

DERMAL EFFECTS OF COMPOSITIONS BASED ON THE EUTECTIC MIXTURE OF LIGNOCAINE AND PRILOCAINE (EMLA)

Studies in Volunteers

H. EVERS, O. VON DARDEL, L. JUHLIN, L. OHLSÉN AND E. VINNARS

Although it is the cationic form of a local anaesthetic agent which blocks the transmission of impulses within the nerve (Covino and Vassallo, 1976), it is the uncharged (base) form which penetrates and diffuses into the tissues after topical or parenteral administration. Effective tissue penetration can be achieved after topical application as follows:

(1) *Water soluble salt in water.* In an aqueous solution of the hydrochloride salt of an amino-amide local anaesthetic, the concentration of the uncharged form depends on the pK_a , which means that only a portion of the local anaesthetic is in the form suitable for tissue penetration. Consequently, to be effective on topical application, the local anaesthetic must be used in high concentration, with the attendant risk of toxic complications. On the other hand, the presence of water in such formulations increases the likelihood of penetration through the skin (Monash, 1957).

(2) *Base in alcoholic solvent.* Since local anaesthetic bases are freely soluble in various alcoholic solvents, it is possible to obtain a high concentration of the base by dissolving it in, for example, ethyl alcohol, as in local anaesthetic sprays or ointments. As such compositions do not contain water, which would precipitate the local anaesthetic, they have poor penetrative properties if applied to the intact skin. They are, however, active on application to mucous membranes (Dalili and Adriani, 1971; Adriani and Dalili, 1971).

(3) *Oil-in-water emulsion.* Recent pharmaceutical research has shown that it is possible to combine a high concentration of the local anaesthetic base

SUMMARY

The effects of the cutaneous application of EMLA cream (a eutectic mixture of lignocaine and prilocaine in their base form) were studied in volunteers. When tested by pin-prick, EMLA cream 2.5% and 5% produced analgesia of the area tested, the cream being most effective if left in contact with the skin for 60 min. The pain produced by the insertion of an i.v. cannula was successfully blocked by the application of this formulation, especially if applied to the antecubital area. Temporary blanching of the skin areas was frequently observed on removal of the occlusive tape bandages, but prolonged, or repeated, application of 5% EMLA cream did not produce local skin reactions. Tests for delayed hypersensitivity reactions were negative. Plasma concentrations of lignocaine and prilocaine were low after a standard application.

with a high water content by using oil-in-water emulsions (Broberg and Evers, 1981; Brodin et al., 1984), thus achieving an increased uptake of local anaesthetic via the intact skin (by including water in compositions containing the base). This seemingly paradoxical effect is achieved because the emulsion droplets contain the local anaesthetic base in high concentration (e.g. 20% lignocaine) while the total concentration in the emulsion is low (only 5% lignocaine).

A further increase in analgesic efficacy has been demonstrated with oil-in-water emulsions, based on the eutectic mixture of lignocaine and prilocaine (Broberg and Evers, 1981). Brodin and colleagues (1984) reported that a mixture of lignocaine and prilocaine in base form had a melting point considerably lower than that of each ingredient. This means that two crystalline powders interact in such a way that, when mixed with each other,

HANS EVERS, D.D.S., Pain Control, Astra Läkemedel AB, Södertälje, Sweden. OTTO VON DARDEL, M.D., Department of Anesthesiology, St Göran Hospital, Stockholm, Sweden. LENNART JUHLIN, M.D. (Department of Dermatology); LENNART OHLSÉN, M.D. (Department of Plastic Surgery); University Hospital, Uppsala, Sweden. ERIK VINNARS, M.D., Department of Anaesthesiology, St Erik Hospital, Stockholm, Sweden.

