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Note

Novel wound models for characterizing ibuprofen release from foam dressings

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ABSTRACT

The purpose of the present study was to design and characterize low exudate level wound (LEW) and high exudate level wound (HEW) *in vitro* models by means of investigating therapeutic substance release from exudate-absorbing formulations. Biatain®Ibu foam dressing was used to characterize *in vitro* release of ibuprofen within the models and also for *in vitro–in vivo* correlation (IVIVC) studies. Ibuprofen release was described by zero order rate constants of 0.0147 for 1 day and 0.0038 mg/cm^2 h for 3 days in HEW and LEW models, respectively. The release is suggested to be controlled by ibuprofen diffusion from the dressing in the HEW model, whereas fluid absorption is rate-limiting in the LEW model. Ibuprofen release, from Biatain®Ibu foam dressings *in vivo*, is within the same ranges as *in vitro*. Thus, it is suggested that, depending on the level of exudate, the *in vivo* release of ibuprofen depends on ibuprofen diffusion from and absorption of exudates to the dressings. Consequently, both the HEW and LEW *in vitro* models should be applied in order to fully characterize ibuprofen release from Biatain®Ibu foam dressings. Future studies may show whether these *in vitro* models can be used to characterize therapeutic substance release from exudate-absorbing formulations in general.

1. Introduction

Advanced pharmaceutical formulations, such as matrixes, hydrogels or foam dressings containing pain-relieving or antibacterial therapeutics, have been developed for wound treatments in order to distribute therapeutic substances to exuding wounds in a controlled release manner (Balakrishan et al., 2006; Miyoshi et al., 2006; Vachon and Yager, 2006; Lansdown et al., 2005; Parsons et al., 2005; Shanmugasundaram et al., 2005; Senet, 2004; Cho et al., 2002). Exuding wounds are also treated with exudate-absorbing dressings and it has been shown that it is an advantage to treat low- to high-exuding chronic wounds with exudate-absorbing polyurethane foam dressings, as compared to non-exudate-absorbing dressings (Andersen et al., 2002; Thomas, 1997; Thomas et al., 1996). Consequently, it is believed that exudate-absorbing polyurethane foam dressings, which also release therapeutic substance, would improve the treatment of exuding wounds.

Wound exudate levels in leg ulcers have been quantified by amount of exudate in absorbing dressings and are described as varying from 0 to 1.2 g/cm^2/day (Thomas, 1997; Thomas et al., 1996). Evaporated water loss in the layer close to the evaporate surface in burns and granulating wounds has also been quantified, as being 0.43 and 0.51 g/cm^2/day, respectively (Lamke et al., 1976).

In the pharmaceutical development of wound formulations, *in vitro*-exuding wound models are applied for characterizing and comparing therapeutic substance release, from different formulations, for quality control reasons and/or for *in vitro–in vivo* correlation (IVIVC) studies (Balakrishan et al., 2006; Shanmugasundaram et al., 2005; Bowler et al., 2005). Such *in vitro* release models are generally based on Franz flow through diffusion cells in which the formulation has unlimited access to fluid (Shah et al., 1999; Thomas et al., 2003–2004). However, for exudate-absorbing formulations, the exudate level may have an influence on therapeutic substance release. Consequently, *in vitro* models, in which the formulations have unlimited access to fluid, may not be suited for therapeutic substance release characterization and IVIVC studies of exudate-absorbing formulations. Thus, the purpose of the present study was to design two *in vitro* wound models, i.e. a low exudate level wound (LEW) model and a high exudate level wound (HEW) model and to characterize the models by following fluid access to, as well as release rates of ibuprofen from, the exudate-absorbing polyurethane foam formulation Biatain®Ibu. An additional purpose of the study was to investigate whether the

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models could be used for possible IVIVC of ibuprofen release from Biatain®Ibu.

