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Research Article

In vivo Brain Pharmacokinetics of Dolutegravir Sodium-Loaded Nanostructured Lipid Carrier *in situ* Gel: Comparative Study with an Intravenous Drug Solution

Salam Shanta Taher*, Khalid Kadhem Al-Kinani

Department of Pharmaceutics, College of Pharmacy, University of Baghdad, Baghdad, Iraq

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Abstract

Background: Dolutegravir sodium (DTG), used to treat HIV, faces challenges in delivering effective therapeutic concentrations to the brain due to the blood-brain barrier (BBB). Nanostructured lipid carriers (NLCs) combined with *in situ* gels present a promising strategy for enhancing brain drug delivery via the intranasal route. ***Objective***: To compare brain pharmacokinetics of DTGs delivered via NLC-loaded *in situ* gel intranasal administration with the conventional intravenous (IV) drug solution. ***Methods***: 80 Wistar rats, which were divided into three groups: two groups consisting of 39 animals each and a control group with 2 animals. Rats were administered with a dose of 1.0 mg/kg of DTGs IV, and DTGs NLC-loaded *in situ* gel were administered intranasally. DTGs were determined in rats' plasma and brain tissue by high-performance liquid chromatography (HPLC). ***Results***: Intranasal administration produced significantly higher brain drug concentrations (C_{max} 35344.8ng/ml) compared to the IV solution (C_{max} 4536.85ng/ml). The area under the curve (AUC) for the intranasal formulation was twice that of the IV solution, indicating enhanced bioavailability. Furthermore, the intranasal route exhibited a faster onset (lower T_{max}) and prolonged retention in brain tissue. The developed nanoformulation exhibited a Drug Targeting Efficiency (DTE) of 232.5% and a Drug Targeting Potential (DTP) of 57%, suggesting improved brain targeting efficiency. ***Conclusions***: The DTGs-loaded NLC in situ gel shows superior brain pharmacokinetics compared to IV administration, highlighting its potential as an effective strategy for enhancing brain targeting.

Keywords: Dolutegravir sodium, HIV, *in situ* gel, NLC.

حركية دواء دولوتغرافير الصوديوم في الدماغ المقدم عبر الهلام الموضعي المحمل بالناقلات الدهنية النانوية: تحليل مقارن مع إعطاء محلول الدواء عن طريق الوريد

الخلاصة

الخلفية :يواجه دواء دولوتيجرافير الصوديوم(DTG) ، المستخدم في علاج فيروس نقص المناعة البشرية(HIV) ، تحديات في تحقيق تركيزات علاجية فعالة في الدماغ بسبب حاجز الدماغ الدموي (BBB)، تمثل الحاملات الدهنية النانوية المنظمة (NLCs) عند دمجها مع المواد الهلامية المتشكلة موضعيا استراتيجية واعدة لتعزيز إيصال الدواء إلى الدماغ عبر المسار الأنفي. **الهدف :**مقارنة الحركية الدوائية في الدماغ لعقار DTG المُعطى عبر هلام متشكل موضعيا المحمل بـ NLC عبر الأنف مع محلول الدواء التقليدي المُعطى وريدياً. **الطرائق:** تم استخدام 80 جرذًا من نوع ويستر، مقسمة إلى ثلاث مجموعات: مجموعتان تضمان 39 جرذًا لكل منهما، ومجموعة ضابطة تحتوي على جرذين. تم إعطاء الجرذان جرعة 1.0 ملغم/كغم من DTG وريدياً، بينما تم إعطاء الهلام المتشكل موضعيا المحمل بـ NLC عبر الأنف. تم تحديد تراكيز DTGفي بلازما ودماغ الجرذان باستخدام كروماتوغرافيا السائل عالية الأداء(HPLC) . **النتائج:** أظهر الاعطاء الانفي تراكيز دوائية أعلى بشكل ملحوظ في الدماغ (C_{max}= 35344.8) نانوغرام/مل (مقارنةً بالمحلول الوريدي (C_{max}= 4536.85 نانوغرام/مل. كانت المنطقة تحت المنحنى (AUC) للتركيبة الأنفية ضعف ما هو عليه للمحلول الوريدي، مما يشير إلى تحسين التوافر البيولوجي. أظهر المسار الأنفي بدء تأثير أسرع(T_{max} اقل) واحتفاظًا مطولًا بالدواء في أنسجة الدماغ. أظهرت الصيغة النانوية المطورة كفاءة استهداف دوائي (DTE) بلغت 232.5%وإمكانات استهداف دوائي (DTP) بنسبة **57%**، مما يشير إلى تحسن كفاءة توصيل الدواء إلى الدماغ. **الاستنتاجات:** أظهر الهلام داخل الموقع المحمل بـ NLC لعقار DTG تفوقًا في الحركية الدوائية في الدماغ مقارنةً بالإعطاء الوريدي، مما يؤكد إمكاناته كاستراتيجية فعالة لتعزيز استهداف الدواء للدماغ.