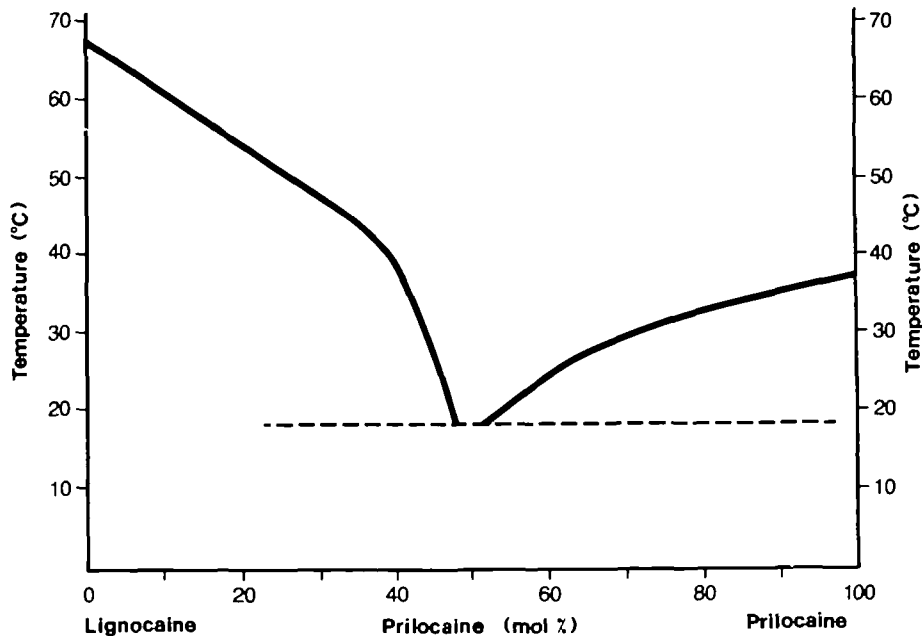


FIG. 1. Phase diagram of the lignocaine/prilocaine system. At around prilocaine 50 mol% mixed with lignocaine, the system is liquid at 18 °C. At lower and higher prilocaine and lignocaine concentrations, the mixture is solid in crystalline form. (Modified from Brodin et al., 1984.)

a liquid is formed at approximately room temperature (fig. 1). This is of significance in the preparation of oil-in-water emulsions as it obviates the need for dissolving the local anaesthetic base in an oil before the addition of the emulsifier. By this method it is possible to increase further the concentration of local anaesthetic in the emulsion droplets from approximately 20% (lignocaine) to about 80% (lignocaine-prilocaine) in eutectic formulations, while still keeping the total concentration of local anaesthetic low (5%).

Although it has been possible to use several different local anaesthetics, or combinations of more than two substances, to make eutectic mixtures (Broberg and Evers, 1981), the choice of lignocaine and prilocaine was obvious for many reasons, particularly in regard to their known margins of safety. The oil-in-water emulsions in our studies were based upon a 50:50 eutectic mixture of lignocaine and prilocaine bases in combination with a conventional and documented emulsifier, Arlatone, and water.

The aim of the present investigation was to study the dermal analgesia produced by the application, to the skin, of compositions based upon the eutectic mixture of lignocaine and prilocaine, and to study the local effects on the skin

after prolonged and repeated application. In addition, evidence of delayed hypersensitivity reactions was sought. Plasma concentrations of lignocaine and prilocaine were measured following a standard application.

SUBJECTS, MATERIALS AND METHODS

Volunteers, between 20 and 40 yr and of either sex, were medically examined before the investigation. A full medical history was obtained and a complete blood analysis was carried out. Any volunteer with a previous reaction to any local anaesthetic was excluded. A double-blind technique with placebo control was used in all the series. Active or placebo formulations were applied to the different test areas according to predetermined randomized patterns.

Dermal analgesia

(1) *Influence of concentration and duration of application on dermal analgesia.* The test compositions of this series consisted of 1.0%, 2.5% and 5% EMLA emulsions and placebo.

