Drug Absorption from Large Intestine: Physicochemical Factors Governing Drug Absorption

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Profiles of absorption versus drug molecular weight and absorption versus drug lipophilicity were investigated in both the small and large intestines of rats by an in situ loop method. The absorption–molecular weight profiles examined using different-sized polyethylene glycols (PEGs) were different between the small and large intestines; the large-intestinal absorption of PEGs with molecular weights larger than 300 was poor, while PEGs with molecular weights up to 600 were relatively well absorbed in the small intestine. It is suggested that the paracellular route for drug penetration in the large intestine is restricted more than in the small intestine.

The absorption–lipophilicity profiles were also examined in various regions (loops of 6 cm) of rat intestine using three acylsalicylic acids, acetyl-, propionyl- and butyrylsalicylic acids. The absorption rates of the acylsalicylic acids were different in the intestinal regions: the jejunum > the ileum > the colon > the rectum. In each region, the absorption rate increased with the drug lipophilicity. However, it was shown that the absorption rates in the small intestine tended to reach a ceiling at the high lipophilicity. To confirm this tendency, the absorption rates of acetaminophen and indomethacin were compared in the four intestinal regions. The absorption rates of highly lipophilic indomethacin were similar in the large and small intestines, while immediately lipophilic acetaminophen was more rapidly absorbed in the small intestine than in the large intestine. A thicker unstirred water layer adjacent to the small-intestinal mucosa would be one of the factors which cause such varying absorption–lipophilicity profiles.

Keywords drug absorption; large intestine; molecular weight; lipophilicity; unstirred water layer; polyethylene glycol

Drug absorption from the large intestine is considered to be important for certain drugs: (1) since protease activities in luminal contents of the large intestine are generally much lower than those in the small intestine, it seems a suitable site for the absorption of protease-labile peptide drugs; (2) whether or not the drug can be absorbed from the large intestine is an important factor affecting the bioavailability of the drug in sustained release dosage forms. However, large-intestinal drug absorption has not been elucidated sufficiently.

Thus, we investigated the physicochemical properties of a drug governing large-intestinal absorption in rats. In this paper, the absorption versus drug molecular weight and the absorption versus drug lipophilicity profiles in the large intestine are reported in comparison with those in the small intestine.

MATERIALS AND METHODS

Materials Propionylsalicylic acid and butyrylsalicylic acid were generous gifts from Taisho Pharmaceutical Co., Tokyo, Japan. [1-14C]Dexy alcohol was purchased from ICN Pharmaceuticals, Irvine, CA, U.S.A. [3H]Dextran and a liquid scintillator, ACS-II, were purchased from Amersham International, Buckinghamshire, England. Polyethylene glycol (PEG)-300, PEG-600 and PEG-1000 (Union Carbide Chemicals and Plastics Co., Danbury, CT, U.S.A.) were mixtures of PEGs with mean molecular weights of approximately 300, 600 and 1000, respectively. All other chemicals were reagent grade commercial products.

Determination of Lipophilic Index The lipophilic index, log(k'<sub>
</sub>o), of each model drug was determined at pH 7.4 by reversed-phase HPLC according to the method of Yamana et al. The chromatograph was an LC-6A (Shimadzu, Kyoto, Japan) equipped with a spectrophotometric detector (SPD-2A, Shimadzu). A column (4.6 mm x 150 mm) was packed with Inertis OD (5 μm; GL Sciences, Tokyo), and all compounds were detected at 254 nm. The mobile phases of methanol–10 mm sodium phosphate buffer solution (pH 7.4) at various volume ratios were run at a flow rate of 1.0 ml/min. Formamide was used as an unretained substance. The log(k') was defined as follows:

\[
\log(k') = \log\left(\frac{t_R - t_o}{t_o}\right)
\]

where \(t_R\) and \(t_o\) are the retention times of a retained peak and of an unretained peak, respectively. The log(k') was plotted against methanol concentration (v/v %), and the lipophilic index, log(k'<sub>0</sub>), was defined as a log(k') value extrapolated to 0% methanol.

