Date of Approval Letter: June 18, 2004

FREEDOM OF INFORMATION SUMMARY

ORIGINAL NEW ANIMAL DRUG APPLICATION

NADA 141-235

EXCEDE for Swine (ceftiofur crystalline free acid)
Sterile Suspension

For the treatment of swine respiratory disease (SRD) associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis*

Sponsored by:
Pharmacia & Upjohn Company
A Division of Pfizer Inc

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1. GENERAL INFORMATION:

a. File Number:	NADA 141-235
b. Sponsor:	Pharmacia & Upjohn Co. 7000 Portage Rd. Kalamazoo, MI 49001-0199 Drug Labeler Code: 000009
c. Established Name:	Ceftiofur crystalline free acid (CCFA)
d. Proprietary Name:	EXCEDE for Swine
e. Dosage Form:	Sterile oil suspension for injection
f. How Supplied:	100 mL glass vial
g. How Dispensed:	Rx
h. Amount of Active Ingredients:	100 mg ceftiofur equivalents (CE) per mL
i. Route of Administration:	Intramuscular (IM) injection in the post-auricular region of the neck
j. Species/Class:	Swine
k. Recommended Dosage:	Single IM injection of 5.0 mg CE/kg (2.27 mg CE/lb) body weight (1.0 mL sterile suspension per 44 lb body weight). No more than 2.0 mL should be injected in a single injection site. Pigs heavier than 88 lb (40 kg) will require more than one injection.
l. Pharmacological Category:	Antimicrobial
m. Indications:	EXCEDE For Swine Sterile Suspension 100 mg/mL is indicated for the treatment of swine respiratory disease (SRD) associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis*.

2. EFFECTIVENESS:

a. Dosage Characterization:

The following dose characterization studies were conducted using a non-final formulation of ceftiofur crystalline free acid (CCFA) sterile suspension. The studies enabled the selection of two doses to be used in the dose confirmation (field) studies, which were conducted using the final CCFA formulation.

1. "The Efficacy of Five Dose Levels of a Sterile Suspension of Ceftiofur Crystalline Free Acid Administered Intramuscularly in Swine Inoculated with *Actinobacillus pleuropneumoniae*." Report No. 829-9690-95-002.

The objective of this study was to evaluate the effectiveness of a single intramuscular (IM) injection of five dose levels of CCFA compared with a placebo (vehicle) control. The investigators were J.W. Hallberg, R.A. Rzepkowski, R.J. Yancey, R.K. Frank, R.A. Evans, B.J. Hanson, K.J. Dame, M.J. Duke, G.M. Baird, and G.R. Bos. The study was conducted in Richland, Michigan. One hundred and forty-four healthy crossbred castrated male pigs, weighing 6.4-28.7 kg, were used in the study. Following a 12-day acclimation period, all pigs were challenged intra-tracheally with approximately 1 x 10^6 cfu of *Actinobacillus pleuropneumoniae*. Pigs were assigned to one of six treatment groups, and received a single IM injection of CCFA at 1.0, 3.0, 6.0, 9.0, or 12.0 mg ceftiofur equivalents (CE)/kg of body weight (BW), or a single IM injection of vehicle (placebo control). Pigs were monitored daily for 14 days post-injection. At the end of the study, all surviving pigs were sacrificed to obtain lung lesion data.

No adverse events were reported for this study. The mortality rates for all CCFA-treated groups were lower than the control group mortality rate. The lung lesion scores for all CCFA-treated groups were lower than the control group lung lesion scores. A polynomial (quadratic) model was used to determine a dose response curve and estimate the dose where mortality and lung lesion scores were minimized. Using the model, 5.0 mg CE/kg BW was defined as the effective dose.

2. "Field Dose Confirmation for Ceftiofur Crystalline Free Acid Sterile Suspension Administered as a Single Intramuscular Dose of 3.0, 5.0, or 7.0 mg Ceftiofur/kg BW, for the Treatment of Bacterial Swine Respiratory Disease." Report No. 829-9690-97-002.

The objective of this multi-location field study was to evaluate the effectiveness of a single IM injection of CCFA sterile suspension at 3.0, 5.0, and 7.0 mg CE/kg BW compared with a placebo (vehicle) control. The investigators were A. Beyer (W. Branch, IA), C. Rowles (Carroll, IA), K. Lorenzen (Sutton, NE), Dr. R. Bush (Flora, IN), J. Harker (Frankfort, IN), M. Rodibaugh (Frankfort, IN), S. Dudley (Worthington, MN), P. Yeske (St. Peter, MN), R. Blomme (Audubon, IA), J. Connor (Carthage, IL), W. Hollis (Carthage, IL), J. Hoffman (Avoca, IA), G. Schultz (Avoca, IA), R. Evelsizer (Fairmont, MN), and L. Hoffman (Ames, IA). Four hundred and seventy eight pigs, with clinical signs of bacterial respiratory

disease - rectal temperature ≥103.5°F and/or respiratory signs (dyspnea, increased respiratory rate, or open mouth breathing and/or signs of pneumonia) - were enrolled in the study. At the time of enrollment, pigs were randomly allocated to treatment groups, and received CCFA sterile suspension at 3.0, 5.0, or 7.0 mg CE/kg BW or a placebo (vehicle) injection. All treatments were administered as a single IM injection in the post-auricular region of the neck. Pigs were evaluated for general appearance and respiratory signs on Days 1-7 and Day 14. Lung lesion data was collected from all pigs that died during the study. All surviving pigs were sacrificed at the end of the study to obtain lung lesion data.

The primary decision variables for evaluating effectiveness were mortality, lung lesion scores, and percent growers (defined as pigs surviving 14 days and gaining at least 2.5 kg during the study period). The mortality rates for all CCFA-treated groups were lower than the control group. The lung lesion scores for all CCFA-treated groups were lower than the control group. The mean percentage of growers was higher for all CCFA-treated groups than the control group. Bacterial culture of the lungs showed the presence of *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis*. No adverse events were reported for this study.

The results demonstrate that a non-final formulation of CCFA, at doses of 5.0 and 7.0 mg CE/kg BW, was effective for the treatment of bacterial swine respiratory disease associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis*. Because the study was conducted with a non-final formulation, the study was not accepted as substantial evidence.

b. Substantial Evidence:

1. "Field Dose Confirmation of a Single Intramuscular Dose of the Revised Formulation of Ceftiofur Crystalline Free Acid Sterile Suspension (CCFA-SS) at 5.0 or 7.0 mg Ceftiofur Equivalents (CE) Per Kg of Body Weight for the Treatment of Naturally Occurring Bacterial Swine Respiratory Disease." Report No. a0100830.

a. Type of Study: a multi-location field study.

b. Investigators:

K. Lorenzen, J. Waddell. Sutton Veterinary Clinic, Sutton, NE
R. Harrison, D. Bryant. Professional Veterinary Research, Inc., Brownstown, IN
J. Harker. Swine Health Service, Frankfort, IN
S. Menke. Ottumwa Veterinary Clinic, Ottumwa, IA
S. Schmitz. Audubon/Manning Veterinary Clinic, Audubon, IA
R. Evelsizer. Fairmont, MN
N. Winkelman. Swine Services Unlimited, Morris, MN
M. Mohr, P. Yeske. Swine Vet Center, P.A., St. Peter, MN

J. Connor. Carthage Veterinary Service, LTD, Carthage, IL
J. Trimble. Robinson Hospital for Animals, LTD, Robinson, IL
J. Kober. Swine Veterinary Services of Michigan, LLC, Holland, MI
L. Hoffman. Diagnostic Laboratory, College of Veterinary Medicine, Iowa State University, Ames, IA

c. Study Design:

1) *Objective:* To investigate the effectiveness of a single IM injection of CCFA (final formulation) at doses of 5.0 or 7.0 mg CE/kg BW for the treatment of naturally occurring bacterial SRD when compared to a placebo-treated control.

2) *Experimental Animals:* A total of 780 pigs were enrolled in the study, conducted at a total of 14 swine farms in Nebraska, Indiana, Iowa, Illinois, Michigan, and Minnesota. Pigs were commercial crossbred or purebred pigs typical of U.S. swine operations. Each site enrolled 45-60 study pigs and three sentinel pigs. Pigs were enrolled if they showed signs of bacterial respiratory disease and met the entrance criteria – a combined general appearance and respiratory index score of 2 or greater, and a rectal temperature of ≥104.1°F.

