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Transdermal lidocaine patch 5% and lidocaine cream 5%: a PK/PD approach in the horse

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Summary

Transdermal absorption of four lidocaine (L) patches 5% was compared to the transdermal absorption of L cream 5% to evaluate the pharmacokinetics of the two formulations applied on the same anatomical region under dressing in eight horses. The animals were also assessed for the adequacy of analgesia after patches and cream removal, using a psychophysical method by pricking the patient's skin to assess the response to pain according to a visual analogue scale.

Horses were randomly assigned to four treatment groups: in groups I and II four L patches were applied for a period of 24 hours with and without alcohol pre-cleaning respectively; in group III, 5% L cream was applied every two hours over a period of 24 hours on the same anatomical site. Group IV was the control. No clinical side effects were noted with both formulations. L was detectable in plasma within 6 and 24 hours after patch application and the highest plasma concentrations were reached between 12 and 18 hours. The use of alcohol to pre-clean the skin appeared to reduce the transdermal drug absorption over time. The plasma drug level, after L cream application, reached the highest plasma concentration after 24 hours. Pain assessment after L patch or L cream application, revealed a decreased response when the L cream was used. The result of this study shows that there is an overall lower absorption from the L patches compared with the L cream in horses. Also the L cream treatment reduces significantly the intensity of pain quality as measured by the visual analogue scale.

Keywords: lidocaine, transdermal patch, 5% cream, horses

Lidocaine (L), brand name Xylocaine, is a widely used anaesthetic in veterinary medicine and especially in equine practice (4, 32). When used intravenously, it is 60% protein bound (19). It is usually administered as a perineural infiltration to produce nerve block when diagnosing limb lameness; its beneficial effect has been reported to decrease the duration of ileus after abdominal surgery (5) due to the prokinetic effect of the drug. Another topical preparation is the eutectic mixture of local anaesthetic (EMLA) cream, containing L and prilocaine (2.5% each), commonly used in veterinary medicine prior to painful minor procedures such as episioplasty, placement of jugular catheters in hospitalized patients and a pain-free venepuncture in laboratory animals (7, 8, 29).

Several studies on humans show that L can also be applied to the skin as a topical preparation (18, 28). The exact mechanism is unknown, but it has been suggested that topical L formulations deliver adequate amounts of L to block sodium channels on small damaged or dysfunctional pain fibers but insufficient amounts to block sodium channels on large myelinated A-β fibers (11).

Recently, a transdermal drug delivery system has been described in feline (15), canine (14, 33) and equine species (1). The L patch is considered to be a single-layer matrix patch that allows the drug to diffuse across the upper layers of skin via passive diffusion from an area of high concentration to an area of low concentration (6). The L patch is a topical analgesic transdermal formulation approved for use in 1999 to treat post-herpetic neuralgia (PHS) in human medicine (10). This new technique has gained wide acceptance in small animal pain management when applied on the skin of patients that have undergone abdominal surgery, limb amputation, thoracotomy, haemilaminectomy, cruciate repair, ear canal ablation (14, 33), close to the site of pain, to function locally reducing both the generation and conduction of peripheral pain impulses in dysfunctional or damaged nociceptors

situated directly under the patch application site. One of the reasons why the L patch has gained acceptance both in human and veterinary medicine is because of its low systemic absorption and therefore less side effects (10, 11, 14, 33). The L patch is associated with local analgesia rather than local anaesthesia, as humans do not report numbness or loss of sensitivity to touch, pressure, or temperature (10). Limited options for analgesia in equine patients and the importance of systemic effects caused by opioid drugs make the transdermal technology an attractive feature in this species. An important characteristic is related to the skin differences between species, which could impact on the design of transdermal delivery patches for veterinary species. The efficacy of a transdermal system is primarily dependent upon the barrier properties of the target species's skin, as well as the ratio of the area of the transdermal patch to the species's total body mass needed to achieve effective systemic drug concentrations (27). A recent publication (1) showed that the L patch 5% applied proximally to the carpus for 12 hours did not result in a detectable systemic plasma concentration.

Therefore the purposes of the current study were to evaluate the plasma level of L after the L patch 5% application with extended dosing, with and without a previous skin treatment with alcohol, in comparison with L cream 5% applied on the same anatomical region under dressing, and finally to determine the adequacy of analgesia.

Material and methods

L patches 5% 10 × 14 cm (Neurodol® Tissugel, IBSA, Lugano, Switzerland) and L cream 5% (Ortodermina®, lidocaine chlorhydrate, SOFAR Spa, Milan, Italy), were obtained from IBSA (www.ibsa.ch) and common pharmacy, respectively. The ELISA kit test was purchased from the Neogen corporation (Lexinton, KY, USA). Pure standard L chlorhydrate was purchased from LGC Promochem (Milano, Italy). Ethanol (EtOH) analytical grade was purchased from Sigma-Aldrich (St. Louis, MO, USA).

