British Journal of Plastic Surgery (1993), **46**, 576–579

BRITISH JOURNAL OF PLASTIC SURGERY

Release of Ofloxacin from silicone gel sheet

Y. Sawada, I. Hatayama and K. Sone

Departments of Plastic and Reconstructive Surgery, Second Department of Biochemistry, and Pharmacy, Hirosaki University School of Medicine, Japan

SUMMARY. **From a drug delivery system using silicone gel, the amount of Ofloxacin (OFLX) released or transferred to a wound and blood was measured over 2 weeks. From three types of silicone gel containing 2, 0.2 and 0.02% OFLX respectively, levels from that with 2% OFLX were highest, approximately two to five times higher than that with 0.02% OFLX. Statistically significant differences were found between the three types ($P < 0.01$, Student's t-test).**

When used in partial thickness skin wounds on rats, only an extremely small amount of OFLX was detected in the serum, being higher under gel containing 2% OFLX. In a clinical study, however, no drug was detected either in the blood or the wound after 1 week.

We have previously described a drug delivery system of silicone gel sheet, which releases its contained antimicrobial agent to prevent wound infection.[1–8] It can be used not only as a wound dressing to prevent wound infection but also for other purposes by varying the contained agents, such as to promote experimental flap survival or to prevent obstruction of the suction drainage tube.[6–8] We studied the release and penetration of Ofloxacin (OFLX) through the dermis from the gel sheet. OFLX is an antimicrobial agent, a quinoline carboxylic acid compound, that exhibits broad spectrum *in vitro* bactericidal activity against both gram-positive and gram-negative bacteria.[1]

Materials and methods

Silicone gel was obtained from Dow Corning K.K. (Tokyo, Japan) and OFLX from Daiichi Seiyaku (Tokyo, Japan). A cellulose acetate membrane filter, 90 mm in diameter (Toyo Roshi, Tokyo, Japan), was used to evaluate the released silicone oil and OFLX. Silicone gels containing 0.02 (Type A), 0.2 (Type B) and 2% (Type C) OFLX retrospectively were prepared as described previously.[1] To measure OFLX in the filter papers, the tissues and the sera, High Performance Liquid Chromatography (HPLC) was used.

The HPLC system used in this study consisted of a solvent delivery system (Spectra Physics, San Jose, U.S.A.), a sample injector (EIE 005, Senshu kagaku, Tokyo, Japan) fitted with a 50 micro litre loop, a guard filter, a prepacked stainless steel column packed with 5 micron metre particles of octadecylsilica and a variable wave length spectrofluorimeter (F1000, Hitachi, Tokyo, Japan).[9]

To 0.5 ml of the serum in a 12 ml glass centrifuge tube, 0.02 ml of internal standard solution and 1.5 ml of 0.1 M phosphate buffer (pH 7.0) were added and mixed on a vortex mixer. Then, a sample was extracted with 5 ml of chloroform. After centrifugation at 1500 g for 10 min, the chloroform layer was separated and evaporated at 40°C. The residue was reconstituted in 0.4 ml of mobile phase, and 20 micro litres were injected into the HPLC column.

Tissue samples were homogenised in 4 volumes of phosphate buffer with a disperser, and the supernatants after centrifugation of the homogenates were treated like serum samples. Each membrane filter was cut into small pieces, and shaken with 5 ml of the phosphate buffer and 2 ml of petroleum ether. After centrifugation at 3000 rpm for 5 min, OFLX in the aqueous layer was measured.

First study

Each of eight silicone gel sheets of types A, B and C were cut 5 × 5 cm. Before study, the filters were

Table 1 Released OFLX and silicone oil from the silicone gel sheet (mean ± S.D., /day/sheet)

		Initial 3 *days*	*Next* 4 *days*	*Next* 3 *days*	*Next* 4 *days*
OFLX with Silicone Oil (mg ± S.D.)	Type A	9.5 ± 0.7	6.2 ± 0.2	6.9 ± 0.3	3.8 ± 0.4
	Type B	9.9 ± 1.1	7.5 ± 0.2	7.9 ± 0.4	4.9 ± 0.1
	Type C	9.6 ± 0.2	6.8 ± 0.5	5.6 ± 0.2	4.3 ± 0.1
OFLX (10^{-3} mg ± S.D.)	Type A	104.3 ± 32.0	94.6 ± 22.1	115.7 ± 28.3	100.0 ± 20.7
	Type B	258.8 ± 110.6	171.6 ± 54.1	186.7 ± 39.9	194.1 ± 38.9
	Type C	517.2 ± 121.8	316.3 ± 195.8	326.7 ± 47.2	266.7 ± 54.1

