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Evaluating Transdermal Delivery of Labetalol Hydrochloride: Role of Penetration Enhancers

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Abstract
Transdermal drug delivery has gained attention as a viable alternative to traditional oral and parenteral routes, especially for drugs with significant first-pass metabolism such as labetalol hydrochloride. This study investigates the feasibility of delivering labetalol hydrochloride through the transdermal route and examines the impact of various chemical penetration enhancers on its skin permeability. Labetalol hydrochloride, a widely used antihypertensive agent, suffers from reduced oral bioavailability, making controlled transdermal administration a potential strategy to maintain therapeutic plasma concentrations with improved patient compliance. In this research, in vitro permeation studies were conducted using excised rat skin in Franz diffusion cells. Penetration enhancers such as ethanol, propylene glycol, oleic acid, and dimethyl sulfoxide (DMSO) were incorporated into the drug formulation to assess their effect on transdermal flux. The cumulative drug permeation and enhancement ratios were calculated to evaluate efficacy. Results demonstrated a significant increase in permeability with selected enhancers, particularly oleic acid and DMSO, without causing notable skin irritation. The findings suggest that transdermal delivery of labetalol hydrochloride is feasible and can be significantly improved with appropriate penetration enhancers. This approach may provide a more effective and patient-friendly alternative for long-term hypertension management.

Keywords: Transdermal delivery, Labetalol hydrochloride, Penetration enhancers, Franz diffusion cell, Skin permeability

Introduction

Transdermal drug delivery systems (TDDS) have emerged as a promising alternative to conventional oral and parenteral routes, offering several advantages such as controlled release, improved patient compliance, and avoidance of first-pass metabolism. Among various therapeutic agents, labetalol hydrochloride, a combined α- and β-adrenergic blocker commonly

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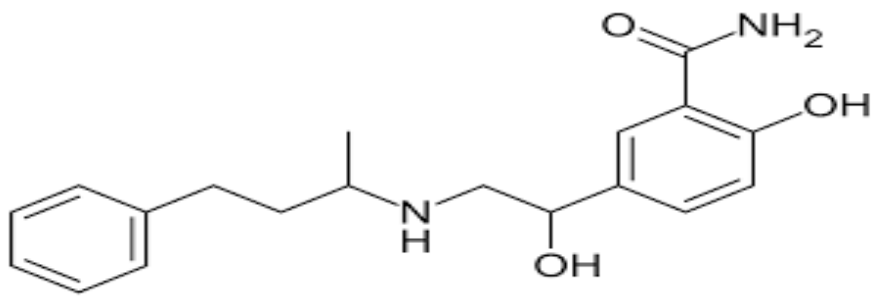
used in the management of hypertension, presents an interesting candidate for transdermal administration. Despite its efficacy, the oral bioavailability of labetalol hydrochloride is relatively low due to extensive first-pass hepatic metabolism. This pharmacokinetic limitation justifies the exploration of alternative routes, such as transdermal delivery, which could provide sustained therapeutic levels, reduce dosing frequency, and minimize systemic side effects. However, the stratum corneum, the outermost layer of the skin, poses a significant barrier to drug permeation, particularly for hydrophilic compounds like labetalol hydrochloride.

To overcome this barrier, the use of chemical penetration enhancers has been widely investigated. These agents temporarily disrupt the skin structure or alter its properties to facilitate increased drug permeation without causing irreversible damage. Various enhancers such as ethanol, propylene glycol, dimethyl sulfoxide (DMSO), and oleic acid have shown potential in promoting drug flux across the skin. The choice and concentration of enhancer, as well as its compatibility with the drug and formulation, play a crucial role in determining the success of transdermal delivery. This study aims to assess the feasibility of transdermal delivery of labetalol hydrochloride and systematically evaluate the influence of selected penetration enhancers on its skin permeability. By comparing the permeation profiles and enhancement ratios, the research seeks to identify optimal conditions under which labetalol hydrochloride can be effectively delivered transdermally, thus offering a novel and patient-friendly route for antihypertensive therapy.

DRUG PROFILE

LABETALOL HYDROCHLORIDE



Labetalol is a medication prescribed for treating high blood pressure and managing angina over

the long term. It's used in various conditions like essential hypertension, hypertensive emergencies, and pregnancy-related hypertension. It can be taken orally or through IV injection, but it's generally not the first choice for treating essential hypertension compared to other available options.

