SCIENTIFIC DISCUSSION

Name of the medicinal product: Draxxin

Marketing Authorisation Holder: Pfizer Ltd
Ramsgate Road,
Sandwich, Kent CT13 9NJ
UK

Active substance: Tulathromycin

ATC Vet Code QJ01FA94

Therapeutic indication(s):

Cattle
Treatment and prevention of bovine respiratory disease (BRD) associated with *Mannheimia haemolytica*, *Pasteurella multocida, Histophilus somni* and *Mycoplasma bovis* sensitive to tulathromycin. The presence of the disease in the herd should be established before preventative treatment.

Treatment of infectious bovine keratoconjunctivitis (IBK) associated with *Moraxella bovis* sensitive to tulathromycin .

Pigs
Treatment and prevention of swine respiratory disease (SRD) associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida, Mycoplasma hyopneumoniae* and *Haemophilus parasuis* sensitive to tulathromycin. The presence of the disease in the herd should be established before preventative treatment. Draxxin should only be used if pigs are expected to develop the disease within 2-3 days.

Target species Cattle and pigs

I INTRODUCTION

Draxxin solution for injection contains 100 mg/ml tulathromycin in a propylene glycol/water for injections vehicle and is packaged in 20, 50, 100, 250 and 500 ml vials. The active substance, tulathromycin, is a semi-synthetic macrolide antimicrobial agent whose proposed use is the treatment of respiratory infections in cattle and swine.

II QUALITY

Tulathromycin drug substance consists of two isomers with a ratio of CP-472,295 (Pfizer code) to CP-547,272 which typically exceeds 99:1. In tulathromycin drug product the ratio of CP-472,295 to CP-547,272 is 9:1.

II.A QUALITATIVE AND QUANTITATIVE PARTICULARS OF THE CONSTITUENTS

II.A.1 Composition of the Veterinary Medicinal Product

The product contains 100 mg/ml tulathromycin and 5 mg/ml monothioglycerol in a buffered solution. The active ingredient complies to Pfizer's in-house specification. All excipients are in compliance with Ph. Eur. and USP/NF quality standards.

II.A.2 Container

The product is packaged in 20, 50, 100, 250 and 500 ml colourless glass vials (Type 1) with 20 mm and 32 mm (250 and 500 ml presentations) respectively chlorobutyl rubber stoppers and aluminium tamper-proof push-off seals. The rubber stoppers are laminated with a Daikyo-Fluoro Resin-D (ETFE) film.

II.A.3 Clinical Trial Formula(e)

The clinical trial formulation is the same as for the final product. Batches used for efficacy testing, drug safety evaluation and microbiological testing have been identified in the dossier. Any differences to the final composition are deemed to have negligible impact.

II.A.4 Development Pharmaceutics

The drug product is a ready-to-use, multi-dose sterile solution of tulathromycin at a concentration of 100 mg/ml, which is the total concentration of the tulathromycin isomers.

The exceptional feature of tulathromycin injectable solution is the usage of an equilibrated mixture of isomers. During the formulation process, it was recognised that CP-472,295 equilibrates with CP-547,272 as well in aqueous as in partially aqueous vehicles. Therefore, the decision was made to develop a drug product with equilibrated isomers so that the ratio between them would be fixed throughout the shelf life of the product. This allowed a consistent product to be evaluated in stability, safety and clinical studies.

A rationale has been provided regarding the function of each of the excipients.

The formulation contains propylene glycol and water for injection, buffered to a target pH of 5.4 $\pm$ 0.3.

The use of monothioglycerol at a level of 5.0 mg/ml as antioxidant has been justified. As an additional precautionary measure the vial headspace is flushed with nitrogen.

It has been shown that the proposed commercial formulation is self-preserving; as formulated, it meets Ph. Eur. Criteria A for antimicrobial efficacy.

The photostability of tulathromycin in solution was investigated under light conditions in accordance with VICH guidelines. No degradation was noted at pH 5.5 through 7.7. Therefore, it is reasonable to use clear type 1 flint glass containers. The vials are sterilised and depyrogenated prior to filling by dry heat.

Stoppers are sterilised by autoclaving. No leaching could be detected when the stoppers were evaluated for extractables by the USP method.

It has been shown that terminal sterilisation with moist heat is unacceptable due to the increase in total degradant level it produces. Moving down the Decision Tree for the Selection of Sterilisation Methods (CVMP/065/99) then a combination of aseptic filtration and aseptic processing has to be used. The proposed filters were found acceptable for bacterial retention and compatibility with the drug product.

