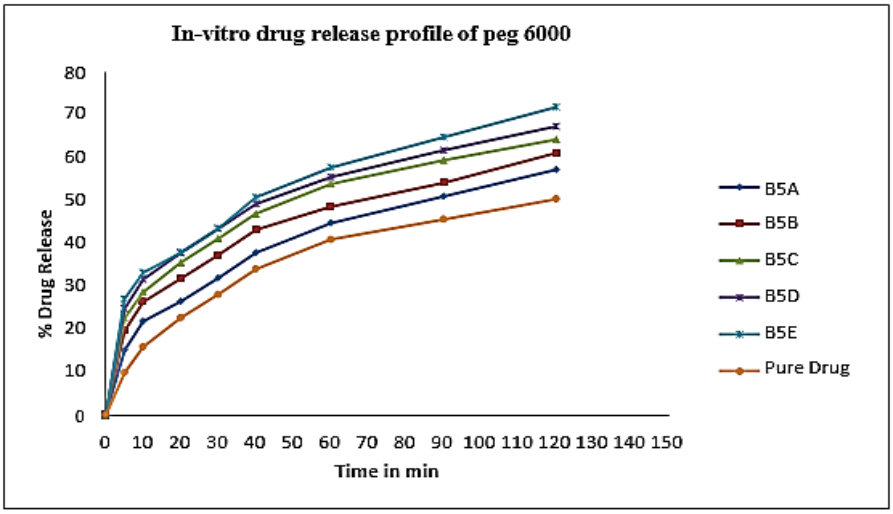

Fig. 10: Ramipril nanocrystals with peg 6000 stabilizer dissolution release profile *in vitro*.

and it provides wettability to the drug particles, which reduces the surface tension between them and the solvent. As a result, the drug dissolving rate is improved. According to Peng Liu *et al*. (2011), the stabilizer PEG 6000 improved the drug dissolution rate by capturing water molecules through hydrogen bonding. This was supported by the increased percentage drug release in formulations B5A to B5E, which were characterized by long hydrophilic chains of PEG.

Determination of Particle size and Zeta potential:

Particle size:

The nanocrystals' particle size, size distribution, and zeta potential are crucial factors that determine other characteristics, such as saturation solubility, dissolution velocity, physical stability, and even biological performances (Dianrui Zhang et al., 2012). The mean diameters and polydispersity index of ramipril nanocrystals are depicted in Figures 11, 12 and 13, respectively. The current study observed that the particle size of ramipril nanocrystals ranged from 80.3 nm to 300.6 nm.

Zeta potential:

The zeta potential of the nanocrystals enabled predictions regarding the storage stability of colloidal dispersions. Typically, nanocrystals can achieve physical stability by maintaining a zeta potential value of at least ±30 mV through the use of ionic surfactants, which create electric repulsion. However, a zeta potential of approximately ±20 mV can also indicate long-term stability of the system when nonionic surfactants are used to create steric hindrance. Furthermore, the specific type of a key variable had a significant impact on the average zeta potential value (Dianrui Zhang et al., 2012). The ramipril nanocrystals were analyzed to assess the impact of stabilizers on the surface charge of the nanocrystals, using various ratios and types of stabilizers. The results are displayed in Figure 13. The formulations code produced with varied revealed negative zeta potential values ranging from -19.7mV to -24.7mV, indicating a stable preparation.

Solubility studies:

The solubility of the pure medication and selected formulation is presented in Figure 14. Nanocrystal formulations exhibited superior solubility in distilled water when compared to the pure medication. The solubility of formulations and pure medication in phosphate buffer at pH 6.8 were 1.177 mg/10 ml, 2.791 mg/10 ml, 5.680 mg/10 ml, 2.294 mg/10 ml, 5.182 mg/10 ml, and 0.547 mg/10 ml, respectively. The solubility of ramipril in nanocrystal formulations was