Procedure of Absorption Experiments Male Wistar rats (180–260 g body weight) were used as the experimental animal. Intestinal absorption was examined by the in situ loop method. PEGs: Briefly, under pentobarbital anesthesia, the intestine was exposed by a midline abdominal incision and a loop of whole small intestine (from the proximal end of the duodenum to the ileo-cecal junction) or large intestine (from the proximal end of the colon to the anus) was formed by cannulating both ends with silicone tubing. Care was taken to exclude major blood vessels from the ligatures. The bile duct was ligated in all the experiments. The contents of the lumen were washed out with saline warmed to 37 °C, then an isotonic aqueous solution of each drug was injected into the loop and each cannula was clamped with forceps. The organs were returned to the abdomen, and the incision was closed. The buffering salts of the isotonic solution were Na₂HPO₄–NaH₂PO₄.
at pH 6.5 for the small intestine and at pH 7.4 for the large intestine. The initial concentrations were 10 mg/ml for PEG-300, and 5 mg/ml for PEG-600 and PEG-1000. The volumes of the solution injected were 5 and 2 ml for the small and large intestine, respectively. At 1 h after the administration, the solution in the segment was withdrawn and the lumen was washed out with the same buffer solution. The solution and the washings were combined and the volume was adjusted to 50 ml. The difference in the amount of the drug between the initial and final solutions was defined as the amount absorbed.

Acetylsalicylic Acids, Acetaminophen and Indomethacin: The absorption of these drugs was examined by the in situ loop method, similarly to that for PEGs, but the loops (6 cm in length) were prepared in the jejunum, the ileum, the colon and the rectum. The drug solution administered was 0.5 ml at a concentration of 5 mm for acetylsalicylic acids, 10 mg/ml for acetaminophen and 1 mg/ml for indomethacin in an isotonic phosphate buffer (pH 7.4). The periods of the absorption experiment were 20 and 60 min in the small and large intestines, respectively, for acetylsalicylic acids, and 10 min in all the segments for acetaminophen and indomethacin. In some of the experiments for acetaminophen and indomethacin, the drugs were administered into each intestinal loop and blood samples (0.2 ml) were collected periodically from the cannula of the carotid artery for 2 h without washing out the drug solution from the loop. The plasma was separated immediately by centrifugation and the drug concentration in the plasma was determined by HPLC.

Pharmacokinetic Analysis of Plasma Concentration–Time Curves Pharmacokinetic evaluations were carried out by non-compartmental analysis of the plasma concentration–time data based on the statistical moment theory. The moments were calculated by the trapezoidal method with a monoexponential extrapolation of the terminal phase. The mean absorption time \( (MAT) \) in the case of the intra-intestinal loop administrations was estimated by applying the following equation:

\[
MAT_{\text{intestinal loop}} = MRT_{\text{intestinal loop}} - MRT_{\text{i.v.}}
\]

where \( MRT \) is the mean residence time of the drug in plasma, and i.v. and intestinal loop are the subscripts for the intravenous administration and the intra-intestinal loop administration in each intestinal segment, respectively. The systemic bioavailability \( (F) \) of a drug was calculated by applying the following equation:

\[
F_{\text{intestinal loop}} = \frac{AUC_{\text{intestinal loop}}}{AUC_{\text{i.v.}}}
\]

where \( AUC \) is the area under the plasma concentration–time curve.

Measurement of Unstirred Water Layer (UWL) Adjacent to Mucosal Surface The incubation chamber was similar to that used by Lukie et al. with a slight modification. The design and its dimensions are shown in our previous paper. The tissue was mounted between the circular platform and the retaining lid, with the mucosal surface of the tissue exposed to the mucosal incubation solution. The exposed surface area was 0.28 cm\(^2\) and the distance between the upper surface of the stirring bar and the tissue surface was kept at 2 cm. The rate of stirring was varied by changing the rotation rate of the magnetic stirrer.

