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Randomized clinical comparisons of diclofenac concentration in the soft tissues and blood plasma between topical and oral applications

Shin Miyatake, Hiroki Ichiyama, Eiji Kondo & Kazunori Yasuda

Department of Sports Medicine and Joint Reconstruction Surgery, Hokkaido University Graduate School of Medicine, Sapporo, Japan

Correspondence
Professor Kazunori Yasuda, MD, PhD, Department of Sports Medicine and Joint Reconstruction Surgery, Hokkaido University School of Medicine, Kita-15 Nishi-7, Kita-ku, Sapporo 060-8638, Japan.
Tel: + 81 1 1706 7211
Fax: + 81 1 1706 7822
E-mail: yasukaz@med.hokudai.ac.jp

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WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

- Efficacy of oral administration of nonsteroidal anti-inflammatory drugs (NSAIDs) on the synovial and muscle tissues has been established.
- However, efficacy of percutaneous administration of NSAIDs has not sufficiently been established because of lack of scientific data.

WHAT THIS STUDY ADDS

- The diclofenac concentration in the muscle and fat at 12 h after topical application with a pair of tapes containing a total of 30 mg diclofenac sodium was significantly higher than that after oral application of a capsule containing 37.5 mg diclofenac sodium, whereas there was no significant difference in the plasma diclofenac level between the two applications.
- The concentration in the synovial membrane and fluid was significantly lower after topical application than after oral application.

AIMS

To compare tissue concentrations of diclofenac resulting from topical and oral applications of diclofenac according to clinically recommended prescriptions.

METHODS

Diclofenac sodium was applied to 14 subjects (four male and 10 female), who were scheduled to undergo knee arthroplasty due to osteoarthritis, according to the oral or topical prescription (a capsule containing 37.5 mg diclofenac sodium or two tapes containing a total of 30 mg diclofenac sodium). At 12 h after prescription, the diclofenac concentration in the fat, muscle and synovial tissues was measured with liquid chromatography and mass analysis.

RESULTS

The diclofenac concentration in the muscle was significantly higher ($P = 0.0196$) after topical application (average 9.29 ng ml^{-1}) than after oral application (0.66 ng ml^{-1}), whereas there was no significant difference in the plasma diclofenac level (4.70 and 6.63 ng ml^{-1}) between the two applications. The concentration in the synovial membrane was significantly ($P = 0.0181$) lower in the topical application (4.99 ng ml^{-1}) than in the oral application (15.07 ng ml^{-1}).

CONCLUSIONS

Whereas plasma levels resulting from topical and oral applications of diclofenac according to clinically recommended prescriptions were comparable, concentration levels in the muscle and synovial tissues were different.

Introduction

Efficacy of oral administration of nonsteroidal anti-inflammatory drugs (NSAIDs) on synovial and muscle tissues has been established [1]. However, efficacy of topical administration of NSAIDs has not sufficiently been established because of lack of scientific data, although it is expected to reduce the adverse reactions in the gastrointestinal tract due to oral administration [2]. There have been many studies on the tissue concentration of NSAIDs after topical administration. Concerning pharmacokinetics of topical application of diclofenac, Muller *et al*. [3] monitored the concentration in the human skin and subcutaneous tissues for 5 h after administration of a single dose (gel preparation) of approximately 300 mg 100 cm^{-2}, using microdialysis probes. The concentration peaked at 1 h and then decreased gradually with time, reaching approximately one-quarter or one-fifth of peak value at 4 h. Assandri *et al*. [4] reported that the peak plasma concentration of diclofenac after plaster application was about 15 ng ml^{-1}, much lower than that reached by oral administration (approximately 1500 ng ml^{-1}), but similar to that reached with a topical gel or cream application. Concentrations of NSAIDs in a targeted tissue are one of the most fundamental data in evaluating the efficacy of NSAIDs. Then, it is an important strategy to compare the tissue concentration between topical and oral applications in order to establish the efficacy of topical administration. Tegeder *et al*. [5, 6] compared the concentration of ibuprofen and ketoprofen in the muscle and fat tissues between topical and oral applications, using microdialysis. However, no studies have compared the concentration of diclofenac sodium in the subcutaneous, muscular and synovial tissues between topical and oral applications. We conducted a randomized clinical study to compare the diclofenac concentration in the targeted tissues between topical and oral applications according to the commonly used clinical prescriptions. In this study, we have clarified that the diclofenac concentration in the muscle was higher at 12 h after topical application than after oral application.

