Permeation of pharmaceutical compounds through silanised poly(dimethylsiloxane)

Laura J. Waters*and Sani Sabo

School of Applied Sciences, University of Huddersfield, Queensgate, Huddersfield, HD1 3DH, UK,

*Corresponding author: l.waters@hud.ac.uk

Abstract

This work evaluates permeation of twelve model pharmaceutical compounds through a chemically modified form of poly(dimethylsiloxane) (PDMS), whereby the polymer surface had undergone silanisation. Standard polymer membrane has been widely used as a simplified skin model to investigate transdermal permeation yet does not fully mimic human skin. The surface chemistry of modified polymer was investigated such as the ability to bind to drugs, hydrophobicity and pore size using optical microscopy, the Brunauer-Emmett-Teller (BET) technique and Fourier-transform Infrared Spectroscopy (FTIR), followed by permeation analysis with UV spectroscopy. For eleven of the twelve compounds an appreciable increase in the extent of permeation was observed after six hours when using the silanised polymer compared with the standard PDMS. Furthermore, a correlation was found between the degree of permeation increase and hydrophobicity (logP) of the drug (R^2 = 0.90). These findings indicate that permeation can be controlled by modifying the membrane surface, although the hydrophobicity of the permeant also plays a vital role in the extent of permeation observed. This concept study presents a potential alternative membrane for pharmaceutical transdermal analysis, providing many benefits over existing options.

Keywords

Membranes; PDMS; permeation: poly(dimethylsiloxane); silanisation; transdermal

1. Introduction

Several alternatives to using human or animal skin when analysing the rate and extent of permeation of compounds have been proposed in recent years. Examples include mathematical

models (Mitragotri et al., 2011), human skin equivalents (Schmook et al., 2001) and chromatographic based methods (Waters et al., 2013b), a summary of the main proposed methods can be found in (L Waters, 2015). A reliable membrane to employ as a skin mimic is poly(dimethylsiloxane), also known as PDMS, which has been the focus of several research groups in recent years (Bhuiyan and Waters, 2017; Luo et al., 2016; Rodríguez-López et al., 2019; Shahzad et al., 2014; Waters and Bhuiyan, 2016; Waters et al., 2013a; Watkinson et al., 2009). PDMS has a silicon-oxygen backbone with a very high bond energy providing high thermal stability, i.e. it does not easily degrade. The general properties of PDMS are a consequence of the surface alkyl groups. This results in the conformational freedom experienced by the silicon-oxygen backbone, lack of steric hindrance, resistance to oxidation, all as a result of the small molecular volume of the alkyl substitute (De Jaeger and Gleria, 2008). Although the high degree of stability can be seen as advantageous, PDMS's poor wettability and low surface energy (i.e. it is relatively inert), have made it difficult to introduce aqueous compounds onto its surface thus it cannot be easily modified from its standard chemical state. However, recent studies have employed plasma treatment on the surface, thus transforming available methyl groups to hydroxyl groups, modifying the chemistry of the surface and consequently the permeability of selected model compounds (Waters et al., 2017).

In this work, after a simple plasma treatment was undertaken, a separate second chemical process was attempted, namely a silanisation reaction. This second reaction step involves reacting the oxidised PDMS polymer (containing surface hydroxyl groups), with the alkoxysilane dimethylphenylsilanol (DMPS). Alkoxysilanes have been widely accepted to constitute a stable membrane. Characteristic stability of these alkoxysilanes is mostly attributed to the presence of the Si-O-Si bond (Arkles et al., 1992). The organosilicon compound has lipophilic properties and can impart these properties to a hydrophilic surface (Virtanen et al., 1988), these properties guided the selection of the active silane reagent for this study. DMPS has a silanol 'tail' (Si-OH), the hydrolysed plasma treated membrane also contains silanol groups, and hence, a condensation reaction can occur. For this study hydroxyl groups present from both reactants (DMPS and plasma treated membrane) were reactive, consequently, a condensation reaction could occur, leading to attachment of the silane reagent to the membrane surface, creating a Si-O-Si bond. Such a membrane surface has not previously been proposed for the purposes of investigating permeation and it was therefore of interest to see how the modified membrane compared with standard, non-modified membrane regarding permeation for a set of model compounds.