(Type A: n = 7, Type B: n = 8, Type C: n = 6)

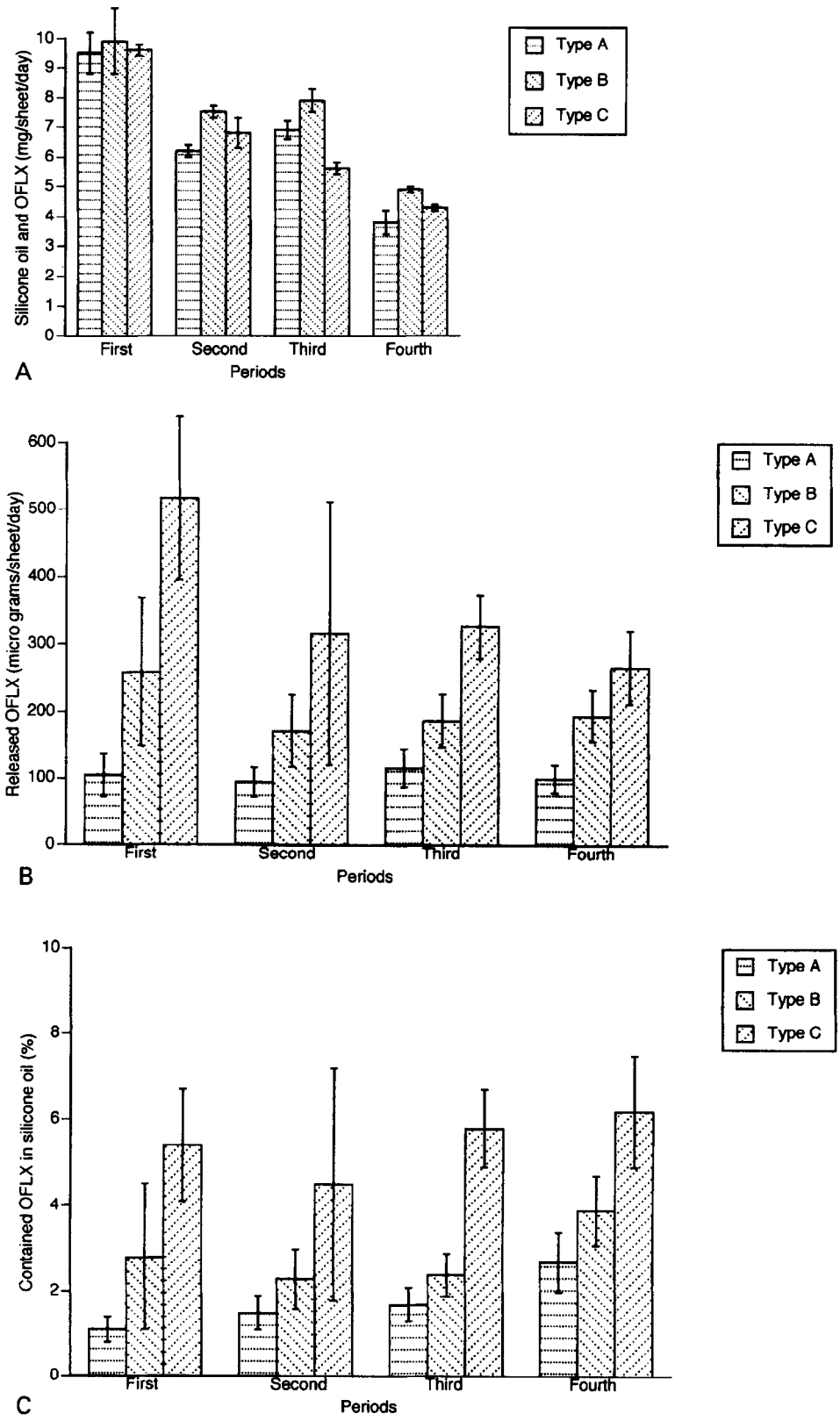

Fig. 1

Figure 1—(A) Released silicone oil and OFLX from silicone gel of types A, B and C, during 2 weeks in the first study. (B). Released OFLX from types A, B and C of silicone gel sheet during 2 weeks. Although the amount of the released silicone oil decreased gradually, that of released OFLX was relatively steady, especially in type A silicone gel. (C). Concentration of OFLX in the released silicone oil was relatively steady during 2 weeks.

