Journal of Population Therapeutics & Clinical Pharmacology

RESEARCH ARTICLE
DOI: 10.47750/jptcp.2023.30.13.028

Enhancement of silymarin solubility and dissolution by nicotinamide-based solid dispersion employing the kneading method

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Submitted: 24 March 2023; Accepted: 12 April 2023; Published: 13 May 2023

ABSTRACT

Silymarin (SM) is a herbal medicine extracted from the plant Silybum marianum. SM has many potent pharmacological activities among which are antioxidant, anti-inflammatory, and possible wound-healing properties. It belongs to BCS class II thus the drug is practically water insoluble with poor dissolution. In this paper the main aim is to modify solubility and dissolution of SM , one SD preparation method is used which is the Kneading method (Kn). The carrier that has being used is nicotinamide(NA) , Nicotinamide shows promise for the treatment of a wide range of dermatological conditions including autoimmune blistering disorders, acne, rosacea, ageing skin and atopic dermatitis. In particular, recent studies have also shown it to be a potential agent for wound healing, reducing actinic keratoses and preventing skin cancers. Solid dispersion (SD) was characterized for percentage yield, drug content, saturation solubility, and in-vitro dissolution study in comparing to pure drug. Formulation evaluated by Fourier-transform infrared spectroscopy, differential scanning colorimetry, X-ray diffraction.

Keywords: *pharmaceutical; silymarin solubility; nicotinamide-based*

INTRODUCTION

Solid dispersions are a valuable pharmaceutical approach for improving drug solubility, absorption, and therapeutic effectiveness in dosage forms. It was first suggested by Sekiguchi and Obi in the early 1960s (1).

A type of solid product termed a solid dispersion has at least two components, often a hydrophilic matrix and a hydrophobic drug (2).

Milk thistle seeds contain the highest concentration of silymarin than any part of the plant (Silybum marianum), making it an ideal source for silymarin extraction (3).

Silymarin belongs to class II of the biopharmaceutical classification system (BCS) meaning it is practically insoluble in water which represent a challenge when formulating a dosage form for this drug so the method chosen to enhance the solubility and dissolution is solid dispersion (4).

The carrier chosen to proceed with is nicotinamide (NA) , an amide of vitamin B3 (niacin) hydrophilic endogenous substance which has been demonstrated to have anti-inflammatory, antioxidative, and regulatory effects on the production of immunomodulatory proteins (5).

This main study objective is an enhancement of solubility, the dissolution rate of poorly water-soluble silymarin by using a hydrophilic carrier. The SDs are prepared by Kneading technique by using three drug: carrier ratios (1:1 , 1:2 , 1:3).

MATERIALS AND METHODS

Materials

Silymarin , Hyperchem supplied the drug (purity: 98%) (China), nicotinamide (Thomas baker chemicals (India)).

All of the remaining materials and solvents that were used in this paper of analytical grade.

SM solid dispersion preparation via kneading (Kn)

Three distinct formulations (SM-SD1, SM-SD2, and SM-SD3) were made using Kn method. In a glass mortar, SM and NA were blended in weight proportions of 1:1, 1:2, and 1:3, then moistened with water drops. This process took 30 minutes. In an oven at 50°, the paste was dried for 24 hr. When completely dry, the powder was sieved through a No. 60 mesh and placed in a desiccator for further analysis (6).

TABLE 1: SD Formulas of SM Composition

Formula no.	Drug: carrier ratio
SM-SD1	1:1
SM-SD2	1:2
SM-SD3	1:3

Evaluation of SD:

Percent yield

The yield was determined to determine if the preparation method is efficient, the following equation is used (7).

$$\textbf{\% yield} = \frac{\textit{Practical yield}}{\textit{Theoretical wt}(\textit{drug}+\textit{carrier})} \times 100$$

Drug Content:

A quantity of the produced SD equal to 10 milligrams of SM was accurately weighed, and then stirred in 20 ml of methanol. The solution was then filtered and diluted appropriately with the same solvent. A UV spectrophotometer with a wavelength of 288 was used to determine the medication concentration, and the experiment was repeated three times (8).

$$\textit{drug content} = \frac{\textbf{Practically obtained mass}}{\textbf{Theoretical mass}} \times 100$$

Saturation solubility studies

The shake–flask technique was used to conduct saturation solubility investigations using distilled water as the solvent. At 25 °C, an excess quantity of SM pure powder, SD formulae, was added to 10 ml of water with nonstop shaking for a full two days. Afterward, the samples were screened through a filter. By the use of Millipore filter paper (0.45 m). Distilled water was utilized to dilute the filtrate and examined at 287 nm to quantify the amount of SM that had dissolved (9).

In vitro study of drug release

Utilizing USP type II (paddle type) dissolving test device, the drug release of two formulations of SM- SD was investigated in vitro. As a dissolving medium, Preheated to 37 C°, 900 ml of phosphate buffered saline (pH 7.4) and 50 mg of drug samples were thrown over its surface. At regular intervals (10,20,30,40, up to 60min), 5ml aliquots were taken and replaced with new dissolving media. At 287 nm, the materials were examined spectrophotometrically. This test was performed three times.

Using the following equation, a similarity factor was computed and used for statistical analysis of the dissolution profiles.

$$f2 = 50 \cdot \log\left\{100 \cdot \left[1 + \frac{1}{n}\sum_{t=1}^{n}(Rt - Tt)^2\right]^{-0.5}\right\}$$

(Rt) is the reference dissolution value at time t , (Tt) is the test dissolution value, and (n) is the number of dissolution time points. If *f*2 is greater than or equal to 50 the dissolution profile is deemed similar. If *f*2 is less than 50, the dissolution profiles are not considered similar (10).

Selection of the best formula

The selection of best formula is based on the results of solubility and dissolution profile .

Evaluation of the selected formula

FT-IR Spectroscopy

FTIR was used to collect the (FTIR) spectra (Shimadzu 8300 Japan). Bromide of potassium was used to compress samples of untreated SM, NA, chosen formula, and PM. Within a scanning range of 4000-0 cm-1, spectra were recorded (11).

Differential Scanning Calorimetry (DSC):

5mg samples of SM, NA, the chosen formula, and its PM were heated at scanning range of 10 degrees Celsius per minute in an aluminum pan in a nitrogen environment in the 50-300 C

temperature range using DSC-60 plus (Shimadzu, Japan). A comparable empty pan was used as a comparison(11).

X- ray diffraction studies on powders (PXRD)

X- ray diffraction studies were used to evaluate the crystalline state of pure SM, NA, a particular SD formula and the PM of the same SD formula. using a PXRD diffractometer (XRD-6000, Shimadzu, Japan) The PXRD was carried out under the subsequent circumstances: target metal Cu, filter K, voltage 40kV, and current 30mA. The samples were scanned at 1.5406 wavelength across a range of 2 including 10-60° (11).

Statistical analysis

The results of the experiments were analyzed using (ANOVA) and the outcome was reported as the mean of triplicate samples standard deviation (std). Using SPSS version 29, we determined that a significance level of ($p \leq 0.05$) was statistically significant.

RESULTS AND DISCUSSION

Evaluation of SD

The percentage yield of prepared SD

The prepared formulas showed good percentage yield as shown in table 2.

Drug content

The drug content was found to be within the acceptable range as will be shown in table 2.