Animal treatment. Experiments were performed at the Faculty of Veterinary Medicine, University of Pisa. Animal care and handling conformed to the provision of the EC council Directive 86/609 EEC, recognised and adopted by the Italian Government (DL 27/1/1992, n° 116). The study protocol was approved by the ethical committee of the University of Pisa (CASA) authorization n° 18340 and transmitted to the Italian Ministry of Health.

The study was carried out using the same horses. Animals were randomly assigned to four treatment groups, using an open, single-dose, four-treatment, four-period, randomized crossover design with a 30-day washout. All the animals received four treatments.

Eight male trotter horses, with a mean age of 11 (9-13) years and a mean weight of 485 (455-540) kg were included in the study. All the tested animals were kept in individual box stalls with an ambient temperature between 10 and 20 degrees Celsius, fed hay *ad libitum*, and allowed unlimited access to water. The horses were previously determined to be clinically healthy based on full chemistry hematologic analyses and physical examinations (including rectal temperature, heart rate and murmurs, respiratory rate, attitude, appetite, water consumption) performed before and during the study.

The day before the drug administration, the horse's limbs were clipped on the medial aspect above the carpus and the anatomical regions were gently washed with water and dried; the left jugular furrow was clipped and surgically prepared for a percutaneous placement of a 80 mm 12 SWG intravenous catheter (Intraflon 2 VYGON®, Padua, Italy). The jugular catheter was secured in place via suture (2-0 3 metric Monosof® nylon, Gosport, UK) and bandage, and heparinised saline was used to maintain the patency of the catheter throughout the course of blood sampling.

Treatment I. The horses were treated through the application of two L patches 5% on the medial aspect just proximal to each carpal joint of both forelimbs. Light bandage (Soffban®, BSN medical Ltd., England) and elastic adhesive bandage (Elastoplast® BSN medical Ltd., England) were used to keep them in place. The L patch is constructed of an adhesive material that contains 5% L (11, 24). It is applied to a non-woven, polyester felt backing, and covered with a polyethylene film-release liner. The release liner must be removed prior to its application to the skin. Following removal of the liner, the L patch is self-adhesive (24). The patch is 10 × 14 cm in size and contains 700 mg of L (50 mg of L per g of the adhesive patch) in an aqueous base. The patch is clean but not sterile. Although the L patch is self-adhesive, an adhesive, non-woven fabric retention dressing was used to cover and secure the L patch in site, in order to maintain full contact with the skin. The patches were kept in place for 24 hours.

Treatment II. The horses were treated as in the treatment I, but at this time the area of application was pre-cleaned with EtOH, before the patch application. The patches were kept in place for 24 hours.

Treatment III. The horses were treated through the application of L cream 5%, applied every two hours (233.3 mg L corresponding to 4.67 g of cream) along a period of 24 hours. The L cream was applied on the medial aspect of both forelimbs, just proximal to each carpal joint, spread on an area of 2800 cm^2 and protected by a polyethylene film (Tegaderm®, 3M™, Milan, Italy).

Treatment IV. The horses received only the light bandage but no drug (control).

At the end of the experiment, pain was assessed in all the treatment groups by two observers blinded to the treatment protocol; both were veterinary clinicians, trained to assess pain in horses using behavioural observations. Horses were assessed for excitement and adequacy of analgesia using a psychophysical method by pricking the patient's skin to test the animal's response to pain, using a visual analogue scale (VAS) pain score (100 mm scale, with 0 indicating no pain and 100 indicating the maximum possible pain) at the end of treatments (24 hr) (3, 23) (fig. 1). One month washout period was completed between the phases.

Fig. 1. Visual analogue scale (VAS). A mark is placed along the line to indicate the current pain level of the horse

The blood samples were collected via catheter at 0, 6, 10, 12, 14, 18, 24 and 36 h after treatment by removing an initial 1 ml of blood, and then collecting 5 ml of blood for analysis in heparinised vials. The catheter was finally flushed with heparinised saline solution. The blood was centrifuged the following hour after collection, for 5 minutes at 3000 rpm and the plasma stored at –80°C until analysis. The plasma was tested in duplicate using L ELISA kit test according to the manufacturer procedure. The L sensitivity (limit of quantisation, LOQ) of the test was 1 ng/mL. A calibration curve was made in triplicate at concentrations of 1, 5, 10, 20, and 50 ng/mL adding pure L chlorhydrate to the drug-free horse plasma. At the end of the test procedure, the plate was read by a spectrophotometer plate reader (SPECTROSTAR, Offenburg, Germany) at 450 nm.

Statistical analysis. Pain scores were described as mean ± standard deviation, non-parametric data were evaluated using the Kruskal-Wallis test. Such a test was also used to evaluate L plasma concentrations that were expressed as both mean ± standard deviation and median (maximum-minimum) values. The level of significance was $p < 0.05$.