2. Materials and Methods

2.1 Materials

Silicone membrane material 0.3 mm, was purchased from Silex UK, antipyrine (≥ 98 %), benzocaine (≥ 99 %), caffeine (≥ 99.5 %), diclofenac sodium salt (≥ 99 %), dimethylphenylsilanol (≥ 99 %) ethyl-4-hydroxylbenzoate (≥ 99 %), ibuprofen (≥ 99 %), lidocaine (≥ 99 %), procaine hydrochloride (≥ 97 %), potassium phosphate dibasic (≥ 98 %), potassium phosphate monobasic (≥ 99 %), and salicylic acid (≥ 99.5 %), were purchased from Sigma Aldrich Ltd., Dorset, UK. Ethanol (≥ 99.8 %), hydrochloric acid, ketoprofen (≥ 99 %), pentoxifylline (≥ 99.5 %), and sodium hydroxide were purchased from Fluorochem Ltd. Sodium chloride (≥ 99.5 %) from Acros Organics Ltd, and tetracaine (≥ 98 %) from Tokyo chemical industry, UK. All items were used as purchased unless stated otherwise.

2.2 Membrane Synthesis

Plasma treatment was performed using a benchtop Henniker Plasma machine (HPT-100). The process was carried out with air as the process gas in the gas chamber, at a power of 100 W, 68 mbar and flow rate of 8 standard cubic centimetres per minute (SCCM). A treatment time of 90 seconds was used throughout the process, selected based upon an optimisation study carried out (data not shown) whereby it was realised that treatments above 90 seconds affected the integrity of the membrane, making it not suitable for this study. Treatment was only undertaken on a single side of the membrane. Pieces of the membrane were cut into square forms sufficient enough to cover the diffusion area of the diffusion cell (~ 0.54 cm^{-2}) for subsequent permeation analysis. The top right corner of the square shaped membranes were marked, making it possible to identify the plasma treated side, when used for further analysis. Plasma treatment was followed by a silanisation process similar to that reported by (Sui et al., 2006) with minor modifications as listed. Dimethylphenylsilanol (DMPS) was used as the silane reagent, with solution phase oxidation undertaken in an acidic medium on the surface of the plasma treated PDMS. The dynamic nature of plasma treated PDMS, i.e. to avoid progressive recovery, meant that the membranes were immediately introduced into a closed reactor equipped with a magnetic stirrer. The reactor contained dimethyphenylsilanol DMPS (active silane reagent) (~1 mL) and ethanol (~5 mL) which was used as the solvent, with a reaction time of 30 minutes at 40 ˚C. Approximately 1-2 drops of dilute hydrochloric acid were

added to lower the pH, consequently enhancing the condensation process. Treated membranes were removed from the reactor, and dried under a nitrogen atmosphere for 10 minutes.

2.3 Membrane Analysis

Optical microscopic analysis was performed on unmodified and modified samples using a Keyence digital microscope VHX-2000 equipped with VH-z250R sensor, set at a multi-scan mode. Samples were placed in a sterilised glass plate to maintain a flat surface which enabled autofocus on the machine. The stage was also adjusted to a 20 mm working distance and x 250 magnification was maintained throughout the process.

The surface area of PDMS (before and after chemical modification) was obtained using a Micromeritics ASAP 2020 physisorption analyser. Samples were degassed at 50 ^{o}C under vacuum for 30 minutes, and then conditioned with nitrogen as the processed gas for 6 hours. After outgassing nitrogen, pore size, pore volume and consequently surface area were determined using nitrogen vapour adsorption data (77 K) and were evaluated by the Brunauer-Emmett-Teller model.

Comparative analysis specifically related to bond changes of modified and unmodified membranes was carried out using an Attenuated Total Reflectance – Fourier Transform Infrared Spectrometer (ATR-FTIR), Nicolet FTIR 380. The machine was in attenuated total reflectance mode with a diamond crystal as the sensor, Omnic software and the wavelength was adjusted from 400 – 4000 cm^{-1}.

Permeation analysis using diffusion cells in a controlled hydrostatic and hydrodynamic donor and receptor compartment respectively, was experimentally performed in triplicate, using twelve model pharmaceutical compounds. Each saturated solution of drug was prepared using pH 7.4 phosphate buffered saline. Saturated drug solutions were prepared by adding an excess amount of drug in 10 mL sample vials containing buffer. Sample vials containing a micro-size magnetic stirrer were mounted in a water bath maintained at ~32 ˚C for 24 hours, equipped with a magnetic stirrer plate. The resulting solution was filtered, and added via a syringe, in the donor compartment to avoid bubbles. Openings were occluded to maintain constant temperature and prevent evaporation. The donor compartment was mounted on a solid support which was equipped with a hollow passage where pre-heated water of approximately 35.5 ˚C passed through to help maintain a stable temperature within the donor compartment. Samples were collected from the receptor arm at 30-minute intervals for 6 hours and analysed using UV spectroscopy. A standard calibration graph of each drug ($R^2 \geq 0.9$.) was used to calculate resultant concentration for each permeation experiment. Membrane samples were also analysed after permeation analysis had been completed using BET and FTIR to ensure

membrane integrity had been maintained. All samples displayed no differences in results compared with those undertaken prior to permeation analysis implying the membrane integrity had been maintained for the duration of the experiment.

