

Formulation and Evaluation of Chitosan Based Sofosbuvir Solid Lipid Nanoparticles

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Abstract

Sofosbuvir is used to suppress hepatitis C virus (HCV). Sofosbuvir, a Biopharmaceutical Classification System (BCS) class III drug, is highly soluble and impermeable. This study focuses on preparation of sofosbuvir polymeric nanoparticles with improved permeability. Sofosbuvir penetrates better into solid lipid nanoparticles (SLNPs). The chitosan polymer loaded the drug into a single particle microemulsion system and analyzed by zeta measurement, transmission electron microscopy (TEM), and entrapment efficiency (EE). *In vitro* drug delivery was tested on optimized SLNPs. The non-Fickian transport release profile of formulated SLNPs was 60 - 73 nm. The RP-HPLC approach of sofosbuvir used a Phenomenex Luna C18 column (4.6 x 150 mm, 5) and a mobile phase of acetonitrile (ACN):phosphate buffer (60:40 v/v) at 1.0 ml/min, 260 nm detected. Sofosbuvir's verified analytical method is linear, precise, accurate, and reliable. All parameters had relative standard deviation (RSD) values less than 5 and stability indicated International Conference on Harmonization (ICH) Q1A R (2) study. Improved oral bioavailability and drug permeability.

Keywords

Bio-polymer, Chitosan, RP-HPLC, Solid lipid nanoparticles, Sofosbuvir

Introduction

Sofosbuvir, is chemically isopropyl(2S)-2-[[[(2R,3R,4R,5R)-5-(2,4dioxypyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-yl]methoxyphenoxyphosphoryl]amino propanoate as in figure 1 and has a molecular weight of 529.453 g/mol. Sofosbuvir is a non-structural nucleotide polymerase inhibitor of the HCV NS5B protein [1, 2]. Sofosbuvir is a BCS class-III drug, having high solubility and low permeability.

Worldwide, 170 million people who are infected with the HCV. Both hepatocellular carcinoma and liver diseases at their end-stage are associated with the HCV [3]. Standard HCV drugs are ribavirin and PEGylated interferon, both of which have low sustained virology response and significant side effects [4]. Several drugs like telaprevir and boceprevir, which are NS3/4a protease inhibitors, were proved for their efficacy in the treatment of HCV genotype 1 infections during the period weeks with PEGylated interferon/ribavirin. Despite major changes in HCV treatment increasing the rate of sustained virology response, some adverse events, such as severe rash/pruritus, anemia and dyspepsia, have led to high rates of early treatment discontinuation. In addition to their limited antiviral activity

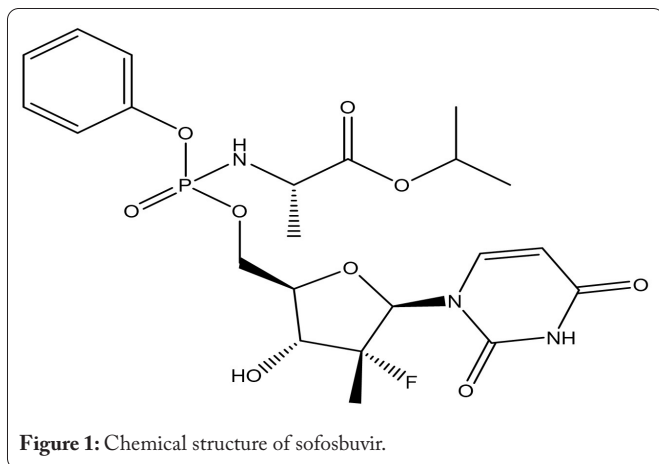


Figure 1: Chemical structure of sofosbuvir.

against HCV genotype 1, they had weak drug interactions, a high number of pills and complex treatment algorithms [5]. With the Food and Drug Administration approval of more effective DAAs such as sofosbuvir and daclatasvir, and their exceptional tolerance, the field of HCV treatment has entered a new era [6]. Sofosbuvir used alone and in combination with other antiviral agents such as ribavirin, simeprevir, ledipasvir, daclatasvir, and velpatasvir for HCV treatment. The oral route of administration is the preferred route of administration. Sofosbuvir disrupts viral RNA synthesis by inhibiting the HCV protein 5B. Due to its rapid absorption with reaching C_{max} in 30 - 60 min, the drug is rightly added to the World Health Organization list of essential medicines [7].