Results and discussion

The patches remained in place on all horses with the bandage application. At the removal most of each patch was completely adherent to the skin, though in some instances the edge of the patch was loosened. Following cream administration no drug was observed beyond the bandage's edge, while in the last treatment few residues of unabsorbed cream were seen on the horses' forelimbs. Although L patches have sometimes shown to induce local erythema (1, 6, 14), in the present study no adverse effects such as behaviour changes, variations in clinical examinations, erythema on the treated anatomical area or pruritis, were shown. The L plasma concentrations are reported in table 1. In the patch treatment groups (I and II) plasma level concentrations were widely variable between the subjects; furthermore not all the horses showed detectable L plasma concentrations through the 24 hours of drug application. One of the eight horses in the patches + alcohol group showed L plasma levels below the LOQ. Although the differences shown in L plasma levels between groups I and II were not significant (except in 18 and 24 h values), the use of alcohol seems to reduce the absorption of the drug and plasma concentration over time. Groups I and II did not show a clear trend of increasing L plasma concentrations over time, unlike group III. In this group, L plasma levels were already high at the first time of blood withdrawal, significantly different from the patch groups, and they rose in the following hours. These subjects showed a lower variability of values compared to the other groups. However at the 36th hour after treatment, L plasma concentrations were non-detectable in all the groups.

Two blinded observers with comparable experience assessed the horses for pain after patch and cream removal. Horses were assessed for adequacy of analgesia using a psychophysical method by pricking the patient's skin to test the animal's response to pain. The mean pain rating in group I, group II and the control group (group IV) were 40, 37.5 and 42 mm, respectively ($p > 0.05$), while it was 20 mm in group III showing a significant reduction of the pain (fig. 2).

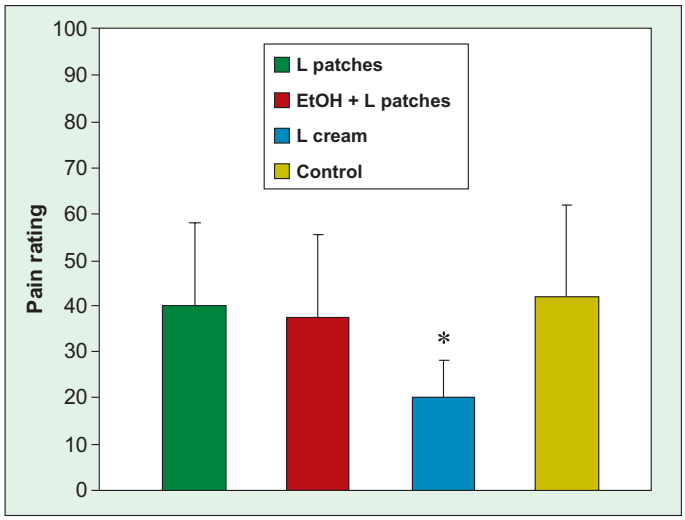

Fig. 2. Pain rating evaluated after the four treatments
Explanation: – 4 × 5% patches; – alcohol + 4 × 5% patches; – L 5% cream; – control; * $p < 0.05$

Tab. 1. Plasma level (ng/ml) of lidocaine expressed as mean ± standard deviation and median (maximum-minimum values) following the three different administrations of lidocaine to horses

Time	Patches		Patches + alcohol§		Cream	
	Mean ± sd	Median (max-min)	Mean ± sd	Median (max-min)	Mean ± sd	Median (max-min)
6	4.3 ± 3.5	4.3 (6.8-1.9)	8.0 ± 6.4	6.5 (15.0-2.4)	26.4 ± 5.6*	25.4 (29.2-23.6)
10	1.5 ± 0.5	1.5 (1.9-1.1)	6.9 ± 9.5	2.3 (17.1-1.0)	29.4 ± 7.3*	30.1 (31.9-22.3)
12	8.1 ± 3.1	8.1 (10.3-6.0)	7.1 ± 7.4	4.3 (15.2-1.0)	30.6 ± 8.2*	30.7 (33.1-25.0)
14	9.0 ± 5.5	7.6 (16.5-4.2)	14.2 ± 11.3	9.8 (21.2-2.9)	32.7 ± 5.1*	31.6 (35.6-25.1)
18	7.3 ± 8.4	4.6 (19.4-1.0)	n.d.°	n.d.°	32.4 ± 6.8*	32.9 (33.8-28.7)
24	4.8 ± 7.0	1.4 (12.9-1.0)	n.d.°	n.d.°	35.2 ± 4.0*	36.2 (37.4-32.1)
36	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.

