Effects of Physicochemical Properties of Constituent Ions of Ionic Liquid on Its Permeation through a Silicone Membrane

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Ionic liquids (ILs), defined as liquid salts composed of anions and cations, have the advantage of allowing constituent ions to be stably absorbed through biological membranes, such as skin. However, limited information is currently available on the effects of the physicochemical properties of constituent ions on the membrane permeation of ILs. Therefore, we herein investigated the effects of the polarity of constituent cations on the membrane permeation of each constituent ion from IL. Various ILs were prepared by selecting lidocaine (LID) as a cation and a series of p-alkylbenzoic acids with different n-octanol/water partition coefficients (Kow) as anions. These ILs were applied to a skin model, a silicone membrane, and membrane permeability was investigated. The membrane permeabilities of p-alkylbenzoic acids from their single aqueous suspensions were also measured for comparison. The membrane permeability of p-alkylbenzoic acid from the aqueous suspension increased at higher Kow. However, the membrane permeability of ILs was similar regardless of the Kow of the constituent p-alkylbenzoic acid. Furthermore, the membrane permeability of the countercation LID remained unchanged regardless of the constituent p-alkylbenzoic acid. These results suggest that even when the Kow of IL constituents markedly differs, the resulting IL does not affect membrane permeability.

Key words ionic liquid; silicone membrane; permeability coefficient; n-octanol/water partition coefficient

Introduction

Oral preparations, such as tablets and capsules, and intravenous, subcutaneous, and intramuscular injections are the predominant administration sites and formulations for drugs. High-molecular-weight drugs, such as antibodies, and medium-molecular-weight drugs, including peptides, have been used in addition to conventional small-molecule-weight drugs, resulting in a variety of therapeutic modalities. High- and medium-molecule-weight drugs are mainly administered by injections. However, injections generally cannot be administered by patients themselves or by their caregivers. Furthermore, the administration of oral preparations is limited by very low absorbability. Although transdermal drug delivery systems, which are the third route and method of drug administration, are convenient, the drug absorption rate is generally lower than that of oral preparations. A number of physical approaches, such as microneedles and iontophoresis, were recently shown to increase the skin permeation of high- and medium-molecular-weight drugs. The development of physical approaches has been advantageous, particularly for high- and medium-molecular-weight drugs, resulting in breakthroughs in drug formulations for these drugs.

While the potential to overcome a low absorption rate has emerged, the decomposition and inactivation of drugs in topically applied formulations, and decreases in solubility due to, for example, polymorphisms have yet to be resolved. The use of ionic liquid (IL) has been paid attention as a method to overcome these issues. IL is defined as a liquid salt composed of anions and cations that is absorbed through the skin barrier.1) The membrane permeability of IL may be calculated based on thermodynamic activity, similar to that of drugs.2) Nevertheless, difficulties are associated with accurately measuring oil-water partition coefficients, such as the n-octanol/water partition coefficient (Kow) of IL, because IL divides into anions and cations when it makes contact with water.

Efficacy and safety evaluations of topically applied drugs have generally been performed based on the skin permeation rate and concentration of a drug. The skin permeation of a drug is a physicochemical phenomenon, and its rate is mainly explained by Fick’s diffusion law. When a drug is applied, it distributes throughout and diffuses into the skin. The diffusion rate of a drug in the skin barrier largely depends on its molecular weight, whereas its distribution is measured by the ratio of the activity coefficient of the drug between the topical formulation and the skin barrier. The permeability of a drug with different physicochemical properties in the same formulation or that of the same drug from a different formulation increases with elevations in the thermodynamic activity of the drug in the formulation. Furthermore, if the same drug is suspended in a formulation, skin penetration becomes constant even if the formulation is changed. Human or animal skin is mainly used to assess the skin permeability of drugs. However, due to animal ethics issues and the ease of experimental operations, model membranes, such as a three dimensional (3D) cultured human skin model, Strat-M®, and silicone membranes, are also used. Moreover, in silico methods and in vitro skin permeation tests are available in addition to Kow.

Studies to evaluate the relationship between the physicochemical properties of drugs and skin permeability have often been performed by dissolving one drug in a vehicle, such
as water. Since IL separates into anions and cations when it makes contact with water, difficulties are associated with accurately measuring $K_{o/w}$. Therefore, it is also challenging to predict the membrane permeability of IL using $K_{o/w}$.

