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Research Article

PREPARATION OF IBRUTINIB-LOADED NANOSUSPENSION: *IN VITRO* AND *IN VIVO* PHARMACOKINETIC EVALUATION FOR ENHANCED DISSOLUTION AND BIOAVAILABILITY

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ABSTRACT

Objectives: The objective of this study was to develop and optimize an ibrutinib (IBR) nanosuspension (NS) formulation using the wet media milling technique and to evaluate its pharmacokinetic (PK) performance in comparison with the plain drug (PD) formulation under both fed and fasted conditions.

Methods: The IBR NS was formulated using a wet nano ball milling technique (Fritsch Pulverissette 7, Germany) with stabilizers such as Tween 80, sodium lauryl sulfate, and hypromellose. The optimized formulation was further processed using spray drying. The formulation was characterized for size, potential, and polydispersity using dynamic light scattering. The Fourier-transform infrared spectroscopy, differential scanning calorimetry, X-ray diffraction, and scanning electron microscopy techniques were employed for physicochemical characterization. Saturation solubility, dissolution, and stability studies were conducted to assess the formulation's performance. PK studies were performed on the IBR NS and PD (IBR PD) formulations in both fed and fasted conditions to evaluate key parameters such as C_{max}, T_{max}, half-life, area under the curve ($AUC_{0-\infty}$) and mean residence time.

Results: After spray drying, the IBR NS showed a notably lower particle size of 135.6 nm with a polydispersity of 0.389 and a zeta potential of −27.1 mV. The formulation showed a 3.786-fold surge in C_{max} and a 2.996-fold rise in $AUC_{0-24\ h}$ in comparison to the drug in fasting conditions. The IBR NS maintained consistent PK characteristics across fed and fasted conditions, demonstrating improved bioavailability. Saturation solubility experiments also indicated a 12.96-fold rise in solubility for the IBR NS versus to the typical medication.

Conclusion: The IBR NS formulation exhibited enhanced solubility, stability, and bioavailability compared to the PD formulation. The significant increase in PK parameters such as C_{max} and $AUC_{0-24\ h}$ underscores the potential of NS technology in proving the p.o bioavailability of poorly soluble drugs. This formulation can provide a more reliable therapeutic effect and has the potential for further clinical application in the treatment of chronic conditions.

Keywords: Ibrutinib, Nanosuspension, Bioavailability, Pharmacokinetics, Dissolution, Stability.

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INTRODUCTION

Cancer continues to be one of the most significant global health challenges, ranking among the leading causes of death worldwide. The uncontrolled growth, invasion, and, in some cases, metastasis of cancerous cells distinguishes malignant tumors from benign tumors, which remain localized. According to the International Agency for Research on Cancer, cancer claims around 8 million lives annually, with more than 14 million new diagnoses each year [1]. This number is expected to rise dramatically, with projections indicating 23.6 million new cases by 2030. The high mortality rate associated with cancer has spurred significant efforts to develop effective and rapid theranostic strategies to enhance cancer treatment [2].

Chemotherapy, radiation therapy, and surgery remain the primary treatment options for cancer. While these therapies can be effective at inhibiting tumor growth, they often come with severe side effects, including systemic toxicity, recurrence, and physiological complications. Despite these limitations, chemotherapy continues to be the first-line treatment approach [3]. However, chemotherapy's lack of specificity for cancer cells results in substantial toxicity to normal cells, leading to various adverse effects on the body [4,5].

An important challenge in cancer chemotherapy is the issue of drug resistance, which remains the leading cause of treatment failure and contributes significantly to cancer-related deaths [6]. Multidrug resistance (MDR) can arise from several mechanisms, including alterations in cell cycle regulation, reduced drug absorption, enhanced drug efflux, and suppression of apoptosis. The overexpression of P-glycoprotein (P-gp), a membrane transporter responsible for drug efflux, is a major contributor to MDR. This transporter actively pumps chemotherapeutic drugs out of cancer cells, preventing drugs from reaching therapeutic intracellular concentrations, thereby diminishing their efficacy [5].

