7 November 2024
EMA/CVMP/490617/2024
Veterinary Medicines Division

# Opinion of the Committee for Veterinary Medicinal Products on the establishment of maximum residue limits

## Procedure no: EMEA/V/MRL/003652/MODF/0005

## Name of the substance: Ketoprofen (INN)

### Basis for the opinion

Pursuant to Article 11 of Regulation (EC) No 470/2009 of 6 May 2009, European Commission submitted to the European Medicines Agency on 01 March 2023 a request to assess the appropriateness of the "No MRL required" status of ketoprofen in bovine, porcine and *Equidae*.

On 18 April 2024 the Committee for Veterinary Medicinal Products adopted a call for data inviting all interested parties to submit scientific data for use in the assessment and not already reported when a "No MRL required" status was agreed by the CVMP for ketoprofen in the aforementioned species (with reference to EMEA/MRL/020/95 and EMEA/MRL/076/96-FINAL). The call for data was closed on 31 August 2024.

### Recommendation

The Committee, having considered the request and having evaluated the data provided during the call for data, recommends by consensus the establishment of numerical maximum residue limits for ketoprofen in bovine and porcine. Furthermore, with reference to Article 5 of Regulation (EC) No. 470/2009 and in line with the criteria laid down in Commission Regulation (EU) 2017/880, the Committee agreed that the proposed maximum residue limits could be extrapolated to all ruminants and *Equidae*. Therefore, the amendment of the entry for ketoprofen in table 1 of the Annex to Regulation (EU) No 37/2010 of 22 December 2009, is recommended as follows:

| Pharmaco-logically active substance | Marker residue | Animal species | MRLs | Target tissues | Other provisions | Therapeutic classification |
|---|---|---|---|---|---|---|
| Ketoprofen | Ketoprofen | All ruminants, porcine, *Equidae* | 50 µg/kg<br>20 µg/kg<br>20 µg/kg<br>50 µg/kg<br>20 µg/kg | Muscle<br>Fat<br>Liver<br>Kidney<br>Milk | For porcine species the fat MRL relates to 'skin and fat in natural proportions' | NO ENTRY |
| | Ketoprofen | Poultry | 10 µg/kg<br>30 µg/kg<br>10 µg/kg<br>10 µg/kg | Muscle<br>Skin and fat in natural proportion<br>Liver<br>Kidney | Not for use in animals from which eggs are produced for human consumption | NO ENTRY |

The Norwegian CVMP member agrees with the above-mentioned recommendation of the Committee.

The scientific conclusions of the Committee are presented in the European public MRL assessment report (EPMAR), provided in Annex I of this opinion.

The present opinion is forwarded to the European Commission and to the applicant together with its appendices.

# Annex I

# European public MRL assessment report (EPMAR)

Ketoprofen (bovine, porcine and *Equidae*)

## Summary of the scientific discussion for the establishment of MRLs

| | |
|---|---|
| Substance name: | Ketoprofen |
| Therapeutic class: | NO ENTRY |
| Procedure number: | EMEA/V/MRL/003652/MODF/0005 |
| Requestor: | European Commission |
| Target species: | Bovine, porcine and *Equidae* |
| Intended therapeutic indication: | Inflammatory and painful conditions of the bones, joints and muscular-skeletal systems in cattle, horses, dogs and cats; alleviation of pain associated with colic in horses and cattle; for reducing pyrexia and respiratory rate in case of respiratory infections in pigs and as supportive treatment of mastitis-metritis-agalactia syndrome in the sow. |
| Route(s) of administration: | Intravenous or intramuscular routes |

## 1. Introduction

Ketoprofen is a non-steroidal anti-inflammatory drug belonging to the arylpropionic acid group. It is used in human and veterinary medicine for its anti-inflammatory, analgesic and antipyretic purposes. It is indicated for inflammatory and painful conditions of the bones, joints and muscular-skeletal systems in cattle, horses, dogs and cats, for alleviation of pain associated with colic in horses and cattle, for reducing pyrexia and respiratory rate in case of respiratory infections in pigs and as supportive treatment of mastitis-metritis-agalactia syndrome in the sow. The dose in these food-producing species is 2 to 3 mg/kg bw by intravenous or intramuscular route. It is also intended for use in chickens for the treatment of inflammatory and painful conditions at an oral dose of 10 mg/kg bw/day for 5 days, administered in drinking water.

Ketoprofen was previously assessed by the CVMP and a toxicological (overall) ADI of 5 µg/kg bw/day (0.3 mg/person) and a pharmacological ADI of 7 µg/kg bw (0.42 mg/person) were established (EMA/CVMP/94885/2020).

