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Effect of Nonsteroidal Anti-inflammatory Drugs on the Permeability of the Rectal Mucosa¹⁾

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The effect of nonsteroidal anti-inflammatory drugs (NSAIDs) on the permeability to marker drugs, sulfanilic acid and creatinine, was investigated in rat rectum using the *in situ* perfusion technique. Indomethacin (IM), phenylbutanone (PB), diclofenac sodium (DF), and aspirin (ASA) were used as NSAIDs. The permeability of rectal mucosa to the marker drugs was increased by NSAIDs. The permeability-enhancing effect increased in the following order: ASA < PB < DF < IM. The effects were reversible. No marked histological changes were observed and, furthermore, protein release from the rectal mucosa was not increased. On the other hand, there was a good correlation between the permeability-enhancing effect and the NSAID amount accumulated in the rectum. Consequently, it is suggested that the accumulation of NSAID in the rectum plays an important role in the enhancement of the rectal permeability to drugs.

Keywords—nonsteroidal anti-inflammatory drug; sulfanilic acid; creatinine; rectal permeability; accumulation; enhancing effect; histological change

In our previous papers of this series,²⁻⁶⁾ we have shown that surfactants, EDTA, and polyethylene glycol (PEG) 400 enhance the rectal permeability to marker drugs, sulfaguanidine and sulfanilic acid, and cause histological changes in the rectum.²⁾ It was considered that membrane permeability is influenced by the interaction between the adjuvant and protein or lipid components in the rectal membrane.

Nonsteroidal anti-inflammatory drugs (NSAIDs) bind strongly to serum albumin in the circulation and induce gastrointestinal mucosal irritation after oral administration.⁷⁻¹¹⁾ Recently, rectal administration has been studied as a delivery route to overcome the problem of mucosal irritation. Yaginuma¹²⁾ and Nishihata¹³⁾ have shown that some NSAIDs promote the absorption of drugs from the rectum, and that the promoting effects may depend on the extent of binding of the promoters to the rectal tissue.¹³⁾ However the direct cause of the promoting effect is not yet fully understood.

In this study, the effect of NSAIDs, indomethacin (IM), phenylbutazone (PB), diclofenac sodium (DF), and aspirin (ASA) on the rectal permeability was investigated in order to elucidate the mechanism of this action, with sulfanilic acid and creatinine as marker drugs.

Experimental

Materials—IM (Medicel Research Laboratory, Japan), PB (Sigma, U.S.A.), DF (Kodama Pharmaceutical Co., Ltd., Japan), ASA (Ebisu Pharmaceutical Co., Ltd., Japan), sulfanilic acid (SA), and creatinine (CR) (Tokyo Kasei Co., Ltd., Japan) were used as supplied. All other reagents used in these experiments were of the finest grade available.

Preparation of Drug Solution—The marker drug (SA: final concentration, 3 mg/ml) was dissolved in isotonic phosphate buffer solution (pH 7.4) and NSAIDs were added at various concentrations.

Absorption Experiments—Male Wistar rats (180–250 g) were used in all experiments. The absorption of SA from the rectum was measured by the *in situ* single perfusion technique as described previously.²⁾ The drug solution

was perfused through the whole rectum at a rate of 20 ml/15 min. Blood samples (0.3 ml) were taken just before the start of the perfusion and at intervals of 15 min thereafter. SA concentration in the blood was determined.

Exsorption Experiments (The Determination of Apparent Rectal Clearance (ARC))—The exsorption of CR to the rectal lumen was examined by the method of Kakemi *et al.*¹⁴⁾ Absorption and exsorption experiments were carried out simultaneously. The rectum was perfused with the drug solution at a rate of 20 ml/15 min, and at 7.5 min, CR (50 mg in 0.5 ml saline solution) was administered intravenously to the rat through the jugular vein. Fractions of 20 ml of the perfusate were collected continuously after CR administration. The amounts of CR in the perfusate were determined. Blood samples were taken in the same way as in the absorption experiments, and CR concentration in the blood was determined. ARC was calculated as follows:

$$ARC \text{ (ml/min)} = \frac{\text{amount of CR excreted in the perfusion media in 15 min/15}}{\text{blood concentration of CR at an intermediate time}} \quad (1)$$

Elution of Protein from Rectal Mucosa—The elution of proteins from the rectal mucosa was estimated by determining the protein concentration in the perfusate in the exsorption experiments described above.

