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Effect of nonionic surfactants on transdermal drug delivery: II. Poloxamer and poloxamine surfactants

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Summary

Poloxamer [poloxamer 188 (Pluronic F68^{TM}), poloxamer 407 (Pluronic F127^{TM}), poloxamer 338 (Pluronic F108^{TM}), and poloxamer 184 (Pluronic L64^{TM})] as well as poloxamine [poloxamine 304 (Tetronic 304^{TM}), poloxamine 904 (Tetronic 904^{TM}), and poloxamine 908 (Tetronic 908^{TM})] surfactants – the latter category reflecting the physical-chemical properties of the first group of compounds – were studied with respect to their influence on the permeability of methanol through hairless mouse skin as well as through silicone elastomer sheeting and were shown to have basically no effect. The permeability of octanol decreased with increasing surfactant concentration. Determination of thermodynamic activity unveiled that the effects observed were due to entrapment of the lipophilic test permeant in micelles, a finding which was in excellent agreement with the permeation data obtained with silicone elastomer sheeting. Poloxamer 188 (Pluronic F68^{TM}) was the only member of the screened surfactants that demonstrated no negative effect on the permeability of octanol through hairless mouse skin. Despite a moderate decrease in thermodynamic activity as a function of poloxamer 188 (Pluronic F68^{TM}) and concentration the permeability of octanol remained almost invariant at all concentrations studied. This is most likely due to the effect that this surfactant does not form micelles in water. All effects of poloxamer or poloxamine surfactants on full thickness hairless mouse skin as well as silicone rubber membrane were totally reversible when the respective surfactant solution was removed from the donor compartment. In conclusion, neither differences in molecular weight nor varying HLB values of poloxamers and poloxamines appeared to play a major role in affecting the barrier properties of hairless mouse skin.

Introduction

Since many drugs do not permeate the skin in amounts necessary to initiate therapeutic effects, transport can be facilitated by employing penetration enhancers. A fundamental requirement is that the promoter does not cause permanent damage to the skin but alters the barrier properties of the stratum corneum temporarily. Many attempts have been made to find the ideal non-toxic, non-irritating and pharmacologically inert penetration enhancer. Certain nonionic surfactants such as poloxamer and poloxamine surfactants meet many of the above-listed requirements.

The strikingly low toxicity of poloxamer surfactants has been clearly demonstrated by the use of poloxamer 188 in intravenous pharmaceuti-

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cal preparations. Interest in the use of these low toxicity products will be further stimulated by their inclusion in the listing of the US National Formulary. Other poloxamers are used as emulsifiers and dispersants in medicated skin creams and ointments, as solubilizers and detergents in surgical scrubs and contact lens cleansing solution as well as dispersants and stabilizers in antibiotics and vitamins dispersions.

The general intent of the present study was to evaluate by in vitro methods the penetration enhancing effect of the above-mentioned surfactants. Methanol and octanol were chosen as test permeants since their permeation through the skin as well as related factors such as hydration, age, sex of animal and anatomical site of skin have been very well documented in the literature (Behl et al., 1982, 1983, 1985).

Materials and Methods

Chemicals

Methanol (E. Merck, Darmstadt, Germany), octan-1-ol (BDH Chemicals Ltd, Poole, U.K.), xylol (E. Merck, Darmstadt, Germany), tritiated methanol (spec. act., 5 mCi/mmol; tot. act., 25 mCi) (Du Pont de Nemours, Biotechnology Systems Division, Paris, France), [1-^{14}C]octanol (spec. act., 1 mCi/mmol; tot. act., 500 μCi) (ICN Biomedicals GmbH, Eschwege, Germany), and sterile 0.9% sodium chloride solution for irrigation (Fresenius AG, Bad Homburg, Germany) were used. Poloxamer 188 (Pluronic F68^{TM}), poloxamer 338 (Pluronic F108^{TM}), poloxamer 407 (Pluronic F127^{TM}), poloxamer 184 (Pluronic L64^{TM}), poloxamine 304 (Symperonic T304^{TM}), poloxamine 904 (Symperonic T904^{TM}), poloxamine 908 (Symperonic T908^{TM}) were provided by C.H. Erbslöh (Düsseldorf-Hafen, Germany). All organic solvents were HPLC grade or of equivalent quality.

The experimental methods were described in detail previously (Behl et al., 1980; Durrheim et al., 1980; Cappel and Kreuter, 1991). Briefly, stock solutions in saline were formed for both test permeants, i.e. [^{3}H]methanol and [^{14}C]octanol. The actual alcohol concentration in the donor chamber was less than 10^{-4} mol. Thus, any penetration enhancing effect by the two alkanols could be excluded.

In order to distinguish between changes in thermodynamic parameters and effects due to biological consequences two different membranes were employed in this study: (i) silicone elastomer sheeting (Silastic^{TM} sheeting Q7-4840, 175 μm thick, Dow Corning Corp., Midland, MI, U.S.A.) and full thickness hairless mouse skin (strain hr/hr-C3H/TifBom, Bommice, Bomholtgård Breeding and Research Centre Ltd, Ry, Denmark). The female hairless mice were older than 120 days and were killed with CO_2. Both abdominal and dorsal skin was used in the experiments.

Two sequential diffusion experiments were carried out. The first sequential run was initiated to assess the effect of the surfactants on the permeation of [^{3}H]methanol and [^{14}C]octanol. Surfactant solutions were prepared in 0.9% sodium chloride solution for irrigation. This constituted the medium in the donor compartment of a two compartment diffusion cell which had a half cell volume of 1.5 ml. Saline was the medium in the receiver chamber. A constant temperature of 37°C was maintained throughout the entire course of the permeation experiment. The contents of each half-cell were stirred at 150 rpm. Samples of the receiver chamber were taken after predetermined time intervals and replaced with neat saline. The samples were processed for liquid scintillation counting and counted in a Beckman LS 1501 (Beckman Instruments, München, Germany) scintillation counter.

After finishing the first set of experiments, care was taken to remove the surfactant quantitatively. The second sequential run was carried out in order to determine the reversibility of the previously observed surfactant effect. With the same membrane still mounted in the diffusion cell, saline constituted the medium in both the donor and the receiver chamber. The diffusion experiment was carried out as described previously.