Currently ketoprofen is included in Table 1 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 |
|---|---|---|---|---|---|---|
| Ketoprofen | NOT APPLICABLE | Bovine, porcine, *Equidae* | No MRL required | NOT APPLICABLE | NO ENTRY | NO ENTRY |
| | Ketoprofen | Poultry | 10 µg/kg<br>30 µg/kg<br>10 µg/kg<br>10 µg/kg | Muscle<br>Skin and fat in natural proportion<br>Liver<br>Kidney | Not for use in animals from which eggs are produced for human consumption | NO ENTRY |

The MRLs for poultry were established as a result of an extension application, submitted at the end of 2020 (EMA/CVMP/206677/2023-Corr.1). In its initial opinion, dated 12 May 2022, the CVMP recommended the establishment of a "No MRL required" status for poultry tissues. However, four CVMP members signed a divergent position and, as a result, the Standing Committee on Veterinary Medicinal Products was not able to support the CVMP opinion. Therefore, in a letter to the Agency dated 1 March 2023, the European Commission requested the CVMP to reconsider its opinion and indicated that, if the "No MRL required" status for poultry could not be further substantiated, the CVMP was requested to recommend numerical MRLs. The European Commission further indicated that, if the eventual outcome of the procedure was the establishment of numerical MRLs for poultry tissues, a review of the MRL status for the substance in bovine, porcine and *Equidae* should follow. The final CVMP opinion for poultry, dated 16 May 2023, did recommend numerical MRLs, and these MRLs were subsequently adopted by the European Commission[1]. Consequently, the current procedure was initiated to allow a review of the appropriateness of the "No MRL required" status of ketoprofen in bovine, porcine and *Equidae*, in accordance with Article 11 of Regulation (EC) No 470/2009.

# 2. Scientific risk assessment

## *2.1. Safety assessment*

The CVMP has previously assessed the consumer safety of ketoprofen and established an overall ADI of 0.005 mg/kg bw, i.e. 0.3 mg/person, based on the BMDL5 of 0.04 mg/kg bw/day for kidney lesions in rats and a chemical specific uncertainty factor of 7.6 (EMA/CVMP/94885/2020). Therefore, no further assessment regarding the consumer safety of the substance was required for the purpose of this modification request.

## *2.2. Residues assessment*

The CVMP previously reported residue depletion data in bovine, porcine and *Equidae*, and concluded on a "No MRL required" status for ketoprofen in these species (EMEA/MRL/020/95 and EMEA/MRL/076/96-FINAL). On 16 May 2023 the Committee adopted an opinion recommending the extension of maximum

[1] Commission Implementing Regulation (EU) No 2023/2194, O.J., of 20 October 2023

residue limits for ketoprofen to poultry (EMA/CVMP/206677/2023-Corr.1). The current report relates specifically to a request for the revision of MRLs for bovine, porcine and *Equidae* and only data relevant to this specific request are described.

No new pharmacokinetic or residue data in target animal species were submitted with this modification request. The CVMP therefore issued a call for data, inviting all interested parties to submit scientific data not already reported when a "No MRL required" status was agreed by the CVMP for ketoprofen and a number of additional studies were retrieved. Most of them were deemed of low relevance for the current modification request, however, some useful residue depletion data was submitted and used in the assessment, as presented below. Additionally, a systematic literature search was performed to address data gaps identified.

### 2.2.1. Pharmacokinetics in target species

The CVMP previously reported the following pharmacokinetic data in target animals.

Ketoprofen is converted into a carbonyl-reduced derivative, the RP 69400 (2-(phenyl 3-alpha-hydroxybenzoyl) propionic acid).

Ketoprofen is strongly bound to proteins (97% in cattle). After single intramuscular administration of 3 mg of ketoprofen/kg bw supplemented with $^{14}C$ ketoprofen, plasma data showed a good relationship between total radioactivity and the sum of ketoprofen and RP 69400.

Ketoprofen represents about 50% of total radioactivity from 4 to 8 hours post treatment.

In cattle after intramuscular administration, ketoprofen is rapidly absorbed ($t_{1/2}$ ka = 0.15-0.25 h). Bioavailability ranged from 85% to 100%. After a single intramuscular administration of 3 mg of ketoprofen/kg bw (with $^{14}C$ methyl-ketoprofen), 90% of the radioactivity was recovered in the urine within 96 hours: 90-93% of the radioactivity was due to the metabolite RP 69400, whereas ketoprofen only represented 1%. The hydroxyl derivatives in position 3 and 4 represented only 0.5 to 2.7% of the urine metabolites. About 6% of the administered dose was recovered in the faeces.

After a single intramuscular administration of 3 mg/kg bw of $^{14}C$-ketoprofen to calves, the ratio ketoprofen/total residue could be evaluated: 56% for muscle, 35% for fat, 2% for liver and 56% for kidney. At the injection site, the ratio was close to 85%.

In horses, after intravenous administration of 2.2 mg of ketoprofen/kg bw/day, ketoprofen and RP 69400 were no longer detected in plasma three hours after injection. In urine, concentrations of metabolite RP 69400 (free and conjugated forms) were lower than those of ketoprofen. Metabolites 3- and 4-hydroxy-ketoprofen could not be detected either in plasma or in urine.

