RESEARCH ARTICLE

Nanocrystal Formulation of Manidipine HCl: An Attempt to Enhance Solubility of Poorly Soluble Drug

Jadhav A B[1*], Nawale R B[2], Thorat S[3]

[*1]*Department of Pharmaceutics, Government College of Pharmacy, Chatrapati Sambhajinagar, 431005, Maharashtra, India.*
[2]*Government College of Pharmacy, Chatrapati Sambhajinagar, 431005, Maharashtra, India.*
[3]*Department of Research and Development, Paonta Sahib, Sirmour, Himachal Pradesh, India.*

Received: 28th Oct 2024; Revised: 29th Nov 2024; Accepted: 6th Dec 2024; Available Online: 25th Dec, 2024

ABSTRACT

Background: Manidipine Hydrochloride (MND) is a long-acting dihydropyridine-type calcium antagonist and antihypertensive drug categorized under the Biopharmaceutics Classification System (BCS) Class II. Despite its therapeutic benefits, MND suffers from poor physicochemical and biological properties due to its high lipophilicity and low aqueous solubility, which pose challenges to formulation and drug delivery. Improving the solubility of such hydrophobic drugs is crucial for enhancing their bioavailability. While conventional methods for solubility enhancement exist, drug nanocrystal systems have emerged as a promising approach for addressing solubility issues in poorly soluble drugs.

Objective: To formulate and evaluate nanocrystals of MND using spray-drying technology to enhance its solubility and address limitations associated with its hydrophobicity.

Method: MND nanocrystals were prepared using a spray-drying technique, a cost-effective and scalable method that converts liquid formulations into dry powder through atomization and drying processes. The spray-dried nanocrystals were characterized by their particle size, morphology, solubility, and dissolution rate. Comparative studies evaluated the solubility enhancement achieved with the nanocrystals versus raw MND.

Result: The nanocrystals demonstrated a significant reduction in particle size and enhanced surface area, resulting in improved aqueous solubility and dissolution rates compared to raw MND. Spray-drying facilitated the uniform formation of nanocrystals, ensuring better dispersion and dissolution performance.

Conclusion: Spray-dried MND nanocrystals proved to be a viable strategy for overcoming solubility and bioavailability challenges. This approach offers a promising avenue for enhancing the clinical efficacy of poorly soluble drugs like MND.

Keywords: Nanocrystals, Spray drying, Solubility, Manidipine.

How to cite this article: Jadhav A B, Nawale R B, Thorat S. Nanocrystal Formulation of Manidipine HCl: An Attempt to Enhance Solubility of Poorly Soluble Drug. International Journal of Drug Delivery Technology. 2024;14(4):2379-86. doi: 10.25258/ijddt.14.4.59

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

Traditionally, hypertension has been described as persistently high (BP). The main reason for early mortality and morbidity worldwide is that blood pressure is 140/90 mm Hg for all reasons. Calcium antagonists have been utilized extensively since their introduction to treat a variety of cardiovascular disorders, including hypertension.[1] MND dihydropyridine Ca channel antagonist is a 3rd generation that is lipophilic and highly selective for the vasculature, leading to significant peripheral vasodilation and minimal cardio-depression. Solubility is the capacity of a chemical molecule, known as a solute, to dissolve solvent and produce a uniform mixture of the substance in the vehicle.[2] It may be measured in terms of the solute's ability to dissolve in the solvent. Several methods and techniques are applied to get better solubility of drugs belonging to the Biopharmaceutics Classification System 2 category, which shows poor water solubility. Nanotechnology may be applied to enhance the solubility of BCS class II drugs.[3]

MATERIAL AND METHOD

MND was obtained as a gift sample from Manus Aktteva, Biopharma LLP, Ahmedabad, Gujarat India.

List of Instruments

HPLC (Agilent 1260 Infinity II), Column (Agilent Technologies), Ultra sonicator (Labman Scientific Instruments), UV Spectrophotometer (Labman 1900), Spray Dryer B-90.

Methodology

Physiochemical characterization of MND solubility enhancement development

MND API powder was kept on a watch glass and observed for its color against a dark background. Its texture and color were scrutinized.