* ***Corresponding author***: Salam S. Taher, Department of Pharmaceutics, College of Pharmacy, University of Baghdad, Baghdad, Iraq; Email: sallam.hashem@copharm.uobaghdad.edu.iq

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INTRODUCTION

The central nervous system (CNS), alongside the peripheral reservoir, represents a significant target for HIV, contributing to various neurological disorders, including neuroAIDS. The infection of the central nervous system by HIV is linked to the emergence of asymptomatic neurocognitive impairment, HIV-associated mild neurocognitive disorder (HAND), and HIV-associated dementia (HAD). These conditions collectively manifest as a clinical syndrome characterized by cognitive, motor, and behavioral dysfunction [1]. Antiretroviral therapy (ART) is the cornerstone of managing HIV/AIDS. Antiretroviral therapy (ART) has demonstrably enhanced the morbidity and mortality rates among individuals infected with HIV-1 by lowering plasma viral loads and restoring immune function. Nevertheless, despite the overall improvement in outcomes for AIDS patients undergoing ART, neurocognitive impairments persist in approximately up to 50% of individuals suffering from HIV-1 encephalopathy [2]. These observations may be linked to the development of resistance to or failure of ART in AIDS patients [3]. Additionally, various physiological barriers significantly impede the effectiveness of CNS-

targeted therapeutics. The blood-brain barrier is particularly notable as it serves as a major obstacle for treating neurological conditions, as it limits the transfer of current antiretroviral (ARV) medications from the bloodstream to the brain, thereby diminishing the efficacy of existing treatments. Consequently, addressing the challenges associated with drug delivery to the brain is essential for the advancement of therapeutic strategies aimed at treating CNS disorders [4]. In recent years, intranasal (IN) drug administration has emerged as a non-invasive and accessible route capable of delivering medications for both localized and systemic effects [5]. This approach facilitates the direct transport of drugs from the nasal cavity to the brain due to the anatomical connections between the central nervous system (CNS) and the nasal passage [6]. Compared to other administration routes, IN delivery offers several advantages, including bypassing the blood-brain barrier (BBB), enabling quicker drug delivery to the brain, and improving drug targeting as well as bioavailability. Additionally, intranasal (IN) administration bypasses gastrointestinal and hepatic metabolism, reduces drug deposition in non-target tissues, and minimizes undesirable systemic adverse effects [7]. These features make the nose-to-brain pathway a promising strategy for managing CNS disorders, including neuroaids [8]. However, there are challenges associated with developing novel nasal formulations for effective nose-to-brain delivery. First, the volume that can be administered per nostril in humans is limited to under 200 µL, making it difficult to use this route for drugs that require larger doses. Second, mucociliary clearance, a protective mechanism that eliminates bacteria, toxic substances, and inhaled particles from the body, can lead to the rapid loss of delivered drugs, restricting their retention time in the nasal passage to less than 30 minutes. Third, nasal enzymes can metabolize certain drugs, necessitating protective strategies against enzymatic degradation [9]. In recent years, various formulation strategies, including nanovesicles, spanlastics, microemulsion, and lipid-based nanoformulations (solid lipid nanoparticles and nanostructured lipid carriers), have been designed to overcome these challenges [10-12]. NLCs show great potential for nose-to-brain drug delivery by enhancing the absorption and bioavailability of poorly soluble drugs through intranasal administration. NLCs offer several benefits, including the avoidance of toxic organic solvents during formulation, enabling controlled drug release, ensuring biocompatibility, and protecting the encapsulated drugs from pre-systemic metabolism [13,14]. Additionally, nano-sized NLCs enable efficient transport of drugs and internalization into the brain via endocytosis and transcytosis mechanisms. The rapid elimination of the drug from the nasal cavity is attributed to the high turnover rate of mucus. Therefore, the drug delivery system must possess mucoadhesive characteristics to prevent the concurrent elimination of the drug with mucus**.** One of the most frequently employed approaches is the *in situ* gelling formulation. Upon intranasal administration, this system transitions from a solution to a gel in response to physiological triggers such as temperature, ion concentration, or pH, facilitating more accurate dosing [15]. Among all in situ gelling systems, thermosensitive formulations are particularly advantageous for drug delivery, as they undergo gelation upon administration due to temperature changes. The most commonly used mucoadhesive polymer in these formulations is poloxamer 407 (Pluronic F127), which micellizes and gels as the temperature increases. To formulate thermosensitive *in situ* gels for nasal application, it is essential to establish the transition temperature within the range of 30-34°C corresponding to the temperature of the nasal mucosa [16]. Thermosensitive in situ gels are designed to remain in a liquid form at lower temperatures and transition into a gel upon contact with the nasal mucosa, allowing for easy administration and extended retention in the nasal cavity. Even if the product changes to a gel state during shipping or storage due to temperature fluctuations, it will revert to a dispersion upon cooling and then transition back into a gel when exposed to body temperature, without compromising its performance or efficacy. Embedding NLC into *in situ* gels presents a promising approach for nose-to-brain delivery of neurotherapeutics by merging the benefits of nanostructured lipid carriers and gel-based delivery systems [17]. Dolutegravir sodium, an HIV integrase inhibitor, is used in combination with other antiretroviral drugs for the management of HIV infection. It has a molecular weight of 441.4 g/mol, is highly lipophilic (log P 2.2), and exhibits low aqueous solubility (95 mg/L at 25°C) [18]. The therapeutic effect and safety profile of dolutegravir, when used alongside other anti-HIV drugs, have been validated in numerous clinical studies. However, dolutegravir demonstrates limited suppression of HIV within the brain, even when administered as part of well-tolerated combination antiretroviral therapy (cART). This limitation may be attributed to the tight junctions of the blood-brain barrier and the activity of efflux transporters on brain cells, such as astrocytes and microglia, indicating that these cells could contribute to suboptimal drug concentrations in the brain [19,20]. Therefore, an effective delivery system is essential to address these obstacles and facilitate the transport of dolutegravir into the central nervous system. Sahoo and collaborators developed dolutegravir-loaded NLCs to improve the drug's bioavailability. The pharmacokinetic profile of the optimized NLC demonstrated significant advantages over the aqueous suspension of the pure drug, offering potential for therapeutic and commercial applications [21]. Devika Sonawane and Varsha Pokharkar successfully formulated a donepezil-loaded NLC *in situ* gel, enhancing both pharmacokinetic and pharmacodynamic performance. When compared to the intravenous drug solution, the in situ gel exhibited improved pharmacokinetic properties. Notably, the designed donepezil in situ gel exhibited a Drug Targeting Efficiency (DTE) of 232.5% and a Drug Targeting Potential (DTP) of 57%, highlighting its exceptional brain-targeting ability through the use of NLCs [22]. The study aims to assess the in vivo brain