2. Materials and methods

2.1. Materials

Ibuprofen Ph. Eur. grade of a purity of 98.5–101.0% (99.8%) was obtained from Albemarle Corporation (South Carolina, USA). Acetonitrile (HPLC-grade) and phosphoric acid 85% (analytical grade) were obtained from Bie Berntsen A/S (Roedovre, Denmark). Nunc 25 mm tissue culture inserts 0.2 μm anopore membrane was obtained from Nunc A/S (Roskilde, Denmark) and agar (Ph Eur. 3rd ed, swelling index 14) from UniKem (Copenhagen, Denmark). Biatain®-Ibu 0.5 mg ibuprofen/cm^2 polyurethane foam wound dressing was donated by Coloplast A/S (Humlebeak, Denmark). High density polyethylene net (HDPE net) was purchased from Smith & Nephew medical.

2.2. Apparatus

Franz diffusion flow through cells with a 2.0 cm^2 open area between the donor and receptor compartment was purchased from Permegear (PA, USA). A Dionex HPLC system, applied to analyze ibuprofen, consists of a Photodiode Array Detector (PDA-100), a two-pump system (P580), an auto-sampler (ASI-100), and a column oven (STH585).

2.3. Methods

2.3.1. HPLC analysis

Reversed phase HPLC was setup as described by Jørgensen et al. (2006). In short, 10 μl samples were injected in to X-terra MSC8, 100 mm × 3.9 mm; 3.5 μm column from Waters (Hedehusene Denmark). The mobile phase consisted of 0.12% aqueous phosphoric acid (pH 2.0) and 80% acetonitrile (50:50), and the flow rate was of 0.9 ml/min. The effluent was detected at 220 nm and the retension time for ibuprofen was 8.26 min. The detection limit was calculated according to ICH guideline, based on the calibration curve, to 1.2291 ppm (ICH, 1998).

2.3.2. Preparation of agar-anopore membranes for 500 and 1000 μl LEW models

The agar-anopore membranes are made with a 2% agar solution in USP phosphate buffer pH 7.4 autoclaved at 121 °C for 20 min. The agar is cooled to 40–50 °C before 500 or 1000 μl agar is applied to the tissue culture insert anopore membrane to be applied in the 500 and 1000 μl LEW Franz diffusion cell models, respectively. The tissue culture insert agar-anopore membranes are then placed at 5 °C overnight. The next day whole tissue culture inserts with the agar-anopore membranes are applied in to the *in vitro* LEW model.

2.3.3. In vitro HEW and LEW Franz diffusion cell models

In the classic HEW model, wound dressings have unlimited access to fluid. A HDPE net is placed on top of the receptor compartment of the Franz diffusion cell in order to keep the Biatain®-Ibu wound dressing in place. The Biatain®-Ibu sample is then surrounded by a silicone gasket and cowered with a flexible impermeable latex lid before the empty donor compartment is clamped to its receptor illustrated in Fig. 1.

In the 500 and 1000 μl LEW models, Fig. 1, wound dressings have limited access to fluid due to the agar-anopore membrane. Thus, in the LEW models, the HDPE net in the HEW model, is exchanged by an 500 or 1000 μl agar-anopore membrane, placed on a silicone gasket. The Biatain®-Ibu wound dressing is placed on top of the agar-anopore membrane followed by a silicone dot. The donor compartment is closed by a latex lid and clamped to its receptor by means to ensure that the Biatain®-Ibu wound dressing stays in contact with the agar membrane through out the experiment. For simplicity the clamp is not shown in Fig. 1.

2.3.4. Characterization of the in vitro models

2.3.4.1. Receptor fluid access to dressings. The 500 μl LEW model is characterized and compared to the classic HEW model by following the receptor fluid access to Biatain®-Ibu wound dressings. To follow the fluid access the models are arranged as described in Section 2.3.3 by placing a weighed Biatain®-Ibu wound dressing (dia. 1.8 cm) on the HDPE net or on the 500 μl agar-anopore membrane in the HEW and 500 μl LEW models, respectively. In both the HEW and LEW models the receptor medium is 15 ml of USP phosphate buffer pH 7.4 that is stirred continuously with a magnetic bar. The flow-through-rate of the receptor medium is 0.79 ml/h. The temperature of the receptor medium is maintained at 37 ± 0.1 °C by an external, constant temperature circulator water bath. At 1, 5 and 16 h and 1, 2, 3, 5 and 7 days a Biatain®-Ibu wound dressing is removed in order to weigh the dressing and calculate the amount of absorbed receptor medium. Numbers of replicates are 9.