To counteract MDR, research has focused on downregulating P-gp expression or inhibiting its activity to reduce drug efflux and maintain therapeutic drug levels inside cancer cells [7]. P-gp inhibitors have shown promise in enhancing the efficacy of chemotherapeutic agents by preventing their expulsion from cancer cells, but careful management of drug interactions and toxicity is crucial [8].

Ibrutinib (IBR), a targeted therapy, has shown efficacy in treating cancers such as chronic lymphocytic leukemia (CLL) and mantle cell lymphoma (MCL). It works by inhibiting Bruton's tyrosine kinase, a key enzyme involved in cancer cell proliferation [9]. Despite its therapeutic potential, IBR's effectiveness is compromised by its status as a P-gp substrate [10]. The active efflux of IBR from cancer cells through P-gp reduces its intracellular concentration, limiting its therapeutic benefits. Furthermore, the low oral bioavailability of IBR, which is around 2.9%, further impairs its clinical application, particularly as it falls under Class II of the biopharmaceutical classification system due to its low solubility and high permeability [11,12].

To address these challenges, our study explores the use of nanotechnology combined with P-gp inhibition to improve the pharmacokinetics (PK) of IBR and enhance its therapeutic potential. The strategy involves the formulation of IBR -loaded nanosuspensions (NS) using techniques such as nano milling and spray drying. In addition, we aim to incorporate P-gp inhibitors such as Vitamin E d-alpha-tocopherol polyethylene glycol 1000 succinate (TPGS), which has been shown to inhibit P-gp and improve the intracellular accumulation of drugs. By combining these approaches, we aim to increase the bioavailability of IBR, combat P-gp-mediated drug resistance, and ultimately improve treatment outcomes for cancer patients.

PK is a critical aspect of drug development, particularly for anticancer agents. PK studies involve the assessment of drug kinetics in the body. These studies provide vital information on how a drug behaves in the body, including its bioavailability, half-life, peak plasma concentration, and overall therapeutic efficacy. In the case of IBR, PK studies are essential to understanding the impact of its low bioavailability and the role of P-gp in limiting its therapeutic potential. By exploring the PKs of IBR in the context of nanoparticle delivery systems and P-gp inhibition, we can develop strategies that optimize drug delivery and improve patient outcomes [13-15].

In our investigation, we utilize NS technology to enhance the bioavailability of IBR. The ball milling technique, followed by spray drying, is employed to reduce the particle size (PS) and improve the dissolution rate of the drug [4]. We further stabilize the formulation using P-gp inhibitors, such as Vitamin E TPGS, which inhibit the efflux of IBR from cancer cells. The optimized formulation is then evaluated through various characterization techniques, including Fourier-transform infrared spectroscopy (FT-IR), differential scanning calorimetry (DSC), X-ray diffraction (XRD), and scanning electron microscopy (SEM). *In vitro* dissolution tests are performed to assess the release profile of the NS and its potential to enhance bioavailability [11]. Furthermore, PK studies are conducted to evaluate the *in vivo* performance of the formulation and its ability to improve drug absorption and therapeutic efficacy.

NSs, stabilized by excipients such as hydroxypropyl methylcellulose (HPMC) E15, sodium lauryl sulfate (SLS), and Vitamin E TPGS, are created using a 3-level, 3-factor Box–Behnken design (BBD) followed by spray drying. The characterization of these NSs is crucial in understanding their stability, morphology, and release behavior [16,17]. The PK studies, which focus on the drug's absorption, distribution, and metabolism, will offer valuable insights into the usefulness of the NS in overcoming P-gp-mediated resistance and improving the bioavailability of IBR. This approach aims to offer a scalable, cost-effective solution to enhance the treatment of cancers such as CLL and MCL, ultimately improving patient outcomes by providing more efficient drug delivery and better PK properties [18,19].