3) *Study Schedule*: After a 14-day acclimation (antibiotic withdrawal and observation) period, pigs were eligible for enrollment. Assigned treatments were administered on the day of enrollment (Day 0). The first three enrolled pigs on each farm were designated as sentinel pigs and were sacrificed and necropsied. All pigs were examined daily on Days 1-7 for general appearance and respiratory signs. Rectal temperatures were taken on Days 0, 1, 3, and 6. Pigs were observed daily from Days 8-14 for general health. Necropsies were performed on all pigs that died during the study. On Day 14, all remaining pigs were weighed, sacrificed, and necropsied. The total study duration, including acclimation, treatment, and post-treatment, was 29 days.

4) *Test Article Administration:* CCFA sterile suspension (100 mg CE/mL, final formulation) was used as the test article. Pigs in the CCFA treatment groups received either 5.0 or 7.0 mg CE/kg (2.27 mg or 3.18 mg CE/lb) BW. A vehicle control injection was administered to the pigs allocated to the placebo control group. All treatments were administered as a single IM injection in the post-auricular region of the neck.

5) *Experimental Design:* The study was conducted as a randomized complete block design over 14 sites. The pig was the experimental unit. A replicate consisted of one block of three pigs (one pig of each treatment group). A maximum of 20 replicates (60 pigs) were assigned at each location. The investigators who made and recorded clinical assessments, and the laboratory personnel were masked to treatment.

6) *Measurements and Observations:* Mortality, clinical cure rate, and lung lesion scores were the primary decision variables. General appearance was evaluated according to the following scale: 0=normal, 1=mildly depressed, 2=moderately depressed, 3= severely depressed, 4= moribund. Respiratory function was evaluated according to the following scale: 0=normal, 1=mild dyspnea, 2=moderate dyspnea, 3=severe dyspnea. Treatment was considered a success (clinical cure) if the sum of the general appearance score and respiratory index was 0 or 1 and body temperature was ≤104.0°F, on both Days 3 and 6. Percent "growers" (defined as pigs surviving 14 days and gaining at least 2.5 kg during the study period) was evaluated as an ancillary variable.

   To determine lung lesion scores, lungs were visually observed, the percentage of each lobe containing lesions was recorded, and a formula was used to calculate the lung lesion score for each pig. Lungs from all study pigs were submitted to a diagnostic laboratory for bacterial etiologic determination.

d. Results: Analysis was conducted using 706 pigs. Thirty-two study pigs were not included in the analyses due to incorrect enrollment or incomplete data. The sentinel pigs (42) were not included in the analyses. The proportion of animals that died within each location x treatment group and the proportion of animals that were clinically cured within each location x treatment group were transformed using the Freeman-Tukey arcsine transformation. Transformed values were analyzed using weighted ANOVA. The percentage of growers was analyzed using SAS MIXED procedures. Treatment groups were compared using a one-sided test and a significance level of 5%. The results are shown in Table 2.1.

Table 2.1. Results of Study 829-7922-O-DMM-00-001

Variable	Placebo control	CCFA 5.0 mg CE/kg BW	CCFA 7.0 mg CE/kg BW
% Mortality	6.3%	4.3%	4.2%
% Clinical Cure Rate (p-value vs. control)	17.7%	24.8% (0.0343)	26.4% (0.0128)
Mean Lung Lesion Scores	9.2%	10.4 %	10.4%
% Growers	84.2%	88.8%	88.4%

The results show that there was a statistically significant improvement in clinical cure rates (treatment success) in the 5.0 mg CE/kg BW and 7.0 mg CE/kg BW treatment groups compared with the control group. Lung lesion scores were numerically lower in the control group, compared with the CCFA-treated groups. There were a higher percentage of growers in both CCFA treatment groups compared with the control group. Bacterial culture identified the following ceftiofur-sensitive respiratory pathogens: *Actinobacillus*

pleuropneumoniae, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis*.

e. Adverse Reactions: No adverse effects were reported in this study.

f. Conclusions: Based on the results of the study, the final CCFA formulation administered as a single IM injection of either 5.0 or 7.0 mg CE/kg BW is effective for the treatment of bacterial swine respiratory disease associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis*.

c. Pharmacokinetics:

"Plasma Pharmacokinetics and Residue Decline of Ceftiofur in the Injection Site of Swine Administered a Sterile Suspension of Ceftiofur Crystalline Free Acid (PNU-064279) at 100 mg Ceftiofur Equivalents (CE)/mL by Intramuscular Injection in the Neck at a Dose of 5.0 mg CE/kg Body Weight." Study number 2000-0305.

A pharmacokinetic (PK) study was conducted to characterize the kinetics of ceftiofur and desfuroylceftiofur-related metabolites in the plasma following administration of the final formulation of CCFA sterile suspension once at 5.0 mg CE/kg BW (2.27 mg CE/lb BW) IM in the neck of swine. The results of this investigation provide both pharmacokinetic and pharmacodynamic evidence supporting product effectiveness for the proposed indications and the CCFA component of a relative bioavailability analysis to justify the extrapolation of swine target animal safety data generated with ceftiofur sodium to the current application.

Materials and Methods

Thirty, 40 to 60 kg pigs were administered the final formulation of CCFA sterile suspension once in the neck at a dose of 5.0 mg CE/kg BW (2.27 mg CE/lb BW). Blood samples were taken from all animals at 0 (pretreatment), 6, 12, 24, 48, 72, 96, 120, 168, and 240 hours after treatment administration. Ceftiofur and desfuroylceftiofur-related metabolites were quantitated using a validated HPLC-DCA assay. The limit of quantitation (LOQ) and limit of detection (LOD) of the method were 0.15 and 0.054 μg CE/mL plasma, respectively. PK analyses of the plasma data were conducted.

Results

The plasma concentration data are plotted in Figure 2.1. PK variables are summarized in Table 2.2. Results are based on actual observed values, using noncompartmental methods.

Figure 2.1: Mean Plasma Concentrations (mcg CE/mL) of Ceftiofur and Desfuroylceftiofur after Administration of the Final Formulation of CCFA-SS to Swine IM at 5.0 mg CE/kg (2.27 mg CE/lb) BW

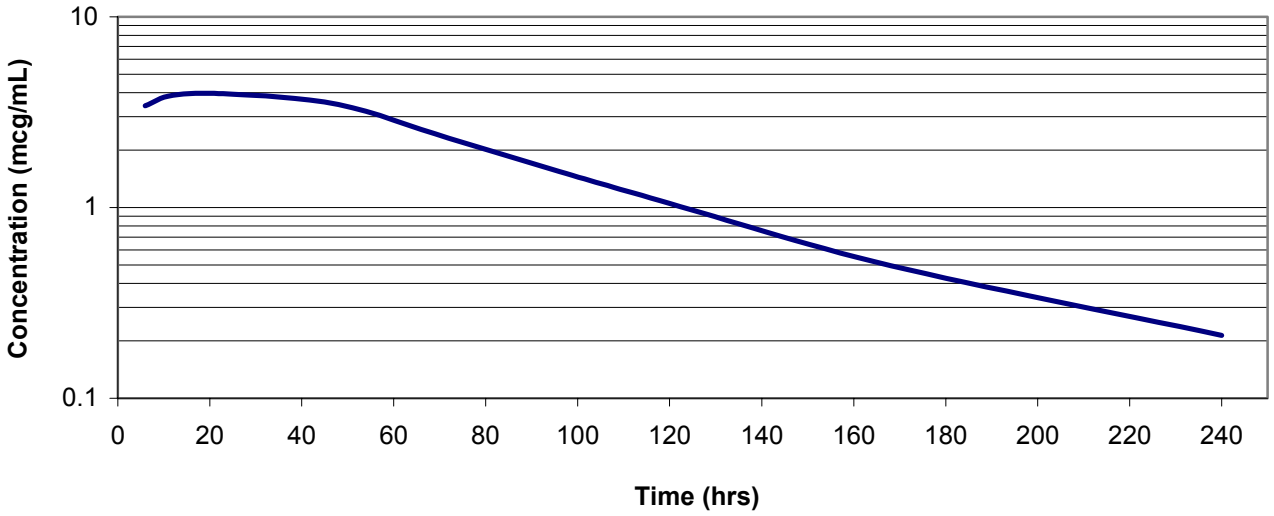


Table 2.2: Pharmacokinetic parameters in swine after a single IM administration of the final formulation of CCFA sterile suspension at 5.0 mg CE/kg (2.27 mg CE/lb) BW

Pharmacokinetic Parameter	Mean Value ± Standard Deviation
C_{max} (µg/mL)	4.17 ± 0.92
t_{max} (h)	22.0 ± 12.2
AUC $_{0\text{-}LOQ}$ (µg•h/mL)	373.0 ± 56.1
$t_{½}$ (h)	49.6 ± 11.8

C_{max} = maximum plasma concentration (in µg CE/mL)
t_{max} = the time after injection when C_{max} occurs (in hours)
AUC $_{0\text{-}LOQ}$ = the area under the plasma concentration vs. time curve from time of injection to the limit of quantitation of the assay (0.15 µg CE/mL)
$t_{½}$ = terminal phase biological half life (in hours)

CCFA is a beta lactam antimicrobial, thereby exhibiting time dependent killing. With regard to the kinetic/dynamic support of product effectiveness, to date, the evaluations in cattle have been based on the duration of time above which plasma concentrations of total ceftiofur equivalents remain above 0.2 µg/mL (refer to the FOI Summary for NADA 140-890 dated July 26, 1998). Justification for using 0.2 µg CE/mL as the target concentration in swine is justified on the basis of similar ceftiofur microbial susceptibility for the pathogens associated with the label claim in bovine and swine. The ability of the individual subjects to maintain total ceftiofur plasma concentrations at or above 0.2 µg/mL is demonstrated in Figure 2.2.

