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Veterinary Medicines and Inspections

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COMMITTEE FOR MEDICINAL PRODUCTS FOR VETERINARY USE

EUROPEAN PUBLIC MRL ASSESSMENT REPORT (EPMAR)

DICLOFENAC (2)
Bovine milk

INTRODUCTION

On 3 July 2009 the European Commission adopted a Regulation[1] establishing maximum residue limits for diclofenac in bovine milk, 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.

Diclofenac is intended for use in bovine species for anti-inflammatory, analgesic and antipyretic therapy by intramuscular injection.

Diclofenac had maximum residue limits already established for tissues in bovine and porcine species.

FATRO S.p.A. submitted the application for the extension of maximum residue limits to the European Medicines Agency (EMEA), on 30 October 2008.

Based on the original and complementary data in the dossier, the Committee for Medicinal Products for Veterinary Use concluded on 11 February 2009 that the entry for diclofenac in Annex I of Council Regulation (EEC) 2377/90, should be extended to milk in accordance with Article 2 of the Regulation. Subsequently the Commission recommended on 5 June 2009 that maximum residue limits in bovine milk are established. This recommendation was confirmed on 14 June 2009 by the Standing Committee on Veterinary Medicinal Products and adopted by the European Commission on 3 July 2009.

EXPLANATORY NOTE

The European Public MRL Assessment Report is a public document giving the overview of the assessment carried out by the Committee for Medicinal Products for Veterinary Use (CVMP) of an application submitted for the establishment of maximum residue limits (MRLs). The document is based on the CVMP assessment report of the application from which confidential information has been deleted.

KEY WORDS: MRLs, diclofenac, bovine,

[1] Commission Regulation (EC) No 582/2009, O.J. L 175, of 04.07.2009

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SUMMARY OF THE SCIENTIFIC DISCUSSION FOR THE ESTABLISHMENT OF MRLs

Substance name: **Diclofenac**

Procedure number: **EU/08/164/FAT**

Applicant: **FATRO S.p.A.**

Target species: **Bovine**

Intended therapeutic indication: **Anti-inflammatory, analgesic and antipyretic therapy**

Route (s) of administration: **Intramuscular**

1. Introduction

Diclofenac sodium, sodium 2-[(2,6-dichlorophenyl)amino]phenylacetate (CAS No 15307-79-6), belongs to the non-steroidal anti-inflammatory drugs (NSAIDs) and, more specifically, to the phenyl acetic acid derivatives. Diclofenac sodium is intended for treatment in cattle and swine as an anti-inflammatory agent at doses of 2.5 mg/kg bw/day by intramuscular route. The proposed duration of treatment is 1 to 3 days.

Currently, diclofenac is included in Annex I of Council Regulation (EEC) No 2377/90 in accordance with the following table:

Pharmacologically active substance(s)	Marker residue	Target species	MRLs	Target tissues	Other provisions
Diclofenac	Diclofenac	Bovine	5 µg/kg 1 µg/kg 5 µg/kg 10 µg/kg	Muscle Fat Liver Kidney	Not for use in animals from which milk is produced for human consumption
		Porcine	5 µg/kg 1 µg/kg 5 µg/kg 10 µg/kg	Muscle Skin+fat Liver Kidney	

An application has now been submitted for the extension of the entry in Annex I of Council Regulation (EEC) No 2377/90 for diclofenac to include bovine milk. The proposed indication for dairy cattle is the same as before and the recommended doses have not changed.

2. Safety assessment

The CVMP has previously assessed the consumer safety of diclofenac and established an overall pharmacological-toxicological ADI of 0.5 µg/kg bw (30 µg per person). Therefore, no further assessment regarding the consumer safety of the substance is required for the purpose of this extension application.

The applicant supplied additional information addressing risk management consideration on the potential effects on dairy starter cultures. It was clearly demonstrated that diclofenac does not exert any inhibitory activity against any of the 16 commercial dairy starter cultures tested at concentrations of up to 256 µg/ml.

2.1 *Overview of evaluation by other international committees*

No MRLs for diclofenac have been established by JECFA/Codex Alimentarius.

3. Residues assessment

3.1 Pharmacokinetics in target species

In cattle ^{14}C-diclofenac sodium was administered by the intramuscular route, at a dose rate of 2.5 mg/kg per day for 3 consecutive days to 16 calves. Absorption of radioactivity into the systemic circulation was rapid, with the peak mean concentration (C_{max} equals 7009 µg equivalents/kg) observed at 2 hours following the first administration. Low levels of radioactivity (1252 µg equivalents/kg) remained in the plasma at 24 hours after treatment. Following administration of the final administration (dose 3), concentrations of radioactivity present in the plasma increased gradually, with highest concentrations (C_{max} equals 9006 µg equivalents/kg) observed at 2 hours after the last administration. By 48 hour after the last administration, mean concentrations of radioactivity had fallen to lowest levels of 411 µg equivalents/kg. The major route of elimination was via urine, which accounted for a mean of 61% and 80% in male and female cattle, respectively. Diclofenac, 4'-5-dihydroxy diclofenac, 5-hydroxy diclofenac 4'-hydroxy diclofenac and 3'-hydroxy diclofenac were observed in urine. Excretion in faeces accounted for a mean of 29% and 16% in male and female cattle, respectively. Diclofenac, 5-hydroxy diclofenac and 4'-hydroxy diclofenac were observed in faeces.

