SCIENTIFIC DISCUSSION

This module reflects the scientific discussion for the assessment of Naxcel and was last updated in October 2009. For information on all changes to the marketing authorisation, please refer to module 8.

1. SUMMARY OF THE DOSSIER

Naxcel is a suspension for injection containing ceftiofur as the active substance in the form of the free acid. The product is intended for use in pigs for the treatment of bacterial respiratory disease associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis* and for the treatment of septicaemia, polyarthritis and polyserositis associated with *Streptococcus suis* infection. In cattle, it is indicated for the treatment of acute interdigital necrobacillosis (also known as *Panaritium* or foot rot).

Ceftiofur, the active substance of Naxcel is a third generation cephalosporin, which is active against Gram-positive and Gram-negative pathogens. Ceftiofur inhibits bacterial cell wall synthesis, thereby exerting bactericidal properties.

The most common side effects noted during the safety and efficacy studies in pigs and cattle are transient local swellings and other mild reactions at the injection site.

The product is intended for single use. The dose in pigs is 5 mg/kg body weight via intramuscular injection and in cattle 6.6 mg ceftiofur/kg body weight administered subcutaneously at the base of the ear.

Ceftiofur is included in Annex I of Council Regulation (EEC) 2377/99, as amended.

The withdrawal period for pigs is 71 days for meat and offal.

In cattle, the withdrawal period for meat and offal is 9 days and for milk zero days. It is essential that Naxcel is only administered subcutaneously at the base of ear location in non-edible tissue in order to comply with the cattle meat withdrawal period.

2. QUALITY ASSESSMENT

COMPOSITION OF THE VETERINARY MEDICINAL PRODUCT

Naxcel is a suspension for injection and contains 100 mg/ml (pigs) and 200 mg/ml (cattle) of ceftiofur (as ceftiofur crystalline free acid micronized) as the active substance, in an oily vehicle carrier. Nitrogen (Ph. Eur.) is used to purge the vial headspace during filling.

CONTAINER

The suspension for injection is presented in a colourless Type I glass vial of 100 ml, with a chlorobutyl-isoprene rubber stopper and an aluminium overseal, with plastic flip-off cap.

Fragmentation and self-sealing testing following the maximum number of punctures per closure, according to the Ph.Eur., were conducted on 3 lots of stoppers and all these lots met their specifications. In a broached vial study it was shown that no microbial contamination of the vials occurred during the 28 day in-use shelf-life period.

The vials are tested for appearance, cleanliness, damage, foreign material, dimensions and confirmation that the glass conforms to the Ph. Eur. type specified.

The closures are tested for appearance, cleanliness, damage, foreign material, dimensions and identification of the elastomeric composition (ash or specific gravity may be used for the identification test).

The specifications and routine tests for the aluminium overseal were provided along with certificates of analysis.

CLINICAL TRIAL FORMULA

The composition of the batches used in the clinical studies is identical to the formulation proposed for the market place, except for the quantity of overage. This should have no impact on the efficacy of the final product.

DEVELOPMENT PHARMACEUTICS

The development pharmaceutics of this suspension for injection are well described. A long-acting single dose formulation was developed using the sustained release activity of ceftiofur crystalline free acid suspension (compared to other ceftiofur products). The concentration of the active is adjusted according to the potency of the drug substance.

The principal factor contributing to the sustained release activity is the "maturation" of the oily suspension formulation. "Maturation" is defined as controlled induction of a progressively slower drug release rate via controlled manipulation of different variables during the manufacture of the product. The main objective of the formulation development was to control the extent of maturation during manufacture into a defined timeframe. Drug release characteristics of the suspension were correlated to the *in vivo* performance of the product in swine. Information on the physical attributes of the drug on the maturation process was provided.

No preservative is added to the formulation as this product is oil based and lacks sufficient water to promote microbial growth.

METHOD OF MANUFACTURE

The product is manufactured according to standard processes used to manufacture sterile oily suspensions for injection.

The manufacturing formula for the proposed batch size was presented. The product is manufactured in three steps: vehicle preparation, drug addition and heating, filling and terminal sterilisation. Detailed descriptions of the method of manufacture, including terminal sterilisation and in-process controls were provided.

In-process controls monitored for the product include drug substance weight, identity and weight of components, temperature during drug addition, suspension recirculation time through blender, nitrogen purge flow rate, suspension temperature during heating phase, temperature at the end of manufacture, density of the sample prior to filling, vial fill weight and yield reconciliation.

Satisfactory process validation data and the validation protocol were presented.

CONTROL OF STARTING MATERIALS

Active substance

The active substance is ceftiofur crystalline free acid micronised, a white to tan crystalline powder composed of generally spherical aggregates of small birefringent crystallites. The active substance is not detailed in any pharmacopoeia. However, full specifications were provided in a monograph such as purity, appearance, water content, impurities, endotoxins content, residual solvents, identity, specific rotation and particle size.

All steps of the manufacture are well described. Starting materials are either synthesised or purchased and full details have been provided. Ceftiofur crystalline free acid results from neutralisation of the hydrochloride salt with a polyvinylpyridine resin followed by precipitation. The crystalline free acid is then micronised with suitable particle size reduction equipment. Structural characterisation (elemental analysis, MS, IR, UV, NMR, X-Ray diffraction spectroscopy) is provided along with a detailed physico-chemical characterisation.

The active substance was subjected to various stress conditions, including (solid state) thermal and photochemical degradation, (solution state) oxidation, acid and base degradation, in order to qualitatively determine which impurities increased under these severe conditions. The conditions of this study were severe and cases resulted in extensive degradation. Real life samples are unlikely to suffer from degradation as observed in these severe treatments. Inorganic impurities, volatile impurities and organic impurities were assessed and they are in compliance with the limits in the specifications for the active substance.

Batch data for several pilot and full scale lots of the active substance were provided and they were in compliance with the proposed specifications.

The long term stability data (60 months) support a 24 month reassay interval when stored in a freezer. Based on the photostability studies, the drug substance should be protected from light. The proposed specifications are the same for time of release and at reassay for all parameters. The stability test on the active substances is in accordance with the current guidelines on active substance stability studies.

The active substance is packaged within 2 sealed, LDPE bags (the first bag which contains the active substance is sealed and put into the second bag) in a plastic drum. The following characteristics are checked for the bags: appearance, cleanliness, damage, foreign material, dimensions, and identification by IR of the polyethylene.

Excipients

The excipients used in the product are: Miglyol (Triglycerides, medium chain, Ph. Eur.), Nitrogen, Ph. Eur., Cottonseed oil, NF all of which comply with the current monograph of the relevant pharmacopoeia.

Materials of animal origin

No materials of animal origin are contained or used in the manufacture of the product apart from milk powder, which is used in the manufacture of the active substance. A TSE declaration concerning the milk powder was provided and the CVMP agreed that the product would comply with the current Note for Guidance on Minimising the Risk of Transmitting Animal Spongiform Encephalopathy Agent via Human and Veterinary Medicinal Products (EMEA/410/01-Rev.2).

Control tests during production

The non-sterilised product is considered as an isolated intermediate. The isolated intermediate is tested for microbial count using a validated method. All other tests are performed on the product after its terminal sterilisation.

CONTROL TESTS ON THE FINISHED PRODUCT

The specifications and details of routine tests on the finished product includes parameters appropriate for this type of product such as appearance, identity, assay, impurities, content uniformity, drug release, volume of injection, resuspendability, sterility and endotoxins content. All tests were suitably validated.

The proposed limits for drug release test and for impurities assay (particularly for gradient impurities) are acceptable at the end of shelf-life, but these specifications were tightened at release.