The thermodynamic activity of methanol and octanol in the respective surfactant solutions was determined by assessing the vapor pressure of each alkanol in the gaseous headspace at 37°C (Cappel and Kreuter, 1991). The ratio of the de-

termined vapor pressure of the individual alkanol in the respective surfactant solution over the vapor pressure of the neat alkanol yielded the thermodynamic activity.

Results and Discussion

Influence of poloxamers on the permeation of methanol and octanol

Methanol Poloxamer 188 (Pluronic F68™) (Fig. 1) does increase the permeability of hydrophilic methanol through full thickness hairless mouse skin at higher concentrations, i.e. 5, 10 and 25% respectively. However, lower concentrations (0.05, 0.5 and 1%) of poloxamer 188 (Pluronic F68™) do not reveal a statistically significant increase of methanol's permeability. The permeability coefficients of methanol through silicone elastomer membrane are basically all of the same order of magnitude, ranging from 40 to 60×10^{-3} cm/h. The permeation behavior of methanol is in pretty good agreement with the thermodynamic activity of methanol in the respective poloxamer 188 (Pluronic F68™) solution. The thermodynamic activity of the hydrophilic alcohol which was determined in separate experiments appears to be invariant at all concentrations of the surfactant.

The other three poloxamers evaluated in this study, i.e poloxamer 338 (Pluronic F108™) (Fig. 2), poloxamer 407 (Pluronic F127™) (Fig. 3), and poloxamer 184 (Pluronic L64™) (Fig. 4) do not enhance the permeability of methanol through hairless mouse skin significantly. Figs 2–4 clearly depict that the permeability coefficients are of a similar order of magnitude, ranging from 2 to 4×10^{-3} cm/h. Notwithstanding the trend that might be suggested by the graphical presentation, the permeability of methanol is not enhanced significantly by any of the three poloxamers. The permeation pattern of methanol through silicone elastomer membrane when applied in poloxamer 338 (Pluronic F108™) (Fig. 2), poloxamer 407 (Pluronic F127™) (Fig. 3), or poloxamer 184 (Pluronic L64™) (Fig. 4) solutions is equivalent to that seen with poloxamer 188 (Pluronic F68™) (Fig. 1).

All effects induced by the poloxamers on hairless mouse skin as well as on silicone elastomer membrane are fully reversible. This finding holds true for methanol in all poloxamer surfactants evaluated in this investigation, i.e. poloxamer 188 (Pluronic F68™), poloxamer 407 (Pluronic F127™), poloxamer 338 (Pluronic F108™), and poloxamer 184 (Pluronic L64™), respectively.

Octanol The thermodynamic activity of octanol decreases slightly as a function of poloxamer 188 (Pluronic F68™) concentration (Table 1). This decrease does not seem to influence the permeability behavior through hairless mouse skin. Here, nearly the same amount of octanol diffuses through hairless mouse skin at higher concentrations of poloxamer 188 (Pluronic F68™). This trend is not supported by the experiment with silicone elastomer membrane. Here, a decrease of octanol's permeability as a function of poloxamer 188 (Pluronic F68™) concentration is evident. The onset of the decrease of the permeability of octanol correlates with the onset of the decline of

TABLE 1

Thermodynamic activity of methanol and octanol in the respective surfactant solution

Concentration (%)	Poloxamer 188		Poloxamer 407		Poloxamine 304		Poloxamine 908	
	MeOH Therm. Act.	OcOH Therm. Act.	MeOH Therm. Act.	OcOH Therm. Act.	MeOH Therm. Act.	OcOH Therm. Act.	MeOH Therm. Act.	OcOH Therm. Act.
0.05	0.0269	0.1266	0.0273	0.1154	0.0268	0.0860	0.0272	0.0670
0.5	0.0286	0.1282	0.0264	0.0767	0.0264	0.0770	0.0286	0.0687
5	0.0277	0.0939	0.0307	0.0181	0.0307	0.0502	0.0282	0.0250
10	0.0309	0.0796	0.0296	0.0053	0.0296	0.0541	0.0295	0.0202

thermodynamic activity of the same solutions of poloxamer 188 (Pluronic F68™). At a concentration of 5, 10 and 25% of poloxamer 188 (Pluronic F68™) the permeability coefficient of octanol through hairless mouse skin is of the order of $50–60 \times 10^{-3}$ cm/h.

This permeation pattern of octanol also distinguishes poloxamer 188 (Pluronic F68™) from