Solubility studies

**Author for Correspondence: amrapalipharma@gmail.com*

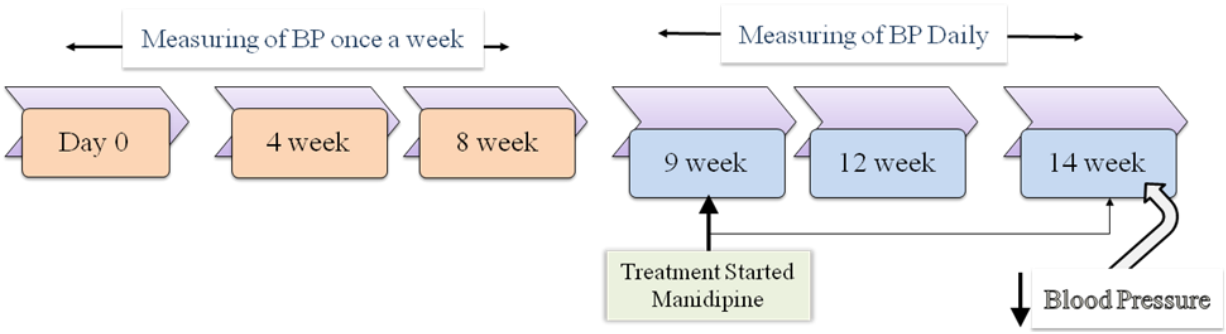

Figure 1: Study design intervention

Table 1: Solubility studies

Sr no.	Solubility	mg/Lit
a.	Water	0.995
b.	Ethanol	15640
c.	Methanol	14978
d.	Acetone	12637
e.	Isopropyl Alcohol	9745
f.	Toluene	6783
g.	Acetonitrile	11012
h.	Tetrahydrofuran	8930
i.	0.1N Hydrochloric acid	215
j.	Phosphate Buffer	15

Table 2: Particle size, zeta potential, and polydispersity index[20]

Batch	Mean Particle Size (nm)	Zeta Potential (mV)	PDI
F1	512±137	16.53±17	0.563
F2	450±126	11.36±07	0.341
F3	476±87	11.76±17	0.245
F4	756±291	12.49±10	0.637
F5	391±125	28.44±22	0.294
F6	183±56	35.21±13	0.114
F7	623±162	6.45±08	0.422
F8	259±97	32.14±15	0.248
F9	689±314	5.23±11	0.345

N=3

Weighed amounts of API were added to an amount of solvent or solvent mixture until either saturation was reached or the target concentration for the drug product was achieved by using sonication and data was collected as solubility in gm per liter. Solubility was performed in different solvents.[4]

Preparation of nanocrystals of MND

As the instrument has different settings for feed rate temperature control and drying rate, a lot of data can be obtained with different temperatures, feed rates, and mist types. In the current study, the pharmaceutical product was dissolved using ethanol as a solvent, and the Büchi® Spray Dryer B-90 was used.

The response surface central composite model was used to illustrate technical analysis,[5] and the design expert system was used to generate variables and critical evaluation parameters. With the design name L9 (3^2), the central composite concept offered a nine-run prototype creation formulation using two categories and three levels.[6]

Differential scanning calorimetry (DSC)

Samples were analyzed on DSC for their properties on polymorphism and phase transitions of nanocrystal state.[7]

Drug and excipient compatibility studies

Drug excipient compatibility studies were performed to know if there was any degradation of API during the process of making nano-sized MND powder. Compatibility studies were performed as the "n+1" technique. API was kept with used excipients during prototype development and API assay was performed and compared against a control MND sample. As MND comes in contact with different solvents, the solubility stability will be performed during the HPLC Method validation technique.[8]

Scanning electron microscopy, or SEM

SEM was used to examine the size and form of the collected particles (SEM; JEOL-6510LA). More than 300 particles were counted from the acquired SEM images to determine the average size and particle size distribution.[9]

Diffraction of X-rays in powder

X-rays are electromagnetic radiations with a wavelength of about 1.54°A or the size of an atom. It is used to analyze crystalline materials at the atomic level. X-ray powder scattering (PXRD) measurements were carried out using an X-ray diffractometer (X: Pert PRO with X-Pert Data Collector, PAN analytical). Monochromatic CuK-radiation 1.54°A at 200 mA and 45 kV was used in the studies, which were carried out at room temperature throughout a 2-range of 7 to 80° with a continuous scanning speed of 4°/min. By using a glass slide, the evaluated samples were tightly crammed into the sample holder.[10]

Dissolution studies - capsule based

The dissolving test was conducted using the USP II paddle method for 12 hours at 75 rpm. 900 having 1.2 pH. The dissolving medium's temperature was maintained at 37±0.5 ^{0}C. The sink condition was maintained after a specific time interval.[11]

Animal Study

Four groups of six female Wistar rats [weight tentatively 150–200 g] were selected for the in-vivo study. The

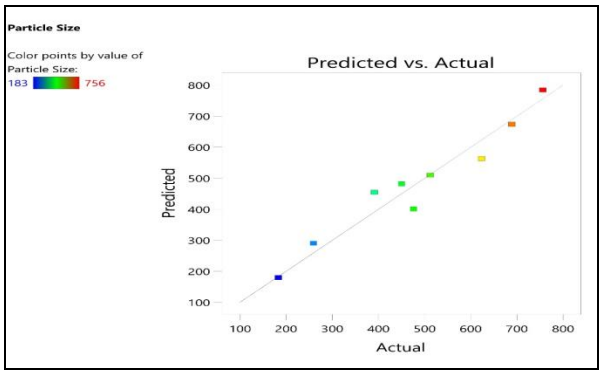

Figure 2: Predicted vs. actual particle size plot.