Table 2 Results of the second study

	Serum (ng/ml)	*Tissue (ng/g)*	*Epithelialisation rate (%)*
Group 1 (n = 12)	17.5±13.6	194.2±231.2	79.2±22.2
Group 2 (n = 9)	4.4±6.8	2534.4±1514.5	94.2±15.6

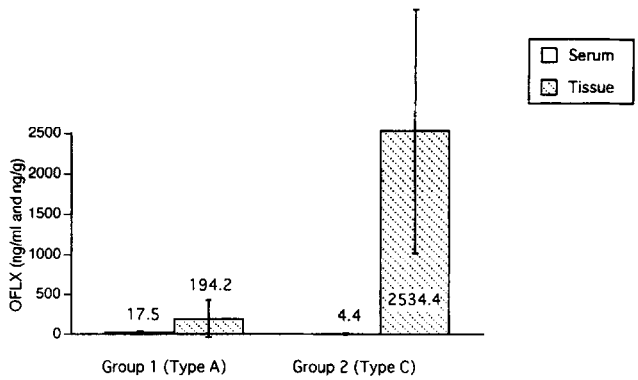

Fig. 2

Figure 2—Tissue and serum concentration of OFLX in the experiment using rats.

weighed on an electronic weighing scale (Sartorius, Germany), the range being from 10 to 10^{-5} g. A silicone gel sheet was placed on a cellulose acetate membrane filter, and stored at room temperature. After starting the study, on the 3rd, 7th, 10th and 14th day, the filter was changed, to evaluate the released silicone oil and OFLX during these four test periods, namely first, second, third, and fourth periods. However, the silicone gel sheet used at the start of this study was used continuously during 2 weeks. The membrane filters were weighed correctly, and stored at −85°C wrapped in aluminum foil until OFLX was measured.

Second study

24 female Wistar rats, weighing from 250–350 g, were used. A 3 × 3 cm split thickness wound approximately 250 micron deep was made on the back of each rat. In preparing the wound, a procedure described previously was used. [10] The rats were divided into two groups 1 and 2, and type A silicone gel was used in group 1 rats, and type C in group 2 rats. The whole wound was covered with the silicone gel sheet. The silicone gel was attached by 8 sutures to the wound. On the seventh day after surgery, blood was taken from the femoral vein of the rat. The blood sample was centrifuged at 3000 rpm for 5 min, and the supernatant serum was stored at −85°C until study. After removing the silicone gel on the back of the rat, the wound was wiped with fresh gauze to remove OFLX on the wound surface. The epithelialised area of the wound was traced using tracing paper to assess the percentage of the epithelialised area in the wound. Then the whole wound beneath the silicone gel was biopsied to the deep fascia. The serum and the biopsied specimen were stored at −85°C until study.

Third study

After obtaining informed consent from four patients with burns to whom silicone gel was applied for a week previous to surgery, blood and tissue specimens were removed at surgery and OFLX levels were measured. OFLX was not otherwise given to these patients. The details of each case are shown in Table 3. After removing silicone gel and wiping the wound surface with fresh gauze to remove OFLX, the wound was biopsied to a level deep to the subcutaneous fatty tissue. The tissue and the blood samples were treated as described above to measure OFLX. Bacteriological examination was carried out on the exudate beneath the silicone gel.

Results

First study

Among eight filter papers in each group, one of type A and two of type C were damaged, so that seven filter membranes of type A, eight of type B, and six of type C were available for study. The released amount of silicone oil and OFLX decreased gradually, but that of OFLX from type A silicone gel was relatively steady over 2 weeks (Table 1, Fig. 1). Although concentrations of OFLX in type C silicone gel were 100 times more than that in type A silicone gel, those of OFLX from type C silicone gel were approximately 2 to 5 times more than that from type A silicone gel. A statistically significant difference was found between type A and B, and type B and C of silicone gel in

Table 3 Clinical results using silicone gel containing 0.02% OFLX

	Age	*Sex*	*Site*	*Wound*	*Size of silicone gel*	*Tissue OFLX*	*Serum OFLX*	*Epithelialisation*
Case 1	17	M	Back	DDB	100 by 100 cm	none	none	none
Case 2	25	M	Thigh	DDB	120 by 150 cm	none	none	none
Case 3	42	F	Thigh	DDB	120 by 150 cm	none	none	none
Case 4	45	F	Abdomen	DDB	120 by 150 cm	none	none	none

M: male, F: female.

respect of released OFLX over the 2 week period ($P < 0.01$, Student's t-test).

