All of the conventional anti-Alzheimer’s medicines are having side effects such as nausea, vomiting, diarrhea. The major problem associated with current anti-Alzheimer’s drugs is gastrointestinal side effects encountered with them. For example Tacrin (one of the most commonly used conventional AD medicines) has shown 40% liver toxicity, hormonal agents have shown prostate and breast cancer, acne, hair loss, hepatic dysfunction and insulin resistance, Huperzine A (one of the most famous herbal anti-Alzheimer’s) has shown nausea, vomiting, diarrhea, sweating, blurred vision and increased of urination, increasing its dose causes broncho spasm.

Oral therapy of vinpocetine causes some side effects including stomach pain, nausea, sleep disturbances, headache, dizziness, nervousness, and flushing of the face, and lastly Memantine which causes constipation on oral administration. Therefore there is a need for a delivery system of these drugs with enhanced GI tolerability, which retaining their efficiency.

The above side effects of anti-Alzheimer’s drugs can be minimized by TDD as it avoids first pass metabolism, improving the short half-life and low oral bioavailability of anti-Alzheimer’s drugs and the other good advantage of transdermal route specifically for Alzheimer’s patients is patient compliance which reduces the chances of missing doses by them as transdermal patches will be used once in two days with no need of further dosing for next 48 hours. Thus it is expected that transdermal delivery of anti-Alzheimer’s drugs through transdermal ethosomal and transfersomal formulations will result in the release of drug at a controlled and suitable rate to maintain appropriate plasma drug level for the therapeutic efficacy by using skin route.

The present study was an attempt to design, develop and evaluate the appropriate transdermal dosage forms (ethosome and transfersome) for enhancement of skin permeation, half-life and bioavailability of vinpocetine (VIN) as Anti-Alzheimer’s drug.

This study was followed up with the twin objectives of investigating the potential of ethosome and transfersome vesicular systems, as drug carriers vis-à-vis other vesicular and conventional modes of drug administration and to explore the practicability of transdermal delivery of vinpocetine by developing transdermal vesicular patch formulations of VIN.
The study was conducted by preparing two nanovesicular formulations of vinpocetine named ethosome and transfersomes, using solvent evaporation method applying factorial design using Box-Behnken method for optimization of drug loaded novel vesicular drug delivery systems.

Further, to select the optimized vesicles, the prepared novel vesicular systems were characterized for various parameters such as higher % entrapment efficiency, elasticity and lower particle size. The optimized vesicular drug delivery systems were then converted into gel formulation by adding carbopol 940 as a gelling agent which acts as rate controlling membrane too. Later on the best vesicular gel formulation was selected according to higher flux resulted from in vitro release studies. The final optimized vesicular gel formulations were then converted into patch and used for pharmacodynamic and pharmacokinetic studies.

Following are the results obtained and conclusions drawn from the lab experiments:

- On the basis of physical characterization and identification tests, it was concluded that the drug sample of Vinpocetine was authentic, pure and confirming to the prescribed standards.

- Through the developed HPLC method, we could significantly improve the retention time of the drug from 6.5 (reported value) 1.5 minutes. The developed method was found to be accurate, sensitive, reproducible, precise and stable.

- Both ethosomal and transfersomal formulations of vinpocetine drug were successfully developed and converted to the gel formulations to incorporate into the patch.

- The mean particle size for optimized VIN ethosome and VIN transfersome formulations was found to be 93.54±1.54 nm and 94.66±1.43 nm respectively.

- All the ethosomal and transfersomal formulations could produce significant increase in the VIN flux via rat skin over their respective control groups.
Phospholipon 90G showed better results in characterization of both vesicles as compare to Phospholipon 75, therefore it was selected as lipid component for the final formulation used for in vivo studies.

Out of the two used surfactants for transfersomal formulations, sodium cholate presented better results in characterization of the vesicle as compare to the sodium deoxycholate, thus it was used as the optimized surfactant for transfersomal formulation used for in vivo studies.

Increasing Tween-80 concentration in ethosome formulations, caused increase in the size of the vesicle.

Changes in concentrations of phospholipid affects EE% of the ethosomal vesicles very significantly. On increasing phospholipid concentration from 75 to 90 mg, the EE% was significantly increased, though on further increase to 105 mg, the EE% decreased. Other two factors (surfactant and ethanol) had the similar effect as their optimized concentration was found to be 50 mg and 60% respectively.

Factors affecting flux and elasticity of the ethosomes were changes in concentrations of phospholipid, surfactant and ethanol ratio. All three had a similar effect as by increasing their concentrations up-to their optimized ones of 90 mg, 50 mg and 60% respectively, the flux and elasticity were increasing whereas further increase of these factors, caused decrease in the flux and elasticity of the vesicles.