Common side effects of labetalol include orthostatic hypotension (a blood pressure drop when standing), dizziness, fatigue, and nausea. More serious side effects may include severe hypotension, liver problems, heart failure, and bronchospasm. It's generally considered safe for use in later stages of pregnancy and doesn't pose significant risks during breastfeeding. Labetalol works by blocking the activation of β- and α-adrenergic receptors.

Labetalol was first patented in 1966 and became available for medical use in 1977. It's also available as a generic medication. In the United States in 2020, it was the 210th most commonly prescribed medication, with over 2 million prescriptions filled.

Medical uses

Labetalol demonstrates efficacy in the control and management of various medical conditions characterized by hypertension. It is notably effective in addressing hypertensive emergencies, postoperative hypertension, hypertension associated with pheochromocytoma, and hypertension that arises as a rebound effect following the withdrawal of beta-blockers.

One of its specific and crucial indications lies in the treatment of pregnancy-induced hypertension, particularly when linked to pre-eclampsia, a condition that can pose serious risks during pregnancy. Additionally, labetalol serves as a valuable alternative for the treatment of severe hypertension, offering a therapeutic option in cases where blood pressure control is of utmost importance.

Special populations

Pregnancy: While studies conducted in laboratory animals have not indicated any harm to the developing fetus, there is a lack of comparable, well-controlled studies conducted in pregnant women. Therefore, caution is advised when considering the use of labetalol during pregnancy.

Nursing: Labetalol has been detected in small quantities in breast milk (approximately 0.004% of the original dose). As a result, healthcare providers should exercise caution when prescribing labetalol to breastfeeding mothers.

Pediatric: There is a dearth of studies that have established the safety and efficacy of labetalol in the pediatric population. Therefore, its use in children and adolescents is not well-documented.

Geriatric: Elderly individuals are more prone to experiencing dizziness as a side effect when taking labetalol. As such, labetalol dosing in the elderly should be approached with caution, and patients should be counseled regarding this potential side effect.

Side effects

This data provides information on the side effects and adverse reactions associated with the use of a medication, possibly labetalol, which is commonly prescribed for conditions like hypertension. Let's break down and explain the data:

1. **Neurologic Side Effects:**
    - **Headache (2%):** About 2% of individuals who take this medication may experience headaches as a side effect.
    - **Dizziness (11%):** Dizziness is more common, affecting around 11% of users. This side effect can make individuals feel unsteady or lightheaded.
2. **Gastrointestinal Side Effects:**
    - **Nausea (6%):** Nausea is reported in approximately 6% of users, indicating a sensation of feeling sick to the stomach.
    - **Dyspepsia (3%):** Dyspepsia, which refers to indigestion or discomfort in the upper abdomen, affects around 3% of users.
3. **Cholinergic Side Effects:**
    - **Nasal Congestion (3%):** About 3% of individuals may experience nasal congestion, which is a stuffy or blocked nose.
    - **Ejaculation Failure (2%):** This side effect, affecting 2% of users, refers to difficulty or the inability to ejaculate.
4. **Respiratory Side Effect:**
    - **Dyspnea (2%):** Dyspnea, or difficulty breathing, is reported by 2% of users.
5. **Other Side Effects:**
    - **Fatigue (5%):** Fatigue, or a feeling of tiredness, is experienced by approximately 5% of individuals.
    - **Vertigo (2%):** Vertigo, characterized by a spinning sensation or dizziness, affects around 2% of users.

- **Orthostatic Hypotension:** Orthostatic hypotension refers to a drop in blood pressure when standing up. This side effect is noteworthy, as it can be more severe and more common with the intravenous (IV) formulation (58%) compared to the oral formulation (1%). This is an important consideration because the severe drop in blood pressure when standing can limit the use of higher doses of the oral formulation.

this data provides insights into the potential side effects associated with the use of this medication. It also highlights a significant difference in the occurrence and severity of orthostatic hypotension between the intravenous and oral formulations, which can impact the dosing and management of the medication for certain individuals. Patients should be aware of these potential side effects and discuss them with their healthcare providers.