Explanations: § – values deriving from 7 horses; ° – significant value between group I and II; * – significant value between cream and patches groups; n.d. = not detectable

The results of this study confirm that an extended dosing regimen of the L patch 5% produces a detectable plasma concentration of the drug, as does the use of L cream 5%. Treatment with L cream 5% significantly reduced pain quality.

After the application of patches, L plasma level concentrations were variable and did not show any initial increase followed by a plateau as showed in small animals (14, 15, 33) and humans (10, 11). The results of this study extend current knowledge on L plasma concentration in the case of L patches 5% from a regimen of 2 patches applied for 12 hours (1) to 4 patches applied for 24 hours.

As previously confirmed by another study (6), 5% patches are only able to deliver 3% ± 2% of the 700 mg of L contained in the patch across the epidermis of human patients. If absorption is similar across equine skin, only 28-140 mg of L would cross the epidermis of the horse when four patches are used for a period of 24 hours. This amount of the drug for about 450-550 kg of equine body weight might be too little to detect relevant plasma concentrations.

The primary role of the stratum corneum is to provide a substantial diffusional barrier and thereby protect the body from xenobiotics. The dermal stratum of the body is known to be a complex, dynamic, biochemical environment that responds to ambient conditions to maximise the protective skin barrier (30). Diffusional resistance is known to reside in the stratum corneum and is constituted by a complex interaction of, especially, lipid and proteinaceous components, which creates fairly distinct hydrophilic and lipophilic penetration pathways. The biochemical order of the intercellular lipid matrices of the stratum corneum (12) or keratinized environment of the corneocytes must be altered to allow the penetration of compounds at a suitable rate to the desired site of activity (24). The removal of upper layers of the stratum corneum due to the application of polar solvents, and the related changes in the constituents of the epidermis, could interfere with the drug absorption as reported by another study (22), but it may also interfere with drug delivery to the systemic circulation. The effect of alcohols can influence transdermal penetration by delipidising the membrane and disrupting the stratum corneum (2). Disruption of the epidermis integrity through the extraction of biochemicals by more hydrophobic alcohols contributes to enhance mass transfer through this tissue (9, 21). In this study, the alteration of the horses' skin by EtOH pre-cleaning did not result in a faster and higher absorption. On the other hand, the EtOH treatment produced a reduction of L plasma concentration over time.

In humans, systemic L is metabolised by the liver to a number of metabolites including monoethylglycinexylidide (MEGX) and glycinexylidide (GX), with a similar pharmacological activity, but less potent than the parental drug. Alternatively, drugs penetrating through the skin will be subjected to both Phase I and Phase II metabolic processes, leading to a first-pass effect as the parent drug and/or metabolite reach dermal vasculature (13). The metabolic enzymes, including hydroxylases, deethylases, hydrolases, esterases and peptidases, reside in the skin and appendages, mostly in the epidermal layer (17). The total amount of the drug that is allowed to enter the systemic circulation will vary between species, individuals and even regions of the body. There is also a marked species variation in cutaneous enzyme activity (13, 20, 21).

As for most topical creams, likewise for L, the dose of the drug which provides for effective analgesia depends on the duration of the application over the treated area; in the present study the cream application area (280 cm^2 for the forelimb) and the dose (2800 mg/horse) were calculated to be the same as for the patches. The sustained drug release from the patches was reproduced in the cream treatment by administering 1/12 of the whole dose every 2 hours. Moreover to reproduce the polyethylene covering on the top of the patch, an occlusive polyethylene dressing was applied upon the drug-treated area. This technique is well known to enhance skin hydration capturing insensible water loss, effectively hydrating the skin (27). Human clinical studies indicate that layers of L cream 5% 1-2 g/10 cm^2 are adequate to produce analgesia (25); the dose administered every 2 hours in this study was 4.1 g/10 cm^2. The rising L plasma concentrations show that in the horse there is a positive correlation between the dose and the L plasma level as in human studies (16). A high L plasma concentration level (8 μg/ml) has been reported to induce toxic effects (34), however, the maximum plasma amount showed in this study was about 250 folds lower.

The clinically observed analgesic effect of L was mainly through local penetration (32); therefore transdermally applied L acts mainly through local analgesic effects, rather than through systemic action. Although the present study did not provide data on L skin concentrations, the cream was more absorbable than the patch treatment, showing different L plasma concentrations. Moreover analysing the pharmaco-economy of the treatments, L cream was about 30-fold cheaper than patches.

In conclusion, the application of L cream 5% has been established to be both more effective against pain and cheaper than the application of L patches, despite its multiple dosages over the 24 hours. The L plasma levels due to this formulation remain well below the toxic concentration. The dose extrapolated from human studies, although applied in multiple units (n = 4), seems to be too small for analgesia in horses. A higher dose patch should be produced for clinical use in equines. Nevertheless, further studies will be necessary to determine an effective role of transdermal L use for therapeutic evaluations also in diseased horses since the action of L might be different in healthy animals.

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