Porous cellulose-fibre pads (2 × 3 cm) soaked with the different formulations were applied on the volar side on both arms. Occlusive tape bandages

(Blenderm (3M)) were applied to cover the pads. The bandages were removed after 30 min on one arm, and after 60 min on the other arm. Dermal analgesia was tested with a modified pin-prick technique, using the relatively blunt reverse side of a disposable 27-gauge sterile dental needle (Monoject, Sherwood). Each needle was used on one test occasion and then discarded. Each skin test area was pin-pricked 10 times, the needle pricks covering the whole test area. This test was carried out immediately after removal of the bandage, and at 30-min intervals for up to 240 min. A score index was obtained from the volunteer, by noting the number of painfree pin-pricks out of the 10 in each area. The volunteers were fully instructed before each study on how to differentiate between the perception of touch and that of sharp pain. During the investigation each volunteer's ability to discriminate between these two types of perception was assessed by pin-pricking outside the treated skin areas also. All pin-prick studies were carried out by the same investigator (H.E.).

(2) *Influence of viscosity on dermal analgesia.* The technique used in this series was identical to that used in the first series, with one exception: instead of pin-pricking the whole test area at each 30-min test, the test skin area was divided in six equal areas, each of which was pin-pricked on each test occasion. In this way a fresh test area was pin-pricked each time, minimizing the local circulatory effects produced by the needle. Using 2.5% and 5% EMLA compositions, the viscosity was modified using Carbopol in three different concentrations. This provided three different viscosities in the test formulations: an emulsion (no viscosity increasing agent), a lotion and a cream formulation. A 5% lignocaine ointment (Xylocaine 5% ointment, Astra) and a placebo were included in the series for comparison. Application time was 60 min.

(3) *Test of dermal analgesia by skin penetration and intravascular placement of i.v. needles.* After application of 5% EMLA or placebo, and following the use of an occlusive dressing for 60 min, the pain on needle insertion through the skin, and the subsequent intravascular placement of standard i.v. needles (Venflon 140, 17 gauge, 1.4 mm o.d., 45 mm b.s. Luer-lock) was assessed.

The skin areas studied were the dorsum of the hand, the lower arm area and the antecubital fossa. The pain experienced by the volunteers was recorded when inserting the needle, immediately

after removal of the occlusive bandages and after intravascular placement of the needle. The second procedure, moving the needle to an intravascular position after insertion, was made after the initial pain of skin penetration had receded completely. The pain produced by the two procedures was classified as no pain (= 0), slight pain (= 1) and painful (= 2).

Tolerance studies

(4) *Prolonged application.* EMLA 5.0% cream or placebo were applied under occlusive tape bandages on the skin of the back. Application time was 24 h. Signs of local irritation were recorded immediately after removal of the bandages as: - = no reaction, + = erythema, ++ = erythema and papules, +++ = vesicles.

(5) *Repeated application to the same area of skin.* EMLA 5.0% cream or placebo cream was applied on marked areas of the skin on the volar surface of the arms. The same areas were treated with one, two and three applications (60 min) with 24-h intervals. Recording of signs of local irritation was made on three different occasions, immediately on removal of the bandages. The scale used by the recording of local effects was the same as that used in the second series of investigations.

Signs of local irritation were recorded after one, two and three applications.

(6) *Test of delayed hypersensitivity reaction.* EMLA 5.0% cream, EMLA 2.5% cream, 5.0% lignocaine cream or placebo cream were applied in a standard epicutaneous skin test procedure (AL-test, Imeco), on the skin of the back. Application time was 48 h and the skin reactions were observed 50 h after application.

All volunteers taking part in the study of delayed hypersensitivity had been previously exposed to EMLA cream on several occasions, in other series of investigations.

Pharmacokinetic studies

(7) *Percutaneous absorption of lignocaine and prilocaine.* EMLA 5% cream was applied to the skin on the lateral side of one thigh with an application time of 60 min. To ensure a standard area of application, a plastic frame with a central opening of 300 cm² was applied to the skin of the thigh, enabling a thick layer of the cream to cover an identical area in all subjects. In each volunteer, 20 g of the cream, containing 500 mg of each local anaesthetic, was used. After 60 min application, the occlusive bandage was removed and pin-

pricking of the test areas was carried out every 30 min for up to 240 min using the same technique as in the previous series. Blood samples were drawn from an antecubital vein at 30, 60, 90, 120, 180 and 240 min after the application of the bandage. Plasma concentrations of lignocaine and prilocaine were analysed by mass fragmentography, carried out at the Department of Analytical Chemistry, Astra Läkemedel AB, Södertälje, Sweden.