The rationale for determining the new impurity limits at release in the drug product takes into account impurity limits in the drug substance, the potential for increases in impurities due to the drug product manufacturing process and subsequent terminal sterilisation and assay variability.

STABILITY

The finished product was stored under long term and accelerated storage conditions of 25°C/60% RH (24 or 30 months, upright and inverted), 30°C/60% RH (24 months inverted) and 40°C/75% RH (12 months inverted). Five supplementary stability studies were conducted: freeze / thaw; syringeability; photostability; and in-use studies during shelf life and at the end of shelf life.

Results for potency, resuspendability, impurities, sterility and appearance were within the specifications for all lots and at all intervals after 24 months of storage at 25°C/60% RH, 30°C/60% RH and 40°C/75% RH. In the freeze / thaw study, no significant differences were seen. The results of the syringeability study are comparable to similar use products with similar viscosities, showing that the syringeability of the product is acceptable.

In the in-use study (within shelf-life), the broached vials show very little change over a 3-month period and there is no significant change in either potency or impurities content. For drug release, broached vials remain within the acceptable range for up to 3 months after broaching. All broached vials meet the test for appearance and resuspended within 30 seconds throughout the study period. Broached vials also meet the test for sterility at the 3-month time point.

In the in-use study (at the end of shelf-life) of the 100 mg/ml pig formulation, all broached vials meet the test for appearance and resuspended within 30 seconds throughout the study period. Broached vials meet the test for sterility at the 6-week time point. Intact vials show very little change over the 24 to 30-month period. The broached vials behave similarly and there is no clear trend for either potency or gradient impurities as compared to the control sample. For the 200 mg/ml cattle formulation, results for two batches were provided and the applicant commited to provide a further in-use stability study close to the end of the shelf-life.

Regarding photostability, all exposed and wrapped vials meet the test criteria for appearance and resuspended within 30 seconds. The data obtained are consistent with the stability data and are not likely to be biologically significant.

The stability data demonstrate that the product, in the proposed market package, maintains acceptable quality for at least 30 months when stored at 25°C/60%RH and the claimed shelf-life of 2 years is therefore justified. All proposed specifications were met, except drug release in one lot. The overage amount of 5% in the stability lots, that was added to account for observed potency drops with terminal sterilisation, was reduced to 3%.

Based on the present primary stability lot data, a 24 month expiration date is recommended for the product when stored at 25°C/60%RH. The product does not need to be protected from light when in the proposed market package.

The stability studies are in accordance with the relevant current guidelines. The shelf-life (2 years) and the storage conditions (do not store above 25 °C and keep the product in the proposed market package) proposed are reflected in the SPC and product literature.

According to the note for guidance "Maximum shelf-life for sterile medicinal products after first opening or following reconstitution" (EMEA/CVMP/198/99), the in-use shelf-life of sterile products should be limited to 28 days. The in-use shelf-life for this product was therefore restricted to 28 days at the request of the Committee.

OVERALL CONCLUSION ON QUALITY

The quality data provided are satisfactory and adhere to current guidelines. Details of the manufacturing process are provided and show that product of the desired quality is consistently produced. Detailed descriptions of the method of manufacture, including terminal sterilisation, and in-process controls are presented. The production and control of starting materials of animal origin are in accordance with the recommendations of the EU note for guidance on minimising the risk of transmitting animal spongiform encephalopathy agents via human and veterinary medicinal products (EMEA/410/01-Rev.2).

The stability studies support a shelf-life of two years for the finished product when stored at 25°C/60%RH. The product does not need to be protected from light in the proposed market package. An in-use shelf-life of 28 days for the broached vials was agreed by the Committee.

3. SAFETY AND RESIDUE ASSESSMENT

SAFETY ASSESSMENT

PHARMACOKINETICS

See part 4.

TOXICOLOGICAL STUDIES

Single dose toxicity

Ceftiofur is of low acute oral and parenteral toxicity and is well tolerated in laboratory rodents following oral, intramuscular, intravenous or intraperitoneal administration. The acute oral LD_{50} is much higher than the acute intravenous LD_{50} indicating its poor absorption from the gastrointestinal tract. The LD_{50} for ceftiofur sodium and ceftiofur hydrochloride following intraperitoneal injection in rats was 927 and 881 mg/kg bw/day, respectively.

Repeated dose toxicity

Six repeated dose toxicity studies of up to 90 days duration were carried out in rats and dogs. The NOEL following oral administration of ceftiofur was 30 mg/kg bodyweight per day in both species. Results from these toxicity studies indicated that the gastrointestinal tract was the target organ. In the dog, the haematopoietic system was also affected.

Reproductive toxicity

In mice, no evidence of teratogenicity or foetotoxicity was observed following oral administration of ceftiofur at up to 4000 mg/kg bw/day from days 6 to 15 of gestation.

In rats, ceftiofur administered orally did not affect male or female fertility, growth or survival of their offspring in a 2-generation study. However, a rat teratogenicity study revealed dose-related maternotoxic effects and reduced foetal weights, and a warning has, therefore, been included in section 5.6 of the SPC (Use during pregnancy and lactation) to use the product only following the benefit/risk assessment by the responsible veterinarian.

Mutagenicity

In one *in vitro* cytogenetics assay in Chinese hamster ovary (CHO) cells, ceftiofur increased significantly the number of cells with chromosomal aberrations, but only in the absence of metabolic activation. However, other *in vitro* assays and *in vivo* mutagenicity tests gave negative results on both ceftiofur and its furoic acid metabolite.

The CVMP concluded that ceftiofur was not mutagenic when taking into account the possibility of exposure of the gastrointestinal tract of the consumer, or the target animal, by unmetabolised ceftiofur.

Carcinogenicity

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

Studies of other effects

Ceftiofur is well tolerated in laboratory animals after intraperitoneal administration.

The excipients and impurities present in the final product do not seem to present any particular risk for either animals or humans.

Inhalation of ceftiofur at high doses did not lead to any respiratory or systemic effects in rats. However, because of the possibility of cephalosporin allergic reactions, it cannot be concluded that small amounts of dust might provoke a reaction either acutely or over time.

USER SAFETY

Ceftiofur is developed for veterinary use only. “Users” are considered to be veterinary surgeons and farmers. The potential user exposure will depend on the number of animals and on the weight of the pigs and cattle being treated.

Accidental self-injection:

The risk encountered from a single accidental self-injection is considered low while repeated injections may present some risk to sensitive individuals. However, the CVMP considered accidental self-injection of considerable amounts of the product as very unlikely.

Skin and eye irritation

In studies in guinea pigs and rabbits, ceftiofur proved to be non-irritant to intact skin, and slightly irritating to abraded skin. Following repeated topical challenge, the substance showed a potential for mild skin sensitisation. Minimal irritation of rabbit eyes was observed after administration to the conjunctival sac.

Results indicated that irritation of abraded skin and mild delayed-type dermal sensitisation may occur in the user when product is (repeatedly) spilled onto the (abraded) skin.

Immunotoxicity, hypersensitivity

Hypersensitivity reactions have been observed among workers who have repeated exposure to ceftiofur. Positive cutaneous anaphylaxis tests in guinea pigs indicated that a metabolite might have the potential to elicit hypersensitivity reactions. This was, however, not confirmed in tests in passively sensitised guinea pigs. Also, i*n vitro* tests on the serum of penicillin-allergic human individuals suggested that residues of ceftiofur were unlikely to present a significant risk to penicillin-allergic persons.

Cross-reactions cannot be ruled out between different beta-lactam drugs with structural similarities and it is considered that ceftiofur, like other cephalosporins and penicillins, might cause hypersensitivity or allergic reactions following injection, inhalation, ingestion or skin contact. Therefore, persons with a known hypersensitivity should not handle the product. Persons developing symptoms following exposure should seek medical advice or, in the case of serious symptoms, urgent medical attention.