2.3.4.2. Ibuprofen release. In a similar setup as described under Sections 2.3.3 and 2.3.4.1, the 500 and 1000 μl LEW as well as the HEW models were further characterized and compared by following the ibuprofen release from Biatain®-Ibu wound dressings to the receptor medium. Thus as described above, a weighed Biatain®-Ibu wound dressing (dia. 1.8 cm) was placed on top of the agar-anopore membrane in the LEW model or on the HDPE net in HEW model. The receptor medium of 15 ml of USP phosphate buffer pH 7.4 is stirred continuously with a magnetic bar and has a flow-through-rate of 0.79 ml/h, at which sink condition is obtained through out the study. As describe in Section 2.3.4.1, the temperature of the receptor medium is maintained at 37 ± 0.1 °C. Samples are collected over a period of 7 days and ibuprofen concentrations in the receptor medium are then analyzed by HPLC as described in Section 2.3.1. Numbers of replicates are between 6 and 9.

2.3.4.3. Ibuprofen diffusion across agar-anopore membranes. The 500 μl LEW and HEW models are also characterized and compared by adding 1.0 ml ibuprofen control formulation (1 mg/ml ibuprofen solution in USP phosphate buffer pH 8) to the donor compartment instead of the Biatain®-Ibu wound dressing formulation. Thus, the control formulation is added directly on to the HDPE or agar-anopore membrane, in HEW and 500 μl LEW models, respectively. Receptor medium samples are collected every second hour for 24 h and then analyzed for ibuprofen by HPLC as described in Section 2.3.1. Number of replicates are 9.

2.4. Comparison of ibuprofen in vitro and in vivo release

In vivo data was taken from a study by Jørgensen et al. (2006). In this study 10 patients with venous leg ulcers were treated with Biatain®-Ibu wound dressings. The dressings were changed every second or third day. Each patient was treated with five dressings. The applied dressings were frozen and analyzed for buprofen content at the separate area that covered the wound and at the separate area that covered the intact skin. The ibuprofen content in the dressings was extracted by methanol containing ketoprofen as an internal standard. The extracts were analyzed by HPLC as described in Section 2.3.1. % ibuprofen release was calculated as described in Section 2.5. % ibuprofen released *in vitro* was then compared to % ibuprofen release *in vivo*.

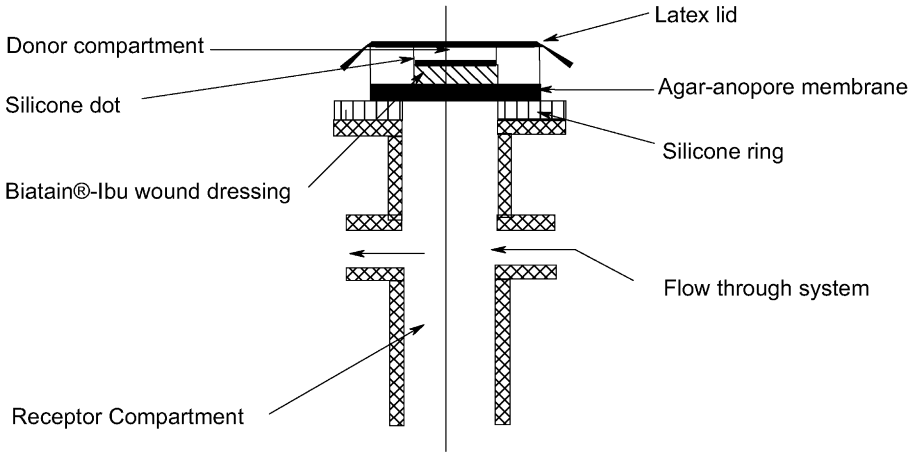

Fig. 1. Illustration of the low exudate level wound (LEW) model. The settings are the same in the high exudate level wound (HEW) model except that agar-anopore membranes in the LEW models are replaced by a HDPE net in the HEW model. In both the LEW and HEW models, the donor compartments are fixed with a clamp, which is not illustrated.