Mechanism of action

Labetalol is a medication that acts as a beta-blocker by blocking β-adrenergic receptors, specifically both β1 and β2 receptors. Additionally, it has intrinsic sympathomimetic activity and blocks α1-adrenergic receptors, making it also an alpha-blocker. Labetalol's antagonistic actions on these receptors are competitive, potent, and reversible. Labetalol exhibits variable effects on α to β blockade depending on its route of administration. When taken orally, it has a ratio of α to β blockade at 1:3, whereas intravenous (IV) administration yields a ratio of 1:7. Therefore, labetalol is considered a beta blocker with some alpha-blocking properties. However, its beta-blocking potency is lower compared to propranolol, and it has weaker affinity for α-adrenergic receptors compared to phentolamine.

The dual antagonism of both α and β adrenergic receptors by labetalol results in different physiological effects in short-term and long-term scenarios. In acute situations, labetalol reduces blood pressure primarily by decreasing systemic vascular resistance, with minimal impact on heart rate, stroke volume, and cardiac output. In long-term usage, labetalol can reduce heart rate during exercise while maintaining cardiac output by increasing stroke volume. Labetalol's structural features include a substituted benzene ring containing a primary or secondary amine, which contributes to its antagonist activity. The molecule's size, particularly the presence of a 1-methyl-3-phenylpropyl substituted amine, primarily drives its antagonist properties. The overall structure of labetalol is highly polar, characterized by an aralkyl group, a carboxamide group in the meta position, and a hydroxyl group in the para position.

Labetalol exists as four stereoisomers, with only the (S,R)-isomer being pharmacologically active. The racemic mixture of labetalol, containing both active and inactive isomers, is commonly used to achieve its desired alpha and beta receptor blocking effects.

MATERIALS AND METHODS

The preformulation phase is critical in pharmaceutical development as it lays the foundation for successful drug formulation by identifying the physicochemical properties of the active pharmaceutical ingredient (API). In this study, preformulation work began with assessing the melting point of labetalol hydrochloride (LHCl) using the capillary method with an MR-VIS visual melting range apparatus (Lab India Analytical Instruments Pvt. Ltd., India). Determining the melting point helps in confirming the identity and purity of the drug substance. The solubility study of LHCl was performed across different solvents to identify suitable media for drug formulation. Saturated solutions were prepared by adding excess LHCl to 10 ml of each solvent, followed by sonication in a temperature-controlled water bath for 6 hours at 25 ± 0.5°C. After filtration through a 0.45 µm Whatman nylon membrane filter, drug concentrations were quantified using Reverse Phase-High Performance Liquid Chromatography (RP-HPLC), and results were averaged across three replicates (n=3) to ensure accuracy. This comprehensive solubility profiling allows the identification of optimal solvents for drug delivery systems.

The partition coefficient (PC) of LHCl was also evaluated to predict its lipophilicity and potential for transdermal absorption. The PC, defined as the ratio of the drug's concentration in n-octanol (organic phase) to that in water (aqueous phase), provides insight into the drug's distribution behavior in biological systems. A high PC generally indicates good membrane permeability, which is crucial for transdermal drug delivery. The PC was calculated using the equation:

$PC = Co / Cw,$

where *Co* represents the concentration in n-octanol and *Cw* is the concentration in water. Additionally, the apparent partition coefficient (APC) of LHCl was determined using phosphate buffer (pH 7.4) and n-octanol. A 20 µg/ml LHCl solution in buffer was mixed with an equal volume of n-octanol in a sealed tube, agitated at 37 ± 0.5°C for one hour, and left to equilibrate for 24 hours. Following phase separation, the drug concentration in the buffer was measured spectrophotometrically at 302 nm, and the concentration in the n-octanol phase was inferred. The APC was calculated as:

$APC = Co / Cb,$

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where *Co* is the drug concentration in n-octanol and *Cb* in buffer.

Advanced characterization techniques such as Attenuated Total Reflection-Fourier Transform Infrared (ATR-FTIR) spectroscopy and Differential Scanning Calorimetry (DSC) were employed to further evaluate LHCl. ATR-FTIR provides valuable insights into the molecular structure and potential interactions between LHCl and excipients by detecting functional groups via infrared absorption. This technique is especially useful for analyzing solid-state drug samples with minimal preparation. DSC analysis was used to examine thermal behaviors such as melting points and phase transitions, aiding in the assessment of drug purity and compatibility with formulation excipients. Lastly, RP-HPLC was again utilized for purity determination, a critical step to ensure consistent drug quality and effectiveness. The analytical method involved preparing a calibration curve using serial dilutions from 2 to 24 µg/ml, and UV absorbance was measured at λmax 302 nm, following Beer–Lambert's law with a correlation coefficient of 0.9981, indicating high linearity and accuracy.