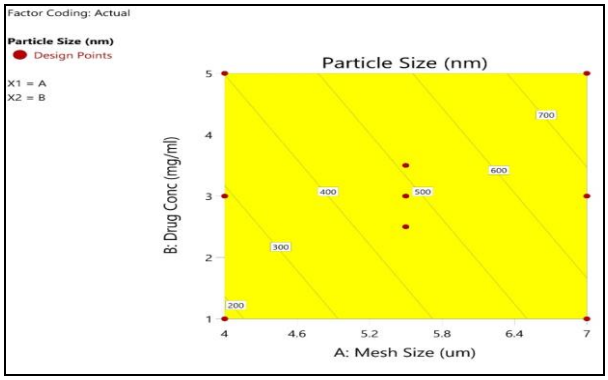

Figure 3: Contour plot effect on particle size.

Table 3: Dissolution data.

Time (min.)	F1	F2	F3	F4	F5	F6	F7	F8	F9
0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
15	8.24	8.32	9.07	5.09	12.78	11.08	9.48	10.82	11.64
30	20.10	20.45	19.12	15.83	21.28	25.87	19.61	22.76	17.46
45	31.19	30.12	31.23	25.35	29.09	38.12	27.68	36.17	33.03
60	39.18	41.08	40.73	33.97	43.78	50.12	37.91	43.65	36.18
90	61.04	63.78	67.11	58.09	59.88	76.34	55.97	63.10	57.12
120	79.01	84.12	81.29	67.06	86.33	91.62	75.88	88.17	72.05

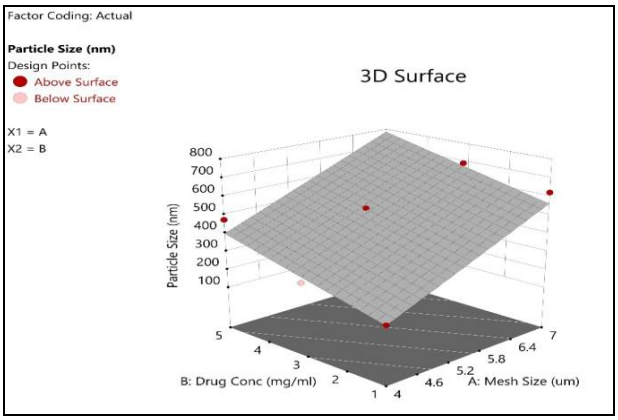

Figure 4: 3D Model graph of mesh size, drug conc. and particle size.

standard conditions i.e. (ad libitum) to the animal during the 12:12 light-to-dark cycle at room temperature.[12]

Drug and animal groups

Animals were divided into four groups (n = 6 for each group) as described below:

Group I: SHAM (normotensive rats)

Group II: DOCA (Deoxycorticosteroneacetate)

Group III: DOCA + MND nanoparticles.

Group VI: DOCA + MND nanoparticles (High Dose 3 mg/kg/Day/Oral)

DOCA (Sigma, 20 mg/kg, St. Louis, MO, USA) was administered subcutaneously twice a week for 14 weeks.

The drinking water for the DOCA and MND groups contained 0.1% KCl and 1.0% NaCl, resulting in hypertension. In a regular saline solution, rats were given both low and high doses of the MND nanoparticle formulation orally. Ad libitum food and saline injections were administered to the rats in the SHAM group. Systolic blood pressure (SBP) measurement: For the first nine weeks, the tail-cuff method was used to assess the systolic blood pressure (SBP) once a week before drug delivery. All groups' systolic blood pressure was checked every day during therapy, and variations in BP (Δ) were recorded.[13,14]

Data analysis

Figure 1, a statistical analysis was performed using the paired t-test to compute and illustrate the change in systolic blood pressure.

RESULTS AND DISCUSSION

Physiochemical characterization of MND

Description

MND powder appeared to be Crystalline and off-white. The powder was light in weight and light protected.

Solubility studies

Solubility studies of MND were performed in water, ethanol, methanol, and acetone[16]. The below table shows the results of solubility studies. (Table 1)

It was observed that MND is insoluble in water (<0.01 mg/ml) and basic buffer (phosphate buffer pH adjusted to 7.4 with 0.1 N NaOH). There was an increase of solubility in an acidic medium (0.1 N HCL) of 0.2 ml/ml (i.e. 200 mg/L). It is highly soluble in ethanol, methanol, Isopropyl Alcohol, Toluene, Acetonitrile, Tetrahydrofuran, and acetone. As the title of the research study describes, the preparation of nanocrystals of MND, after the prototype development, the formulation was analyzed for its solubility in water and buffer.[15,16]

Preparation of nanocrystals

The following nine analyses were done on the spray dryer with defined drug concentration in solvent ethanol. It was ensured that the powder was completely dissolved in a solvent before injecting in the spray dryer. The dried

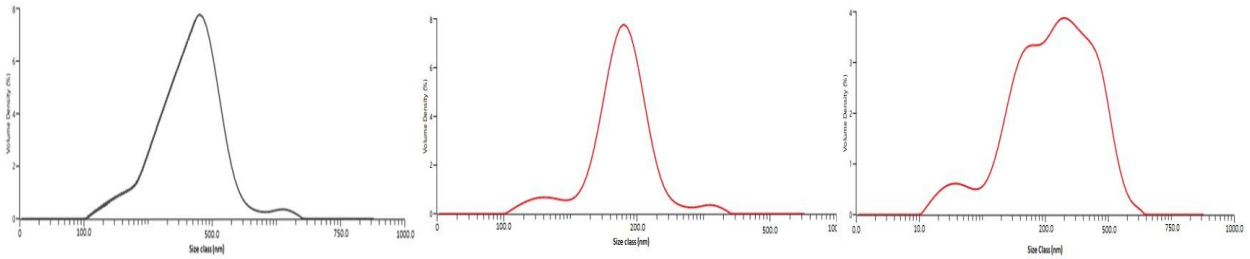

Figure 5: Particle Size distribution of formulation F5 B. Particle Size distribution of formulation F6.