II.B METHOD OF PREPARATION

II.B.1 Manufacturing Formula and Batch Size

The manufacturing formula is given for two proposed commercial batch sizes.

II.B.2 Manufacturing Process and In-process Controls

Stoppers are sterilised by moist heat (121 °C/30 min). Empty vials are sterilised and depyrogenated prior to filling in a dry heat sterilising tunnel under specified and validated conditions. Minimum times for all vial sizes were specified.

Conventional pharmaceutical equipment and procedures are used in the manufacture of the drug product. Stainless steel or glass-lined compounding tanks will be used. Nitrogen is used to provide an inert atmosphere because monothioglycerol is sensitive to oxygen.

During validation of the manufacturing process analytical controls ensure that the isomers are equilibrated prior to proceeding to the next step. Once sufficient experience is gained this testing will no longer be required.

The bioburden of the excipients is controlled at a level of 10 cfu/100 ml for the pre-filtered bulk solution and batch data has shown that the active substance has a consistently low bioburden.

Samples for in-process controls (assay, monothioglycerol content, pH, bioburden) are taken. The solution is filtered (0.2 micron) into a sterilised receiving tank before being filled into sterilised vials. After the insertion of sterilised stoppers the vials are sealed and samples for release testing are taken.

II.B.3 Validation of Manufacturing Process

The following manufacturing steps have been identified as critical: dissolution of the active ingredient, equilibration of the tulathromycin isomers, pH adjustment and sterilisation procedure.

The suitability of the method of manufacture is further confirmed by batch results. They show that the manufacturing process leads to the production of a consistent drug product.

The intended site for commercial manufacture, packaging and batch release is in Europe. Scale-up lots were made at a manufacturing site in the USA. These lots were used for the VICH stability programme. Three drug product lots of at least 10% of the maximum scale envisaged at the European facility were compounded. Each lot was filled into 20, 100 and 500 ml vials.

To date, the Applicant has not manufactured production scale batches at the proposed commercial manufacturing site. Experience gained during manufacture of the VICH lots at the US facility provides a satisfactory basis for transfer of the product to the commercial manufacturing site in France. The Applicant has committed to a validation programme comprising three consecutive lots at production scale manufactured at the proposed commercial manufacturing site and to validate the filling process on each vial size. The process validation scheme of the manufacturing process that will be conducted on three production scale batches at the French site was submitted. The product is manufactured using standard manufacturing and sterilising processes and there are appropriate controls in place to monitor the critical processes. The evaluation of the Follow Up Measure will specifically seek to verify that scale-up does not affect the reproducibility of the process. The Applicant additionally committed to reporting any out of specification results.

II.C CONTROL OF STARTING MATERIALS

II.C.1 Active Substance

The Applicant agreed to revise the tulathromycin assay limits (94.0 – 103.0%) and the limits for total impurities (6.0% maximum) in tulathromycin based on batch analysis data as it becomes available. The Applicant has committed to provide a timetable for this within 3 months of the Opinion.

II.C.1.1 Specification and routine tests

1.1.1 Active ingredients listed in a Pharmacopoeia.

Not applicable.

1.1.2 Active ingredients not listed in a Pharmacopoeia.

Tulathromycin is a macrolide (triamilide) antibiotic that consists of two isomers, CP-472,295 and CP-547,272. Tulathromycin drug substance typically contains < 1% CP-547,272, whereas tulathromycin drug product contains 8 to 13% CP-547,272. The equilibrated mixture of the two structural isomers, CP-472,295 and CP-547,272, is also referred to as CP-472,295(e).

The specification of the active substance contains tests for appearance, identity, assay and purity. The routine tests and specification limits defined in the Applicant's testing instructions show conformity to pharmacopoeial standards and are considered sufficient to assure constant quality of the drug substance.

Specifications for individual residual solvents are in agreement with the CVMP VICH guideline 18 "Impurities: residual solvents in new veterinary medicinal products, active substances and excipients". Clarification was provided that the solvent used in the final purification step is a mixture of heptane isomers. The specified limit of heptanes at 0.5 % in tulathromycin active substance is considered qualified.

Limits for specified and unspecified impurities are justified by batch history and safety considerations. A qualification limit for impurities of 0.5 % has been retained though tulathromycin is a semisynthetic drug and is therefore not covered by the CVMP VICH guideline on impurities.