As occasional skin or eye contact may occur while handling the product, relevant warnings regarding user safety with respect to hypersensitivity, skin sensitisation and eye irritation have been included in the product literature.

IMPACT OF RESISTANCE-DEVELOPMENT FOR HUMANS

The resistance of target, commensal and food-borne pathogens against ceftiofur and its main metabolite was documented in line with requirements according to VICH GL27.

Ceftiofur has been registered for use in food-producing animals in the EU since 1989. The applicant provided MIC data of ceftiofur for relevant food-borne pathogens (*E. coli* and *Salmonella*) isolated 1999-2001 from colon content of healthy pigs at slaughter, which showed no resistance.

Ceftiofur has a poor or no activity against species of *Enterococcus* and *Pseudomonas* and is considered inactive against these organisms. Ceftiofur and other 3rd generation cephalosporins have poor activity against *Campylobacter*. Ceftiofur is active against *Escherichia coli* and *Salmonella*. *E. coli* and salmonella species are, therefore, still the relevant foodborne organisms for the extension for use in cattle.

Among *Salmonella* and *Escherichia coli*, resistance due to acquired β-lactamases is by far the most prevalent resistance mechanism. Acquired resistance to 3rd generation cephalosporins may be mediated by genes encoded on chromosomal or extrachromosomal genetic elements, although plasmid encoded β-lactamases are more commonly reported. With few exceptions, *Escherichia coli* and *Salmonella* that have acquired resistance to 3rd generation cephalosporins are resistant to other drug classes, including phenicols, aminoglycosides sulfonamides and tetracyclines. There have been some reports in Europe of *E. coli* resistance in cattle; however, the numbers of isolates are too low to draw any meaningful conclusions on resistance patterns and epidemiological factors. Two cases in cows were noted in Spain and two in France. Beyond these sporadic cases, cephalosporin-resistant *E. coli* isolates were reported by Liebana et al. in scouring calves in 2006. The reported prevalence rates of *salmonella* in beef meat at slaughter and retail are also very low in Europe.

The potential for exposure of enteric organisms in the animal environment to microbiologically active drug was assessed. It was considered transient and low because microbiologically active ceftiofur residues in the urine and in urine plus faeces are readily inactivated. This inactivation of ceftiofur also occurred in the presence of human faecal slurries. Ceftiofur is rapidly inactivated in aerobic soil systems and mineralized. Ceftiofur is also degraded by photolytic and hydrolytic processes.

Data in different species show that faecal excretion is a minor pathway. Only a very small percentage (0.08-6%) of the total residue in the faeces is estimated to be microbiologically active and it is rapidly inactivated in manure.

Taking also into account that the product is likely to be used in individual animals rather than for herd treatment, the route of administration of the product and the low excretion via faeces, the Committee concluded that the level of exposure of human gut flora to ceftiofur could be classified as "low potential exposure".

However, during the assessment of the cattle extension, the CVMP acknowledged the concerns and recommendations summarised in the CVMP "Reflection Paper on the use of 3rd and 4th generation cephalosporins in food-producing animals in the European Union: Development of resistance and impact on human and animal health" (EMEA/CVMP/SAGAM/81730/2006/Rev.1) and comprehensive prudent use warnings have been included in section 4.5 of the SPC.

ENVIRONMENTAL RISK ASSESSMENT

A Phase I assessment was performed in order to assess if the use of the product could have potential harmful effects on the environment, and to identify any precautionary measures which may be appropriate to reduce any harmful effects.

In pigs, Predicted Environmental Concentration estimates in soil (PEC_{soil}) were provided for piglets and adult pigs encompassing a range of sizes of animals, and to make the assessment with the largest number of animals potentially raised per year per housing location.

All metabolites were incorporated in total for the PEC_{soil} calculations, assuming that 100% of the excreted ^{14}C-labeled material is unchanged ceftiofur. Taking into account that the amount of ^{14}C-ceftiofur and residues excreted from piglets was 88% after post-injection time of 10 days, values of 88% were used in the calculation of the PEC_{soil}. It was also assumed that 50% of the swine herd is treated with Naxcel and that the proportion of the number of animal cycles receiving ceftiofur treatment will be 100%. The amount of ceftiofur excreted is assumed to be the total of that found in urine plus faeces, i.e. 88% of the initial dose. The rate of manure application to soils is assumed to be 170 kg manure N/ha/year. The infiltration depth of ceftiofur residues was 5 cm for grassland soils and 20 cm for ploughed arable soils.

The PEC_{soil} values were at least 80-fold lower than the Phase I trigger value for soil of 100 µg/kg, and therefore no Phase II assessment was considered necessary.

For cattle, a total residue approach was taken in the Phase I assessment, wherein it was assumed that the total amount of the dose applied is excreted from the animal and data on metabolism, excretion and biodegradation are not taken into account. Additionally, it is assumed that 50% of the herd is treated for foot rot. In practice, this rate of treatment is expected to be considerably lower than the incidence of bovine respiratory disease. Thus, the Phase I PEC_{soil} calculations presented are worst-case.

Calculated PEC_{soil} values were provided for different types (calf, dairy cows, beef cattle) and age groups ("0-1 years" and "more than 2 years") under different animal husbandry conditions (intensively reared, pasture). Results showed PEC_{soil} values between 9.2 (dairy cow, pasture) to 19.2 µg/kg (intensively reared animals, more than 2 years).

The Committee agreed that the PEC_{soil} values for all of the target animals are less than the Phase I trigger value of 100 µg/kg soil and that based on these calculations, no further environmental assessment was considered necessary for Naxcel 200 mg/ml Suspension for Injection for Cattle, when used as recommended. Additionally, there are no other factors known for ceftiofur that would require further investigation in a Phase II assessment.

The CVMP concluded that ceftiofur should have no impact on the environment when used in the form of Naxcel by the recommended dosing instructions.

RESIDUE ASSESSMENT

DEPLETION OF RESIDUES

In pigs, three depletion studies were provided, each following a single intramuscular injection of Naxcel at the recommended dosage of 5 mg ceftiofur/kg bw in pigs. The samples were analysed using a validated HPLC-DCA method with a limit of quantification (LOQ) of 100 µg/kg.

In the pivotal residue depletion study, samples of muscle, liver, kidney, fat, injection site and skin/fat were collected up to 70 days post injection. The ceftiofur and desfuroylceftiofur-related injection site tissue residues had depleted to less than 500 µg CE/kg by 70 days after treatment. By 14 days after treatment, all tissue residues except injection site depleted to a concentration below the LOQ (100 µg/kg) of the assay. In the injection site, all marker residue levels were below the muscle MRL at the last sampling time (i.e. 70 days following the injection).

In a further study, samples of muscle, liver, kidney, fat, injection site and skin/fat were collected up to 42 days post injection. The ceftiofur and desfuroylceftiofur-related residue depleted to concentrations below the LOQ in all tissues, except injection site, by 14 days post-treatment. In the injection site, marker residue levels were above the limit of detection (LOD) of the analytical method (50 µg/kg) and at the last sampling time (i.e. 42 days following the injection), the marker residue levels were above the muscle MRL.

In a third study, samples of kidney and injection site were collected up to 12 days post-injection. In the injection sites, the residue concentrations were highly variable at all time points, decreasing from a mean of 28300 ± 19900 µg/kg at 12 days after treatment. Kidney residues were much lower with a mean residue concentration below the LOQ of the analytical method 12 days after treatment. In the injection site, the marker residue levels were above muscle MRL in all samples at the last sampling time (i.e. 12 days following the injection).