Corresponding estimates of μg CE/mL plasma at 72, 96, and 120 hours post dose (mean, standard deviation about the lower limit of mean concentrations estimated with 95% confidence) are provided in Table 2.3. Based upon these evaluations, the pharmacokinetics support the dosage regimen of CCFA in swine.

Figure 2.2: Maintainance of individual total ceftiofur plasma concentrations above 0.2 mcg/mL

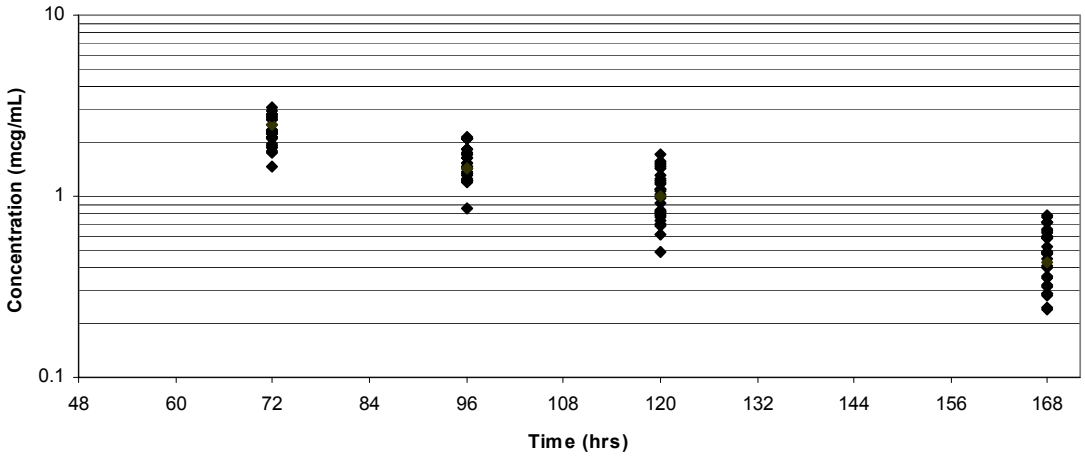


Table 2.3: Plasma concentrations (mean, standard deviation and lower confidence limit about the mean, estimated with 95% confidence*)

Hour of Blood Sample	Mean	Standard Deviation	Lower Confidence Limit
72	2.31	0.41	2.16
96	1.54	0.32	1.42
120	1.05	0.31	0.94
168	0.495	0.17	0.43

* where the lower confidence limit estimated as mean – ($t_{0.05, n-1}$* SEM), and

SEM = $mean/\sqrt{n}$

d. Microbiology:

1. Ceftiofur minimum inhibitory concentration (MIC) values from field studies evaluating SRD in the U.S.

"Minimum Inhibitory Concentration Determination for Ceftiofur Against Target Respiratory Disease Pathogens Isolated from the Lungs of Pigs Included in the Pivotal U.S. Clinical Field Efficacy Studies for Ceftiofur Crystalline Free Acid Sterile Suspension 100 mg/mL." Report No. 0829-7922-2003-011.

Clinical isolates derived from the two pivotal field effectiveness studies were tested for susceptibility to ceftiofur *in vitro*. The investigators were E.S. Portis, J.P. Crane, C.J. Lindeman, and S.A. Salmon, Pharmacia & Upjohn, Kalamazoo, Michigan. Ceftiofur MICs were determined using a commercially available microdilution system according to National Committee for Clinical Laboratory Standards (NCCLS) procedures. No standard method exists for MIC testing of *Haemophilus parasuis*. Modifications similar to those that NCCLS has recommended for *Haemophilus* spp. were made for testing *H. parasuis* isolates (NCCLS M7-A6, 2003). Quality control strains were included in each run and results were within acceptable ranges. Ceftiofur MIC distributions for the SRD pathogens *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Streptococcus suis* and *Haemophilus parasuis* are presented in Table 2.4.

Table 2.4. Ceftiofur MIC values from field studies evaluating swine respiratory disease (SRD) in the U.S. (1996-1997 and 2000-2001)

Pathogen	Number of isolates	MIC_{90}* (μg/mL)	MIC range (μg/mL)
Actinobacillus pleuropneumoniae	28	≤0.03	≤0.03-0.06
Pasteurella multocida	58	≤0.03	≤0.03**
Streptococcus suis	41	0.12	≤0.03-0.5
*Haemophilus parasuis****	72	0.06	≤0.03-0.25

* Minimum inhibitory concentration (MIC) for 90% of the isolates.
** No range; all isolates yielded the same value.
*** These MIC data were obtained using NCCLS procedures but quality control values for *H. parasuis* had not been standardized.

2. Ceftiofur MIC values from U.S. and Canadian diagnostic laboratory survey data during 1997 to 2001.

MIC Survey: "Results of 1997-98 Resistance Monitoring Program for Premafloxacin with Veterinary Pathogens." Report No. a0032820.

MIC Survey: "Results of 1998-99 Resistance Monitoring Program for Premafloxacin with Veterinary Pathogens." Report No. a0086065.

MIC Survey: "Results of 2000 Susceptibility Monitoring Program for Ceftiofur with Veterinary Pathogens." Report No. a0097495.

MIC Survey: "Results of 2001 Susceptibility Monitoring Program for Ceftiofur with Veterinary Pathogens." Report No. 0829-7922-2002-006.

These studies were conducted to determine the *in vitro* activity of veterinary antimicrobials, including ceftiofur, against veterinary pathogens isolated from food animals, including swine, in the U.S. and Canada (1997-2001). The investigators included E.S. Portis, S.A. Salmon, C.A. Case, J.L. Watts, and C.J. Lindeman, Pharmacia & Upjohn, Kalamazoo, Michigan. Isolates were

obtained annually from accredited veterinary diagnostic labs. Ceftiofur MICs were determined using a commercially available microdilution system according to NCCLS procedures. Quality control strains were included in each run and results were within acceptable ranges. Ceftiofur MICs for the SRD pathogens *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, and *Streptococcus suis* are presented in Table 2.5.

Table 2.5. Ceftiofur MIC values from U.S. and Canadian diagnostic laboratory survey data* during 1997 to 2001

Pathogen	Year Tested	Origin of Isolates	Number of Isolates	MIC_{90} ** (μg/mL)	MIC Range (μg/mL)
Actinobacillus pleuropneumoniae	1997-1998	U.S.	97	≤0.03	≤0.03***
Pasteurella multocida	1997-1998	U.S.	114	≤0.03	≤0.03-1.0
Streptococcus suis	1997-1998	U.S.	106	0.50	≤0.03-4.0
Actinobacillus pleuropneumoniae	1998-1999	U.S.	111	≤0.03	≤0.03-0.25
Pasteurella multocida	1998-1999	U.S.	147	≤0.03	≤0.03-0.50
Streptococcus suis	1998-1999	U.S.	142	0.25	≤0.03-1.0
Actinobacillus pleuropneumoniae	2000	U.S.	126	≤0.03	≤0.03-0.06
Pasteurella multocida	2000	U.S.	173	≤0.03	≤0.03-0.06
Streptococcus suis	2000	U.S.	146	0.06	≤0.03-4.0
Actinobacillus pleuropneumoniae	2000-2001	U.S.	89	≤0.03	≤0.03-0.06
Pasteurella multocida	2000-2001	U.S.	186	≤0.03	≤0.03-0.12
Streptococcus suis	2000-2001	U.S./Canada	167	0.06	≤0.03-4.0

* The following *in vitro* data are available, but their clinical significance is unknown.
** Minimum inhibitory concentration (MIC) for 90% of the isolates.
*** No range; all isolates yielded the same value.