pharmacokinetics of dolutegravir sodium-loaded nanostructured lipid carriers incorporated into an *in situ* gel formulation following nasal administration in comparison to the intravenous (IV) administration of a dolutegravir solution.

METHODS

Materials

Dolutegravir sodium was purchased from Xi'an Healthful Biotechnology Co., Ltd. (China). HPLC-grade acetonitrile and water were purchased from Chem-Lab (Belgium). Soluplus and Kolliphor® P 407 were obtained from D-BASF Co., Ltd. Paracetamol (internal standard, IS) was supplied by the College of Pharmacy, University of Baghdad.

Preparation of dolutegravir sodium-NLC in situ gel

Firstly, dolutegravir-loaded nanostructured lipid carriers (DTG NLCs) were prepared using the melt emulsification-ultrasonication technique. GMS (solid lipid) and Triolein (liquid oil) were melted at 80°C, and the drug was dissolved in the molten lipid phase. An aqueous phase containing 4.0% w/v Tween-40, 1% PEG 400, and 50 mg of Soluplus was added dropwise while stirring at 850 rpm for 30 minutes to form a hot pre-emulsion. After cooling to room temperature, the pre-emulsion was sonicated for 2.5 minutes to obtain the NLC dispersion. The in situ gel formulation was prepared by incorporating a specified amount of poloxamer 407 into the prepared NLC dispersion., stirred continuously for one hour, and then stored in a refrigerator (4ºC) to allow the mucoadhesive components to fully dissolve. Moreover, benzalkonium chloride at a concentration of 0.01% w/v was incorporated into the final formulation to prevent the growth of microorganisms [23]. The final compositions of the optimized in situ gel formulation were depicted in Table 1.

Table 1: Composition of *in situ* nasal gel of DTGs NLC

Drug/Excipients	Compositions
NLC dispersion	0.3 %
Poloxamer 407 (%w/v)	19%
Carbopol 934 (%w/v)	0.1 %
Benzalkonium chloride (%w/v)	0.01%
D.W q. s.	10 mL

In vivo pharmacokinetic study

Female Wistar albino rats weighing between 200 and 250 grams were obtained from the National Cancer Institute and the College of Pharmacy at the University of Baghdad for use in the *in vivo* studies. Prior to the study, we gave the selected animals a one-week period to adapt to the experimental conditions of temperature and humidity. During the experiment, they were housed in groups within plastic cages under controlled temperatures (27±2°C) and a 12-hour light/dark cycle. The animals did not receive any food the night before the experiment. The *in vivo* experiments performed on the rats were authorized by the Research Ethics Committee for Experimental Investigations, College of Pharmacy, Baghdad University, Iraq, under protocol number REC03024108A.