In cattle, a number of residue depletion studies were provided, each following a single subcutaneously injection of Naxcel at the recommended dosage of 6.6 mg ceftiofur/kg bw.

In the pivotal GLP-compliant **tissue** residue depletion study, the recommended dosage of 6.6 mg ceftiofur/kg bw was subcutaneously injected at the recommended site of injection (base of the ear). The product used in this study is the final formulation of CCFA-SS with a mean *in vitro* release rate of 72%. The study was undertaken in adult animals (highest bodyweight) using the highest recommended volume (30 ml). Samples of muscle, liver, kidney, fat) were investigated up to 7 days post injection. and analysed using a new validated LC/MS-DCA method with a limit of quantification (LOQ) of 100 µg/kg. At day 7, all samples are below the MRL and LOQ.

Three **milk** depletion studies of ceftiofur were performed in cattle following single subcutaneous administration of CCFA-SS at the base of the ear or in the middle of the ear at the recommended dose (i.e. 6.6 mg ceftiofur/kg bw). The product used in these studies was the final formulation of CCFA-SS. The mean *in vitro* release rates of the test formulations was about 90%. Samples were taken twice daily (every 12 hours) for 7 days post treatment. All data at all time point for all three studies with animals dosed at the base of the ear were below the MRL.

MRL

MRLs for Ceftiofur have been included in Annex I of Council Regulation (EEC) No. 2377/90 as follows:

Pharmacologically active substance(s)	Marker residue	Animal species	MRLs	Target tissues	Other provisions
Ceftiofur	Sum of all residues retaining the betalactam structure expressed as desfuroylceftiofur	Porcine	1000 µg/kg 2000 µg/kg 2000 µg/kg 6000 µg/kg	Muscle Fat Liver Kidney	

All other constituents of Naxcel are included in Annex II of Council Regulation (EEC) No 2377/90 or are considered as not falling within the scope of the Regulation.

WITHDRAWAL PERIOD

The pivotal study for the determination of the withdrawal period used Naxcel at the lower limit of the release rate. The residue concentrations in injection site muscle from all five slaughter points were used for the calculation of a withdrawal period using the statistical approach proposed by the Note of Guidance 'Approach towards Harmonisation of Withdrawal Periods' (EMEA/CVMP/036/095), and a withdrawal period of 71 days for "meat and offal" in pigs was established for Naxcel.

In the pivotal residue depletion study in cattle, at day 7 all samples are below the limit of quantification of the analytical method. The proposed withdrawal period of 9 days for **meat and offal** was adopted, this withdrawal period includes a 28% safety span to account for study deficiencies. According to the residue levels observed in the three depletion studies, a withdrawal period for **milk** of zero days was established.

ANALYTICAL METHOD

In support of the pig application, the applicant provided a description of the regulatory analytical method, which was considered by the CVMP during the MRL application. For the cattle presentation, two analytical methods were provided. The regulatory analytical method (HPLC-DCA, tissue and milk depletion) considered during the MRL application and a new method (LC-MS/MS-DCA tissue depletion).

OVERALL CONCLUSION ON SAFETY AND RESIDUES

Ceftiofur is of low acute oral and parenteral toxicity. Results from repeated dose toxicity studies indicated that the gastrointestinal tract is the target organ.

Reproductive toxicity studies, including teratogenicity, indicated that ceftiofur was not reprotoxic or teratogenic in mice. However, a rat teratogenicity study revealed maternotoxic and foetotoxic effects and the SPC, therefore, includes a warning that the product should be used in pregnant or lactating sows only following the benefit/risk assessment by the responsible veterinarian.

A battery of mutagenicity tests indicated that ceftiofur sodium and its metabolite, furoic acid, were non-mutagenic. Ceftiofur is capable of inducing chromosomal aberrations *in vitro* at cytostatic high doses but not *in vivo* at achievable dose levels. Therefore, an initial *in vitro* observation of the induction of aberrations in CHO cells was considered not relevant for the risk assessment, and the CVMP concluded that ceftiofur was not mutagenic. The CVMP also agreed that ceftiofur is not a potential carcinogen.

Ceftiofur is slightly irritant to abraded skin and minimal irritating on rabbit eyes. Like other cephalosporins, ceftiofur has a potential to cause hypersensitivity or allergic reactions following injection, inhalation, ingestion or skin contact. Under field conditions, while handling the product, accidental skin or eye contact may occur. Adequate precautionary warnings are included in the product literature and these are in line with other comparable authorised products.

Resistance emergence has been low or nil among major food-borne bacteria. Taking also into account the product posology and the low excretion via faeces, the Committee concluded that the level of exposure of human gut flora to ceftiofur could be classified as "low potential exposure". However, animals can serve as reservoirs of resistance determinants, including multiresistant organisms that acquire Expanded spectrum β-lactamases ESBLs or AmpC cephalosporinases. During the assessment of the cattle extension, the CVMP acknowledged the concerns and recommendations summarised in the CVMP "Reflection Paper on the use of 3rd and 4th generation cephalosporins in food-producing animals in the European Union: Development of resistance and impact on human and animal health" (EMEA/CVMP/SAGAM/81730/2006/Rev.1) and agreed to strengthen the prudent use warnings, in accordance with the recommendations of the reflection paper.

The predictions of environmental exposure (PEC) support that ceftiofur should have no impact on the environment when used as recommended for the product. Since Naxcel is below the Phase I trigger value of 100 µg ceftiofur /kg soil, no Phase II assessment was required.

In pigs, three depletion studies were provided following intramuscular injection of Naxcel at the recommended dosage (5 mg ceftiofur/kg bw) in swine confirming that 70 days following the injection, all marker residue levels were below the muscle MRL in the injection site. The pivotal study for the determination of the withdrawal period used Naxcel at the lower limit of the release rate. MRLs for Ceftiofur have been included in Annex I of Council Regulation (EEC) No. 2377/90.The residue concentrations in injection site muscle from all five slaughter points were used for the calculation of a withdrawal period using the statistical approach proposed by the Note of Guidance 'Approach towards Harmonisation of Withdrawal Periods' (EMEA/CVMP/036/095), and a withdrawal period of 71 days for "meat and offal" in pigs.

In cattle, one pivotal tissue depletion study and three milk depletion studies were provided following the recommended route of administration and dose confirming that 7 days following the injection, all marker residue levels were below the MRL in all tissues. For milk, marker residue levels were below the MRL at all time points. The CVMP concluded on a withdrawal period of 9 days for meat and offal and zero days for milk, provided that animals are treated in the recommended way (at the base of the ear).

4. EFFICACY ASSESSMENT

PHARMACODYNAMICS

Ceftiofur is a 3rd generation cephalosporin. Cephalosporins have a bactericidal mechanism of action similar to that of other β-lactam antibiotics. They disrupt bacterial cell wall synthesis, targeting the penicillin-binding proteins (PBPs) located in the bacterial cell wall.

Ceftiofur and to a lesser extent its main metabolite, show activity against a variety of gram-negative and gram-positive bacteria in a time-dependent manner.

Ceftiofur is highly active against a number of swine pathogens isolated in Europe (*Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus spp., Streptococcus suis, Escherichia coli, Klebsiella pneumoniae, Mannheimia haemolytica)*. The highest MIC_{90} values of ceftiofur are 0.125, 0.03, 0.03 and 0.13 µg/ml, respectively, against *Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus parasuis* and *Streptococcus suis*. All strains yielded MIC values below 0.5 µg/ml and would be classified as susceptible to ceftiofur using the CLSI breakpoint. Ceftiofur was not active against *Bordetella bronchiseptica*.