Nanoparticles are another important type of nanocarriers whose formulation mainly relies on the use of compostable and biocompatible polymers. Due to the biodegradation of the polymer and the diffusion of the drug from the polymer medium, a controlled release of the drug is achieved. Through natural metabolic pathways, all polymers used in the formulation of these polymeric nanoparticles are eliminated from the body [8]. Both natural and synthetic polymers are used. The biocompatible and biodegradable nature is mainly attributed to natural polymers. However, its use is limited due to variation in properties from batch to batch and may be somewhat immunogenic. There are two main types of polymeric nanoparticles namely polymeric nanocapsules and nanospheres. The first consists of the drug encapsulated in the aqueous or non-aqueous core. While in the latter case, the drug is adsorbed on a polymer surface, or it can be placed in any space of the particle [9].

The high-performance liquid chromatography (HPLC) approach has been used in a number of studies to quantify sofosbuvir, both alone and in combination with other antiviral tablets. SLNPs were a type of commonly used drug delivery devices that are made using non-toxic, biocompatible, and biodegradable polymers. The current study focuses on formulation of sofosbuvir SLNPs and the development of the reverse phase (RP)-HPLC technique for the evaluation of sofosbuvir in whole and prepared SLNPs, as well as the formulation of sofosbuvir SLNPs with increased permeability. The developed method has been validated by the ICH Q1AR (2) guidelines. The permeability of sofosbuvir SLNPs was determined using the reverse chicken gut technique [10].

Materials and Method

Materials

Sofosbuvir, chitosan, stearic acid and ACN were acquired from Aurobindo Laboratories Private Limited, Hyderabad. Chitosan, stearic acid purchased were from Hi-Media, Mumbai. ACN (HPLC grade) was purchased from AR Rankem Ltd, New Delhi, India. Ultrapure water is made using the Milli-Q® refining system. The rest of the chemicals and solvents remained of analytical quality.

Methodology

Chromatographic conditions

The isocratic elution mode was used in the experiment. A 60:40 (v/v) ACN:phosphate buffer mixture was used as the bipartite mobile phase. A vacuum filter was used to filter out acetic acid, which was used to adjust the pH to 3. Each cycle involved approximately 20 μ l of sample injection and ultraviolet (UV) detectors with a wavelength of 260 nm detected the eluate.

Solubility studies by a shake-flask method

Sofosbuvir solubility studies have been performed with various solvents such as methanol, ethanol, water, and phosphate buffer solution ranging from pH 1.2 to 6.8. Approximately 10 mg of sofosbuvir was withdrawn into a standard 25 ml vial. To this was added 10 ml of solvent and kept at 25 - 30 °C for 48 h in an isothermal shaker. The drug is added until the stage of saturated equilibrium is reached. The solution was removed and centrifuged at 4000 rpm for 5 min, and the supernatant was filtered. The solubility was studied in UV-Visible spectroscopy at 260 nm [11].

Determination of wavelength by UV spectroscopy

A standard 10 ml vial containing approximately 10 mg of drug was weighed. To this was added 5 ml of ACN and the mixture was sonicated to completely dissolve the drug. ACN is used to make the final volume, which is 10 ml. 1 ml of the above stock solution is transferred to a second 10 ml bottle and the capacity is topped up with solvent. Between 200 and 400 nm, this final solution was scanned in a UV spectrophotometer.

Preparation of a stock solution and a working standard solution

Exactly 10 mg of sofosbuvir was calibrated in a 10 ml vial and dissolved with 4 - 6 ml of ACN. To produce a standard stock solution with a concentration of 1000 μ g/ml, the drug was first dissolved and then subjected to a 2 - 3-min sonication process. This process was followed by the addition of ACN to the final product. To determine the linearity of the substance, working standard solutions were developed by gradually diluting a stock solution produced in the concentration range of 10 - 60 μ g/ml.