Animal grouping and administration of medication

The selected rats were randomly assigned to three groups. The first group, which included two rats, was used as a negative control and was killed early so that plasma and brain tissue could be used for HPLC calibration. This made sure that the drug concentration was measured correctly. The second group, consisting of 39 rats, was administered an in situ gel of DTG NLC intranasally at a dose of approximately 1 mg/kg. A micropipette instrument was used to deliver 40 µL of the formulation into each nostril. The third group (39 rats) received the intravenous form of dolutegravir sodium solution at the same dose (about 1.0 mg/kg). After the completion of the treatment, the experimental animals were anesthetized via intraperitoneal (IP) injection with a dose of 80 mg/kg of ketamine and 10 mg/kg of xylazine. Once the animals were fully anesthetized, they were sacrificed through cardiac puncture [24,25]. Following the experimental period for each group, the euthanized rats were dissected, and their brain tissues were harvested and weighed for subsequent analysis. Prior to weighing, the entire brain tissue was rinsed with normal saline to eliminate any adhering tissue or fluids. Each weighed rat brain was then preserved in a cold solution of physiological saline at a 1:6 dilution by weight. The brain tissue was then homogenized using a homogenizer and subsequently stored at -21°C for HPLC analysis. Two ml of blood samples were withdrawn from the heart via puncture at certain time points spanning from 0 to 48 hours. Rat blood samples were collected into EDTA-coated tubes and immediately separated. Plasma samples were acquired using the centrifugal process (Hettich, Germany), where blood samples were spun at a speed of 4000 revolutions per minute for a duration of 15 minutes. We obtained plasma samples from the clear portion (supernatant), transferred them into Eppendorf tubes, and stored them in the freezer for further analysis [26].

HPLC method

Isocratic chromatographic separation was carried out using a C18 Luna column (5 µm, 150 x 4.6 mm, Phenomenex) with a reversed-phase stationary phase. The mobile phase was a mixture of 50% acetonitrile (ACN) and 50% OPA buffer (adjusted at pH 3). The flow rate of the mobile phase was controlled at 0.8 mL/min, and the column temperature was maintained at 30°C. The effluent was monitored at a wavelength of 258 nm using a PDA detector. The total run time was 10 minutes [27].

Pharmacokinetic and neuropharmacokinetic parameter estimation

Pharmacokinetic parameters were analyzed utilizing Excel PK-Solver® by plotting DTG concentration against time [28]. Non-compartmental analysis was applied to derive parameters such as C_{max}, T_{max}, and AUC. Furthermore, neuro-pharmacokinetic

parameters , including the percentage of targeting efficiency (%DTE) and the percentage of drug targeting potential (% DTP), were also evaluated [29]. %DTE is an indicator of drug accumulation in the brain after intranasal (IN) administration compared to intravenous (IV) administration and is expressed by the following equation:

$$\% \text{ DTE} = \frac{(AUC \text{ brain IN} / AUC \text{ plasma IN})}{(AUC \text{ brain IV} / AUC \text{ plasma IV})} \times 100$$

A %DTE value exceeding 100% indicates effective targeting of the brain.

%DTP represents the proportion of a drug that reaches the brain through direct pathways, such as the olfactory and trigeminal routes, and is described as shown below.

$$\%DTP = \frac{AUC_{brain,\ IN} - \left(\frac{AUC_{brain,\ IV}}{AUC_{blood,\ IV}} x AUC_{blood,\ IN}\right)}{AUC_{brain,\ IN}} \times 100$$

A positive value of %DTP signifies that direct pathways substantially contribute to the overall drug delivery to the brain.

Statistical analysis

The results were presented as mean values with their standard deviation ± SD (n = 3). A difference was considered statistically significant if the P-value was below 0.05. The pharmacokinetic parameters, including Cmax, Tmax, and AUC0-48, undergo a statistical analysis using a student's t-test.

RESULTS

The HPLC method was used to determine the amount of dolutegravir sodium in plasma and brain tissue. The HPLC chromatograms for the drug and internal standard are illustrated in Figure 1.

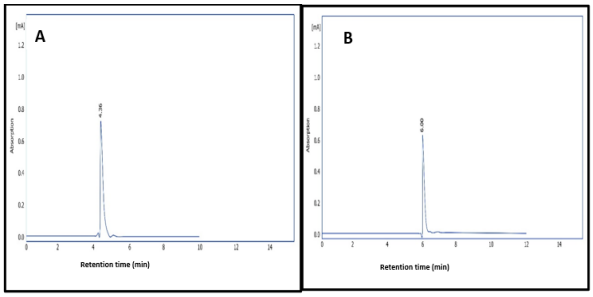

Figure 1: Chromatogram of **A**) Dolutegravir sodium and **B)** Internal standard.

HPLC method validation was conducted in accordance with International Conference on Harmonization (ICH) guidelines, evaluating linearity, accuracy, precision, limit of detection (LOD), and limit of quantification (LOQ). The method showed a linear response across six concentration levels, ranging from 50 to 2000 ng/mL. The correlation coefficients (R^2) of the standard drug demonstrated a good linear correlation (R^2 of 0.999), as presented in Figure 2.