Histological Examination—Immediately after the perfusion experiments, rectal tissues were surgically excised and washed with cold saline solution. The excised tissues were then fixed with 10% formaldehyde in saline solution and cut into slices. The slices were stained with hematoxylin–eosin solution. The tissues were observed under an optical microscope.

Surface Tension Measurement—The surface tension of the drug solution was measured at 25 °C by using a Du Nöuy tensiometer (Shimadzu, Japan).

Apparent Partition Coefficient—The apparent partition coefficient was determined by the method of Suzuki *et al.*¹⁵⁾

Accumulation of NSAID in the Rectal Membrane—Immediately after the perfusion experiments, the rectal lumen was washed with 20 ml of cold saline solution. The rectum was carefully resected and blotted with a filter paper to remove adhering moisture, and the wet weight was measured. The tissue was homogenized in a Potter-type homogenizer with saline solution and the volume of the homogenate was adjusted to 5 ml. NSAID contents were determined in both the homogenate and the washing solution (20 ml).

Analytical Methods—(a) SA and CR were determined spectrophotometrically by the method described previously.³⁾ (b) The protein concentration was determined by the method of Bradford¹⁶⁾ with bovine serum albumin as the standard. (c) NSAID: IM and ASA were determined fluorometrically by the method of Hukker *et al.*¹⁷⁾ and Schachter *et al.*,¹⁸⁾ respectively. PB was determined spectrophotometrically by the method of Wallance.¹⁹⁾ DF was determined by high performance liquid chromatography (HPLC). A known amount of *N*-phenylanthranilic acid was added to the sample solution as an internal standard and was extracted with benzene after acidification with 1 N HCl. The organic layer was evaporated to dryness. The residue was redissolved in distilled water and the aliquot was applied to HPLC. HPLC (JASCO FAMILIC-100, JASCO, Japan) equipped with a variable-wavelength ultraviolet (UV) detector (JASCO UVIDEC-100 II, JASCO) was used in a reversed phase mode with a stationary phase of μ -Finepak SIL C₁₈ (JASCO) packed in 0.5 mm i.d. $\times$ 200 mm tubing and operated at ambient temperature. A mixture of acetonitrile–water (1 : 1, v/v) containing 0.2% phosphoric acid was used as the mobile phase at a flow rate of 16 μ l/min. The effluent was monitored by measurement of the UV absorption at 282 nm. The drug concentration was determined from the peak height by using the calibration curve.

Results

Effect of NSAIDs on SA Absorption

Figure 1 shows the blood concentration–time curves of SA, illustrating the effects of IM (a), DF (b), PB (c), and ASA (c) on the absorption of SA in the rat rectum. As is evident from the figure, the absorption of SA was significantly increased by the presence of these NSAIDs. The extent of the increase was dependent on both NSAID level in the perfusate and the exposure period of the rectal mucosa to the NSAID. For instance, while the absorption of SA was influenced neither by NSAIDs at low perfusate level (2 mM) nor by a 15 min exposure even at high NSAIDs level, a significant increase was observed in the case of high NSAIDs level (10 mM) and 90 min perfusion. The degree of SA absorption was examined by comparing the area under the concentration–time curve during 90 min perfusion, AUC_{90} . The maximum increase was observed in the case of 10 mM IM. The enhancing effect increased in the following order: ASA < PB < DF < IM. The addition of 10 mM IM showed a fifty-fold increase in AUC_{90} of SA as compared with the control value (Table I).

Fig. 1. Effect of NSAIDs on the Absorption of Sulfanilic Acid

(a) indomethacin: —■—, 2 mM; —▲—, 5 mM; —●—, 10 mM; —○—, control.
 (b) diclofenac sodium: —■—, 2 mM; —▲—, 5 mM; —●—, 10 mM; —○—, control.
 (c) phenylbutazone: —△—, 5 mM; —▲—, 10 mM; —○—, control. aspirin: —□—, 80 mM; —■—, 200 mM.

Each point represents the mean value of 3 to 5 rats with the standard error shown as a bar.