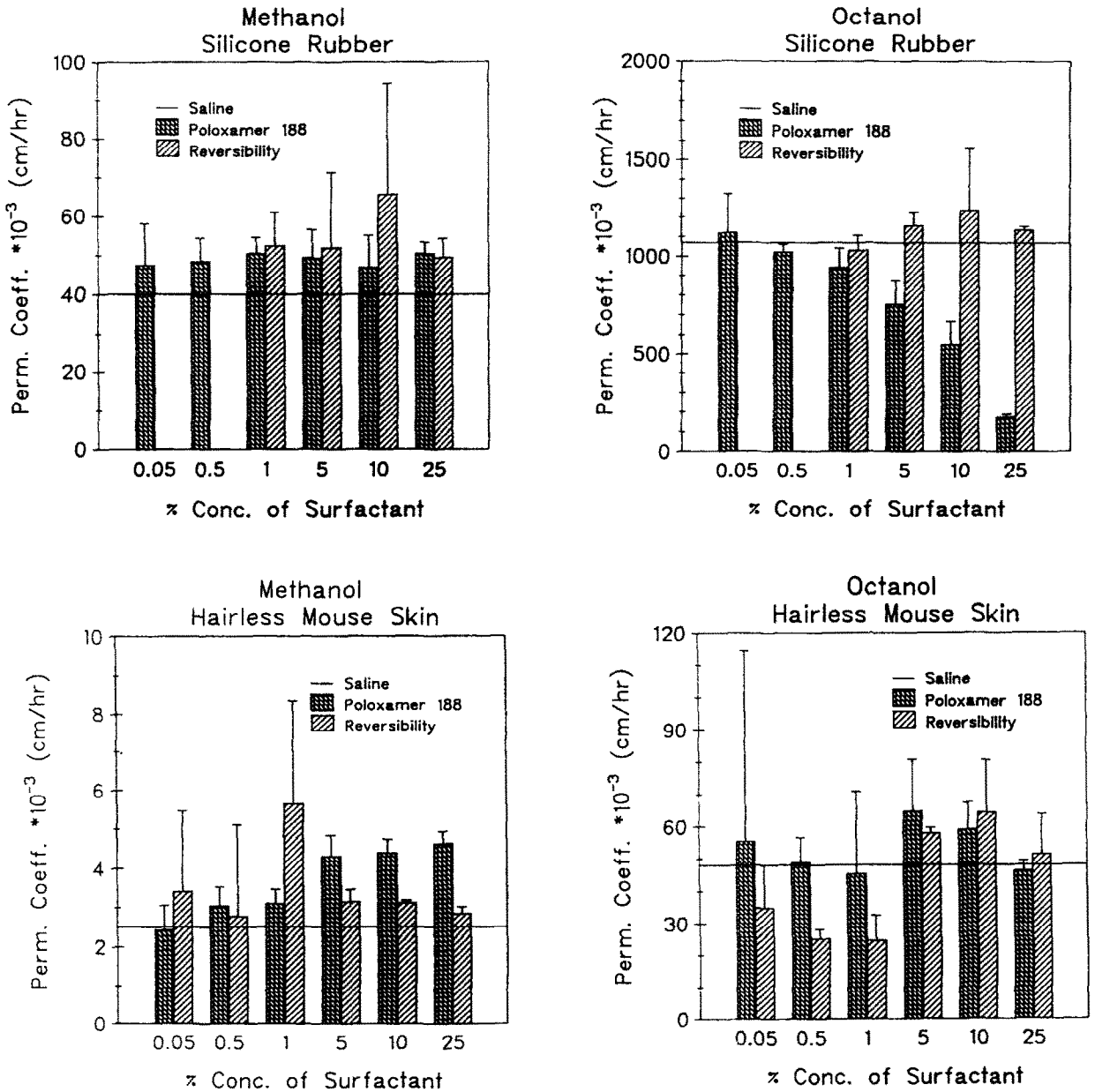

Fig. 1. Effect of different concentrations of poloxamer 188 (Pluronic F68™) on the permeation of methanol and octanol through either silicone elastomer membrane or full thickness hairless mouse skin. All data represent the mean of three experiments ± S.D. The straight line depicts the permeability coefficient of the test permeants when neat saline constitutes the medium in the donor compartment.

the other three poloxamer surfactants tested, i.e. poloxamer 407 (Pluronic F127^{TM}) (Fig. 3), poloxamer 338 (Pluronic F108^{TM}) (Fig. 2), and poloxamer 184 (Pluronic L64^{TM}) (Fig. 4). Here, the permeability coefficient of octanol decreases as a function of surfactant concentration. The permeability coefficients at the equivalent concentrations of 5, 10 and 25% surfactant are of the order of

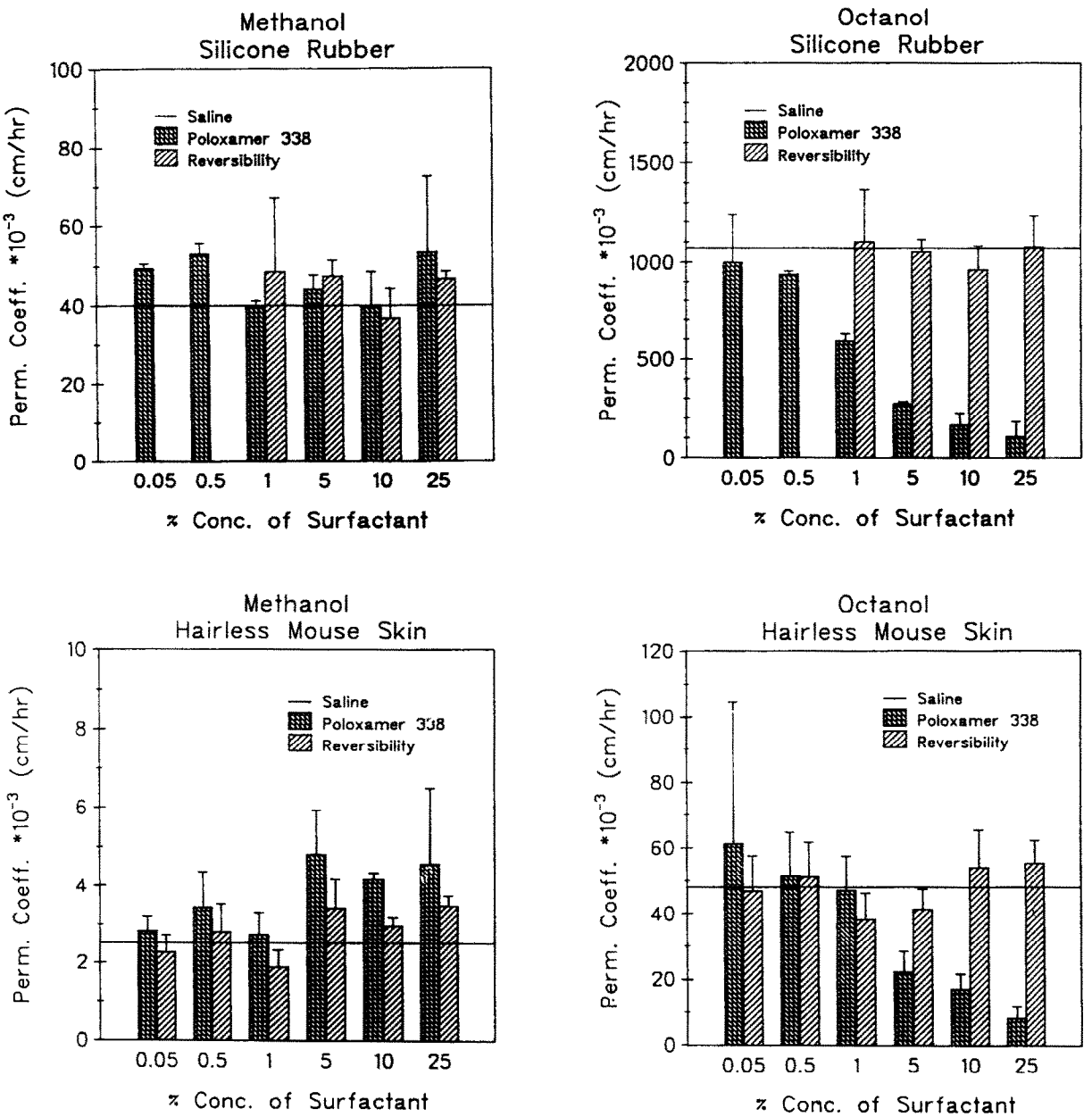

Fig. 2. Effect of different concentrations of poloxamer 338 (Pluronic F108^{TM}) on the permeation of methanol and octanol through either silicone elastomer membrane or full thickness hairless mouse skin. All data represent the mean of three experiments ± S.D. The straight line depicts the permeability coefficient of the test permeants when neat saline constitutes the medium in the donor compartment.

