16 February 2021
EMA/CVMP/393160/2020
Committee for Medicinal Products for Veterinary Use

# European public MRL assessment report (EPMAR)

## Lidocaine (porcine species)

On 16 November 2020, the European Commission adopted a Regulation[1] establishing maximum residue limits (no MRL required classification) for lidocaine in porcine species (piglets up to 7 days of age only), valid throughout the European Union. These maximum residue limits were based on the favourable opinion and the assessment report adopted by the Committee for Medicinal Products for Veterinary Use.

Lidocaine is intended for cutaneous and/or epilesional use in piglets up to 7 days of age for local anaesthesia during and after castration at a maximum dose of 100 mg/animal.

Maximum residue limits had previously been established for lidocaine (no MRL required classification) in *Equidae* (for local/regional anaesthesia only)[2].

Medical Ethics UK Ltd submitted to the European Medicines Agency an application for the extension of maximum residue limits on 6 September 2018.

Based on the original and complementary data in the dossier, the Committee for Medicinal Products for Veterinary Use recommended, on 16 July 2020, the extension of maximum residue limits for lidocaine to porcine species.

Subsequently, the Commission recommended, on 6 October 2020, that maximum residue limits (no MRL required classification) in porcine species are established. This recommendation was confirmed on 27 October 2020 by the Standing Committee on Veterinary Medicinal Products and adopted by the European Commission on 16 November 2020.

[1] Commission Implementing Regulation (EU) No 2020/1712, O.J. L 384, of 17 November 2020
[2] Commission Regulation (EU) No 37/2010

**Official address** Domenico Scarlattilaan 6 ● 1083 HS Amsterdam ● The Netherlands
**Address for visits and deliveries** Refer to www.ema.europa.eu/how-to-find-us
**Send us a question** Go to www.ema.europa.eu/contact **Telephone** +31 (0)88 781 6000 An agency of the European Union

# Summary of the scientific discussion for the establishment of MRLs

| | |
|---|---|
| Substance name: | Lidocaine |
| Therapeutic class: | NO ENTRY |
| Procedure number: | EMEA/V/MRL/003649/EXTN/0002 |
| Applicant: | Medical Ethics UK Ltd |
| Target species: | Piglets up to 7 days of age |
| Intended therapeutic indication: | Local anaesthesia |
| Route(s) of administration: | Cutaneous and/or epilesional |

# 1. Introduction

Lidocaine was assessed by the CVMP in 1999 with a view to establishing MRLs. Based on the limited data package available at that time, the Committee was unable to establish an ADI and thus unable to establish MRLs in species other than horses. In relation to horses (a minor species), the evaluation concluded that numerical MRL values were not required for the protection of the consumer.

Currently, lidocaine is included in Table 1 (allowed substances) of the Annex to Commission Regulation (EU) No 37/2010 of 22 December 2009, in accordance with the following table:

| **Pharmaco-logically active substance** | **Marker residue** | **Animal species** | **MRLs** | **Target tissues** | **Other provisions** | **Therapeutic classification** |
|---|---|---|---|---|---|---|
| Lidocaine | NOT APPLICA-BLE | *Equidae* | No MRL required | NOT APPLICA-BLE | For local/ regional anaesthesia only | NO ENTRY |

In 2015, lidocaine was assessed by the CVMP in accordance with Article 30(3) of Regulation (EC) No 726/2004, with regards to safety concerns due to consumer exposure to 2,6-xylidine, a genotoxic metabolite of lidocaine. Concerns were raised in relation to potential consumer exposure to 2,6-xylidine from food derived from treated horses as well as from cattle and pigs treated with lidocaine-containing products in line with the provisions of Article 11 of Directive 2001/82/EC (under the so-called 'cascade'). Concerning the use in horses, the CVMP concluded that, based on the limited use, extensive metabolism and considering available in vitro metabolism data, the risk to the consumer was negligible. In relation to the use in cattle and pigs, the CVMP concluded that, taking into account the required withdrawal periods for animals treated under the cascade, together with the extensive metabolism in pigs and cattle, the risk to consumers from lidocaine-treated animals was negligible. This conclusion was not applicable to cattle milk, for which the required withdrawal period from treated cattle under the cascade would not be sufficient for the protection of the consumer (EMA/CVMP/124059/2015).

Medical Ethics submitted an application for the extension of MRLs for lidocaine to porcine species, in the form of lidocaine hydrochloride monohydrate (50 g/l lidocaine) on 6 September 2018. Lidocaine is intended to be used as a local anaesthetic for topical use during and after castration of piglets, at a maximum dose rate of 2 ml of the product (100 mg) in piglets under 7 days of age.

# 2. Scientific risk assessment

## *2.1. Safety assessment*

### 2.1.1. Overview of pharmacological properties

**Pharmacodynamic properties including mode of action**

The major clinical use of lidocaine in human and veterinary medicine is as a local anaesthetic. Transmission of peripheral nociceptive stimuli depends on the presence of voltage-gated sodium ion ($Na^+$) channels in peripheral neurons. Two types of channels occur on peripheral neurons (NaV 1.8 and NaV 1.9), while a third type occurs in sensitive neurons of the sympathetic nervous system (NaV 1.7). Lidocaine and its active metabolite monoethylglycinexylidide as well as other amide anaesthetics interact with peripheral and central voltage-gated $Na^+$ channels in the cell membrane of neurons. Lidocaine has more affinity to the open ion channel which can be found in this state during depolarisation. Thus, local anaesthetics such as lidocaine block nerve impulse generation and conduction by decreasing or preventing the large but transient increase in the permeability of excitable membranes to $Na^+$, normally produced by a slight depolarisation of the membrane. As these change increases in the nerve, the threshold for electrical activity increases, the rate of increase of the action potential declines, impulse conduction slows and ultimately, nerve conduction fails. This failure of nerve impulse generation and conduction leads to numbing and analgesia at the site of local administration.

In humans, when given intravenously, lidocaine affects peripheral and central nerves. It is used intravenously for a number of indications, but largely for the control of neuropathic pain. Following intravenous administration, different mechanisms of analgesia may come into effect. Lidocaine increases the concentration of acetylcholine and activates inhibitory pain pathways, which results in analgesia, possibly by binding to muscarinic receptors, by inhibiting glycine receptors and inducing the release of endogenous opioids, thus providing pain relief. Lidocaine also reduces the inflammatory response to tissue ischaemia and reduces tissue damage induced by endothelial and vascular cytokines.

Lidocaine is also used as a ventricular antiarrhythmic drug. It is effective in terminating ventricular premature beats and ventricular tachycardia, can shorten ventricular arrhythmias as well as the QTc interval and provide haemodynamic effects, such as lowering of blood pressure. These effects are mediated by the action of lidocaine on cardiac pacemaker and Purkinje cells.

The scientific literature on the pharmacological actions of lidocaine and related compounds is extensive. Oral doses up to 250 mg (3–4 mg/kg) do not result in therapeutic blood levels, or in any subjective or objective clinical effects, and lidocaine has therefore proven ineffective as an oral antiarrhythmic at these doses. Oral doses of 250 mg or less result in peak serum levels above the therapeutic range of < 1–2 µg/ml. Higher oral doses of 500–750 mg may result in transient neurological symptoms such as dizziness, light-headedness, and circumoral numbness. Clinically, 500 mg/day had some inconsistent short-term clinical benefit, although 250 mg/day were without therapeutic efficacy. In view of its rapid elimination half-life, studies indicate that lidocaine, up to 300 mg, may be administered orally every 3 hours without inducing toxic effects. An absence of therapeutic or toxic systemic effects was observed when a 300 mg oral dose was administered every 3 hours on 8 occasions (a total dose of 2,400 mg was given during 24 hours).

In humans, the tissues that may be exposed to local pharmacological effects of lidocaine in food are the mouth and upper GI tract. Several studies have examined local anaesthetic efficacy of lidocaine

lozenges, lollipops, topical applications, sprays gels for pre-operative dental or pharyngeal anaesthesia. Pain relieving effects are documented with single and multiple doses of lozenges containing 8 mg lidocaine as a single agent (up to 8–10 times per day), or single doses of 300 mg lollipops, in all cases requiring a 5–10 minute suck and dwell time in the mouth. The dose-response effect was evident with lidocaine lozenges containing 2, 4 and 8 mg, with the 8 mg tablet tending to be the most effective. With respect to experienced adverse events, no differences were observed between the active and the placebo groups. The most frequently reported adverse events were headache, hypoesthesia, nausea, sore throat and voice alteration.

**Pharmacokinetic properties (mainly in laboratory animals)**

No original studies on pharmacokinetics in laboratory animals using the oral exposure route are available for lidocaine.

**Absorption**

When rats were given oral doses of 10 mg/kg bw 1% $^{3}$H-lidocaine hydrochloride by gavage, the highest levels of radioactivity were detected in the intestine (47.5%) 4 hours after administration, and in the stomach (13.7%), liver (11,7%) and in the kidney (2.0%) 30 minutes after administration.

Absorption after oral administration of lidocaine hydrochloride to beagle dogs is extensive. When given intravenously, plasma lidocaine concentrations peaked at 15 minutes after administration. The plasma half-life was 0.86 hours.