Formation of SLNPs

Sofosbuvir SLNPs were primed by the microemulsion technique. About 30 mg of the drug was dissolved in 100 - 200 mg of stearic acid, which was kept at 70 °C in the magnetic stirrer (solution A). On the other hand, solution B

was prepared by mixing 10 to 40 mg of chitosan with 20 ml of distilled water in a 100 ml beaker. Solution B is added drop wise to solution A while using magnetic stirring to maintain a constant temperature of 70 °C. After complete mixing of solution B with solution A, the mixture is set aside with stirring to obtain a homogeneous solution [12]. The resulting mixture is poured dropwise into 250 ml of distilled water stirred in a Remi stirrer at a speed of 2000 rpm and a temperature range of 0 to 100 °C. The solution is stirred for 30 min after the previous solutions have been completely added. After 30 min, the resulting solution was frozen and dried in a freeze dryer [13].

Characterizations of SLNPs

A dynamic laser light scattering method was designed to control the zeta potential (ZP) and particle size (PS) of SLNPs [14]. To estimate the %EE, the SLNPs dispersion was centrifuged at 20,000 rpm at 10 °C for 120 min in a cooling centrifuge. The %EE was then calculated using equation 1.

$$\%EE = \frac{\text{Total quantity of drug} - \text{Quantity of drug present in supernatant}}{\text{Total quantity of drug}} \times 100 \quad (1)$$

The polydispersity index (PDI) is used to study the degree of PS dispersion of prepared SLNPs. The ideal PDI should be between 0 and 1. PS, PS distribution, morphological, topographical, compositional, and crystalline information were studied using a powerful high-resolution TEM microscope. Pre-dried SLNPs were imaged using TEM (200 kV TEM JEOL 2000FX) [15].

Cumulative drug release

Percentage cumulative drug release (%CDR) was calculated by applying the dialysis bag method. A dialysis bag was filled with 5 ml of SLNPs, and the ends were tied. The dialysis bag was immersed in 100 cm³ of pH 7.4 phosphate buffer containing 0.2% SLNPs while rotating at 75 rpm. During drug release tests, the dissolution medium was maintained at 37 °C ± 0.5 °C. At different intervals of time, up to 72 h, same volume of fresh dissolution medium was added in place of the 5 ml sample, and the absorbance was monitored by UV-Visible spectroscopy at 260 nm [16]. Calculation of %CDR of various formulations was calculated by using equation 2.

$$\%CDR = \frac{C_t}{C_0} \times 100 \quad (2)$$

Where C₀ is the quantity of SLNPs laden, and the amount of drug released at time t is represents as C_t.

Permeability study

The reverse chicken gut technique was used to perform permeability experiments for sofosbuvir SLNPs. An outer intestinal tract connected tubes A and B. The ends of the intestines were connected with a thread. The entire apparatus was submerged in the dissolving vessel. The study of *in vitro* drug dissolution and absorption patterns was performed at the same time as sampling. Dissolution and absorption studies have been performed. The temperature of the 1 L dissolution medium with deionized water was maintained at 37 ± 0.5 °C.

A fresh piece of intestine was attached to the infusion set. In the absorption compartment consisting of tubes A and B of the infusion set, the Krebs-Ringer solution had a volume of 35 ml [17]. The drug moved from the mucosal side of the dissolution medium to the serosa or absorption compartment [18]. The prepared sofosbuvir-SLNP4 was delivered at the scheduled expiration date of the system. 75 rpm was the rotational speed of the device. Two milliliter samples of the solution were taken every hour for up to three hours. After collecting solution samples with replacements at 15, 30, 45, 60, 75, 90, 105, 120, and 180 min, the released sofosbuvir was measured spectrophotometrically at a wavelength of 260 nm. A replacement solution (Krebs-Ringer solution) was used to remove the drug transported from the absorption compartment. New dissolution medium was used in three replicates (n = 3) of the study.

Analytical method validation

Linearity

Linearity test is carried out by comparing the observed results with reference standard values. Linearity response measurements were performed between 250 ng/ml and 60 µg/ml. To provide a range of concentrations, the stock solution was diluted with the mobile phase of ACN:phosphate buffer (60:40 %v/v). Data evaluation was performed using the linear regression equation. The peak area ratios of the reference compounds were developed based on the concentrations of the reference compounds.

Precision

Three separate doses (250 µg/ml, 10 µg/ml, and 50 µg/ml) were administered on the same day to assess intra-day precision. %RSD calculation has been performed ant this study was conducted over several days to confirm inter-day accuracy.