In the present study, we investigated the effects of the $K_{o/w}$ of the constituent acid in IL on membrane permeability. Lidocaine (LID) was selected as the cationic substance (base), and a series of $p$-alkylbenzoic acids with similar molecular weights (MW: 136.15 to 206.28) and markedly different log$clogP$ were selected as anionic substances (acid). In addition, a silicone membrane was used as the skin model, and the silicone membrane permeation rates of these ILs were assessed.

**Experimental**

**Materials** LID and $p$-methylbenzoic acid were purchased from Tokyo Kasei Co., Ltd. (Tokyo, Japan). $p$-Ethylbenzoic acid was purchased from APOLLO SCIENTIFIC (East Hyde, U.K.). $p$-Butylbenzoic acid and $p$-hexylbenzoic acid were purchased from Kanto Chemical Co., Inc. (Tokyo, Japan). $p$-Pentylbenzoic acid was purchased from FUJIFILM Wako Pure Chemical Corporation (Osaka, Japan). Other reagents and solvents were used as is for commercial liquid chromatographs or special grade products without purification. Table 1 shows the physicochemical properties of each model substance to make ILs. The table also shows the abbreviations of the materials. A silicone membrane (thickness: 75 µm) was provided by LINTEC Corporation (Tokyo, Japan).

**IL Preparations** The cationic substance LID and the anionic substance $p$-alkylbenzoic acid were mixed at a molar ratio of 1:1 and dissolved in methanol. After dissolution, IL was obtained by completely removing methanol under reduced pressure using an evaporator. Table 2 shows the prescription of each IL. The obtained ILs were confirmed to be liquid and further evaluated for an endothermic peak by DSC measurement (Fig. 1). The DSC measurements were performed using the following conditions: temperature range from room temperature to melting point temperature of each material, heating rate at 10°C/min, in an atmosphere of N$_2$ gas at a flow rate of 50 mL/min, sample amount about 4 mg, and an aluminum seal cell for the sample cell. The endothermic peak disappeared by this IL formation.

**Preparation of Each IL Solution in 1, 3-Butandiol Solution (IL/1,3-BG)** IL/1,3-BG solution was prepared by dissolving the prepared IL in 1,3-BG. The concentrations of the anion ($p$-alkylbenzoic acid) and cation (LID) in IL were both 0.1 mol/L.

**Preparation of Each Compound-Suspended Aqueous Solution (Suspended Solution)** Approximately 3-fold the amount of $p$-alkylbenzoic acid with saturated solubility was placed in purified water and stirred at 37°C for 24 h to obtain a suspended solution of $p$-alkylbenzoic acid.

**Membrane Permeation Experiments** A silicone membrane was set in a horizontal 2-chamber diffusion cell (permeation area: 0.95 cm$^2$). Approximately 3.0 mL of the $p$-alkylbenzoic acid-suspended solution, neat IL, or IL/1,3-BG and isotonic phosphate-buffered saline (PBS) pH 7.4 were then added to the donor and receiver cells, respectively, to start the permeation experiment. During the experiment, water maintained at a temperature of 37°C was circulated outside the double cells, and the donor and receiver solutions were both stirred with a magnetic stirrer. The receiver liquid (2 mL) was collected at the sampling time, and the same amount of PBS was returned to maintain the sink condition of the receiver solution.

**Assessment of Each Compound** A HPLC system (Shimadzu Corporation, Kyoto, Japan) was used. TSKgel ODS-80Ts QA (TOSOH, Tokyo, Japan) was used as the separation column, and the column temperature was maintained...
at 40°C. The mobile phase was prepared in a solution of acetonitrile : purified water = 1:1 with a final concentration of 5 mM sodium dodecyl sulfate and 0.05% phosphoric acid. UV measurements were conducted at 249 nm.

**Results and Discussion**

Figures 2a and b show the cumulative amount of \( p \)-alkylbenzoic acid and LID that permeated through the silicone membrane from its aqueous suspension. Figures 2c and d show the cumulative amounts of \( p \)-alkylbenzoic acid and LID, respectively, from neat IL. Figures 2e and f show the cumulative amounts of \( p \)-alkylbenzoic acid and LID, respectively, from IL in 1,3-BG solution. The cumulative amount that permeated through the silicone membrane was normalized to the applied drug concentration and shown in %/cm². A lag time was not observed in most datasets due to the high diffusion rate of drugs through the silicone membrane, and an almost steady-state flux was noted from time zero. The cumulative amounts of \( p \)-alkylbenzoic acids that permeated through the membrane from its aqueous suspension increased as the log \( K_{ow} \) of \( p \)-alkylbenzoic acids became higher. However, the permeabilities of \( p \)-alkylbenzoic acids and LID were similar when neat IL and IL/1,3-BG were applied to the silicone membrane even when different \( p \)-alkylbenzoic acids were used.