By employing nanotechnology and P-gp inhibition strategies, this research seeks to make significant advancements in the field of cancer therapy, addressing the challenges of drug resistance and low bioavailability associated with conventional chemotherapeutic agents such as IBR. The successful creation of an optimized NS could open the path for more efficient, targeted, and patient-friendly cancer therapies.

METHODS

Materials and methods

Dr Reddys Laboratories Pvt. Ltd., Hyderabad, India, provided us with IBR. Sigma-Aldrich® in India provides a variety of chemicals, such as TPGS, Tween®80, Hypromellose HPMC with viscosity of between 2.4 and 3.6 cP, SLS, and polyvinyl pyrrolidone K 30. Lactose monohydrate (pharmattose 200) was acquired from Tokyo Chemical Industry chemicals. Solvents of analytical grade were purchased from Merck Pvt Ltd, India.

Formulation and optimization of NS

The formulation and optimization of IBR -loaded NSs (IBR NS) were carried out using a two-stage process. In Stage A, IBR NS was prepared using a nano ball mill (Fritsch Pulverissette 7, Germany). To begin, 100 mg of the drug was added to 2 mL of an aqueous stabilizer solution (as outlined in Table 1), and the mixture was stirred with a vortex mixer to ensure complete wetting of the drug particles. The well-mixed dispersion was then transferred into a milling chamber containing 2g of 0.3 mm stainless steel balls. Milling was performed at varying rpm with 5-min on and 5-min off cycles to achieve the desired PS. PS, zeta potential (ZP), and polydispersity index (PDI) were measured in the resultant suspension. In Stage B, the formulated NS was processed using a Buchi mini spray dryer (B-190), which uses a cocurrent flow mechanism for atomization through a two-fluid nozzle. The NS was mixed with 5% w/v Vitamin E TPGS and 10% w/v lactose monohydrate before being sprayed. Operational parameters comprised an input temperature of 110°C, output temperature of 55°C, aspiration flow rate for air of 1400 rpm, and feed pump speed of 16 rpm. A cyclone separator was used to isolate the particles, which were then captured in a filter bag. The final product, SDP-F, was sealed in airtight containers for further analysis, focusing on PS and production yield [19].

Formulation by designing (FbD) method

Endorsed by regulatory authorities for its methodical and goal-oriented character, the creation of IBR-loaded NS follows a quality-by-design (QbD) methodology. Emphasizing risk awareness, data analysis, and experimental design (design of experiments [DoE]) during the product lifetime, this approach fits ICH Q8 recommendations. Compared to conventional approaches, QbD provides reasonably priced quality assurance and better process understanding. Beginning with the definition of a quality-targeted product profile and a risk analysis to identify critical success factors, critical process parameters (CPPs), and critical quality attributes (CQAs), the formulation by design (FBD) process systematically advances toward optimized product development. Experimental planning and factor screening optimization define possible product development regions. The Quality Target Product Profile points forth important qualities guaranteeing quality, safety, and effectiveness. Other influencing factors are classified as complete Freund's adjuvants and CPPs; CQAs have to stay within set limitations. This methodical technique helps to better grasp product creation. Under this system, regulatory bodies acknowledge operations.

Pre-screening investigations

Many different factors might affect CQAs, and carefully considering them throughout the design phase will require some time. This process was simplified by filtering the variables with the one-factor-at-a-time approach, therefore, lowering the influence of individual factors and enhancing the anticipated accuracy of the statistical model. The selected CQAs were PDI, PS, and ZP.

Experimental design

A BBD is used in the experimental setup to assess how milling duration, stirring rate, and stabilizer-to-drug concentration influence PS and PDI in NS formulations. Supported by Design Expert® software, response surface analysis allows us to see the correlations between parameters and important quality characteristics (CQAs). While checkpoint validation guarantees model correctness and product quality in line

Table 1: Information on DOE with variables and responses

Identification	Variables	Levels		
		−1	0	+1
A	Stabilizer to drug ratio	1.0	1.50	2.0
B	Stirring rate (rpm)	600	800	1000
C	Milling time (min)	30	45	60
Responses		**Constraints**		
Y1	Particle size (nm)	Minimize		
Y2	PDI	Minimize		

PDI: Polydispersity index, DOE: Design of experiments

with specified quality standards, a desirability function and visual optimization direct the formulation process.