Accuracy

Three different point percentages (50, 100, and 150%) were used to assess accuracy. The samples were made by mixing the SLNPs formulation with regular sofosbuvir. Added chemical is recovered.

Limit of detection (LOD) and limit of quantification (LOQ)

LOD is the minimum concentration at which the analyte can be consistently identified. LOQ is the lowest amount of analyte in a sample that can be quantitatively measured with sufficient precision and accuracy for a given analytical process. LOD and LOQ were estimated by signal-to-noise ratio by comparing the measured signals of samples with known low analyte concentrations to those of pure samples; one can determine minimum concentration of the drug that can be identified and quantified. For assessing LOD, a signal-to-noise ratio between 3 and 2:1 and for that of LOQ 10:1 is considered sufficient. In accordance with ICH recommendations, the equation 3 and equation 4 can be used to determine LOD and LOQ, respectively [19].

$$LOD = 3.3 \times N/S \quad (3)$$

$$LOQ = 10 \times N/S \quad (4)$$

Where S is the slope of the associated calibration curve and N is the standard deviation of the compound's peak regions.

Stability study

The resilience of the standard samples produced was evaluated at room temperature (25 ± 0.5 °C) after an 8-h analysis of the solutions. The safety of samples during analysis is ensured by ensuring that the time spent setting up and analyzing samples has not led to degradation of the drug [20].

Ruggedness

Varying experimental conditions such as instruments, operators, solvents, reagent sources, and columns of similar types assessed the ruggedness of the method. A number of chromatographic parameters were evaluated, including asymmetric factor, capacity factor retention time, and selectivity factor.

Robustness

By injecting the standard solutions under optimal conditions, i.e., $\pm 1\%$ less ACN in the mobile phase and 0.1 ml less flow rate, the robustness of the approach was studied.

Forced degradation studies

As part of the degradation procedure, standard samples of sofosbuvir were subjected to acidic, alkaline, oxidative, thermal, and photostability conditions, as well as neutral conditions [21]. Acid degradation was obtained by refluxing the samples with 0.1 N HCl and alkaline degradation by refluxing them with 0.1 N NaOH at 60 °C for half an hour, respectively. To suppress oxidative degradation, 30% v/v HClO₄ was mixed and heated under reflux at 60 °C for 30 min. The sample was subjected to three different types of degradation: thermal, photostable, and neutral. The sample was heated in an oven at 60 °C for 72 h for thermal degradation; for photodegradation, a sample was exposed to 200 Wh/m² in a photostability chamber for 7 days; and the drugs were refluxed in water at 60 °C for six hours for neutral degradation. A final concentration of 50 µg/ml sofosbuvir was prepared by appropriate dilution. Sample stability can be determined by injecting 10 µl of the samples into the system and observing the chromatograms [22].

Results and Discussion

Sofosbuvir has been shown to be more stable, less toxic, biodegradable, more targeted and more biocompatible in solid lipid nanoparticles than in other approved carriers [23].

Preparation of SLNPs

The formulation of SLNPs was performed using the microemulsion method. The method is optimized based on the variation between drug, stearic acid, and chitosan. The F4 formulation showed a smaller particle size attributed to a lower lipid concentration. As the lipid concentration increased, the particle size increased. This is due to the surface coating of lipids to the drug on the surface. The formulation table is shown in table 1.

Table 1: Preparation of SLNPs of sofosbuvir.

S.No.	Content	Drug (mg)	Stearic acid (mg)	Chitosan (mg)	Speed (rpm)	Time (min)
1	F ₁	30	400	40	2000	30
2	F ₂	30	300	30	2000	30
3	F ₃	30	200	20	2000	30
4	F ₄	30	100	10	2000	30

Characterization of sofosbuvir SLNPs

ZP and PS

The PS of sofosbuvir SLNPs varied between 60.3 ± 2 nm and 72.4 ± 2 nm. In the middle of the preparations, the optimized nanoparticles with a sofosbuvir:stearic acid:chitosan ratio of 3.10:1 had the smallest PS with a typical diameter of 60.3 nm (Figure 1). Changing the mass ratio of stearic acid and chitosan leads to a noticeable change in PS. The smaller particle size is attributed to the lower lipid concentration. As the lipid concentration increased, the PS increased; this is due to the surface coating of lipids to the outer layer of the drug. The ZP of SLNPs ranged from -50 ± 1.5 to -67 ± 2.1 mV. Due to electrostatic repulsion, negative ZP values demonstrate nanoparticle stability. The PDI ranged from 0.862 to 1.125. The PDI indicate the homogeneous molecular weight distribution of the polymer chain in the SLNPs, the uniformity of the cross-linking and the formation of the network with a more ordered arrangement. The results of PS and ZP are presented in figure 2.