Absence of bioburden testing has been justified by:

- antimicrobial nature of solvents used in the terminal steps of the drug substance manufacturing process,
- hostile nature of the drug product to micro-organisms,
- historical data demonstrating consistently low bioburden levels, and
- routine performance of bioburden and bacterial endotoxin testing on each drug product batch.

Historical batch data provided to the Committee confirmed the microbial purity (both aerobic plate count and combined yeast and mould count were < 1 cfu/g) of tulathromycin active substance.

All testing methods have been described and validated where required. An isocratic HPLC method with UV detection at 205 nm has been described for identity, assay and purity testing. Two sets of validation data have been provided for assay and determination of impurities. Validation has been performed according to the requirements of the VICH guidelines. Validation data was provided for the determination of Limit of Detection (LOD) and Limit of Quantification (LOQ) respectively of each specified impurity in tulathromycin active substance.

The method for testing of residue on ignition can be regarded as equivalent to the Ph. Eur. method for sulfated ash. The test for the determination of heavy metals used in the dossier was shown to be equivalent to the Ph. Eur. method after testing three lots.

1.1.3. Physico-Chemical Characteristics liable to affect bioavailability

Since tulathromycin is formulated as a ready-to-use injectable solution, physicochemical characteristics of the drug substance are not expected to have an impact on bioavailability.

II.C.1.2 Scientific data

1.2.1. Nomenclature

INN: tulathromycin

Chemical Name of **CP-472,295**: (2R,3S,4R,5R,8R,10R,11R,12S,13S,14R)-13-[[2,6-dideoxy-3-C-methyl-3-O-methyl-4-C-[(propylamino)methyl]-α-L-ribo-hexopyranosyl]oxy-] 2-ethyl-3,4,10-trihydroxy-3,5,8,10,12,14-hexamethyl-11-[[3,4,6-trideoxy-3-(dimethylamino)-β-D-xylo-hexopyranosyl]oxy]-1-oxa-6-azacyclopentadecan-15-one

Chemical Abstracts Services Number of CP-472,295: 217500-96-4

Chemical Name of **CP-547,272**: (2*R*,3*R*,6*R*,8*R*,9*R*,10*S*,11*S*,12*R*)-11-[[2,6-dideoxy-3-*C*-methyl-3-*O*-methyl-4-*C*-[(propylamino)methyl]-□-*L-ribo*-hexopyranosyl[oxy]-2-[(1*R,*2*R*)-1,2-dihydroxy-1-methylbutyl]-8-hydroxy-3,6,8,10,12-pentamethyl-9-[[3,4,6-trideoxy-3-(dimethylamino)-□-*D-xylo*-hexopyranosyl]oxy]-1-oxa-4-azacyclotridecan-13-one

Chemical Abstracts Services Number of CP-547,272: 280755-12-6

Proposed Proprietary or Invented Name: Draxxin

USAN , WHO or BNF: tulathromycin

Pfizer Laboratory Codes: CP-472,295
CP-547,272
CP-472,295(e)
triamilide

1.2.2. Description

Physical form: White to off-white powder, essentially free from visible contaminants
Molecular Formula: $C_{41}H_{79}N_3O_{12}$
Molecular Weight: 806.23
Stereochemistry: CP-472,295 is synthesised from erythromycin A. The chirality of the macrocycle and the desosamine sugar is unaffected by the synthetic steps.

1.2.3. Manufacture

Tulathromycin drug substance is manufactured, tested and released by Pfizer Inc, Eastern Point Road, Groton, Connecticut 06340,USA

The manufacture of tulathromycin drug substance involves 4 steps, starting from CP-449,197-51. Full details of manufacture and control (including in process tests) were provided and deemed satisfactory.

The starting material CP-449,197-51 is manufactured from CP-60,273 which is derived from erythromycin A. CP-60,273 can be synthesised by either Pfizer, Inc or Pliva Pharmaceutical Industry, Inc.

The Applicant committed to provide confirmation that both manufacturers of CP-60,273 will use the same synthetic route, the same quality control procedures, the same specifications and the same supplier of the erythromycin A.

Batches of tulathromycin active substance produced to date were at least 10% of the proposed commercial batch size so the Applicant agreed to verify that scale-up will not affect the reproducibility of the process and to provide any out of specification results along with proposed action.

1.2.4 Quality control during manufacture

For the starting material, CP-449,197-51, a comprehensive specification (appearance, identity, assay, purity) has been provided. For step 2 and step 3 intermediates, limits for assay and purity (solvents, related substances) have been specified. All test methods have been described and validated where required.