Methods

This randomized clinical study was conducted with prior approval from the Investigational Review Board, the Ethics Committee of Hokkaido University School of Medicine. The subjects were patients with osteoarthritic knee who were scheduled to undergo total knee replacement. Exclusion criteria used in this study are shown in Table 1. Prior to the start of the study, informed consent was obtained from each patient with a signed document. Finally, 14 patients with osteoarthritis of the knee were enrolled, consisting of four men and 10 women, ranging in age from 57 to 88 years (mean 75.8 years).

Table 1

Exclusion criteria from this study

Number	Criteria
1	Patients who had received any diclofenac sodium or mefenamic acid preparation within 2 weeks before the start of the study
2	Patients who had received an injection of a steroid, sodium hyaluronate, etc., into the affected joint within a week before the start of the study
3	Patients with wounds or diseases in the skin around the knee
4	Patients with a history of hypersensitivity to a certain drug
5	Patients suffering from a peptic ulcer
6	Patients with severe underlying internal diseases (heart disease, renal disease, coronary artery disease, haematological disease, etc.) or history of such diseases
7	Patients with a history of hypersensitivity to a certain nonsteroidal anti-inflammatory drug (especially diclofenac sodium)
8	Pregnant or possibly pregnant women
9	Nursing women
10	Other patients judged, for any reason, to be unsuitable for the study by the attending physician

The 14 subjects were randomly divided into two groups, a topical application group (Group T) and an oral application group (Group O). We prepared diclofenac sodium tapes (Voltaren Tape®; Novartis Parma Co., Tokyo, Japan), which has been clinically used in Japan since 2004, and diclofenac sodium slow-release capsules (Voltaren SR Capsule®; Novartis Parma Co.), which has been used clinically in Japan since 1990. The tape consisted of 70-cm^2 flexible backing cloth (7×10 cm) and 1.5 g of adhesive in which 15 mg of diclofenac sodium was dissolved as an active pharmaceutical ingredient. The slow-release capsule was made from gelatin and contained immediate-release granules and sustained-release granules mixed at a ratio of 3:7. The capsule contained a total of 37.5 mg of diclofenac sodium. In Group T, two diclofenac sodium tapes were patched on the medial and lateral aspects, respectively, 12 h before surgery (in the evening of the previous day). In Group O, a slow-release capsule was orally administered 12 h before surgery. These applications were performed according to the clinically recommended prescriptions. Then, at the time of surgery (in the morning), 5-ml blood samples were collected with heparinized vacuum syringes from each patient. During surgery, after >5 ml of synovial fluid had been aspirated, the subcutaneous fat, the medial vastus muscle and 1–2 g of synovial tissue were resected from each subject. All samples were stored at −40°C until measurement.

To measure diclofenac concentrations, 0.1-g tissue samples were homogenized with methanol. A solution portion was dried, diluted in phosphoric acid and mixed with cyclohexane and diethyl-ether. An organic portion of the extract was dried, and the residue was dissolved again in methanol. The diclofenac level was then measured with liquid chromatography and mass spectrometry. Liquid

Table 2

The diclofenac concentration in each tissue

Group	Patient	Fat	Muscle	Synovial membrane	Plasma	Synovial fluid
Topical application	1	4.757	<LOQ	3.15	6.209	2.994
	2	2.45	10.776	6.626	3.566	1.718
	3	20.47	<LOQ	<LOQ	3.364	1.8
	4	6.03	8.911	6.844	5.133	2.851
	5	6.953	6.632	2.886	1.783	1.325
	6	32.655	23.359	11.899	7.608	1.524
	7	20.894	15.381	3.535	5.252	1.504
	Mean	13.46	9.29	4.99	4.70	1.96
	SD	11.31	8.34	3.84	1.95	0.68
	5% CI	3.35	1.84	1.55	2.96	1.35
	95% CI	16.81	11.14	6.55	7.66	3.31
Oral application	8	4.236	<LOQ	6.695	4.242	8.301
	9	2.946	<LOQ	11.242	5.069	15.747
	10	6.494	3.167	16.166	5.255	7.858
	11	1.857	<LOQ	12.413	3.415	17.233
	12	<LOQ	<LOQ	4.787	2.37	2.655
	13	5.907	<LOQ	24.232	11.728	32.485
	14	5.276	<LOQ	29.965	14.335	33.064
	Mean	3.85	0.66	15.07	6.63	16.76
	SD	2.28	1.11	9.17	4.54	12
	5% CI	1.72	–0.62	6.87	2.57	6.04
	95% CI	5.54	–0.17	21.95	9.20	22.80
Comparisons between the two groups	*P*-value	0.0476	0.0196	0.0181	0.6547	0.004