TABLE I. Effect of NSAIDs on *AUC* of Sulfanilic Acid and the Average Apparent Rectal Clearance of Creatinine

NSAID	Concn. (mM)	<i>AUC</i> ± S.E. ^{a)}	Ratio ^{b)}	<i>ARC</i> ± S.E. ^{c)}	Ratio ^{d)}
Control		61 ± 9	1	1.16 ± 0.2	1
Indomethacin	2	276 ± 39	4.5	5.53 ± 0.5	4.8
	5	1844 ± 119	30.2	28.80 ± 2.4	26.6
	10	3100 ± 178	51.0	39.40 ± 1.5	33.9
Phenylbutazone	5	91 ± 11	1.5	2.40 ± 0.6	2.1
	10	506 ± 73	8.3	9.59 ± 1.5	8.3
Diclofenac sodium	2	145 ± 21	2.4	2.36 ± 0.3	2.0
	5	498 ± 58	8.2	8.31 ± 1.1	7.2
	10	1220 ± 113	21.7	15.50 ± 2.2	13.4
Aspirin	80	43 ± 7	0.7	1.35 ± 0.3	1.2
	200	499 ± 67	8.2	4.47 ± 0.4	3.9

a) $\mu\text{g/ml} \cdot 90 \text{ min}$.

b) $\frac{\text{AUC of sulfanilic acid with NSAID}}{\text{AUC of sulfanilic acid without NSAID}}$.

c) Apparent rectal clearance; 10^{-3} ml/min .

d) $\frac{\text{average apparent rectal clearance of creatinine with NSAID}}{\text{average apparent rectal clearance of creatinine without NSAID}}$.

Effect of NSAIDs on CR Exsorption

Figure 2 shows the *ARC*–time curve of CR, illustrating the effects of NSAID, IM (a), DF (b), PB (c), and ASA (c), on the exsorption of CR to the rectal lumen. The exsorption of CR was significantly increased by adding NSAID. The increase of *ARC* was dependent on the perfusate levels of NSAID. Interestingly, it was found that the clearance shows a ceiling effect at 5 and 10 mM IM and at 10 mM DF. The maximum increase in the *ARC* of CR over 90 min (average *ARC*; *ARC*₉₀) was observed in the case of 10 mM IM. The order of magnitude of the enhancing effect was similar to that in the case of SA absorption (Table I).

Fig. 2. Effect of NSAIDs on the Apparent Rectal Clearance of Creatinine

- (a) indomethacin: —■—, 2 mM; —▲—, 5 mM; —●—, 10 mM; —○—, control.
 (b) diclofenac sodium: —■—, 2 mM; —▲—, 5 mM; —●—, 10 mM; —○—, control.
 (c) phenylbutazone: —△—, 5 mM; —▲—, 10 mM; —○—, control. aspirin: —□—, 80 mM; —■—, 200 mM.

Each point represents the mean value of 3 to 5 rats with the standard error shown as a bar.

Fig. 3. Effect of NSAIDs on the Ratio of Apparent Absorption Rate of Sulfanilic Acid

- (a) indomethacin: —■—, 2 mM; —▲—, 5 mM; —●—, 10 mM.
 (b) diclofenac sodium: —■—, 2 mM; —▲—, 5 mM; —●—, 10 mM.
 (c) phenylbutazone: —△—, 5 mM; —▲—, 10 mM. aspirin: —□—, 80 mM; —■—, 200 mM.

$$\text{ratio of apparent absorption rate} = \frac{\text{apparent absorption rate with NSAID}}{\text{apparent absorption rate without NSAID}}$$

Ratio of Apparent Absorption Rate (RAAR) and Ratio of Apparent Permeation Rate (RAPR)

RAAR was determined by the method described previously.²⁻⁴⁾

$$\text{control: } C_0 \xrightarrow{k_c} C_c \xrightarrow{K} C_c = \frac{k_c}{VK} (1 - e^{-Kt}) \quad (2)$$

$$\text{sample: } C_0 \xrightarrow{k_s} C_s \xrightarrow{K} C_s = \frac{k_s}{VK} (1 - e^{-Kt}) \quad (3)$$

Where V (ml) is the volume of distribution, K (min^{-1}) is the elimination rate constant (first-order), C_0 ($\mu\text{g/ml}$) is the concentration of SA in the perfusate (constant), the subscripts c and s specify the perfusate without and with NSAID, respectively, and k 's (k_c and k_s , $\mu\text{g/ml}$) and C 's (C_0 , C_c and C_s , $\mu\text{g/ml}$) are the absorption rate constant (zero-order) and the blood concentration of SA at the time t specified by the subscripts, respectively. By using Eqs. 2 and 3, RAAR may be written as follows:

$$k_s/k_c = C_s/C_c$$

(4)

namely, $RAAR$ (k_s/k_c) can be represented as the ratio of blood concentration (C_s/C_c) of SA at time t . Figure 3 shows $RAAR$ -time curve of SA; the pattern is similar to that of the ARC -time curve of CR (Fig. 2). That is to say, the rectal permeability changed in the same manner with respect to absorption and exsorption. Therefore, $RAPR$ ²⁾ ($RAAR/RARC$; ratio of apparent absorption rate (k_s/k_c)/ratio of apparent rectal clearance (CL_s/CL_c)) may be constant in all cases.