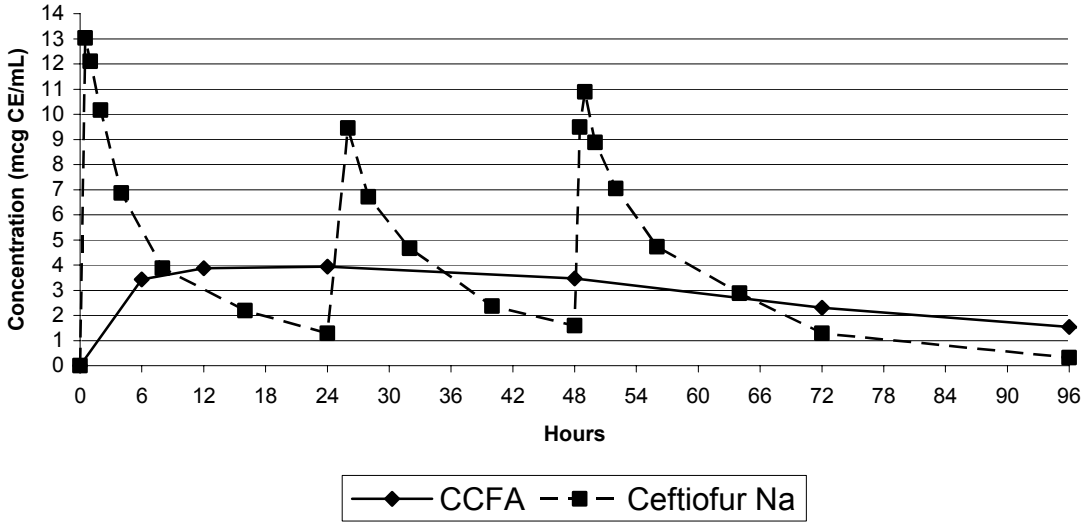
3. TARGET ANIMAL SAFETY:

The safety of ceftiofur sodium (NAXCEL Sterile Powder) was demonstrated in target animal safety and field effectiveness trials, which are discussed in the FOI Summary for NADA 140-338 dated August 4, 1992. A five-day tolerance study (TR 7220-89-002) in healthy feeder pigs indicated that ceftiofur sodium produced no overt adverse signs of toxicity and was well tolerated when administered at 125 mg CE/kg (57 mg CE/lb) BW (25 times the proposed dosage of CCFA) for five consecutive days. In an additional dose toxicity study (TR 7220-89-041), ceftiofur sodium was administered intramuscularly at 0, 5, 15, and 25 mg CE/kg (0, 2.27, 6.81, and 11.36 mg CE/lb) BW (0, 1, 3, and 5 times the proposed dosage of CCFA) for 15 consecutive days. There were no adverse systemic effects observed, indicating that ceftiofur sodium has a wide margin of safety when administered intramuscularly in feeder pigs.

a. Confirmation of Systemic Safety

After parenteral administration, CCFA, ceftiofur sodium, and ceftiofur hydrochloride are metabolized to the same principal metabolite, desfuroylceftiofur. CCFA administered as a single IM injection of 5.0 mg CE/kg (2.27 mg CE/lb) BW provides peak plasma concentrations of desfuroylceftiofur and related metabolites not exceeding those from IM administration of ceftiofur sodium and ceftiofur hydrochloride (EXCENEL RTU Sterile Suspension) at their approved dosages of 3.0 to 5.0 mg CE/kg (1.36 to 2.27 mg CE/lb) BW (Figure 3.1). The total amount of ceftiofur administered by the single dose regimen of CCFA in swine (5.0 mg CE/kg BW) is less than the cumulative amount administered over the three-day treatment regimens (9.0 to 15.0 mg CE/kg BW) for the ceftiofur sodium and ceftiofur hydrochloride formulations currently approved in swine.

Figure 3.1: Comparison of 1X CCFA vs. 3X Ceftiofur Na given to Swine



Considering these points, the remaining information needed to justify an extrapolation of the existing ceftiofur sodium target animal safety data to CCFA, thereby fulfilling the requirement to demonstrate target animal safety, is as follows:

- A determination of whether a molar correction is needed for appropriate comparison of the administered dose.

  ANALYSIS: All doses of CCFA and ceftiofur sodium are expressed as ceftiofur equivalents. Accordingly, there is no need to correct for molar differences in amount of active moiety administered.

- Assurance that the absolute bioavailability of ceftiofur sodium (F) in swine is at least 100%/3 or 33.3%. If this is the case, then multiples of the 1X dose (administered for 3 sequential days) of ceftiofur equivalents, as used in the original TAS study, will adequately cover a 1X dose of CCFA.

  ANALYSIS: The absolute bioavailability of ceftiofur sodium in swine was determined in a study by Banting et al., (TR #796-7926-91-002 dated 2 April 1991: *Determination of Pharmacokinetic Parameters of Ceftiofur Given at 3 Mg/Kg to Pigs)*. In this investigation, the relative bioavailability of an intramuscular (IM) injection of ceftiofur sodium was determined (where F = absolute bioavailability = $AUC_{0\text{-}inf}$ IM / $AUC_{0\text{-}inf}$ IV). The IM route of administration is identical to the method of administration used in the original target animal safety study. The results of this pharmacokinetic study confirmed that ceftiofur sodium is nearly 100% bioavailable in swine. Based upon this investigation, we conclude that ceftiofur sodium is extremely well absorbed in swine and that the existing safety data adequately cover the extent of drug exposure anticipated with a single administration of CCFA.

- Assurance that the duration of drug exposure associated with the original TAS study is adequate to cover the prolonged exposure to ceftiofur and its metabolites associated with the release of CCFA from this new formulation.

  ANALYSIS: The supplemental approval for the use of NAXCEL Sterile Powder in swine (NADA 140-338, approved August 4, 1992) contains a safety evaluation conducted with three dosages of ceftiofur sodium (5, 15, and 25 mg CE/kg BW) administered as a once daily injection for fifteen consecutive days. Considering that the CCFA terminal elimination half-life in swine is estimated as ~49.6 hours, we conclude that the drug and its metabolites will be effectively eliminated from the body by 496 hours after administration (10 times the estimated terminal elimination half-life). In contrast, the estimated terminal elimination half-life for ceftiofur sodium in swine (IM injection) is 14.3 hours (Brown, et al., 1999, *J. Vet. Pharmacol. Therap*., 22:35-40). Accordingly, when dosed for 15 days, the drug will be effectively eliminated from the body within 503 hours following the first administration [(15 days x 24 hours) + (10 x 14.3 hours)]. Based on this information, we conclude that on the average, the duration of drug exposure during the ceftiofur sodium target animal effectiveness trial was as long or longer

than the duration of exposure anticipated with a single dose of CCFA. Therefore, from a duration of exposure perspective, the existing target animal safety information is adequate to cover effects that may be attributable to the duration of drug exposure following administration of CCFA.

Based upon our evaluation, we conclude that the systemic safety of CCFA in swine is adequately covered by the information contained within the target animal safety study originally submitted in support of the approval of ceftiofur sodium use in swine.

b. Injection Site Tolerance Study:

"PNU-64279: Intramuscular Injection Site Local Tolerance Study to Evaluate Ceftiofur Crystalline Free Acid Sterile Suspension (CCFA-SS) in Swine." Report No. a0103691.

1. Type of Study: Injection site tolerance study. The study was performed in accordance with Good Laboratory Practice (GLP) requirements.

2. Study Director: W.J. Seaman. Senior Veterinary Pathologist/Toxicologist, Pharmacia Corp., Kalamazoo, MI.

3. Study Design:

   a) *Objective:* To provide information on injection site tissue reaction and resolution following IM administration of the final formulation of CCFA sterile suspension (100 mg CE/mL) in the neck of mixed-breed swine at a dose volume of 2 mL/injection site.

   b) *Experimental Animals:* Six mixed-breed swine (3 castrated males and 3 females) were used. Animals weighed between 36.2 and 44.0 kg at injection. Each animal was randomized to two of three possible injection time points using a random draw. The two injection sites per animal were assigned to different time points.

   c) *Test Article Administration:* CCFA sterile suspension, (100 mg/mL) was used as the test article (GMP lot number 40773). Pigs received one IM injection of CCFA on each side of the neck at a standardized dose volume of 2 mL/injection site. Injections were given approximately 3-4 cm behind the bottom of the base of the ear, using a 1.5 inch 18-gauge needle. The injections were given on Day 1, 4, or 8 in order to observe injection sites at 3, 7, or 10 days post-injection.

   d) *Measurements and Observations:* Monitored variables included daily clinical evaluations and palpation of injection sites, periodic body weights, and postmortem gross evaluation of injection sites.