2.4.1. Ethical assurances

The study was conducted as described by Jørgensen et al. (2006). Thus, in accordance with the Declaration of Helsinki II 1964 as amended in Scotland, October 2000, and in accordance with Council Directive 93/42/EEC of 14 June 1993, concerning medical devices (commonly known as the Medical Device Directive), the Council Directive 95/46/EC on the protection of individuals with regard to the processing of personal data and the International ISO standard ISO/DIS 14155-1:2000 Clinical investigation of medical devices on human. All approvals were obtained prior to inclusion of patients. Written informed consent was obtained from all patients after written and verbal information about the study, procedures, potential risks or inconvenience and/or expected benefits.

2.5. Data analysis

Regarding *in vitro* studies, accumulative amount of ibuprofen released per cm^2 Biatain®-Ibu wound dressing is calculated by the following equation:

$$C_c = \frac{C_n + TF}{V} \frac{\sum C_n + C_{n-1}}{2} \quad (1)$$

C_c is the accumulative concentration of ibuprofen in the receptor medium. C_n is the actual concentration of ibuprofen in the sample number n. T is the time for each sample collection (h). F is the constant flow through rate of the receptor medium (0.79 ml/h). V is the volume of the receptor compartment (15 ml). $\sum((C_n + C_{n-1})/2)$ is the sum of the ibuprofen concentration in all previous samples, in which it is anticipated that there is a linear release of ibuprofen between two following samples taken in one sequence (Briggs and Nelson, 2003).

The *in vitro in vivo* correlation (VIVIC) is prepared by comparing the calculated % ibuprofen release from the Biatain®-Ibu wound dressings when investigated *in vitro* and *in vivo*.

% *In vitro* release is calculated by the following equation:

$$C\,(\%) = \frac{C_c}{C_T} \times 100 \quad (2)$$

in which C_T is the total accumulated concentration of ibuprofen in the receptor compartment at time 168 h corresponding to 100% release when measured in the HEW model; C_c is the accumulated concentration at the time t measured in h.

% *In vivo* release is calculated by the following equation:

$$C\,(\%) = \left(\frac{A_{skin} - A_{wound}}{A_{skin}}\right) \times 100 \quad (3)$$

In which A_{skin} is ibuprofen content in the area of the Biatain®-Ibu wound dressing covering intact skin and A_{wound} is the area of the Biatain®-Ibu wound dressing covering the wound.

Correlation between fluid absorption and ibuprofen release *in vitro* is at corresponding times t, calculated by estimating the fluid absorbed at time t $(Abs)_t$ in mg/cm^2 (Eq. (4)) and amount ibuprofen released (C_n) in μg/cm^2 (Eq. (1)).

$$Abs_t = \frac{W_t - W_0}{A} \quad (4)$$

Abs_t is the amount absorbed (μg/cm^2) at the time t. W_t is the weight (mg) of the sample at the time t. W_0 is the weight at time $t = 0$. A is the area of the sample 2.54 cm^2.

3. Results

3.1. Characterization of the in vitro models

3.1.1. Receptor fluid access to dressings

In Fig. 2 is shown the amount of receptor medium absorbed by the Biatain®Ibu foam dressing in g/cm^2 as a function of time in days, when measured in the *in vitro* HEW and 500 μl LEW models. It can be seen that the access of fluid to the Biatain®Ibu foam formulation

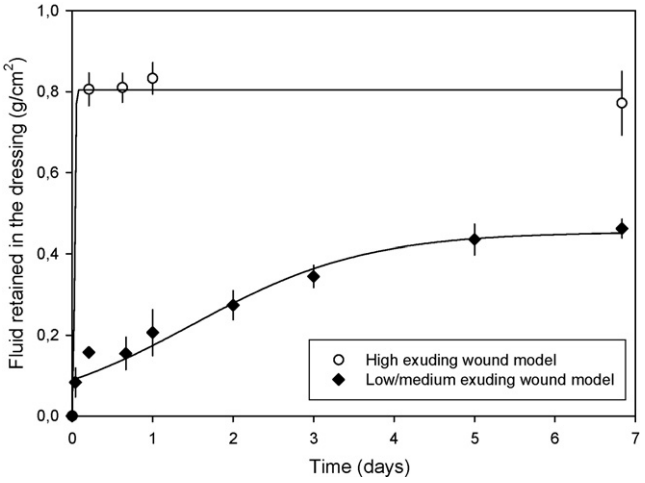

Fig. 2. Amount of fluid absorbed by Biatain®Ibu foam dressings (g/cm^2) versus time in days when investigated with the high exudate level wound (HEW) and low exudate level wound (LEW) models.