In transfersomes, increasing the concentration of Sodium cholate from 10 to 15 mg caused decrease in particle size, increase in EE%, elasticity and flux, however further increase of it to 25 mg caused increase in particle size, decrease in EE%, elasticity and flux of the vesicles. This parameter was more prominent to affect the vesicle particle size, EE%, elasticity and flux of the vesicles as compared to other factor viz. phospholipid concentration and sonication time changes, though their effects were also similar to surfactant concentration changes.
Based on higher drug permeation, lowest droplet size, best surfactant selection, higher EE% and elasticity, ethosomal formulation containing Phospholipon 90G-(90 mg), Tween 80-(50 mg) and Ethanol ratio (60%) was optimized as the best composition of vinpocetine ethosomal formulation and for transfersome, the formulation having Phospholipon 90G-(90 mg), Sodium cholate-(15 mg) and Sonication time (15 minutes) were selected as the optimized vinpocetine transfersomal formulation.

TEM and SEM studies on both the vesicular formulations confirmed well-identified and spherical shape of both vesicles.

Confocal spectroscopy has shown us the highly significant drug permeation property of both the vesicular formulations as compared to their control groups. Drug penetration capacity in all four groups was found in the descending order as:

Ethosome > Transfersome > Liposome > Hydroethanolic solution

Pharmacodynamics studies on rats (by elevated plus maze test) revealed that anti-Alzheimer effects of the vesicular transdermal formulation of vinpocetine have been achieved successfully for a period of 48 hours.

Ethosomal formulations were more effective in alleviation of AD effects than transfersomal and both above formulations were more effective than marketed preparation.

Histopathological examination of ethosomal patch treated rat skin showed the disturbances by ethanol to the lipid bilayer structure of stratum corneum (SC) therefore enhancing its lipid fluidity to facilitate VIN permeation. The flexible ethosome vesicles can then penetrate the disturbed SC and even forge a pathway through the skin due to their particulate nature. The same effect is presented by surfactant in the case of transfersome vesicles to pass over SC easily and reach the systemic circulation. Moreover, the special surfactant like sodium cholate makes the vesicles ultradeformable and highly flexible thus, facilitating the permeation of the active substance.
The results from pharmacokinetic studies revealed that all the kinetic parameters were significantly improved in the case of both transdermal formulations as compared to the marketed one. Tmax for ethosomal & transfersomal formulations was found to be ~12 hr. while for Neurovin Tab. it was found to be 2 hr. Percent (%) Relative bioavailability of VIN via ethosomal and transfersomal formulations against Neurovin tab was significantly increased to 314.75 and 262.39 respectively.

As we discussed earlier, oral form of VIN is having short half-life of 2.5 hrs. while according to our study, it was significantly improved in case of ethosomal and transfersomal transdermal patches to 18.82 and 10.39 hours respectively. Elimination rate constant of both transdermal patches were found to be decreased significantly as compared to marketed tablet as ethosomal and transfersomal showed elimination rate of 0.036 and 0.06 per hour whereas for Neurovin Tab. it was found to be 0.26 per hours.

Brain histopathology studies confirmed neuronal density was higher in ethosomal and transfersomal formulations with reference to the marketed formulation. The result was as follows:

Normal control > Ethosome > Transfersome > Neurovin Tab. > Toxic control

According to the ELISA studies, the minimum amount of β-Amyloid protein in the brain was found in ethosomal group followed by transfersomal group while the marketed formulation showed less improvement.

No significant skin irritation (redness, swelling) was observed on treatment of skin with optimized ethosomal and transfersomal patch formulations of VIN as evident by visual and histopathological examinations of treated and untreated rat skin specimens.

Patch adhesion test showed good adhesion (i.e. ≥ 90 % adherence of patch, no lift off of the skin) of the patches to the skin area of each rat.
Stability studies of the optimized formulations showed that there were no significant changes in vesicle size and entrapment efficiency % values after a period of 180 days at room temperature (25 °C) which indicated good stability of formulations.

Shelf lives of ethosomal and transfersomal formulations have been calculated to be 1.88 and 1.98 years respectively by accelerated stability studies.

From both in vitro and in vivo data it can be concluded that the developed ethosomal and transfersomal formulations of vinpocetine drug, have great potential for transdermal drug delivery for better management of AD symptom.

Conclusion:

The present research furnishes strong evidence that our in-house ethosomal & transfersomal formulations provide superior pharmaceutical advantages as compared to the currently available marketed oral formulation of Vinpocetine for management of Alzheimer’s disease symptoms. Further research is recommended for establishing the therapeutic utility of these formulations by clinical studies on human volunteers and AD patients.