The reduction of ketoprofen into metabolite RP 69400 was demonstrated in an in vitro metabolism study of $^{14}C$-ketoprofen by the microsomal and cytosol fractions of pig liver. However, this reduction is about two-fold lower than that in bovine species.

In pigs, after a single intravenous administration of 3 mg/kg bw, ketoprofen is poorly distributed, the steady-state volume of distribution (Vdss) being low (= 0.17 $\pm$ 0.02 l/kg). The mean residence time (MRT) was 2.32 $\pm$ 0.41 h.

Thirty minutes after a single intramuscular injection of 3 mg/kg bw of $^{14}C$-ketoprofen to pigs, a peak mean plasma concentration of 12.74 ± 2.50 mg equivalents $^{14}C$-ketoprofen/l was observed. The concentrations decreased rapidly to reach 0.07 ± 0.01 mg equivalents $^{14}C$-ketoprofen/l twenty-four hours later. For later collecting points, $^{14}C$-Ketoprofen was detected at trace amounts.

It was shown that about 84% of the plasma radioactivity in pigs was due to the parent compound, and 7% to RP 69400. The remaining radioactivity was due to unidentified polar materials accounting for about 8% of radioactivity.

In pigs, a balance study after a single intramuscular injection of 3 mg/kg bw of $^{14}$C-ketoprofen showed that 72% of the total radioactivity administered was excreted via urine within 96 hours, a majority being excreted within 24 hours. Only 20% were recovered in faeces.

In pig urine, about 30% of the radioactivity was due to RP 69400, 12% to the parent compound, and approximately 45% to polar components.

The concentrations of metabolite RP 69400 were below the limit of quantification of 0.1 mg/kg in all tissues, at all times and in all animals.

Due to very fast elimination of the radioactivity, the ratio of ketoprofen/total residues and RP 69400/total residues could only be evaluated in samples collected 3 hours after a single intramuscular administration of 3 mg/kg bw of $^{14}$C-ketoprofen to pigs.

The percentage of ketoprofen residues with respect to total residues were as follows: 31.5% for kidney, 0.3% for liver, 72% for fat, 94% at the injection site.

For RP 69400, the figures were respectively: 29% for kidney, 78% for liver, 10% for fat, and 3% at the injection site.

A number of pharmacokinetic studies performed in cattle and pigs were submitted as a result of the call for data. However, these data do not alter the previous conclusions on the absorption, distribution, metabolism and excretion of ketoprofen in bovine, *Equidae* and porcine species. To note, in the pig several polar metabolites were observed but not identified.

Since the CVMP is of the opinion that numerical MRLs should be established (see below) and, because metabolites may contribute to toxicity, a marker to total ratio should be established.

## 2.2.2. Residue depletion studies

The CVMP previously reported the following residues depletion data in target animals.

In cattle, after a single intramuscular administration of 3 mg/kg bw with $^{14}$C-ketoprofen, only trace levels were detected at the injection site, 96 hours after treatment. After repeated administrations of 3 mg/kg bw/day for 3 days, ketoprofen and RP 69400 could only be measured in kidneys, 24 hours after the third injection: 0.19 ± 0.14 µg/g for ketoprofen and 0.24 ± 0.17 µ/g for RP 69400. In other tissues, they were either non detectable (<0.025 µg/g for ketoprofen and 0.05 µg/g for metabolite RP 69400) or lower than the quantification limit (0.05 µg/g for ketoprofen, 0.1 µg/g for RP 69400). At the injection site, corresponding to the 3$^{rd}$ injection, only ketoprofen was detectable, the mean concentration being 1.51 ± 1.68 µg/g.

Ketoprofen and its metabolite RP 69400 could not be detected (<0.025 µg/ml) in the milk at any milking both during and after treatment by ketoprofen at the recommended dosage.

No residue depletion study was carried out in the horse. However, from the comparison of basic pharmacokinetic parameters obtained for cattle and horses, it can be concluded that tissue depletion of ketoprofen in horse will be faster than in cattle.

A radiometric depletion study was carried out in pigs with $^{14}$C-ketoprofen at the recommended intramuscular dosage of 3 mg/kg bw:

- Three hours post injection, the concentrations were the following: kidney: 11.63 mg equivalents-ketoprofen/kg, liver: 3.02 mg equivalents ketoprofen/kg, injection site: 12.40 mg equivalents ketoprofen/kg. In the samples of skin plus fat and in muscle, the amounts were close to 1 and 0.5 mg equivalents ketoprofen/kg;
- Twenty four hours post dose, the radioactivity levels had already decreased to 2.07 mg equivalents ketoprofen/kg in kidney and to 0.24 mg equivalents ketoprofen/kg in liver. In other edible tissues, the concentrations were close to the limit of quantification (0.03 and 0.09 mg equivalents ketoprofen/kg for muscle and skin plus fat respectively);
- Ninety six hours post dose, $^{14}$C-ketoprofen could only be measured in kidney (0.82 mg equivalents ketoprofen/kg) and in liver (0.07 mg equivalents ketoprofen/kg).

