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Original Article

POTENTIAL HERB-DRUG INTERACTION OF *DECALEPIS HAMILTONII VIA* P-GP MEDIATED PHARMACOKINETIC INTERACTION WITH FEXOFENADINE IN RATS: AN *IN SITU* AND *IN VIVO* STUDY

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ABSTRACT

Objective: The objective of this study was to investigate the influence of *Decalepis hamiltonii* (*D. hamiltonii*), a traditional plant used in herbal medicine, on the intestinal absorption and pharmacokinetics of fexofenadine, a substrate of P-glycoprotein (P-gp), in rats.

Methods: *In situ* intestinal perfusion tests were conducted to assess the intestinal permeability of fexofenadine. P-gp ATPase activity was also evaluated to understand the modulatory effects of *D. hamiltonii* on P-gp. An *in vivo* pharmacokinetic investigation was performed by administering oral fexofenadine to rats.

Results: The *in situ* study results revealed that the effective permeation (Peff) of fexofenadine was significantly diminished ($p<0.001$***) in aqueous extract of *D. hamiltonii* (AREDH, 200 mg/kg p. o.) pretreated group compared to normal control indicating modulation in absorption. Further, there was significant augmentation ($p<0.01$**) of P-gp ATPase activity in AREDH pretreated group (200 mg/kg p. o.) compared normal control indicating P-gp inductive potential of *D. hamiltonii*. Pharmacokinetic study results revealed that the peak plasma concentration (Cmax) and the area under the concentration-time curve (AUC) of fexofenadine was significantly downregulated ($p<0.001$***) in AREDH pretreated group (200 mg/kg p. o.) compared to the normal control group indicating the compromised absorption and bioavailability. However, no significant changes were observed in fexofenadine half-life (T1/2 k10), time to reach peak plasma concentration (Tmax), or elimination rate constant (k10).

Conclusion: In conclusion, *D. hamiltonii* significantly reduced the oral bioavailability of fexofenadine by promoting P-gp-mediated drug efflux during intestinal absorption. This suggests that the modulatory characteristics of *D. hamiltonii* may lead to herb-drug interactions when taken in combination with xenobiotics, emphasizing the importance of considering such interactions in clinical practice and further research.

Keywords: *Decalepis hamiltonii,* Fexofenadine, P-glycoprotein, Intestinal permeability, Pharmacokinetics

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INTRODUCTION

Traditional knowledge of several native local herbs in India led to the preparation of several herbal health drinks, which are only limited to the location from which they originated. Nannari sharbat is one such ancient herbal beverage made from the roots of the herb *Decalepis hamiltonii,* which is native to the southern Indian states of Andhra Pradesh, Karnataka, Kerala, and Tamil Nadu. In the summer, beverages are used to satisfy thirst and function as a hepatoprotective agent, which is beneficial to stomach health. The herb is known as Ananthamula in Ayurveda. It belongs to the family Apocynaceae. The roots have high antioxidant activity and are used in the preparation of pickles [1, 2]. Traditional uses of *D. hamiltonii* root include blood cleansing, the management of intrinsic haemorrhage, fever, Kushtha, wound healing, bronchial asthma, erysipelas and food poisoning. Moreover, numerous pharmacological effects of *D. hamiltonii* have been reported, including antidiabetic activity, hypolipidemic, antioxidant activity, and antimicrobial activity against food-borne pathogens [3, 4]. Secondary metabolites are abundant in *D. hamiltonii* tuberous roots, particularly 2-hydroxy-4-methoxybenzaldehyde (HMB) [5]. HMB is a volatile compound that is an isomer of vanillin, the main flavour compound found in the roots of *D. hamiltonii*, and it has abundant medicinal properties. Moreover, aqueous and methanolic root extracts were identified to possess additional phenolic acids, including caffeic acid, cinnamic acid, syringic acid, ferulic acid, protocatechuic acid, p-coumaric acid, gallic acid, gentisic acid, ellagic acid and 4-hydroxy isophthalic acid [6, 7]. Furthermore, the volatile oils in *D. hamiltonii* tuberous roots include 0.17% benzaldehyde, 0.018% salicylaldehyde, 0.081% 2-phenyl ethyl alcohol, 0.044% methyl salicylate, 0.01% p-anisaldehyde, 0.016% benzyl alcohol, 0.45% vanillin and 0.038% ethyl salicylate [3]. Though it has not been proven that *D. hamiltonii* is toxic to humans, various studies show that micromolar concentrations of phenolic chemical constituents such as 2-Hydroxy Methyl Benzoic Acid and vanillin are toxic to *Haplochelius panchax*, a freshwater fish.