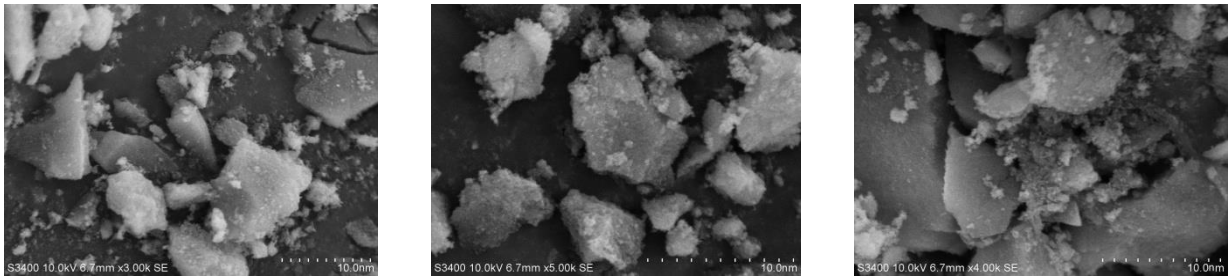

Figure 6: Formulation F5 - SEM image MND spray dried powder B. Formulation F6 - SEM image MND spray dried powder C. Formulation F8 - SEM image MND spray dried powder.

Table 4: The measurement of the systolic blood pressure in different groups of rats.

Groups	Systolic Blood Pressure (mmHg)				
	Day 0	4 Weeks	8 Weeks	12 Weeks	14 Weeks
Group I: SHAM (normotensive rats)	107±3.2	108±3.1	117±3.4	106±2.9	108±3.2
Group II: DOCA*	110±4.6	133±5.6	159±4.9	165±5.6	178±2.1
Group III: MND [0.3 mg/Kg/Day/Oral) (Low dose)	118±3.3	129±2.8	161±4.5	157±5.7	154±2.6
Group IV: MND [3 mg/Kg/Day/Oral] (High dose)	115±5.4	135±4.3	171±4.7	127±3.3	118±4.4

*(DOCA-salt-induced hypertensive rats without treatment) $p< 0.05$, compared with the systolic blood pressure at 8 weeks. (Actual measurements were done once a week in all groups before treatment and after starting the treatment daily measurements were done.

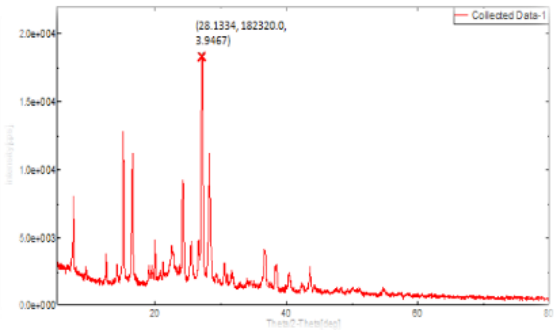

Figure 7: XRD of Pure drug MND

powder was collected through a spatula and analyzed for its particle size using a malvenzetasizer advanced range with technology.[17] From figure no.2 it can be incurred that the actual measured particle size compared to the predicted value from the design expert software are very close. Implying the Quality by Design by Design expert.

Contour plot for effect on particle size

Figure 3. implies that the concentration of the drug and mesh size play important roles in making a nanocrystal of any drug. Lowering the mesh size with low drug concentration gives small-sized nanoparticles.[18] Figure 4. can be concluded from the 3D model graph that the lower the mesh size and drug cons., the lower the particle size.[19]

Physiochemical characterization of spray-dried powder

Analysis of all prototypes of formulation of MND for its particle size, zeta potential, and polydispersity index was done[20] and the results are as per Table No.02. Zeta can be used to forecast a particle's long-term stability. The measurement range is between -60 and +60 mV. In the -10 mV to +10 mV range, zeta potential tends to group and form a cluster. Formulations F5, F6, and F8 have an excellent zeta potential above 25 mV according to our findings. This indicates that there is little probability that they may clump together or agglomerate. Formulations F5, F6, and F8 have mean particle sizes between 180 and 400 nm. The initial particle size of MND was 2 microns before any processing. The particle has significantly shrunk from its original size as a result of the spray dryer process. The PDI value is 1.0 for a highly polydisperse sample with many particle size populations, and 0.0 for a sample with a perfectly uniform particle size distribution. In practice, values of 0.2 or less are usually considered acceptable for compositions containing polymer-based nanoparticles. With a PDI of less than 0.2, Formulation F1 exhibits better and more consistent compatibility.