Impurity specifications have been justified by batch history and by demonstrating the fate of impurities during synthesis in form of an "Impurity Control Grid". Determination of the chemical fate of an impurity in the reaction or subsequent processing steps shows that the specified impurity limits for intermediates are suited to make sure that the specifications for tulathromycin drug substance will be met.

For all reagents, solvents and auxiliary materials used during synthesis, routine tests and specifications have been provided.

The residual solvents to be controlled in the specifications of CP-449,197-51 provided was justified by the Applicant. Methylene chloride and isopropyl alcohol are the solvents used to manufacture CP-449,197-51. Isopropyl alcohol is specified at 1.5 % maximum. Methylene chloride is not specified as the level was consistently below the limit of detection (0.1 %). The Applicant committed to provide infomation on methanol residues in the starting material CP-449,197-51.

1.2.5. Development Chemistry

Evidence of structure:
Data have been provided for the two isomers of tulathromycin, CP-472,295 and CP-547,272.

Characterisation and proof of structure of **CP-472,295**:
The structure of CP-472,295 reference standard, lot 44145-84-3, was determined by mass spectrometry (MS), infrared (IR) spectroscopy and ^{1}H and ^{13}C nuclear magnetic resonance (NMR) spectroscopy. All of the spectroscopic data were consistent with the proposed structure.
HPLC/positive ion electrospray MS was used to confirm some structurally significant fragment ions. Inter-atomic connectivities were identified using multiple nuclear magnetic resonance spectroscopy (NMR) analyses. No suitable crystals of the anhydrous form could be obtained, but a single crystal X-ray was obtained for the monohydrate form, confirming the structure of CP-472,295. The sample was also characterised by elemental analysis.

Characterisation and proof of structure of **CP-547,272**:
The isolation of high purity samples of CP-547,272 is a technical challenge because it equilibrates to CP-472,295 in solution. While conclusive assignment of the structure of CP-547,272 is presented, complete characterisation of its physicochemical properties has not been performed.

The structure of CP-547,272 was determined by ^{1}H and ^{13}C NMR, mass spectrometry (MS) and infrared (IR) spectroscopy. All of the spectroscopic data were consistent with the proposed structure.

HPLC/positive ion electrospray MS was used to confirm some structurally significant fragment ions. Inter-atomic connectivities were identified using NMR. The sample was also characterised by ultraviolet (UV) absorption spectroscopy and elemental analysis.

Physicochemical properties of CP-472,295:
The physico-chemical properties characterised are solubility, ionisation constants (pKa), melting point, and hygroscopicity.

CP-472,295 has been found to exist in anhydrous, monohydrate and sesquihydrate crystalline forms, each of which was found as a single form in polymorph screening experiments. The commercial process of tulathromycin synthesis consistently produces the anhydrous form (water content below 0.5%).

Reference standard:
Complete analytical test results according to the drug substance specification are presented. Additional tests have been performed (DSC, TGA, MP, Elemental analysis, ^{1}H and ^{13}C NMR, Mass spectroscopy, X-Ray powder diffraction), confirming the proposed structure. Potency has been calculated by subtracting impurities, residual solvents and residue on ignition from 100 %.

1.2.6. Impurities

Since tulathromycin is a semi-synthetic drug substance, it is beyond the scope of the CVMP VICH guideline 18 "Impurities: residual solvents in new veterinary medicinal products, active substances and excipients". Nevertheless, the Applicant has adopted impurity thresholds that parallel the CVMP VICH impurity guideline for synthetic drug substances, which are report at 0.1%, identify at 0.2%, and qualify at 0.5%. The Applicant proposes to retain the 0.5% level for qualification, but use levels of 0.2% to report and 0.5% to identify impurities. Due to the complex nature of the drug substance, these limits can be regarded as acceptable.

Four individual process-related impurities are specified for tulathromycin drug substance. Individual, unspecified impurities are limited to 0.5% and the sum of individual, unspecified impurities is limited to 3.0%. The total level of impurities is limited to 6.0%. It should be noted that the majority of unspecified impurities have been identified. They are similar to CP-472,295 in structure, containing the intact macrolide ring system.

1.2.7. Batch analysis

Batch analysis data for 20 lots of tulathromycin drug substance manufactured using the commercial process have been provided. All batches were produced by Pfizer, Croton. Additionally, data for 4 developmental lots used in clinical and toxicological studies are included in the dossier. Testing results show that the drug substance can be manufactured to an adequate and constant quality.