NS characterization

The characterization of the IBR NS involved several analytical techniques to assess key properties. Dynamic light scattering with a Malvern ZetaSizer (Malvern design, UK-based company with model no: Nano ZS) was used to quantify PS, PDI, and ZP. The samples were reconstituted ten times with triple-distilled water before being examined at 25°C [19]. FT-IR Perkin-Elmer, model no: 1600 was used to evaluate the drug and excipients' interactions, with spectra recorded from 4000 to 400 cm^{-1} [23]. DSC (DSC-60, Shimadzu, Kyoto, Japan) was employed to assess thermal behavior and potential interactions between the drug and its excipients, with thermograms recorded under a nitrogen atmosphere [24]. XRD - model no. 6000 analysis was performed to examine the crystallinity of the formulations, with patterns recorded using a Philips X-ray diffractometer [27]. SEM was utilized to visually observe the surface morphology of the formulations at various magnifications [25]. Saturation solubility studies were conducted to compare the solubility of IBR, its physical mixture, and the NS formulations, with ultraviolet-visible spectrophotometry (V 650, Jasco, Japan) used to measure drug concentration [26]. Dissolution testing was carried out in biorelevant media to evaluate drug release in conditions mimicking *in vivo* environments, and stability studies were performed under different temperature conditions to assess the formulation's stability over 3 months [22,27].

Studies on PKs

The study's male Wistar rats were from Telangana, India's Nutrition National Institute. They weighed 200±20 g on average and were 4–5 weeks old. All animal experiments were conducted in strict accordance with the "Guidelines for the Care and Use of Laboratory Animals," and the protocols were officially approved by the Institutional Animal Ethics Committee (IAEC) under an assigned protocol number. Before the study, the animals were housed for 1 week in a natural light/dark cycle to acclimatize to a controlled environment at 20±2°C with a relative humidity of 40–60%. Six animals each were placed in four groups and labeled as a, b, c, and d. The animals in the first two groups were assigned to fast for the entire night. On the other hand, the remaining two groups were given meal access up to 30 min before to taking the drug. Groups B and D were given an optimized NS, whereas Groups A and C were given a normal IBR suspension dissolved in 0.5% w/v water-soluble polymer such as methylcellulose. A dose of 20 mg of IBR/kg of body weight was administered to each group [28].

High-performance liquid chromatography (HPLC) analysis

Analytical method development using reverse-phased HPLC (RP-HPLC)

Chromatographic analysis of IBR was performed on a Develosil ODS HG-5 RP C18 (150 × 4.6 mm, 5 μm) utilizing a RP-HPLC (Waters, Milford, USA model no: 2707) procedure. The mobile phase with the composition of 0.1% orthophosphoric acid: methanol with a 35:65 ratio at a 1.0 mL/min flow rate was used.

Accurately weighed the pure drug and sorafenib (IS) to produce a primary stock solution of 1 mg/mL. A calibration curve with a range of 0.250–200 ng/mL was then established using a secondary stock solution at a concentration (100 μg/mL).

Sample extraction for bioanalysis

Protein precipitation was utilized to extract the drug from plasma samples. In particular, rat plasma (50 μL) and acetonitrile (250 μL) were mixed and vortexed. After centrifuging the mixture for 10 min at 8500 rpm, the supernatant was subjected to chromatographic analysis at a wavelength of 287 nm.

Information analysis

We used WinNonlin (version 3.1; Pharsight Co., Mountain View, CA, USA) to look at the data and judge the concentration-time patterns. A non-compartmental approach was used to predict the PK parameters. This method analyses drug concentration-time data without making assumptions about specific patterns of distribution or elimination. This approach offers a simple and trustworthy way to assess important kinetics of NS and drugs.

Analytical statistics

The standard deviation (SD) of the PK variables was given as mean±SD. GraphPad Prism (GraphPad Software, version 8.05, CA), a statistical program, was used to perform additional analysis on the variables.