(8) *Influence of preoperative skin cleaning on plasma concentrations of lignocaine and prilocaine.* The application procedures and amounts of 5% EMLA cream used were identical to those used in

the previous series, as were the assessment by pin-prick and the blood sampling procedure. Immediately after removal of the occlusive bandage, the application site was cleansed for 2 min by a qualified nurse, who simulated the procedure of regular preoperative cleaning. The disinfectant used contained 0.5% chlorhexidine digluconate in 70% alcohol.

RESULTS

Dermal analgesia

(1) *Influence of concentration and duration of application on dermal analgesia.* EMLA 2.5% and

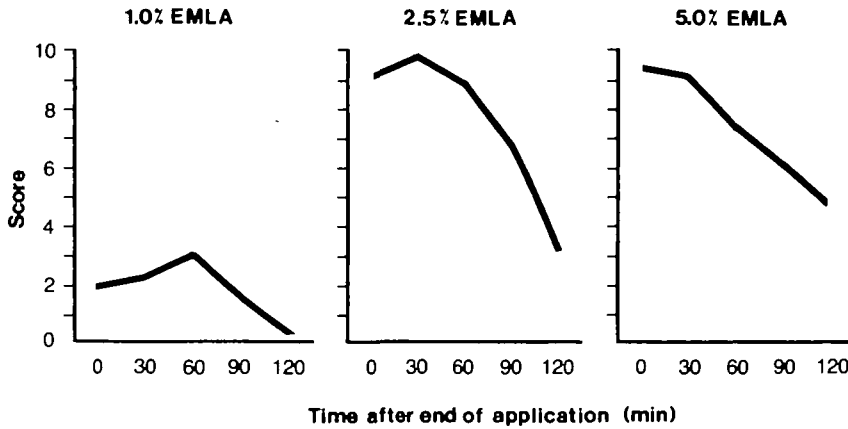


FIG. 2. Influence of concentration on dermal analgesia. Mean painfree scores at different times of observation, after 60 min applications of 1.0%, 2.5% and 5.0% EMLA emulsions. Number of volunteers = 12.

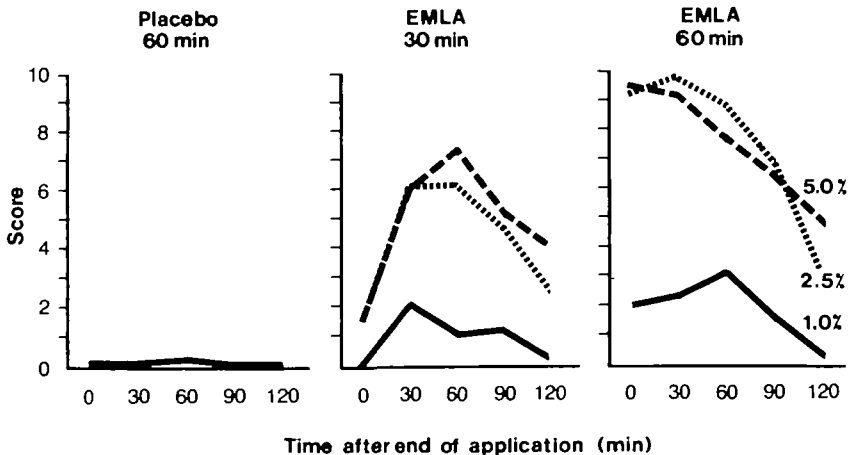


FIG. 3. Influence of duration of application on dermal analgesia. Mean painfree scores at different times of observation after 30- and 60-min applications of 1.0%, 2.5% and 5.0% EMLA emulsions or placebo. Number of volunteers = 12.