The tissue was prepared as follows. Under sodium pentobarbital anesthesia, the upper half of the jejunum, the ileum, the upper half of the colon and the rectum (including the lower colon), approximately 6 cm in length, were removed and rinsed with saline. Each segment was then opened along the mesenteric border and the mucosal surface was blotted on a filter paper. An approximately 10 mm square piece was cut from the segment.

To estimate the effective resistance of UWL, the tissue uptake of decyl alcohol was measured according to the method of Thomson. The tissue specimen was mounted in the chamber and 1 ml of Krebs–bicarbonate buffer (pH 7.4) was added to the serosal compartment. The chamber was preincubated in the same buffer, which was previously oxygenated by a stream of 5% CO\(_2\)-95% O\(_2\) for 5 min at 37°C. Then the chamber was transferred to another beaker with 70 ml of the same buffer containing both \[^{14}C\]decyl alcohol (0.1 mm, 0.01 μCi/ml) and a trace amount of the radiolabeled volume marker, \[^{3}H\]dextran (1.25 μCi/ml). Each solution for preincubation and incubation was stirred at an identical stirring rate. The stirring rates are expressed as the revolutions per minute (rpm) at which the stirrer was driven. The uptake experiments were carried out for 6 min at 37°C. When the incubation was completed, the chamber was removed from the beaker and the exposed tissue was immediately rinsed with cold saline for approximately 5 s. The tissue was blotted on a filter paper and cut with a circular steel punch (5 mm i.d.). The tissue fragment was placed in a vial and saponified with 3 N KOH at 55°C for 3 h. After decolorization with 30% H\(_2\)O\(_2\) and neutralization with 3 N HCl, a scintillation fluid (ACS-II) was added. Radioactivity was determined by means of an external standardization technique to correct for the quenching. The rate of tissue uptake of \[^{14}C\]decyl alcohol, \( J_d \), was calculated after correcting the radioactivity originated in the adherent mucosal solution with \[^{3}H\]dextran. It has already been demonstrated that the rate of tissue uptake of decyl alcohol is limited by diffusion across UWL and, thus, the uptake rates at different stirring rates were used to estimate the effective resistance of UWL \( (R_w) \). According to Thomson, \( R_w \) can be calculated by the following equation:

\[
R_w = d(S_a \cdot D) = C_1 / J_d
\]

where \( d \) and \( S_a \) are the effective thickness and the effective surface area of UWL, respectively, \( D \) is the free diffusion coefficient of decyl alcohol and \( C_1 \) is the concentration of decyl alcohol in the bulk phase.

Analytical Methods i) PEG-600 and PEG-1000: These PEGs were determined according to the method of Tagesson et al. with a slight modification. An aliquot of the sample solution was extracted with chloroform. After shaking and centrifugation, the chloroform layer was sampled out and was evaporated to dryness. The residue was redisolved in 40% (v/v) methanol solution for PEG-600 and in 46% (v/v) methanol solution for PEG-1000. An aliquot of the sample solution was filtered through a 0.45 μm pore-size filter (Nihon Millipore Kogyo, Yonezawa, Japan), and was used as a sample for HPLC analysis. A high pressure liquid chromatograph (model...
570, Gasukuro Kogyo, Tokyo) equipped with a refractive index detector, ERC-7510 (Erma Optical Works, Tokyo, Japan) was operated at the detector temperature of 40 °C. The analytical column was a reversed-phase Shim-pack CLC-C8 (5.0 mm x 150 mm, Shimadzu). The mobile phase was composed of methanol–water (40:60 for PEG-600 and 46:54 for PEG-1000 by volume), and the flow rate was 1.8 ml/min. The molecular weight for each peak was identified by the mass spectroscopy measurement. The concentration of each molecular weight of PEG was quantified by the area of the corresponding peak.