RESULTS AND DISCUSSION

Formulation of IBR NS

In the present study, the IBR NS was formulated using a top-down wet nano ball milling technique (Fritsch Pulverissette 7, Germany), which has garnered significant attention in recent years for its effectiveness in producing NSs [29,30]. Various stabilizers, including Tween 80, SLS, and hypromellose, were employed to stabilize the suspension, with a combination of hypromellose and SLS yielding the most promising results. To come up with the optimal NS formulation, the DoE was utilized with the usage of surface response graphs. Following this, the optimized formulation was subjected to spray drying for further processing [31-34]. The combination of wet milling and optimization through DoE has proven to be an efficient strategy for formulating NS s with improved stability and performance [19,23].

Characterization of IBR NS

The IBR NS was successfully formulated using a wet nano ball milling technique, resulting in a PS of 307.5±12.98 nm and a PDI of 0.084±0.013, indicating a homogeneous system. After spray drying, the PS was reduced to 135.6 nm, with a PDI of 0.389, and the ZP improved from −15.3 mV to −27.1±0.26 mV, confirming the stabilizing effect of steric stabilizers [35]. FT-IR, DSC, and XRD analyses revealed no significant chemical interactions between the drug and stabilizers, and the drug showed no melting peak in the NS formulation, indicating a molecular solid-state complex [13,36,37]. Saturation solubility studies demonstrated that the NS had a 12.96 times higher solubility compared to the pure drug [38], and dissolution tests showed improved drug release in both fasted and fed state conditions, with the NS exhibiting superior release profiles [39].

Stability experiments showed that while PS rose after 90 days, it stayed constant for 60 days at 2–8°C, suggesting a decline in stability with time that could be caused by loss of integrity of polymeric stabilizer with time. Ostwald ripening would cause a rise in PS by way of the lower surface covering [39]. The formulation showed improved solubility, dissolution, and stability relative to the pure drug, as evidenced by the increase in particle size from 135.6 ± 8.89 nm to 146.25 ± 10.22 nm at 40°C [40].

The faster rate at which the NS dissolves than the PD can be elucidated using Noyes-Whitney formulae. As per the same, the rate at which any drug dissolves will go up if the particles are smaller and the surface area is bigger. Size, form, state (amorphous or crystalline), and habit (needle or spherical) of the particles, as well as their state, mostly affect how quickly and easily a medicine dissolves in physiological settings.

PK studies

Analytical and bioanalytical method development using reverse-phased high-performance liquid chromatography

Using RP-HPLC at a wavelength of 287 nm, a mobile phase of 0.1% orthophosphoric acid and methanol in a 35:65% v/v ratio at a flow rate of 1.0 mL/min was used to conduct the study. 0.997 was the regression coefficient (R^2). By contrasting the sample's chromatogram with a blank one, specificity was evaluated. Analyzing the analyte response extracted from the biological matrix and comparing it to the pure standard yielded the percentage recovery. Using the protein precipitation procedure, 250 μL of acetonitrile was added to extract IBR from plasma samples. The drug's HPLC chromatogram and the internal standard are shown in Supplementary Fig. 1 [14].