The pharmacodynamic data provided for cattle pathogens in Europe associated with bovine interdigital necrobacillosis were limited. MIC_{90} values ranged between 0.03 to 8.0 µg/ml. A high variability was observed.

Ceftiofur related breakpoints approved by the Clinical and Laboratory Standards Institute (CLSI; formerly known as NCCLS) for "susceptible" bacteria are ≤ 2.0 µg/ml and for "resistant" bacteria ≥ 8 µg/ml. However, these values do not include *Haemophilus parasuis*.

PHARMACOKINETICS

Naxcel is a long acting formulation and its pharmacokinetics are, therefore, not comparable with other ceftiofur salt formulations.

In pigs, one hour after a single administration, plasma concentrations are above 1 µg/ml with maximum concentrations (4.2 ± 0.9 µg/ml) reached at approximately 22 hours after administration. Plasma concentrations above 0.2 µg/ml are maintained for at least 158 hours.
Approximately 70% of ceftiofur-related molecules are reversibly bound to plasma proteins in pigs. Ceftiofur is quickly metabolised to its main metabolites, which also express antibacterial properties.
Ceftiofur in healthy animals has affinity for the respiratory tract, kidney, skin and synovial fluid, but not for cerebrospinal fluid. Accumulation of free ceftiofur and active metabolites in infected tissue has been demonstrated *ex vivo* in calves using tissue cages, with concentrations in the infected area exceeding those in plasma and non-infected areas.
Approximately 60% and 15% of the dose are excreted in the urine and faeces respectively, within 10 days after administration.

In cattle, two non-GLP studies were provided using radiolabelled ceftiofur in the final formulation following single subcutaneous injection at 6.6 mg/kg bw/day. The excreta and the edible tissues were assayed every 24 hours for 10 days and at 7 and 10 days, respectively. More than 95% of the dose of ceftiofur sodium is excreted within the first 24 hours, with 61-77% of the total drug (parent & metabolites) excreted through the urine and the remainder in the feces. 90% of administered ^{14}C dose was recovered from animals sacrified at both 7 and 10 days post-treatment, primarily in excreta.

Four GLP pharmacokinetic studies were performed in cattle following subcutaneous injection of Naxcel 200 mg/ml for cattle. The $AUC_{infinity}$ and Cmax increased in a dose related manner following administration of CCFA dosed at 3.3 – 6.6 and 13.2 mg/kg bw.

IMPACT OF RESISTANCE DEVELOPMENT TO EFFICACY IN TARGET ANIMALS

Resistance against macrolides, lincosamides and streptogramin B in *Streptococcus suis* is mediated by ribosome methylation, encoded by the ermB gene. Resistance to ceftiofur is relatively rare in Europe but reports from the USA indicate that bacteria resistant to ceftiofur often express plasmid mediated multiresistance to other antimicrobial substances.

The susceptibility of 135 *Streptococcus suis* strains isolated from pigs in France to 13 antimicrobial agents was studied in a survey going up to 2000. All strains were susceptible to ceftiofur. 124 and 180 *Streptococcus suis* strains isolated from pigs in Denmark showed little (4.8%) or no resistant strains before 1992 and 1995-96, respectively.

An assessment of the potential for resistance in cattle resulting from the use of Naxcel for the treatment of acute interdigital necrobacillosis associated with *Fusobacterium necrophorum* and *Bacteroides spp* in cattle was provided.

Taking into account the low incidence of resistance, the likely use of the product in individual animals rather than herd treatment, and the limited amount of microbiologically active residues in faeces and their rapid inactivation in manure, the Committee concluded that the impact of the use of Naxcel on the development of resistance in relation to efficacy in the target animals would be low.

TARGET ANIMAL SAFETY

In pigs, four studies were conducted to address target animal safety when ceftiofur was administered intramuscularly to swine.

Two of these studies (using ceftiofur sodium) demonstrated that ceftiofur was well tolerated in pigs following daily intramuscular doses up to 5 times the recommended dose for 15 days, and 25 times the recommended dose for 5 days, respectively. There were no clinical signs except for local transient tissue reactions at the injection sites.

Two further studies, using ceftiofur free acid formulation in the recommended dosage, revealed transient palpable swellings at the injection sites, and local tissue reactions classified as foreign body granulomas, with or without cystic cavities, which could be found up to 42 days post-injection. In some animals at the injection site a single small area (less than 6 cm^2) of tan discoloration in the fascia separating muscle bundles could be observed. Resolution of these reactions has been observed at 56 days post-injection.

In cattle, three studies were presented to evaluate target animal safety when ceftiofur (as ceftiofur sodium) was administered intramuscularly in doses up to 25x the recommended therapeutic dose. Although the active substance administered in these studies was presented slightly different (salt) to the final presentation (free acid), both dissociate into the same ceftiofur anion when in solution. Furthermore, pharmacokinectic data show that when ceftiofur is administered by the intramuscular or subcutaneous route, plasma-time concentration profiles are broadly similar. Consequently, it is accepted that use of data from the three early studies (ceftiofur sodium administered by the intramuscular route) in support of the systemic tolerance of the formulation proposed for marketing (ceftiofur crystalline free acid for administration by the subcutaneous route) was justified. The Committee agreed that that the active substance, ceftiofur, has a wide systemic margin of safety in cattle. It was therefore not deemed necessary to conduct further specific target animal safety studies with the final formulation in cattle.

In order to demonstrate local tolerance, a pivotal GLP study was provided using Naxcel as recommended in animals up to approximately 900 kg bodyweight (using a maximum volume of 30 ml). No signs of systemic intolerance were detected. Swelling was a common occurrence at sites where the test item was administered. Typically, these lesions were not painful and resolved over a period of weeks (swelling was not detected clinically after day 23). Histopathological examination of injection site tissue revealed discolouration of the fat pad and signs of inflammation characterised by fibrosis, granulomas and perivascular lymphocyte infiltrates. These findings indicate local irritation at the site of injection.

Target animal safety was also evaluated as part of the clinical efficacy study. In this study, there were no treatment-related systemic adverse effects. However, reactions at the injection site were common. Typically, these reactions were characterised by swelling and, in a proportion of cases, pain. The frequency of observation of pain at the injection site was higher in the field trial compared with the local tolerance study. The reason for this is unclear, but may be related to differences in the method of assessing pain and that the local tolerance study was targeted specifically at assessing occurrence of local reaction, including pain. Another reason may have been the careful preparation of the "base of the ear" sites prior to injection in the local tolerance studies by which injection site reactions may have tended to become less severe when compared to the conditions in the field trial. Notwithstanding the differences in frequency of pain observations between the target animal safety study and the field study, the results clearly indicate that in the field pain is detected at the injection site in a relatively large proportion of treated animals. Consequently, the potential for such lesions to be painful and their frequency are reflected in section 4.6 of the SPC.

Based on pharmacovigilance experience from the US, rare cases of sudden death have been reported, which might be in relation to intra-arterial injection or anaphylaxis. The potential for such effects is reflected in section 4.6 of the SPC.

The CVMP concluded that ceftiofur in pigs and cattle was well tolerated systemically and showed mainly adverse effects at the injection sites. In cattle, very rare cases of sudden death have been reported. Appropriate warnings have therefore been included in the SPC and product literature under "Undesirable effects (frequency and seriousness)".
It is recommended to limit injection volumes to a maximum of 4 ml (pigs) and 30 ml (cattle) per injection site.

CLINICAL DOCUMENTATION

Pigs

Bacterial Swine Respiratory Disease (SRD)

The majority of respiratory problems in pigs is due to a combination of disease pathogens. The clinical signs of swine respiratory disease may include: coughing, sneezing, nasal discharge, tear stains under the eyes, lethargic pigs that are often off-feed, have elevated rectal temperatures, and laboured breathing. In severe cases cyanosis in the tips of the ears can be seen.