Result And Discussion

PREFORMULATION STUDY

Description: -

Parameter	Specification	Result
Description	White crystalline powder	White crystalline powder

Melting point: -Melting Point of Labetalol HCl was found to be 195°C.

Solubility study: -

Parameter	1 Trial	2 Trial	3 Trial	Mean
Solubility(mg/ml)	20	19.5	20.5	20

Solubility in water at 25°C

A significant difference ($P < 0.05$) was noted when comparing the skin permeation rates between the distilled water solution and the isotonic phosphate buffer with a pH of 7.4. However, the inclusion of a co-solvent, specifically ethanol at a concentration of 95%, resulted in a slight increase in the permeability coefficient, which was found to be statistically insignificant ($P > 0.05$) according to the data presented in Table 2. This suggests that the addition of ethanol did not lead to a significant alteration in the skin permeation characteristics when compared to the isotonic phosphate buffer.

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Table:1:-The effect of various drug solutions on the skin permeability of LHCl

Study code	Drug solution	[+]Permeability coefficient $K_p \times 10^2$ (cm hour^{-1})	Enhancement factor (EF)
[+] PS1	Solution in distilled water	6.322 (± 0.013)	1.000
PS2	Solution in IPB pH 7.4	4.458[s] (± 0.015)	0.705
PS6	Solution in ethanolic IPB pH 7.4 (1:9)	3.285 (± 0.015)	1.00

Partition Coefficient: - Partition Coefficient of Labetalol HCl was found to be 7.08.

Attenuated total reflection Fourier transfer infrared analysis (ATR-FTIR)