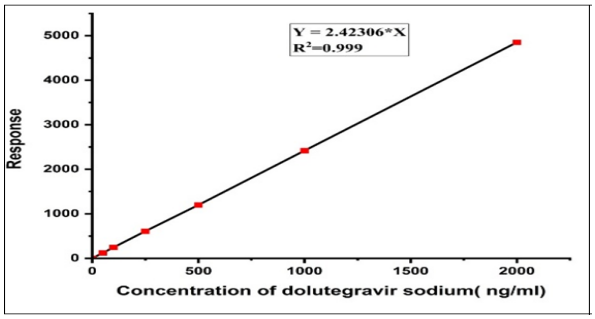

Figure 2: Calibration curve for the standard solution of Dolutegravir sodium.

The LOD and LOQ values were determined from the calibration curve: 10 and 33.3 ng/mL, respectively. The intra-day and inter-day precision, assessed with quality control samples and reported as the relative standard deviation (RSD), ranged from 0.63% to 1% (Table 2).

Table 2: Intra-day and inter-day precision of dolutegravir sodium

Drug	Concentration (ng/ml)	Intraday conc. (ng/ml)	%RSD	Interday conc. (ng/ml)	%RSD
Dolutegravir sodium	50	49.97 50.78 49.85	1%	50.10 49.57 50.13	0.63%

Validated HPLC analysis yielded retention times of 4.2 and 6.0 minutes for DTGs and the internal standard, respectively, confirming successful chromatographic separation. The chromatograms obtained from the analysis of rat plasma and rat brain tissue demonstrated no interference from endogenous substances with the analytes and the internal standard (IS) at their respective retention times, as shown in Figure 3. The concentration-time profile of DTGs in the brain tissue, following a single dose of nasal in situ gel of the optimized NLC and intravenous administration of the drug solution in rats, is depicted in Figure 4.

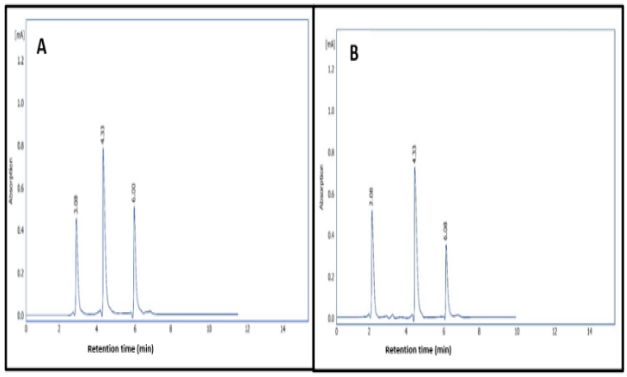

Figure 3: Chromatogram of **A)** plasma spiked with dolutegravir sodium and IS and **B)** Tissue spiked with dolutegravir sodium and IS.

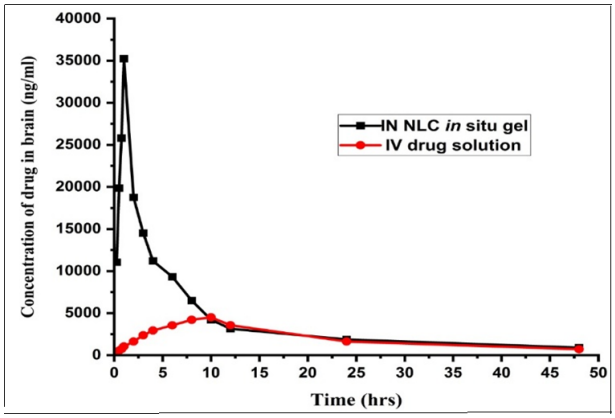

Figure 4: Drug brain levels following a single dose of Dolutegravir sodium (1 mg/kg) after intravenous administration (0.5 mL) or optimized *In* Situ Gel (40 µL) by nasal route. Measurements are reported as mean±SD (n = 3).

The principal pharmacokinetic parameters are outlined in Table 3. Two distinct kinetic profiles were observed for both IN and IV administrations, with nasal delivery yielding significantly higher ($p< 0.005$) drug concentrations in the brain. The highest concentration was observed in the brain region after IN administration; the C_{max} was 35344.8±3626 ng/mL at a T_{max} of 60 min, whereas the C_{max} was 4536.85±102 ng/mL at a T_{max} of 9.33±1.1 hr after IV administration. Additionally, the area under the curve ($AUC_{0-\infty}$) was markedly higher for dolutegravir NLC-loaded in situ gel ($p< 0.001$) compared to its free form. DTGs-loaded NLC showed the highest DTE% and DTP% of 232.5 and 57, respectively, which confirms that nanocarrier-loaded in situ gel enhanced the brain-targeting efficiency of the incorporated drug compared to the free drug solution.