To support the indication "bacterial respiratory disease associated with *Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus parasuis,* and *Streptococcus suis",* the Applicant provided three dose determination studies and three further dose confirmation studies.

Challenge studies

In a GCP-compliant (placebo controlled, randomised, blinded) dose determination study in USA, pigs aged 6 to 8 weeks were challenged with *A. pleuropneumoniae* via intratracheal inoculation. Three hours after challenge, the animals were treated intramuscularly with up to 12 mg ceftiofur crystalline free acid/kg bodyweight. Mortality rates and lung lesion scores were significantly less for all treatment groups than for the placebo group. The percentages of culture positive lungs for *A. pleuropneumoniae* for the groups treated with 6 mg/kg or more were significantly smaller than for the groups treated with 0, 1 or 3 mg/kg.

In a GCP compliant (randomised, blinded) dose confirmation study in USA, pigs aged 6 to 8 weeks were challenged with *A. pleuropneumoniae*. Three hours after challenge, the animals were treated intramuscularly with 5 mg ceftiofur crystalline free acid (final formulation)/kg bodyweight, a placebo or a positive control (authorised in the EU). Mortality and lung lesion scores were the primary decision variables for the assessment of any efficacy differences between the groups. Ancillary variables included rectal temperature, clinical scores and bacteriological results.
The high mortality, number of pathogens isolated from lungs, and lung lesion scores in the placebo group confirmed the validity of the experimental model. There was no statistical difference between ceftiofur crystalline free acid (final formulation) and the positive control confirming the efficacy of Naxcel at the dosage of 5 mg/kg bodyweight in the treatment of respiratory disease associated with *A. pleuropneumoniae*.

Field studies

In a European GCP multicentre (Germany, France, Denmark) dose determination study, the efficacy of a single intramuscular dose of 3, 5 or 7 mg ceftiofur/kg bodyweight for the treatment of bacterial swine respiratory disease was compared to a positive control containing amoxicillin. The pigs involved in the study were naturally infected and diagnosed with clinical respiratory disease including fever, dyspnoea, increased respiratory rate or open mouth breathing. All three treatment levels were not significantly less efficacious than the positive control.

In another multicentre GCP dose determination study conducted in the USA, naturally infected pigs with diagnosed clinical respiratory disease were treated with a single intramuscular dose of 3, 5 or 7 mg ceftiofur/kg bodyweight and compared to a negative control. Bacteriological findings on Day 14 included *Actinobacillus pleuropneumoniae, Haemophilus parasuis, Pasteurella multocida* and/or *Streptococcus suis.* The mean lung lesion scores for the 5 and 7 mg/kg groups were significantly less than the placebo group and the mean percent "growers" (i.e. pigs gaining more than 2.2 kg bodyweight/14 days) for the 5 mg/kg treatment was significantly greater than the placebo group, demonstrating the efficacy of the ceftiofur free acid formulation.

In another multicentre GCP field study conducted in the USA, naturally infected pigs with diagnosed clinical respiratory disease were treated with a single intramuscular dose of 5 or 7 mg ceftiofur/kg bodyweight and compared to a negative control. The main pathogens identified at the initiation of the study (*Actinobacillus pleuropneumoniae, Haemophilus parasuis, Pasteurella multocida* and/or *Streptococcus suis*) confirmed the presence of Swine Respiratory Disease (SRD). Regarding the mortality and mean lung lesion scores, no significant difference was observed between the two dosage groups and the placebo. However the clinical cure rate of Naxcel treated pigs was significantly better than that of control pigs, establishing the efficacy as defined in the protocol. For the mean rectal temperature and weight gain ancillary variables, significant difference were also observed. The mean rectal temperatures in the Naxcel groups were significantly less than in the placebo group whilst the percentage of pigs gaining more than 2.2 kg bodyweight/14 days was improved in the 5 mg Naxcel group. The study supports the efficacy of Naxcel as a single injection at a dose of 5 or 7 mg ceftiofur /kg bodyweight under field conditions.

A GCP compliant (controlled, randomised, blinded) multicentre European (Denmark, France, Germany) dose confirmation study was performed in naturally infected pigs weighing 18 to 80 kg. The primary criteria for any site entering the study was the occurrence of SRD in a group of pigs with a morbidity of 10% or more. Pigs were treated with either a single injection of 5 mg ceftiofur crystalline free acid (final formulation)/kg bodyweight or a positive control containing amoxicillin. On the basis of the lung lesion scores and the percentage of pigs gaining more than 2.2 kg bodyweight/14 days, it was concluded that the efficacy of Naxcel is statistically comparable to the positive control. The mortality rate was relatively low, which was attributed to an early treatment or to a mild severity of the disease.

Conclusions:

Although the three dose determination studies had not been performed with the final formulation but with an earlier one using different excipients, the Committee concluded that the challenge study together with the field studies confirm the efficacy of a single dose of 5 mg ceftiofur crystalline free acid/kg bodyweight for the treatment of bacterial respiratory disease in swine associated with *Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus parasuis* and *Streptococcus suis*.

***Streptococcus suis* Disease Complex**

Infection with *Streptococcus suis* occurs via inhalation, through contact, or via small wounds. Pigs of all ages may be infected with *S. suis*. In animals with a well-functioning immune system there will be no clinical signs, and the pathogen resides in the tonsils. It has been demonstrated that in Western Europe, nearly 100% of pigs are carriers of one or more serotypes of *S. suis* in their tonsils. However, the overall disease incidence averages only 5%.

Disease can be triggered by external factors (e.g. stress), or by infection with other pathogens. In older pigs, *S. suis* will be found predominantly as a co-pathogen in respiratory infections, while in young piglets, the pathogen can become septicaemic and may cause disease and sudden death, or may be spread throughout the piglet's body. Target organs and tissues include the joints, serosa and the meninges. Piglets may therefore exhibit signs of septicaemia, (poly)arthritis, polyserositis and/or meningitis. Pneumonia may also be present as a mixed infection.

Challenge Study

Efficacy of Naxcel has been demonstrated in a GCP-compliant experimental dose confirmation study using a valid model. Piglets of about 2 weeks of age were infected intravenously using an *S. suis* serotype 2 inoculum, mimicking the septicaemic part of the pathogenesis of the disease. Only piglets with clear clinical signs associated with *S. suis* infection were included. The animals were then treated either with Naxcel or with a negative control.

Following treatment, animals in the Naxcel group had less severe lameness scores than the piglets in the placebo group. Incidence and severity of inflammation scores were lower for the Naxcel group than for the placebo group. A significant difference was found in "mortality" with 21% in the Naxcel group versus 75% in the placebo group. About 10% of all the piglets showed signs of meningitis at the time of enrolment. The number of animals with CNS scores increased at a later stage but then in combination with other signs. 63% of the ceftiofur-treated animals never showed any sign of meningitis, compared to 15% in the placebo-treated control group. Minor lesions at injection site were seen on a number of piglets.

It was concluded that under the conditions of this experiment, Naxcel was efficacious at the therapeutic dosage in the treatment of clinical signs associated with *S. suis* infection. However, since piglets under field conditions might be treated at a later stage, this study was not considered sufficient to prove the efficacy of Naxcel under field conditions.

Field study

A GCP-compliant (controlled, randomised, single blinded) multicentre European (Belgium, France, Netherlands, Germany) field study was conducted with naturally infected pigs aged about 4 weeks from commercial farms with a history of *Streptococcus suis* infection. The inclusion criteria for piglets entering the study were the acute onset of one or more of the followings clinical signs associated with polyarthritis, septicaemia and/or meningitis, i.e. CNS symptoms indicating meningitis; mild lameness in one or more legs together with fever or moderate to severe lameness in at least one leg with or without swelling of the joint(s) addressing polyarthritis; mild to moderate depression together with fever or severe depression indicative for septicaemia. Pigs were treated with either a single injection of 5 mg ceftiofur crystalline free acid/kg bodyweight or a positive control containing amoxicillin.