$5–20 \times 10^{-3}$ cm/h, which is approximately one third of the values observed for octanol applied in a poloxamer 188 (Pluronic F68™) vehicle at equivalent concentrations.

Whereas the permeability coefficient of octanol decreases significantly for both poloxamer 407 (Pluronic F127™) and poloxamer 184 (Pluronic L64™), poloxamer 338 (Pluronic F108™) reveals

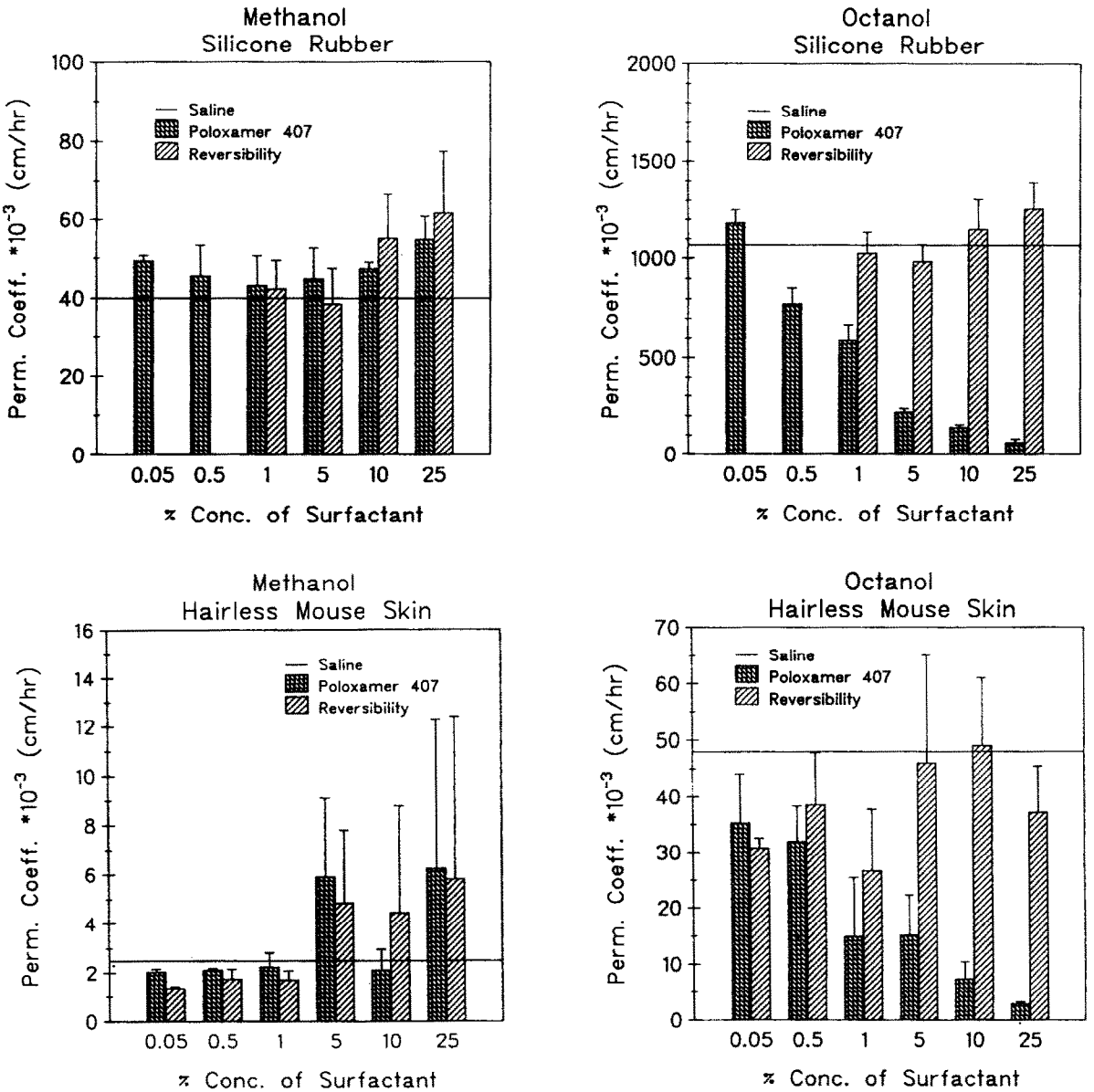

Fig. 3. Effect of different concentrations of poloxamer 407 (Pluronic F127™) on the permeation of methanol and octanol through either silicone elastomer membrane or full thickness hairless mouse skin. All data represent the mean of three experiments ± S.D. The straight line depicts the permeability coefficient of the test permeants when neat saline constitutes the medium in the donor compartment.

a moderate decline. Up to a concentration of 1% poloxamer 338 (Pluronic F108™) there are only minor effects on the permeation of octanol. At higher concentrations a decrease is evident.

The reason for the different effect of poloxamer 188 (Pluronic F68™) on the permeability of octanol through hairless mouse skin might be associated with two possible modes of action of non-

