Complex Formation with Layered Double Hydroxides for the Remediaion of Hygroscopicity

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Layered double hydroxides (LDHs) have been used commercially as antacids, to stabilize drugs, to allow the controlled release of incorporated drugs, and to act as drug carriers to reduce drug accumulation within the body. Several types of LDH were investigated: nitrate type (LDH-NO₃); chloride type (LDH-Cl); and carbonate type (LDH-CO₃). Each type was added to an aqueous or methanol (MeOH) solution containing a drug (pravastatin or nateglinide). With pravastatin sodium, the interlayer distance expanded after reaction with LDH-NO₃ and LDH-CI in aqueous solution. In contrast, the interlayer distance of LDH-CO₃ increased in methanol with nateglinide. Each drug was intercalated into the interlayer space of LDH by ion exchange. The hygroscopicity of the drug substances, complexes, and physical mixtures were determined at 70% relative humidity. Increases in weight (%) of the complexes were less than those of the physical mixtures, which demonstrates that hygroscopicity was reduced upon complexation with LDH due to the layer of LDH over the drugs.

Key words layered double hydroxide; hygroscopicity; drug

Layered double hydroxides (LDHs) have been used as matrices for drug delivery systems and as coating materials to protect compounds against external influences such as light and humidity. For example, complexation with LDH improved the thermostability of 5-fluorouracil. Additionally, LDH complexed with methotrexate, an anticancer drug, provided a lower IC₅₀ value compared to methotrexate alone, LDH complexed with methotrexate, an anticancer drug, and carbonate (CO₃²⁻) in the interlayer region. However, the carbonate ion is the most stable in the interlayer space due to its high affinity for the layer. Therefore, decarbonation of LDH-CO₃ has been considered difficult, and LDH-CO₃ has been transformed to calcined-LDH by heating in an oven at 500°C for reconstruction reactions. Iyi et al. reported that decarbonation occurred in mixed solutions of hydrochloric acid and sodium chloride, as well as in acetic acid and sodium chloride at controlled pH under N₂ flow. A recent study demonstrated that carbonate ion in LDH-CO₃ could be exchanged easily with organic and inorganic anions by using methanol as the solvent. This method is applicable to the uptake of organic compounds that have only slight solubility in water.

Pravastatin sodium is a drug with high hygroscopicity used to treat hyperlipidemia. Nateglinide is a drug used to treat diabetes mellitus; hygroscopicity was induced using an additive (crsopovidon) common in formulation. The hygroscopicity of these drugs would be reduced by incorporating them into the LDH interlayer space. It opens the application of biocompatible hydrotalcite.

Experimental

Chemicals Carbonate-type MgAl-LDH (LDH₃-CO₃) [Mg₀.75Al₀.25(OH)₂(CO₃)₀.125·0.52H₂O, Mg/Al=3.0] and chloride-type MgAl-LDH (LDH₂-Cl) [Mg₀.66Al₀.34(OH)₂·0.3Cl₀.26(CO₃)₀.01·0.48H₂O, Mg/Al=2.2] were obtained from Kyowa Chemical Industry Co., Ltd. (Kagawa, Japan), and Tomita Pharmaceutical Co., Ltd. (Tokushima, Japan), respectively. Nitrate-type MgAl-LDH (LDH₂-NO₃) [Mg₀.66Al₀.33(OH)₂·0.3NO₃₀.25(CO₃)₀.02·0.32H₂O, Mg/Al=2.0] was synthesized by coprecipitation and was contaminated with CO₃²⁻ during synthesis. Other reagents, obtained from Wako Pure Chemical Industries, Ltd. (Osaka, Japan), were used without further purification.

Procedure for Complex (LDH-Drug) Synthesis The ion-exchange reaction was performed by mixing (500rpm) 0.025 or 0.05g of LDH and 10mL of solvent (methanol or...
Reactions were conducted using different drug concentrations, reaction temperatures, and reaction times. The suspension was centrifuged (3500 rpm) and the solid (complex) obtained was washed with the same solvent used for the reaction and dried under vacuum for 3 h at room temperature.

**Hygroscopicity Test** Hygroscopicity testing was performed on the following powder samples: intercalation compound (complex), original pravastatin sodium or nateglinide, crospovidone, LDH3-CO$_3$, and a physical mixture of the two (pravastatin and LDH, nateglinide and LDH or nateglinide and crospovidone). The powder samples were dried in vacuum and stored in a desiccator at 70% relative humidity, maintained by a saturated sodium chloride solution. Any increase in weight was determined over time.

