COMMITTEE FOR VETERINARY MEDICINAL PRODUCTS

CEFTIOFUR

SUMMARY REPORT (1)

1. Ceftiofur is a cephalosporin antibiotic which is administered to cattle (including lactating cattle) and swine for control of bacterial infections of the respiratory tract. The formulations include a powder which is re-constituted in sterile water prior to intramuscular injection.

2. Ceftiofur was poorly absorbed after oral administration and rapidly absorbed after intramuscular administration. In all species, ceftiofur was rapidly metabolised to (initially) desfuroylceftiofur (DFC) and furoic acid. Maximum blood concentrations (ceftiofur + metabolites) were achieved within 0.5 and 2 hours of dosing. Unmetabolised ceftiofur was generally undetectable in blood within 2 - 4 hours of dosing. Blood concentrations declined biphasically.

3. More than 95% of the administered dose was excreted within 24 hours of administration; 60 - 80% of this was in the urine and the remainder in the faeces. Most of the excreted material was desfuroylceftiofur and desfuroylceftiofur cysteine disulphide with small amounts of unmetabolised ceftiofur.

4. Ceftiofur was of low acute oral and parenteral toxicity. The acute oral LD_{50} was >7760 mg/kg bw in rats. The acute intravenous and intramuscular LD_{50}s in rats were 2156 and 1250 mg/kg bw respectively.

5. Repeat-dose studies of up to 90 days duration were carried out in various species. Gastrointestinal disturbances, reduced erythrocyte counts, reduced serum glucose levels and electrolyte imbalance were observed in a 90-day study in which rats were given up to 3000 mg/kg bw per day; compound-related histopathological findings were observed at the higher dose levels and reflected the poor nutritional status of these animals. The no-observed-adverse-effect level was 30 mg/kg bw per day. In the dog, repeated administration of doses of up to 3000 mg/kg bw per day of ceftiofur produced adverse effects on the haematopoietic system, including extramedullary haematopoiesis in the kidney, liver and spleen. The no-observed-adverse-effect level in dogs was 30 mg/kg bw per day.

6. In studies in which the target species were given many times the recommended dose, transient injection site reactions were the most notable findings.

7. Ceftiofur did not affect male or female fertility or reproductive performance in a 3-generation study in the rat in which doses of up to 1000 mg/kg bw per day were employed. Ceftiofur was not teratogenic in the rat or the mouse.

8. In an *in vitro* cytogenetics assay in CHO cells, ceftiofur induced a significant increase in the numbers of cells with structural aberrations, but only in the absence of metabolic activation. Other *in vitro* assays, including bacterial mutation assays and mammalian cell gene mutation assays on both ceftiofur and the furoic acid metabolite all gave negative results. An *in vitro*/*in vivo* UDS assay, an *in vivo* micronucleus test, and two *in vivo* chromosome aberration assays with ceftiofur also gave negative results. A radio-labelled study showed that ceftiofur or its metabolites reached the bone marrow following intraperitoneal injection, confirming the validity of the *in vivo* assays which used this tissue. Overall, it was concluded that ceftiofur was not mutagenic.

9. No data on carcinogenicity were presented. However the lack of structural alerts for ceftiofur, the absence of pre-neoplastic lesions in the repeat-dose studies and the evidence from the mutagenicity assays provide adequate reassurance that ceftiofur is not a potential carcinogen.

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10. Positive cutaneous anaphylaxis (PCA) tests in guinea pigs indicated that the desfuroylceftiofur metabolite had the greatest potential to elicit hypersensitivity reactions. However negative PCA responses were obtained in passively sensitised guinea pigs which were challenged with extracts containing 300 µg ceftiofur-related residues obtained from injection site muscle and kidney of bovines. In an *in vitro* radioallergosorbent test (RAST) inhibition assay, no IgE antibodies from the serum of 17 penicillin-allergic individuals were bound to ceftiofur, whereas one first generation cephalosporin did partially inhibit the binding of penicillin antibody from one patient.