Moreover, research suggests that most of these phenolic acids isolated from plant resources have the potential to interfere with the pharmacokinetics and pharmacodynamics of several xenobiotics, resulting in herb-drug interactions via modulating P-gp function [8]. P-gp (170-kDa) is a protein associated with plasma membrane belonging to the ABC protein family and is present in the blood-brain barrier, liver, kidney, and small intestine of humans and rodents [9]. P-gp, like several other ABC proteins, has a diverse substrate range that includes xenobiotics, hazardous metabolic by-products, and several chemotherapeutic agents used in the treatment of various illnesses (e. g., reverse transcriptase inhibitors, antibiotics, narcotics, antineoplastics, and antidepressant drugs) [10]. P-gp, found on the villus tip of gut enterocytes' apical brush border membrane, actively promotes substrate efflux from gut epithelial cells back into the intestinal lumen. This phenomenon results in modifications in the absorption of several drugs that act as P-gp substrates [11]. Additionally, P-gp also confers resistance by preventing sufficient accumulation of drugs in the cells, which causes failure in the cytotoxic activity of certain anticancer medications like tamoxifen [12]. All these functions of P-gp attribute their importance to the fate of drugs. Fexofenadine, a non-sedating antihistamine that is used to treat seasonal allergic rhinitis, is one of the known substrates of P-gp [13]. Fexofenadine has a low oral bioavailability (approximately 33%) in humans due to P-gp-driven drug efflux and might be used as a useful probe substrate drug to examine the significance of P-gp-mediated drug interactions [14]. On the contrary, phenolic acids isolated from *D. hamiltonii,* like caffeic

acid, vanillin, syringic acid, and ferulic acid, are demonstrated as potential P-gp inhibitors by various studies and are also reported for their ability to attenuate multidrug resistance in various chemotherapy therapies [8, 15-17]. Further, *D. hamiltonii* is also reported to possess modulating properties towards cytochrome P450 enzymes [18] and human organic anion transporter 1, which result in substantial herb-drug interactions when ingested with xenobiotics. Drug resistance mediated through P-gp is one of the major culprits responsible for chemotherapy failure, and these phenolic acids have been shown to reverse this problem of resistance when administered alongside treatment [15-19]. Though *D.* Hamiltonii is an important ingredient in the most famous herbal drink of peninsular India, its possible herb-drug interactions are yet to be effectively explored. In the present research, we sought to explore the modulatory property of *D. Hamiltonii* on the activity of P-gp protein and pharmacokinetic parameters of fexofenadine.

MATERIALS AND METHODS

Experimental chemicals

Fexofenadine hydrochloride was acquired from TCI Chemicals (India) Pvt. Ltd. HPLC-grade solvents were employed for quantifiable analysis (Merck, India), and all other reagents and chemicals used in the investigation were of analytical quality. Chemicals such as sodium bicarbonate, sodium chloride, potassium chloride, magnesium sulfate, sodium dihydrogen orthophosphate, calcium chloride, di sodium ethylene diamine tetra acetic acid, D-glucose, potassium dihydrogen phosphate, sodium hydroxide, zinc acetate, sodium azide, sodium orthovanadate, magnesium, ATP, EGTA, disodium hydrogen phosphate, potassium.

Preparation of the plant extracts

D. hamiltonii roots were purchased from a merchant in Kurnool town and gathered in December from Nallamalla woodland, Nandyal district, and confirmed by Dr K. Madhava Chetty, Plant Taxonomist, SV University, Tirupati, with voucher number 0549. The obtained roots were ground into a coarse powder, mixed with nine parts demineralized water at 500 °C, steeped overnight, and filtered the next day. The resulting filtrate was recognised as an aqueous root extract of *D. hamiltonii*. Weight per millilitre was calculated using loss on drying.

Experimental animals

Male albino rats of the strain Wistar, weighing 150-200 gm were chosen. Raghavendhra Enterprises Pvt. Ltd. in Bangalore, India, provided the animals. The animals were kept in a conventional environmental laboratory setting and provided laboratory meals and water. Before the trial, all of the animals were adapted to housing conditions for a week. The Sri Padmavathi School of Pharmacy's institutional animal ethics committee (IAEC) authorised all operations (approval number: SPSP/1016/PO/Re/S/06/CPCSEA/2022/02).