Table 1
Abs_{TOTAL} is maximum fluid absorbed to Biatain®Ibu, $k_{release}$ is zero order rate constant representing ibuprofen release from Biatain®Ibu, $t_{50\%}$ is the time after which 50% ibuprofen is released

Model	Abs_{TOTAL} (g/cm^2)	$k_{release}$ (mg/cm^2 h) (r^2)	$t_{50\%\ released}$	
			Control formulation	Biatain®-Ibu
In vitro HEW model	0.82 ± 0.004	0.0147 (0.983)	1.2 h	16 h
In vitro 500 μl LEW model	0.46 ± 0.0023	0.0038 (0.998)	3.6 h	3 days

dressing is significantly lower when investigated by 500 μl LEW than by the HEW model.

During the 7 days of investigation, the final fluid content in the Biatain®Ibu foam dressings was approximately twice as high when investigated by HEW as compared to the 500 μl LEW model. In the HEW model, the foam formulation is saturated in less than 1 h, a steady state which is maintained throughout the study. As illustrated by Fig. 2, and shown in Table 1, column 1, the mean total amount of fluid absorbed by the formulation (Abs_{TOTAL}) is 0.82 g/cm^2 when investigated in the HEW model. In the 500 μl LEW model, the foam formulation does not reach the same level of fluid saturation as observed in the HEW model. After 5 days, the Abs_{TOTAL} (mean) is 0.46 g/cm^2 (Table 1), i.e. 56% of the maximal amount absorbed, when using the HEW model.

3.1.2. *Ibuprofen release from Biatain®Ibu foam dressings*

Fig. 3 shows % ibuprofen release from Biatain®Ibu foam dressings as a function of time in days when investigated *in vitro* by the 500 and 1000 μl LEW as well as by the HEW models. The ibuprofen release rate from dressings investigated by the HEW model is significantly faster than the corresponding release rate investigated by both the LEW models. There are no significant differences between the release rates of ibuprofen from dressings investigated using the 500 and 1000 μl LEW models. As can be seen in Fig. 3, the initial linear releases of ibuprofen from Biatain®Ibu dressings follow zero order kinetics with all the models. A zero order rate constant of 0.0147 mg/cm^2 h for 1 day is seen when ibuprofen release is investigated with the HEW model and a zero order rate constant of 0.0038 mg/cm^2 h for nearly 4 days is seen when release is investigated using either of the LEW models (Table 1, column 2).

The upper dotted line in Fig. 3 is the 95% release line, which shows that 95% ibuprofen is released from Biatain®Ibu foam dressings at approximately 4 and >7 days when investigated in the LEW and HEW models, respectively. The lower dotted line in Fig. 3 is the 50% release line which shows that 50% ibuprofen is released from Biatain®Ibu foam dressings within 16 h and 3 days when investigated in the HEW and LEW models, respectively.

Fig. 4 shows the % ibuprofen diffusion during 24 h from an ibuprofen control formulation solution to the receptor medium when investigated with the HEW and 500 μl LEW models. Here it can be seen that, as expected, ibuprofen immediately diffuses to receptor medium when investigated with the HEW. However, the diffusion rate of ibuprofen from the control solution to the receptor medium is much lower when investigated with the 500 μl LEW than with the HEW model, the half lives being approximately <1.2 and 3.6 h, respectively. This shows that the diffusion across the agar membrane with the LEW model is rate-limiting for diffusion of ibuprofen from the control solution to the receptor medium when investigated using the LEW model.

Table 1 columns 3 and 4 show the time at which 50% ibuprofen reached the receptor medium from ibuprofen control formulations and from Biatain®Ibu foam dressings, respectively when investigated using HEW and LEW models. It can be seen that the release of ibuprofen is significantly lower from Biatain®Ibu foam dressings than from the control formulation when investigated using either of the models. Furthermore, that the release of ibuprofen from either of the formulations is lower when investigated with the LEW model than with the HEW model, and that the lowest release of ibuprofen is seen from the Biatain®Ibu foam dressings when investigated using the LEW model.