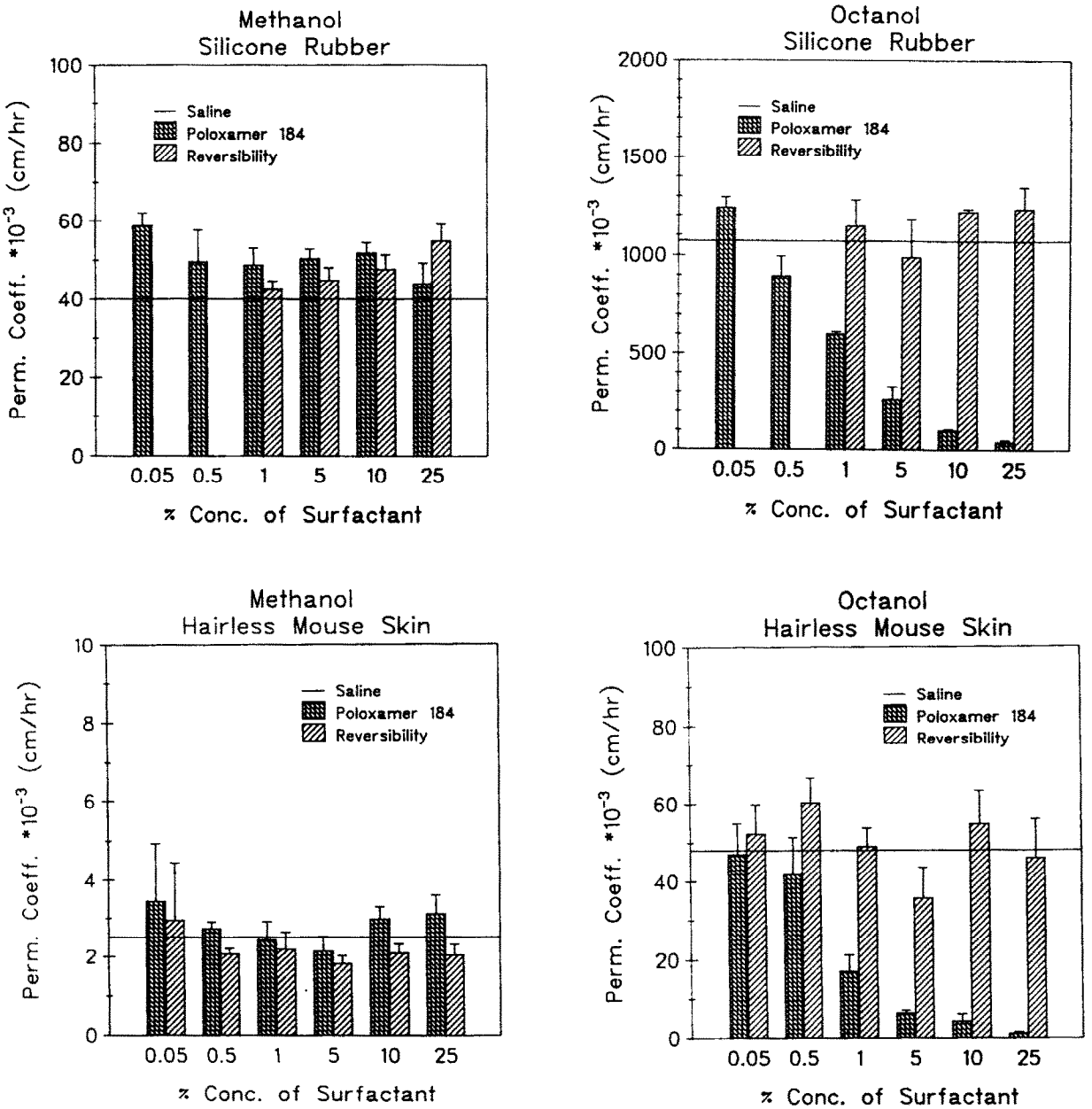

Fig. 4. Effect of different concentrations of poloxamer 184 (Pluronic L64™) on the permeation of methanol and octanol through either silicone elastomer membrane or full thickness hairless mouse skin. All data represent the mean of three experiments ± S.D. The straight line depicts the permeability coefficient of the test permeants when neat saline constitutes the medium in the donor compartment.

ionic surfactants. Stratum corneum lipids might be extracted to a certain degree which yields a nearly invariant permeability coefficient of octanol from poloxamer 188 (Pluronic F68™) solutions. Another mode of action of poloxamer 188 (Pluronic F68™) might be denaturation of skin proteins. However, both possibilities are unlikely, since neither extraction of skin lipids nor denaturation of skin proteins is reversible in in vitro experiments. The reversibility experiments indicate that the permeability of hairless mouse skin returns back to a normal level. Most likely, poloxamer 188's (Pluronic F68™) effect on the permeability of octanol may be attributed to a change of the vehicle/skin partition coefficient in favor of the skin.

Octanol depicts a decrease in permeability through both types of membranes, i.e. hairless mouse skin and silicone elastomer membrane when applied in a poloxamer 407 (Pluronic F127™) vehicle (Fig. 3). This effect can be clearly attributed to a decrease in thermodynamic activity as a function of poloxamer 407 (Pluronic F127™) concentration.

Table 1 depicts the thermodynamic activity of octanol in poloxamer 188 (Pluronic F68™) and in poloxamer 407 (Pluronic F127™). The thermodynamic activity of octanol in poloxamer 188 (Pluronic F68™) solution decreases only marginally whereas it declines significantly in poloxamer 407 (Pluronic F127™) solutions. Unlike poloxamer 188 (Pluronic F68™), poloxamer 407 (Pluronic F127™) forms micelles. One might speculate that octanol is enclosed in micelles thus being unavailable for diffusion. The latter surfactant normally forms a gel in water at concentrations of 20%. Also, the increase in viscosity of the poloxamer 407 (Pluronic F127™) preparation has to be taken into account as a possibility for the observed decrease of octanol's permeability. Evaluating and comparing the trends of all permeation experiments of octanol through silicone elastomer sheeting demonstrates that poloxamer 407 (Pluronic F127™) as well as poloxamer 338 (Pluronic F108™) and poloxamer 184 (Pluronic L64™) retard octanol's permeation due to enclosing the lipophilic solvent in micelles.

Ranking all three poloxamer surfactants which reduce the penetration of octanol through hairless mouse skin in a decreasing order yields the following sequence: (1) poloxamer 338 (Pluronic F108™), (2) poloxamer 407 (Pluronic F127™), (3) poloxamer 184 (Pluronic L64™).

Summarizing the effects of poloxamer surfactants the expectation that they might be promising candidates as skin penetration enhancers has not been verified. Table 2 clearly demonstrates that only poloxamer 188 (Pluronic F68™) enhances significantly the permeation of hydrophilic compounds such as methanol by approximately a factor of 2.

Concurrent application of poloxamer surfactants with lipophilic compounds such as octanol shows no enhancing effect at all. Even a concentration as low as 0.05%, surfactant retards the permeation of octanol. Detailed information on enhancement factors * of poloxamer surfactants is given in Table 2.

Furthermore, variation in neither the molecular weight of poloxamer surfactants, which ranges from 2900 to 14000, nor the HLB values, which range from 15 to 25, seems to affect transdermal drug delivery.