In situ intestinal perfusion study

The rats were randomized into five groups (n = 6), with the first receiving distilled water and the second receiving Rifampicin p. o. (inducer 13.5 mg/kg), the third and fourth receiving AREDH p. o. (Test-1 200 mg/kg) and AREDH in the chamber (Test-2 1 mg/ml), and the fifth receiving ketoconazole in the chamber (1 g/ml) for 14 d. After completion of the treatment, on the 15th day, the invasive technique of the *in-situ* rat gut technique was executed, rendering to the formerly reported methods [20-23]. Xylazine (10 mg/kg) and Ketamine (100 mg/kg) were used to induce anaesthesia in rats via intramuscular injection, and then the small intestine was uncovered by a midline incision. The jejunum was externalised for about 15 cm, for enabling accessibility to the lumen at both the proximal and distal ends of the jejunum section. After that, the intestinal contents were washed out with warm normal saline before being cannulated with a Teflon tube. Perfusate was pumped using a peristaltic pump through the lumen of the inlet cannula at 0.6 ml/min from an encased reservoir that maintained the perfusate's temperature at 37 °C. The perfusate reservoir initially contained PBS buffer pH-7.2 with glucose maintained at 37 °C without drying. The exposed jejunum section was wrapped with saline-soaked cloth gauze and a transparent plastic sheet. the body temperature was kept stable with the use of a heating light. After the entire process had stabilised for 30 min, the perfusate that was leaving the jejunal section was dumped to waste. After completion of stabilization, the perfusate reservoir was replaced with a precise volume of 200 ml of a 10 µM fexofenadine solution. An initial 1 ml sample was taken from the reservoir before the instigation of perfusion through the jejunal segment. The outlet cannula was directed to collect the perfusate for 15 min after 45 seconds of perfusion [20, 22-23].

Q_{OUT} was calculated by weighing the collected perfusate from the outlet in the first 15 min.

$$Q_{IN} = \text{Measure flow entirely to the intestine}$$

(Weight of 9 ml perfusate from inlet) $(0.6\ \text{ml/min} = 0.6\text{ml} \times 15 = 9\text{ml})$

C_{OUT} is the concentration of the drug after 15 min (1 ml sample collected immediately after first 15 min)

$$C_{COR} = \text{concetration} \times Q_{OUT}/Q_{IN}$$

Immediately after 15 min, a 1 ml sample was collected to find Cout (concentration of drug); after 15 min, Cout was used to calculate Ccor after 15 min.

Calculations

Estimation of water flux correction factor by gravimetric method

$$C_{COR} = C_{OUT} \times Q_{OUT}/Q_{IN}$$

Estimation of effective permeability:

$$\text{Peff} = \frac{\left[Q_{IN} \times \ln\left(\frac{C_{COR}}{C_{IN}}\right)\right]}{2\pi RL}$$

All plasma samples (1 ml) and *in situ* perfusions were extracted with acetonitrile (10 ml) using a simple protein precipitation procedure. Before being centrifuged for 10 min at 10,000 rpm, the samples were vortexed for 1 min. The produced clean supernatant (20 µl) was injected and assessed using the HPLC method. The assay range was 0.01-10 µg/ml, while the detection limit was 0.01 µg/ml.

P-gp ATPase activity assessment

The rats' small intestines were detached, and the length was measured. The cells of the intestine were scraped with the help of the slide and homogenised with Tris-HCL buffer (0.1 M: pH 7.4) in a chilled milieu using a Teflon homogenizer. The obtained homogenate was subjected to centrifugation at 1000 rpm at 40 °C for 15 min, and the isolated supernatant was utilised for the estimation of inorganic phosphate. The amount of total protein present in the homogenate was calculated, and it was diluted to 1 mg/ml with tris-HCL. 40 µl of this solution was taken from the sample, and 20 µl of assay buffer was added. 40 µl of the sample's solution was transferred to another microcentrifuge tube, and 20 µl of buffer solution comprising sodium orthovanadate was added. The reaction was started by adding 10 µl of 0.5 mmol Mg-ATP, and the reaction mixture was incubated at 37 °C for one hour. 20 µl of the sample was taken to determine inorganic phosphate after the reaction was ceased by adding 30 µl of 10% sodium dodecyl sulphate (SDS) after the complete incubation period. P-gp activity was measured in mg/dl and expressed in nanomoles/mg/h.