      All animals were necropsied on Day 11. Injection site areas were dissected, evaluated grossly, and photographed. Injection sites were scored by the Study Pathologist according to the following scale: "-" = No irritation or irritation

repaired with some hyperemia or some fibrotic tissue left (no edema); macroscopically injection site is hardly detectable; the injured area is < 6 cm^2; "±" = Slight irritation: fibrotic tissue with some edema or few hemorrhages or discoloration of connective tissue; almost completely repaired; the injured area is 6 to 30 cm^2; "+" = Moderate irritation: fibrotic tissue, edema, hyperemia or hemorrhages, some muscle degeneration; area of irritation is obvious and detectable; the injured area is > 30 cm^2; or "++" = Severe irritation: extensive discoloration of connective tissue, hemorrhages, edema, muscle degeneration, or necrosis; the injured area is > 30 cm^2.

4. Results:

   a) *Clinical Observations*: No animals died on study. Injection site reactions ranged from nondetectable (6 of 12 injection sites) to a transitory (up to 4 days post-injection) palpable, nonvisible swelling (2 of 12 injection sites) or a small, visible, reddened nodule (4 of 12 injection sites) at the needle-insertion point first noted at one day post-injection. Nodules ranged in size from 0.25 cm^2 to 0.60 cm^2 at one day post-injection and diminished in size and redness until they were non-detectable or the animal was necropsied. There was no clinical evidence of injections at 10 days post-injection.

   b) *Body Weight*: There was no adverse effect on body weight gains attributed to dosing with CCFA.

   c) *Gross Injection Site Observations*: The muscle layer of all the injection sites at 3 and 7 days post-injection had a small area (ranging in size from 0.5 cm^2 to 27 cm^2) of tan discoloration in the muscle or fascia separating muscle bundles (one site at 3 days post-injection had a dark red area). The affected tan area of the muscle in all the sites at 3 days post-injection and in one site at 7 days post-injection also contained red speckles or red foci. About half the affected muscle areas at 3 and 7 days post-injection had very small cysts, and exuded tan or clear oily material from the cut surface. Gross changes were detected in the muscle layer of only 1 of 4 sites at 10 days post-injection (a 6 cm^2 area of tan with red specks in the muscle fascia). Two of four injection sites examined 3 and 7 days after injection were scored as "-" (no irritation), while the other two injection sites for these time periods were scored as "±" (slight irritation). All four injection sites examined 10 days after injection were scored as "-" (no irritation).

5. Conclusions: IM injections of 2 mL CCFA sterile suspension (100 mg CE/mL) per injection site were well tolerated in all pigs. Administration of CCFA may induce a transient reaction at the site of injection and underlying tissues that may result in loss of edible tissues at slaughter.

c. Corroborative Study:

"PNU-64279: Intramuscular Injection Site Local Tolerance Study in Swine." Report No. a0084154.

1. Type of Study: Injection site tolerance study. The study was performed in accordance with GLP requirements.

2. Study Director: W. J. Seaman, Senior Veterinary Pathologist/Toxicologist, Pharmacia Corp., Kalamazoo, MI.

3. Study Design:

   a) *Objective*: To examine the nature and resolution of tissue reaction at the injection site following the IM administration of a single injection of the final formulation of CCFA sterile suspension at a dose level of 5.0 mg CE/kg BW into each side of the neck of swine.

   b) *Experimental Animals*: Eight mixed-breed swine (4 castrated males and 4 females) were used. Animals weighed between 53.1 and 103.8 kg at injection. Each animal was randomized to two different injection time points using a random draw.

   c) *Test Article Administration*: Single dose injections were administered to four different animals on Study Days 1, 15, 29, and 36 so that four injection sites were evaluated for each time point. Each pig received a single IM injection of CCFA sterile suspension into each side of the neck. Injection volume was based on body weight and ranged from 2.7 to 5.2 mL. Animals were killed and the neck injection sites were dissected at 21, 28, 42, and 56 days post-injection (4 injection sites/time point).

   d) *Measurements and Observations*: Monitored variables included daily clinical observations, clinical evaluation and palpation of injection sites, weekly body weights, and postmortem gross and microscopic evaluation of injection sites. Clinical and gross evaluation of the injection sites were conducted as in the previously described study (Study Report a0103691).

4. Results:

   a) *Clinical Observations*: No animals died on study. One animal had palpable mild swelling at the injection site for 6 days post-injection. There were no other clinical observations attributed to injection with CCFA.

   b) *Body Weight*: There was no adverse effect on body weight gains attributed to dosing with CCFA.

   c) *Gross Injection Site Observations*: Injection of CCFA was either undetectable or associated with a single small area (less than 6 cm^2) of tan discoloration in the fascia separating muscle bundles. All injection sites examined were

scored as "-" (no irritation). Areas of discoloration usually contained very small cystic cavities (most cysts were approximately 1 mm) that expressed a clear, oily material from the cavity when sectioned. This reaction was observed in 3 of 4 sites at 21 days post-injection, 2 of 4 sites at 28 days post-injection, and 3 of 4 sites at 42 days post-injection. No gross changes were observed in the 4 sites at 56 days post-injection. No changes in the skin or subcutis were detected in any of the sites.

d) *Histopathological Observations*: The injection sites that appeared normal grossly were also normal microscopically. The injection sites which grossly had tan discoloration and small cystic cavities in the fascia had similar microscopic findings. Microscopically, the cystic cavities were generally empty (the clear, oily contents had probably been extracted during tissue processing) with thick walls composed of macrophages, foreign body giant cells, lymphocytes, and occasional eosinophils, and were classified as cystic foreign body granulomas. Occasionally nodules of similar composition but without the cavity were observed. Dense fibrous connective tissue separated the cysts. Adjacent muscle fibers generally appeared normal, although a few appeared shrunken and occasionally the muscle contained cystic foreign body granulomas.

5. Conclusions: CCFA, administered as a single IM injection of 5.0 mg CE/kg BW per injection site, was well tolerated in all pigs. The study demonstrates that administration of CCFA may induce a transient reaction at the site of injection and underlying tissues that may result in loss of edible tissues at slaughter.

d. Adverse Effects Observed in Multi-location Field Studies:

No adverse effects were observed in multi-location field studies involving more than 1000 pigs.

4. HUMAN FOOD SAFETY:

a. Toxicity

The toxicity testing of ceftiofur is summarized in the FOI Summaries for NAXCEL (ceftiofur sodium) Sterile Powder (NADA 140-338, approved January 25, 1988) and for the original approval of EXCENEL RTU (ceftiofur hydrochloride) Sterile Suspension (NADA 140-890, approved April 26, 1996) for use in swine. The current FOI Summary summarizes the additional studies required to assess the residue decline of EXCEDE For Swine (ceftiofur crystalline free acid) Sterile Suspension and establish the relevant tolerances and withdrawal period.

1. "Comparative Oral Bioavailability Study of Ceftiofur Sodium and Ceftiofur Crystalline Free Acid in Sprague-Dawley Rats." Report No. 829-7926-95-001.

a. Purpose: The objective of this GLP study was to assess the relative oral bioavailability of CCFA and ceftiofur sodium. Bioavailability was assessed by measuring the total ceftiofur residues in plasma, urine, feces, kidney, and liver after oral administration of either ^{14}C-CCFA or ^{14}C-ceftiofur sodium in rats when administered at an oral dose of 100 mg CE/kg BW.

b. Study Director: Gwendolyn D. Fate, Ph.D., Animal Health Drug Metabolism, Pharmacia & Upjohn, Kalamazoo, MI.

c. Study Design: A randomized complete block design was used to evaluate the plasma residues over time, urine and fecal excretion of total residues, and quantitation of total ceftiofur-related residues in kidney and liver 72 hours after drug administration.

1) *Test Animals:* Sprague-Dawley rats [CRL:CD(BD)]; 22 males and 22 females

2) *Dosage Form and Route of Administration:* Ceftiofur sodium and ceftiofur crystalline free acid, both labeled with ^{14}C at the same location in the aminothiazol ring, were administered orally dissolved or suspended, respectively, in sterile water.

3) *Dosage:* All rats were administered 100 mg CE/kg BW by oral gavage, with only one oral dose per test animal.