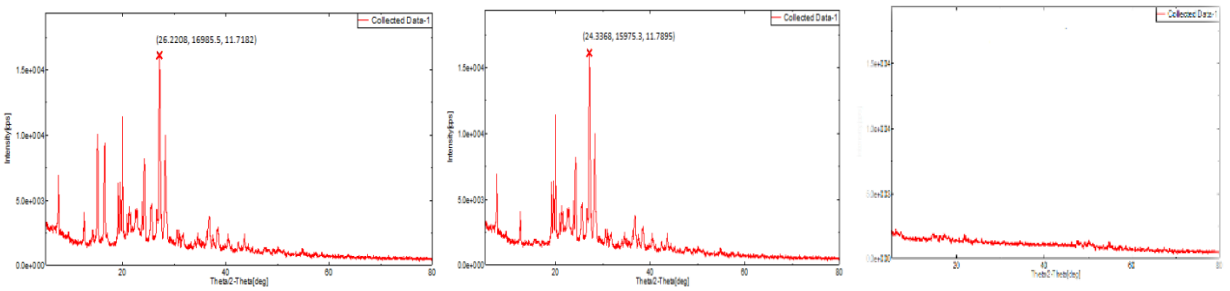

Figure 8: XRD of formulation F1, B. XRD of formulation F2, C. XRD of formulation F6.

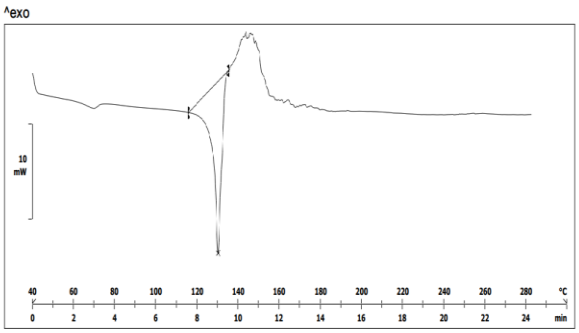

Figure 9: Manidipine HCL DSC graph.

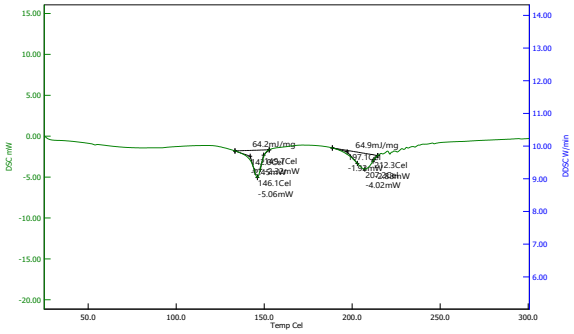

Figure 10: Prototype F6 DSC graph.

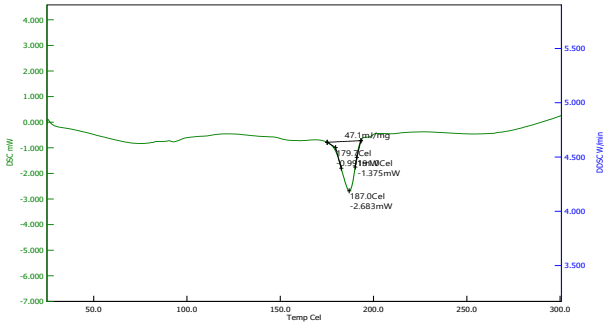

Figure 11: Prototype F7 DSC graph

Particle size distribution

Particle size distribution studies were performed to know the particle size ratio across the sample size.[21] Below are the listed graphs of 3 formulations F5, F6, and F8. (Fig. 5 A-C)

Particle size distribution of formulation F8

The average particle size of each formulation F5 was 391±125, formulation F6 was 183±56, and formulation F8 was 259±97.

Scanning electron microscopy (SEM)

SEM analysis

Using SEM, the shape and size of the collected particles were examined (SEM; JEOL-6510LA). By counting > 300 particles from the acquired SEM images, the average size and particle size distribution were computed SEM analysis was performed for the 3 formulations, and the images are shown below: (Fig. 6A-C). It can be observed that the original MND powder was initially crystalline and after spray-dried processing, the images imply that the nanoparticles are now This was further confirmed by XRD studies.[22]

Powder X-ray diffraction (XRD)

XRD was performed for pure drug material as per the method specified in the material and method section.[23] (Fig. 7) XRD studies were performed to know if the powder is crystalline or amorphous after processing it for particle size reduction by Spray drying method using ethanol as a solvent.[24] (Fig. 8. A-C). Two formulations F1 and F2 have shown crystalline peaks proving the formulation of crystals where F6 did not show any peak, meaning that this batch is amorphous and may enhance solubility.

DSC study

Original Manidipine powder was subjected to DSC to know about the nature of the molecule. The following observation was recorded from the DSC Graph. The sample quantity taken was 7.5 mg for each sample. From Figure 9. It can be observed from the DSC graph that there is no crystalline nature to the powder and shows an endothermic heat change at 130°C causing the melting of the powder. The transition temperature was at around 70°C. The integral energy was found to be at -648.96 mJ.