ii) PEG-300: PEG-300 was determined by gas-liquid chromatography (GLC) according to the method of Chadwick et al.9,10 with a slight modification. An aliquot of the sample solution was extracted with isopropanol–chloroform (9:1 by volume). After shaking, anhydrous potassium carbonate was added to the mixture of aqueous sample and organic solvents, followed by shaking and centrifugation. An aliquot of the organic phase was evaporated to dryness and the residue was redissolved in acetic anhydride containing an internal standard (pentan-erythritol tetraacetate) by vigorous shaking, then left for 45 min at room temperature. The column for GLC was Silicone OV-1 (Gasukuro Kogyo). A flame ionization detector was used and nitrogen gas was used as the carrier gas. After conditioning, the sample was injected at a column temperature of 130 °C and a detector temperature of 350 °C. The column temperature was raised at the rate of 4 °C/min up to 300 °C. The molecular weight of each peak was identified by tetraethylene glycol as a standard. The concentration of each PEG was then calculated from the ratio of the peak area against that of the internal standard.

iii) Acylsalicylic Acids: To hydrolyze the esters to salicylic acid, the sample solution was acidified with 1 N HCl and was placed in a boiling water bath for 1 h (acetyl-) or 2 h (propionyl- and butyryl salicylic acids). After cooling, the resultant solution was extracted with chloroform. Then, salicylic acid in the organic phase was reextracted with 0.1 N NaOH solution, and salicylic acid in the final aqueous layer was determined spectrophotometrically at 295 nm.

iv) Acetaminophen and Indomethacin: The aqueous sample solution was filtered through a 0.45 μm pore-size filter (Nihon Millipore Kogyo) and these drugs were determined by HPLC. In the case of the plasma sample, acetonitrile for acetaminophen and methanol for indomethacin were added to the plasma, and the supernatant was filtered through a 0.45 μm pore-size filter (Nihon Millipore Kogyo). Drug concentrations in plasma were determined by the apparatus of HPLC described in the Determination of Lipophilic Index. For acetaminophen analysis, the mobile phase of methanol–0.1 M acetate buffer solution (pH 3.1) (20:80 by volume) was run at a flow rate of 0.9 ml/min and acetaminophen was detected at 241 nm. For indomethacin analysis, the mobile phase of methanol–water–acetic acid (80:20:1 by volume) was run at a flow rate of 0.9 ml/min and indomethacin was detected at 254 nm.

**Statistical Analysis**

Statistical analysis was carried out by a Student’s t-test.

**RESULTS**

Absorption of Different-Sized PEGs from Small and Large Intestines The absorption of different-sized PEGs from small- and large-intestinal loops is shown in Fig. 1. The extent of absorption of each component of PEGs declined as the molecular weight increased, and the absorption–molecular weight profiles in the small and the large intestines are clearly different. After small-intestinal loop administration of PEG-600, the absorption increased as the molecular weight decreased, and the absorption rate, about 40% in 1 h at a molecular weight of 414, was much higher than the large-intestinal absorption. In addition, when PEG-1000 was administered, the absorption was independent of the molecular size, and the absorption rate was lower than PEG-600: about 15—20% in 1 h. In contrast, PEG-600 administered into the large-intestinal loop was poorly absorbed, and the rate of absorption of each component was independent of the molecular weight, about 10% in 1 h. However, when PEG-300 was administered, the absorption rate increased as the molecular weight decreased: about 30% in 1 h at a molecular weight of 238. Accordingly, it is clear that PEGs with molecular weights larger than 300 is restricted from large-intestinal absorption, while larger molecular sized PEGs, with molecular weights below 600, can be absorbed in the small intestine.