5.0% emulsions were effective in producing analgesia of the skin, while the lower concentrations and the placebo were significantly less effective (fig. 2).

On removal of the occlusive bandage after 30 min application, the dermal analgesia was poor with all formulations. When tested 30 min later, however, dermal analgesia had improved significantly. After 60 min application, both the 2.5% and the 5% EMLA emulsions were found to be highly effective in blocking the pain of pin-prick (fig. 3).

(2) *Influence of viscosity on dermal analgesia.* Dermal analgesia was not influenced by the addition of the two tested concentrations of Carbopol, a viscosity increasing agent (fig. 4).

(3) *Test of dermal analgesia by skin penetration and intravascular placement of i.v. needles.* The pain felt by the volunteers on the insertion of the needle through the skin and on intravascular placement was blocked to a significantly greater extent by 5% EMLA than by placebo in the antecubital area and on the dorsum of the hand. The differences between the two formulations were insignificant when applied on the lower arm area (fig. 5). If applied in the antecubital area, 5% EMLA cream was completely effective in blocking the pain of needle insertion in all volunteers.

Tolerance studies

(4) *Prolonged application.* None of the applied formulations, 2.5% EMLA, 5.0% EMLA or placebo, caused any signs of local irritation in any of the volunteers, with the exception of two areas in which very slight redness was observed after removal of the bandage covering the 2.5% EMLA formulation. Outside these two areas there was some irritation from the tape bandage.

(5) *Repeated application.* The repeated application of either the 5% EMLA cream or the placebo cream did not cause any signs of local irritation in any of the subjects.

(6) *Test of delayed hypersensitivity reaction.* No delayed skin reactions were observed on any of the areas to which 2.5% EMLA, 5% EMLA or placebo was applied.

Pharmacokinetic studies

(7) *Percutaneous absorption of lignocaine and prilocaine.* After removal of the occlusive dressing, applied to the lateral side of the thigh and covering 300 cm² of the skin area for 60 min, the blood concentrations of lignocaine and prilocaine were found to be low (fig. 6). However, the variation between subjects in the concentration of both substances was high (table I).

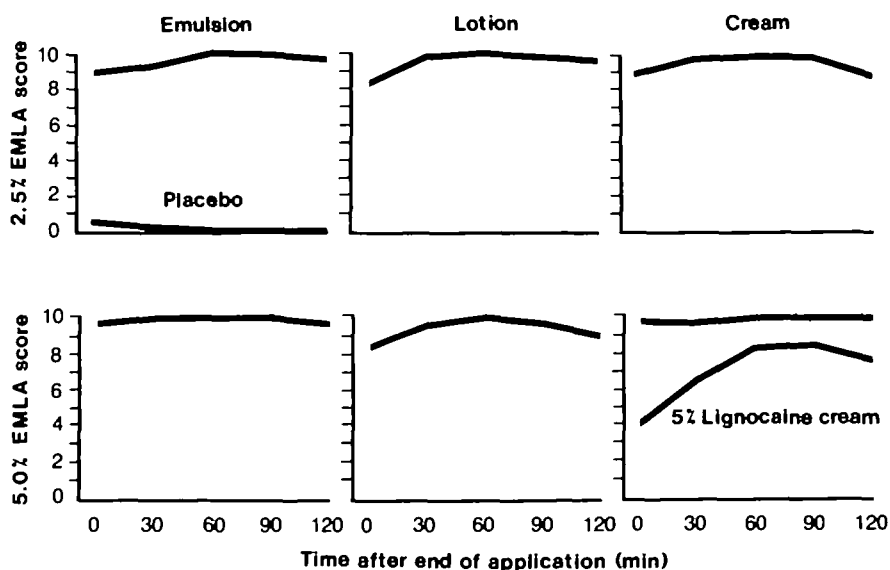


FIG. 4. Influence of viscosity on dermal analgesia. Mean painfree scores at different times of observations after 60-min applications of 2.5% and 5.0% EMLA emulsion, lotion and cream, 5.0% lignocaine cream or placebo. Number of volunteers = 12.