4) *Pertinent Parameters Measured:* Primary decision variables were the plasma concentration-time curve from zero to 72 hours (AUC_{0-72}), excretion of total ceftiofur residues in urine and feces through 72 hours, and total residue levels in liver and kidney at 72 hours post-treatment.

d. Results:

Table 4.1. Pharmacokinetic and Tissue Concentration Data from Rats Orally Dosed with Ceftiofur Sodium or Ceftiofur Crystalline Free Acid (100 mg CE/kg BW)

Value	CCFA (males)	CCFA (females)	Ceftiofur Na (males)	Ceftiofur Na (females)
C_{max} (μg/mL)	3.43 ± 1.31	1.86 ± 0.51	4.78 ± 1.76	2.19 ± 0.60
T_{max} (hours)	3.8	3.5	3.2	2.9
Kidney (μg/g)	0.47 ± 0.12	0.50 ± 0.11	0.18 ± 0.02	0.27 ± 0.04
Liver (μg/g)	0.13 ± 0.02	0.15 ± 0.09	0.02 ± 0.01	0.06 ± 0.01
Urine excretion (μg)	542 ± 178	407 ± 93	549 ± 55	389 ± 43
AUC $_{0\text{-}72}$ (μg•h/mL)	44.44 ± 17.34	23.62 ± 5.09	49.53 ± 11.54	23.53 ± 3.89

Urine: Mean urinary excretion of ceftiofur in males was 542 ± 178 μg and 549 ± 55 μg for ceftiofur crystalline free acid and ceftiofur sodium, respectively. In females, mean urinary excretion was 407 ± 93 μg and 389 ± 43 μg for ceftiofur crystalline free acid and ceftiofur sodium, respectively.

Kidney: Ceftiofur residues in kidney were 0.47 ± 0.12 μg/g (males) and 0.50 ± 0.11 μg/g (females) for ceftiofur crystalline free acid, compared with 0.18 ± 0.02 μg/g (males) and 0.27 ± 0.04 μg/g (females) in ceftiofur sodium-treated rats. This difference is consistent with the slower absorption observed with ceftiofur crystalline free acid than with ceftiofur sodium.

Plasma: Plasma bioavailability (assessed as the pharmacokinetic parameter $AUC_{0\text{-}72}$) was comparable within males and females for ceftiofur sodium and ceftiofur crystalline free acid administered orally.

e. Conclusions: Given that the mean excretion into urine is quite consistent across ceftiofur sodium and ceftiofur crystalline free acid, the AUC data supports equal bioavailability and the kidney residue data are consistent with slower absorption of ceftiofur crystalline free acid than ceftiofur sodium. The data from this study support the conclusion that ceftiofur crystalline free acid and ceftiofur sodium are equally bioavailable when administered orally. The demonstration of equal oral bioavailability demonstrates that the results of toxicology testing conducted for ceftiofur sodium may be applied to the evaluation of ceftiofur crystalline free acid.

2. "Passive Cutaneous Anaphylaxis Study in Guinea Pigs." Technical Report No. 7263/87/077

a. Purpose: Studies conducted in the guinea pig model of ceftiofur hypersensitivity and cross hypersensitivity between penicillin G and ceftiofur were previously submitted to NADA 140-338 (technical report number 7220-88-026). The results

of these studies were reassessed to provide the basis of an acute single daily intake for residues of ceftiofur crystalline free acid at the injection site.

b. Study Director: Terry A. Jackson, Worldwide Toxicology, Pharmacia & Upjohn, Kalamazoo, MI.

c. Identification of Substance and Dosage Form: Ceftiofur (U-64,279E) was tested in four forms:

1) Parent compound (bulk drug)
2) Parent compound conjugated to hen egg albumin as the carrier protein
3) Metabolite II
4) Extract of residue from kidney and injection site muscle of treated animals

In addition to the four forms of ceftiofur, two additional conjugates with hen egg albumin were tested:

1) Compound A - structurally similar to Metabolite VII
2) Compound B - common to parent compound and all metabolites

d. Species and Strain: Hartley Albino Guinea Pigs

e. Number of Animals Per Sex Per Treatment Group: Females only, variable number per group (2 - 12), depending on challenge material and level

f. Route of Drug Administration: IV and Oral

g. Drug Levels Tested and Duration of Dosing: Single challenge doses were given to animals passively sensitized with antibody to benzylpenicillin G and/or ceftiofur. The challenge levels tested were based on anticipated maximum human exposure level (approximated 0.083 mg/kg), varied with the form of the drug and were usually given in 10-fold increments. Routinely, oral challenges were at levels 10X greater than those given IV:

Parent compound (bulk drug):	IV: 0.076, 0.76, 7.6 mg/kg Oral: 0.76, 7.6, 76 mg/kg
Ceftiofur conjugate (ceftiofur to hen egg albumin):	IV: 0.01, 0.10, 1.0 mg/kg Oral: 0.10, 1.0, 10.0 mg/kg
Metabolite II:	IV: 0.076, 0.76, 7.6, 76, 760, 7600 µg/kg Oral: 0.76, 7.6, 76, 760, 7600 µg/kg
Compound A conjugate (to hen egg albumin):	IV: 0.0083, 0.083, 0.83 mg/kg (actual content of Compound A not determined)

Compound B conjugate (to hen egg albumin):	IV: 0.83 mg/kg (actual content of Compound B not determined)
Extract of drug residue from injection-site muscle and kidney:	Oral: 830 µg/kg

h. Parameters Tested: Passive Cutaneous Anaphylaxis (PCA) following IV and/or oral challenge. Briefly, skin sites on guinea pigs were passively sensitized with antibody of desired specificity and five days later the sensitized animals were given Evans blue dye and challenged by either the IV or oral route with appropriate test material. Sensitized skin sites were subsequently examined for evidence of PCA reactions as indicated by the leakage of Evans blue dye from the vascular system into the skin.

i. Significant Findings: Regardless of dose, there were no positive reactions of passive cutaneous anaphylaxis (PCA) when guinea pigs sensitized with penicillin antibody were challenged by either route with any form of ceftiofur.

Guinea pigs sensitized with ceftiofur antibody did not respond to challenges with the parent compound by either route. However, positive PCA reactions occurred following challenge with protein conjugate by both routes. While an IV challenge of 0.01 mg/kg produced positive reactions, oral exposure resulted in PCA activity only at a challenge level of 10 mg/kg.

Metabolite II caused PCA reactions over a wide range of dose levels when given by either the IV or oral route to guinea pigs sensitized with ceftiofur antibody. While as little as 0.076 µg/kg produced positive responses following IV challenge, levels of at least 76 µg/kg were required to induce positive PCA activity as a sequel to oral exposure.

Guinea pigs sensitized with ceftiofur antibody then challenged IV with conjugates of Compound A or Compound B responded to the challenge. These positive responses confirm that the model is able to recognize all metabolites in the extract of kidney or muscle.

Guinea pigs sensitized with ceftiofur antibody and challenged orally at a level of 0.83 mg/kg with acetic acid extract of drug (ceftiofur) residue from kidney and injection-site muscle were uniformly negative.

No Observed Effect Level - 0.83 mg/kg. The relevant NOEL is for oral challenge of sensitized animals with the acetic acid extract of ceftiofur residue from kidney and injection site muscle.

j. Statistical Analysis: Not applicable.

k. Conclusions: Antibody to benzylpenicillin G did not react with any form of ceftiofur tested, regardless of route or dose. Data from the IV challenges indicate that ceftiofur antibody used for this study was capable of detecting the drug as a

conjugated hapten in challenge material given at a level as low as 0.01 mg/kg body weight (lowest level tested). Positive PCA reactions also occurred with IV challenges of Metabolite II at levels as low as 0.076 µg/kg body weight (lowest level tested). The route was of considerable importance for both the protein conjugate and Metabolite II because of the differences in the level of challenge necessary to elicit a positive PCA reaction. With both materials, the oral challenge level necessary to produce a positive response was approximately 1,000X the minimum level required for a PCA reaction following IV challenge. This suggests that the gastrointestinal tract may play an important role in modulating the effect of potential hypersensitive reactions with ceftiofur.

Further, the data suggest that ceftiofur does not exist in residue from injection site muscle or kidney in a form or concentration such that PCA reactions occur following oral exposure of animals sensitized with ceftiofur antibody and challenged with extract of the residue. The NOEL derived from oral challenges of injection site muscle extract (IME) is 0.83 mg/kg BW. Of all the challenge materials tested in this study, IME is considered the most appropriate one to use for evaluating acute exposure. A safety factor of 1 was used in the calculation of an ASDI for residues of ceftiofur at the injection site (see item 5 (**Determination of an ADI and an ASDI**) below).