In a radiolabelled study in humans, when lidocaine hydrochloride monohydrate (250 mg; approximately 4.17 mg/kg bw) was given to a group of 20 healthy male volunteers orally, absorption was extensive. Around 84% of the administered dose was recovered as metabolites in the urine within 24 hours. In a further study in humans similar results were reported: an oral dose of lidocaine hydrochloride monohydrate (250 mg; approximately 4.17 mg/kg bw) administered to two healthy male volunteers resulted in 64–75% of the administered dose being recovered in the urine within 24 hours. In the same study, a male volunteer was given the molar equivalent of lidocaine base following the same route of administration, which resulted with its absorption being much lower. Maximum plasma concentration in the 2 subjects given the hydrochloride monohydrate was approximately 0.08 µg/ml, but in the subject given the base, plasma concentration was not detectable 30 minutes after administration. Administration of ammonia (to decrease urinary pH) has no effects on absorption. In another study in humans, following oral administration of 300 mg lidocaine hydrochloride monohydrate to 3 healthy volunteers (gender not specified), peak lidocaine concentrations were detected in blood from 20 to 60 minutes after dosing. In patients with hepatic cirrhosis or hepatitis, oral doses of 5 mg/kg bw resulted in significantly higher plasma concentrations of lidocaine and monoethylglycinexylidide than were noted in healthy controls given the same oral dose. In a different study with healthy human volunteers given an oral dose of $^{3}$H-labelled lidocaine (500 mg; 8.3 mg/kg bw), urine was collected for 72 hours after dosing from 3 normal subjects (gender not specified); around 50% of the radioactivity was recovered in the urine, with the majority present in the first 8 hours after dosing. Following intravenous injections or infusions of lidocaine, plasma concentrations were relatively high soon after administration. In infants and children (up to 4 years of age) given lidocaine by the epidural route, lidocaine plasma concentrations increased in a linear manner from 2.5 µg/ml at the start of administration to < 3 µg/ml 5 hours later.

Lidocaine is subject to dermal absorption after topical application. When applied as a topical patch containing 46.1 mg lidocaine to the gingiva of healthy human volunteers, the $C_{max}$ was 16.5 ng/ml

and the $T_{max}$ 28.6 minutes. The plasma half-life was 102 minutes. It is likely that the degree and extent of absorption may vary according to the formulation/patch used.

**Distribution**

When rats were treated intramuscularly with $^{14}C$-labelled lidocaine, concentrations of radioactivity were high in the lungs, kidney, spleen, brain and liver 30 minutes after dosing. The highest concentration was found in the kidney, possibly reflecting renal elimination. Following intravenous administration of $^{3}H$-2,6-xylidine to Sprague-Dawley rats, whole-body radioluminography revealed that the highest concentrations of radioactivity after 30 minutes were located in the adipose tissue. A high degree of labelling was also evident in some nasal glands, with moderate labelling in the spinal cord and brain. Autoradiography of freeze dried sections showed a high degree of labelling in the nasal olfactory mucosa and in the mucosa of the of the upper alimentary tract including the tongue, cheek, gingiva, soft palate, oesophagus and forestomach, with lower degrees of labelling in the respiratory, tracheal and bronchial mucosa. Similar findings were reported after oral administration in rats.

Lidocaine is able to cross the placenta in rabbits following intravenous administration and is able to reach the brain in foetal rabbits.

There are few data on the distribution of lidocaine in tissues in humans. *In vivo*, lidocaine binds strongly to serum proteins. It binds weakly to human serum albumin but has a strong binding affinity to α1-acid glycoprotein. The volume of distribution after oral administration to healthy volunteers was between 1.3 to 3.6 l/kg, indicating substantial distribution in tissues. When given by epidural administration to pregnant women, lidocaine, monoethylglycinexylidide and glycinexylidide were detected in foetal blood. The foetal/maternal plasma ratios of lidocaine, monoethylglycinexylidide and glycinexylidide after caesarean delivery were 0.66, 1.45 and 1.03, respectively. Similar findings were reported for women after vaginal delivery. After intravenous administration to the mother, lidocaine has been detected in maternal venous blood, the umbilical vein, amniotic fluid and foetal blood, and has been shown to be able to cross the placenta. Data from human autopsies demonstrates that, at a relatively short time after administration, lidocaine was initially found in cardiac blood, while the highest concentrations were later found in the brain, followed by the kidney and urine. Autopsy data from an oral lidocaine overdose taken by one individual showed residues that were found in the blood, liver, kidney, brain, spleen, lung, heart, urine and bile, suggesting widespread distribution.

**Metabolism**

The biotransformation of lidocaine is extensive in all species tested (rats, dogs and guinea pigs), including humans, and a number of metabolites are formed. Metabolism appears to be mediated through the action of a number of enzymes, including carboxylesterase and cytochrome P450 isoenzymes.

In a study with female Sprague-Dawley rats administered doses of 20 mg/kg bw lidocaine hydrochloride by gavage, the 24-hour urine was examined for lidocaine metabolites. The main compounds found in urine were lidocaine (0.2%), monoethylglycinexylidide (0.7%), glycinexylidide (2.1%), 3'-hydroxylidocaine (31.2%), 3'-hydroxymonoethylglycinexylidide (36.9%), 2,6-xylidine (1.5%) and 4-hydroxyxilidine (12.4%), expressed as percentage of the administered dose. In another study in rats (male Wistar), using an intraperitoneal administration of 27 mg/kg bw lidocaine hydrochloride, the only significant metabolites present in 24-hour urine to be 3'-hydroxylidocaine (6–16% of the dose) and 4'-hydroxylidocaine (3–5% of the dose, with 3'-hydroxylidocaine in the form of its glucuronide conjugate).

In an in vitro study, brain, lung, kidney and liver microsomes from rats either untreated or pre-treated with phenobarbital were incubated with lidocaine. The results strongly suggest that lidocaine metabolism occurs mainly in the liver and that cytochrome P450 is involved. In this study, the cytochrome P450 isozymes CYP2B1 and CYP2C11 were shown to form monoethylglycinexylidide as the only metabolite of lidocaine. Antibodies against CYP2B1 abolished this activity. Thus, in this study, hepatic cytochrome P450 isozymes were shown to have the ability to *N*-deethylate lidocaine to monoethylglycinexylidide.

Following authorisation of lidocaine for nasal application in humans for the treatment of migraine, the metabolism of lidocaine was studied in olfactory and respiratory microsomes derived from Long-Evans rats and in liver microsomes derived from human donors. Affinity for lidocaine was higher in rat nasal microsomes than in rat liver or rat respiratory microsomes. Vmax values (µM/min) for the metabolism of lidocaine to monoethylglycinexylidide were as follows: human liver microsomes: 0.23, rat liver microsomes: 0.41, rat olfactory microsomes: 0.54 and rat respiratory microsomes: 0.09.

Haemoglobin adducts derived from 2,6-xylidine have been detected in rats given lidocaine or 2,6-xylidine intraperitoneally. After 24 hours, around 0.84% of the dose of 2,6-xylidine was covalently bound to haemoglobin. After administration of lidocaine, 0.027% of the dose was bound to haemoglobin as 2,6-xylidine.

When lidocaine was incubated with mouse liver slices, concentrations of 2,6-xylidine increased slowly during a 60-minute period.

Female guinea pigs were administered lidocaine hydrochloride orally and the 24-hour urine collected. They excreted a larger amount of 2,6-xylidine than did dogs, humans or rats. A total of 53.8% of the administered dose were recovered in urine after 24 hours and the major compounds found in this urine were lidocaine (0.5%), monoethylglycinexylidide (14.9%), glycinexylidide (3.3%), 3'-hydroxylidocaine (0.5%), 3'-hydroxymonoethylglycinexylidide (2.0%), 2,6-xylidine (16.2%) and 4-hydroxyxilidine (16.4%), expressed as percentage of the administered dose.

In a study of the metabolism of lidocaine by rabbit S9 liver homogenates, 5 µmol lidocaine were incubated with this homogenate for 30 and 120 minutes. The concentrations of the metabolites, expressed as percentage of lidocaine metabolised for 30 and 120 minutes, respectively, were as follows: monoethylglycinexylidide 67.8 and 37.1%, glycinexylidide 3.1 and 51%, 3'-hydroxymonoethylglycinexylidide 1.6 and 1.1%, 3'-hydroxylidocaine 2.1 and 1.7%, 4'-hydroxylidocaine 0.4 and 0.4%, 2,6-xylidine 7.3 and 35.5%, 4-hydroxyxilidine 4.1 and 7.9% and 2-amino-3-methylbenzoic acid 0.2 and 0.5%. The authors also looked for evidence of the formation of 2,6-dimethylnitrobenzene, lidocaine-*N*-oxide and 2,6-dimethylnitrosobenzene, but these were not detected.