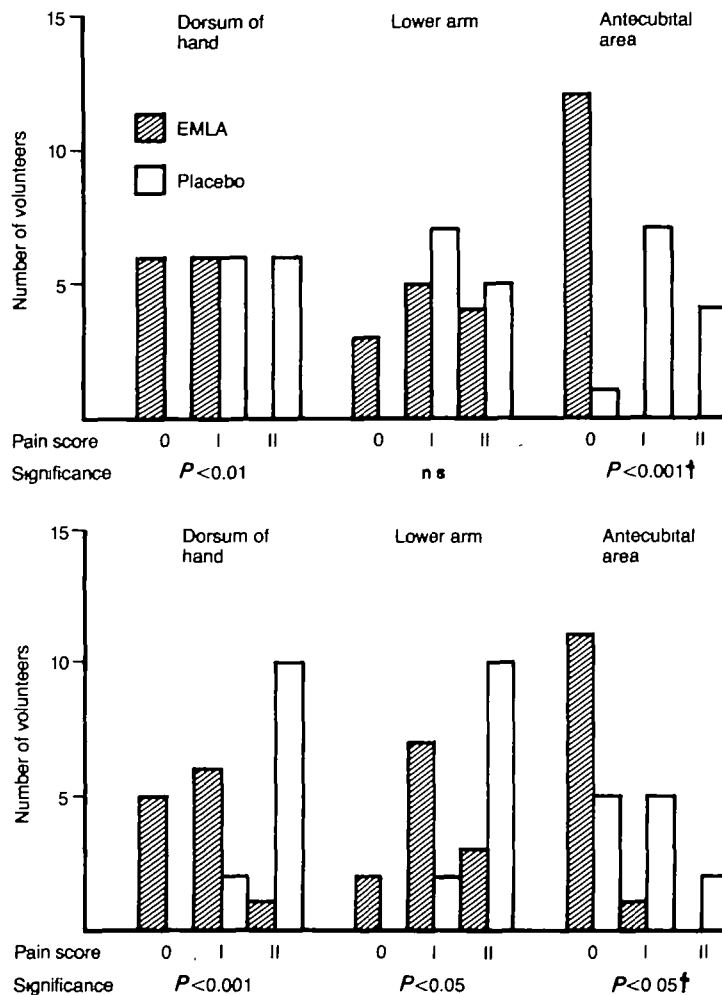


FIG. 5. Pain scores on insertion of i.v. needle: top = skin penetration; bottom = vessel wall penetration. Number of volunteers = 12. \uparrow McNemar test.

TABLE I. Mean anaesthetic concentrations (ng/ml plasma), all volunteers. Statistical comparison was by paired t test

	Time after application (min)					
	30 (n = 20)	60 (n = 20)	90 (n = 20)	120 (n = 20)	180 (n = 19)	240 (n = 20)
Lignocaine (ng ml ⁻¹)	8.2	24.0	24.0	68.4	74.5	65.4
SD	7.02	27.68	27.68	47.95	35.26	25.12
Prilocaine (ng ml ⁻¹)	1.2	3.6	13.4	21.4	24.7	19.9
SD	3.83	7.87	17.00	19.35	14.93	12.19
Lignocaine/ prilocaine						
t	3.91	3.17	1.46	4.07	5.67	7.29
P	< 0.001	< 0.01	ns	< 0.001	< 0.001	< 0.001