Table 3: Main pharmacokinetic parameters of DTGs in the brain after IN administration of DTGs-NLCs, and I.V administration of DTGs-solution

Pharmacokinetic parameters	IN DTGs-NLCs	IV DTGs-solution
Cmax (ng/ml)	35344.8±36	4536.85±102
Tmax (h)	1.25±0.6	9.33±1.1
AUC_{0-48} (ng.h/ml)	193082.6±16	96233.3±921
%DTE	232.5	
%DTP	57	

DISCUSSION

The application of a calibration curve for estimating the Limit of Detection (LOD) and Limit of Quantification (LOQ) enables a comprehensive analysis of the method's detection limits. The high correlation coefficient (R^2 of 0.999) of standard dolutegravir sodium also shows a good linear relationship, which proves the accuracy of the method. The precision, represented by relative standard error (RSD) values below 2%, ensures that the proposed HPLC method is highly precise and appropriate for quantifying the compound in both plasma and brain tissue [30]. When given through the nose, DTGs are absorbed quickly, and a much higher concentration (about 35344.8 ng/ml) reaches the brain within an hour compared to when given through an IV (about 4536.85 ng/ml). The T_{max} for intranasal delivery was lower (1.0 hour) compared to intravenous delivery (~10 hours), suggesting fast drug transport from the nose to the brain. Additionally, intranasal administration resulted in a nearly 9-fold increase in the C_{max} value compared to intravenous administration. The elevated C_{max} observed with the nasal route might be due to direct absorption through the nasal mucosa, including the olfactory region, which provides a direct pathway to the brain, bypassing the blood-brain barrier more efficiently. Additionally, the mucoadhesive properties of *in situ* gel may contribute to decreased mucociliary clearance in rat models, thus prolonging the residence time of the drug in the nasal cavity and enhancing the absorption of DTGs from the nasal epithelium [31]. Finally, intranasal delivery limits the dilution of the drug in systemic circulation, ensuring a higher concentration reaches the brain more rapidly than through intravenous administration, where the drug is distributed more broadly in the body before reaching the brain. These combined factors explain why intranasal administration leads to both a higher peak concentration and a faster onset compared to intravenous administration. The limited quantity of DTGs that reaches the brain during intravenous therapy is due to the restricted permeation of dolutegravir from the circulating plasma into the brain, owing to the highly selective nature of the semipermeable endothelial cells that line the tight junctions of the blood-brain barrier. The levels of DTGs in the brain in both therapies appear to decrease swiftly, indicating the rapid elimination of the drug from the brain tissue [32]. A comparative analysis of the bioavailability of dolutegravir sodium, measured by the AUC, revealed a 2-fold higher concentration in the brain after IN administration than after administration of IV drug solution, thus confirming the potential effectiveness and practical feasibility of intranasal NLC incorporated in situ gel for brain targeting. Overall, the pharmacokinetic findings indicate that intranasal delivery of the optimized in situ gel significantly enhanced the brain bioavailability of dolutegravir sodium in comparison to intravenous administration. High values of DTE and DTP indicate a well-designed colloidal dispersion capable of effectively reaching and engaging the HIV reservoir in brain tissue by trigeminal or olfactory pathways. These findings align with earlier studies that demonstrated nanoformulations improve the direct transport of drugs to the brain, including frovatriptan, asenapine, and buspirone [33-35]. The significant improvement in the pharmacokinetics and neuropharmacokinetics of DTG NLCs is primarily ascribed to the advantageous characteristics of the nano-sized particles and the effective utilization of the nasal route for administration. The nano-sized particles of DTG NLCs significantly enhance transcellular

transport through olfactory neurons to the brain tissue. The carriers also enable targeted drug release, promoting consistent therapeutic concentrations while reducing systemic exposure. Furthermore, the lipid-based matrix of the NLCs enhances drug retention in brain tissues and minimizes premature clearance. Additionally, the nose-to-brain delivery route plays a crucial role in bypassing the blood-brain barrier, facilitating direct drug transport to the brain, and reducing systemic exposure. Together, these factors underscore the potential of NLC-based delivery systems to optimize CNS drug delivery and therapeutic outcomes.

Conclusion

This study explored the efficacy of a nanostructured lipid carrier (NLC) based in situ gel for the delivery of dolutegravir to the brain via nasal administration. A key aspect of this investigation was the pharmacokinetic assessment of the dolutegravir-loaded NLC in situ gel formulation at a nasal physiological pH of 6. The pharmacokinetic and biodistribution studies showed that the optimized formulation led to a much higher concentration of the drug in the brain compared to the drug solution that was given intravenously. A lower T_{max} may also mean that the drug is absorbed more quickly, possibly through the olfactory region in the nose. Therefore, the optimized DTGs-loaded NLC incorporated in the in situ gel could be considered a novel and promising approach for the management of neuro-AIDS.

Conflict of interests

No conflict of interest was declared by the authors.

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The authors did not receive any source of funds.

Data sharing statement

Supplementary data can be shared with the corresponding author upon reasonable request.