Following oral administration of 2% lidocaine hydrochloride to dogs, at a dose of 10 mg/kg bw, the main metabolites in the 24-hour urine were parent drug (2.0%), monoethylglycinexylidide (2.3%), glycinexylidide (12.6%), 3'-hydroxylidocaine (6.7%), 3'-hydroxymonoethylglycinexylidide (3.1%), 2,6-xylidine (1.6%) and 4-hydroxyxilidine (35.2%), expressed as percentages of the administered dose.

A group of 20 healthy male volunteers were given an oral dose of lidocaine hydrochloride monohydrate and urine was then collected over a 24 hour period. The main compounds found in urine, expressed as percentage of the lidocaine dose were monoethylglycinexylidide (3.7%), glycinexylidide (2.3%), 3'-hydroxylidocaine (1.1%), 3'-hydroxymonoethylglycinexylidide (0.3%), 2,6-xylidine (1.0%) and 4-hydroxyxilidine (72.6%). Oral administration of lidocaine hydrochloride monohydrate resulted in recoveries of 64.2 to 75.2% of the administered dose in the urine.

Compounds found in urine, expressed as percentage of the administered dose, were as follows: lidocaine (4.76%), monoethylglycinexylidide (12.68%), glycinexylidide (0.78%), 3'-hydroxylidocaine and/or 4'-hydroxymonoethylglycinexylidide (0.7%), 3'-hydroxymonoethylglycinexylidide and/or 4'-hydroxymonoethylglycinexylidide (0.81%), 2,6-xylidine (0.84%) and 4-hydroxyxilidine (65.98%). *N*-ethylglycine and N,*N*-diethylglycine were detected but not quantified. The phenylhydroxylamine of 2,6-xylidine (*N*-hydroxyxilidine) has also been detected in human subjects given lidocaine hydrochloride orally and quantified at 1% of the administered dose.

In two patients given intravenous lidocaine after suspected myocardial infarction, urine was collected for 72 hours after administration. The following compounds were found in urine, expressed as a percentage of the administered dose: lidocaine (2.1%), monoethylglycinexylidide (1.7%), glycinexylidide (0.55%), 4'-hydroxylidocaine (0.28%), 4'-hydroxymonoethylglycinexylidide (0.06%), 4'-hydroxyglycinexylidide (0.24%), 4-hydroxyxilidine (80.1%), 3'-hydroxylidocaine (0.13%), 3'-hydroxymonoethylglycinexylidide (0.04%). The metabolite2,6-xylidine was not detected.

Monoethylglycinexylidide, glycinexylidide and 4-hydroxyxilidine were detected after lidocaine hydrochloride was incubated with human liver slices. When lidocaine concentrations were increased, concentrations of monoethylglycinexylidide and 2,6-xylidine increased. Lidocaine metabolism by human liver or kidney slices as well as human lung slices was investigated in 3-hour incubations (lung and kidney) or 1-hour incubations (liver). Incubation of lidocaine with kidney cortex slides resulted in the formation of 20 pmol monoethylglycinexylidide/3 h/µg but less than half this amount was produced in the renal medulla. Lidocaine metabolism in human lung resulted in 10 to 50 pmol monoethylglycinexylidide/3 h/µg, whereas in human liver this was approximately 90 pmol monoethylglycinexylidide/1 h/µg.

Studies in rats suggest that 4-hydroxyxilidine is formed from the rearrangement of *N*-hydroxyxilidine. *N*-hydroxyxilidine, also known as *N*-(2,6-dimethylphenyl)hydroxylamine, is, as the name suggests, a phenylhydroxylamine. These can undergo the Bamberger rearrangement to yield the corresponding 4-hydroxy derivative which, in case of *N*-hydroxyxilidine is 4-hydroxyxilidine. However, this appears to be a minor metabolic pathway only observed in *in vitro* studies and at extreme dilutions. At higher dilutions *in vitro*, 4-hydroxyxilidine was the major metabolite. The major pathway of formation of 4-hydroxyxilidine is through direct 4-hydroxylation of 2,6-xylidine.

In humans, CYP1A2 appears to be the main cytochrome P450 isoform responsible for the conversion of lidocaine to monoethylglycinexylidide and glycinexylidide. When human subjects were given a specific inhibitor of this isoform, fluvoxamine, clearance of lidocaine was significantly reduced (up to 60%) with increases in terminal half-lives. The kinetics of monoethylglycinexylidide and glycinexylidide formation were impaired. Cytochrome P450 3A4 is also involved in the human metabolism of lidocaine.

**Excretion**

After oral or intravenous administration of 3H-lidocaine hydrochloride to rats, biliary excretion was extensive. Following an oral dose of 10 mg/kg bw, biliary excretion accounted for 28.5% of the dose, while, after intravenous administration of 5 mg/kg bw, the respective amount was 30% of the dose. The half-life of lidocaine in rats was around 30 minutes compared with 90 minutes in humans. A similar degree (29.9–37.0%) of biliary excretion was reported following an intravenous administration of 14C-lidocaine hydrochloride (4.2 mg/kg bw) to rats. In dogs treated with 10 mg/kg bw lidocaine hydrochloride, urinary excretion was the main route of elimination.

The major route of excretion for lidocaine (and its metabolites) in humans is via the urine. Up to 80% of an oral dose of lidocaine was excreted this way. Faecal elimination is low. It is likely that there is a degree of biliary excretion, but this may be offset by intestinal reabsorption leading to

hepatobiliary recycling. Total clearance of lidocaine was around 15 ml/min/kg after oral administration to healthy volunteers. In a study of a female patient who was anhepatic and waiting for a liver transplant, lidocaine serum concentrations declined much more slowly than those of a healthy subject, and monoethylglycinexylidide concentrations in the anhepatic patient were lower than those reported in the healthy subject.

Urinary excretion of lidocaine may be affected by urinary pH. Higher clearance values were noted when the urine was acidic and it dropped to almost zero when the urine was alkaline.

In neonates from mothers given lidocaine epidurally prior to caesarean or vaginal delivery, the urinary excretion was similar to that of the mother. However, excretion of monoethylglycinexylidide and glycinexylidide in the neonate was around twice that of the mother on day 1 after birth, and 4–5 times that of the mother on the second and third day after delivery. Other studies have shown similar findings, thus confirming that lidocaine can cross the placenta.

## 2.1.2. Calculation of pharmacological ADI

From the review of published data, a pharmacological NO(A)EL of 250 mg/person/day can be established based on the absence of clinical effects and therapeutic blood levels seen in human subjects.

A pharmacological LOEL of 8 mg/person/day was not considered appropriate since the data used to derive this value relate only to a topical application of lidocaine and are not considered representative of a dietary uptake of lidocaine residues.

A pharmacological ADI of 25 mg/person can be established based on the NOAEL and an uncertainty factor of 10 to account for intra-species variation.

## 2.1.3. Overview of toxicology

**Single-dose toxicity**

Published literature shows adverse effects following a single administration of lidocaine in various species and using various exposure routes. Lidocaine treatment can result in central nervous system depression, which causes behavioural effects such as tonic extension, clonic convulsions, a loss of righting reflex and motor ataxia, that can produce a respiratory depression which can result in cardiovascular collapse and death. The severity of acute adverse effects increases with increased systemic availability of lidocaine and is therefore dependent on the route of administration.

Acute toxicity of lidocaine was determined in rats, mice, dogs and monkeys. Oral and subcutaneous $LD_{50}$ values in mice are in the range of 200–400 mg/kg bw, while the intramuscular $LD_{50}$ value of lidocaine in rats is 260 mg/kg bw. Intravenous administration was the most toxic route, with an $LD_{50}$ value of 27.8 mg/kg bw in rats and a value of total toxic dose of 11.09 ± 4.12 mg/kg bw in dogs, where the onset of a muscle tremor was considered a toxic effect. In another study in dogs using the intravenous administration route, convulsions were induced at a rate 8 mg/kg bw/minute, while, in monkeys exposed intravenously to lidocaine, an induction of convulsions occurred at a rate 4 mg/kg bw/minute. An $LD_{50}$ value of 133 mg/kg bw in mice was obtained via the intraperitoneal route.

Regarding the acute oral toxicity of the lidocaine metabolite 2,6-xylidine, several published references are available, indicating an oral $LD_{50}$ value ranging from 840 mg/kg bw to 2042 mg/kg bw in rats. In mice, an $LD_{50}$ value of 720 mg/kg bw was obtained.

**Repeated dose toxicity**

There are no sub-chronic or chronic toxicity studies available for lidocaine, hence a relevant NO(A)EL/LO(A)EL from oral exposure cannot be established.

Published literature on sub-chronic toxicity of the lidocaine metabolite 2,6-xylidine was submitted. In one of the studies, rats were given oral doses of 400 mg of 2,6-xylidine/kg bw/day during the first week, and thereafter 500 mg/kg bw/day were administered for 28 days. The main finding was hepatomegaly with normal appearance of hepatocytes. The hepatomegaly in rats was later shown to be due to a proliferation of smooth endoplasmic reticulum. In a different study, rats given oral doses of 20, 100 or 500 mg/kg bw/day of 2,6-xylidine for 28 days demonstrated decreased haemoglobin and haematocrit concentrations. In a similar study, beagle dogs were given lidocaine at 2, 10 and 50 mg/kg bw/day orally for the same period and fatty degeneration of the liver was observed, although no adverse renal effects were noted at any of the doses tested. Decreases in glycogen content occurred along with reduced glucose-6-phosphate activity in rats. Increases in cytochrome P450 enzyme activity did not occur.