In a non-radiometric study in pigs carried out with 3 x 3 mg/kg bw at 12 hours apart, the concentrations of ketoprofen in the liver, muscle and skin-fat, were below the limits of quantification (0.050 mg/kg) in all animals except one in which the levels in the skin-fat and muscle were 0.058 and 0.060 mg/kg respectively, twenty-four hours after the end of the treatment. In kidney, the levels ranged from 0.067 to 0.097 mg/kg in 3 pigs and for the 4th animal, the concentration was below the limit of quantification (0.050 mg/kg). Twenty-four hours post dosing, the ketoprofen levels at the injection sites ranged from 20.80 to 0.40 mg/kg. Four and ten days after the final injection, the concentrations of ketoprofen were below the limits of quantification of the method (0.050 mg/kg) for all the tissues, the injection site included.

To note, these studies are old and do not meet today's standards. A discrepancy in the residue data precluding a robust determination of marker to total ratios in pig liver was also detected.

As a result of the call for data, no new total residue studies or additional data in *Equidae* were submitted. However, several more recent residue depletion studies in calves and pigs were submitted.

In a residue depletion study in calves administered ketoprofen orally in drinking water with 3 mg/kg bw during three consecutive days, 4 animals were slaughtered at each time point of 12, 24, 48, and 96 hours. Very low levels or no trace of the ketoprofen or its main metabolite (RP 69400) were observed in the muscle and fat tissue samples. Ketoprofen was measured in the liver from two animals at 4 days (28 and 31 µg/kg) and in the kidney in all four animals at 12 h, ranging from 284 to 421 µg/kg. The metabolite was measured in the liver in three animals at 12 h in the range 73 to 103 µg/kg and in the kidney in all four animals in the range 219 to 474 µg/kg. The limits of quantification for ketoprofen and RP69400 were 20 µg/kg and 50 µg/kg, respectively, in liver and kidney, and 10 µg/kg and 40 µg/kg, respectively, in muscle and fat.

In a residue depletion study in pigs administered ketoprofen orally in drinking water with 3 mg/kg bw during three consecutive days, 4 animals were slaughtered at each time point of 8, 24, 48, and 96 hours. Levels of ketoprofen and its main metabolite (RP 69400) were observed in the muscle and fat tissue samples in single animals at different timepoints (up to 34 µg/kg of ketoprofen at 8 h and 89 µg/kg of the metabolite at 24 h). Ketoprofen was measured at 4 days in the liver in three animals ranging from 31 to 67 µg/kg and in the kidney in two animals, at concentrations of 22 and 28 µg/kg. The metabolite was also measured in the liver in three animals at 4 days in the range 73 to 87 µg/kg. The limits of quantification for ketoprofen and RP69400 were 20 µg/kg and 50 µg/kg, respectively, in liver and kidney, and 10 µg/kg and 40 µg/kg, respectively, in muscle and fat.

In a residue depletion study in calves administered ketoprofen intramuscularly with 3 mg/kg bw during three consecutive days, 4 animals were slaughtered at each time point of 12, 24, 48, and 96 hours. The concentrations of ketoprofen in injection site tissue were slightly over the LOQ, ranging from 13 to 46 µg/kg, at 24 h in three animals. Also, kidney tissue showed levels of ketoprofen (47 and 91 µg/kg) and its metabolite (106 and 169 µg/kg) in two animals at 24 h. At 48 h all levels of ketoprofen and its

metabolite fell below the LOQ or were not detectable in any of the tissues. The limits of quantification for ketoprofen and RP69400 were 20 µg/kg and 50 µg/kg, respectively, in liver and kidney, and 10 µg/kg and 40 µg/kg, respectively, in muscle and fat.

In a residue depletion study in sows administered ketoprofen intramuscularly with 3 mg/kg bw during three consecutive days, 4 animals were slaughtered at each time point of 12, 24, 48, and 72 hours. Concentrations of ketoprofen ranging from 0.5 to 3.5 mg/kg and 0.015 to 0.5 mg/kg were found in all 4 animals in the injection site and in 3 animals in the surrounding area, respectively, at 48 h. At 48 h, residues of ketoprofen were also observed in the kidney in 4 animals at concentrations ranging from 21 to 34 µg/kg. At 72 h residues of ketoprofen were observed in the injection site in two animals (13 and 96 µg/kg) and in surrounding tissue in one animal at 18 µg/kg. Residues of the metabolite were found in all 4 animals in the kidney, injection site and in 3 animals in the surrounding tissue at 12 h. At 24 h residues of the metabolite were observed in two animals in the kidney at 73 and 109 µg/kg. The limits of quantification for ketoprofen and RP69400 were 20 µg/kg and 50 µg/kg, respectively, in liver and kidney, and 10 µg/kg and 40 µg/kg, respectively, in muscle and fat.