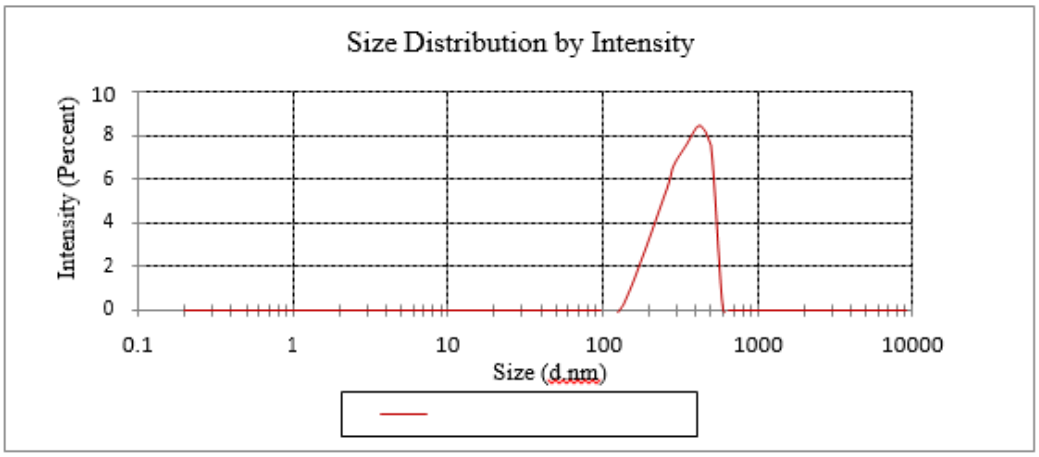

Fig. 11: Formulations B3C, B3D, and B3E particle size distribution curve.

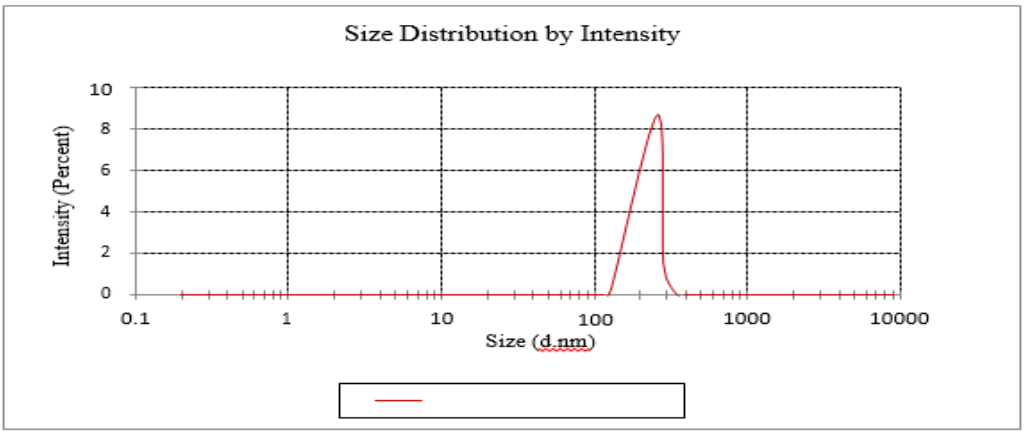

Fig. 12: Formulations B4D and B4E particle size distribution curve.

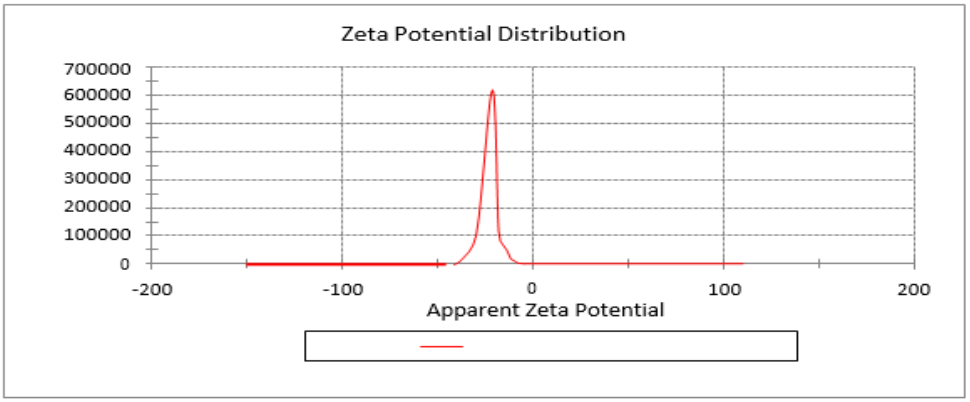

Fig. 13: Potential curve of zeta for formulations.