The following data was recorded for the endothermic peak:

Integral	-648.96 mJ
Onset	115°C
Peak Height	19.18 nW
Peak	131°C
Heating rate	10.00°C per min

The pure drug, Prototypes F6, and F7 were analyzed in Figure 10 and figure11 shows their crystalline nature graph is shown below in:

DSC Studies further summarizes that the spray drying technique caused the crystalline peak to amorphous or semi-crystalline in nature. The Integral energy was in the range of 245.93 mJ to 254.54 mJ for the prototypes.[25, 26]

Dissolution studies

Dissolution studies were performed on the original drug and spray-dried nanoparticle MND drug packed in a size

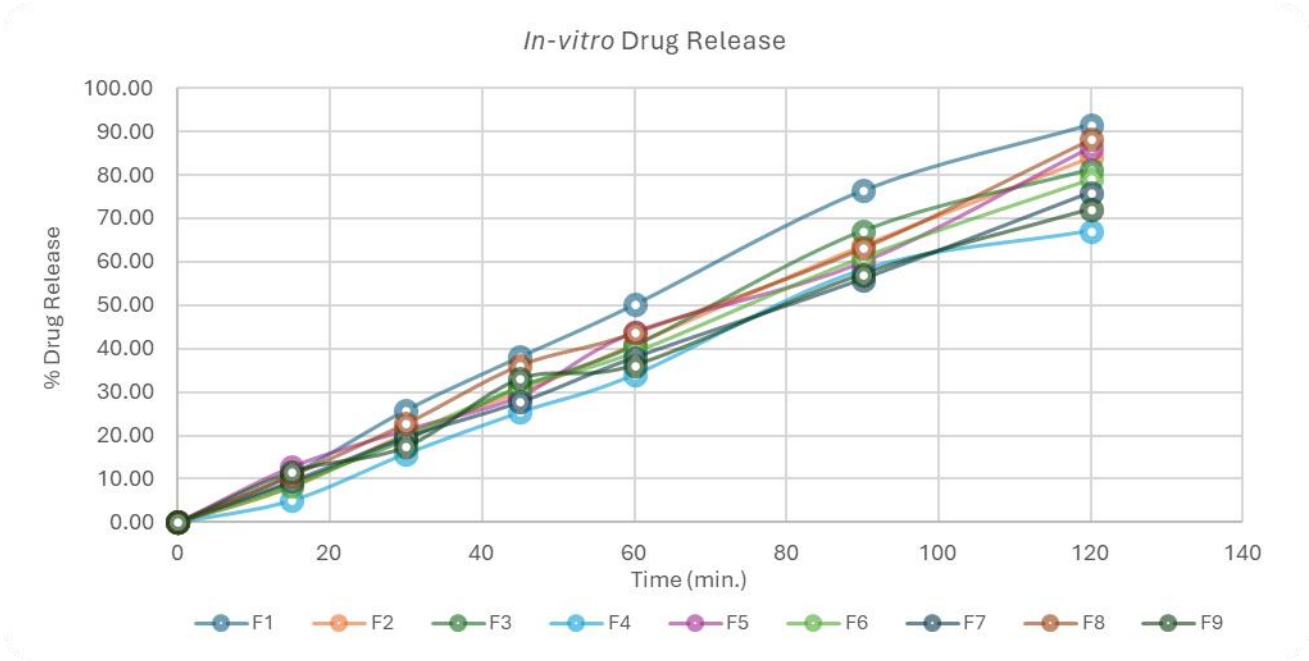

Figure 12: Dissolution Studies of all formulations of MND in capsule.

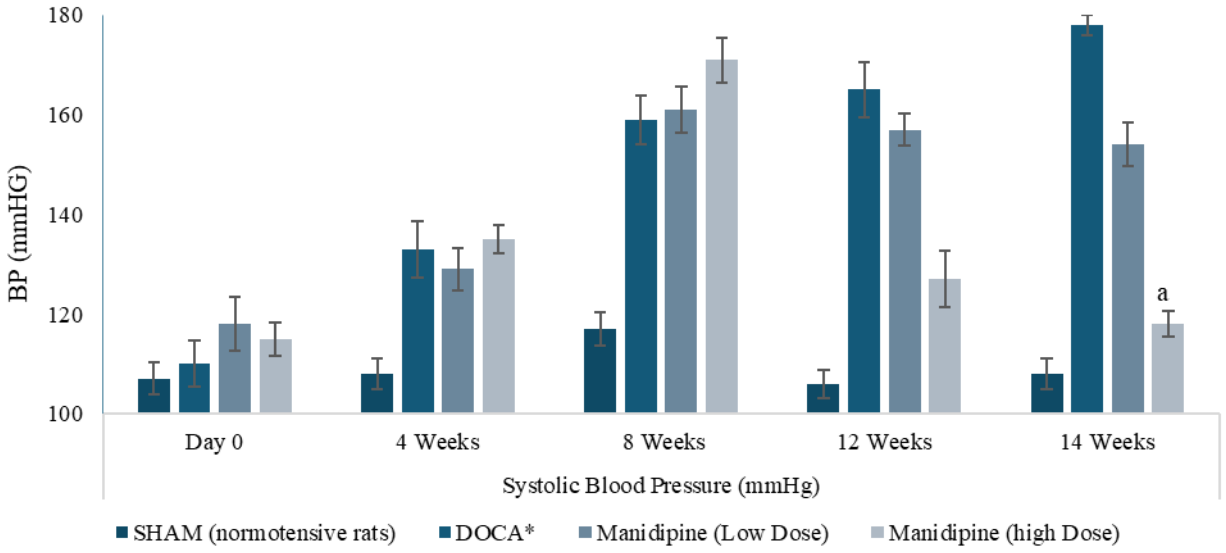

Figure 13: Systolic blood pressure in different groups of rats.