**Relationship between Lipophilicity and Absorption of Acylsalicylic Acids in Different Sites of Intestine**

The lipophilic indexes, \( \log k'_{oc} \), for acylsalicylic acids, i.e. acetyl-, propionyl- and butyryl salicylic acid, at pH 7.4 were 0.778, 1.167 and 1.581, respectively. The absorption of these acylsalicylic acids at pH 7.4 from the jejunum, the ileum, the colon and the rectum, was examined. The absorption rates in the four sites are shown as a function of the lipophilic indices in Fig. 2. The absorption rate constant was calculated by assuming that the absorption is regarded as a first-order rate process. With respect to the site of intestine, the absorption increased in the order of the rectum < the colon < the ileum < the jejunum. In each site, the absorption rate constant increased as the acyl...
chain-length increased. Linear correlations between lipophility and the absorption rate constant are observed in the colon and the rectum, but the absorption rate constant tended to reach a ceiling at the high lipophility in the small intestine, i.e. in the jejunum and the ileum.

Absorption of Acetaminophen and Indomethacin in Different Sites of Intestine  To further clarify the different absorption–lipophility profiles in the small and the large intestines, the absorption of acetaminophen and indomethacin was examined using the loops of the jejunum, the ileum, the colon and the rectum. Plasma concentrations of the drugs were periodically determined after administration into each loop, and the results for acetaminophen and indomethacin are shown in Figs. 3 and 4, respectively. The model independent moment analysis was applied, and the pharmacokinetic parameters estimated for acetaminophen and indomethacin are listed in Tables I and II, respectively. In addition, the rates of absorption, estimated from the disappearance from the lumen of each segment, for acetaminophen and indomethacin, are summarized in Table III. With regard to both the MAT value estimated from the plasma concentration–time data and the rate of absorption estimated by the disappearance from the loop, the absorption rate of acetaminophen was greater in the small intestine than in the large intestine, while that of indomethacin was not significantly different among the sites of administration.

Estimation of Effective Resistance of UWI  The rate of [$^{14}$C]decyl alcohol uptake into the intestinal mucosa was examined in vitro and the results at stirring rates of both
Table III. Absorption of Acetaminophen and Indomethacin from Various Sites of Rat Intestine

<table>
<thead>
<tr>
<th>Site</th>
<th>% absorbed in 10 min</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Acetaminophen</td>
</tr>
<tr>
<td>Jejunum</td>
<td>59.3 ± 9.8</td>
</tr>
<tr>
<td>Ileum</td>
<td>51.5 ± 4.0</td>
</tr>
<tr>
<td>Colon</td>
<td>22.5 ± 5.5</td>
</tr>
<tr>
<td>Rectum</td>
<td>20.6 ± 0.9</td>
</tr>
</tbody>
</table>

Absorption was calculated from the amount of drug, which disappeared from the luminal solution of the loop (6 cm). Results are expressed as the mean ± S.E. of 3–4 experiments.

Table IV. Effect of Stirring of Mucosal Solution on Mucosal Uptake of [14C]Decyl Alcohol in Various Sites of Rat Intestine

<table>
<thead>
<tr>
<th>Site</th>
<th>Stirring rate (rpm)</th>
<th>Uptake rate (mmol/cm²/min)</th>
<th>Effective resistance (cm²·min/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Jejunum</td>
<td>0</td>
<td>0.12 ± 0.01</td>
<td>882 ± 82</td>
</tr>
<tr>
<td></td>
<td>600</td>
<td>2.93 ± 0.92</td>
<td>42 ± 13</td>
</tr>
<tr>
<td>Ileum</td>
<td>0</td>
<td>0.24 ± 0.01</td>
<td>421 ± 20</td>
</tr>
<tr>
<td></td>
<td>600</td>
<td>2.55 ± 0.70</td>
<td>56 ± 21</td>
</tr>
<tr>
<td>Colon</td>
<td>0</td>
<td>0.33 ± 0.06</td>
<td>315 ± 71</td>
</tr>
<tr>
<td></td>
<td>600</td>
<td>1.58 ± 0.69</td>
<td>89 ± 30</td>
</tr>
<tr>
<td>Rectum</td>
<td>0</td>
<td>0.35 ± 0.02</td>
<td>300 ± 18</td>
</tr>
<tr>
<td></td>
<td>600</td>
<td>1.24 ± 0.24</td>
<td>86 ± 15</td>
</tr>
</tbody>
</table>

Results are expressed as the mean ± S.E. of 3–5 experiments.