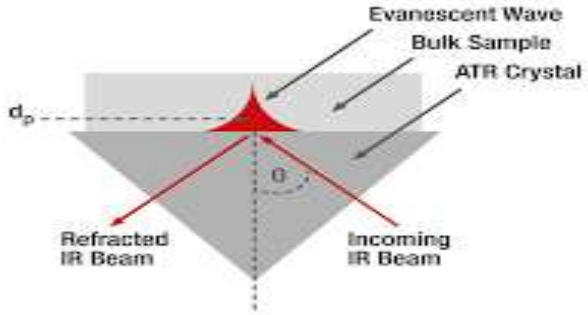

Figure:1 : Attenuated total reflection

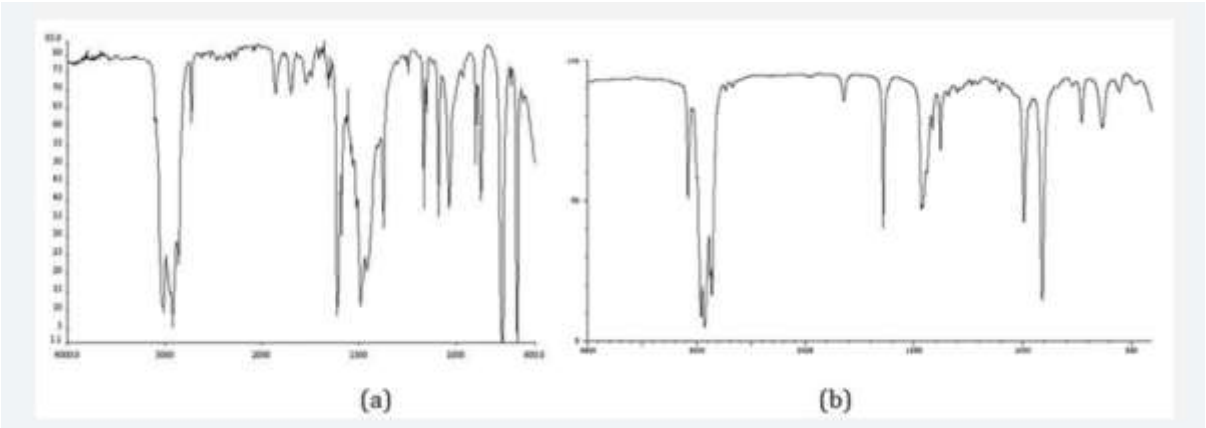

Figure 2 Fourier transfer infrared analysis

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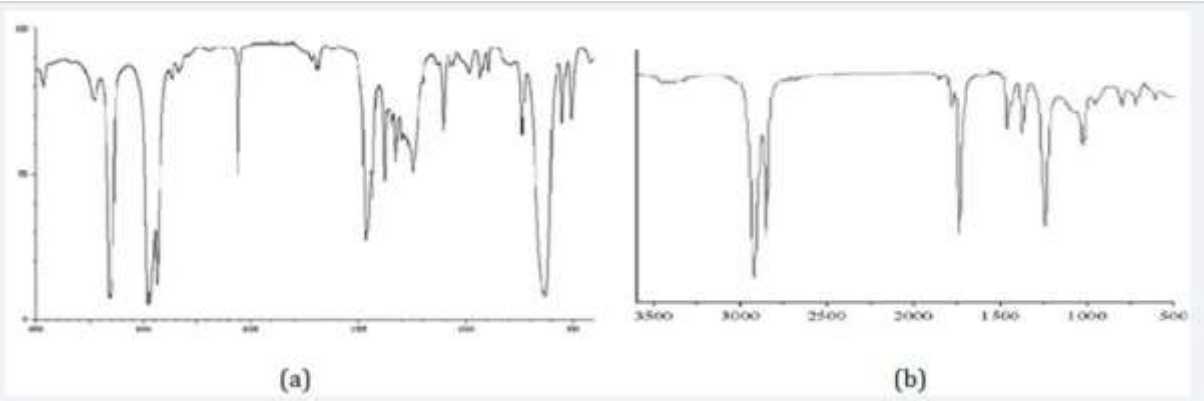

Figure:3: Attenuated total reflection Fourier transfer infrared analysis

.DSC Analysis

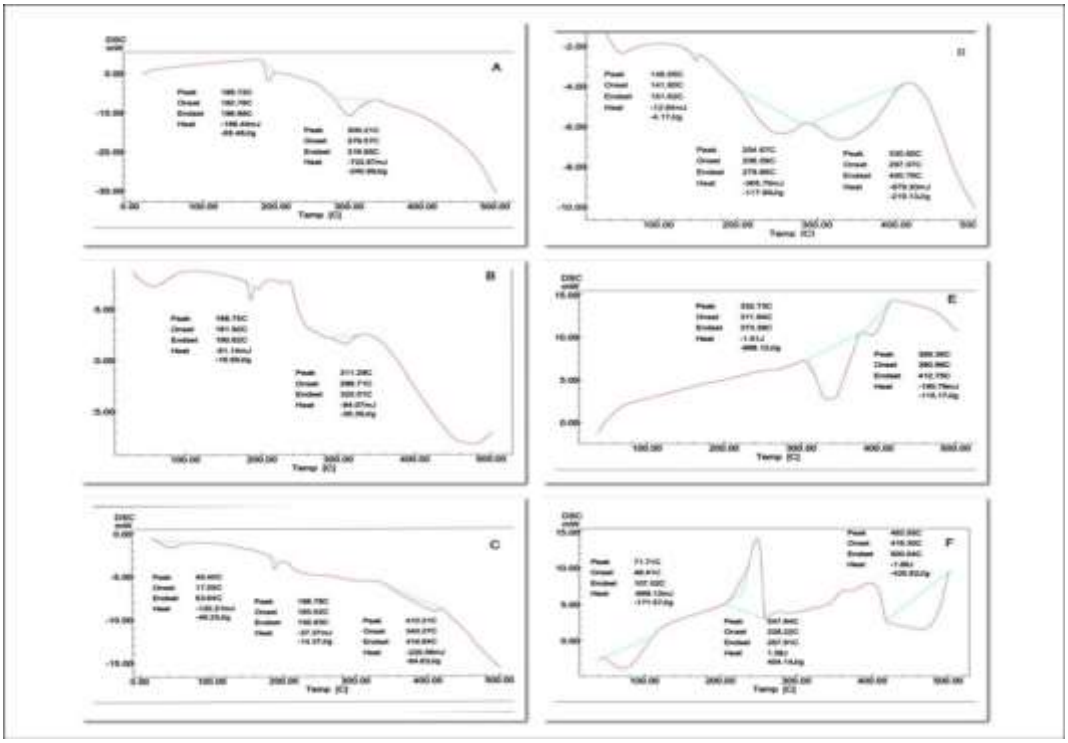

Figure:4 : DSC Analysis

PENETRATION ENHANCERS METHOD

In an effort to improve the permeation of Labetalol HCL (LHCl), various penetration enhancers were initially tested at a concentration of 5% v/v. These enhancers included turpentine oil, dimethyl formamide (DMF), menthol, dimethyl sulfoxide (DMSO), pine oil, and 2-pyrrolidone. The use of each of these enhancers resulted in an increase in the drug's permeability coefficient compared to their respective control groups.