TABLE 2: percentage yield and drug content of prepared SDs

Formula no.	Percentage yield	Drug content
SM-SD1	95	95.57
SM-SD2	99	98
SM-SD3	100%	101

Saturation solubility of pure SM and prepared SM-SDs

The solubility of SM is 0.05 mg/ml in water, this solubility was enhanced by the SDs formulations as shown in the table 3 below

TABLE 3: saturated solubility of prepared SDs in mg/ml

Formula	Saturated solubility mg/ml
SM-SD1	0.235±0.03
SM-SD2	0.8±0.02
SM-SD3	0.806±0.001

When comparing the solubility of pure drug with the SDs formulations we find a significant ($p \leq 0.05$) enhancement due to the hydrophilic nature of the carrier . the increment in solubility is proportional to increasing the ratio of carrier to drug , so we notice as the ratio of carrier increase so does the solubility ; however the increment with 1:2 and 1:3 was not statistically different . The method of preparation (Kn) was successful in enhancing the solubility by 16-fold (12).

***In vitro* dissolution study**

The release of the pure SM was compared with the release of the SM-SD2 and SM-SD3 formulas that showed the most increase in solubility in order to choose the best formula since both of them increased the solubility by almost equal folds. The dissolution of the drug from the SDs formulations (SM-SD2, SM-SD3) was faster, both of them gave 100 % release within 20 min, in comparison to the dissolution of the pure drug in a given time course, with *f*2 values of (9.5, 9.2) respectively. This can be attributed to solubility enhancement, which agrees with the Noyes-Whitney equation(13).

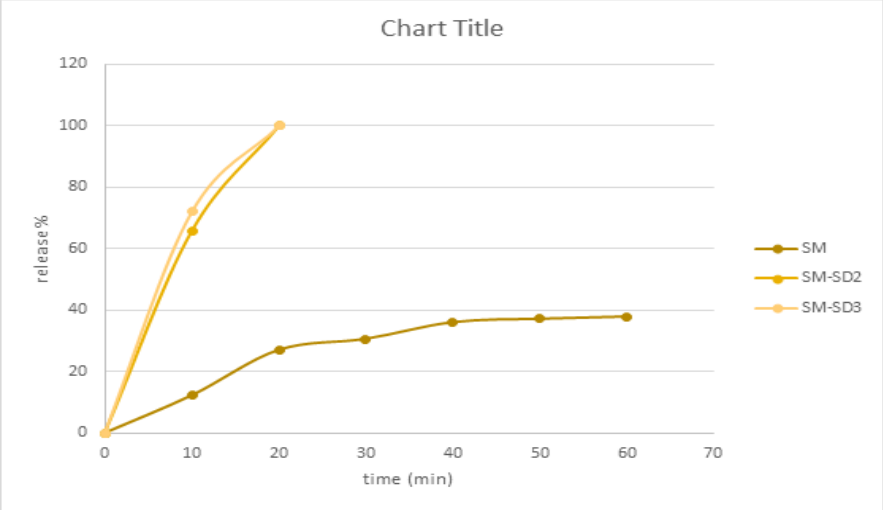


FIGURE 1: *In vitro* dissolution rate of SM , SM-SD2 and SM-SD3 at 37 °and a phosphate buffer saline with a pH of 7.4 .

Best formula selection

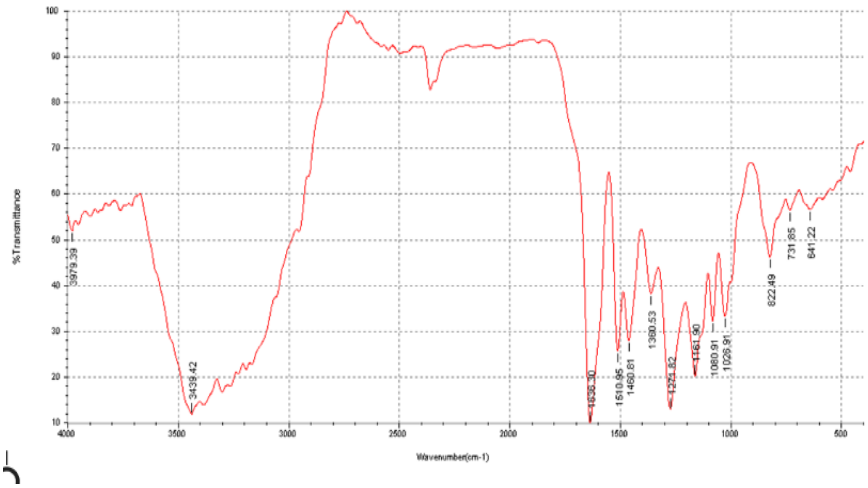
Since the solubility and dissolution profile are very approximate for the SM-SD2 and SM-SD3 , SM-SD3 is the formula that will proceed to be evaluated due to its slight better solubility and dissolution .