3. Microbial Safety

The potential for residues of CCFA to affect the microflora of the human gut was evaluated. The Agency has concluded that the amount of microbiologically active residues of CCFA that reach the colon would most likely not cause adverse effects on the human intestinal microflora of the consumer.

4. Determination of No Observed Effect Level (NOEL)

The lowest No Observed Effect Level (NOEL) for chronic exposure was determined from the 90-day feeding studies in both dogs and rats: 30 mg/kg body weight.

The No Observed Effect Level (NOEL) for acute exposure was determined from challenges of injection site muscle extract (IME): 0.830 mg/kg body weight.

5. Determination of an ADI and an ASDI

The ADIs for chronic exposure are:

0.008 mg/kg BW/day for milk and
0.022 mg/kg BW/day for edible tissues.

The NOEL from the IME challenge materials is considered the most appropriate NOEL to use for the basis of the ASDI. A safety factor of 1 was used in the calculation of an ASDI for residues of ceftiofur at the injection site:

ASDI = 0.830 mg/kg BW/1
= 0.830 mg/kg BW

6. Calculation of the Safe Concentrations

The calculation of the safe concentrations for total residues in edible tissues resulting from chronic exposure is summarized in the FOI Summaries for NADA 140-338 and NADA 140-890.

The calculation of the safe concentration for total residues in injection site tissues resulting from acute exposure is:

Safe Concentration in the injection site = (0.830 mg/kg BW X 60 kg)/0.300 kg* = 166 mg/kg or 166 ppm

*The consumption factor for muscle is used in this calculation

b. Residue Chemistry

The total residue depletion and metabolism in the target species and comparative metabolism in the toxicological species for ceftiofur are summarized in the FOI Summaries under NADA 140-338 and NADA 140-890. The marker residue in edible tissues is the sum of ceftiofur and desfuroylceftiofur-related metabolites, measured by HPLC as the stable derivative desfuroylceftiofur acetamide (DCA). The target tissue for residue monitoring is kidney. The following pivotal studies were conducted to permit decisions on tolerances and the withdrawal period.

1. "The determination of kidney and injection site ceftiofur and desfuroylceftiofur-related residue concentrations in swine at 7 and 10 days after administration of a suspension of ^{14}C-ceftiofur free acid (PNU-064279) at 100 mg/mL by intramuscular injection in the neck at a dose of 5 mg ^{14}C-ceftiofur equivalents/kg body weight." Report No. a0104682.

a. Purpose: This study was designed to determine the relationship between total residue and the residue as measured by the regulatory method, the HPLC-DCA assay, in swine at 7 and 10 days after treatment with 5.0 mg ^{14}C-CCFA/kg BW.

b. Study Director: John L. Nappier, Ph.D., Animal Health Drug Metabolism, Unit 7926, Pharmacia & Upjohn, Kalamazoo, MI.

c. Study Location: The animal phase of this study and analysis of the tissue samples for total ^{14}C-residues of ceftiofur took place at Animal Health Drug Metabolism Laboratories, Pharmacia & Upjohn Research Farm, Richland, MI. The HPLC-DCA assays were done in the laboratories of Pharmacia Animal Health Preclinical Development, Building 300, Kalamazoo, MI.

d. Study Design:

1) *Test Animals:* Twelve pigs (6 male and 6 female), 26.8 to 31.0 kg BW, were treated with ^{14}C-CCFA sterile suspension.

2) *Dosage Form and Route of Administration:* Sterile suspension for injection administered intramuscularly in the neck.

3) *Dosage*: Each animal received a single IM dose of about 0.15 mCi of ^{14}C-CCFA at a nominal dose rate of 5.0 mg ceftiofur equivalents (CE) per kg BW.

4) *Pertinent Parameters Measured*: The injection site and kidney were collected for DCA and total combustion analyses at 7 days and 10 days after injection.

e. Results: In injection site, the HPLC-DCA assay gave mean results that were 57.5 ± 6.4% and 51.8 ± 10.9% of the results as determined by total combustion analysis for the 7-day and 10-day slaughter groups, respectively.

f. Conclusions: At 7 and 10-day post-dosing, the ratio of DCA:total residue approximates to 0.575 and 0.518, respectively. An extrapolation to a ratio of 0.484 is considered appropriate for the designated withdrawal period.

2. "Residue decline (3, 5, 7, 10 and 12 days post-treatment) of ceftiofur and desfuroylceftiofur-related residues in the injection site and kidneys of swine administered a sterile suspension of ceftiofur crystalline free acid (PNU-064279) at 100 mg ceftiofur equivalents (CE)/mL by intramuscular injection at a dose of 5 mg CE/kg body weight." Report No. a0104616.

a. Purpose: To determine the concentration of the marker residue for ceftiofur in kidneys and injection sites at various times after intramuscular administration of CCFA sterile suspension in swine at a dose of 5.0 mg CE/kg BW.

b. Study Director: John L. Nappier, Ph.D., Animal Health Drug Metabolism, Pharmacia & Upjohn, Kalamazoo, MI.

c. Study Location: The animal phase of this study took place at Animal Health Drug Metabolism Laboratories, Pharmacia & Upjohn Research Farm, Richland, MI. The HPLC-DCA assays were done in the laboratories of Pharmacia Animal Health Preclinical Development, Building 300, Kalamazoo, MI.

d. Study Design:

1) *Test Animals:* Thirty Yorkshire/mixed breed pigs, 15 castrated male and 15 females (plus 1 castrated male control and 1 female control), 87.0 to 104.8 kg in weight.

2) *Dosage Form and Route of Administration:* Sterile suspension for injection (GMP lot 40,784) administered intramuscularly in the neck.

3) *Dosage*: Each animal received a single IM dose of 5.0 mg CE/kg BW; total dose was split such that no injection volume was larger than 2 mL (pigs received two 2-mL injection volumes, one on each side of the neck, and the remainder of the dose in the ham).

4) *Pertinent Parameters Measured*: Groups of six animals were sacrificed at 3, 5, 7, 10, and 12 days after injection. The injection sites and kidneys were

collected for analysis of desfuroylceftiofur-related residue by the regulatory HPLC-DCA assay.

e. Results: see Table 4.2 for injection site residue results.

Table 4.2. Tissue Residue Concentration of Ceftiofur and Desfuroylceftiofur-related Metabolites in Injection Sites

Slaughter group (days post-dosing)	Injection site residue results, means (range), μg/g
3	144 (64.6-281)
5	62 (14.5-175)
7	56 (12.5-121)
10	31 (7.66-92.5)
12	22 (3.98-79)

f. Conclusions: The mean for injection site residues was below the research value of 80 ppm DCA by 5 days withdrawal and no single value was above 80 ppm by 12 days withdrawal.

3. "Residue decline of ceftiofur in the injection site and edible tissue of swine administered a sterile suspension of ceftiofur crystalline free acid at 100 mg ceftiofur equivalents (CE)/mL by intramuscular injection at a dose of 5 mg CE/kg." Report No. 0829-7926-2002-004.

a. Purpose: To determine the concentration of ceftiofur and desfuroylceftiofur-related residues in injection site tissue and kidney tissue at various time intervals from 14 to 70 days following administration of CCFA sterile suspension at 5.0 mg CE/kg BW in swine.

b. Study Director: John L. Nappier, Ph.D., Animal Health Drug Metabolism, Pharmacia & Upjohn, Kalamazoo, MI.

c. Study Location: The animal phase of this study took place at Animal Health Drug Metabolism Laboratories, Pharmacia & Upjohn Research Farm, Richland, MI. The HPLC-DCA assays were done in the laboratories of Pharmacia Animal Health Preclinical Development, Building 300, Kalamazoo, MI.

d. Study Design:

1) *Test Animals:* Thirty Yorkshire/mixed breed pigs, 15 castrated male and 15 females (plus 1 castrated male control and 1 female control), 35.0 to 51.0 kg in weight.

2) *Dosage Form and Route of Administration:* Sterile suspension for injection (GMP lot 40,787) administered intramuscularly in the neck.

3) *Dosage*: Each animal received a single IM dose of 5.0 mg CE/kg BW in the neck.

4) *Pertinent Parameters Measured*: Groups of six animals were sacrificed at 14, 28, 42, 56, and 70 days after injection. The injection sites, kidneys, non-injection site muscle, fat, skin/fat and liver were collected for analysis of desfuroylceftiofur-related residue by the regulatory HPLC-DCA assay.

e. Results: The mean for injection site residues at 14 days was 24.4 μg DCA/g, with a range of 4.26 to 37.8 μg DCA/g. DCA was <50 μg/g in all tissues by 14 days.

f. Conclusions: At 14 days withdrawal, no single value for injection site residues exceeded half the research value of 80 ppm DCA.