The quantification limit (<LOQ) value was 0.24 ng g^{-1} for fat, muscle and synovial tissue, and 0.4 ng ml^{-1} for synovial fluid and blood plasma. For samples in which the diclofenac level was below the <LOQ, zero was used to calculate the mean and the standard deviation (SD).

chromatography was performed on an Alliance 2695 Separations Module (Waters Corp., Milford, MA, USA). Chromatographic separations were obtained under isocratic conditions using an Inetetsil ODS-3 column (GL Sciences Inc, Torrance, CA, USA) at a flow rate of 0.2 ml min^{-1}. The temperature was set at 40°C. The mobile phase consisted of acetonitrile and 2.5 mM ammonium acetate in 0.5% acetic acid (90:10; v/v). Mass spectrometry was performed with a Quattro micro API Mass Analyzer (Waters Corp.). The negative mode and the Multiple Reaction Monitoring (MRM) mode were chosen for diclofenac and mefenamic acid (internal standard) detection, respectively. Selected reaction monitoring was employed using nitrogen as the collision gas with collision energy of 10 and 20 eV for diclofenac and internal standard, respectively. Ions monitored in the MRM mode were m/z 293.8 to m/z 249.9 for diclofenac, and m/z 240.0 to m/z 196.0 for internal standard, respectively. The measured diclofenac concentration was calibrated with a calibration curve that was determined using plasma as a blank (control) matrix.

For samples in which the diclofenac level was below the quantification limit, measured values assumed zero. Comparison between groups was made using the Mann–Whitney *U*-test. The significance level was set at $P = 0.05$.

Results

None of the 14 enrolled patients showed any clinical problems due to the topical and oral application, with no abnormal data shown in the laboratory evaluation during the study period. Table 2 shows a comparison of the diclofenac concentration in each tissue between Groups T and O. There was no significant difference in the plasma diclofenac level between the two groups. The diclofenac concentration in subcutaneous fat and muscle were significantly higher in Group T than in Group O ($P = 0.0476$ and 0.0196), and that in the synovial membrane and synovial fluid was significantly lower in Group T than in Group O ($P = 0.0181$ and 0.004).

Discussion

The mean diclofenac concentration in the muscle was significantly higher in the topical application than in the oral application group. Previously, Majima *et al*. [7] reported that the diclofenac concentration in the muscle was below the quantification limit (20.0 ng g^{-1}) at 6 h after oral administration of diclofenac sodium. Their results support our measurement results after the oral application. It has been unknown why the diclofenac concentration in muscle is below the quantification limit after oral administration. However, we can state that topical application is a method to increase the diclofenac concentration effectively in the muscle close to the body surface, which frequently presents inflammatory conditions due to overuse and trauma.

In the present study, diclofenac concentrations in the synovial membrane as well as the synovial fluid were

significantly lower after topical application than after oral application. From the viewpoint of increasing diclofenac concentration in the synovial tissue, oral seems to be superior to topical application. However, the minimal concentration that has anti-inflammatory effects in the synovial tissue remains unknown. Therefore, we cannot conclude that topical application has no anti-inflammatory effects in pathological conditions of the synovial membrane. We therefore speculated why synovial concentrations were different while plasma concentrations were comparable. Fower *et al*. [8] reported that the ratio of plasma to synovial fluid concentrations gradually decreased after administration of 100 mg slow-release formulation of dicrofenac sodium (222 ng ml^{-1} in plasma *vs.* 181 ng ml^{-1} in synovial fluid at 4 h, and 32 ng ml^{-1} *vs.* 118 ng ml^{-1} at 12 h). This study suggested the reason why synovial fluid concentrations after oral administration were much greater than plasma concentrations in the present study. However, we could not verify this explanation in the present study, because we did not determine the concentration–time curves of plasma and synovial tissues or the peak values after administration. This is a limitation of the present study.