In vivo pharmacokinetic studies

Rats were randomised into two groups (n = 6): one that received normal saline for seven days and another that received AREDH (200 mg/kg) for seven days. Rats were deprived of food overnight, roughly 3 h after the last oral dosing. On the 7th day, fexofenadine (10 mg/kg; p. o) was given to both the control and treated groups. Under ether anaesthesia, rats' blood samples (0.25 ml) were drawn from the retro-orbital plexus into heparin-containing micro-centrifuged tubes. Before the dose, as well as 0.25, 0.5, 1, 2, 4, 6, 8, and 12 h later, samples were taken. The tubes were centrifuged at 8000 rpm at 25 °C for 20 min to obtain plasma, and they were stored at-80 °C until HPLC analysis of the samples.

HPLC analysis of fexofenadine

Shimadzu HPLC system with SPD 20A UV visible detector, Dual Pump System, and RP C18 column (Phenomenex Luna, 250 mm x 4.6

mm ID, particle size 5 m) was used to examine the samples. Elution was detected at 195 nm at a flow rate of 1.0 ml/min using methanol and 6.8 g of monobasic potassium phosphate in 1000 ml of water as mobile phase, 35:65 (v/v), adjusted to pH 7.4 [14].

Statistical analysis

The standard deviation (SD) of each mean value is shown along with it (mean±SD). With a significance level of $p<0.05$, statistical analysis was carried out using the student's unpaired t-test and analysis of variance (ANOVA) in version 9.0 GraphPad Prism (GraphPad Software Inc., San Diego, CA, USA).

RESULTS

Effect of AREDH on intestinal permeability of fexofenadine

The modulatory property of AREDH on intestinal permeability of fexofenadine was assessed through an in-*situ* technique. The results of the in-situ rat gut technique involving AREDH in two dosage administrations along with a P-gp inducer (rifampicin) and inhibitor (ketoconazole) are demonstrated in fig. 1. According to the graphical representation, it reveals that the effective intestinal permeability (P_{eff}) of fexofenadine was effectively diminished by inducer and AREDH when given orally at 200 mg/kg when compared to the normal control. Further, there was a significant augmentation in P_{eff} of fexofenadine in animals administered with an inhibitor. On the contrary, when AREDH was given through perfusate at 1 mg/kg, it failed to exhibit any alterations in P_{eff} fexofenadine when compared to the normal control group.

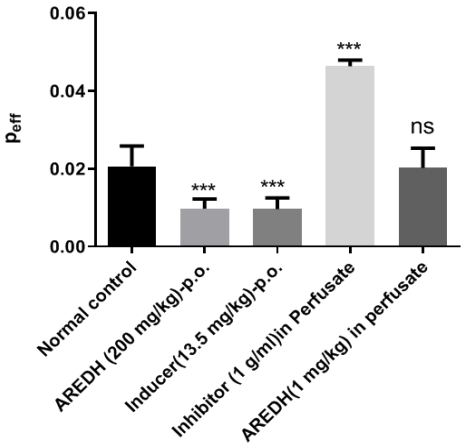

Fig. 1: Effect of AREDH on intestinal permeability of fexofenadine, all the values were expressed as mean±SEM n=6. *p<0.001, ***statistically significant difference in the AREDH group when compared to normal control. *p<0.001, ***statistically significant difference in inducer group when compared to normal control. *p<0.001, ***statistically significant difference in the inhibitor group when compared to normal control. ns no significant difference in AREDH in perfusate when compared to normal control

Effect of AREDH on P-gp ATPase activity of fexofenadine

The measurement of P-gp ATPase activity was measured as the amount of inorganic phosphate produced. Fig. 2 depicts the outcomes of AREDH's P-gp ATPase modulatory activity, as well as the inducer and inhibitor. The graphical representation indicates that administration of AREDH (200 mg/kg; p. o.) and the inducer resulted in significant upregulation of P-gp ATPase activity when compared to normal control. While AREDH administered via perfusate was ineffective in inducing P-gp ATPase activity, the inhibitor group presented a significant diminution in the activity of P-gp ATPase compared to the normal control group.