0 rpm and 600 rpm are summarized in Table IV. The rate of decyl alcohol uptake at a stirring rate of 600 rpm was greater than that at 0 rpm in each site of the intestine, and the rate of uptake at 600 rpm decreased as the site was more distal. Next, the effective resistance of UWL, $R_w$, adjacent to the mucosal surface was estimated. Contrary to the uptake rates, $R_w$ values were reduced with 600 rpm stirring in the respective intestinal segments.

Discussion

Physicochemical properties of a drug which govern its large-intestinal absorption were investigated in rats. At first, the absorption–molecular weight profile in the large intestine was examined in comparison with that in the small intestine. Chardwick et al. introduced low molecular weight PEGs as ideal probe molecules for measuring intestinal permeability; PEGs are inert, water-soluble molecules of varying sizes, nontoxic, not degraded by intestinal bacteria, not metabolized after absorption, and rapidly excreted in the urine.3,9,10 The different-sized PEGs cross the intestinal epithelium at different rates, allowing for the characterization of passive permeability properties of the mucosa. Using PEG-600, Olaison et al. described that the intestinal absorption in Crohn’s disease was greater than in cancer patients, and was similar throughout the weight range, whereas in the cancer patients absorption was inversely proportional to the molecular weight.11 As is evident from Fig. 1, the extent of absorption of each PEG component declines as molecular weight increases, and differences in the absorption–molecular weight profiles between the small and the large intestines can be clearly detected. When PEG-600 was administered in the small-intestinal loop, absorption increased as molecular weight decreased, and the absorption rate, about 40% in 1 h at the molecular weight of 414, was much higher than the rate of large-intestinal absorption. In addition, when PEG-1000 was administered, the absorption of each component was independent of molecular size, and the absorption rate became lower than that of PEG-600, about 20% in 1 h at molecular weights above 634. By contrast, when PEG-600 was administered in the large-intestinal loop, it was poorly absorbed, and the absorption rate of each component of PEG-600 was independent of the molecular weight. However, when PEG-300 was administered, the absorption rate increased as the molecular weight decreased, and it became greater, about 30% in 1 h, at the molecular weight of 238. Accordingly, it is clear that the absorption–molecular weight profile was different between the small and the large intestines; the large-intestinal absorption of PEGs with molecular weights larger than 300 was poor, while PEGs with molecular weights up to 600 were relatively well absorbed in the small intestine.

There are two routes in the transepithelial drug transport by passive diffusion mechanism: the transcellular route through the lipoidal cell membrane and the paracellular route through the tight junction to the lateral intercellular space. Hollander et al. showed using PEG-900 that the transport of PEG takes place mostly through the tight paracellular junctions and that transport directly cross cell membranes is minimal.12 Thus, it can be assumed that the paracellular route contributes to the small-intestinal absorption of PEG-600 and large-intestinal absorption of PEG-300. The paracellular shunt path resistance was about 45 Ω·cm² in rat jejunum13 and around 100 Ω·cm² in rat colon (unpublished data) as calculated in our electrophysiological work. Accordingly, it appears that it is difficult for larger sized PEGs to penetrate the large-intestinal mucosa in comparison with the small-intestinal mucosa. Usually, the contents in the lumen are solid in the large intestine but are liquefied when they reach the small intestine. Thus, it can be considered that the tight junction in the large intestine must be tighter against physical pressure, and that the tight epithelia function as a defense against toxic substances produced by the fermentation of luminal contents in the large intestine.