As regards the differences between NSAIDs and diclofenac, Tegeder *et al*. [5] have reported that the ibuprofen concentration in muscle was greater after oral administration (80 mg) than after topical administration (5% ibuprofen gel of 16 g applied onto the femoral skin of 17 × 19 cm). This result is similar to that in our study. Regarding ketoprofen [6], most dialysate concentrations after topical dosing of ketoprofen (100 mg) were below the quantification limit. This result appears to be different from the result in our study. However, because there are many differences in prescription, formulation and measurement methods between our study and Tegeder's, we cannot directly compare the two.

We tried to compare tissue concentration with the IC_{50} (50% inhibitory concentration for cyclooxygenese-2 inhibition) values of dicrofenac. *In vivo* data are not available. In the present study, the concentration was 4.99 ng ml^{-1} in synovial membrane and 9.29 ng ml^{-1} in muscle at 12 h. However, reported IC_{50} values of dicrofenac have had a wide range of 1–500 ng ml^{-1} [9, 10]. The reason for such high variability is not known, but might be due to differences in the free fraction of dicrofenac or the accessibility of the cyclooxygenases, which might be better with single cells than with tissue fragments [6]. Furthermore, it is not known which of these values most accurately predicts *in vivo* performance and hence clinical efficacy. Therefore, we could not conclude whether single-dose administration of dicrofenac gel led to effective tissue concentrations.

In conclusion, we have compared diclofenac concentrations between topical application with a pair of tapes containing a total of 30 mg diclofenac sodium and oral application of a capsule containing 37.5 mg diclofenac sodium, because we intended to compare the two applications according to clinically recommended prescriptions. There was no significant difference in plasma diclofenac levels between the two applications. The present study has shown evidence that topical application with a tape containing diclofenac sodium is an effective method to deliver diclofenac to the human body, particularly to muscle near the body surface.

Competing interests

None to declare.

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REFERENCES

1 Buckwalter JA, Stanish WD, Rosier RN, Schenck RC Jr, Dennis DA, Coutts RD. The increasing need for nonoperative treatment of patients with osteoarthritis. Clin Orthop Relat Res 2001; 385: 36–45.

2 Henry D, Lim LL, Garcia Rodriguez LA, Perez Gutthann S, Carson JL, Griffin M, Savage R, Logan R, Moride Y, Hawkey C, Hill S, Fries JT. Variability in risk of gastrointestinal complications with individual non-steroidal anti-inflammatory drugs: results of a collaborative meta-analysis. BMJ 1996; 312: 1563–6.

3 Muller M, Mascher H, Kikuta C, Schtier S, Brunner M, Dorner G, Eichler HG. Diclofenac concentrations in defined tissue layers after topical administration. Clin Pharmacol Ther 1997; 62: 293–9.

4 Assandri A, Canali S, Giachetti C. Local tolerability and pharmacokinetic profile of a new transdermal delivery system, DHEP plaster. Drugs Exp Clin Res 1993; 19: 89–6.

5 Tegeder I, Muth-Selbach U, Lötsch J, Rüsing G, Oelkers R, Brune K, Meller S, Kelm GR, Sörgel F, Geisslinger G. Application of microdialysis for the determination of muscle and subcutaneous tissue concentrations after oral and topical ibuprofen administration. Clin Pharmacol Ther 1999; 65: 357–68.

6 Tegeder I, Lötsch J, Kinzig-Schippers M, Sörgel F, Kelm GR, Meller ST, Geisslinger G. Comparison of tissue concentrations after intramuscular and topical administration of ketoprofen. Pharm Res 2001; 18: 980–6.

7 Majima T, Kondo S, Sakiyama N. Concentrations of Voltaren (diclofenac sodium) SR capsule in serum and tissues of patients with rheumatoid arthritis (Japanese text). Enshou (The Official Journal of Japanese Society of Inflammation) 1995; 15: 255–9.

8 Fower PD, Dawes PT, John VA, Shotton PA. Plasma and synovial fluid concentrations of diclofenac sodium and its

hydroxylated metabolites during once-daily administration of a 100 mg slow-release formulation. Eur J Clin Pharmacol 1986; 31: 469–72.

9 Cryer B, Feldman M. Cyclooxygenase-1 and cyclooxygenase-2 selectivity of widely used nonsteroidal anti-inflammatory drugs. Am J Med 1998; 104: 413–21.

10 Mitchell JA, Akarasereenont P, Thiemermann C, Flower RJ, Vane JR. Selectivity of nonsteroidal anti-inflammatory drugs as inhibitors of constitutive and inducible cyclooxygenase. Proc Natl Acad Sci USA 1993; 90: 11693–7.