From Fig. 5 it can be seen that there is a linear relationship between amounts of receptor medium absorbed by the Biatain®Ibu foam dressings in mg/cm^2, and ibuprofen release, measured in μg/cm^2, from the dressings, when investigated with the LEW model for 7 days.

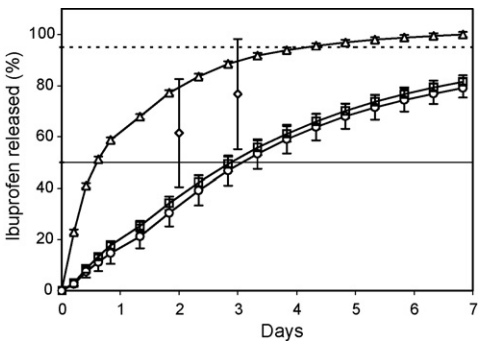

Fig. 3. Accumulated % release of ibuprofen from Biatain®-Ibu foam dressings versus time in days when investigated with low exudate level wound (LEW) and high exudate level wound (HEW) models. (△) represents the release of ibuprofen when investigated with the HEW model; (○) represents the release of ibuprofen when investigated with the 500 μl LEW model; (□) represents the release of ibuprofen when investigated with the 1000 μl LEW model; (◊) represents the ibuprofen release *in vivo* at the second and third day after application. The upper dotted and lower solid lines represent 50% and 95% ibuprofen release, respectively.

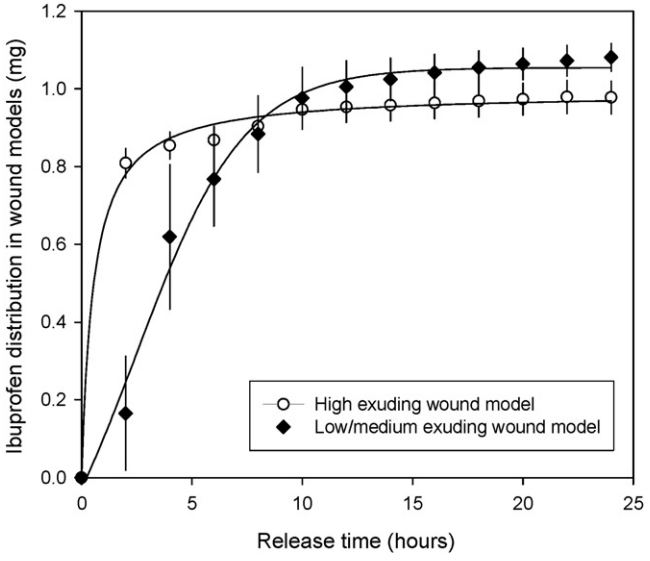

Fig. 4. mg ibuprofen diffusion from an ibuprofen control formulation solution to receptor compartment versus time in hours when investigated with the low exudate level wound (LEW) and high exudate level wound (HEW) models.

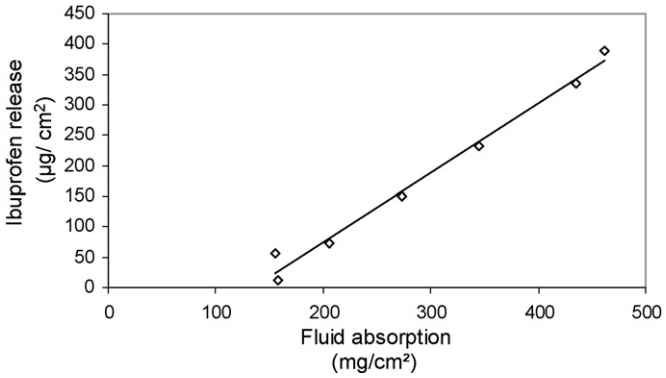

Fig. 5. Ibuprofen release from Biatain®-Ibu foam dressings (μg/cm^2) versus fluid absorbed by the dressings (mg/cm^2). From left to right, the dots represent measured times at 5 and 16 h followed by 1, 2, 3, 5 and 7 days.