3.2 Residue depletion studies

Eight lactating cows were given 3 daily intramuscular doses of 2.5 mg ^{14}C-diclofenac sodium and milk was collected from each animal prior to dosing and at 12 hourly intervals thereafter until 216 hours post first dose (168 h post third dose withdrawal). All samples were analysed for total radioactive residues (TRR) by liquid scintillation counting. Diclofenac was quantified using a validated HPLC-MS/MS method. Maximum total radioactive residues and marker residue concentrations were observed 12 hours post last dose and were 42.5 µg equiv/kg and 4.66 µg diclofenac/kg respectively. The cumulative recovery of total radioactive dose did not exceed 0.04% of administered dose, indicating extremely low uptake into milk. At the last time point examined (168 hours post last dose) total residues were present at 0.002 µg equivalents/g. The ratios of marker residue to total residues were calculated for each time-point until 96 hours after third administration; at the cessation of treatment diclofenac was 11.6% of the total residues, and this declined gradually to 5.9% after 96 hours. At 84 and 96 hours post last dose, six of the eight analysed milk samples were below the limit of quantification (0.2 µg/kg) after administration.

It was demonstrated that, contrary to edible tissues from bovine and porcine, there was not a significant proportion of irreversibly bound residues in milk.

HPLC analysis of milk extracts revealed components with the same retention time as diclofenac, 3-hydroxy diclofenac, 4-hydroxy diclofenac, 5-hydroxy diclofenac and 3-hydroxy-4-methoxy diclofenac and 4,5-dihydroxy diclofenac. Furthermore LC-MS/MS investigation failed to identify the nature of metabolites. However, no single component was present at level greater than 8.6% of total radioactive residues.

In a non-radiolabelled study, diclofenac-sodium was administered for 3 consecutive days intramuscularly at a daily dose of 2.5 mg/kg b.w. (equivalent to 2.33 mg/kg bw of diclofenac) to 8 lactating cows. Milk samples were taken every 12 hours from cessation of dosing to 168 hours after last administration and analysed using a HPLC/MS/MS method. The highest concentrations of diclofenac were present in milk 12 hours after the last dose was administered (mean value 3.76 µg/kg). Residues depleted steadily, until 108 hours (9th milking) when diclofenac concentrations were below the limit of quantification (0.05 µg/kg) or limit of detection (0.03 µg/kg) in milk taken from all animals.

3.3 Selection of marker residue and target tissues

As for bovine and porcine tissues diclofenac was confirmed as the marker residue.

At the time when residue intake is below the allocated ADI from milk, the marker residue is not quantifiable and therefore the ratio marker residue to total residues cannot be established. As the ratio

is decreasing over time and in the last 4 time points the depletion curve is nearly flat, it was possible to extrapolate the ratio value expected at the time at which the residue intake is below the allocated ADI for milk (132 hours, i.e. 11 milkings) resulting in a ratio of marker to total residue of 0.05.

3.4 Elaboration of MRLs

The MRL for milk has been established as 0.1 µg/kg twice the limit of quantification (0.05 µg/kg) of the analytical method.

3.5 Calculation of theoretical daily intake of residues

Based on the MRL of 0.1 µg/kg, the ratio of 0.05 (at 11 milkings) and 1.5 litre milk consumption, the calculated residue intake from milk is 3 µg which is equivalent to 10% of the ADI.

Based on the MRLs for pigs (the species with the highest theoretical tissue residue intake) and milk consumption, the theoretical maximum daily intake was calculated to be 96% of the ADI.

3.6 Analytical method for monitoring of residues

An HPLC/MS/MS analytical method is available and validated according to the requirements of Volume 8 of The Rules Governing Medicinal Products in the European Union to quantify residues of diclofenac in bovine milk in the range 0.05 to 5.0 µg/kg. The limit of quantification is 0.05 µg/kg and the limit of detection is 0.03 µg/kg.

4. Conclusions and recommendation

Having considered that:

- an ADI of 0.5 µg/kg bw (i.e. 30 µg/person) was previously established for diclofenac,
- diclofenac did not exert an inhibitory activity against the commercial dairy starter cultures tested,
- diclofenac was retained as the marker residue in bovine milk,
- the average marker to total residues ratios in tissues 3 days after intramuscular administration in porcine and bovine tissues were 0.035 and 0.11 in liver and 0.12 and 0.20 in kidney, respectively; data were not available for muscle and fat or skin + fat and so a conservative ratio of 0.1 for these tissues was retained,
- the marker to total residues ratio in milk is 0.05 calculated at 11 milkings,
- 50% and 20% of the total residues in liver and kidney were irreversibly bound in pig tissues; 40% and 30% of the total residues in liver and kidney were irreversibly bound in bovine tissues; irreversible bound residues were not identified in milk,
- total radioactive residues in milk did not exceed 0.04% of administered dose,
- a validated routine analytical method for monitoring residues of diclofenac in bovine milk is available;

the Committee for Medicinal Products for Veterinary Use recommends the amendment of the current entry for diclofenac in Annex I of Council Regulation (EEC) No. 2377/90 in accordance with the following table:

Pharmacologically active substance(s)	Marker residue	Target species	MRLs	Target tissues	Other provisions
Diclofenac	Diclofenac	Bovine	5 µg/kg 1 µg/kg 5 µg/kg 10 µg/kg 0.1 µg/kg	Muscle Fat Liver Kidney Milk	
		Porcine	5 µg/kg 1 µg/kg 5 µg/kg 10 µg/kg	Muscle Skin + fat Liver Kidney	

Background information on the procedure

Submission of the dossier:	30 October 2008
Steps taken for assessment of the substance:	
Application validated:	13 November 2008
Clock started:	14 November 2008
CVMP opinion adopted:	11 February 2009