11. A toxicological ADI of 0 - 300 µg/kg bw per day was calculated by applying a "safety factor" of 100 to the NOEL of 30 mg/kg bw per day in the repeat-dose toxicity studies in rats and dogs.

12. Data on the activity of ceftiofur and two of its metabolites towards bacterial strains of human origin were provided. A microbiological ADI was calculated, based on the following assumptions:

    - 2.0 µg/ml is the modal MIC_{50} for the most sensitive strains of human gut flora;
    - a factor of 2 to account for inoculum density effects (giving a no-microbiological-effect level of 4 µg/ml);
    - 150 ml for the human daily faecal bolus;
    - a factor of 0.05 for the assessment of the remaining microbiological activity in the human intestine, based on >95% degradation capacity of the human faeces for ceftiofur;
    - a factor of 10 to cover possible variations in human degradation capacity;
    - 60 kg for the human bodyweight.

    $$\text{Microbiological ADI} = \frac{4 \times 150}{0.05 \times 10 \times 60}$$

    = 20 µg/kg bw per day or 1.2 mg/person/day.

    This microbiological ADI was adopted as the ADI for ceftiofur.

13. Ceftiofur and its metabolites were rapidly depleted from tissues of the target species. Residues of unmetabolised ceftiofur were generally undetectable. Residues depleted slowest from the kidney in which the major "free" metabolite was desfuroylceftiofur cysteine disulphide (DCD). In cows' milk the principal free metabolite was also DCD.

14. Residues of ceftiofur in milk and bovine and porcine tissues can be monitored using an HPLC analytical method in which both free and conjugated residues containing the β-lactam structure are converted to desfuroylceftiofur and stabilised to the acetamide derivative. It was noted that the method was not validated with respect to bovine and porcine liver and fat tissues.

15. In most studies in cattle, the highest residues were found initially in the kidney. However residue depletion from the liver was slow and the liver is one of the "target" tissues in some studies from around day 20 after the cessation of treatment. In swine, the highest residues were consistently found in the kidney. Taking into account the residues depletion profiles the following MRLs for the sum of all residues retaining the beta-lactam structure, expressed as desfuroyceftiofur, were elaborated:

Species	MRL	Target tissue
Bovine	2000 µg/kg	kidney
	2000 µg/kg	liver
	200 µg/kg	muscle
	600 µg/kg	fat
	100 µg/kg	milk
Porcine	4000 µg/kg	kidney
	3000 µg/kg	liver
	500 µg/kg	muscle
	600 µg/kg	fat

Using these MRL values, the theoretical maximum daily intake of ceftiofur, desfuroyl ceftiofur and desfuroyl ceftiofur cysteine disulphide residues would be lower than the ADI adopted above.

Ceftiofur is administered dissolved in water and caused very little tissue irritation. Residues were not persistent at the injection site and depleted to below the above MRLs proposed for muscle with the adoption of a realistic withdrawal period. Residue concentrations at the injection site of 600 µg/kg were found in a study in cattle, 7.5 hours after treatment. In a study in pigs, 190 $\pm$ 110 µg/kg were found at the injection site 12 hours after treatment.

Unmetabolised ceftiofur was generally undetectable in milk. All milk samples from cattle treated according to the recommended dosing schedule gave negative results in a number of screening assays including the Delvotest -P. There were no effects on cheese, yoghurt or buttermilk starter cultures. In a study in which pigs were treated according to the recommended dosing schedule and slaughtered 22 hours after the last dose, all muscle and kidney samples gave negative results in the EEC Four-Plate test.

16. Until the company sufficiently justifies why the same MRLs could not be proposed for bovine and porcine species and until data are available concerning the validation of the HPLC analytical methods to be used to control these MRLs, in accordance with Volume VI of the "Rules governing medicinal products in the European Community", only provisional MRLs can be established. This data shall be provided before 1 July 1996. The provisional MRLs will expire on 1 July 1997.