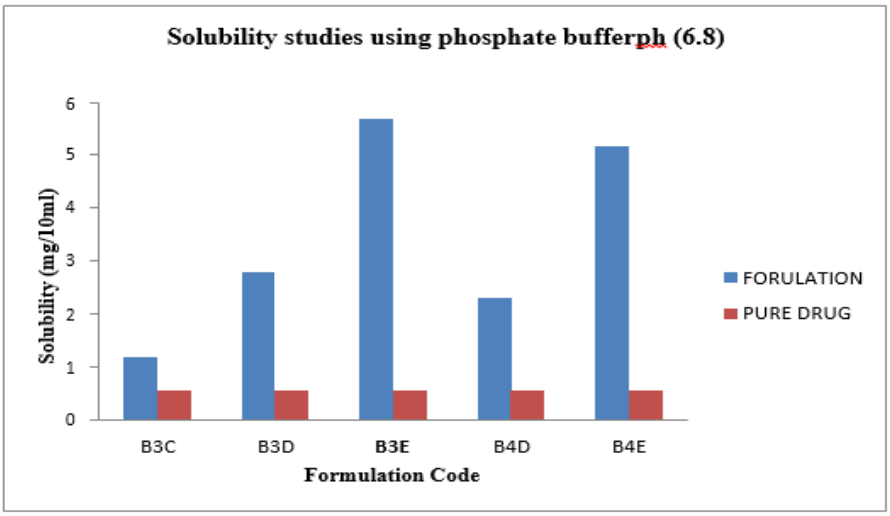

Fig. 14: Solubility of chosen formulations vs. pure medication.

enhanced by a factor of ten compared to the pure drug. Therefore, the significant increase in the solubility of ramipril in the nanocrystal formulation can be primarily due to the reduction in particle size and the corresponding increase in surface area. The results can be elucidated by the Ostwald–Freundlich equation, which illustrates that the saturation solubility of the drug augments as the particle size decreases (Dianrui Zhang *et al*., 2012).

Finding the optimal formulation:

The two formulations listed below were determined to have the most promising properties based on the characterization data provided above.

Infrared spectroscopic studies:

Research using infrared spectroscopy was conducted on various nanocrystal formulations, stabilizers, and pure drugs. Displayed here are the spectra examined between 4000 cm^{-1} and 400 cm^{-1}. The spectra of the medication, stabilizers, and chosen formulations showed no significant shifting and no loss of functional peaks. It was evident from this that the drug was present in its unaltered form and that there was no interaction between it and the polymer.

SEM nanocrystal morphology:

Scanning Electron Microscopy was used to analyze the morphology of specific nanocrystal compositions. The scanning electron microscopy (SEM) picture of the formulation is displayed in Figure 15. The Ramipril nanocrystal formulations (B3E and B4E) exhibited small, homogeneous particles with a crystal shape. The average particle size was less than 1 µm. This data was consistent with the data acquired from the Malvern zeta sizer analysis conducted by Rainer Muller *et al.* (2008).

X-ray Powder Diffraction analysis:

The X-ray powder diffraction (X-RPD) patterns of the pure medication and formulations are shown in Figure 16. The X-ray powder diffraction (X-RPD) patterns of the pure substance exhibited multiple distinct peaks, indicative of its crystalline nature. Formulation also exhibited detectable drug crystallinity peaks. This outcome validated that the distinctive peaks remained intact, signifying that the crystalline state remained unaltered. It is widely understood that the amorphous form of pharmaceuticals can increase their solubility rate and bioavailability because of its high-energy nature. Based on that concept and taking into account the XRPD study, the increased rate of dissolution of Ramipril may be attributed to the decrease in particle size or the impact of stabilizers, rather than the presence of an