REFERENCES

1. Kakad SP, Kshirsagar SJ. Neuro-AIDS: Current status and challenges to antiretroviral drug therapy (ART) for its treatment. *CDTH*. 2020;15(5):469–481. doi: 10.2174/1574885515666200604123046.
2. Taher SS, Al-Kinani KK. Current nanotechnological strategies for delivery of antiretroviral drugs: Overview and future prospects. *CDTH*. 2024;20. doi: 10.2174/0115748855331460241017100207.
3. Nastri BM, Pagliano P, Zannella C, Folliero V, Masullo A, Rinaldi L, et al. HIV and drug-resistant subtypes. *Microorganisms*. 2023;11(1):221. doi: 10.3390/microorganisms11010221.
4. Mastrangelo A, Gama L, Cinque P. Strategies to target the central nervous system HIV reservoir. *Curr Opin HIV AIDS*. 2024. doi: 10.1097/COH.0000000000000847
5. Jaafer H, Al-Kinani KK. Formulation and evaluation of idebenone microemulsion as a potential approach for the transmucosal drug delivery systems. *IJPS*. 2024;33(1):79–88. doi: 10.31351/vol33iss1pp79-88.
6. Erdő F, Bors LA, Farkas D, Bajza Á, Gizurarson S. Evaluation of intranasal delivery route of drug administration for brain targeting. *Brain Res Bull*. 2018;143:155–170. doi: 10.1016/j.brainresbull.2018.10.009.
7. Keller LA, Merkel O, Popp A. Intranasal drug delivery: opportunities and toxicologic challenges during drug development. *Drug Deliv and Transl Res*. 2022;12(4):735–757. doi: 10.1007/s13346-020-00891-5.
8. Sarma A, Das MK. Nose to brain delivery of antiretroviral drugs in the treatment of neuroAIDS. *Mol Biomed*. 2020;1(1):15. doi: 10.1186/s43556-020-00019-8.
9. Patharapankal EJ, Ajiboye AL, Mattern C, Trivedi V. Nose-to-brain (N2B) delivery: An alternative route for the delivery of biologics in the management and treatment of central nervous system disorders. *Pharmaceutics*. 2023;16(1):66. doi: 10.3390/pharmaceutics16010066.
10. Muhammed SA, Al-Kinani KK. Formulation and in vitro evaluation of meloxicam as a self-microemulsifying drug delivery system. *F1000Res*. 2023;12:315. doi: 10.12688/f1000research.130749.1.
11. Jaber SA, Rajab NA. Preparation and In vitro/ex vivo evaluation of nanoemulsion-based in situ gel for intranasal delivery of lasmiditan. *Iraqi J Pharm Sci*. 2024;33(3):128–141. doi: 10.31351/vol33iss3pp128-141.
12. Alkufi HK, Kassab HJ. Soluplus-stabilized nimodipine-entrapped spanlastic formulations prepared with edge activator (Tween20): Comparative physicochemical evaluation. *Pharm Nanotechnol*. 2024;13. doi: 10.2174/0122117385348551241028102256.
13. Shah B, Khunt D, Bhatt H, Misra M, Padh H. Intranasal delivery of venlafaxine loaded nanostructured lipid carrier: Risk assessment and QbD based optimization. *J Drug Deliv Sci Technol*. 2016;33:37–50. doi: 10.1016/j.jddst.2016.03.008.
14. Tamer MA, Kassab HJ. Optimizing intranasal amisulpride loaded nanostructured lipid carriers: Formulation, development, and characterization parameters. *Pharm Nanotechnol*. 2024;13(2):287–302. doi: 10.2174/0122117385301604240226111533.
15. Koo J, Lim C, Oh KT. Recent advances in intranasal administration for brain-targeting delivery: A comprehensive review of lipid-based nanoparticles and stimuli-responsive gel formulations. *Int J Nanomedicine*. 2024;19:1767-1807. doi: 10.2147/IJN.S439181.
16. Alaayedi MH, Maraie NK. Effect of pluronic F127 concentration on gelling temperature and other parameters of lomustine mucoadhesive in-situ gel. *Iraqi J Pharm Sci*. 2024;33(3):63–71. doi: 10.31351/vol33iss3pp63-71.
17. Rajab NA, Jawad MS. Preparation and evaluation of rizatriptan benzoate loaded nanostructured lipid carrier using different surfactant/co-surfactant systems. *Int J Drug Deliv Technol*. 2023;13(01):120–126. doi: 10.25258/ijddt.13.1.18.
18. Belgamwar AV, Khan SA, Yeole PG. Intranasal dolutegravir sodium loaded nanoparticles of hydroxypropyl-beta-cyclodextrin for brain delivery in neuro-AIDS. *J Drug Deliv Sci Technol*. 2019;52:1008–1020. doi: 10.1016/j.jddst.2019.06.014.
19. Patel SH, Ismaiel OA, Mylott WR, Yuan M, Hauser KF, McRae M. Simultaneous determination of intracellular concentrations of tenofovir, emtricitabine, and dolutegravir in human brain microvascular endothelial cells using liquid chromatography-tandem mass spectrometry (LC-MS/MS). *Analytica Chimica Acta*. 2019;1056:79–87. doi: 10.1016/j.aca.2019.01.015.
20. Dharshini KP, Devi DR, Banudevi S, Narayanan VHB. In-vivo pharmacokinetic studies of Dolutegravir loaded spray dried Chitosan nanoparticles as milk admixture for paediatrics infected with HIV. *Sci Rep*. 2022;12(1):13907. doi: 10.1038/s41598-022-18009-x.
21. Sahoo L, Jena GK, Patro CS, Patro CN, Meher NR. In vitro and in vivo characterization of transdermal patch loaded with nanostructured lipid carrier for bioavailability enhancement of dolutegravir sodium using Taguchi and Box-Behnken design. *BioNanoSci*. 2023;13(3):1213–1230. doi: 10.1007/s12668-023-01143-9.
22. Sonawane D, Pokharkar V. Nose to brain targeting of the donepezil nanostructured lipid carrier *in situ* gel: formulation, *in vitro* , *ex vivo* , *in vivo* pharmacokinetic and pharmacodynamic characterization. *RSC Pharm*. 2024;1(4):820–840. doi: 10.1039/D4PM00174E.
23. Mohanty D, Alsaidan OA, Zafar A, Dodle T, Gupta JK, Yasir M, et al. Development of atomoxetine-loaded NLC in situ gel