Fourier transform infrared spectroscopy (FTIR)

It is well known that vibrational changes can serve as a probe of intermolecular interactions in solid materials. Characteristic peaks of silymarin appeared at 3440.39 cm^{-1} (-OH stretching vibration), 2947 cm^{-1} (O-H stretching), 1637.27 cm^{-1} (C=O stretching), 1511–1461 cm^{-1} (skeleton vibration of aromatic C=C ring stretching), 1361 cm^{-1} (-OH in-plane bending), 1272 cm^{-1} (C-O-C stretching), 1081–1161 cm^{-1} (in-plane = C-H bending) and 642.18–823 cm^{-1} (Figure 4). These results agreed with previously reported studies(14).

FTIR spectra of NA (Figure) showed the appearance of the characteristic amino group –NH2 symmetric stretching vibrations at 3364 cm^{-1}. The C=O stretching also appeared at 1618 cm^{-1} and the –CN stretching vibrations also appeared at 1422 cm^{-1} which correspond with the recorded values in the literature (15).

The SM skeleton in the best formula was not altered by the addition of the carrier indicating a lack of interaction. This states that the drug was molecularly dispersed in the carrier without any interactions.



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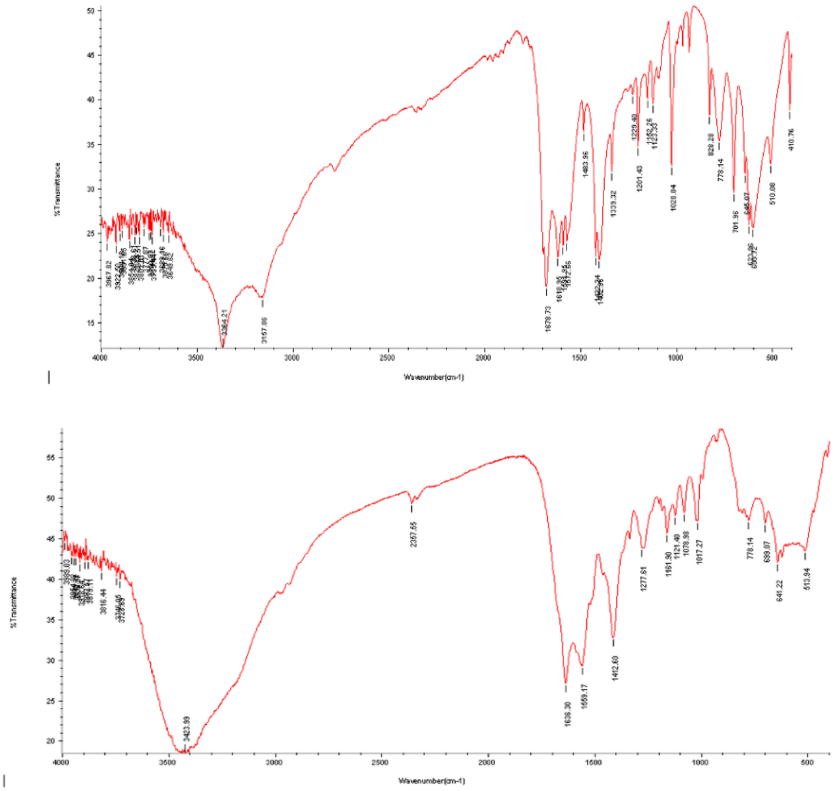
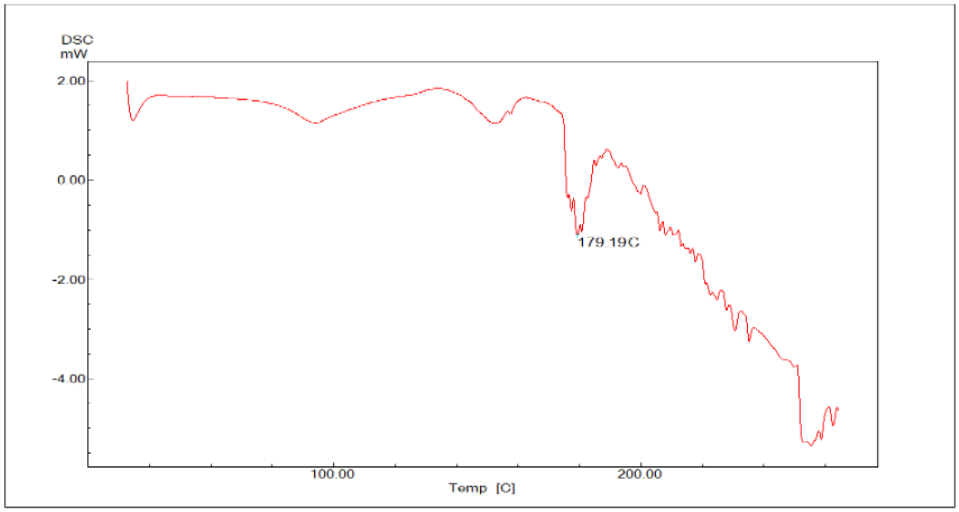