3. Results and Discussion

Characteristic changes in surface appearance were evaluated through comparison of the PDMS surface as purchased (unmodified), following plasma treatment and after modification with the aid of optical microscopy. An example image for each stage in the process can be seen in Figure 1.

From Figure 1 (A) and (B) little difference can be observed indicating that there was little or no physical change following the initial plasma treatment, i.e. that the integrity of the membrane was not compromised as a consequence of the process. A significant physical difference could be seen between the unmodified membrane (A) and the modified (silanised) PDMS (C). Following the silanisation process an increase in surface roughness and the presence of discontinuous patches can be seen, yet no physical alteration or damage within the membrane that might result in an increase in porosity is visible.

BET was used to determine surface area (as a reflection of porosity) of the membrane surface, more specifically comparing between unmodified and modified membrane. Unmodified membrane, was seen to have zero porosity (0 m^2/g) indicating the membrane is non-porous, as expected. Similar results was obtained by others (Nakade et al., 2005) when analysing PDMS samples. The non-porous nature of the membrane makes it a good candidate for use as a skin mimic as substances are forced to permeate through the membrane, rather than through pores. Therefore, it is vital to ensure that this feature is maintained throughout the process of modification rather than creating a membrane that allows compounds to freely pass through a porous structure.

Interestingly, the same value (0 m^2/g) was obtained when analysing the modified membrane, thus confirming that the process has not increased surface porosity, indicating the modification that has occurred was chemical rather than physical. This finding reinforces the assumption that any differences observed in the permeation profiles for a compound when comparing modified and unmodified membranes are not a consequence of a change in surface area or porosity and must involve chemical differences in interactions between the compound and membrane surface.

ATR-FTIR analysis was undertaken to analyse unmodified, plasma treated and modified membrane for confirmation of the presence of functional groups expected on the membrane surface. It is important to realise that only a small fraction of the surface is analysed at any one time, compared with the effective area dictating the overall process when undertaking permeation analysis. Regardless of these limitations, ATR-FTIR analysis was able to detect the presence of a surface hydroxyl group following plasma treatment, as expected following conversion of methyl groups to hydroxyl groups, as shown in Supplementary Information Figure S1. This notable change confirms the effectiveness of the plasma treatment step in the modification process. Furthermore, an increase in peak intensity attributed to the Si-O-Si bond when comparing spectra representing plasma treated PDMS and chemically modified membrane can be attributed to the desired formation of alkoxysilanes. Methyl groups were confirmed by symmetrical stretching vibrations at 2962.4, 2963.0 and 2962.3 cm^{-1} for unmodified, plasma treated and modified membranes respectively. Despite plasma treatment, the presence of methyl groups on the plasma treated membrane was seen and were attributed to underlying methyl groups, because only the membrane surface was targeted with the plasma treatment. Therefore, ATR-FTIR analysis was confirmed as having the ability to detect bonds embedded within the polymer matrix.

Asymmetric deformations of methyl groups from the membranes were also observed at approximately 1411 cm^{-1}, and symmetric deformation of methyl groups for modified, unmodified and plasma treated membranes were at 1257.2, 1257.1 and 1257.8 cm^{-1} respectively. Peaks from stretching vibrations at 1004.5, 1004.7, and 1005.7 cm^{-1} were attributed to Si-O-Si bonds, an increase in intensity of this peak for modified membrane further confirmed alkoxysilanisation had occurred. The presence of $O\text{-}SiCH_3$ was confirmed by peaks at 783.1, 784.4 and 783.7 cm^{-1}, the presence of $C\text{-}SiCH_3$ groups were confirmed with peaks at 699.3, 669.8 and 669.8 cm^{-1}. Peaks observed for the unmodified and modified membrane were in close agreement with those from literature (Bodas and Khan-Malek, 2006; Kim and Jeong, 2011). An absorbance at 3342.1 cm^{-1} indicated the presence of a C-OH group after exposing the membrane to plasma treatment, a similar finding to that reported by (Kim and Jeong, 2011). However, the hydroxyl group was not present in the subsequently modified membrane, yet the Si-O-Si- peaks remained, which was in line with expectations following a successful silanisation process.