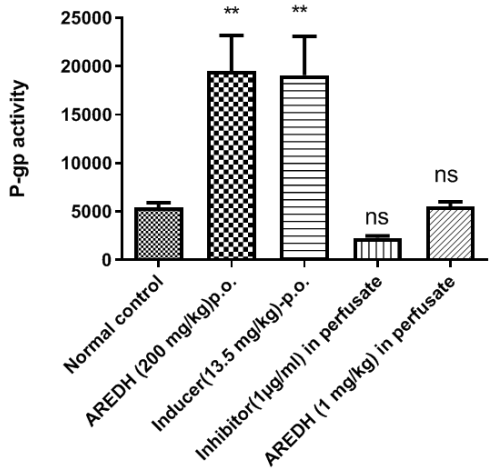

Fig. 2: Estimation of P-gp activity in different groups, all the values were expressed as mean±SEM n=6 *p<0.01, ** statistically significant difference in the AREDH group when compared to normal control. *p<0.01, ** statistically significant difference in inducer when compared to normal control. ns no significant difference in inhibitor and AREDH in perfusate when compared to normal control

Effect of AREDH on pharmacokinetic parameters of fexofenadine

Fig. 3 depicts the plasma concentration-time plots of fexofenadine following fexofenadine (10 mg/kg; p. o) administration in control and pre-treated groups (AREDH 200 mg/kg). While table 1 summarises the mean values of parameters associated with the pharmacokinetics of fexofenadine in both the absence and presence of pre-treatment with AREDH, the oral pharmacokinetics of fexofenadine were shown to be substantially changed by AREDH (200 mg/kg) pre-treatment for 7 d when related to the control group (fexofenadine alone). When fexofenadine was pre-treated with AREDH, the Cmax, AUC_{0-t}, and $AUC_{0-\infty}$ of fexofenadine decreased significantly when compared to the control group. Furthermore, the CL and Vd of fexofenadine were higher in the pre-treatment group compared to the control group, whereas there was no significant difference in Tmax, T1/2, and K10.

Table 1: Effect of AREDH on pharmacokinetic parameters of fexofenadine

Parameter	Mean values±standard error (n=6) (control)	Mean values±standard error (n=6) (test)
Ka (h)	1.321±0.021	1.121±0.028
k10/h	0.229±0.003	0.231±0.005
t1/2 ka (h)	0.524±0.008	0.620±0.016
t1/2 k10 (h)	3.024±0.034	3.002±0.064
Vd (ml/kg)	2.291±0.040	2.711±0.044***
CL (ml/kg/min)	0.525±0.004	0.626±0.006
Tmax (h)	1.604±0.010	1.775±0.017
Cmax (µg/ml)	3.024±0.035	2.447±0.021***
AUC 0-t (µg/ml/min)	17.58±0.171	14.71±0.108***
AUC 0-inf (µg/ml/min)	19.05±0.162	15.97±0.154***

Significant deviation from the control group ($p<0.001$).

Mean data of Pharmacokinetic Parameters (k10 = Elimination rate constant, ka = Absorption rate constant, t1/2 k10 = Elimination half-life, t1/2 ka = Absorption half-life, CL = Clearance, Vd = Volume of distribution, Cmax = Maximum Concentration in the Plasma, Tmax = Time taken to reach Maximum Concentration, AUC 0-inf = Area under the curve from 0 to infinity, AUC 0-t = Area under the curve from 0 to time t) of Fexofenadine and their standard error was calculated when administered via oral route in control and test groups.

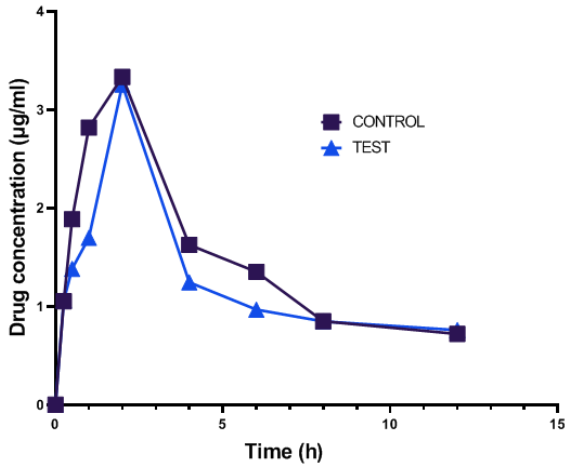

Fig. 3: Fexofenadine (10 mg/kg) plasma drug concentration-time plots in wistar rats. The test group received both AREDH and fexofenadine for 7 d, whereas the control group only received fexofenadine. The data are shown as mean±SD (n = 6)