Histological Changes and Release of Protein

The results of microscopic and histological observation are presented in Fig. 4 and Table II, respectively. In the control group, normal and clear images of epithelial cells, the crypt of goblet cells, and principal cells were observed. In the groups treated with NSAIDs, a slight deficiency of epithelial cells was observed at the luminal border, but there was no change in the crypt region or the submucosal layer. When the histological change in the rectal mucosa was scored by the method of Bhuta *et al.*,²⁰⁾ the disappearance of the epithelial cells amounted to at most about 20% after the treatment with 10 mM IM, and that of goblet cells amounted to about 35% in all cases during the perfusing experiment. Table III shows the amount of protein released from the rectum into the perfusate in 90 min. In the previous paper,²⁾ we showed that there is a good correlation between AUC_{90} of a marker drug and amount of protein released by sodium deoxycholate (SDC), sodium lauryl sulfate (SLS), EDTA, PEG 400, and Tween 80 at various perfusate levels. Although AUC_{90} and ARC_{90} for marker drugs were markedly increased by NSAIDs, only a small amount of protein was released by NSAIDs. The greatest

Fig. 4. Microphotographs of the Rectal Lumen

A, isotonic phosphate buffer; B, 10 mM indomethacin; C, 10 mM phenylbutazone; D, 10 mM diclofenac sodium; E, 200 mM aspirin ($\times 300$).

TABLE II. Histological Study of NSAID Effect on the Structural Integrity of the Rectal Membrane

NSAID	Concn. (mM)	Presence of epithelial cells	Presence of goblet cells	Edema of lamina propria	Change in mucous membrane
Control		100 (%)	100 (%)	—	—
Indomethacin	2	100	70	++	+
	5	95	60	+	+
	10	80	80	+	+
Phenylbutazone	5	100	60	—	+
	10	100	60	—	+
Diclofenac	2	95	70	++	+
sodium	5	100	60	++	+
	10	70	60	+	+
Aspirin	80	90	60	+	—
	200	80	60	+	—

Key: —, normal; +, slight and ++, moderate.

TABLE III. Effect of NSAID on the Release of Protein from the Rat Rectum

NSAID	Concn. (mM)	Protein (mg)/90 min
Control		0.75 ± 0.21
Indomethacin	10	4.11 ± 1.52
Phenylbutazone	10	8.10 ± 2.37
Diclofenac sodium	10	2.28 ± 0.98
Aspirin	200	3.10 ± 1.10

Each result is the mean value ± S.E. of 3 to 5 rats.

protein release (8.1 mg) was that produced by 10 mM PB among the NSAIDs. There was no correlation between AUC_{90} of the marker drug and the amount of protein released in the presence of NSAID.

Surface Tension

One of the physico-chemical properties of NSAIDs that may affect the rectal permeability is surface activity. Figure 5 shows the surface tension values of NSAID solutions. The values of surface tension were similar within the concentration range of 0.01–0.1 mM, but decreased gradually at higher perfusate levels of NSAID. The surface tension of Tween 80 solution was much less than those of NSAID solutions. However, the permeability enhancing effect of NSAID was larger than that of Tween 80. Thus, there appears to be no correlation between surface tension and the magnitude of the permeability enhancement.

Apparent Partition Coefficients of NSAIDs and SA

The apparent partition coefficients of NSAIDs and SA used in this study are summarized in Table IV. Although NSAIDs exist in ionic form in pH 7.4 isotonic phosphate buffer, the apparent partition coefficients of NSAIDs to organic solvents were high and the NSAIDs are clearly lipophilic. On the other hand, the apparent partition coefficient of SA to organic solvents was extremely low. The absorption and partition coefficient of SA may be changed from that of free SA, if an NSAID–SA complex is formed. However, in fact the apparent partition coefficient of SA was found to be the same in the presence or absence of NSAID.