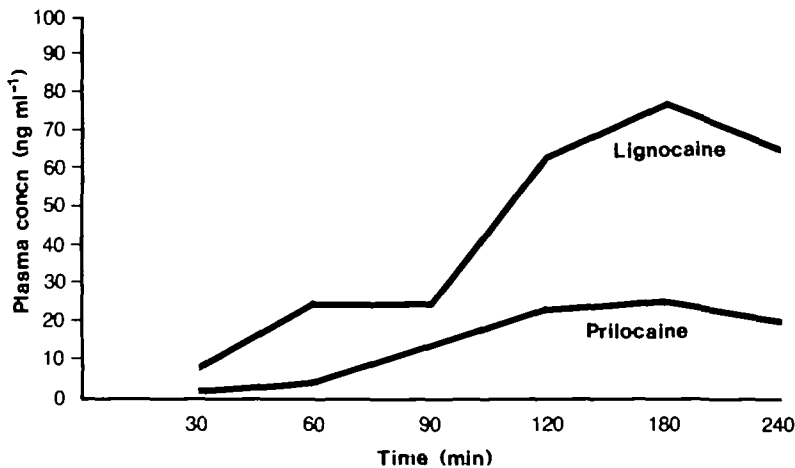


FIG. 6. Mean plasma concentrations of lignocaine and prilocaine. Different times of observation after a 60-min application of 5.0% EMLA cream on a 300-cm² skin area on the lateral side of the thigh. Number of volunteers = 20.

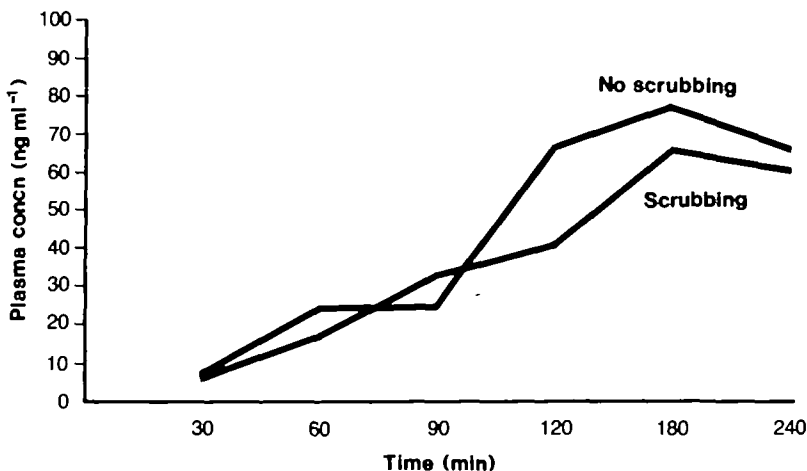


FIG. 7. Mean plasma concentrations of lignocaine. Different times of observation after a 60-min application of 5.0% EMLA cream on a 300-cm² skin area on the lateral side of the thigh, with and without preoperative cleansing of the skin. Number of volunteers = 20.

(8) *Influence of preoperative cleaning upon lignocaine and prilocaine concentrations.* The cleansing procedure immediately after removal of the occlusive bandage decreased the concentrations of lignocaine in the blood samples compared with the values found in the previous series, where no cleansing had been carried out (fig. 7). The prilocaine concentrations were not influenced by the cleansing procedure (fig. 8). As in the previous series, the variation in serum concentrations between subjects was substantial (table II).

DISCUSSION

New formulations based on the eutectic mixture of two, widely used, local anaesthetic substances, lignocaine and prilocaine (in their base form) have been investigated.

The pin-prick studies demonstrated clearly that concentrations lower than 2.5% of lignocaine/prilocaine were significantly less active in producing dermal analgesia than the 2.5% and 5.0% preparations. In addition, it was noted that an