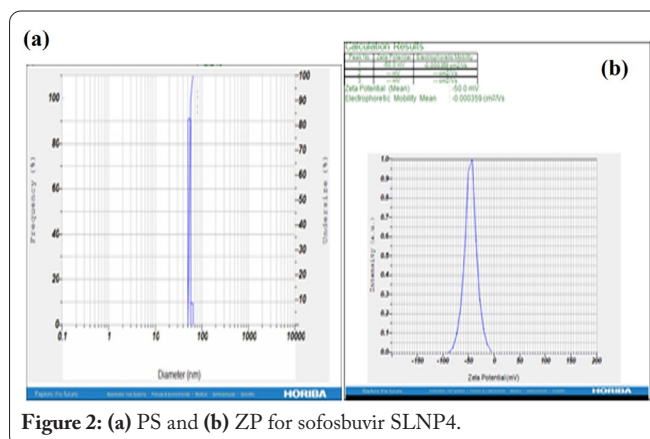


Figure 2: (a) PS and (b) ZP for sofosbuvir SLNP4.

TEM of SLNP4

TEM images of SLNP4 were observed in a spherical shape and uniform in size (Figure 3). No nanoparticle aggregation was observed. However, the PS determined by dynamic laser light scattering technique and the TEM result agreed well. The TEM data showed that the SLNP4 produced had a consistent spherical shape.

%EE

The %EE of SLNPs increased from 82.26 ± 1.9 to $91.43 \pm 1.1\%$. The results showed that the drug was effectively entrapped in the nanoparticles and hindered in the unit mass of the nanoparticles. The amount of solid lipids such as stearic

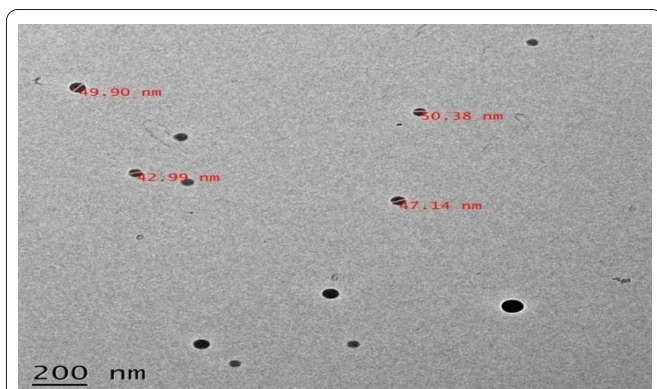


Figure 3: TEM images of sofosbuvir SLNP4.

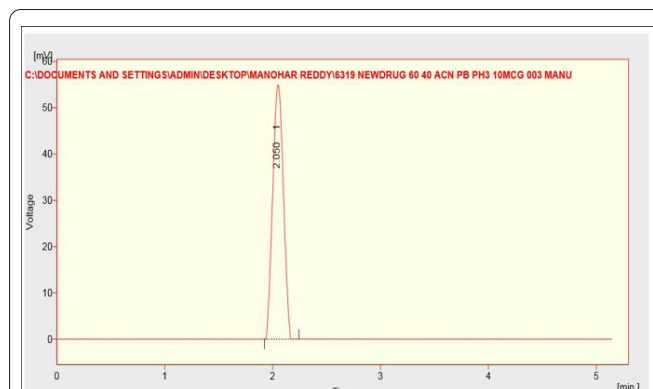


Figure 4: Representative chromatogram of sofosbuvir.

acid, surfactant, and stearic acid, among other variables, can affect the percentage of drug that is trapped in these systems; therefore, the most appropriate combination of lipid drug and surfactant should be chosen to achieve the highest purification efficiency [24]. Increasing the concentration of soy lecithin and stearic acid resulted in a decrease in %EE. The production of aggregates may be the reason why the EE decreased when the stearic acid concentration was increased.