The absorption–lipophilicity profiles were also examined in various regions (loops of 6 cm) of rat intestine using three acylsalicylic acids, acetyl-, propionyl- and butyrylsalicylic acid. The absorption rates of the three acylsalicylic acids were different in the intestinal regions: the jejunum > the ileum > the colon > the rectum. In each region, the absorption rate increased with the lipophilicity of the drug. However, it was shown that the absorption rates in the small intestine tended to reach a ceiling at the high lipophilicity. Bridges et al. investigated the gastrointestinal absorption of a homologous series of alkylcarbamates in rats using an in situ technique.14,15 The absorption rate constants in the colon increased with increasing lipophilicity.15 On the other hand, an increase in the small-intestinal absorption rate constant was
observed when the alkyl chain length of a carbamate was increased from methyl to n-butyl, but a further increase in chain length resulted in a fall in the absorption rate constant.\textsuperscript{14} As a result, in the case of highly lipophilic derivatives, the absorption rate constants in the small and large intestines showed similar values. The absorption–lipophilicity profiles shown in Fig. 2 seem to be a part of this example.

As to the regional variation of the absorption, the difference in the mucosal surface area along the rat intestinal tract was considered to be one of the causes; there are large variations in the structure of the epithelial surface along the intestine. Although in the duodenum the epithelium forms many plicae circulares and villi, which enlarge the epithelial surface, the villi decrease towards the anus. Thus, the epithelial surface is decreasing from the pylorus towards the anus, suggesting that the large intestine has less favorable characteristics for drug absorption. This site-dependent absorption profile could also be caused by differences in the lipid composition along the intestinal tract. Varying lipid fluidity of brush-border membranes prepared from colonocytes\textsuperscript{16} and enterocytes\textsuperscript{17} has been demonstrated. The decrease in fluidity of the brush border membrane of the large intestine in comparison with that of the small intestine resulted from an increase in cholesterol content, cholesterol/phospholipid molar ratio and degree of saturation of the fatty acid residues in the large-intestinal membrane. Accordingly, the difference in surface area and/or the fluidity of the lipid membrane in the intestinal segments would be able to explain the regional variation of the absorption.

To confirm this absorption–lipophilicity-site relation, the site dependency of indomethacin absorption (Fig. 4, Tables II and III) was compared with that of acetaminophen (Fig. 3, Tables I and III). Acetaminophen (lipophilic index, 0.67) is less lipophilic than acetylsalicylic acid, while indomethacin (lipophilic index, 3.1) is a highly lipophilic drug. The \textit{MAT} from the plasma concentration–time curve for acetaminophen was shortened in the order of the rectum > the colon > the ileum > the jejunum, and the absorption rate calculated from the disappearance from the lumen decreased as the site going approached the anus. Since the experimental values are scattered and the absorption is relatively rapid, \textit{MRT} values are not significantly different between the i.v. injection and each intra-luminal administration (Table I). However, the tendency is clear. Thus, the following discussion uses \textit{MAT} values calculated from each mean \textit{MRT} value. It is known that acetaminophen is eliminated by first-pass metabolism, mainly by sulfation in the intestine and/or in the liver, following oral administration in rats.\textsuperscript{18} However, since first-pass metabolism is only around 10% at the high dose,\textsuperscript{18} as in this experiment, it may be all right to consider that the values of \textit{MAT} were not as affected by first-pass metabolism. Thus, neither the \textit{MAT} value nor the absorption rate of indomethacin varied in any of the intestinal segments. Acetaminophen showed site-dependent absorption which was similar to acetylsalicylic acids, and it can be considered that this result is caused by the same factors as mentioned above, \textit{i.e.}, the effective surface area and the lipid fluidity. On the other hand, indomethacin showed site-independent absorption, and was not favored in its absorption by the enlargement of the surface.

In order to clarify this connection, the mucosal uptake rate of \textsuperscript{14}C\textit{decyl alcohol was determined in vitro and the value of the effective resistance of UWL was calculated (Table IV). We have examined the effect of stirring rate on the rate of \textsuperscript{14}C\textit{lauryl alcohol uptake into the hamster cheek pouch and showed that the uptake rate appeared to become constant when the stirring rate was increased higher than 400 rpm.\textsuperscript{23} Thus, present experiment was carried out at 600 rpm, at which time the uptake rate becomes constant. When the uptake rate, \textit{J}_d, is measured for decyl alcohol, whose uptake is totally limited by UWL, the experimentally determined \textit{J}_d can be expressed as follows:

\[
\textit{J}_d = \frac{C_i \cdot S_w \cdot D}{d}
\]

The values of \textit{C}_i and \textit{D} can be fixed to be identical in a series of experiments, and the ratio of the uptake rates at 0 rpm in different sites means the ratio of the \textit{S}_w/\textit{d} values. Thus, since it can be presumed that the effect of UWL is negligible at 600 rpm, the \textit{d} value was regarded as constant at 600 rpm, and the difference in uptake rates originates from the difference in \textit{S}_w values.