In a residue depletion study in pigs administered ketoprofen intramuscularly with 3 mg/kg bw during three consecutive days, 4 animals were slaughtered at each time point of 12, 24, 48, and 96 hours. Concentrations of ketoprofen were observed in two animals at 48 h in injection sites (91 and 102 µg/kg) and in three animals in surrounding tissue (19-30 µg/kg). In the kidney, levels of ketoprofen at about 30 µg/kg were observed in two animals at 24 h. The metabolite was only detected in single animals at 12 h. The limits of quantification for ketoprofen and RP69400 were 20 µg/kg and 50 µg/kg, respectively, in liver and kidney, and 10 µg/kg and 40 µg/kg, respectively, in muscle and fat.

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

Currently, ketoprofen has a "No MRL Required" status for bovine, porcine and *Equidae*. Therefore, selection of a marker residue and determination of ratios of marker to total residues was deemed not necessary.

In its opinion of 16 May 2023 recommending the extension of maximum residue limits for ketoprofen to poultry (EMA/CVMP/206677/2023-Corr.1), the Committee concluded that ketoprofen, as the residue with pharmacological activity, is the appropriate marker residue for monitoring, and agreed that the calculation of consumer intake of residues should take account of total residues (i.e. including all metabolites) and consequently a ratio of marker to total residues was established. From the radiolabel study data in poultry, where ketoprofen was administered in gelatine capsules, the following ratios of marker to total residues of 0.043, 0.031, 0.02, 0.003 for muscle, fat plus skin, liver and kidney observed 6 hours after treatment were retained.

As the CVMP now proposes to also set numerical MRLs for bovine, porcine and *Equidae*, a marker residue should be proposed. The CVMP considers that ketoprofen is the appropriate marker residue in bovine, porcine and *Equidae*. The following ratios of ketoprofen to total residues for cattle were given in the original summary report (EMEA/MRL/020/95): 56%, 85%, 2%, 56%, and 35% in muscle, injection site, liver, kidney, and fat, respectively. For pigs, the ratios were 94%, 0.3%, 31.5%, and 72% in injection site, liver, kidney, and fat, respectively. The ratio in pig muscle could not be determined in the total residue study, so the value for fat (72%), which exhibited the most similar residue levels to muscle, is applied. The very low ratio for pig liver (0.3%) yielded extreme intake estimates, complicating the proposal for MRL values that protect consumer safety. The ratio was adjusted to 2%, matching the lowest marker to total residues ratio established for liver in other species.

### 2.2.3. Monitoring or exposure data

The latest EFSA report on residues of veterinary medicinal products from 2022[2] found that the highest proportion of non-compliant samples among the B2 group of substances ('other veterinary drugs') was 0.19% for NSAIDs (group B2e). Consistent with this finding, tissue samples testing positive for ketoprofen, especially from cattle but also from pigs, have been recorded by the German national residue control program. Residues have been found in all edible tissues, not only in muscle, which might sometimes be the injection site. For example, from 2020-2024 the maximum residue concentration found in cattle muscle was 268 µg/kg, in cattle liver 31.3 µg/kg, and in cattle kidney 7300 µg/kg. With the standard food basket and marker ratio from the original summary report (EMEA/MRL/020/95), this corresponds to an estimated intake of 143.6 µg (268/0.56*0.3), 156.5 µg (31.3/0.02*0.1), and 652.8 µg (7300/0.56*0.05), respectively. The last value indicates that exposure can exceed both the toxicological and the pharmacological ADI through kidney tissue alone.

### 2.2.4. Analytical method for monitoring of residues

The CVMP previously reported that there is an adequate and transferable analytical HPLC method with UV detection technique to ensure the monitoring of ketoprofen residues in the various tissues of cattle and in muscle of horses. The limits of detection and of quantification of this method are 25 and 50 µg/kg respectively. The method would not be sufficiently sensitive for monitoring of residues at the MRLs currently being considered.

The CVMP also previously reported that an analytical method for residues detection in skin plus fat, liver and kidney of pigs has been validated according to the recommendations of Volume VI of the rules governing medicinal products in the European Union (which has been superseded by Regulation (EU) 2018/782) and described according to the format ISO 78/2. The limits of quantification for ketoprofen and RP 69400 were 0.050 and 0.100 mg/kg respectively. The limits of detection ranged between 0.017 to 0.031 mg/kg for ketoprofen and from 0.027 to 0.066 mg/kg for RP 69400. Again, the method would not be sufficiently sensitive for monitoring of residues at the MRLs currently being considered.

According to the EU Reference Laboratory for Residues of Veterinary Drugs, there are methods to detect acidic NSAIDs in muscle and milk with UHPLC-MS/MS. The method was tested with cattle milk and cattle and horse muscle as matrices. The EU Reference Laboratory provided a full description of the analytical methods indicating that ketoprofen can be detected in muscle in the range of 0.5 to 10 µg/kg, and 5 to 60 µg/kg in milk. Further, the methods were robust to the operator, the storage time, solid phase extraction cartridge batch, HPLC column batch changes and other random effects (as observed over a validation period of one month). The increased sensitivity of this state-of-the-art analytical method was taken into consideration when considering MRL values proposed. The methods can be considered to be validated in line with the requirements set out in VICH GL49 and to provide a suitable basis for the official control of veterinary medicinal product residues at the level of the proposed MRLs. While the method has only been validated for quantification of residues in muscle and milk, it is considered to provide an adequate basis for development of methods for monitoring of residues in fat, liver and kidney.