'0' capsule of about 20 mg each. This was analyzed on a UV spectrophotometer.[27] The results of dissolution studies are shown below in drug release. (Table 3 and Fig.12)

As compared to all formulations' medication releases. The maximum release within 120 minutes, or 91.62%, is achieved by prototype F6. Every formulation exhibited a release above 65%, suggesting that the drug's size reduction greatly improved MND's solubility. in contrast to the original medication, which had a drug release of less than 1%.

In-vivo study

The systolic blood pressure of the MND nanoparticle groups (low and high), DOCA (DOCA-salt-induced hypertension rats without therapy), and SHAM (normotensive rats) are among the findings of the *in-vivo* study[28] that are shown in Table No. 4. In a 14-week study protocol, blood pressure was measured systematically. To understand the significance intervals of the change in SBP (Δ) the measurements were done on days 0, week 4, 8, 12, and 14 accordingly. From days 0 to 8 weeks, blood pressure was measured once a week, showing a progressive increase in BP. For careful observation, blood pressure was taken daily from 9 weeks to 14 weeks of the study. When comparing the changes in the ΔSBP from 9 weeks and 14 weeks, it is noticed that there has been a significant decrease in blood pressure i.e., ΔSBP in the MND high dose group as compared to the low dose group. Importantly, at 8 weeks, the group that received MND treatment (high dose) had a blood pressure reading of 171±4.7 and in the same group at 14 weeks BP followed down to 118 ±4.4 indicating significant changes in SBP by the high dose MND nanoparticles.

The nanosizing of MND is responsible[29] for this notable improvement in antihypertensive activity, which was clearly shown (table No. 3 and Fig No. 13). Thus, it was established that MND nanoparticles lower blood pressure the medication amount that reaches the systemic circulation, or bioavailability, can be directly connected with the decrease in pressor effect.[30]

CONCLUSION

According to the current analysis, prototype F6 has a maximum release of 91.62% within 120 minutes. Every formulation exhibited a release above 65%, suggesting that the drug's size reduction greatly improved MND's solubility. in contrast to the original medication, which had a drug release of less than 1%. According to animal tests, the formulation was also successful in lowering blood pressure.

Acknowledgments

The author would like to extend sincere regards and gratitude to the university for permitting them to research conduct and guide for his valuable support.