PKs

The PK comparison between the NS (IBR NS) and conventional drug (IBR PD) formulations, under both fed and fasted conditions, revealed notable differences in drug absorption and bioavailability. Table 2 presents key PK data, and Fig. 1 illustrates the plasma concentration-time profiles for both formulations. For the plain drug (PD), significant changes were observed among fed and fasted states, with a decrease in half-life after food intake, alongside increased C_{max} and area under the curve (AUC_{0-24}) h. By contrast, the PK properties of the NS (IBR NS) did not vary much between fed and fasted states, implying that the formulation is less influenced by food consumption. Under fasting conditions, the NS formulation showed a 3.786-fold increase in C_{max} and a 2.996-fold rise in $AUC_{0-24\ h}$ relative to the PDs. Increased surface area and PS reduction with increased surface area may explain the higher bioavailability of the drug, as they lower diffusion layer thickness, improve drug solubility, and hasten the rate of drug absorption. Increased adhesive surface area between small-sized particles and intestinal epithelium of villi also enables direct contact among the surface and immediate release of the drug, making it more available at the absorption site and resulting in high absorption, distribution, and high bioavailability. Smaller PS increases drug bioavailability by P-gp efflux reduction. Clinically, especially for poorly soluble medications, these changes can enable more efficient drug distribution, hence improving therapeutic results and lowering dose frequency. In the end, this might mean improved patient compliance and maximized therapeutic effectiveness. Our results confirm earlier research showing that NSs increase bioavailability by lowering PS. This method improves solubility and absorption degree, therefore, providing a hopeful solution for improved medication delivery [13,19,24]. These findings support the potential of nanocrystal formulations in enhancing drug bioadhesion and bioavailability, offering significant advantages over conventional drug formulations, particularly in overcoming the impact of food intake [20,21].

The PK parameters presented for the PD (IBR) and the NS formulation (IBR NS) under both fasted and fed conditions provide valuable insights into the bioavailability and drug absorption characteristics of the two formulations.

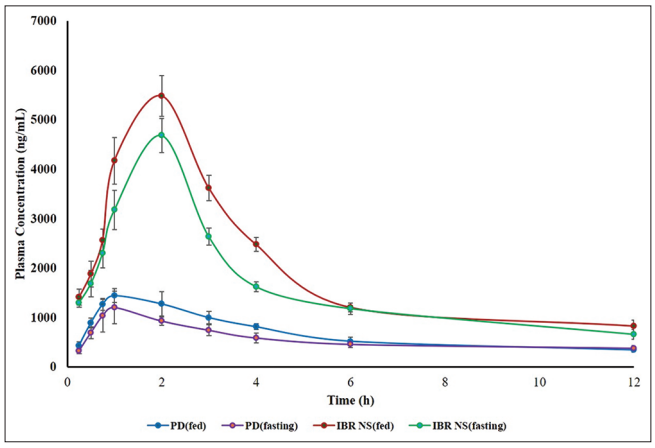

Fig. 1: Pharmacokinetic profiles in rats following oral administration. (n=3) Mean±SD was used to express all of the values

In both fed and fasted conditions, IBR NS's peak plasma concentration (C_{max}) is much higher than that of the ordinary medication. In the fed state, the C_{max} of IBR NS (5484.4±414.66 ng/mL) is more than 3 times higher than the PD (1448.22±142.93 ng/mL). In the fasted state, IBR NS also demonstrates a higher C_{max} (4686.82±348.66 ng/mL) compared to the PD (1204.88±329.91 ng/mL). This suggests that the NS formulation enhances the absorption and bioavailability of IBR, likely due to the reduced PS and increased surface area, which facilitates faster absorption and higher drug concentrations in the plasma.

The time to reach peak concentration (T_{max}) is delayed for IBR NS compared to the PD. While both formulations reach their peak plasma concentration at 1 h in the fed state, IBR NS takes 2 h to reach T_{max} in both the fasted and fed conditions, indicating a slower absorption rate. This could be due to the formulation's longer residence time in the gastrointestinal tract or a more gradual dissolution process of the nanosized particles.

The half-life of IBR in both formulations differs between the fasted and fed states. For the PD, the half-life is longer in the fasted state (7.139±0.38 h) compared to the fed state (5.199±0.82 h), suggesting that the absorption rate is slower when no food is present. Conversely, the half-life of IBR NS is shorter in the fasted state (3.901±0.78 h) than in the fed state (6.368±0.46 h). This may be due to the faster absorption and elimination of the drug in the fasted state, driven by the improved solubility and dissolution properties of the NS.