The permeation of twelve compounds through modified PDMS was measured and analysed with comparison to permeation through unmodified membrane, as presented in Table 1. The percentage increase represents the percentage difference in cumulative mass ($\mu g/cm^2$) permeated for each

compound over a 6 hour period between modified and unmodified membrane. In an attempt to address some of the potential experimental and physicochemical variations an infinite dose method was used. This approach is believed to address donor chamber concentration variabilities by employing super-saturated solutions for all compounds facilitating calculation of cumulative mass permeated. Increases in the extent of permeation observed in Table 1 indicate that even though the modification was only on the membrane surface, the overall effect on membrane permeation was significant for the majority (eleven of the twelve) compounds analysed.

Silane reagent used in the modification protocol was selected to introduce lipophilic properties to the polymer hydrophilic surface (Virtanen et al., 1988). Such a modification had the effect of altering the physiochemical properties of the membrane, as reflected in the effect on permeation in this case. Having created a more lipophilic membrane, log P, known to influence the extent of permeation in epidermal tissues and synthetic lipophilic membranes (Baba et al., 2015; Potts and Guy, 1992) (Zhang et al., 2012), was among physical parameters used for the analysis of the results. It was found that compounds with comparatively high log P values tended to exhibit the greatest increase in permeation and it is not surprising for diclofenac having the highest log P value of 4.98 (amongst the compounds used), displayed the highest percentage increase, as displayed in Figure 2. To confirm that the observed increase in permeation was not simply a consequence of the plasma treatment step, permeation data for plasma treated membrane was also obtained. As exemplified in Figure 2, plasma treated membrane did not display a significant change in permeation compared with unmodified PDMS, in contrast to the effect observed following the silanisation process.

In agreement with this theory, caffeine displayed a similar correlation, as having the lowest log P value also had one of the lowest percentage increases (20.0 %), as displayed in Figure 3. An even more pronounced lack of permeation increase was observed for pentoxifyllinne (-6.0 %) which is also comparatively hydrophilic and can be considered to have a similar extent of permeation using either unmodified or modified membrane.

Overall, after analysing potential relationships between permeation and several physicochemical parameters such as molecular weight, ionisation, hydrogen bond donation/acceptance and polar surface area, a substantially linear correlation was obtained when comparing literature log P values with percentage increase (data presented in Table 1). This correlation can be increased further ($R^2 = 0.90$) through removal of diclofenac from the series where a far more substantial increase in permeation was observed than that expected,

most likely a consequence of the significantly greater hydrophobic nature of the compound compared with the others studied. It is important to note that the infinite dose approach was employed to avoid experimental variations whilst evaluating permeation of *in vitro* models, as highlighted by the work of others (Uchida et al., 2015). This relationship indicates that the change in lipophilicity on the membrane surface has a significant effect on subsequent compound through the membrane over the duration of the study.

4. Conclusion

In conclusion, a modified form of PDMS was successfully created whereby the polymer surface had undergone silanisation, confirmed using optical microscopy, the Brunauer-Emmett-Teller (BET) technique and Fourier-transform Infrared Spectroscopy (FTIR). Permeation analysis for eleven of the twelve compounds indicated an appreciable increase in the extent of permeation after six hours when using silanised polymer compared with standard PDMS. Furthermore, a correlation was found between the degree of permeation increase and hydrophobicity (logP) of the drug. These findings indicate that the degree of permeation can be controlled by modifying the surface, and therefore the hydrophobicity, of the membrane.

References

Arkles, B., Steinmetz, J., Zazyczny, J., Mehta, P.J.J.o.A.S., Technology, 1992. Factors contributing to the stability of alkoxysilanes in aqueous solution. 6, 193-206.

Baba, H., Takahara, J.-i., Mamitsuka, H., 2015. In silico predictions of human skin permeability using nonlinear quantitative structure–property relationship models. Pharmaceutical research 32, 2360-2371.

Bhuiyan, A., Waters, L.J., 2017. Permeation of pharmaceutical compounds through silicone membrane in the presence of surfactants. Colloids and Surfaces A: Physicochemical and Engineering Aspects 516, 121-128.