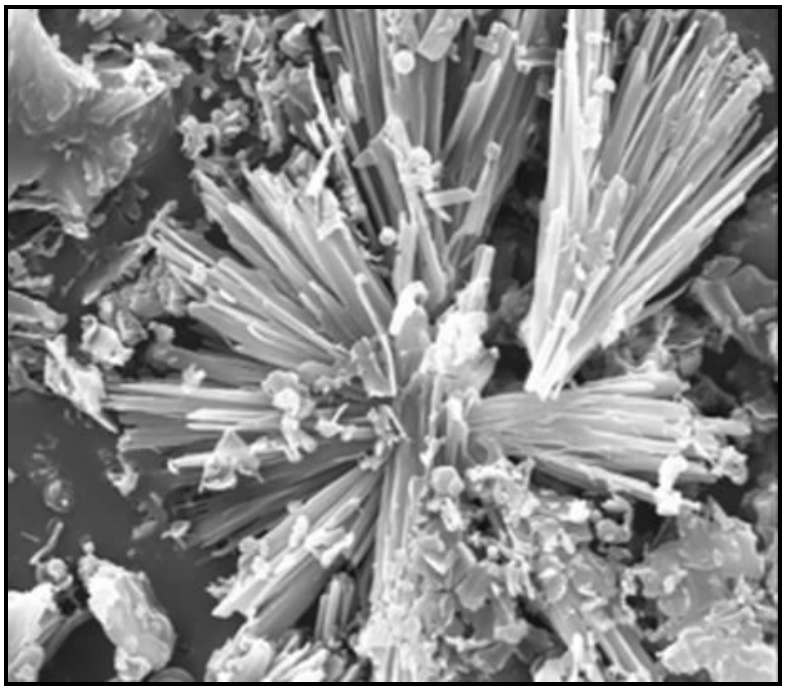

Fig. 15: SEM picture of optimal formulation.

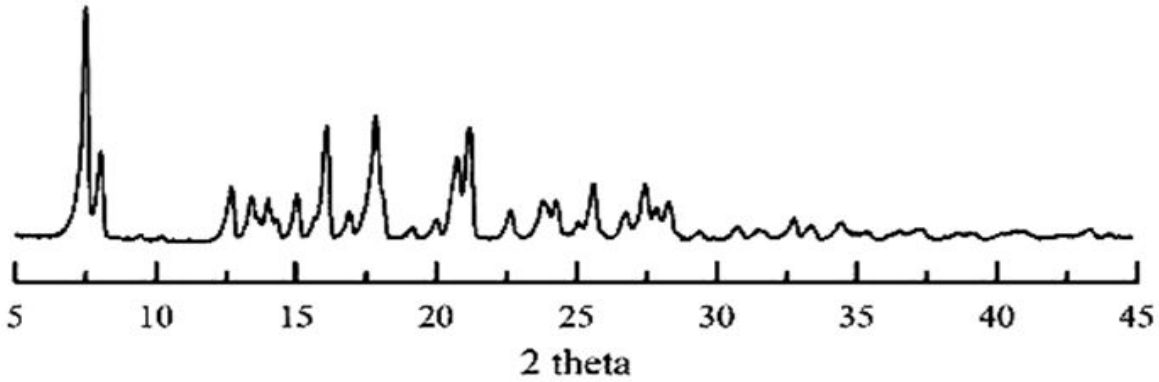

Fig. 16: Ramipril pure medication X-ray diffraction.

amorphous form. In addition, maintaining the crystalline state was shown to be advantageous for long-term stability, as stated by Dianrui Zhang *et al.* (2012) and Keservani *et al*. (2016, 2017).

Conclusion

Therefore, it was determined that nanocrystallization was an effective strategy for improving Ramipril's solubility through the emulsion solvent diffusion process. Nanocrystal formulations of Ramipril have the potential to increase the medicine's bioavailability compared to the pure molecule, according to *in vitro* dissolution and solubility tests. Nanocrystal drug delivery systems can improve the bioavailability of drugs with low solubility, such as ramipril, by increasing their dissolving rate and increasing their solubility.

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