By considering the above, the relative surface area (\textit{S}) on the basis of the rectum (\textit{S} = 1) in a respective intestinal segment is calculated by the following equation:

\[
\textit{S} = \frac{R_{600,i}}{R_{600,1}}
\]

where \textit{R}_{600,i} and \textit{R}_{600,1} are the effective resistance at a stirring rate of 600 rpm for the rectum and for the other intestinal segments, respectively. The \textit{S} values thus calculated are 2.1, 1.5 and 1.0 for the jejunum, the ileum and the colon, respectively. The larger \textit{S} values in the jejunum and the ileum in comparison with those in the large intestine are probably due to the existence of the plicae circulares and the villi. This result agrees extremely well with the relative area per unit length of rat intestines reported by Permezel and Weibling.\textsuperscript{19}

Secondly, the relative thickness of UWL (\textit{T}) on the basis of the rectum (\textit{T} = 1) is calculated using \textit{S} values by the following equation:

\[
\textit{T} = \frac{S \cdot R_{0,i}}{R_{0,1}}
\]

where \textit{R}_{0,i} and \textit{R}_{0,1} are the effective resistance at 0 rpm for the rectum and for the other intestinal segments, respectively. The \textit{T} values thus calculated are 6.1, 2.2 and 1.0 for the jejunum, the ileum and the colon, respectively. The relative thickness of UWL in the jejunum was about 6-fold, and that in ileum was about twice as large as that in the large intestine. Moreover, the thickness of UWL was nearly identical in the large intestine.

These results suggest that the barrier to small-intestinal absorption of highly lipophilic drugs, such as butyrylsalicylic acid and indomethacin, is the UWL rather than the epithelial wall. The pH–absorption profile in various parts of rat intestine has been investigated with salicylic acid and so on.\textsuperscript{20} The absorption from the rectum followed the pH-partition theory, while the absorption from the small intestine was incompatible with the theory.
Although this result was interpreted by the binding process to the mucosal surface, the UWL is also considered as an important factor in this phenomenon.

As we first mentioned, the large intestine is considered to be a favorable site for peptide absorption because of a lack of peptidases in the luminal content. However, large polar drugs like peptide drugs do not seem to penetrate the large-intestinal membrane easily. Thus, enhancers for drug penetration across the large-intestinal membrane have been extensively studied. On the other hand, as to the bioavailability of a drug in sustained release dosage forms, the absorption of moderately lipophilic drugs would be poor in the large intestine, while highly lipophilic drugs seems to be absorbed in the large intestine as well as in the small intestine. However, extremely lipophilic drugs, such as d-α-tocopherol acetate, could not be absorbed from the large intestine (unpublished data).

It is known that tocopherol is absorbed from the small intestine through the lymph system by using chylomicron as the carrier, but the chylomicron cannot be produced in the large-intestinal epithelial cells. This may be the reason for the absence of absorption of tocopherol in the large intestine.

In conclusion, the influence of the physicochemical factors of a drug on large-intestinal absorption differs substantially from factors involved in the small-intestinal absorption. The differences might be caused by morphological differences between the small and the large intestines: the tighter tight junction and thinner UWL adjacent to the mucosal surface in the large intestine. Studies of physiological factors, such as the transit rate along the gastrointestinal tract and the amount of intestinal juice, as well as the capacity for chylomicron formation, governing large-intestinal drug absorption, are in progress.

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