Influence of poloxamines on the permeation of methanol and octanol

Methanol The trends seen with poloxamine 304 (Tetronic 304™) and poloxamine 908 (Tetronic 908™) reflect those of poloxamer 338 (Pluronic F108™), poloxamer 184 (Pluronic L64™), and poloxamer 407 (Pluronic F127™). Actually, all three surfactants, poloxamine 304 (Tetronic 304™) (Fig. 5), poloxamine 904 (Tetronic 904™) (Fig. 6), and poloxamine 908 (Tetronic 908™) (Fig. 7), indicate that they do not influence the percutaneous absorption of methanol significantly. Statistically significant differences appear randomly at some concentrations but no trend is apparent. For this reason, it can be assumed that observed differences are merely the result of biological fluctuations in skin permeability. This finding that the poloxamines do not

* The enhancement factor is defined as the ratio of permeability coefficient with penetration enhancer/permeability coefficient without penetration enhancer.

influence methanol permeability applies for both membranes, i.e. hairless mouse skin and silicone elastomer membrane, respectively. The thermodynamic activity of methanol remains invariant at all concentrations of both surfactants. This result verifies the trend observed in diffusion experi-

TABLE 2

Enhancement factors resulting from surfactant solutions at different concentrations using hairless mouse skin as a membrane

Conc (%)	Poloxamer 188 (Pluronic F68™)				Poloxamer 338 (Pluronic F108™)			
	EF MeOH	*P*	EF OcOH	*P*	EF MeOH	*P*	EF OcOH	*P*
0.05	0.9707	ns	1.1529	ns	1.1267	ns	1.2753	ns
0.5	1.2147	ns	1.0172	ns	1.3720	ns	1.0681	ns
1	1.2413	ns	0.9949	ns	1.0893	ns	0.9797	ns
5	1.7200	s	1.3481	ns	1.9227	ns	0.4635	s
10	1.7533	s	1.2265	ns	1.6627	ns	0.3539	hs
25	1.8440	hs	0.9615	ns	1.8240	ns	0.1711	hs
	Poloxamer 407 (Pluronic F127™)				Poloxamer 184 (Pluronic L64™)			
	EF MeOH	*P*	EF OcOH	*P*	EF MEOH	*P*	EF OcOH	*P*
0.05	0.8133	s	0.7338	ns	1.3800	ns	0.9779	ns
0.5	0.8493	s	0.6649	s	1.0920	ns	0.8710	ns
1	0.9013	ns	0.3106	s	0.9720	ns	0.3570	hs
5	2.3600	ns	0.3159	s	0.8547	ns	0.1305	hs
10	0.8453	ns	0.1542	hs	1.1920	ns	0.0885	hs
25	2.4933	ns	0.0599	hs	1.2453	ns	0.0261	hs
	Poloxamine 304 (Tetronic 304™)				Poloxamine 904 (Tetronic 904™)			
	EF MeOH	*P*	EF OcOH	*P*	EF MeOH	*P*	EF OcOH	*P*
0.05	1.0067	ns	0.9593	ns	0.9600	ns	0.7619	ns
0.5	1.0773	ns	0.6354	ns	1.2267	ns	0.5378	s
1	1.4187	ns	0.7703	ns	1.3640	ns	0.3222	hs
5	0.9880	ns	0.5210	s	0.6667	s	0.0706	hs
10	1.4747	s	0.5817	hs	0.9987	ns	0.0565	hs
25	1.1400	ns	0.1542	hs	1.0093	ns	0.0178	hs
	Poloxamine 908 (Tetronic 908™)							
	EF MeOH	*P*	EF OcOH	*P*				
0.05	1.1840	ns	0.9900	ns				
0.5	1.1347	ns	0.8340	ns				
1	1.1293	ns	0.6197	s				
5	1.0147	ns	0.3858	hs				
10	0.7453	ns	0.1461	hs				
25	1.6960	ns	0.0648	hs				

The enhancement factor (EF) is defined as the ratio of permeability coefficients with/without penetration enhancer. ns, not significant; s, significant ($P \ll 0.05$); hs, highly significant ($P \ll 0.01$).

ments employing silicone elastomer membrane. However, poloxamine 908 (Tetronic 908™) reveals an irregularity in these studies. Apparently, the permeability coefficient of methanol through the silicone elastomer membrane decreases at 0.05, 0.5 and 1% concentration of poloxamine 908 (Tetronic 908™). This trend is consistent neither with the results obtained with other surfactants,