FIGURE 2: FTIR of SM, NA, SM-SD3

Differential scanning calorimetry

The DSC thermograms are shown in Figure 3. For SM powder, a sharp endothermic peak was observed at 179.19 C, corresponding to its melting point (16). The melting endotherm for NA was observed at 130.80 C as in previous studies. The DSC analysis showed a slight decrease in NA m.p. but it's still in crystalline state. However, the endothermic peak of SM was not observed in SDs. These results suggested that SM was present in a different state in the SM-SD, meaning that SM have turned into an amorphous state explaining the increment in solubility and dissolution behavior (17).



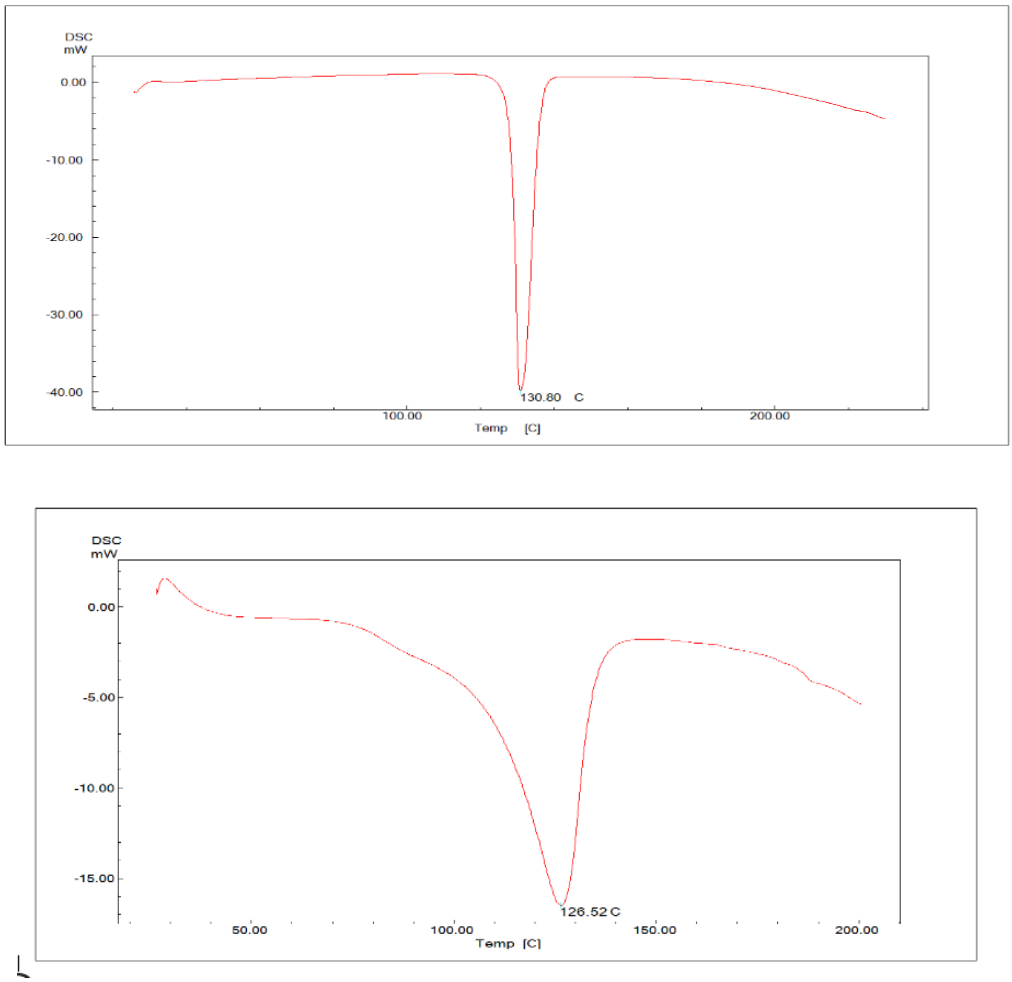