3.2. Comparison of ibuprofen release in vitro and in vivo

In Fig. 3 is shown both % ibuprofen release from Biatain®Ibu dressings investigated *in vitro* as described above and % ibuprofen release at the second and third day after there *in vivo* application to venous leg ulcers (10 patients) (Jørgensen et al., 2006). It can be seen that the *in vivo* release of ibuprofen from Biatain®Ibu dressings varies from approximately 40% to 85% ibuprofen release at the second day after application and from approximately 55% to 100% release at the third day after application. When comparing ibuprofen release from Biatain®Ibu investigated *in vitro* and *in vivo*, the *in vivo* variations, is within the predicted levels of ibuprofen release investigated *in vitro* by the HEW and LEW models. The lowest % ibuprofen release, figures at 40% and 55%, respectively when investigated *in vivo* at the second and third day after application of Biatain®Ibu, correspond well to the % ibuprofen release, at 38% and 55%, respectively, seen on the second and third day when investigated using the LEW *in vitro* model. Similarly, the highest % ibuprofen release at 85% and 100%, respectively, when investigated *in vivo* at the second and third day after application corresponds very well to the % ibuprofen release, at 78% and 90% release seen at the second and third day, respectively, when investigated with the HEW *in vitro* model.

4. Discussion

4.1. Characterization of the in vitro models

4.1.1. Fluid access to dressings

The total amount of fluid absorbed to the Biatain®Ibu foam dressings when investigated with the *in vitro* HEW model is much higher than in dressings investigated with the 500 μl LEW model (Fig. 2). This may be due to the dressings being placed in tissue culture insert donor compartments with room around the sample in the LEW model into which part of the absorbed fluid in the formulation may evaporate. In contrast, there is no tissue culture insert in the HEW model and thus no room around the dressing for evaporation. Consequently, the amount of exudate absorbed to the Biatain®Ibu foam dressings when investigated with the LEW model seem to depend on both the fluid access rate and the evaporation rate, which may explain why the dressings are not saturated by fluid to the same extent as they are when investigated with the HEW model, where limited evaporation occurs.

The maximum fluid access to the Biatain®Ibu foam dressings observed with the LEW model after 7 days is 0.46 g/cm^2 (Table 1, column 1). This is approximately the same as the lowest observed exudate amount to be absorbed by hydrocolloid dressings applied to leg ulcers, which can be calculated to 0.20–0.58 g/cm^2 from data presented by Thomas (1997) and Thomas et al. (1996).

The maximum fluid access to Biatain®Ibu foam dressings observed with the HEW model is 0.83 g/cm^2 (Table 1, column 1), which is similar to the maximum observed levels of exudate absorbed by hydrocolloid dressings applied to leg ulcers, which can be calculated to 0.78–3.60 g/cm^2 from data presented by Thomas (1997) and Thomas et al. (1996).

4.1.2. Ibuprofen release

The times at which 50% ibuprofen is released from Biatain®Ibu foam and from control formulations are shown in Table 1, columns 3 and 4. This shows higher ibuprofen diffusion through the agar-anopore membrane than through the Biatain®Ibu foam dressing formulation. Thus, though the release of ibuprofen from the Biatain®Ibu foam dressings seems to be complex, two rate-limiting factors are fluid access to the formulation and ibuprofen diffusion through the formulation. Ibuprofen release figures from formulations investigated using the LEW models show that the 500 μl and 1000 μl agar-anopore membrane LEW models are similar. This shows that the LEW model is robust within the agar content range of 500–1000 μl, and should thus be easy to reproduce.