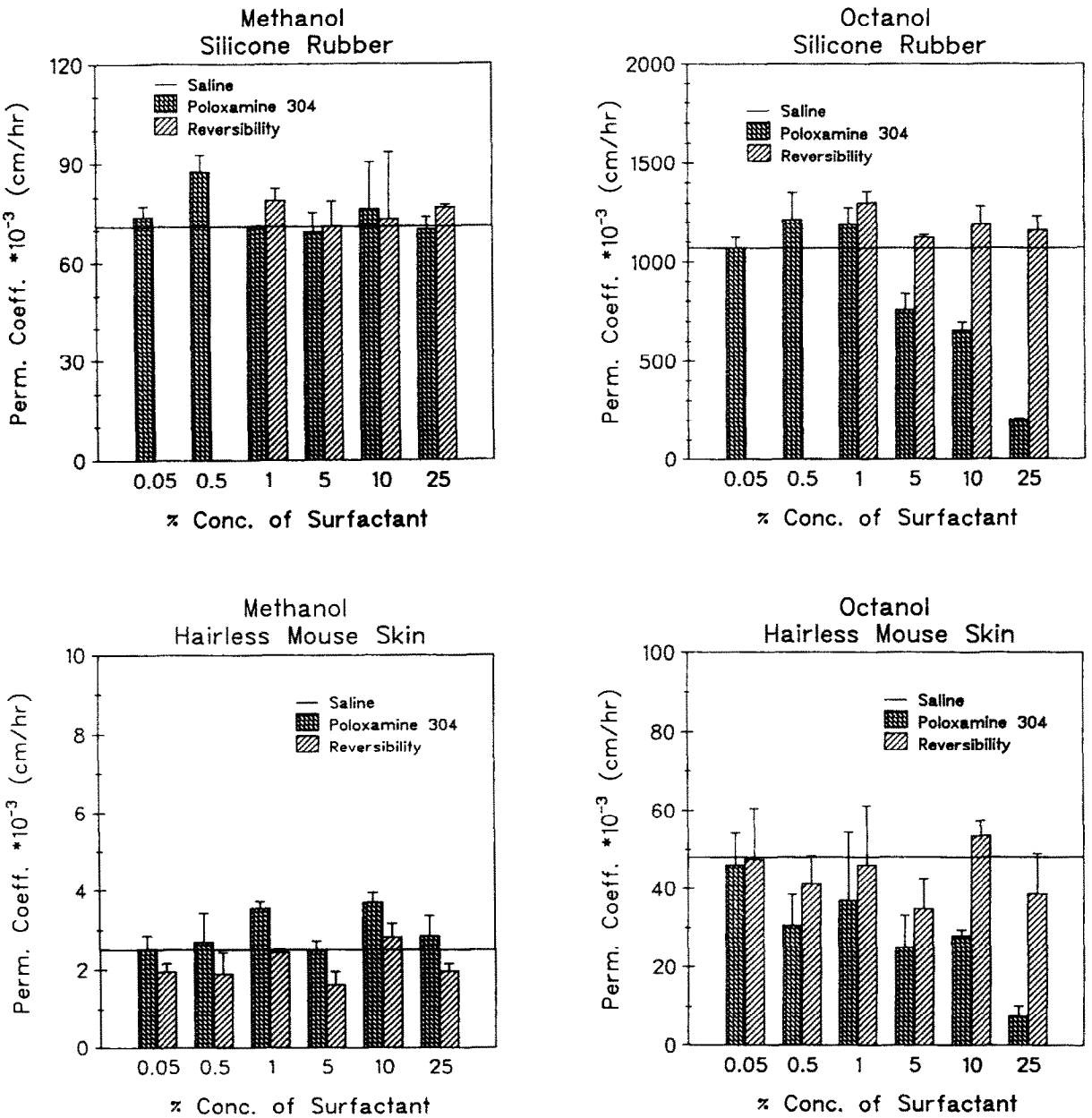

Fig. 5. Effect of different concentrations of Poloxamine 304 (Tetronic 304™) on the permeation of methanol and octanol through either silicone elastomer membrane or full thickness hairless mouse skin. All data represent the mean of three experiments ± S.D. The straight line depicts the permeability coefficient of the test permeants when neat saline constitutes the medium in the donor compartment.

nor with the hairless mouse skin studies nor with the results of the determination of the thermodynamic activity.

Octanol The thermodynamic activity of octanol decreases in poloxamine 304 (Tetronic 304™) as well as in poloxamine 908 (Tetronic

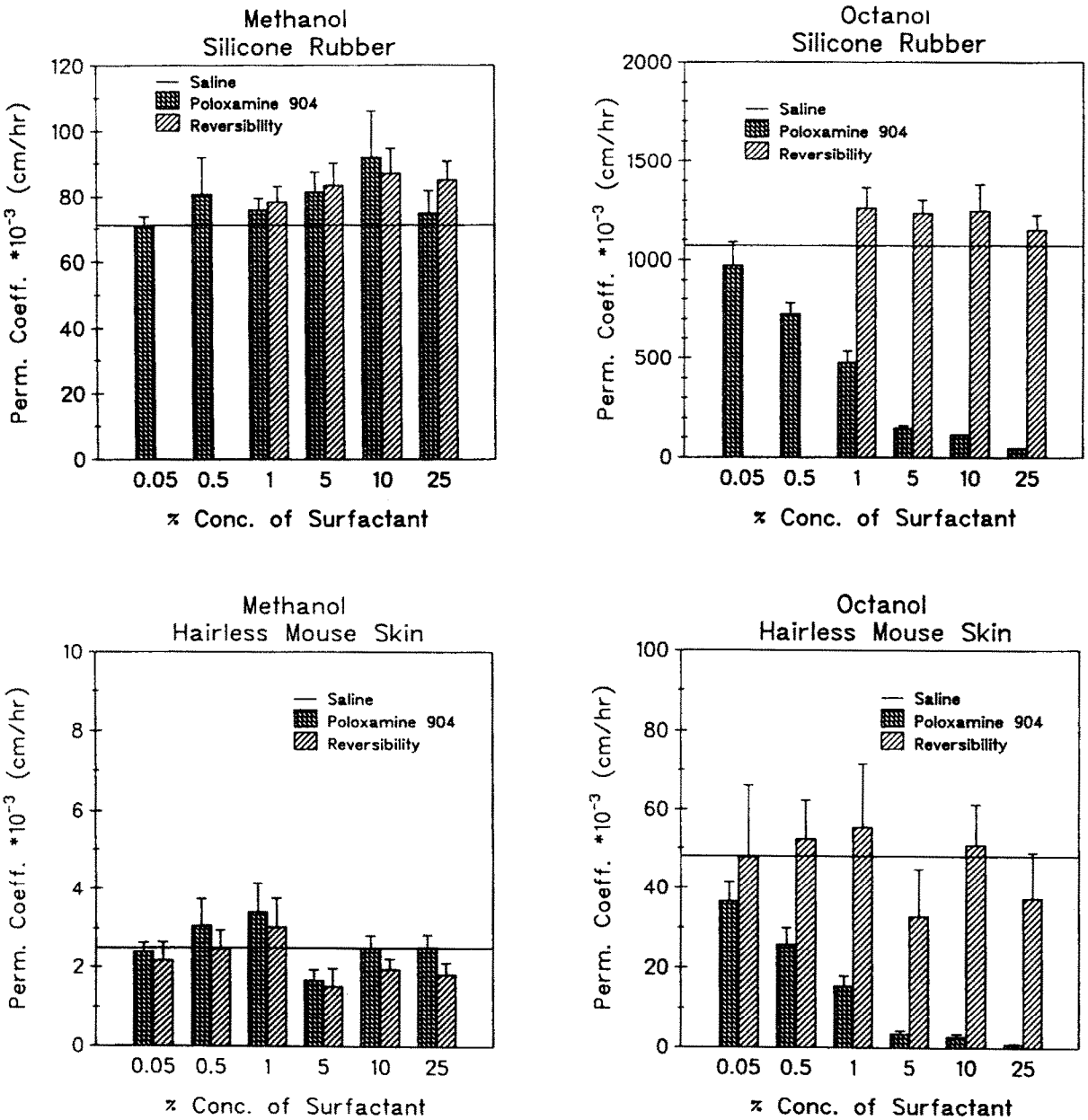

Fig. 6. Effect of different concentrations of poloxamine 904 (Tetronic 904™) on the permeation of methanol and octanol through either silicone elastomer membrane or full thickness hairless mouse skin. All data represent the mean of three experiments ± S.D. The straight line depicts the permeability coefficient of the test permeants when neat saline constitutes the medium in the donor compartment.