Bodas, D., Khan-Malek, C.J.M.e., 2006. Formation of more stable hydrophilic surfaces of PDMS by plasma and chemical treatments. 83, 1277-1279.

De Jaeger, R., Gleria, M., 2008. Silicon-based Inorganic Polymers. Nova Science Pub Incorporated.

Kim, H.T., Jeong, O.C.J.M.E., 2011. PDMS surface modification using atmospheric pressure plasma. 88, 2281-2285.

L Waters, L., 2015. Recent developments in skin mimic systems to predict transdermal permeation. Current pharmaceutical design 21, 2725-2732.

Luo, L., Patel, A., Sinko, B., Bell, M., Wibawa, J., Hadgraft, J., Lane, M.E., 2016. A comparative study of the in vitro permeation of ibuprofen in mammalian skin, the PAMPA model and silicone membrane. International Journal of Pharmaceutics 505, 14-19.

Mitragotri, S., Anissimov, Y.G., Bunge, A.L., Frasch, H.F., Guy, R.H., Hadgraft, J., Kasting, G.B., Lane, M.E., Roberts, M.S., 2011. Mathematical models of skin permeability: an overview. International journal of pharmaceutics 418, 115-129.

Nakade, M., Ichihashi, K., Ogawa, M.J.J.o.s.-g.s., technology, 2005. Preparation of titania/PDMS hybrid films and the conversion to porous materials. 36, 257-264.
Potts, R.O., Guy, R.H., 1992. Predicting skin permeability. Pharmaceutical Research 9, 663-669.
Rodríguez-López, L., Shokry, D.S., Cruz, J.M., Moldes, A.B., Waters, L.J., 2019. The effect of the presence of biosurfactant on the permeation of pharmaceutical compounds through silicone membrane. Colloids and Surfaces B: Biointerfaces 176, 456-461.
Schmook, F.P., Meingassner, J.G., Billich, A., 2001. Comparison of human skin or epidermis models with human and animal skin in in-vitro percutaneous absorption. International Journal of Pharmaceutics 215, 51-56.
Shahzad, Y., Waters, L.J., Barber, C., 2014. Solvent selection effects on the transport of compounds through silicone membrane. Colloids and Surfaces A: Physicochemical and Engineering Aspects 458, 96-100.
Sui, G., Wang, J., Lee, C.-C., Lu, W., Lee, S.P., Leyton, J.V., Wu, A.M., Tseng, H.-R., 2006. Solution-phase surface modification in intact poly (dimethylsiloxane) microfluidic channels. Analytical chemistry 78, 5543-5551.
Uchida, T., Kadhum, W.R., Kanai, S., Todo, H., Oshizaka, T., Sugibayashi, K., 2015. Prediction of skin permeation by chemical compounds using the artificial membrane, Strat-M™. European Journal of Pharmaceutical Sciences 67, 113-118.
Virtanen, J.A., Kinnunen, P.K., Kulo, A.E., 1988. Surface treatment agents and polymers comprising substituted phenyl silanes and siloxanes. Google Patents.
Waters, Bhuiyan, A.K.M.M.H., 2016. Ionisation effects on the permeation of pharmaceutical compounds through silicone membrane. Colloids and Surfaces B: Biointerfaces 141, 553-557.
Waters, Dennis, L., Bibi, A., Mitchell, J.C., 2013a. Surfactant and temperature effects on paraben transport through silicone membranes. Colloids and Surfaces B: Biointerfaces 108, 23-28.
Waters, L.J., Finch, C.V., Bhuiyan, A.M.H., Hemming, K., Mitchell, J.C., 2017. Effect of plasma surface treatment of poly (dimethylsiloxane) on the permeation of pharmaceutical compounds. Journal of Pharmaceutical Analysis 7, 338-342.
Waters, L.J., Shahzad, Y., Stephenson, J., 2013b. Modelling skin permeability with micellar liquid chromatography. European Journal of Pharmaceutical Sciences 50, 335-340.
Watkinson, R.M., Herkenne, C., Guy, R.H., Hadgraft, J., Oliveira, G., Lane, M.E., 2009. Influence of ethanol on the solubility, ionization and permeation characteristics of ibuprofen in silicone and human skin. Skin Pharmacology and Physiology 22, 15-21.
Zhang, K., Chen, M., Scriba, G.K., Abraham, M.H., Fahr, A., Liu, X., 2012. Human Skin Permeation of Neutral Species and Ionic Species: Extended Linear Free Energy Relationship Analyses. Journal of pharmaceutical sciences 101, 2034-2044.