In the range-finding study of 2,6-xylidine in rats given doses of 0, 80, 160, 310, 620 or 1250 mg/kg bw per day for 2 weeks (5 days per week) via oral gavage, bodyweights were reduced at 310 mg/kg bw per day. Deaths and clinical biochemistry changes were also observed at 310 mg/kg bw per day and at higher doses. The NOEL in this study was 160 mg/kg bw per day. As part of the same study, rats were given the substance at 20, 40, 80, 160 or 310 mg/kg bw/day orally, 5 days per week for 90 days. No deaths were reported, while male and female bodyweights were reduced at the highest dose level and in females given 40 or 160 mg/kg bw/day. At 160 and 310 mg/kg bw/day there was an increase in relative liver weights. Histological changes in the form of minimal to moderate inflammatory changes were noted in the nasal mucosa of both sexes, but this was also noted in controls. Based on haematology and clinical chemistry, male rats appeared to be more sensitive than females to the effects of 2,6-xylidine. Significant decreases in leukocyte and lymphocyte counts occurred in males at doses of 40 mg/kg bw/day and above. Also, an increase in the percentage of segmented neutrophils was observed. There were significant decreases in haemoglobin levels at 160 and 310 mg/kg bw in males and females and in the haematocrit at 310 mg/kg bw in males. It is possible that the haematological effects noted in this study were due to methaemoglobinaemia formation. A dose-related increase in polychromasia occurred in males. The NOAEL in this study was 20 mg/kg bw per day.

The toxicity of lidocaine following 30 days of oral administration to rats was investigated in an early study report. Histopathology was performed on the liver, kidney, and spleen of 38 animals dosed up to 100 mg/kg bw and no findings were reported.

A 28-day study in rabbits dosing lidocaine and tetracaine topically confirmed systemic exposure to lidocaine and showed no significant haematological, biochemical or histopathological effects other than possible mild inflammatory reaction at the application sites.

Several published pharmacokinetic and long-term topical human studies are summarised. Patches containing 700 mg of lidocaine are marketed for analgesic use for herpes zoster patients and post herpetic neuralgia patients. Patients included in the studies are dosed with 1 to 3 lidocaine 5% patches (700–2100 mg) for 12 h daily for up to 5 years. The calculated systemic bioavailability of the dermal patches ranges from 1 to 5% which is equivalent to a mean systemic exposure of approximately 64 mg (0.9 mg/kg; range 20–134 mg). $C_{max}$ is around 150 ng/ml (neurologic and cardiac toxic manifestations occur around 5000–6000 ng/ml) and no accumulation is seen. Model simulations predict that plasma levels reach steady state after 3 days, with a mean steady state plasma lidocaine concentration of approximately 45 ng/ml and 2,6-xylidine concentrations of 6–

8 ng/ml in patients wearing up to 3 plasters daily for 1 year. Localised skin reactions are reported and the monitoring of systemic reactions revealed mild to moderate events.

Lidocaine has an oral bioavailability of approximately 35%, hence the level of systemic absorption from approved daily use of lidocaine patches is equivalent to an oral lidocaine dose of 182 mg (range 57–383 mg p.o.). Studies from long-term patch administration in humans (supported by studies in animals reported and reviewed by regulatory authorities) indicate that a systemically absorbed dose of 63.4 mg/day (approximately 1 mg/kg/day) provides a lower bound for a NO(A)EL for repeat dose toxic effects in humans (this is considered conservative as it assumes 100% bioavailability of the dose via the oral route).

**Reproductive toxicity**

No studies on fertility and reproductive performance ([multi-]generation study) and no developmental toxicity study using the oral route of exposure are available, hence a relevant NO(A)EL/LO(A)EL/BMDL from oral exposure cannot be established. Submitted studies on developmental toxicity in rats were performed using the subcutaneous, intramuscular or intraperitoneal exposure route and were available as published literature. One *in vitro* study on developmental toxicity from published literature was also submitted.

In an *in vitro* study in rat embryos, signs of growth retardation at concentrations higher than 250 µM lidocaine were reported. In an *in vivo* study in Sprague-Dawley rats, mean foetal weight was significantly lower than in control animals at 500 mg/kg/day at day 21 after subcutaneous administration by means of an implanted osmotic pump. An increased incidence of incomplete ossification of the *sternebrae* in the high dose group (500 mg/kg/day) was also observed, which was claimed by the study author as not being a teratogenic effect. No NOAEL was established by the author.

A further study in Sprague-Dawley rats investigated maternal and foetal effects after intraperitoneal administration of 56 mg lidocaine per kg bw per day during 3 days at different periods of gestation. The study was carried out with the maximum non-convulsive dose. Experimental data are not significantly different from control values. Neither gross malformations nor histological abnormalities were observed. In an additional study in Sprague-Dawley rats, animals were dosed intramuscularly on days 10 and 11 of gestation. Treated rats were allowed to deliver pups normally and were observed for spontaneous activity, nociception, learning ability and physical development. The author concluded that findings show no evidence of behavioural deficits or consistent alterations as a result of prenatal treatment with lidocaine.

In a study in mice, teratogenic effects were investigated after an intraperitoneal doses of 32, 40, 50, 55, 58, 60, 66, 70 mg/kg bw. Doses of up to 40 mg/kg bw had no notable effects on foetal development, while doses of 50 mg/kg bw and above resulted in central nervous system abnormalities, specifically dilatation of the fourth ventricle. The author concluded the findings reflect direct central nervous system toxicity rather than any specific teratogenic effect.

Reproduction and developmental tests in rats and rabbits were summarised in two pharmacological reviews of the US FDA to confirm safety of daily human oral exposure to lidocaine such as through ‘Oraquix’ (oral lidocaine-prilocaine solution) and high repeat dose chronic daily topical exposure such as via an S-caine patch (lidocaine 7%/tetracaine 7%) (NDA 21-717). While the NDA report cannot be considered to contain sufficient detail be considered pivotal data, the summarised studies suggest little potential for adverse reproductive and teratogenic effects of lidocaine in the standard two species, rat and rabbit, at doses up to 60 mg/kg/day s.c. (NOAELs for these effects usually being the highest doses tested in each study). Maternal acute neurological toxicity was evident from approximately 15 mg/kg/day.

Exposure to lidocaine in pregnancy has also been investigated in humans. Lidocaine is frequently used for pain control during *in vitro* fertilisation procedures. Reports have found no negative impact of lidocaine on human sperm mobility or on fertility as well as early embryonic development with therapeutic doses of 10–200 mg, which translate to mean follicular fluid concentrations of 0.36 ± 1.1 µg/ml, with a range of 0 to 155 µg/ml. Conversely, several investigators have reported improved fertility outcomes with lidocaine treatment under these conditions.

**Genotoxicity**

Genotoxicity studies have been carried out with lidocaine and metabolites. Within the battery of *in vitro* and *in vivo* genotoxicity studies, lidocaine did not induce gene mutations in prokaryotic test systems *in vitro* and was negative in *in vitro* chromosomal aberration tests. Results from an *in vivo* micronucleus tests were animals were dosed by the intraperitoneal route were also negative. A test for gene mutation in *Drosophila* gave negative results for lidocaine.

However, the metabolite 2,6-xylidine showed weakly positive responses in bacterial mutagenicity tests using *Salmonella typhimurium* strain TA100 (and once in TA1535) in the presence of S9 in some tests, while, in other tests, the results were negative. 2,6-xylidine induced chromosomal aberrations in hamster ovary and lung cells *in vitro* at high concentrations or under cytotoxic conditions, and induced gene mutations in eukaryotic test systems *in vitro*. *In vivo*, 2,6-xylidine was considered non-clastogenic in micronucleus assays, in the peripheral blood and bone marrow of mice after oral doses of up to 375 mg/kg bw, although no plasma level was measured in these studies. An oral *in vivo* unscheduled DNA synthesis assay in rats assay using 2,6-xylidine was clearly negative in liver cells up to a dose of 850 mg/kg bw, although the exposure of liver was not monitored.

A transgenic gene mutation assay was positive in nasal tissue of mice after doses of 100 mg of 2,6-xylidine/kg bw administered once weekly by oral gavage during four weeks. DNA extraction was carried out from nasal tissue, bone marrow and liver. A two-fold increase in mutation frequency of lacZ and cII genes in nasal tissue was reported; A:T to G:C base pair transitions and G:C to A:T transversions were observed, but the authors question their biological relevance because statistical significance was not reported and no historical control mutant frequency data were presented. Two *in vivo* comet assays with 2,6-xylidine were reported to be positive as well. In one comet assay study, 2,6-xylidine was orally administered at 200 mg/kg bw four times at weakly intervals and positive results were obtained in the lung, kidney, and liver at 3 hours after the last dosing, but these were not observed 24 hours after the last dosing. In the second comet assay in mice, a single dose of 350 mg/kg was administered orally. Statistically significant increases in comet tail length were seen in the stomach and bladder 8 hours after dosing only, while comet tail length was not significantly increased at 3 and 24 hours after dosing. Significant increases in comet tail length were also seen at 3 and 8 hours (but not 24 hours) in brain, and at 8 and 24 hours in lung tissue.