Figure 3a shows the relationship between the permeability coefficients of \( p \)-alkylbenzoic acids from their suspended solution, neat IL, and IL/1,3-BG and the log \( K_{ow} \) of \( p \)-alkylbenzoic acids. The permeability coefficient was calculated by dividing the steady-state flux obtained in Fig. 2 by the applied concentration. The permeability coefficients of a series of \( p \)-alkylbenzoic acids from aqueous suspensions increased as their log \( K_{ow} \) became higher. On the other hand, the permeability coefficients of \( p \)-alkylbenzoic acids when neat IL and IL/1,3-BG were applied to the silicone membrane remained almost constant, even when the log \( K_{ow} \) of \( p \)-alkylbenzoic acid differed. In addition, the permeability coefficient of \( p \)-alkylbenzoic acid was significantly lower when neat IL and IL/1,3-BG were applied to the silicone membrane than with the application of the aqueous suspension of \( p \)-alkylbenzoic acids.

Figure 3b shows the log \( K_{ow} \) of \( p \)-alkylbenzoic acids and the permeability coefficients of LID from neat IL and IL/1,3-BG. The permeability coefficient of LID remained almost constant even when the log \( K_{ow} \) of \( p \)-alkylbenzoic acid increased, similar to the permeability coefficient of \( p \)-alkylbenzoic acid. By converting from an anion and cation with different physicochemical properties to IL, the permeation coefficient became constant. The relationship was determined between the log \( K_{ow} \) of IL and the membrane permeability of IL. Unfortunately, it is difficult to directly determine the log \( K_{ow} \) of IL, because IL returns to the original anion and cation when contacted with water. It was speculated from Fig. 3 that even when the log \( K_{ow} \) of the anion component of IL is significantly different, the log \( K_{ow} \) of IL probably becomes similar. Therefore, it was speculated the physicochemical properties of IL are generally independent of the physicochemical properties of the anion or cation constituents in IL. In addition, the permeability coefficient of LID was significantly lower when neat IL and IL/1,3-BG were applied to the silicone membrane than with the application of the aqueous suspension of LID, i.e., the permeability coefficient of LID from neat IL and IL/1,3-BG was about twice as high as that of \( p \)-alkylbenzoic acid. Thus, both LID and \( p \)-alkylbenzoic acid have similar permeability through the membrane as the IL. However, IL was probably divided into LID and \( p \)-alkylbenzoic acid after being distributed to the silicone membrane and permeated through the membrane, because the permeability coefficients of LID and \( p \)-alkylbenzoic acid were
In a comparison of the permeability coefficients of \( p \)-alkylbenzoic acids and LID from neat IL and IL/1,3-BG (see Figs. 3a and b), the permeability coefficients of \( p \)-alkylbenzoic acids and LID were higher from IL/1,3-BG than from neat IL. IL was released from the 1,3-BG vehicle in the case of IL/1,3-BG, whereas IL was released from the neat IL vehicle in the case of Neat IL. In other words, the thermodynamic activities of IL in the 1,3-BG vehicle were estimated to be higher than those of IL itself. Therefore, the permeability coefficients of \( p \)-alkylbenzoic acids and LID were higher from IL/1,3-BG than from neat IL.

**Conclusion**

In the present study, a series of \( p \)-alkylbenzoic acids with similar structures were selected to prepare IL with LID, and the permeabilities of \( p \)-alkylbenzoic acids and LID from IL through the silicone membrane were examined. Regardless of the \( \log K_{ow} \) of \( p \)-alkylbenzoic acids, similar membrane permeabilities were observed. In the future, we intend to perform silicone membrane permeation experiments on ILs prepared using several anions and cations with different physicochemical properties in order to investigate the relationship between the physical properties of IL and silicone membrane permeation.

**Conflict of Interest** The authors declare no conflict of interest.

**References**