The release of ibuprofen from Biatain®Ibu foam dressings is suggested to occur by simple passive diffusion of solute ibuprofen through the foam dressings when investigated with either the HEW or LEW *in vitro* models. Since the solubility of ibuprofen is determined to be 4.43 mg/ml at pH 7.4 (data not shown), the dressings have absorbed more fluid than is needed for dissolving the amount of ibuprofen enclosed in the Biatain®Ibu foam dressing formulation, whichever model is used for investigation. It may therefore not be expected that the release of ibuprofen is controlled by limited solubility of ibuprofen. From Figs. 2 and 3 it can be seen that the dressings investigated using the HEW model are capable of releasing ibuprofen in a controlled manner for approximately 3 days, even though they are saturated with fluid instantly. Thus, it is suggested that a rate-limiting diffusion process of dissolving ibuprofen within unstirred water layers in the foam, or a rate-limiting diffusion of dissolved ibuprofen within the polyurethane foam, takes place. Rate-limiting diffusion within the polyurethane foam layer would give rise to square-root-of-time kinetics, which is not seen in the present experiments. An unstirred water layer may control a rate-limiting diffusion in the aqueous fluid within the formulation, where sink condition is not obtained. Consequently, when investigated with the HEW model the ibuprofen concentration gradient between the unstirred water layer and the receptor compartment may explain the initial linear zero order release of ibuprofen. However, this is not the case when the dressings are investigated with the LEW model. Here a linear relationship between fluid absorption and ibuprofen release is shown (Fig. 5). Consequently, when dressings are investigated for 7 days with the LEW model, absorption of fluid is shown to control the release of ibuprofen. In summary, the rate-limiting *in vitro* release of ibuprofen from Biatain®Ibu is controlled by exudate level and ibuprofen diffusion from the formulation. At high fluid levels, which are obtained with the HEW model where unlimited access to fluid takes place, the diffusion of ibuprofen from formulation is rate-limiting. When fluid access levels are low, as is the case with the LEW model, fluid access to the formulation is rate-limiting.

4.2. Comparison of in vitro and in vivo ibuprofen release

Maximum and minimum observed exudate levels from leg ulcers are 0.78–3.60 and 0.20–0.58 g/cm^2, respectively, when calculated from data presented by Thomas (1997) and Thomas et al. (1996). These data correspond very well to fluid access levels in HEW and LEW models of 0.82 and 0.46 g/cm^2, respectively. Based on the *in vitro* investigations, it is suggested, that ibuprofen release *in vivo* is dependent on wound exudate levels. Thus, it is suggested that in highly exuding wounds the diffusion of ibuprofen from dressings is rate-limiting, while for low to medium exuding wounds the exudate level are rate-limiting. This corresponds well with *in vivo* studies of silver-release into wounds from similar polyurethane foam dressings carried out by Lansown et al. (2003), in which an exudate access rate-dependent silver release was demonstrated (Briggs and Nelson, 2003). When Biatain®Ibu foam dressings are applied *in vivo* to wounds of medium to high exudate levels, the % ibuprofen release corresponds very well with the obtained *in vitro* ibuprofen release results in the LEW and HEW models (Fig. 3) (Jørgensen et al., 2006). As shown in Fig. 3, the average *in vivo* release of ibuprofen measured after 2 and 3 days of treatment is in the same range as *in vitro* release of ibuprofen seen with the LEW and HEW models. Unfortunately, the exudate levels of the *in vivo* wounds were not availably from reference (Jørgensen et al., 2006), but experienced professionals rated the wounds as being medium- to high-exuding wounds. The patient experienced a continuous pain-relieving effect when treated with the Biatain®Ibu foam dressings (Jørgensen et al., 2006). This supports the hypothesis that ibuprofen is released from the formulation during the entire period of wearing the dressing, i.e. 2–3 days.

5. Conclusions

Two *in vitro* models, i.e. HEW and LEW, were characterized by following the fluid access to and the release rate of ibuprofen from the exudate-absorbing polyurethane foam formulation Biatain®Ibu, as well as by studying IVIVC. The study shows that ibuprofen release from foam dressings investigated in the *in vitro* models follows zero order kinetics for 1 and 3 days with the HEW and LEW models, respectively. Ibuprofen release is suggested to be controlled by ibuprofen diffusion from the dressings with the HEW model, whereas fluid absorption is rate-limiting with the LEW model. The average *in vivo* release of ibuprofen after 2 and 3 days of treatment correlate very well with the release of ibuprofen in the two *in vitro* models and fluid access correlates well with exudate rates observed in the clinical situation of medium- to highly-exuding wounds. Thus, either of the LEW and the HEW models can be used for quality control of Biatain®Ibu and both should be used to fully characterize ibuprofen release from Biatain®Ibu foam dressing formulations *in vivo*, i.e. for IVIVC. Future studies may reveal whether the two *in vitro* release models are useful for investigating exudate-absorbing formulations as well as exudate level-dependent release in general.

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