**Analysis** The interlayer distance in the solid obtained was measured by X-ray diffractometry using a Rint 2000 diffractometer (Rigaku Co., Ltd., Tokyo, Japan) with Ni-filtered CuKα radiation. The carbon content of the complex was determined by elemental analysis using a Sumigraph NC-80 analyzer (Sumika Chem. Anal. Service, Ltd., Osaka, Japan). The $^{13}$C cross polarization/magic angle spinning (CP/MAS) NMR spectra were obtained using a Varian NMR System 500 instrument (Agilent Technologies Inc., Santa Clara, CA, U.S.A.) with a recycle time of 5 s and accumulations of up to 5470 scans. IR spectra were obtained using a Thermo-Electron FT-IR 200 instrument (Thermo Fisher Scientific Inc., Waltham, MA, U.S.A.) by KBr tablet method.

**Results and Discussion**

The structures of the drugs used in the study are shown in Fig. 1. Pravastatin sodium contains a sodium carboxylate group while nateglinide contains a carboxyl group. These groups can interact with the positively charged brucite-like layer. The LDHs (LDH2-NO$_3$, LDH2-Cl, and LDH3-CO$_3$) were added to a solution of one of the drugs in water or methanol (MeOH), and reactions were performed at different drug concentrations, temperatures, and times.

**Intercalation of Pravastatin Sodium** Pravastatin sodium is soluble in water, and could be intercalated into LDH2-Cl and LDH2-NO$_3$, which was confirmed by an increase in interlayer distance in the X-ray diffraction (XRD) patterns. For example, XRD results show that the interlayer distance of LDH2-NO$_3$ increased from 0.89 nm before reaction to 3.2 nm after reacting with pravastatin sodium (Fig. 2), and the peak intensity corresponding to $d=3.2$ nm was enhanced slightly with increased reaction temperature. The layered structure of LDH was maintained after the reaction and pravastatin was arranged regularly in the interlayer region. In addition, a secondary peak was observed in the XRD patterns. The arrangement of pravastatin in the interlayer region was suggested by the increase in interlayer distance and the size of the pravastatin molecule. The increase in interlayer distance was calculated to be 2.72 nm by subtracting the thickness of the brucite-like layer (0.48 nm) from the interlayer distance of the complex (3.2 nm). In contrast, the size of pravastatin (the long axis length from carboxylate group) was 1.65 nm. It is noted that the carboxylate group (1560–1570 cm$^{-1}$) exists in the interlayer space from their IR spectra (Fig. S1). And it interacts with Al$^{3+}$ in the brucite-like layer. Because the increase in interlayer distance was greater than the size of the pravastatin, the pravastatin molecule must exist as a bilayer structure without decomposition. This trend also was observed for LDH2-Cl.

The amount of pravastatin uptake was calculated using carbon content (Fig. 3). Results showed that LDH2-Cl was easily influenced by reaction temperature when compared with LDH2-NO$_3$. The LDH2-NO$_3$ incorporated a sufficient amount of pravastatin, even at 30°C. As shown in Fig. 3b, pravastatin was rapidly incorporated into LDH and the amount of uptake in 1 h was approximately 2 and 1 mmol/g by LDH2-NO$_3$ and LDH-Cl, respectively. Uptake amount increased with pravastatin concentration and reached a maximum of 3.4 mmol/g for LDH2-NO$_3$ and 3.1 mmol/g for LDH2-Cl (Fig. 3c). These uptake amounts of LDH2-NO$_3$ and LDH2-Cl were 80 and 84%, respectively, of the ion-exchange capacity of LDH. Uptake amount did not reach 100% due to the steric hindrance of pravastatin. One large pravastatin molecule was placed on the adjacent binding site in the brucite-like layer. Optimal reac-
tion conditions for intercalation of pravastatin in both LDHs included reaction at 60°C for 24 h in 20 mM of pravastatin concentration. Reactions with LDH depended mainly on drug concentration.

The test for hygroscopicity monitored the weight of sample powders over time at 70% relative humidity. Figure 4 shows the increase in sample weights over time. The testing samples are 0.017 g of pravastatin, 0.03 g of the complex, and 0.034 g of the physical mixture (0.017 g of pravastatin plus 0.017 g of LDH2-Cl). The testing sample amount of 0.017 g of pravastatin corresponds to the amount of it within 0.03 g of complex. The weight of pravastatin alone increased rapidly by approximately 18% in 24 h. The weight of the physical mixture increased by 12%. In contrast, increases in the weights of the LDH2-Cl and LDH2-NO3 complexes were less than 5%. Thus, the hygroscopicity of pravastatin was depressed upon complexation with LDH.