A combined acute bone marrow micronucleus/comet study in nasal tissue and liver using the oral route in Sprague-Dawley rats was performed with 2,6-xylidine by the applicant. In the main study, 2,6-xylidine was orally administered daily at 15, 50, 125, 250, 500 mg/kg bw for males and 15, 50, 100, 200, 400 mg/kg bw for females for 3 days. No increase in the frequency of MN-PCE in bone marrow was observed for male or female rats administered 2,6-xylidine. In the comet-assay, no significant increases in % tail DNA in either nasal or liver tissue of female rats or in nasal tissue of male rats exposed to 2,6- xylidine were reported. However, an equivocal response was evident in comet results from hepatic tissue in male rats by virtue of finding a positive dose-response, although values were still within the historical negative control range. An *in vivo* comet assay was considered inconclusive and the liver of male rats exposed to 2,6-xylidine was thus microscopically evaluated for changes indicative of tissue toxicity. Microscopic findings in the liver of male rats

exposed to 2,6-xylidine for 3 days with up to 500 mg/kg/day were minimal. In male liver, 2,6-xylidine was metabolised to two known metabolites, DHMA and DMAP-glucuronide. There were no changes in liver glutathione in male rats. The author considered that the lack of genotoxicity in bone marrow and nasal tissues of male and female rats as well as livers of female rats administered 2,6-xylidine is due to efficient detoxification by glucuronidation of DMAP, thus preventing the formation of reactive iminoquinones, as evidenced by the unchanged levels of nasal or liver glutathione. In male liver, the author hypothesized that the observed dose-response is driven by non-statistically significant increases at 250 and 500 mg/kg/day and is due to these high doses overwhelming biochemical defence systems sufficiently to produce minor increases in % tail DNA, although these doses are not sufficient to produce a statistically significant increase in % tail DNA. Clinical signs of toxicity were mainly present at the 250 and 500 mg/kg/day doses for males and 400 mg/kg/day dose for females. The weight loss was dose-dependent and statistically significant compared to the concurrent control at 250 (3%) and 500 mg/kg/day (7%) dose groups in males. The weight loss in females showed dose-dependence, although this was not statistically significant.

*In vitro* studies with the metabolite *N*-(2,6-dimethylphenyl)hydroxylamine showed that genotoxic effects are present in prokaryotic and eukaryotic test systems using synthesized *N*-(2,6-dimethylphenyl)hydroxylamine. In one study in mammalian cells, 4-amino-3,5-dimethylphenol was far more toxic and induced more mutations than *N*-(2,6-dimethylphenyl)hydroxylamine and both hydroxy derivatives were more toxic than activated 2,6-xylidine. A comet assay was positive for *N*-(2,6-dimethylphenyl)hydroxylamine and 4-amino-3,5-dimethylphenol. The authors attributed DNA damage to redox cycling of intracellularly bound aminophenol/quinonimine structures generating reactive oxygen species.

Haemoglobin-adduct formation has been documented in *in vivo* studies with lidocaine and 2,6-xylidine in humans and rats. Weak haemoglobin adduct and DNA adduct formation was observed in *in vitro* and *in vivo* studies with 2,6-xylidine. A number of findings cast doubt on *N*-hydroxylation as the putative mechanism of 2,6-xylidine DNA adduct formation. DNA-adduct characterization studies found a different pattern of adduct formation *in vitro* from reacting DNA with synthesized *N*-acetoxy-2,6-xylidine. Although C8dG and C8dA adducts were present, this was only in minor proportion, and ring-derived as opposed to *N*-derived adducts were found to predominate. 2,6-xylidine haemoglobin adducts have not been characterised and consequently the significance of these adducts in relation to the elucidation of the genotoxic mechanism of action remains uncertain. Low levels of lidocaine-related DNA adducts (0.6–0.7 per $10^{-7}$ normal nucleotides) were observed in male F344 rats after repeated administration of 144 mg/kg/day by gavage for 7 days. In the same study, DNA adducts of 2,6-xylidine (60.6 mg/kg bw, given by oral gavage once or daily for 7 days) were observed. The test was positive in cells from nasal mucosa for single dose (1.12 per $10^{-7}$ normal nucleotides) and repeated dose administration (23.1 per $10^{-7}$ normal nucleotides). Low levels of 2,6-xylidine DNA adducts were also reported in liver and urinary bladder cells. In male Wistar rats, low levels of DNA adducts were also detected at 310 mg/kg bw administered by the oral route during 7 days. In a further study, 2,6-xylidine bound covalently to the DNA of the ethmoid turbinate tissue of the nose of rats after a 9-day oral pre-treatment followed by a single high dose (262.5 mg/kg/day). A study performed in C57BL/6 mice dosed at 83 µg/kg bw 2,6-xylidine via the intraperitoneal route was positive for DNA adduct formation, mainly in urinary bladder and liver cells.

Regarding the mechanism of action*, N*-hydroxylation of 2,6-xylidine (with subsequent esterification and DNA adduct formation) has been referenced as a presumed putative mechanism of carcinogenicity in the rat, in line with that of heterocyclic aromatic amines. It is assumed that, *in vivo*, 2,6-xylidine undergoes *N*-oxidation to *N*-hydroxy-2,6-xylidine (*N*-hydroxyxylidine; 2,6-dimethylhydroxy aniline) and that subsequent co-oxidation reactions with haemoglobin result in

methaemoglobin and nitrenium ion formation. Nitrenium ions (via esterification) are believed to form irreversible covalent bonds with haemoglobin and DNA. DNA adducts resulting from carcinogenic heterocyclic aromatic amines are typically formed via covalent linkage of the amide nitrogen to the C8 of 2'-deoxyguanosine (dG), leading to C8-dG adducts, which in turn result in a variety of mutagenic outcomes. The most common involve frameshift mutations and base substitutions, predominantly G to T, and also G to C transversions. *N*-hydroxyxylidine formation has been observed *in vitro* and synthesized *N*-acetoxy metabolites do form C8-dG adducts when reacted with DNA. However, there is little evidence of *N*-hydroxyxylidine production and/or esterification to *N*-acetoxy derivatives *in vivo*.

An alternative potential mechanism of genotoxicity is via metabolic activation of 4-hydroxy-2,6-xylidine (4-hydroxyxylidine) to 2,6-dimethylquinonimine. 4-hydroxyxylidine, a major metabolite of 2,6-xylidine, is an aminophenol. *In vivo*, 4-hydroxyxylidine undergoes extensive and rapid glucuronic and/or sulphonic acid conjugation. At high doses however, phase 2 metabolism may saturate in such a way that haem co-oxidation may occur, resulting in methaemoglobin and 2,6-dimethylquinonimine formation. A statistical increase in methaemoglobin formation is seen following intravenous doses of 30 mg/kg bw 2,6-xylidine in cats and 80 mg/kg bw in rats. Repeat dose toxicity studies identify 50 mg/kg bw 2,6-xylidine orally administered as NOAEL for methaemoglobin formation in rats. 2,6-dimethyl quinonimine-glutathione conjugates are evident following intraperitoneal doses of 90 mg/kg bw in rats. 2,6-dimethylquinonimine has been detected in urine and liver of rodents *in vivo*, following high doses of 2,6-xylidine administered intraperitoneally. Oxygen radicals are known to cause DNA damage (G:C to A:T transitions) and to induce C8 DNA adducts, which can be a cause of spontaneous mutations. Chronic dosing or acute overdosing thus has the potential to saturate glutathione binding and deplete glutathione, leading to cytotoxic and clastogenic effects. *In vitro* systems do not contain co-factors necessary for phase 2 metabolism and may be deficient in glutathione, methaemoglobin reductase and/or other antioxidants. Hence, reactive oxygen species formation and associated clastogenic effects may occur *in vitro* that are not seen *in vivo* except at high doses.

Hence, 2,6-xylidine is believed to exhibit genotoxic (clastogenic) effects only under certain activation conditions and beyond certain levels of exposure. Based on the overall weight of evidence and especially on the data provided for metabolism in the liver, it is concluded that detoxification of 2,6-xylidine is saturated above a certain dose *in vivo*, resulting in metabolites that potentially may drive genotoxicity through reactive oxygen species production. Regarding the most sensitive tissue for carcinogenicity, i.e. the nasal cavity, 2,6-xylidine metabolism data are lacking and a threshold dose for genotoxicity in this tissue cannot be derived from the above data. Overall, consumer risk assessment is based on the carcinogenicity data for 2,6-xylidine assuming a threshold mechanism of action.