A significant number of animals enrolled in the study showed clinical signs associated with polyarthritis and/or septicaemia, however, the number of animals showing clinical signs associated with meningitis was relatively low.

The main parameter to analyse the efficacy of the treatment was the mortality rate. Other parameters were lameness, depression, rectal temperature and relapse rate. There was no statistically significant difference between the Naxcel group and the control group with respect to mortality and most of the other parameters.

The CVMP therefore concluded that the data demonstrated sufficiently the efficacy of Naxcel in the treatment of septicaemia, polyarthritis and/or polyserositis associated with *Streptococcus suis* infections.

With regard to the treatment of meningitis, the Committee acknowledged the positive results of the challenge study and the difficulties encountered under field conditions in demonstrating the efficacy of the treatment of meningitis. The Committee considered that the result of the challenge study indicated that the disease is expressed firstly in the joints, and only at a later stage in the brain, and that early treatment of piglets might prevent the onset of clinical signs of meningitis. However, it was concluded that the limited number of animals with clinical signs of meningitis enrolled in the field study was too low to demonstrate the efficacy of Naxcel in the treatment of meningitis associated with infection by *Streptococcus suis*.

Cattle

Dose determination / justification

No classical dose determination studies were performed. The marketing authorisation holder used a PD/PK approach to select a dose of product to use in a clinical efficacy study: The main criteria to justify the selected dose rate were the requirements to maintain ceftiofur levels above 0.2 µg/ml (a concentration higher than the MIC of the majority of target pathogens) for a sufficient period. Doses of 3 mg/kg and 6.6 mg/kg bodyweight were selected for *in vivo* clinical efficacy testing. However, this approach was not considered pivotal in establishing the dosing regime. In the clinical study, the dose of 6.6 mg/kg was retained because it performed better than the lower dose. The PD/PK approach was not considered as fully robust in concluding at the dose, but good clinical efficacy at 6.6 mg/kg justified the dosing regime as retained.

Field trials (including dose confirmation)

A well conducted multi-centre GCP compliant field study was conducted in several European countries between 2006 and 2008. The objective of the study was to evaluate the field safety and efficacy of Naxcel administered subcutaneously at the base of the ear as a single dose of 3 mg or 6.6 mg ceftiofur per kg bodyweight in the treatment of naturally occurring bovine interdigital necrobacillosis associated with anaerobic Gram-negative bacteria including *Fusobacterium necrophorum* and *Bacteroides melaninogenicus* in adult dairy cattle compared with a positive control, a product authorised for the intended indication containing containing ceftiofur. The study was

conducted using the formulation in animals that are representative of the target population. Inclusion and exclusion criteria were clearly defined. The major pathogens associated with this clinical condition were equally represented in all three treatment groups.

Interim statistical analysis was performed approximately 50% of animals had been enrolled in the study: the findings of this analysis was that animal withdrawal related to inter-digital necrobacillosis in the low-dose test product group (3 mg/kg) was more than four times the rate of withdrawal of animals assigned to the positive control. The difference in the rate of withdrawals was not statistically significant but, on welfare grounds, was considered to be unacceptably high. Enrolment of animals into the low-dose test product group was therefore terminated at this time.

Based on the final analysis of efficacy, administration of ceftiofur crystalline free acid at a dose of 6.6 mg/kg bodyweight by single subcutaneous injection for the treatment of interdigital necrobacillosis was shown to be non-inferior to the positive control. Generalised mixed linear model analysis for non-inferiority established the following back-transformed lower and upper 95% confidence limits, and least square means (LS means) for cure rates: 63.3 to 86.6% (LS mean = 76.9%) for the low-dose test product group; 73.4 to 89.1% (LS mean = 82.6%) for the high-dose test product group and 71.1 to 89.9% (LS mean = 82.4%) for the positive control group. Based on the lower 95% confidence limits, and using 15% difference as the acceptable margin, non-inferiority was demonstrated for the high-dose test product group (6.6 mg/kg).

No treatment related systemic adverse reactions were recorded during the clinical efficacy study although local reactions at the injection site (swelling and pain) were recorded in some animals.

The CVMP did not consider that the dose has been confirmed since it has been tested in only one clinical trial. However, given the multicentered nature of the trial, no further clinical data were considered necessary to further optimise the dose.

OVERALL CONCLUSIONS ON EFFICACY

Cephalosporins, like other β-lactam antibiotics, disrupt bacterial cell wall synthesis, targeting the penicillin-binding proteins (PBPs) located therein. Ceftiofur has a time-dependent bactericidal activity. *In vitro* data were provided, demonstrating the susceptibility of relevant pathogens against ceftiofur. The highest MIC_{90} values of ceftiofur are 0.125, 0.03, 0.03 and 0.13 µg/ml, against *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis* and *Streptococcus suis*, respectively. In cattle, MIC_{90} values ranged between 0.03 to 8.0 µg/ml, with a high variability.

In pigs, one hour after a single administration, plasma concentrations are above 1 µg/ml, with maximum concentrations reached at approximately 22 hours after administration. Plasma concentrations above 0.2 µg/ml are maintained for at least 158 hours. With a view to the claimed indications there is sufficient evidence that ceftiofur and/or its metabolites reach relevant target tissues. Accumulation of free ceftiofur and active metabolites in infected tissue has been demonstrated *ex vivo* in calves, with concentrations in the infected area exceeding those in plasma and in the non-infected areas.
In cattle, ceftiofur is well absorbed following base of the ear injection and quickly metabolised to desfuroylceftiofur, the principal active metabolite. Protein binding of ceftiofur and its major metabolite is approximately 70-90%. One hour after a single administration, plasma concentrations are greater than 1 µg/ml. Maximum concentrations in plasma (about 5 µg/ml) occurred from 12 hours following administration. Plasma concentrations above 0.2 µg/ml of ceftiofur and its metabolite are maintained for at least 7 days. Excretion is mainly via urine.

Development of resistance with regard to efficacy of the product and the potential impact on human safety were sufficiently addressed by the Applicant. Taking into account the low incidence of resistance in Europe, despite the use of ceftiofur for more than 10 years, the likely use of the product in individual animals rather than herd treatment, the limited amount of microbiologically active residues in faeces and their rapid inactivation in manure, the Committee concluded that the impact of

the use of Naxcel on the development of resistance in view of efficacy in the target animal would be low.

Ceftiofur was well tolerated in pigs and cattle systemically. Local effects (swelling and pain) were seen at the injection sites. In cattle, very rare cases of sudden death have been reported. Appropriate warnings have therefore been included in the SPC and product literature under "Undesirable effects (frequency and seriousness)".
It is recommended to limit injection volumes to a maximum of 4 ml (pigs) and 30 ml (cattle) per injection site

In order to demonstrate efficacy of Naxcel in the treatment of Bacterial Swine Respiratory Disease, three dose determination studies (using a formulation with slightly different excipients to the final formulation) and three dose confirmation studies have been conducted using the final formulation: one challenge study (positive and negative control), one field study in the EU (positive control), and one field study in the US (negative control). All of these studies confirm the efficacy of a single 5 mg/kg dose for the treatment of bacterial respiratory disease in swine associated with *Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus parasuis* and *Streptococcus suis*.