The IBR NS has a much larger area under the concentration-time curve from time 0 to the last dose that can be measured (AUC_{0-t}) than the PD. In the fed state, the AUC_{0-t} of IBR NS (24246.53±520.44 ng.h/mL) is more than 3 times higher than the PD (8174.50±408.12 ng.h/mL), and in the fasted state, it is also significantly increased (19787.76±329.26 ng.h/mL vs. 6832.74±533.61 ng.h/mL). This increased AUC_{0-t} indicates greater overall exposure to the drug in the body, which is a direct consequence of the enhanced solubility and bioavailability of the NS.

The $AUC_{0-\infty}$, which represents the total drug exposure over time, is also higher for IBR NS in both fed and fasted states. In the fed state, IBR NS has an $AUC_{0-\infty}$ of 28941.56±959.76 ng.h/mL, while the PD has 10756.22±844.92 ng.h/mL. In the fasted state, the $AUC_{0-\infty}$ for IBR NS is 25864.82±1288.041 ng.h/mL, compared to 10788.86±799.623 ng.h/mL for the PD. The larger $AUC_{0-\infty}$ for IBR NS further confirms the increased bioavailability and extended exposure to the drug due to the NS formulation [40].

The mean residence time (MRT) for IBR NS is shorter than that for the PD in both the fasted and fed states. In the fasted state, IBR NS has a lower MRT (6.28±4.68 h) compared to the PD (11.29±4.26 h). Similarly, in the fed state, IBR NS has a lower MRT (6.28±4.68 h) compared to the PD (8.08±3.26 h). A lower MRT indicates faster elimination of the drug from the body, which could be related to the enhanced dissolution rate and faster absorption of the NS formulation.

Table 2: Pharmacokinetic parameters

Parameters	Plain drug (IBR)		Formulation (IBR NS)	
	FED	FAST	FED	FAST
Cmax (ng/mL)	1448.22±142.93	1204.88±329.91	5484.4±414.66	4686.82±348.66
Tmax (h)	1	1	2	2
Half-life (h)	5.199±0.82	7.139±0.38	3.901±0.78	6.368±0.46
AUC_{0-t} (ng.h/mL)	8174.50±408.12	6832.74±533.61	24246.53±520.44	19787.76±329.26
$AUC_{0-\infty}$ (ng.h/mL)	10756.22±844.92	10788.86±799.623	28941.56±959.76	25864.82±1288.041
MRT (h)	8.08±03.26	11.29±4.26	6.28±4.68	8.146±5.20

IBR: Ibrutinib, NS: Nanosuspension, AUC: Area Under The Curve, MRT: Mean Residence Time, SD: Standard Deviation. All The Values Were Expressed In (n=3) Mean±SD

The findings show generally that the NS formulation of IBR greatly increases the bioavailability of the medication in both the fed and fasted conditions. The formulation leads to higher plasma concentrations, increased AUC values, and improved dissolution characteristics, making it a promising approach for improving the therapeutic efficacy of IBR. The slower T_{max} and lower MRT observed for IBR NS may reflect the prolonged absorption and increased drug residence time in the system, which may improve the drug's therapeutic window[40].

CONCLUSION

The development of the IBR NS significantly enhanced the bioavailability of the drug compared to the PD formulation (IBR PD). The NS demonstrated a marked increase in C_{max} and $AUC_{0-24\ h}$, irrespective of fed or fasted conditions, indicating its superior and stable absorption profile. A small-sized particle will have increased surface area, and the enlarged diffusion layer improving interaction with the intestinal lining accounts for this change. The formulation's ability to maintain consistent PK characteristics across different feeding states highlights its potential to overcome food-related variations in drug absorption. All things considered, IBR NS offers a hopeful way to increase the bioavailability of badly soluble medications such as IBR, thereby providing more therapeutic potential and consistency.

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AUTHOR'S CONTRIBUTION

VD and PR did the research, execution, and writing; the work plan, review, and revisions were done by others. The two writers support the submission and publishing. Every author has read and consented to the published version of the paper.

ETHICAL APPROVAL

IAEC authorized the animal protocol under no: 1447/PO/Re/S/11/CPCSEA-80/A.

CONFLICT OF INTEREST

The authors utter no conflict of interest.

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