Fig. 5. Surface Tension of NSAID and Tween 80 Aqueous Solutions

—○—, indomethacin; —●—, phenylbutazone;
—△—, diclofenac sodium; —□—, Tween 80.

NSAID dissolved in pH 7.4 isotonic phosphate buffer.

The surface tension of pH 7.4 phosphate buffer was 71.5 dyn/cm.

TABLE IV. Apparent Partition Coefficients of NSAIDs, Sulfanilic Acid and Creatinine

Drugs	CHCl ₃ /buffer ^{a)}	Isoamyl alcohol/buffer	Cyclohexane/buffer
Indomethacin	7.82	15.6	0.01
Phenylbutazone	605.0	19.7	1.94
Diclofenac sodium	2.21	16.5	0.02
Aspirin	0.001	—	0.001
Sulfanilic acid	0.001	0.001	0.001
Sulfanilic acid with NSAID	0.001	0.001	0.001
Creatinine	0.03	0.11	0.03
Creatinine with NSAID	0.03	0.11	0.04

a) pH 7.4 phosphate buffer. Drug concentration was used 0.5 mM.

Thus, the effect of NSAID on the permeability of the rectal membrane to marker drugs is considered to be mainly due to their direct action on the rectal membrane components.

Accumulation of NSAID

To clarify the mechanism of this acceleration effect, the accumulated amount of NSAID in the rectal membrane was determined after the perfusion experiments. As shown in Fig. 6, there is a good correlation between the amount of NSAID accumulated and ARC_{90} of CR values (IM, $r=0.9899$, $p<0.01$; DF, $r=0.9039$, $p<0.01$; PB, $r=0.9600$, $p<0.01$; ASA, $r=0.9879$, $p<0.01$).

Figure 7 shows the accumulated amount–time curve of IM; the pattern is similar to that of the ARC –time curve of CR (Fig. 2 (a)). Thus, the change of rectal permeability is closely related to the amount of NSAID accumulated.

Recovery Experiments

To confirm that the bidirectional permeability to marker drugs was enhanced by the direct action of NSAID on the rectal membrane, the perfusion solution was changed from a marker drug solution containing NSAID to the control solution at 45 min. The results are shown in Fig. 8. As the perfusion solution was changed to the control solution, ARC of CR fell gradually to the control level. The accumulated amount of NSAID in the rectal membrane

Fig. 6. Relationship between Apparent Rectal Clearance of Creatinine and Accumulated NSAID in the Rectal Tissue

Indomethacin: ●, 2 mM; ○, 5 mM; ●, 10 mM ($r=0.9899$).
 Phenylbutazone: ▲, 5 mM; △, 10 mM; ▲, 15 mM ($r=0.9600$).
 Diclofenac sodium: ■, 2 mM; □, 5 mM; ■, 10 mM ($r=0.9039$).
 Aspirin: ▽, 80 mM; ▼, 200 mM ($r=0.9879$).

Fig. 7. Plot of Indomethacin Accumulated in the Rat Rectal Tissue *versus* Time

Each point represents the mean value of 3 rats with the standard error shown as a bar.

Fig. 8. Effect of Pretreatment with NSAIDs for 45 min on the Apparent Rectal Clearance of Creatinine

—○—, control; —●—, 10 mM indomethacin; —△—, 10 mM phenylbutazone; —▲—, 10 mM diclofenac sodium; —■—, 200 mM aspirin.
 Each point represents the mean value of 3 rats with the standard error shown as a bar.

decreased similarly. These results suggest that the enhancement of the permeability is reversible, and again correlated well with the accumulation of NSAID in the rectal membrane.

Discussion

The effect of NSAID on the permeability of the rectal membrane was evaluated. Changes in the bidirectional membrane permeability to marker drugs (rectal lumen $\rightleftharpoons$ blood) were determined simultaneously. The bidirectional membrane permeability to the drugs was increased by four NSAIDs tested and the effects were in the following order: IM > DF > PB > ASA.

Under the present experimental conditions, two mechanisms by which the permeability to marker drugs may be altered can be considered. A change in the physico-chemical properties of the marker drug due to the presence of NSAID, an intraluminal effect, could enhance or reduce the permeation rate, as could an alteration in the permeability of the membrane itself. However, SA and CR apparently have little physico-chemical interaction with NSAIDs, since SA and CR have low partition coefficients to organic solvents at pH 7.4 regardless of the presence of NSAID. Also, there was no correlation between surface tension and the permeability enhancement. Therefore, the enhancement of the permeability to marker drugs is considered to be due to the direct action of NSAID on rectal membrane components, not to physico-chemical interaction between NSAIDs and marker drugs.