To accommodate enhancers that were not soluble in the isotonic phosphate buffer (IPB) with a pH of 7.4, a co-solvent, specifically 95% ethanol, was combined with IPB in a 1:9 ratio. This

approach allowed the inclusion of DMSO, DMF, and 2-pyrrolidone as experimental substances, while distilled water served as the control. For enhancers such as turpentine oil, menthol, and pine oil, an ethanolic IPB with a pH of 7.4 was used as the control group. This methodology enabled the investigation of a diverse range of enhancers, ensuring that those not readily soluble in the buffer were still included in the experimental setup.

The effectiveness of various penetration enhancers was assessed by calculating their enhancement factor (EF). Among the enhancers tested, DMSO demonstrated the highest EF when compared to turpentine oil, DMF, pine oil, 2-pyrrolidone, and menthol (as indicated in Table 3). Notably, the application of 5% DMSO resulted in an approximately 17% increase in the permeability coefficient, establishing it as the most efficient penetration enhancer in this study.

DMSO's effectiveness in improving skin permeation can be attributed to several potential mechanisms. Firstly, it may bring about a reversible alteration in the protein structure within the stratum corneum, the skin's outermost layer, which in turn facilitates enhanced drug permeation. Additionally, DMSO can elevate the drug's thermodynamic activity, promoting its movement through the skin barrier. Furthermore, DMSO has the capability to induce swelling in the stratum corneum, leading to the creation of channels that reduce resistance to diffusion, ultimately facilitating greater drug permeation. Collectively, these mechanisms contribute to the significant enhancement in drug permeation observed when DMSO is used as a penetration enhancer in this study.

Table:2 :-Influence of various penetration enhancers on the permeability of LHCl

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Study code	Penetration enhancer	*Permeability coefficient $K_p \times 10^2$(cm hr^{-1})	Enhancement factor (E.F.)
PS1	Distilled water Control-1	6.322 (0.013)	1.00
PS6	Ethanolic IPB Control-2	6.490 (0.019)	1.00
PS7	Turpentine (5%)	7.450 (0.021)	1.15
PS4	DMSO (5%)	7.491 (0.032)	1.185
PS8	Pine oil (5%)	7.018 (0.036)	1.08
PS9	Menthol (5%)	7.183 (0.044)	1.11
PS5	2- Pyrrolidone (5%)	6.844 (0.052)	1.08
PS3	DMF (5%)	6.422 (0.071)	1.01

The table presents experimental data on the impact of different penetration enhancers on the transdermal delivery of an undisclosed substance. Each condition (represented by a study code) involves the use of a specific penetration enhancer at a 5% concentration. The permeability coefficient (Kp × 10^2 cm/hr^1) measures how effectively the substance can pass through the skin under each condition, and the enhancement factor (E.F.) quantifies the improvement in substance permeation compared to control conditions (PS1 and PS6).

Several penetration enhancers, including Turpentine, DMSO, Pine oil, and Menthol, exhibit higher permeability coefficients and enhancement factors compared to the control conditions. This suggests that these enhancers are effective in enhancing the transdermal delivery of the substance, potentially making them valuable components in the development of transdermal drug delivery systems.

In summary, this data underscores the significance of different penetration enhancers in facilitating the efficient delivery of substances through the skin. It provides valuable insights for researchers and formulators aiming to optimize transdermal drug delivery systems for improved therapeutic outcomes.

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SKIN PERMEATION EXPERIMENTS

The impact of various penetration enhancers on the permeability of Labetalol HCL (LHCl) was investigated. Additionally, the influence of an elevated drug concentration in the donor phase, ranging from 100 to 200 µg/ml, was studied in both distilled water and isotonic phosphate buffer at pH 7.4. In both scenarios, it was observed that an increase in drug concentration led to a corresponding increase in the permeability coefficient, as detailed in Table 4. Furthermore, another study explored the effect of varying the concentration of DMSO from 5% to 7.5% and up to 10%. This investigation revealed an augmentation in the permeability coefficient of the drug as the concentration of DMSO increased, as depicted in Table 5.