Chromatographic conditions

Using the right combination of mobile phases, pH, erratic flow rate, elution type which is isocratic or gradient, type of column, temperature and wavelength, preliminary work was done to make the liquid chromatography method easier, more sensitive and cost effective to separate and quantify the drug. In the first step, the mobile phase combination consisted of ACN and phosphate buffer (60:40 v/v), the pH was adjusted between 2.5 and 4 and the mobile phase velocity was adjusted between 0.75 and 1, 2 ml/min. The mobile phase of ACN in combination with phosphate buffer at pH 3 and Phenomenex Luna C18 column (4.6 x 150 mm, 5) was chosen from the pilot study as it allows faster separation of chromatographic with appropriate peak symmetry and reduced peak tailing.

Solubility studies

The solubility study of sofosbuvir was performed by the shake flask method with various solvents such as chloroform, ACN, ethanol, methanol-ACN (50:50), water, methanol-water (50:50) [25]. Solubility results showed that in ACN the maximum solubility was observed at 9.27 mg/ml and the lowest in water, 0.13 mg/ml, respectively.

Method development

According to the ICH requirements (Q2R1), the technique has been validated for several parameters such as precision, linearity, specificity, accuracy, stability, LOQ, LOD and robustness [26].

Sofosbuvir had a retention time of 2.050 min as represented in figure 4. With concentrations between 10 and 100 µg/ml, the linearity of the method was assessed (Figure 5). The calibration curve was plotted using the analyte concentration and the response factor. This demonstrates that within the chosen analyte concentration range of 10 to 100 µg/ml, concentration and peak area show a linear relationship.

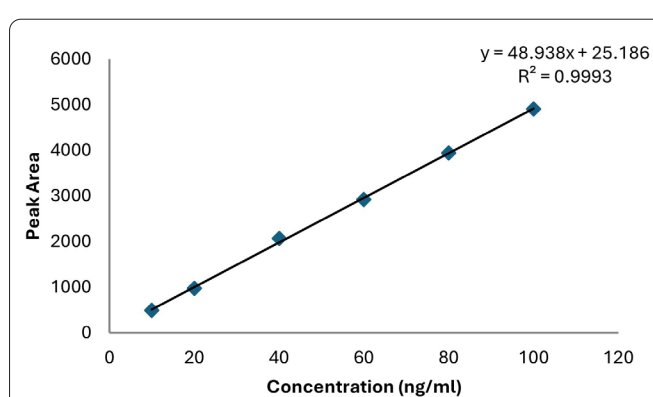


Figure 5: Linearity graph.

The results for slope, correlation coefficient and intercept were found to be 48.938, 0.993 and 25.186, respectively.

By adding standard solutions to previously examined sample solutions at three different levels of LQC, MQC and HQC, precision was assessed. A high recovery rate of 99.12 to 99.94% was obtained from the samples analyzed using the developed method.

For estimation of accuracy (Table 2) both intra-day (Table 3) and inter-day (Table 4) precisions were used. Repetitions of six at three different levels were prepared to assess intra-day precision. The table below contains the observed %RSD values. An inter-laboratory test was used to determine inter-day precision. According to table 3 and table 4, the RSD value was found to be less than 2%.

The signal-to-noise ratio was used to calculate the LOQ and LOD, which were estimated to be 10 ng/ml and 30 ng/ml, respectively. The %RSD was found to be less than 2%. The approach proved to be reliable and sustainable as the %RSD was less than 2%. During the investigation, the solution was found to remain stable.

All parameters related to system suitability are within acceptable ranges and comply with ICH guidelines. ICH guidelines require a plate count greater than 2000, a resistivity factor less than 2, and a resolution greater than 2. There were no deviations from limits in any of the appropriate system's suitability parameters. A study was conducted, and ICH (Q2R1) guidelines were followed to validate the method [27].

When blank and placebo are injected at the retention time

Table 2: Accuracy studies for the sofosbuvir.

S. No.	Concentration (µg/ml)		Measured concentration (µg/ml)	Mean	STDEV	% Nominal (%w/w)
	Actual concentration	Added concentration				
1	10	20	30.25	29.74	0.476	99.12
			29.31			
			29.65			
2	60	20	80.12	79.79	0.418	99.74
			79.32			
			79.93			
3	100	20	120.43	119.93	0.457	99.94
			119.54			
			119.81			

Table 3: Intra-day precision of sofosbuvir.