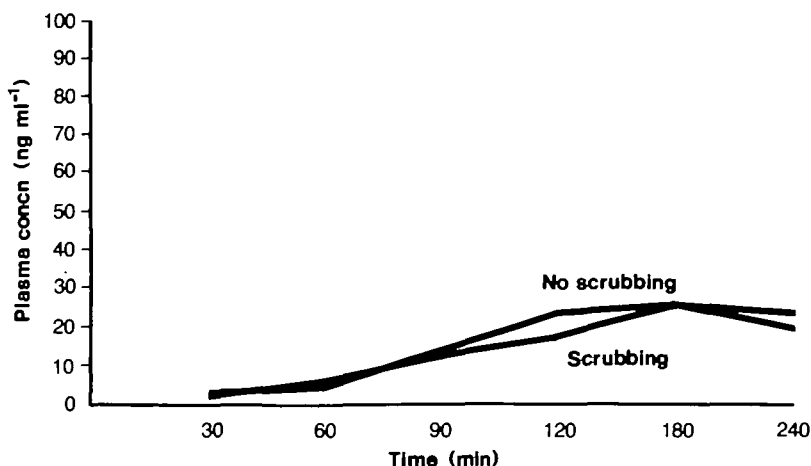


FIG. 8. Mean plasma concentrations of prilocaine. Different times of observation after a 60-min application of 5.0% EMLA cream on a 300-cm² skin area on the lateral side of the thigh, with and without preoperative cleansing of the skin. Number of volunteers = 20.

TABLE II. Mean anaesthetic concentrations (ng/ml plasma), all volunteers, after the cleansing procedure

	Time after application (min)					
	30 (n = 20)	60 (n = 20)	90 (n = 20)	120 (n = 20)	180 (n = 20)	240 (n = 20)
Lignocaine (ng ml ⁻¹)	6.3	18.2	31.9	40.8	66.0	59.5
SD	9.82	24.86	29.20	30.87	34.32	28.90
SEM	2.1958	5.5589	6.5293	6.9027	7.6742	6.4622
Prilocaine (ng ml ⁻¹)	1.0	4.4	12.5	17.5	24.6	22.0
SD	2.58	11.65	15.80	15.18	13.40	11.03
SEM	0.5769	2.6050	3.5330	3.3944	2.9963	2.4664

application time of 60 min under an occlusive dressing was desirable to achieve a high frequency of painless pin-pricks. An application time of 30 min produced limited analgesia, although, when tested 30 min later, the analgesia scores had increased to values similar to those after 60 min application time. A preparation with a higher viscosity than that of the emulsion can be of clinical interest from a practical point-of-view; for example, ease of application. In our studies it was found that the production of dermal analgesia to pin-prick was not altered by the addition of a viscosity increasing agent. In those series of experiments where the pin-pricks had been distributed evenly over the whole test area, the

duration of dermal analgesia was comparatively short. When the pin-prick testing was modified, and a fresh area was used at each test, the duration of analgesia was found to be much longer. This may have clinical consequences, since any disturbance of the local circulation, by scratching or rubbing, may decrease the duration of dermal analgesia.

In the series in which the pain induced by the introduction of an i.v. needle and its subsequent placement in a vein was investigated, the difference between the active formulation and placebo was insignificant in the lower arm area. In the two other areas studied (the antecubital vein and the dorsum of the hand) the difference was significant

in favour of the 5% EMLA cream. It is interesting to observe that these differences were also observed as the needle penetrated the vessel wall, at a distance of about 10 mm from the point of skin puncture.

Skin reactions to the clinical use of 5% EMLA, applied to intact skin, are unlikely. The temporary pallor of the skin treated with 5% EMLA cream seems to be caused by constriction of the peripheral capillary bed although the precise mechanism is unknown at present. In pilot studies, using a conventional skin temperature probe, the temperature of the skin in the areas showing a blanching effect was not found to be lower than that of the untreated skin areas outside the site of application. Investigations aimed at finding an explanation of this phenomenon are under way at present.

When 5% EMLA cream 20 ml was applied to the lateral surface of the thigh for 60 min, the plasma concentrations in blood sampled from an antecubital vein were found to be significantly lower than those considered likely to cause toxic effects. The prilocaine concentrations were found to be lower than those of lignocaine and so accord with previously investigated distribution properties of the two agents (Scott et al., 1972).

It was interesting to find that the concentrations of lignocaine were decreased more after the pre-operative cleansing procedure than the prilocaine concentrations, when one considers that identical amounts of both agents were applied. The phenomenon is difficult to explain as both local anaesthetics are freely soluble in ethyl alcohol.

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