In order to demonstrate efficacy of Naxcel in the treatment of infection associated with the *Streptococcus suis* disease complex, the Applicant provided a challenge study performed in the Netherlands using a negative control group, and one European multicentre field study using a positive control group study containing amoxicillin. In both the challenge and the field studies, lameness was the most important condition encountered at enrolment. A significant number of animals also showed clinical signs associated with polyarthritis and/or septicaemia, however, the number of animals in the field study showing clinical signs associated with meningitis was relatively low and, therefore, considered by CVMP as insufficient to support the proposed indication of "treatment of meningitis associated with *S. suis* infection". The main parameter to analyse the efficacy of the treatment was the "mortality rate". In both studies, treatment with Naxcel resulted in either a significantly lower (compared to the negative control) or a comparable mortality rate (compared to the positive control). The CVMP therefore, concluded that the data demonstrated sufficiently the efficacy of Naxcel in the "treatment of septicaemia, polyarthritis or polyserositis associated with *Streptococcus suis* infections".

In cattle, ceftiofur crystalline free acid administered once subcutaneously at the base of the ear, at a dose rate of 6.6 mg/kg in the treatment of interdigital necrobacillosis was efficacious. The test product was shown to be non-inferior to treatment with ceftiofur sodium administered intramuscularly on three consecutive days at a dose rate of 1 mg/kg bodyweight.

5. BENEFIT RISK ASSESSMENT

Naxcel is an oily, non-aqueous suspension for injection containing a third generation cephalosporin, ceftiofur, as active ingredient. It is used in pigs for the treatment of bacterial respiratory diseases, septicaemia and polyarthritis/polyserositis and in cattle for the treatment of acute interdigital necrobacillosis (panaritium or foot rot). In pigs, the withdrawal period in meat and offal is 71 days. In cattle, the withdrawal period in meat and offal is nine days, and in milk it is zero days.

Benefit assessment

Direct therapeutic benefits

Naxcel contains ceftiofur which is present in a number of products that are already licensed for use in pigs and cattle. Ceftiofur's use and activity as a third-generation cephalosporin are well known. Its mode of action is typical of a late generation beta-lactam-type antibiotic, with activity against a large number of Gram-negative and Gram-positive bacteria. *In vitro* efficacy against pathogens relevant for the indications in cattle and pigs has been demonstrated.

In pigs, the product is used in the treatment of bacterial respiratory diseases caused by a variety of bacteria, septicaemia and polyarthritis/polyserositis. Treatment resulted in direct benefits to the animals' welfare, i.e. the improvement of clinical conditions and reduction of mortality in affected herds.

In cattle, the product is used in the treatment of interdigital necrobacillosis, which is a serious disease causing lameness. The direct benefits to the animal include improved, less painful ambulation. Systemic antibiotic treatment is accepted practice for treatment of cattle suffering from interdigital necrobacillosis. Beyond the direct benefits to the animal, if treated early, this disease is not debilitating and cows will not have to be culled because of it. It is therefore expected that the life expectancy of the treated cow is increased.

Additional benefits

Indirect benefits due to treatment of pigs were seen in commercial benefits for the farmer, i.e. improved gain in bodyweight in treated pigs, reduction in mortality and improved quality of carcass.

Naxcel 200 mg/ml for cattle is effective after a single administration whereas currently, ceftiofur must be given for 3 days for treatment of interdigital necrobacillosis. Other treatments, including local paring of the hoof, are very labour-intensive. Reduction in animal handling and increased compliance are expected to provide an additional advantage to the treatment by this veterinary medicinal product.

RISK ASSESSMENT

Main potential risks

Quality

The product is a sterile, non-aqueous, oily, injectable suspension.

In early development studies the drug release characteristics of the suspension (with potential impact on both efficacy and residues) were found to be dependent on several product manufacturing variables, however the tight manufacturing controls developed as a result of recognising this has lead to a well defined, controlled manufacturing process leading to a consistent product.

The quality data provided are satisfactory and comply with current guidelines.

Safety

Ceftiofur is of low acute oral and parenteral toxicity, and is non-mutagenic or carcinogenic and reproductive toxicity studies in mice indicated that ceftiofur was not reprotoxic or teratogenic. However, a teratogenicity study in rats revealed maternotoxic and foetotoxic effects, and a warning has therefore been included in the product literature to use the product in pregnant animals only following a benefit/risk assessment by the responsible veterinarian.

Ceftiofur is slightly irritant to abraded skin and minimal irritating on rabbit eyes. Like other cephalosporins, ceftiofur has a potential to cause hypersensitivity or allergic reactions following injection, inhalation, ingestion or skin contact. Under field conditions, while handling the product, accidental skin or eye contact may occur. Adequate precautionary warnings are, therefore, included in the SPC.

In cattle, the at the ear injection does not seem to pose an additional risk to the user and consequently no particular warning in regards to this route of injection is necessary in the SPC and product literature. Proper restraint which such an injection necessitates is considered to be part of good veterinary practice and no particular mention of the need for this has been listed in the SPC nor in the product literature.

The environmental risk assessment allows halting the assessment in Phase I, since exposure is below the Phase I trigger value of 100 µg ceftiofur /kg soil.

The excipients used are no cause for concern in terms of animal, user and/or consumer safety.

MRLs have been established for ceftiofur and are listed in Annex I of Council Regulation (EEC) No. 2377/90. A withdrawal period of 71 days for meat and offal was established in pigs. In cattle, the product is injected into the base of the ear, a location which is considered non-edible tissue in Europe. Therefore, provided the product is used as recommended, a withdrawal period of 9 days for meat and offal and zero days for milk was established. In order to avoid injection into edible tissues, which users might be tempted to do, detailed description is given of the correct route of administration as well as an appropriate warning to emphasize that the withdrawal period is only valid if the product is administered by the recommended route and at the correct location.

Efficacy

In terms of tolerance, ceftiofur is well tolerated systemically in pigs and cattle. Local effects (swelling and pain) were seen at the injection sites. In cattle, very rare cases of sudden death have been reported. Appropriate warnings have therefore been included in the SPC and product literature under "Undesirable effects (frequency and seriousness)". Also, it is recommended to limit injection volumes to a maximum of 4 ml (pigs) and 30 ml (cattle) per injection site.

Specific potential risks - resistance

Third generation cephalosporins are listed by WHO as critically important antimicrobials for human use and recent monitoring data indicate increased frequency of resistance due to ESBL. Therefore, a number of prudent use warnings have been added to the SPC (see section 4.5 which is in line with the CVMP reflection paper on late-generation cephalosporins (EMEA/CVMP/SAGAM/81730/2006-Rev.1).

Development of resistance with regard to efficacy of the product and the potential impact on human safety were sufficiently addressed. Taking into account the low incidence of resistance, the likely use of the product in individual animals rather than herd treatment, and the limited amount of microbiologically active residues in faeces and their rapid inactivation in manure, the Committee concluded that the impact of the use of Naxcel on the development of resistance would be low.

EVALUATION OF THE BENEFIT RISK BALANCE

Naxcel has been shown to have a positive benefit risk balance overall. It has been shown to be efficacious in pigs and cattle in a number of indications. The formulation and manufacture of Naxcel are well described and specifications set will ensure that the manufactured product is of a consistent high quality. The product is well tolerated by the target animals and presents a low risk for users and the environment and appropriate warnings have been included in the SPC and product literature. Sufficient withdrawal periods have been set to ensure consumer safety.

CONCLUSION

The overall benefit risk evaluation is deemed positive with a sufficiently clear and complete SPC and product literature.

Based on the original and complementary data presented the Committee for Medicinal Products for Veterinary Use (CVMP) concluded that the quality, safety and efficacy of Naxcel were considered to be in accordance with the requirements of Directive 2001/82/EC, as amended.