The current assessment is not the result of an application from a company to establish MRLs, so no commitment has been made to provide standards. However, ketoprofen has been authorised for use in food producing animals for many years and the availability of standards is not considered to be a problem.

[2] European Food Safety Authority (EFSA), 'Report for 2022 on the results from the monitoring of veterinary medicinal product residues and other substances in live animals and animal products', EFSA J, Vol. 21, No. 3, 2024, e8669. DOI: https://doi.org/10.2903/sp.efsa.2024.EN-8669.

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

As the substance is not expected to possess antimicrobial activity no effects on microorganisms used for industrial food processing are expected.

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

Numerical MRLs for ketoprofen have been set by the Australian Government (cattle: meat: 50 µg/kg; milk: 50 µg/kg; edible offal: 50 µg/kg) and the Government of Canada (cattle: kidney: 450 µg/kg, muscle: 150 ppm, fat - not required, liver - not required; pigs: kidney: 400 µg/kg, muscle 100 µg/kg, fat - not required, liver - not required).

# 3. Risk management recommendations

Ketoprofen is currently included in Table 1 of the Annex to Commission Regulation (EU) No 37/2010 of 22 December 2009, with a 'No MRL required' status for bovine species, porcine species, and *Equidae*. The Committee, in its original MRL evaluations for bovine, porcine and *Equidae*, recommended a withdrawal period of 4 days for edible tissues to ensure adequate depletion of residues at the injection site.

Annex II, section I.2. of Commission Regulation (EU) 2018/782 states that "the consumer exposure to residues shall always remain at safe levels (below the ADI or alternative limit) in order for a 'No MRL required' classification to be recommended." Consumer exposure to ketoprofen residues was calculated to exceed the ADI at 4-12 hours post treatment in cattle and 3-12 hours post treatment in pigs. The ADI was also exceeded at 24-48 hours when the injection site was considered.

The available data indicates that a 4-day withdrawal period is likely sufficient for residues to deplete to safe levels, but this does not imply that no MRL is required. MRL status is not conditional on withdrawal periods. Rather, withdrawal periods are established on the basis of MRL values, as stated in Regulation (EC) No 470/2009 (recital 7). Further, without MRL values, there is not a legal framework within which to monitor compliance, prosecute illegal use, or ensure consumer safety if higher doses are used in the EU or in third countries.

Accordingly, numerical MRL values are now proposed. The proposals were derived using all available data, including the residue depletion studies submitted in response to the call for data.

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

Various non-steroidal anti-inflammatory drugs (NSAIDs) are available for food producing species in the EU.

**Feasibility of controls**

Without numerical MRL values, control of residues is not possible. Because monitoring has revealed residues of ketoprofen at levels that could lead to consumer exposure exceeding the ADI (see section 2.2.3 above), the establishment of numerical MRLs is considered appropriate.

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

- likelihood of misuse or illegal use:

According to information received from farmers and competent authorities of the Federal States in Germany, there is a strong suspicion of illegal ketoprofen use to deliver sick animals without pain symptoms for ante-mortem inspection at slaughterhouses. There is also information available that ketoprofen is used instead of antibiotics to circumvent the surveillance measures for antimicrobial substances. Both issues may result in increased residue concentrations in food commodities and a possible unacceptable risk for consumers.

Without numerical MRL values, prosecution/penalization of misuse is difficult under national law (e.g. German Law Section 10(3) of the LFGB) because the criteria for assessing compliance are lacking. Normally, substances with the status "No MRL required" are considered to pose no risk to consumer safety, regardless of the residue concentration measured in food. However, as discussed above, this is not the case for ketoprofen.

Numerical MRL values would enable assessment of compliance and thereby contribute to the protection of consumer safety.

## *3.2. Elaboration of MRLs*

Theoretical maximum daily intake (TMDI) of residues was calculated for cattle and pigs based on available data. The following two assumptions were made: (i) the upper 95% confidence interval of the mean was used, when possible, as the residue concentration to account for uncertainty in the estimated means, and (ii) half the LOQ was entered for values below the LOQ, and zero was entered for values below the LOD.

Additionally, two other assumptions were made about the marker ratio when calculating TMDI for pigs. First, the ratio for pig muscle could not be determined in the total residue study, so the ratio for skin/fat (i.e. 0.72) was used. Second, the total residue study produced a very low estimate for the liver marker ratio in pigs (0.3%); the ratio was adjusted to 2%, matching the lowest marker to total residues ratio established for liver in other species.