Second study

Silicone gels of three rats in group 2 were damaged during the experiment so evaluation was carried out on 12 rats of group 1 and 9 rats of group 2. The wounds were not infected macroscopically. The percentage epithelialisation of the wound was $79.2 \pm 22.2\%$ in group 1 and $94.2 \pm 15.6\%$ in group 2 (not statistically significant). The OFLX values in the serum and the tissue are listed in Figure 2. Although statistically significant differences were detected in the tissue concentration of OFLX ($P < 0.01$, Student's t-test), significant differences were not found in the serum concentration (Table 2, Fig. 2).

Third study

During the test period, little epithelialisation of the wound was seen in all cases. No wound infection under the silicone gel sheet developed in these patients during the test period. In all serum and tissue specimens, no meaningful value of OFLX was detected (Table 3).

Discussion

In our present study, only an extremely small amount of drug or no drug was detected in the serum in the second and the third study. This seems to be due to low concentration of drug released, as shown in the first study. The drug is released from the gel sheet into the discharge on the wound surface. As the silicone gel is usually in close contact with the wound surface, abundant discharge together with OFLX will escape from beneath the silicone gel, so that the drug can scarcely transfer across the wound. We believe that although the released concentration of OFLX is low, it will be sufficient to prevent wound infection in the extremely thin layer of discharge from our clinical and experimental studies.[3,4]

On the contrary, the concentration of the drug was relatively high in the tissue in group 2 of the second study. It might be postulated that moderate absorption of the drug into the tissue occurs, as most of the wound had already been epithelialised in rats of group 2. When occlusive dressing techniques are used on healthy skin, an increase of drug absorption through the skin has been well demonstrated.

Consequently, when applying silicone gel to prevent wound infection, close contact with the wound surface is essential. Further, we advise removing the gel sheet when wound epithelialisation is complete. Although further study is necessary, these occlusive dressing materials with continuous drug release should be an effective dressing material in the future.

Acknowledgement

The authors wish to thank Dow Corning K.K. and Daiichi Seiyaku, Tokyo, Japan for supplying the materials used in this study.

References

1. Sawada Y, Suzuki T, Hatayama I, Sone K. Silicone gel including antimicrobial agent. Br J Plast Surg 1990; 43: 78–82.
2. Sawada Y, Yotsuyanagi T, Hatayama I, Sone K. A new system of treating wounds by a continuous topical application of medication. Br J Plast Surg 1990; 43: 83–7.
3. Sawada Y, Yotsuyanagi T, Sone K. A silicone gel sheet dressing containing an antimicrobial agent for split thickness donor site wounds. Br J Plast Surg 1990; 43: 88–93.
4. Sawada Y, Ara M, Yotsuyanagi T, Sone K. Treatment of dermal depth burn wounds with an antimicrobial agent-releasing silicone gel sheet. Burns 1990; 16: 347–52.
5. Sawada Y, Yotsuyanagi T, Ara M, Sone K. Experiences using silicone gel tie-over dressing following skin grafting. Burns 1990; 16: 353–7.
6. Sawada Y, Kudo H, Sone K. Cause of closed suction drainage tube obstruction and its prevention using a tube coated on the inside with a silicone gel drug delivery system. Br J Plast Surg 1991; 44: 589–92.
7 Sawada Y, Yotsuyanagi T, Hatayama I, Sone K. The relationship between Prostaglandin E1 applied area and flap survival rate. Br J Plast Surg 1992; 45: 465–8.
8. Sawada Y, Hatayama I, Sone K. The effect of continuous topical application of heparin on flap survival. Br J Plast Surg 1992; 45: 515–8.
9. Matsubayashi K, Une T, Osada Y. Determination of ofloxacin in bronchoalveolar fluid by high-performance liquid chromatography and fluorimetric detection. J Chromatogr 1989; 495: 354–7.
10. Suzuki T, Sawada Y. A dermatome for experiments with small animals. Eur J Plast Surg 1989; 12: 143–4.

The Authors

Yukimasa Sawada, MD*, Associate Professor.
Ichiro Hatayama, MD**, Assistant Professor.
Ken Sone, MD***, Research Instructor.
Department of Plastic and Reconstructive Surgery*, Second Department of Biochemistry**, and Pharmacy***, Hirosaki University School of Medicine, 53 Hon-cho, Hirosaki City, Aomori Prefecture 036, Japan.

Requests for reprints to Dr Yukimasa Sawada.

Paper received 26 March 1992.
Accepted 19 May 1993, after revision.