**Carcinogenicity**

No carcinogenicity studies using lidocaine are available. 2,6-xylidine, an *in vivo* metabolite of lidocaine, has been associated with carcinogenicity in rats. In a carcinogenicity study, rats were given 2,6-xylidine by oral gavage at doses of 0, 300, 1000 and 3000 mg/kg per day for two years. Statistically significant decrease in mean body weight gains were seen at 3000 mg/kg in both sexes and at 1000 mg/kg in females. Increases in the number of adenomas and carcinomas of the nasal cavities were found in rats of both sexes of the 1000 and 3000 mg/kg groups. Unusual rhabdomyosarcomas and malignant mixed tumours of the nasal cavity were observed in both sexes of the high dose group. No relevant subcutaneous fibromas and fibrosarcomas were noted at any dose, although subcutaneous fibrosarcomas were observed in three high dose females, one high dose male, one mid dose female, one low dose male and one control female. Also, an increased

incidence of neoplastic nodules in the liver of female rat→ (but not in males) was observed. Hepatocellular carcinomas were observed in one control, one mid-dose and one high dose female rat. In males, a high dose rat had a neoplastic nodule, and a control and a mid-dose male had a hepatocellular carcinoma. No NOEL was established in the study. Benchmark dose analyses were performed using data for overall tumour incidence, separately for males and for females. A $BMDL_{10}$ of 51.4 mg/kg bw per day was obtained for males as a worst-case estimation, generated using the Log-Probit model. It was noted that actual exposure levels of the rats are likely to be below the nominal doses in feed. It was evident in a stability test that open storage of the feed at room temperature over 7 days resulted in losses of 9% (day 1) to 43.9% (day 7) of the test substance. The nominal doses and the resulting $BMDL_{10}$ must therefore be corrected for the possible losses of 2,6-xylidine in feed over the full study duration. Although it is not known which is the most relevant value, a correction for the range of losses would result in a $BMDL10_{corrected}$ range of 28.8 to 46.8 mg/kg bw per day as points of departure for risk assessment (the lower value being the most conservative).

**Studies of other effects including immunotoxicity and neurotoxicity**

No studies have been carried out with lidocaine investigating potential immunotoxicity. However, these studies are not considered necessary because there is no initial concern of immune system damage according to available literature.

Neurotoxicity studies have not been performed with lidocaine. However, observations in humans do not raise a concern of neuropathological effects after prolonged exposure.

## 2.1.4. Calculation of the toxicological ADI or alternative limit

A toxicological ADI is established from a value derived from human data based on the systemic dose that is achieved when human subjects are topically exposed to lidocaine for which chronic use has been widely reported. Administered topically in humans, lidocaine doses (via medicated plasters) for up to 5 years do not show systemic toxicity or drug-related adverse systemic effects. The systemic NOAEL is 1 mg/kg/day, which, by applying a 10-fold factor for individual variation, results in a toxicological ADI of 0.1 mg/kg/day or 6 mg/person/day. This is considered to be a conservative value, since the NOAEL is based on the systemically absorbed dose of lidocaine, whereas oral exposure would result in a lower bioavailability. This value is derived from long-term exposure studies in humans and results in a more conservative assessment than available toxicity studies performed in animals using lidocaine. The toxicological ADI of 6 mg/person/day or 0.1 mg/kg/day is established for the risk assessment of lidocaine.

Given the occurrence of the metabolite 2,6 xylidine as residue in edible tissues as well as from human metabolism of lidocaine, a toxicological reference value for exposure to this metabolite is required. 2,6-xylidine is believed to exhibit genotoxic (clastogenic) effects only under certain activation conditions and beyond certain levels of exposure. Based on the overall weight of evidence and especially on the data provided for metabolism in the liver, it is concluded that detoxification of 2,6-xylidine is saturated above a certain dose in vivo, resulting in metabolites that potentially may drive genotoxicity through reactive oxygen species formation. Therefore, consumer risk assessment is based on the carcinogenicity study using 2,6-xylidine and assuming a threshold mechanism of action. Based on a $BMDL_{10\ corrected}$ range of 28.8 to 46.8 mg/kg bw per day from the carcinogenicity study and using an interspecies uncertainty factor of 10, an intraspecies uncertainty factor of 10 and a further uncertainty factor of 10 to account for severity of effect and the quality of the database, a range of the acceptable exposure levels for 2,6-xylidine of 1728 to 2808 µg/person per day was obtained.

### 2.1.5. Overview of microbiological properties of residues

Lidocaine was shown to have antimicrobial activity towards bacterial strains which may colonize the human intestine. Mean MIC concentrations ranged from 2 to 8 mg/ml in a study where antimicrobial activity of lidocaine in Xylocaine was tested mainly in oral and a few intestinal bacterial strains. With pure lidocaine hydrochloride, a MIC of 8.0 mg/ml was observed in *Fusobacterium nucleatum*. No studies on disruption of the colonisation barrier and the increase of the population of resistant bacteria were provided. The MIC values reported for lidocaine are clearly outside the range of MICs that would normally be considered relevant for the calculation of a microbiological ADI.

Nevertheless, the microbiological ADI was calculated based on the approach described in VICH GL 36 and the disruption of the colonisation barrier endpoint. Taking into account 36% as the fraction of lidocaine potentially available to microorganisms, in accordance with 24-hour urine recoveries between 64 and 75% in human following oral doses of lidocaine, and the mean MIC values of the most susceptible species from those considered as relevant or related to the human gastrointestinal tract microbiome (2 mg/kg), a microbiological ADI of 20.37 mg/person was calculated.

Further assessment of microbiological activity of lidocaine is considered unnecessary.

### 2.1.6. Calculation of microbiological ADI

The MIC values reported for lidocaine are clearly outside the range of MICs that would normally be considered relevant for the calculation of a microbiological ADI. Nevertheless, a microbiological ADI of 20 mg/person was derived.

### 2.1.7. Observations in humans

Multiple applications of lidocaine 5% medicated plasters for up to 14.5 months from two phase III clinical trials (up to 2.5 months in the first trial and up to 12 months in a follow-up trial) in post-herpetic neuralgia patients (212 patients) showed that no tissue accumulation is expected according to computer-based simulations. Long-term safety of 5% lidocaine medicated plasters was investigated in 102 patients suffering from post-herpetic neuralgia during 4 years. Drug-related adverse events were reported in 19 of 102 patients, mainly mild to moderate localized skin reactions. In an additional 2-year study with lidocaine 5% medicated plasters, 249 patients suffering from post-herpetic neuralgia showed similar drug-related local adverse events, for instance skin reactions. Another study also investigated the effects of 5% lidocaine medicated plaster in 20 patients with localised neuropathic pain. Reversible erythema occurred in two patients wearing the plaster for more than 16 hours. There were no systemic side-effects observed during 5 years.

In a carcinogenicity case-control study, authors utilised data from two Kaiser Permanente medical care programmes to evaluate risks of hematopoietic and lymphoproliferative (HLP) malignancies after use of 14 common medications. The lidocaine group contained 112 cases (non-Hodgkin lymphoma, multiple myeloma and leukaemia combined) and 138 matched controls. Using a minimum 1-year lag between first notation and malignancy diagnosis there was no increase in risk associated with exposure to lidocaine. In a clinical trial, 63 patients undergoing surgery for breast cancer were dosed at 1.5 mg/kg/h lidocaine intravenously. It was found that the neutrophil-to-lymphocyte ratio changes were smaller in breast cancer patients who had received a lidocaine infusion during surgery, compared to the control group, showing that lidocaine has considerable anti-inflammatory effects.

Methaemoglobin levels were investigated in 90 healthy children undergoing dental treatment with one single dose of 2.5 mg/kg of 2% lidocaine in conjunction with 1:100000 adrenaline by the intravenous route. The mean peak methaemoglobin level of the lidocaine group was 1.63% while peak methaemoglobin level of the no local anaesthetic group was 1.60%. These results suggest that lidocaine with adrenaline does not increase methaemoglobin levels above physiologic levels.

### 2.1.8. Findings of EU or international scientific bodies

In 2005, the Norwegian Scientific Committee for Food Safety published the manuscript 'Risk assessment of lidocaine residues in food products from cattle, swine, sheep and goats: Withdrawal periods for meat and milk' to determine whether withdrawal periods for lidocaine-containing products used in cattle, swine, sheep and goat under the cascade could be shortened. As a NOEL could not be established for lidocaine, alternative methods for the evaluation of human health safety after ingestion of food containing residues of the lidocaine metabolite 2,6-xylidine were considered. The T25 dose from the NTP rat 2,6-xylidine carcinogenicity study was calculated as 63.5 mg/kg bw. The Norwegian Scientific Committee for Food Safety was of the opinion that the MOE approach of 25,000, suggested by EFSA in 2005 for compounds that have both genotoxic and carcinogenic properties could be used for the risk characterization of 2,6-xylidine. All calculated MOE values of 2,6-xylidine with different scenarios of daily intake of milk and meat were higher than the estimated MOE.In 2006, the Office of Chemical Safety of Australia (OCS) performed human food safety assessments for lidocaine and bupivacaine. In the absence of suitable NOELs from animal or human studies, the OCS considered human therapeutic doses as an appropriate endpoint from which to derive a LOEL for establishing an ADI. The ADI for lidocaine was established at 0.009 mg/kg bw/day based on route-to-route extrapolation from a LOEL derived from human therapeutic use. It was determined considering the low end of 1–10 mg/kg bw as the typical therapeutic range for cardiac treatment in humans by injection, a 35% oral bioavailability and a safety factor of 1000 due to intraspecies variability, inadequate toxicological database and the fact that the point of departure represents a LOEL.