Table: 3 Effect of the quantity of drug concentration in donor cell on skin permeation of LHCl (n = 3)

Study code	Drug concentration (µg/ml)	[+] Permeability coefficient $K_p \times 10^2$ cm/hr[1]	Enhancement factor (E.F.)
PS1 (D/W*)	100	6.322 (± 0.013)	1.00
PS10 (D/W)	200	7.367 (±0.016)	1.16
PS2 (IPB)	100	4.458 (± 0.015)	1.00
PS11 (IPB)	200	5.537 (±0.011)	1.24

The table provides data from experiments investigating the transdermal delivery of Labetalol Hydrochloride (LHCl) using different conditions and LHCl concentrations. Permeability coefficients (Kp × 10^2 cm/hr^1) were measured to gauge LHCl's skin penetration, and enhancement factors (E.F.) were calculated to compare the effectiveness of various conditions and penetration enhancers. Two sets of conditions were examined: PS1 and PS10 (D/W*) with LHCl concentrations of 100 and 200 µg/ml, and PS2 and PS11 (IPB) with the same LHCl concentrations. The terms "D/W*" and "IPB" likely represent different formulations or vehicles employed for drug delivery. The findings indicate that, for both LHCl concentrations, PS10 and PS11 generally exhibited higher permeability coefficients and enhancement factors in comparison to PS1 and PS2. This suggests that the conditions associated with PS10 and PS11,

combined with the use of higher LHCl concentrations, were more effective in facilitating the permeation of LHCl through the skin.

Table:4 Influence of concentration of penetration enhancer on the skin permeability of LHCl

Study code	Concentration of enhancer (%)	*Permeability coefficient K_p × 10^2 cm/hr[1]	Enhancement factor. (E.F.)
PS4	5	7.491 (0.032)	1.00
PS12	7.5	8.010 (0.045)	1.07
PS13	10	9.292 (0.052)	1.240

The table presents data from experiments examining the effect of different concentrations of a penetration enhancer on drug permeation through the skin. The penetration enhancer is represented by varying concentrations in different conditions: PS4 with 5% enhancer, PS12 with 7.5% enhancer, and PS13 with 10% enhancer.

The permeability coefficient (Kp × 10^2 cm/hr^1) measures the rate at which the drug can penetrate the skin under each condition. The enhancement factor (E.F.) is calculated to assess how much the permeability of the drug is improved in comparison to a control condition, which in this case is PS4 (without the enhancer).

The results indicate that as the concentration of the penetration enhancer increases, both the permeability coefficient and the enhancement factor generally rise. This suggests that higher concentrations of the enhancer enhance the drug's ability to permeate through the skin. In particular, PS13 with a 10% enhancer concentration exhibited the highest permeability coefficient and enhancement factor, indicating the most significant improvement in drug permeation. These findings emphasize the importance of the selected concentration of penetration enhancers in transdermal drug delivery systems. Increasing the enhancer concentration appears to enhance the efficiency of drug delivery through the skin, which can have implications for designing more effective and efficient transdermal drug delivery formulations.

Conclusion

The present study successfully evaluated the transdermal delivery potential of labetalol hydrochloride and emphasized the critical role of chemical penetration enhancers in enhancing its permeation through the skin. Labetalol hydrochloride, with its known limitations in oral bioavailability due to first-pass metabolism, demonstrated significant promise for transdermal administration as a means of achieving sustained therapeutic levels and improved patient compliance. The use of excised rat skin in Franz diffusion cells provided a reliable in vitro model to study drug permeation, and the incorporation of enhancers such as ethanol, propylene glycol, oleic acid, and dimethyl sulfoxide (DMSO) markedly improved the drug's flux across the skin barrier. Among these, oleic acid and DMSO showed superior enhancement effects, likely due to their ability to disrupt the stratum corneum lipid structure, facilitating increased drug diffusion. Preformulation studies, including solubility profiling, partition coefficient measurement, ATR-FTIR, DSC, and RP-HPLC analysis, confirmed the physicochemical suitability of labetalol hydrochloride for transdermal application. The comprehensive data generated from this research support the feasibility of formulating an effective transdermal drug delivery system for labetalol hydrochloride. These findings pave the way for further in vivo studies and formulation optimization, potentially offering a non-invasive, controlled-release therapy for hypertensive patients that reduces dosing frequency and minimizes systemic side effects commonly associated with oral administration.

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