REFERENCES

1. Dharmashankar K, Widlansky ME. Vascular endothelial function and hypertension: insights and directions. Current hypertension reports. 2010 Dec; 12:448-55.
2. Judd E, Calhoun DA. Apparent and true resistant hypertension: definition, prevalence, and outcomes. Journal of human hypertension. 2014 Aug; 28(8):463-8.
3. Gao Q, Xu L, Cai J. New drug targets for hypertension: a literature review. BiochimicaetBiophysicaActa (BBA)-Molecular Basis of Disease. 2021 Mar 1; 1867(3):166037.
4. Ikemura N, Yamaori S, Kobayashi C, Kamijo S, Murayama N, Yamazaki H, Ohmori S. Inhibitory effects of antihypertensive drugs on human cytochrome P450 2J2 activity: Potent inhibition by azelnidipine and manidipine. Chemico-Biological Interactions. 2019 Jun 1; 306:1-9.
5. Macedo LD, Barbosa EJ, Löbenberg R, Bou-Chacra NA. Anti-inflammatory drug nanocrystals: state of art and regulatory perspective. European Journal of Pharmaceutical Sciences. 2021 Mar 1; 158:105654.
6. 12. Chen, H., Khemtong, C., Yang, X., Chang, X., Gao, J., Nanonization strategies for poorly water-soluble drugs. Drug Discov. Today 2011; 16:354–360.
7. Brough, C., Williams III, R.O., Amorphous solid dispersions and nano-crystal technologies for poorly water-soluble drug delivery. Int. J. Pharm. 2013; 453:157–166 14. Loftsson, T., Brewster, M.E., Pharmaceutical applications of cyclodextrins: basic science and product development. J. Pharm. Pharmacol. 2010; 62:1607–1621.
8. Manjusha Bhange et.al. Emerging therapies and innovations in vitiligo management: a comprehensive review, Journal of Immunoassay and Immunochemistry, 2024: DOI: 10.1080/15321819.2024.2412528.
9. Brouwers, J., Brewster, M.E., Augustijns, P., Supersaturating drug delivery systems: the answer to solubility-limited oral bioavailability? J. Pharm. Sci. 2009; 98:2549–2572.
10. Liu, P., Rong, X., Laru, J., van Veen, B., Kiesvaara, J., Hirvonen, J., Laaksonen, T., Peltonen, L., Nanosuspensions of poorly soluble drugs: preparation and development by wet milling. Int. J. Pharm. 2011; 411:215–222.
11. Borchard, G., 2015. Drug nanocrystals. In: In: Crommelin, D., de Vlieger, J. (Eds.), Non-Biological Complex Drugs. AAPS Advances in the Pharmaceutical Sciences Series, vol. 20. Springer, Cham, pp. 171–189.
12. Kesisoglou, F., Panmai, S., Wu, Y., Nanosizing – oral formulation development and biopharmaceutical evaluation. Adv. Drug Deliv. Rev. 2007; 59:631–644.
13. Gao, L., Liu, G.Y., Ma, J.L., Wang, X.Q., Zhou, L., Li, X., Drug nanocrystals: in vivo performances. J. Control. Release 2012; 160:418–430. 20 Lu, Y., Li, Y., Wu, W., Injected nanocrystals for targeted drug delivery. Acta Pharm. Sin. B 2016; 6:106–113.
14. R.R. Sevda, A.S. Ravetkar, P.J. Shirote, UV spectrophotometric estimation of rosuvastatin calcium and fenofibrate in bulk drug and dosage form using simultaneous equation method, Int. J. Chem. Tech. Res. 2011; 3:629–635.
15. Fu, Q., Sun, J., Zhang, D., Li, M., Wang, Y., Ling, G., Liu, X., Sun, Y., Sui, X., Luo, C., Sun, L., Han, X., Lian, H., Zhu, M., Wang, S., He, Z., Nimodipine nanocrystals for oral bioavailability improvement: preparation, characterization and pharmacokinetic studies. Colloids Surf. B Biointerfaces 2013; 109:161–166.
16. Oussoren C1, Storm G. Liposomes to target the lymphatics by subcutaneous administration. Adv Drug Deliv Rev. 2001; 50(1 2):143-56
17. Dufek M, Hayles M. The Quanta FEG 200, 400, 600 User's Operation Manual. 4022 290 22211 1st Edition. 2003.
18. Cheer SM, McClellan K. Manidipine: a review of its use in hypertension. Drugs. 2001 Oct; 61:1777-99.
19. SaizSatjes M, Martinez-Martin FJ. Manidipine: an antihypertensive drug with positive effects on metabolic parameters and adrenergic tone in patients with diabetes. Drugs in Context. 2018;7.
20. Detroja C, Chavhan S, Sawant K. Enhanced antihypertensive activity of candesartan cilexetil nanosuspension: formulation, characterization and pharmacodynamic study. Scientiapharmaceutica. 2011 Sep;79(3):635-52.
21. Manjusha Bhange et. al. Niosomes a promissing nanocarrier: a review, International Journal of Applied Pharmaceutics, 2023: DOI: 10.22159/ijap.2023v15i6.47969.
22. Porteri E, Rizzoni D, Piccoli A, Castellano M, Bettoni G, Muiesan Ml, Pasini G, Guelfi D, Zulli R, Rosei E. Effects of hypotensive and non-hypotensive doses of

manidipine on structure, responses to endothelin-1 and ICAM-1 production in mesenteric small resistance arteries of spontaneously hypertensive rats. Blood pressure. 1998 Jan 1; 7(5-6):324-30.
23. Chen, H., Khemtong, C., Yang, X., Chang, X., Gao, J., Nanonization strategies for poorly water-soluble drugs. Drug Discov. Today 2011; 16:354–360B.
24. rough, C., Williams III, R.O., Amorphous solid dispersions and nano-crystal technologies for poorly water-soluble drug delivery. Int. J. Pharm. 2013; 453:157–166.
25. Manjusha A. Bhange et. al., Design and Development of Phytosomal Soft Nanoparticles for Liver Targeting. 2023, Doi: Https://Dx.Doi.Org/10.22159/Ijap.2023v15i1.
26. Brouwers, J., Brewster, M.E., Augustijns, P., Supersaturating drug delivery systems: the answer to solubility-limited oral bioavailability? J. Pharm. Sci. 2009; 98:2549–2572.
27. Liu, P., Rong, X., Laru, J., van Veen, B., Kiesvaara, J., Hirvonen, J., Laaksonen, T., Peltonen, L., Nanosuspensions of poorly soluble drugs: preparation and development by wet milling. Int. J. Pharm. 2011; 411:215–222.
28. Manjusha a. Bhange et.al., Formulation and development of Gallen gum loaded self-assembled mixed micelles system based on flavonoid phospholipid complex, International journal of applied pharmaceutics, 2023: Doi: 10.22159/ijap.2023v15i3.46795.
29. Chen Y, Liu J, Zhao X, Xu H (2005) Oleanolic acid nanosuspensions: preparation, in-vitro characterization and enhanced hepatoprotective effect. J Pharm Pharmacol 57(2):259–264.
30. Chen JM, Wang ZZ, Chaun BJ, Li S, Lu T-B (2012) Crystal engineering approach to improve the solubility of mebendazole. CrystEngComm 14:6221–6229.