**Intercalation of Nateglinide**

Nateglinide is insoluble in water, but is soluble in methanol and ethanol. Intercalation of etodolac, which is slightly soluble in water, was achieved by mixing it with LDH3-CO3 in methanol. Therefore, a methanol solution of nateglinide was stirred with three different types of LDH at 50°C. An increase in interlayer distance was observed for the solid obtained after stirring with LDH3-CO3, which indicated successful intercalation of nateglinide into LDH by ion exchange with carbonate ion. Nateglinide also formed a complex with LDH in methanol. The 13C CP/MAS NMR spectrum of the complex did not contain a methanol peak, confirming that the complex was not contaminated with methanol. The XRD patterns of solids obtained at different concentrations of nateglinide are shown in Fig. 5. The interlayer distance of the complex with nateglinide is 2.3 nm which expanded from 0.78 nm of host LDH3-CO3. An increase in nateglinide concentration resulted in an increase in intensity of the peak corresponding to \( d = 2.3 \) nm, while the intensity of the peak at 0.78 nm decreased. Uptake amount increased with nateglinide concentration and reached a maximum of 2.2 mmol/g for 40 mM (Fig. 6a), which corresponds to 73% of the ion-exchange capacity of LDH3-CO3 (3.30 mmol/g). Uptake amount also depended on reaction temperature and...
time (Figs. 6b, c). Nateglinide was intercalated rapidly into LDH; its uptake amount became saturated at 3 h. Optimal reaction conditions for intercalation of nateglinide were 20 mM nateglinide concentration in MeOH, 50°C, and 24 h. Although the intercalation reaction also was performed using ethanol, the reaction occurred more readily in methanol. A previous report indicated that carbonate ion (CO$_3^{2-}$) in LDH$_3$-CO$_3$ was released as HCO$_3^-$ by abstracting a proton from methanol (MeOH).$^{21}$) Because the pK$_a$ of MeOH and ethanol (EtOH) is very similar (15.5 and 16, respectively), the effect of a proton on reactivity is likely to be small. Perhaps the difference in polarity of the alcohols (dielectric constants of 33 and 24 for MeOH and EtOH, respectively) influences the amount of anionic nateglinide (i.e., nateglinide easily deprotonates the carboxyl group and intercalates into LDH in methanol with greater polarity).

Information about the arrangement of nateglinide in the interlayer region was suggested by the results. In their IR spectra (Fig. S2), a peak of carboxyl group (1714 cm$^{-1}$) shifted to 1590 cm$^{-1}$ and carbonate ion (1371 cm$^{-1}$) disappeared through the intercalation. This result indicated that anionic nateglinide exchanged with carbonate ion in MeOH and interacted with Al$^{3+}$ in brucite-like layer. The interlayer distance increased by 1.82 nm, which was determined by subtracting the thickness of the brucite-like layer (0.48 nm) from the interlayer distance of the complex (2.3 nm). In this case, it is feasible that nateglinide exists as a bilayer structure with the short axis from the carboxylate group perpendicular to the plane of the LDH layer. However, since the cyclohexane portion of nateglinide was on an adjacent binding site in the brucite-like layer, the uptake amount of nateglinide did not reach 100%.

Hygroscopicity was determined by monitoring the weight of a powder sample over time at 70% relative humidity. Results indicated an increase in sample weights from the initial weight of 0.025 g crospovidone or nateglinide or LDH$_3$-CO$_3$, 0.04 g complex, and 0.05 g of the physical mixture of crospovidone and nateglinide or LDH$_3$-CO$_3$ and nateglinide (0.025 g crospovidone plus 0.025 g nateglinide or 0.025 g LDH$_3$-CO$_3$ plus 0.025 g nateglinide) (Fig. 7). Crospovidone was used because the addition of crospovidone causes hygroscopicity to the preparations of nateglinide (starch). These sample amounts were determined based on the amount of nateglinide (0.025 g).
in 0.04 g complex. The nateglinide sample did not exhibit any hygroscopicity. In contrast, crospovidone exhibited deliquescence after 3.3 h, with a weight gain greater than 20%. Although weight of the physical mixture with crospovidone increased by 6%, weight gain of the complex was less than 2%, indicating that hygroscopicity of the crospovidone additive was suppressed by complexation with LDH. The weight gain of physical mixture with LDH3-CO3 was 2.5% similarly to complex. In addition, the fluidity of nateglinide, which tended to clump, was improved by intercalation into LDH as shown in Fig. S3.

Conclusion
Pravastatin was incorporated into the interlayer region of nitrate- and chloride-type LDH in aqueous solution through an ion-exchange reaction, resulting in an interlayer distance of the complex of 3.2 nm. The amount of pravastatin uptake increased with its concentration in solution. Hygroscopicity of pravastatin was suppressed by intercalation into LDH (i.e., through coating with the LDH layer).

Nateglinide formed a complex with carbonate-type LDH in methanol, with an interlayer distance of 2.3 nm. Nateglinide uptake amount increased with its concentration in solution and with reaction temperature, reaching one-half of the ion-exchange capacity of carbonate-type LDH at 1 h. Hygroscopicity of additives could be suppressed by intercalation of nateglinide into LDH without using crospovidone. The fluidity of the intercalation compound also was better than that of nateglinide alone.

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Conflict of Interest
The authors declare no conflict of interest.

Supplementary Materials
The online version of this article contains supplementary materials.

References