***Bovine***

Four studies in cattle provided the full set of residue data needed to calculate the TMDI. At 4 hours post treatment, the TMDI was 6.5 times higher than the ADI, although these data were from a single calf given an elevated dose. At 12 hours, the TMDI exceeded the ADI (4 times higher) in one study but not in a second study (56.5% of the ADI). By 24 hours, the TMDI was below the ADI in all studies, accounting for 15.5-50.6% of the ADI. With the injection site, though, intake at 24 hours was seen to still exceed the ADI (3.6 times the ADI). All TMDI estimates, including those with the injection site, were below the ADI by 2 days post treatment.

Thus, the available data suggest that TMDI may exceed the ADI until 24 hours post treatment and that considering injection site residues prolongs this by a day.

***Porcine***

Five studies in pigs reported the full set of residue data needed for calculating the TMDI. At 3 hours post treatment, the TMDI was 4.2 times higher than the ADI. At 8 to 12 hours, the TMDI exceeded the ADI in one study (53.8 times the ADI) but not in two other studies (55.9-74.8% of the ADI). By 24 hours, the TMDI was below the ADI in all studies, accounting for 11.7-32.8% of the ADI. With the injection

site, though, the TMDI exceeded the ADI at 24 hours in all studies that administered ketoprofen intramuscularly, with the TMDI being 2.5 to 18.7 times higher than the ADI. All TMDI estimates, including those with the injection site, were below the ADI by 3 days post treatment.

Thus, analogous to cattle, the available data suggest that the TMDI may exceed the ADI until 24 hours post treatment and that this is prolonged by 1-2 days when considering injection site residues.

***Equidae***

While no residue study was carried out in the horse, comparison of pharmacokinetic parameters suggests faster tissue depletion than in cattle.

Milk was not included in the TMDI calculations above. Ketoprofen and its metabolite RP 69400 could not be detected (<25 µg/l) in the milk at any milking both during and after treatment in a residue depletion study. There were detectable residues in milk (57 µg/l) when a single dairy cow was given an elevated dose (6 mg/kg) of ketoprofen shortly (2 hours) before milking. Two studies identified in the literature search also found very low or undetectable residue levels in milk under standard milking regimes. Given that (i) residues in milk were only slightly above the LOQ when the dose was doubled (to 6 mg/kg) and the milking interval shortened, and that (ii) residues were not detectable under standard dosing and milking regimes, an MRL value of 20 µg/kg is proposed for milk.

**Calculation of the theoretical daily intake of residues**

For edible tissues, TMDI estimates were below the ADI by 24 hours after treatment for both cattle and pigs. So, for each study, residue concentrations at this time point were used to derive potential MRL values following Regulation (EU) 2018/782. This yielded the following MRL values for cattle: 20, 20, 40, and 40 µg/kg for muscle, fat, liver, and kidney respectively, as well as 20 µg/kg for milk. The values for pigs were: 20, 20, 40, and 50 µg/kg for muscle, skin/fat, liver, and kidney respectively.

So, values were identical for muscle, fat, and liver, but not for kidney. The higher value for kidney (50 µg/kg) is selected as the proposed MRL because it is closer to the kidney MRLs yielded by most other studies, both in cattle and pigs, and this value remains safe. There are also reasons to reduce the liver MRL. First, like muscle and fat, liver residues were commonly below the LOQ at 24 hours, but in several studies, the LOQ was higher for liver (20 µg/kg) than for muscle and fat (10 µg/kg), with the result that the MRL based on twice the LOQ would be 40 µg/kg instead of 20 µg/kg, but this is a consequence of the LOQ rather than of actual residue levels. Second, a liver MRL of 40 µg/kg results in a consumer exposure estimate of 200 µg based on residues in liver alone, representing 66% of the ADI (300 µg/kg), which is disproportionate; in the total residue study in pigs, liver residues accounted for 28.5% of TMDI. Thus, the recommended MRL value for liver was reduced to 20 µg/kg, which, given the method described in section 2.2.4, is still applicable for monitoring purposes.

Finally, the available data suggested that injection site residues do not always deplete below the muscle MRL of 20 µg/kg by 4 days post treatment, which was the withdrawal period previously recommended by the CVMP for ketoprofen. When increased from 20 to 50 µg/kg, no data points exceeded the MRL at 4 days. Therefore, the muscle MRL was increased to 50 µg/kg, with the result that a 4-day withdrawal period will remain appropriate. This also eliminates the need to propose an injection site residues reference value (ISRRV).