4. "Determination of ceftiofur and desfuroylceftiofur-related metabolite concentrations in injection sites and kidneys of swine at 10 days after two 2-mL intramuscular injections of ceftiofur crystalline free acid suspension (PNU-064279, 100 mg/mL) in the neck using three experimental lots and one clinical supplies lot." Report No. a0105976.

a. Purpose: To evaluate injection site and kidney residues in swine at 10 days after intramuscular injection of three research lots and one clinical supply lot of CCFA sterile suspension at 5.0 mg CE/kg BW in swine.

b. Study Director: Dawn A. Merritt, Ph.D., Animal Health Drug Metabolism, Pharmacia & Upjohn, Kalamazoo, MI.

c. Study Location: The animal phase of this study took place at Animal Health Drug Metabolism Laboratories, Pharmacia & Upjohn Research Farm, Richland, MI. The HPLC-DCA assays were done in the laboratories of Pharmacia Animal Health Preclinical Development, Building 300, Kalamazoo, MI.

d. Study Design:

1) *Test Animals:* Twenty-four Yorkshire/mixed breed swine (68.2 to 90.0 kg) were assigned to four pens of six animals each (3 males and 3 females). Each pen of animals was treated with a different lot of CCFA sterile suspension.

2) *Dosage Form and Route of Administration:* Sterile suspension for injection administered intramuscularly in the neck. Three laboratory-scale research lots (Research Lots 32544-CCM-141-1, 32544-CCM-141-2 and 32544-CCM-141-3) and one clinical lot of CCFA sterile suspension were evaluated.

3) *Dosage*: All animals received a 2-mL injection in each side of the neck by IM injection (2 injections per animal). Animals weighing ≥85 kg received an additional IM injection of CCFA in the hindquarter/ham for a total dose of 5.0 mg CE/kg BW.

4) *Pertinent Parameters Measured*: The animals were sacrificed 10 days after injection. The two injection sites in the neck and the kidneys were collected at necropsy. These tissue samples were assayed for ceftiofur- and desfuroylceftiofur-related residues using the validated HPLC-DCA assay.

e. Results: See Table 4.3. for injection sites and kidney results. Only the data from lots within specification are reported (Research Lot 32544-CCM-141-3 was below specifications).

Table 4.3. Concentrations of Ceftiofur- and Desfuroylceftiofur-Related Residues in Kidneys at 10 Days After Administration of CCFA at 5.0 mg CE/kg BW

CCFA-SS Group (release specification)	Injection Sites, means (μg CE/g)	Kidney, means (μg CE/g)
Research lot 32544-CCM-141-1 (at lower limit)	47.6 ± 26.0	0.144 ± 0.062
Research lot 32544-CCM-141-2 (at lower limit)	68.5 ± 46.3	0.126 ± 0.048
Clinical lot 40,784 (middle)	35.3 ± 25.1	0.110 ± 0.054

f. Conclusions: The results support the assignment of a kidney tolerance of 0.25 ppm DCA.

5. Tolerances for the Marker Residue

The target tissue for residue monitoring is kidney. Based on the results in item 4.B.4., a tolerance of 0.25 ppm DCA in the kidney has been selected to take into account the sustained release characteristics of the CCFA sterile suspension formulation and the assigned withdrawal period.

In addition, tolerances of 3 ppm for liver and 2 ppm for non-injection site muscle have been established for DCA. Furthermore, for research purposes a value of 80 ppm DCA (i.e., 166 ppm x 0.484, the ratio of DCA:total residue as determined in item 4.B.1.) has been established for making decisions regarding the safety of the injection site.

A summary of approved safe concentrations and tolerances for ceftiofur in edible tissues of swine is presented in Table 4.4.

Table 4.4. Consumption Factors, Ceftiofur Safe Concentrations, and Tolerances for Edible Tissues in Swine

Tissue	Daily Consumption (g)	Safe Concentration (mg/kg)	Tolerance (mg/kg)
Muscle (non-injection)	300	4.40	2.0
Liver	100	13.2	3.0
Kidney	50	26.4	0.25

Fat	50	26.4	NE*

* NE – Not Established

6. Withdrawal Period

The injection site tissue determines the withdrawal time. Two residue depletion studies (items 4.B.2 and 4.B.3) have been conducted using CCFA sterile suspension at 5.0 mg CE/kg BW in the neck of swine. The results of these studies support the assignment of a 14-day withdrawal period.

c. Microbial Food Safety

The Agency used a qualitative risk assessment to evaluate the available microbial food safety information for ceftiofur crystalline free acid (CCFA). This risk assessment procedure involved conducting: 1) a release assessment to describe the probability of emergence of resistant bacteria in swine under proposed conditions of use; 2) an exposure assessment to describe the likelihood of human exposure to the resistant bacteria through consumption of edible products from treated animals (specifically, pork); and 3) a consequence assessment to describe the potential human health consequences of exposure to the defined resistant bacteria by considering the human medical importance of 3rd generation cephalosporins in the treatment of human infectious diseases.

It was determined that the risk associated with the use of this product is MEDIUM. An overall risk of MEDIUM is compatible with the proposed conditions of use for CCFA in swine, i.e. 5.0 mg of ceftiofur equivalents/kg body weight as a single injection for the treatment of bacterial respiratory disease in swine.

d. Regulatory Method for Residues

The regulatory method for determination of DCA in swine kidney and muscle, and bovine kidney, muscle, and milk is the HPLC-DCA assay which successfully completed a sponsor-monitored multi-laboratory method trial. The method is on file with the Center for Veterinary Medicine, Food and Drug Administration, 7500 Standish Place, Rockville, MD 20855.

5. USER SAFETY:

Studies to evaluate the safety of ceftiofur to users are discussed in detail in the FOI Summary for NADA 140-338 (NAXCEL Sterile Powder, ceftiofur sodium, approved January 25, 1988).

Human warnings are provided on the product labeling as follows:

For use in animals only. Not for human use. Keep out of reach of children. Restricted drug (California) - Use only as directed.

Penicillins and cephalosporins can cause allergic reactions in sensitized individuals. Topical exposures to such antimicrobials, including ceftiofur, may elicit mild to severe allergic reactions in some individuals. Repeated or prolonged exposure may lead to sensitization. Avoid direct contact of the product with the skin, eyes, mouth and clothing. Sensitization of the skin may be avoided by wearing latex gloves.

Persons with a known hypersensitivity to penicillin or cephalosporins should avoid exposure to this product.

In case of accidental eye exposure, flush with water for 15 minutes. In case of accidental skin exposure, wash with soap and water. Remove contaminated clothing. If allergic reaction occurs (e.g., skin rash, hives, difficult breathing), seek medical attention.

The material safety data sheet contains more detailed occupational safety information. To report adverse effects in users, to obtain more information or to obtain a material safety data sheet, call 1-800-253-8600.

6. AGENCY CONCLUSIONS:

The data submitted in support of this NADA satisfy the requirements of section 512 of the Federal Food, Drug, and Cosmetic Act and 21 CFR Part 514 of the implementing regulations. The data demonstrate that EXCEDE for Swine (ceftiofur crystalline free acid) sterile suspension, when administered as a single intramuscular injection in the post-auricular region of the neck, is safe and effective for the treatment of swine respiratory disease (SRD) associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Haemophilus parasuis*, and *Streptococcus suis*.

Labeling restricts this drug to use by or on order of a licensed veterinarian. This decision was based on the following factors: (a) adequate directions cannot be written to enable lay persons to appropriately diagnose and subsequently use this product to treat swine respiratory disease, (b) restricting this drug to use by or on order of a licensed veterinarian should help prevent indiscriminate use which could result in violative tissue residues, and (c) the rate of emergence of ceftiofur-resistant organisms may be reduced by the involvement of veterinarians in product use.

Under section 512(c)(2)(F)(ii) of the Federal Food, Drug, and Cosmetic Act, this approval qualifies for THREE years of marketing exclusivity beginning on the date of the approval.

The application contains investigations conducted or sponsored by the applicant that demonstrate animal safety and substantial evidence of effectiveness.

No patents were submitted with this application.

7. ATTACHMENTS:

Facsimile labeling is attached as indicated below.

A. EXCEDE for Swine - Vial Label
B. EXCEDE for Swine - Package Insert