Concentration (µg/ml)	Peak area			%RSD (%)
	1 h	2 h	3 h	
10	510.04	510.12	511.01	0.48
60	2923.01	2923.15	2623.21	0.06
100	4914.03	4914.58	4914.88	0.04

Table 4: Inter-day precision of sofosbuvir.

Concentration (µg/ml)	Peak area			%RSD (%)
	1 st day	2 nd day	3 rd day	
10	504.98	506.81	504.80	0.81
60	2923.74	2933.108	2928.26	0.15
100	4912.85	4919.7234	4915.23	0.24

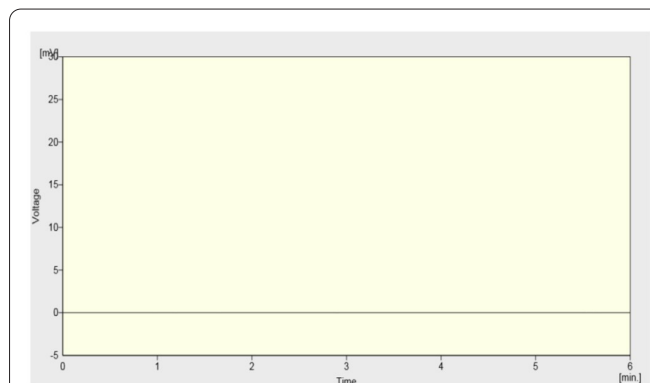


Figure 6: Chromatogram of blank.

of the drug, the method shows no interfering spikes as shown in figure 6 and figure 7, respectively.

Force degradation studies

Force degradation studies revealed that maximum degradation was observed in alkaline hydrolysis with 79.69% w/w at 24 h, with minimum degradation observed in thermal conditions at 40.91% w/w at 72 h, respectively (Table 5). Alkaline hydrolysis, oxidative degradation, photolytic degradation, acid degradation, and thermal degradation were found to be the order of deterioration. The studies suggest the analyte solution was established to be more stable in thermal degradative environment and highly susceptible to alkaline degradation.

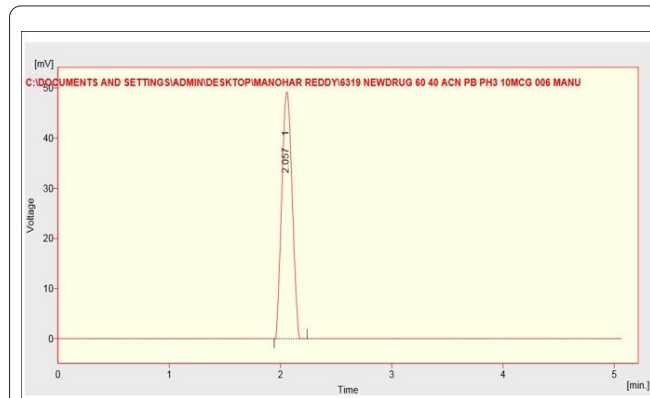


Figure 7: Chromatogram of standard.

Table 5: Forced degradation studies for sofosbuvir.

S. No.	Time (h)	Acid hydrolysis % degradation (%w/w)	Alkaline hydrolysis % degradation (%w/w)	Photolytic % degradation (%w/w)	Thermal % degradation (%w/w)	Oxidative % degradation (%w/w)
1	0	9.096386	39.36747	40.72289	35.93373	14.30723
2	1	36.77711	66.89759	37.13855	35.93373	58.10241
3	2	39.33735	69.45783	32.83133	35.93373	66.92771
4	3	39.27711	69.57831	41.86747	36.74699	69.36747
5	4	41.86747	77.95181	42.16867	38.88554	71.23494
6	6	50.8426	78.69124	54.1768	39.5000	72.2658
7	24	52.6543	79.69873	60.29697	40.1579	73.27869
8	48	51.85542	79.8253	63.57887	40.81325	74.24699
9	72	53.85542	80.63253	64.21687	40.91385	74.56588

Table 6: Drug release kinetics.