908^{TM}). There are differences between both surfactants since it is evident that the thermodynamic activity decreases more when octanol is present in poloxamine 908 (Tetronic 908^{TM}) solutions. This may be attributed to octanol being enclosed in micelles. This finding is in good agreement with the permeation experiments using silicone rubber membrane.

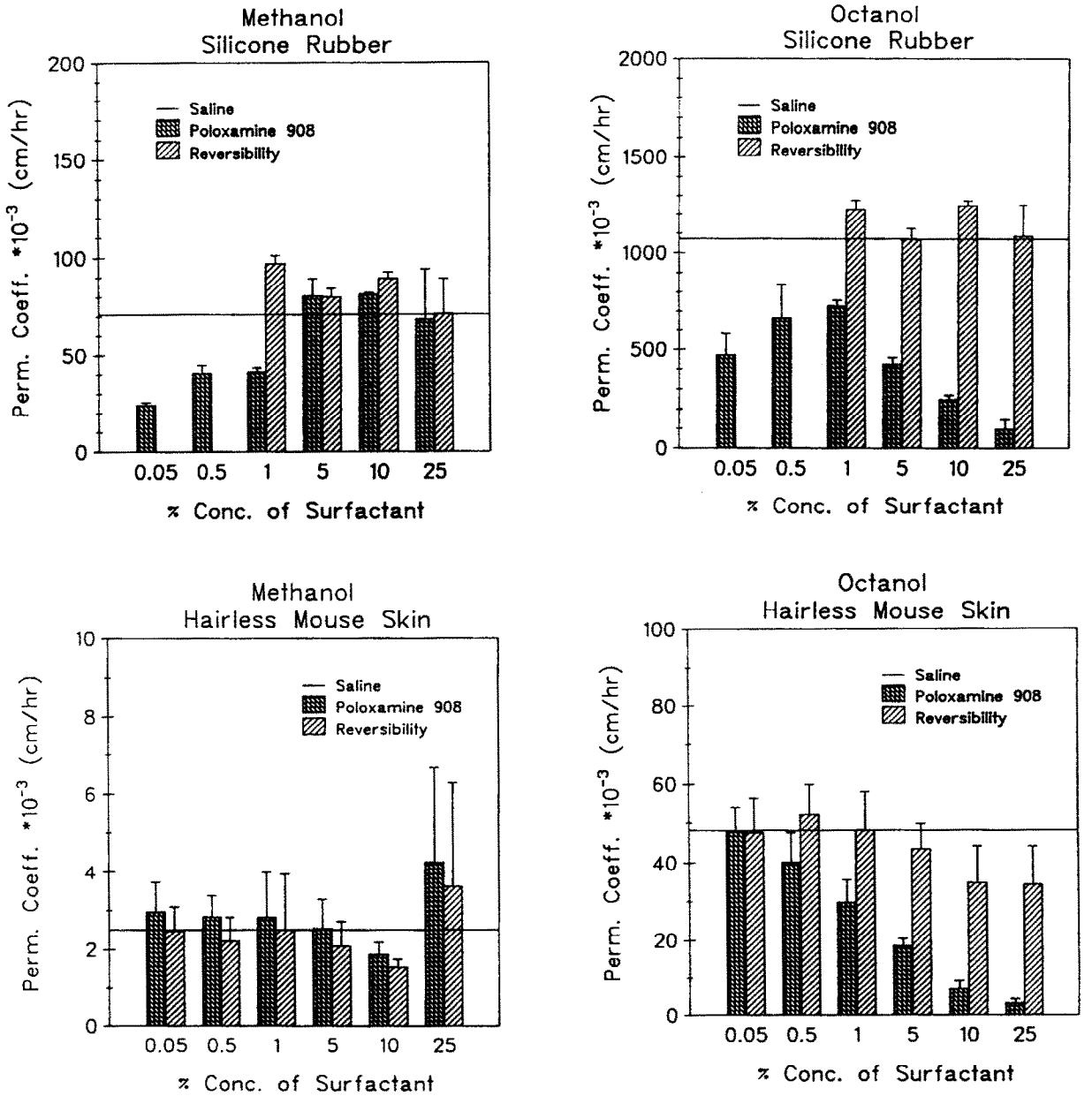

Fig. 7. Effect of different concentrations of poloxamine 908 (Tetronic 908^{TM}) on the permeation of methanol and octanol through either silicone elastomer membrane or full thickness hairless mouse skin. All data represent the mean of three experiments ± S.D. The straight line depicts the permeability coefficient of the test permeants when neat saline constitutes the medium in the donor compartment.

Octanol's permeability decreases as a function of poloxamine 304 (Tetronic 304™) (Fig. 5) concentration when applied to hairless mouse skin. This suggests that poloxamine 304 (Tetronic 304™) does not have the same effect on the permeability of octanol through hairless mouse skin as observed with poloxamer 188 (Pluronic F68™). A comparison between these two nonionic surfactants is interesting since both poloxamer 188 (Pluronic F68™) and poloxamine 304 (Tetronic 304™) showed only a moderate decrease of the thermodynamic activity of octanol at higher surfactant concentrations. Analogous to poloxamer 407 (Pluronic F127™) the permeability decreases as a function of either poloxamine 904 (Tetronic 904™) (Fig. 6) and as well as poloxamine 908 (Tetronic 908™) (Fig. 7) concentration. The permeability coefficients are of the same order of magnitude, ranging from 5 to 15 $\times$ 10^{-3} cm/h.

In conclusion, poloxamine surfactants do not reveal a significant impact on the permeation of hydrophilic compounds such as methanol. The maximum enhancement is approximately by a factor of 1.5. The overall trend shows no influence on methanol at all. However, the permeation of lipophilic octanol through hairless mouse skin as well as through silicone elastomer sheeting is significantly reduced by all poloxamine surfactants. It appears that the molecular weight of the screened poloxamine surfactants as well as the HLB value play no major role in affecting membrane properties of hairless mouse skin. It is evident from all permeation results of poloxamine surfactants that they mirror the physico-chemical as well as the biological properties of their poloxamer counterparts.

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