Our previous report²⁾ showed that marked histological changes and significant protein release were induced by surfactants, a chelating agent, and PEG 400, leading to changes of the rectal membrane permeability. However, in spite of a general increase in the membrane permeability by NSAIDs, the histological changes and the protein release were much less than those produced by the adjuvants tested previously.²⁾ Furthermore, in contrast to the nature of the permeability enhancement by those adjuvants, which was irreversible,⁴⁾ the effects of NSAIDs were reversible. Consequently, the observed enhancement of the permeability by NSAID could not be explained in terms of solubilization of membrane components or histological changes caused by the adjuvants.^{2,21,22)} It is well known that mucosal irritation occurs on both oral and rectal suppository administration of NSAID. Yaginuma *et al.* demonstrated that the NSAID-induced enhancement of the permeation of antibiotics through the rectal membrane is closely related to irritation of the rectal mucosa.¹²⁾ Our results, however, showed that the rectal mucous membrane was irritated rather little by NSAID. The conflicting results might be due to differences in the experimental conditions and in the concentration of NSAIDs.

Nishihata *et al.* reported a correlation between the extent of binding of salicylate analogues to the rectal tissue and the absorption promoting effect.¹³⁾ To determine what caused the enhancement of permeability of the rectal membrane by NSAIDs, we investigated the accumulation of NSAIDs in the rectal membrane. As shown in Fig. 6, a good correlation between ARC_{90} of CR and the accumulated amount of NSAID in the rectum was demonstrated. Further, the accumulated amount-time curve of IM (Fig. 7) and the ARC of CR-time curve (Fig. 2(a)) showed similar patterns. The enhancing effect of NSAID on the rectal permeability was also reversible. Thus, NSAID accumulation in the rectal tissue appears to be important for the enhancement of rectal permeability to marker drugs under the conditions of this study.

In order to clarify whether the enhanced membrane permeability was caused by the accumulation of NSAID or not, we investigated the effect of simultaneous treatment with combinations of NSAIDs on the rectal clearance of CR. As shown in Fig. 9 and Table V, the values of enhanced permeability and accumulated amount of NSAID were not equal to the sum of those of the individual NSAIDs. However, it was found that there was a good agreement between the calculated enhancement effect based on the accumulated amount of individual NSAIDs (from Fig. 6) and those actually determined in this experiment. Thus, it appears that the accumulation of NSAID is necessary for the facilitation of rectal membrane

Fig. 9. Effect of Simultaneous Treatment with Indomethacin and Phenylbutazone or with Phenylbutazone and Diclofenac Sodium on the Apparent Rectal Clearance of Creatinine

—●—, perfused with indomethacin (5mM) and phenylbutazone (10mM); —▲—, perfused with phenylbutazone (10mM) and diclofenac sodium (10mM); ---○---, perfused with indomethacin (5mM) alone; ---□---, perfused with phenylbutazone (10mM) alone; ---△---, perfused with diclofenac sodium (10mM) alone.
Each point represents the mean value of 3 rats with the standard error shown as a bar.

TABLE V. Relationship between Apparent Rectal Clearance of Creatinine and Amount of NSAID Accumulated in the Rectal Membrane

Treatment	NSAID	Accumulated amount ($\mu\text{g/g}$)	Clearance (10^{-3} ml/min)	
			Measured	Calculated
5 mM indomethacin and 10 mM phenylbutazone			21.9 ± 2.0	19.10
	Indomethacin	415 ± 51	—	9.93
	Phenylbutazone	469 ± 23	—	9.17
10 mM diclofenac sodium and 10 mM phenylbutazone			22.0 ± 0.9	21.51
	Diclofenac sodium	427 ± 41		15.22
	Phenylbutazone	368 ± 57		6.29
5 mM indomethacin 10 mM phenylbutazone 10 mM diclofenac sodium	Indomethacin	890 ± 92	30.9 ± 2.3	—
	Phenylbutazone	550 ± 71	16.1 ± 1.1	—
	Diclofenac sodium	568 ± 73	20.2 ± 1.3	—

permeability to marker drugs. Additional studies are in progress to clarify the mechanism of alteration of membrane permeability by NSAIDs.

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