Formulation number	Zero order		First order		Higuchi's		Korsemeyer-Peppas's		
	K_0	R^2	K_1	R^2	KH	R^2	N	K_p	R^2
SLNP1	11.25	0.829	0.218	0.940	31.24	0.970	0.429	39.11	0.916
SLNP2	12.60	0.803	0.299	0.947	35.40	0.960	0.437	38.80	0.932
SLNP3	13.57	0.788	0.393	0.964	41.15	0.948	0.432	34.66	0.951
SLNP4	14.46	0.772	0.674	0.935	41.15	0.948	0.408	32.11	0.961

%CDR and release kinetics

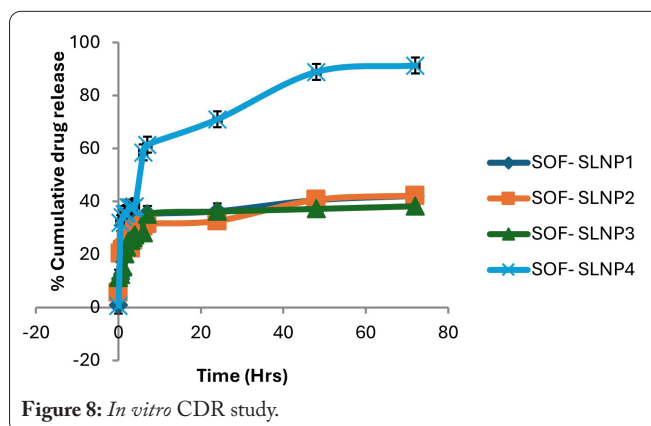
The %CDR is shown in figure 8 and ranged from 42.37 ± 1.3 to $91.26 \pm 1.8\%$ over a 72-h period. Compared to previous formulations, the sofosbuvir SLNPs4 formulation showed an excellent %CDR ($91.261.8\%$) at 72 h. With increasing drug concentration and a fixed stearic acid:chitosan release ratio of 10:1, marked variations in drug release were observed. Although the stearic acid:chitosan ratio (SLNP1 to SLNP4) changed, this also affected the amount of drug released. The result clearly shows that complex formation can temporarily delay drug release. The sofosbuvir:stearic acid:chitosan ratio (3:10:1) showed the maximum drug release, which can be attributed to the large surface area, small particle size and greater interaction with the dissolution medium. The sofosbuvir contained in the nanoparticles was dissolved by the delivery media. Large sized nanoparticles can create more vacuum space, slowing down the discharge of the particles. According to the %CDR, the drug was uniformly distributed or dissolved in the lipid surfactant matrix, and its release was caused by diffusion from the matrix. Table 6 presents the results of the kinetic drug release studies. The results of released kinetics of all formulations described how the Higuchi model and first order kinetics were used to predict the release pattern. Kinetic data showed that the SLNP4 formulation followed the Higuchi model ($R^2 = 0.948$ and "n" value = 0.408), demonstrating that the diffusion mechanism controls drug release from the nanoparticles.

Permeability study by everted chick intestinal sac method

To ensure that drugs are bioavailable, their permeability is crucial. Permeability was investigated using an inverted gut model [28]. A number of advantages can be derived from this model, including fewer samples required for the study, shorter experiment times, and reduced first-pass metabolism. According to the permeability data, the SLNPs produced had a permeability of 89.87%, compared to 40.76% for the pure drug.

Conclusion

Sofosbuvir is a low permeability drug that falls into BCS class-III. The use of sofosbuvir has increased in the global market. This drug shows less therapeutic efficacy when taken orally, which may be due to low permeability issues. The present work highlights the physical modification approach to increase the transmissivity of sofosbuvir. The SLNPs formulation of sofosbuvir has been developed and characterized. A permeability study showed that the SLNPs formulation of sofosbuvir has 2.2 times the permeability of the drug. A robust method of RP-HPLC was established and according to ICH

**Figure 8:** In vitro CDR study.

guidelines (Q2R1) was validated, where the results showed that all validation parameters were within limits. The formulation showed a smaller PS attributed to a lower lipid concentration. The established RP-HPLC method could be effectively used to estimate sofosbuvir in bulk and nanoformulations. This study helps in emerging nanoformulations of BCS class-III drugs with accurate, reliable, and well-developed methods, as well as increased permeability and bioavailability, helping to reduce dosages and minimize toxicity caused by excessive doses.

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Conflict of Interest

None.

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