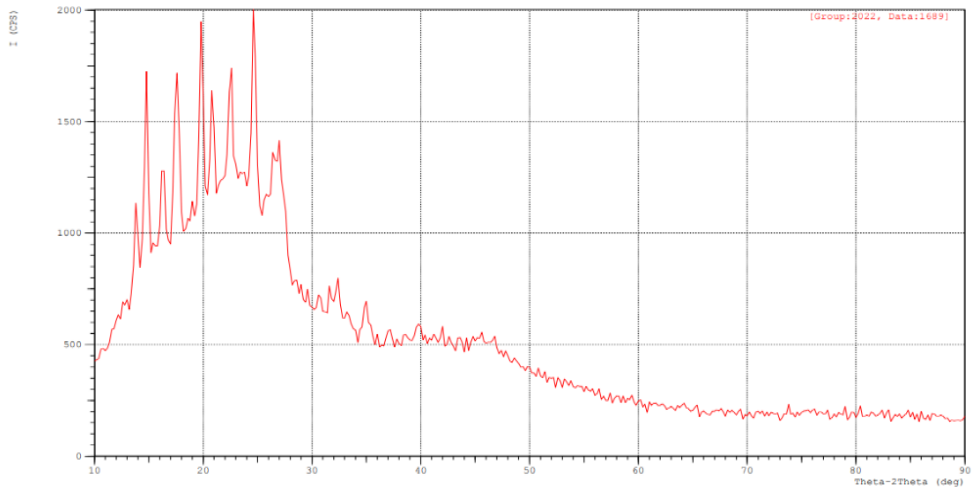


FIGURE 3: DSC of SM , NA , SM-SD3

Powder X-Ray Diffraction (PXRD)

pattern of silymarin, NA and solid dispersions prepared by Kn are shown in Fig2. The locations of the most prominent peaks at **2Θ** of 14 °, 17 °, 19 °, and 24° (Figure 3-A), which confirm the crystalline structure of the drug. The pattern of NA showed characteristic diffraction peaks at **2Θ** Θ of 15∘, 24.81∘, 26.43∘. indicating that the carrier was also in crystalline state. the pattern of best formula showed disappearance of the characteristic peaks meaning the conversion of the drug to amorphous form which helps in explaining the solubility enhancement(118).