In 2015, the CVMP performed an assessment '[...] pursuant to Article 30(3) of Regulation (EC) No 726/2004 in relation to the potential risk for the consumer resulting from the use of lidocaine in food producing species' (EMA/CVMP/124059/2015). At that time, the Committee concluded that 2,6-xylidine is a genotoxic carcinogen in rats and assumed that no threshold exists for genotoxicity. A no observed effect level (NOEL) was not established for carcinogenicity from the NTP carcinogenicity study (see above) and the Threshold of Toxicological Concern (TTC) approach was used for the risk assessment of 2,6-xylidine. In the frame of the present procedure, the genotoxicity/carcinogenicity of 2,6-xylidine was further evaluated and it was concluded that the mechanism of genotoxic/carcinogenic action is threshold-based. Based on this and taking a worst-case approach, the CVMP considers that the data from this same NTP carcinogenicity study is adequate for risk assessment, although it acknowledges deficiencies regarding data quality.

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the CVMP considers that the data from this same NTP carcinogenicity study is adequate for risk assessment, although it acknowledges deficiencies regarding data quality.

### 2.1.9. Overall conclusions on the ADI

A pharmacological ADI of 25 mg/person was established based on NO(A)EL of 250 mg/person seen in humans and an uncertainty factor of 10 to account for intraspecies variation.

A toxicological ADI of 6 mg/person/day or 0.1 mg/kg/day is established based on the systemic NOAEL of 1 mg/kg/day in human patients and applying a 10-fold factor for individual variation. This is a conservative value, since the NOAEL is based on the systemically absorbed dose of lidocaine, whereas oral exposure would result in lower bioavailability.

Given the occurrence of the metabolite 2,6-xylidine as residue in edible tissues as well as from human metabolism of lidocaine, a toxicological reference value for exposure to this metabolite is required. Based on a $BMDL_{10\ corrected}$ range of 28.8 to 46.8 mg/kg bw per day from a carcinogenicity study and using an interspecies factor of 10, an intraspecies factor of 10 and a severity of effect and the quality of the database factor of 10, the acceptable exposure level for 2,6-xylidine was calculated to be 1728 µg/person per day.

MIC values of lidocaine are in a range (mg/ml) not considered relevant for consumer risk assessment. Nevertheless, a microbiological ADI of 20 mg/person was derived.

Therefore, an overall ADI for lidocaine of 6 mg/person/day and an acceptable exposure level of 1728 µg/person/day for 2,6-xylidine are established.

## *2.2. Residues assessment*

### 2.2.1. Pharmacokinetics in target species

One GLP-compliant metabolism study was provided, in which piglets were treated following the intended therapeutic indication of a combination product (Tri-Sofen) containing lidocaine as well as bupivacaine, adrenaline and cetrimide. Bupivacaine and lidocaine were metabolised to similar metabolites and therefore no conclusion can be drawn regarding the exclusive metabolism of lidocaine in pigs.

In this study, thirty entire male piglets (1.35–2.91 kg bw, commercial hybrid) were randomly allocated to six groups of four animals each, and one group of six animals. Piglets were treated with Tri-Solfen (lidocaine 50.5 g/l; bupivacaine 5.02 g/l; cetrimide 5.06 g/l) topically for either tail docking or castration.

For castration (groups 1–5), piglets up to 4 kg bw were treated at a dose of 0.6 ml/kg bw divided equally per side of the castration. Piglets of 4 kg bw and above were treated at a fixed volume of 2.4 ml divided equally per side of the castration. The product was applied into the incised scrotum and piglets were castrated 20 seconds later.

In a separate group (group 6), Tri-Solfen was applied to the wound within one minute after tail docking at a dosage of 0.3 ml.

Several samples from blood, urine and faeces were collected from animals in groups 5 and 7 in a time interval from 5 minutes up to 4 days post-treatment. Additionally, blood, urine and faecal samples were collected from all animals at sacrifice.

Triplicate samples of skeletal muscle (loin), liver, kidney and skin with subcutaneous fat in natural proportions from over the shoulder were collected from each animal at 5 minutes (group 1), 4 hours (group 2), 12 hours (group 3), 24 hours (groups 4 and 6) and 7 days (group 5) after treatment. Additional sampling from treated spare animals (group 7) at 42 days post treatment was conducted.

Samples were analysed for parent compound and the metabolites 2,6-xylidine, 3-OH lidocaine, monoethylglycinexylidide, lidocaine-*N*-oxide and glycinexylidide. At the same time, bupivacaine and its metabolites as well as cetrimide were also quantified in the samples.

Lidocaine and its metabolites were detected in plasma, faeces and urine until 24 hours post treatment. In plasma, the main analyte found was the parent compound followed by lidocaine-*N*-oxide and 2,6-xylidine. After 7 days of treatment only the parent compound, monoethylglycinexylidide, lidocaine-*N*-oxide and glycinexylidide were detected.

In urine, the parent compound and the metabolites 3-OH lidocaine, monoethylglycinexylidide and lidocaine-*N*-oxide were the analytes found in high concentrations but glycinexylidide and 2,6-xylidine were also found until 7 days and 24 hours, respectively.

In faeces, the pattern of excretion of the lidocaine and its metabolites were similar to the urine excretion but in lower amounts.

A second metabolism study was also conducted with the combination product (Tri-Solfen) containing bupivacaine plus lidocaine, adrenaline and cetrimide and applied to 9 study groups and with 0.5% bupivacaine (as hydrochloride) solution applied to 3 study groups. Lidocaine and its metabolites as well as cetrimide residues were found in samples from bupivacaine-only treated animals, indicating cross-contamination of samples. Results from this study were considered unreliable and not taken further into account.

In addition to the study in piglets, some pharmacokinetic data relating to lidocaine in cattle are available.

After lactating cows were treated with lidocaine by the subcutaneous and epidural routes, residues of the drug were detected in serum and milk. No attempt was made to identify or quantify any metabolites. In a separate study, when cattle were treated with lidocaine prior to surgery, 2,6-xylidine was found in the milk. In cattle treated with lidocaine intramuscularly, the injection site, meat, liver and kidney were analysed for concentrations of lidocaine, 2,6-xylidine, 3'-hydroxyxilidine and other metabolites. 2,6-xylidine was found in meat and milk from treated cows while monoethylglycinexylidide, 3'-hydroxylidocaine, 4-hydroxyxilidine and lidocaine-*N*-oxide were also identified. 4-hydroxyxilidine was the most important metabolite in urine. Concentrations of all compounds decreased rapidly after administration.

In cattle treated subcutaneously with 10 mg/kg bw lidocaine, the major metabolite in plasma was 2,6-xylidine. It constituted up to 98% of the urinary metabolites. However, in sheep treated in the same manner with the same dose, 2,6-xylidine was only a minor plasma metabolite and accounted for only 3.6% of the urinary metabolites. In both species, recovery of lidocaine metabolites was low at 9.0% of the administered dose in cattle and 8.4% in sheep, respectively. 3-hydroxyxilidine and 4-hydroxyxilidine were present at very low concentrations. In contrast to that, 2,6-xylidine was not a metabolite of bupivacaine in these species. 4-hydroxyxilidine was a minor plasma metabolite in cattle, with even lower concentrations of lidocaine-*N*-oxide and monoethylglycinexylidide observed, while in sheep, 4-hydroxyxilidine was a minor plasma metabolite, with lower concentrations of monoethylglycinexylidide, 3-hydroxylidocaine, 2,6-xylidine, 3-hydroxyxilidine and lidocaine-*N*-oxide measured. Residues of lidocaine were determined in muscle, fat, liver, kidney and brain 72 hours and 7 days after administration. The major metabolites found were lidocaine and glycinexylidide as

well as some monoethylglycinexylidide. Residues at 72 hours were very low in all tissues, while, after 7 days, they were either non-detectable or extremely low in concentration. Fat appeared to be the target tissue in sheep and fat and muscle appeared to be the target tissues in cattle.

## 2.2.2. Residue depletion studies

**Depletion in tissues**

No radiometric residue depletion studies were conducted. The applicant provided results from one non-radiolabelled residue depletion study conducted with a combination product (Tri-Solfen) containing bupivacaine plus lidocaine, adrenaline and cetrimide. For information on the study design see section 2.2.1.

Residues of lidocaine and its metabolites were found in all edible tissues, with slightly higher concentrations in skin and fat. Residues of lidocaine and its metabolites were low 24 hours after treatment but residues persisted at slightly higher concentrations in skin and fat. Residues of one lidocaine metabolite, lidocaine-*N*-oxide, were not determined in liver and kidney tissues. Residues of 2,6-xylidine were higher than 100 µg/kg in all tissues during the first 24 hours after treatment. Seven days after treatment the residues of 2,6-xylidine were higher than 0.1 µg/kg in all the edible tissues.