So, taking into consideration all the available data, MRL values could be derived, and the theoretical daily intake of residues calculated as follows:

| Edible tissue or products | Daily consumption (kg) | MRL proposal µg/kg) | Ratio of the marker/total residue (bovine; porcine) | Amount per edible tissue or product (bovine; porcine) |
|---|---|---|---|---|
| Muscle | 0.30 | 50 | 0.56; 0.72[a] | 26.8; 20.8 |
| Fat or Skin/Fat | 0.05 | 20 | 0.35; 0.72 | 2.9; 1.4 |
| Liver | 0.10 | 20 | 0.02; 0.02[b] | 100.0; 100.0 |
| Kidney | 0.05 | 50 | 0.56; 0.31 | 4.5; 7.9 |
| Milk | 1.50 | 20 | 1; - | 30.0; 30.0 |
| | | | | |
| SUM | | | | 164.1; 160.1 |
| % of the ADI | | | | 54.7%; 53.4% |

[a]The ratio for muscle could not be established in the total residue study, so the value for skin/fat was used

[b]The ratio for liver from the total residue study (0.3%) resulted in extreme intake estimates, so it was raised to 2% to match that seen in other species

Based on the above figures, the maximum theoretical consumer intake represents 54.7% of the ADI (164 of 300 µg/person) for cattle and 53.4% of the ADI (160 of 300 µg/person) for swine. In both cases, a portion of the ADI is available for a possible future extension to eggs.

# 4. Considerations on possible extrapolation of MRLs

In line with Article 5 of Regulation (EC) No 470/2009, the CVMP considered the possibility of extrapolating its recommended MRLs to other food producing species and commodities. Taking into account the provisions laid down in Commission Regulation (EU) 2017/880, and current scientific knowledge, the recommendations on extrapolation are justified as follows:

| Animal species/ food commodities | Extrapolation possible (Yes/No) | Justification |
|---|---|---|
| All food producing species | No | Pharmacokinetic data were available in bovine, porcine, *Equidae* and poultry and residue data were only available in bovine, porcine and poultry. In the absence of supporting data demonstrating similarity of metabolism, and in line with Commission Regulation (EU) 2017/880, extrapolation to unrelated species is not recommended. |
| All ruminants | Yes | Existing data indicate that the pattern of metabolites seen across species is similar as identical MRLs have been recommended with regards to bovine and porcine. In line with the provisions of Commission Regulation (EU) 2017/880 extrapolation to all ruminant species is therefore recommended. |
| Horses (including milk) | Yes | While no residue study was carried out in the horse, comparison of pk parameters suggests faster tissue depletion than in cattle. Horses are a minor species, and in |

| | | |
|---|---|---|
| | | line with Commission Regulation (EU) 2017/880, extrapolation of the MRLs to *Equidae* can be accepted. |

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

Having considered that:

- the toxicological ADI of 0.005 mg/kg bw (i.e. 0.3 mg/person) was established as the overall ADI for ketoprofen,
- the previous CVMP conclusions on the absorption, distribution, metabolism and excretion of ketoprofen in bovine, *Equidae* and porcine species remains unchanged,
- the theoretical intake of residues from bovine, porcine and *Equidae* tissues represents less than 55% of the ADI,
- extrapolation to *Equidae* and all ruminants is appropriate,
- with a view to facilitating official residue control activities numerical MRLs are recommended,
- ketoprofen is retained as the marker residue,
- the ratios of marker to total residues for cattle were 0.56 in muscle, 0.35 in fat, 0.02 in liver and 0.56 in kidney and for pigs, 0.72 in fat plus skin, 0.02 in liver, and 0.31 in kidney,
- an appropriately validated analytical method for the determination of ketoprofen in muscle and milk is available and suitable for the official control of residues at the level of the proposed MRLs,

the Committee recommends the establishment of numerical maximum residue limits for ketoprofen in bovine and porcine. Furthermore, with reference to Article 5 of Regulation (EC) No. 470/2009 and in line with the criteria laid down in Commission Regulation (EU) 2017/880, the Committee agreed to that the proposed maximum residue limits could be extrapolated to all ruminants and *Equidae*. Therefore, the amendment of the entry for ketoprofen in table 1 of the Annex to Regulation (EU) No 37/2010 of 22 December 2009, is recommended as follows:

| Pharmaco-logically active substance | Marker residue | Animal species | MRLs | Target tissues | Other provisions | Therapeutic classification |
|---|---|---|---|---|---|---|
| Ketoprofen | Ketoprofen | All ruminants, porcine, *Equidae* | 50 µg/kg<br>20 µg/kg<br>20 µg/kg<br>50 µg/kg<br>20 µg/kg | Muscle<br>Fat<br>Liver<br>Kidney<br>Milk | For porcine species the fat MRL relates to 'skin and fat in natural proportions' | NO ENTRY |
| | Ketoprofen | Poultry | 10 µg/kg<br>30 µg/kg<br><br>10 µg/kg<br>10 µg/kg | Muscle<br>Skin and fat in natural proportion<br>Liver<br>Kidney | Not for use in animals from which eggs are produced for human consumption | NO ENTRY |

The theoretical intake of residues from bovine, porcine and *Equidae* tissues represents less than 55% of the ADI. A portion of the ADI is available for a possible future extension to eggs.

# 6. Background information on the procedure

| | |
|---|---|
| Submission of the request | 01 March 2023 |
| Steps taken for assessment of the substance | |
| Clock started: | 20 December 2023 |
| List of questions adopted: | 18 April 2024 |
| End of consultation of the call for data | 31 August 2024 |
| Clock restart: | 9 September 2024 |
| Opinion: | 7 November 2024 |