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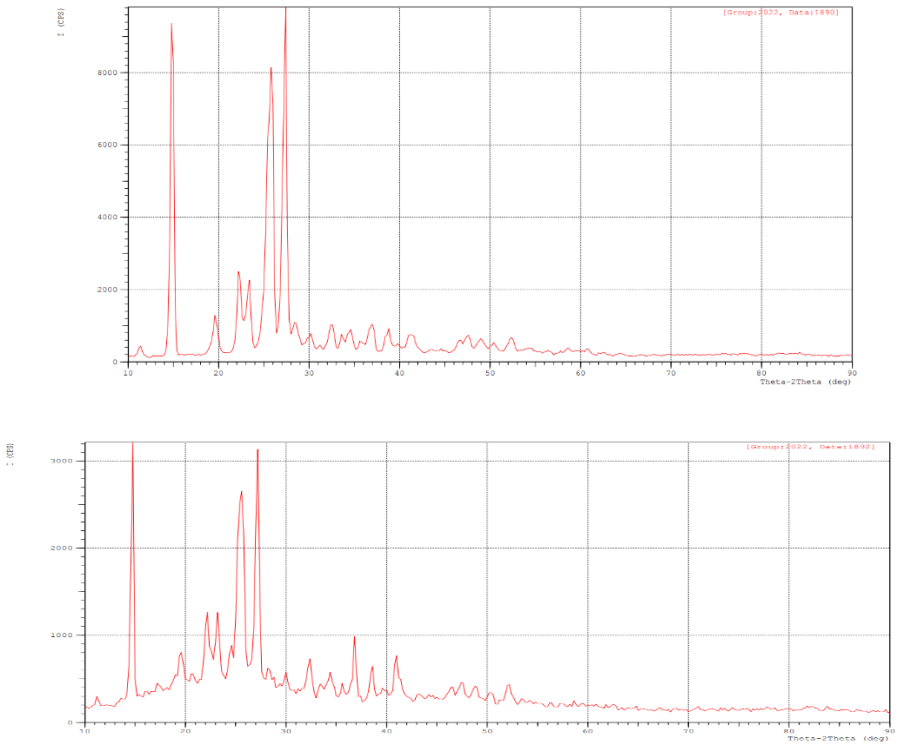

FIGURE 4: PXRD of SM, NA, SM-SD3

CONCLUSION

The solubility and dissolution of SM-SD showed great improvement compared to unprocessed SM. The SM-SD3 which was prepared by Kn method in 1:3 drug: carrier ratio showed 16-fold increase in solubility and great enhancement in dissolution rate.