for nose-to-brain delivery: Optimization, in vitro, and preclinical evaluation. *Pharmaceutics*. 2023;15(7):1985. doi: 10.3390/pharmaceutics15071985.

24. Yaribeygi H, Hemmati MA, Nasimi F, Maleki M, Jamialahmadi T, Reiner I, et al. Sodium glucose cotransporter-2 inhibitor empagliflozin increases antioxidative capacity and improves renal function in diabetic rats. *J Clin Med*. 2023;12(11):3815. doi: 10.3390/jcm12113815.
25. Underwood W, Anthony R. AVMA guidelines for the euthanasia of animals: 2020 edition. Retrieved in March. 2020 Mar;2013(30):2020-1.
26. Hamzah M, Kassab H. Formulation and characterization of intranasal drug delivery of frovatriptan-loaded binary ethosomes gel for brain targeting. *Nanotechnol Sci Appl*. 2024;17:1–19. doi: 10.2147/NSA.S442951.
27. Al-Tamimi D, Al-Kinani K, Taher S, Hussein A. Effect of food on the pharmacokinetics of fluoxetine in healthy male adult volunteers. *Iraqi J Pharm Sci*. 2023;31(Suppl.):153–161. doi: 10.31351/vol31issSuppl.pp153-161.
28. Ramyasree A, Umadevi S. An efficient RP-HPLC-PDA method for estimating dolutegravir and lamivudine in combined pharmaceutical formulations using a Box-Behnken design approach. *Int J Pharm Qual Assur*. 2023;14(03):507–513. doi: 10.25258/ijpqa.14.3.08.
29. Abdelbary GA, Tadros MI. Brain targeting of olanzapine via intranasal delivery of core–shell difunctional block copolymer mixed nanomicellar carriers: In vitro characterization, ex vivo estimation of nasal toxicity and in vivo biodistribution studies. *Int J Pharm*. 2013;452(1–2):300–310. doi: 10.1016/j.ijpharm.2013.04.084.
30. Bhandari R, Kuhad A, Paliwal JK, Kuhad A. Development of a new, sensitive, and robust analytical and bio-analytical RP-HPLC method for in-vitro and in-vivo quantification of naringenin in polymeric nanocarriers. *J Anal Sci Technol*. 2019;10(1):11. doi: 10.1186/s40543-019-0169-1.
31. Nair AB, Chaudhary S, Shah H, Jacob S, Mewada V, Shinu P, et al. Intranasal delivery of darunavir-loaded mucoadhesive in situ gel: Experimental design, in vitro evaluation, and pharmacokinetic studies. *Gels*. 2022;8(6):342. doi: 10.3390/gels8060342.
32. Nair AB, Chaudhary S, Jacob S, Patel D, Shinu P, Shah H, et al. Intranasal administration of dolutegravir-loaded nanoemulsion-based in situ gel for enhanced bioavailability and direct brain targeting. *Gels*. 2023;9(2):130. doi: 10.3390/gels9020130.
33. Hamzah ML, Kassab HJ. Frovatriptan succinate intranasal delivery for brain targeting – in vivo study. *Iraqi J Vet Med*. 2023;47(2):101–109. doi: 10.30539/me2mm152.
34. Singh SK, Dadhania P, Vuddanda PR, Jain A, Velaga S, Singh S. Intranasal delivery of asenapine loaded nanostructured lipid carriers: formulation, characterization, pharmacokinetic and behavioural assessment. *RSC Adv*. 2016;6(3):2032–22045. doi: 10.1039/C5RA19793G.
35. Noorulla KM, Yasir M, Muzaffar F, Roshan S, Ghoneim MM, Almurshedi AS, et al. Intranasal delivery of chitosan decorated nanostructured lipid carriers of Buspirone for brain targeting: Formulation development, optimization and In-Vivo preclinical evaluation. *J Drug Deliv Sci Technol*. 2022;67:102939. doi: 10.1016/j.jddst.2021.102939.

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