DISCUSSION

In a country like India, the general populace extensively utilises several herbal products on their own or in conjunction with prescription and non-prescription treatments, thinking that they are safe since they are derived from natural rather than synthetic sources. Certain herbal products are consumed daily, such as basil, blueberries, garlic, and ginger, and are used as ingredients in cooking. Nannari sharbat is one such ancient herbal beverage made from the roots of the herb *D. hamiltonii,* which is widely consumed in southern India. In the summer, beverages are used to satisfy thirst, and they also function as hepatoprotective agents, which are beneficial to stomach health. Herbs, like any other synthetic medication, have a dose-dependent impact. A precise dose of an herbal substance may have a medicinal effect, but going above or below the amount may have negative consequences. Though most herbs are not used for life-threatening disorders, they may be used intentionally or unintentionally by the general population in conjunction with synthetic pharmaceuticals to enhance the bioavailability, minimise toxicity, or activate other functions through synergistic action. Herbal medications, like synthetic medicines, interact with pharmaceuticals in two ways: pharmacokinetically and pharmacodynamically. Pharmacokinetic interactions affect medication or herbal medicine absorption, distribution, metabolism, or excretion. These interactions impact pharmacological action by either increasing or decreasing the free drug accessible to have a therapeutic effect. Pharmacodynamic interactions have a qualitative influence on a drug's activity, either by increasing (synergistic or additive activities) or antagonising it. Before oral drugs get to the capillaries that lead to the portal vein, they have to navigate through the mucosal layer of the gut wall. The multidrug resistance-associated protein family (MRP; ABCC), MDR1 drug-transporting p-glycoprotein (P-gp; ABCB1), oligopeptide transporters (PEPT; SLC15A), organic anion transporters (OAT; SLC22A) and breast cancer resistance protein (BCRP; ABCG2) are a few of the drug transporters that have been identified in this mucosal barrier. However, only a small number of these are known to be involved in intestinal drug absorption. The organic anion peptide transporter (OATP), organic anion transporter (OAT), and permeability-glycoprotein (P-gp) transporters have all been reported to interact with a variety of phenolic acids, which has led to their involvement in drug-herb interactions.

D. hamiltonii roots are used in many ayurvedic remedies and have been traditionally consumed by tribal people as pickles and to preserve food grains (as a bio-insecticide). The main ingredient in the famous herbal drink Nannari is the aqueous root extract of *D. hamiltonii*. It has been reported that *D. hamiltonii* contains several phenolic acids like caffeic acid, vanillin, syringic acid, and ferulic acid, which have been demonstrated as potential P-gp modulators by various research studies. Due to their modulating property, the goal is to investigate the interaction of P-gp substrates (many prescription and non-prescription drugs) and modulators (phenolic compounds of AREDH) to recognize the possible consequences in pharmacokinetics and to predict the clinical outcome. Therefore, in the present study, the drug fexofenadine has been used as a presumed substrate of P-gp to investigate the herb-drug interactions of AREDH due to its possible modulatory effect on the ABC protein P-gp, which predominantly orchestrates the pharmacokinetic fate of fexofenadine. Previous research evidenced that there is a significant expression of P-gp (mdr 1a) mRNA from the duodenum to the ileum, implying that P-gp plays a crucial role in the transport of fexofenadine from the intestine [24]. Additionally, research shows that the rat model for permeation investigations is suitable for human prediction since MDR1 in rats expresses P-gp with a similar level of affinity and expression [25]. Furthermore, it has been demonstrated that oral xenobiotic absorption in rats and humans is similar [26]. Thus, rat gut permeability studies would be more useful for forecasting *in vivo* P-gp substrate absorption. Studies suggested computing the passive permeability of actively transported molecules using a non-everted gut sac model [27, 28]. As a result, in the current study, an i*n-situ* perfusion assessment was performed to investigate the role of P-gp in fexofenadine intestinal transport [29].