The lidocaine metabolites analysed (2,6-xylidine, monoethylglycinexylidide, glycinexylidide, 3-OH-lidocaine and lidocaine-*N*-oxide) in the study would represent the major metabolite profile of lidocaine in pigs and humans. Taking into consideration the nature of the minor metabolites not analysed in this study, it can be concluded that they can be considered as being substances of minimum impact on consumer safety.

As a combination product was used in the residue depletion study and as both lidocaine and bupivacaine may be metabolised to 2,6-xylidine, some uncertainties remain regarding the proportion of 2,6-xylidine attributable to the metabolism of lidocaine as well as the proportion resulting from metabolism of bupivacaine. Due to the low biotransformation of bupivacaine to 2,6-xylidine and the low bupivacaine dose, the concentrations of 2,6-xylidine found in the metabolism study would be mainly of lidocaine provenance.

From a pharmacological and toxicological point of view, the main relevant substances are lidocaine and the lidocaine metabolite 2,6-xylidine. Total residue intake can be assumed using the residue depletion data available for lidocaine and 2,6-xylidine. In the case of 2-6-xylidine, several issues have to be considered in the frame of a worst-case scenario situation: The first point to consider is that all 2,6-xylidine precursors are converted to 2,6-xylidine after human consumption of lidocaine residues. The second point to consider is that all potentially consumed 2,6-xylidine originates from ingested lidocaine residues. Finally, the third point to consider is the estimated amount of 4-OH-xylidine residues. Taking these points into account, the total calculated intake of 2,6-xylidine is presumed to be 883 µg/person/day, 64.5 µg/person/day and 3.5 µg/person/day, 12 hours, 24 hours and 7 days after treatment with lidocaine, respectively. These amounts of 2,6-xylidine are below the acceptable level of exposure of 1728 µg/kg/day established in section 2.1.9.

**Selection of marker residue and the ratio of marker to total residues**

Selection of a marker residue and of a ratio of marker to total residues are not needed, as the utilized portion of the ADI is sufficiently low to allow for a 'No MRL required' status (see section 3.2 below).

### 2.2.3. Monitoring or exposure data

No monitoring or exposure data other than that described elsewhere in this report were available.

### 2.2.4. Analytical method for monitoring of residues

The applicant developed a residue analytical method allowing for the determination of lidocaine and its metabolites and provided a validation report.

Lidocaine and its metabolites 3-OH lidocaine, monoethylglycinexylidide and lidocaine-*N*-oxide are quantified using ultra-high performance liquid chromatography (UHPLC) coupled to a tandem mass spectrometer. For 2,6-xylidine and glycinexylidide quantification other chromatography conditions were used.

### 2.2.5. Potential effects on the microorganisms used for industrial food processing

The substance is not intended for use in dairy animals and therefore potential effects in dairy products were not investigated.

### 2.2.6. Findings of EU or international scientific bodies

No relevant evaluations by EU or international scientific bodies were identified.

# 3. Risk management recommendations

## *3.1. Availability of alternative medicines and other legitimate factors*

**Availability of alternative medicines**

The intended use of lidocaine in porcine species is for local anaesthesia during and after castration of piglets. This would result in a potentially wide use in a major species. However, the treated animals are unlikely to be sent to slaughter immediately after treatment.

There are limited options available which can be used for anaesthesia/treatment of pain associated with the castration of piglets. These include local anaesthetic products for injection and NSAIDs.

**Technological aspects of food and feed production (potential effects on the microorganisms used for industrial food processing)**

Piglet meat is not further processed to food or feed products using microorganisms.

**Conditions of use**

Use of lidocaine needs to be restricted to piglets up to 7 days of age. Available data indicate that pigs at higher age more extensively metabolise the substance in question, potentially resulting in higher concentrations of 2,6 xylidine. Furthermore, the use needs to be restricted to the cutaneous and epilesional routes as this is the scenario used in the available studies and assessed by the CVMP. Use of lidocaine via injection may require higher doses or may lead to a different residue depletion pattern and may, therefore, require additional risk management measures in order to ensure consumer safety.

**Other factors that should, if applicable, be taken into consideration in support of the MRL recommendation**

No other relevant factors were identified for consideration of the risk management recommendations.

## *3.2. Elaboration of MRLs*

The toxicological ADI for lidocaine is considered the overall ADI to be used for the risk assessment and is 0.1 mg/kg bw per day or 6 mg/person per day. It is considered appropriate to reserve 20% of the ADI in case future uses of the substance would result in residues in other food commodities, particularly in milk. Consequently, the maximum amount of lidocaine that can be accepted in relation to residues in tissues is 4.8 mg/person/day. Using the same reasoning, the maximum acceptable amount of the genotoxic metabolite 2,6-xylidine that may be present in edible porcine tissue is calculated to be 1382 µg/person per day (worst-case scenario).

Residue depletion data demonstrate that a total intake of residues, including lidocaine plus its metabolites at 12 hours post treatment would equal 1029 µg/person per day based on the standard food basket and 84 µg/person per day at 24 hours. The margin between the amount of residues per food basket and the proportion of the ADI that can be considered in relation to residues in tissues (4.8 mg/person/day) is approximately 4.5 at 12 hours after treatment and approximately 57 at 24 hours after treatment.

Concerning risk assessment of the carcinogenic metabolite 2,6 xylidine, the exposure to 2,6-xylidine at 12 hours after treatment (883 µg/person/day) and at 24 hours of treatment (64.5 µg/person/day) is compared to the substance-specific reference value of 1728 µg/person per day or 1382 µg/person per day when reserving 20% of the reference value for residues in milk.

In view of the fact that exposure to residues of lidocaine can be expected to be substantially below the ADI at 12 hours after dosing and that, at this time point, exposure to the genotoxic metabolite 2,6 xylidine will also be below the relevant reference value, a 'No MRL required' classification is considered appropriate for lidocaine. However, this conclusion can only be applied considering the conditions used in the residue studies provided. Consequently, the use of lidocaine should be restricted to cutaneous and epilesional use in piglets up to 7 days of age only.

# 4. Considerations on possible extrapolation of MRLs

In line with Article 5 of Regulation (EU) No 470/2009, the CVMP considered the possibility of extrapolating the maximum residue limits established for lidocaine on the basis of residue data in piglets, to other food producing species. As discussed in section 3.1, lidocaine can be metabolised to a different extent in pigs of different ages. In view of the fact that lidocaine can be metabolised to a carcinogenic metabolite, extrapolating the current recommendation to species other than pigs would be associated with excessive uncertainty and consequently, based on the available data, the Committee considers that extrapolation of MRLs to other food producing species cannot be recommended.

# 5. Conclusions and recommendation for the establishment of maximum residue limits

Having considered that:

- the toxicological ADI of 6 mg/person/day was derived as the overall ADI and used for quantitative risk assessment of consumer exposure to lidocaine residues,
- an acceptable exposure level for 2,6-xylidine of 1728 µg/person per day based on substance-specific data was derived,
- the quantitative risk assessment based on lidocaine residues in piglet tissues resulted in a consumer exposure estimate well below the toxicological ADI at all timepoints investigated (i.e. from 12 hours after dosing),
- the quantitative risk assessment based on levels of 2,6 xylidine in piglet tissues plus levels of 2,6-xylidine produced by metabolism of lidocaine in humans resulted in a consumer exposure estimate below the acceptable exposure level at all timepoints investigated (i.e. from 12 hours after dosing),
- metabolism of lidocaine in neonatal piglets is considerably lower than that in pigs of a higher age and a corresponding restriction to use in neonatal animals is therefore considered appropriate,
- the available data only allowed assessment of residues resulting from epilesional and cutaneous application and a restriction to this route of administration is therefore considered appropriate,

the Committee concludes that the establishment of maximum residue limits for lidocaine in porcine is not necessary for the protection of human (consumer) health and therefore recommends the inclusion of lidocaine in Table 1 (allowed substances) of the Annex to Commission Regulation (EU) No 37/2010 of 22 December 2009 as follows:

| **Pharmacologically active substance** | **Marker residue** | **Animal species** | **MRLs** | **Target tissues** | **Other provisions** | **Therapeutic classification** |
|---|---|---|---|---|---|---|
| Lidocaine | NOT APPLICABLE | Porcine | No MRL required | NOT APPLICABLE | For use in piglets up to 7 days of age only<br>For cutaneous and epilesional use only | Local anaesthetic |

The theoretical maximum daily intake of residues from porcine tissues represents approximately 1.5% of the ADI 24 hours post administration. There is a margin of approximately 25 between the potential total consumer exposure to 2,6-xylidine and the reference value of 1728 µg/person per day.

# 6. Background information on the procedure

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| Submission of the dossier | 6 September 2018 |
| Steps taken for the assessment of the substance: | |
| Application validated: | 26 September 2018 |
| Clock started: | 27 September 2018 |
| List of questions adopted: | 24 January 2019 |
| Consolidated response to list of questions submitted: | 17 January 2020 |
| Clock re-started: | 20 January 2020 |
| List of outstanding issues adopted: | 18 March 2020 |
| Consolidated response to outstanding issues submitted: | 28 May 2020 |
| Clock re-started: | 17 June 2020 |
| CVMP opinion adopted: | 16 July 2020 |