REFERENCES

1. Sinha S, Ali M, Baboota S, Ahuja A, Kumar A, Ali J. Solid dispersion as an approach for bioavailability enhancement of poorly water-soluble drug ritonavir. *AAPS PharmSciTech*. 2010;11(2):518-527. doi:10.1208/s12249-010-9404-1.
2. Tekade AR, Yadav JN. A Review on Solid Dispersion and Carriers Used Therein for Solubility Enhancement of Poorly Water Soluble Drugs. *Adv Pharm Bull*. 2020;10(3):359-369. doi:10.34172/apb.2020.044.
3. Karimi G, Vahabzadeh M, Lari P, Rashedinia M, Moshiri M. "Silymarin", a promising pharmacological agent for treatment of diseases. *Iran J Basic Med Sci*. 2011;14(4):308-317.
4. Yousaf AM, Malik UR, Shahzad Y, Mahmood T, Hussain T. Silymarin-laden PVP-PEG polymeric composite for enhanced aqueous solubility and dissolution rate: Preparation and in vitro characterization. *J Pharm Anal*. 2019;9(1):34-39. doi:10.1016/j.jpha.2018.09.003.
5. Verma, M Mohan et al. "Dissolution, bioavailability and ulcerogenic studies on piroxicam-nicotinamide solid dispersion formulations." *Bollettino chimico farmaceutico* vol. 142,3 (2003): 119-24.
6. Ghareeb MM, Abdulrasool AA, Hussein AA, Noordin MI. Kneading technique for preparation of binary solid dispersion of meloxicam with poloxamer 188. *AAPS PharmSciTech*. 2009;10(4):1206-1215. doi:10.1208/s12249-009-9316-0.
7. Hassani, Hiba & Al-khedairy, Eman. (2021). Formulation and In-Vitro Evaluation of Meloxicam Solid Dispersion using Natural Polymers.. Iraqi Journal of Pharmaceutical Sciences (P-ISSN: 1683 - 3597 , E-ISSN : 2521 - 3512). 30. 169-178. 10.31351/vol30iss1pp169-178.
8. Sharma A, Jain CP. Preparation and characterization of solid dispersions of carvedilol with PVP K30. *Res Pharm Sci*. 2010;5(1):49-56.
9. Arora SC, Sharma PK, Irchhaiya R, Khatkar A, Singh N, Gagoria J. Development, characterization and solubility study of solid dispersions of Cefuroxime Axetil by the solvent evaporation method. *J Adv Pharm Technol Res*. 2010;1(3):326-329. doi:10.4103/0110-5558.72427
10. Dalwadi, Dr. Sonali & Soni, Tejal & Thakkar, Vaishali & Tejal, Gandhi. (2010). Silymarin-solid dispersions: Characterization and influence of preparation methods on dissolution. Acta

pharmaceutica (Zagreb, Croatia). 60. 427-43. 10.2478/v10007-010-0038-3.
11. Song, I.-S.; Nam, S.-J.; Jeon, J.-H.; Park, S.-J.; Choi, M.-K. Enhanced Bioavailability and Efficacy of Silymarin Solid Dispersion in Rats with Acetaminophen-Induced Hepatotoxicity. *Pharmaceutics* **2021**, *13*, 628. https://doi.org/10.3390/pharmaceutics13050628
12. Dispersions S. Effect of Hydrophilic Carriers for Solubility and Dissolution Enhancement of Effect of Hydrophilic Carriers for Solubility and Dissolution Enhancement of Sulfamerazine by Solid Dispersions Technique. 2021;(December).
13. Michael E. Aulton KMGT. Aulton-Pharmaceutics-The-Design-and-Manufacture-of-Medicines-5th-Edition. Vol. 5, Aulton's Pharmaceutics The Design and Manufacture of Medicines. 2018. 933 p.
14. Saiyyad, Irfan & Bhambere, Deepak & Kshirsagar, Sanjay. (2017). Formulation and Optimization of Silymarin Loaded PLGA Nanoparticle for liver targeting. 10.13140/RG.2.2.35383.16809.
15. Yadav, Dattatraya & Savjani, Jignasa & Savjani, Ketan & Kumar, Aakash & Patel, Snehal. (2022). Pharmaceutical Co-crystal of Antiviral Agent Efavirenz with Nicotinamide for the Enhancement of Solubility, Physicochemical Stability, and Oral Bioavailability. AAPS PharmSciTech. 24. 10.1208/s12249-022-02467-7.
16. Gandey S, Aparna V, Kandarapu R. Preparation and Biological Evaluation of Silybin Liposomes for the Treatment of Liver Disorders. J Pharm Res Int. 2021;(July):186–200.
17. Oryan A, Tabatabaei Naeini A, Moshiri A, Mohammadalipour A, Tabandeh MR. Modulation of cutaneous wound healing by silymarin in rats. J Wound Care. 2012;21(9):457–64.
18. Sharifi R, Pasalar P, Kamalinejad M, Dehpour AR, Tavangar SM, Paknejad M, et al. The effect of silymarin (Silybum marianum) on human skin fibroblasts in an in vitro wound healing model. Pharm Biol. 2013;51(3):298–303.