In situ perfusion assessment in the rat jejunum was executed to confirm the role of P-gp activity modulation in fexofenadine intestinal transport. One of the key biopharmaceutical processes used to measure the rate and degree of gastrointestinal (GI) medication absorption is intestinal Peff, which was estimated through this *in situ* study in rats that were pre-treated with AREDH along with an inducer and inhibitor of P-gp for comparison. The effective permeability of fexofenadine was considerably downregulated in the AREDH (200 mg/kg) and inducer (rifampicin) (13.5 mg/kg) pre-treated groups when compared to the normal control group. This was also supported by the results of the P-gp ATPase activity assessment. AREDH 200 mg/kg, like the inducer (rifampicin), increased P-gp ATPase activity compared to normal controls, indicating a direct relationship to altered Peff of fexofenadine in pre-treated groups. On the contrary, when AREDH was given through perfusate, Peff fexofenadine showed no significant modulation and was comparable to normal control. Thus, the decrease in fexofenadine intestinal permeability observed in AREDH 200 mg/kg pre-treated rats is attributable to AREDH-induced P-gp ATPase activity. These findings also revealed how fexofenadine was effluxed back to the gut via P-gp. Furthermore, in groups where AREDH (Test-2) and an inhibitor (ketoconazole) are given through perfusate, the permeability values of the AREDH group couldn't display any statistically significant difference when compared to the control. This effect was in direct relation to P-gp ATPase activity modulation in the AREDH (Test-2) group. The activity of P-gp ATPase was like that of the normal control group. However, the inhibitor group exhibited significantly augmented PEF of fexofenadine when compared to the normal control, which slightly correlated with its respective P-gp ATPase activity modulation, albeit the downregulation of the activity was not statistically significant. There, it indicates that AREDH possessed substantial inducing properties towards P-gp ATPase rather than inhibition, which resulted in altered absorption of fexofenadine *in vitro* and *in situ* studies.

To further comprehend the impact of AREDH on the pharmacokinetic characteristics of fexofenadine, an *in vivo* pharmacokinetic study of fexofenadine was carried out in rats pre-treated with AREDH for 14 d. Though the *in vitro* and *in situ* studies

proposed the probable influence of AREDH pre-treatment on the transport and permeation of fexofenadine, there was no significant change in the pharmacokinetics of fexofenadine except for a few parameters. AUC and Cmax were suggestively repressed in the AREDH pre-treated group when compared to the control group, likely indicating diminished absorption of fexofenadine due to AREDH pre-treatment via induction of P-gp ATPase. The absorption half-life was also slightly repressed. It also indicates that the systemic exposure to fexofenadine was also slightly restricted with AREDH pre-treatment. Furthermore, the CL and Vd of fexofenadine were slightly higher in the AREDH pre-treated group compared to the control group, whereas parameters like Ka, k10, and T1/2 k10 were almost similar in both groups. Furthermore, these findings show that AREDH pre-treatment had a limited effect on hepatic drug elimination as well as an effect on absorption.

Finally, herbal products contain several active ingredients with varied or comparable pharmacological effects. Previous studies indicate that the occurrence of phenolic acids and various other chemical constituents in herbs are capable of binding to nuclear receptors like the constitutive androstane receptor (CAR), the vitamin D-binding receptor (VDR) and pregnancy and xenobiotic receptor (PXR), which influence the expression of enzymes associated with drug metabolism and drug transporters [30]. Any of these receptors can be activated or inhibited by the binding of an herbal constituent as a ligand. This increases or decreases the metabolism or transport of the co-administered conventional drug(s), which can result in diminished therapeutic efficacy or augmented toxicity of the drugs. The present study suggests that the chemical constituents of *D. hamiltonii* modulating the receptors responsible for influencing and inducing P-gp ATPase activity is the possible mechanism for alterations in the pharmacokinetic properties of fexofenadine, indicating a potential herb-drug interaction. To assess the likelihood that these products will interfere when used along with prescription medication, patients should be questioned about any usage of herbal remedies or natural product(s) and other medications.

CONCLUSION

In essence, the current investigation demonstrates that *D. hamiltonii* is a possible P-gp inducer, which may result in medication ineffectiveness, resistance, or even tolerance development. As a result, caution should be exercised while co-administering *D. hamiltonii* medicines that are efficiently transported by P-gp. It is highly advised that herbal remedies be assessed, monitored, and tested when taken in conjunction with other pharmaceuticals to avoid potential interactions. Further studies are required to isolate specific constituents responsible for P-gp induction and elucidate the underlying mechanism orchestrating this interaction. A comprehensive approach and understanding will help combat undesirable clinical difficulties.

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AUTHORS CONTRIBUTIONS

All the authors have contributed equally.

CONFLICT OF INTERESTS

The authors declare no conflict of interest

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