II.C.2 Excipients

C.2.1 Specifications and routine tests

2.1.1. Excipients described in a Pharmacopoeia

All excipients are of compendial grade (Ph. Eur. or USP/NF). Monothioglycerol is tested according to NF specifications since a corresponding Ph. Eur. monograph does not exist. For all excipients, certificates of analyses have been provided. Possible microbiological contamination of the excipients used in the manufacture of this sterile product was discussed. However, as each batch of bulk solution is tested for bioburden and must meet the specification of "not more that 10 cfu/100 ml", this ensures that minimal bioburden is present in the final solution prior to sterilising filtration.

II.C.3 Packaging Material (Immediate Packaging)

The 20, 50, 100, 250, and 500 ml Type I flint glass vials and the chlorobutyl stoppers comply with Ph. Eur. requirements. Revised drawings of the overseals were provided.

II.D CONTROL TESTS ON INTERMEDIATE PRODUCTS

Not applicable.

II E CONTROL TESTS ON FINISHED PRODUCT

II.E.1 Product Specification and Routine Tests

1.1. Product specifications and tests for release at time of manufacture (general characteristics, specific standard)

The finished product specification contains tests/limits for appearance, particulate contamination (visible particles), identity, assay and isomer ratio of tulathromycin, degradation products, content of monothioglycerol, pH, sterility, bacterial endotoxins and extractable volume.

The specification of active substance content was tightened to 95 % - 105 % of label claim at time of release. Limits for specified degradation products and for total degradation products were tightened as well.

The specifications and routine tests are regarded as sufficient to ensure adequate and consistent quality of the finished product.

1.2. Control methods

1.2.1 Tests procedure for identification and quantitative determination for the active subtance(s)

Positive identification of tulathromycin is confirmed using reversed phase HPLC and TLC.

Potency is determined by an isocratic reversed phase HPLC procedure with UV detection which is stability indicating. The method separates the two isomers of tulathromycin from potential degradants, drug substance impurities and the formulation excipients. Ultraviolet detection is at 205 nm to achieve the necessary sensitivity given that tulathromycin lacks a strongly absorbing UV chromophore.

1.2.2 Identification and determination of excipient(s)

Monothioglycerol content is determined by iodometric titration. The release specification level is 90 to 110% of label claim and the end-of-shelf-life specification is 50% minimum to allow for losses due to reaction with residual headspace oxygen and to provide adequate protection during the in-use period.

1.2.3 Safety tests

Impurities: The dossier provides reasonable explanations for setting the specifications of specified, unspecified and total degradation products. Limits are in line with current VICH recommendations and are supported by batch history.

Sterility: The membrane filtration method is used to determine sterility of the finished product. Testing is validated according to pharmacopoeial requirements.

Bacterial endotoxins: The level of bacterial endotoxins is demonstrated using a Ph. Eur. test. The specification is set at 200 EU/ml.

II.E.2 Scientific Data

2.1. Analytical validation of methods and comments on the choice of routine tests and standards

Identity, Assay, Purity and isomer ratio by RP-HPLC: The method was validated in the presence of excipients and related substances. Specificity, linearity, accuracy (recovery and range), precision (repeatability) and ruggedness of the method have been demonstrated. The criteria satisfy the requirements of the corresponding CVMP VICH Guidelines (EMEA/CVMP/590/98 and EMEA/CVMP/591/98).

Therefore, it is concluded that the ratio of tulathromycin isomers as well as related substances/degradation products can be determined reliably.

2.2. Batch analyses

Analytical data are presented for the nine stability lots (made from three compounded lots of solution) manufactured at the applicant's production facility at Lee's Summit, Missouri, USA. All results are in accordance with the specification and show uniformity from batch to batch.

The specification of active ingredient content is set to 95 % - 105 % of label claim at time of release based on batch history and in response to a question posed by the Committee.

II F STABILITY

II.F.1 Stability Tests on the Active Substance

Stability testing has been performed on 6 lots of drug substance in 2 stability programmes. Samples were stored in double polyethylene bags inside a high density polyethylene drum, representing the commercial package.

Appearance, assay, impurities and water content have been determined. Testing methods were identical to those described for the drug substance (cf. II.C.1) with the exception of assay and purity testing at the initial time point in program I, where "scientifically equivalent" procedures ("same preparation procedures", "same chromatographic parameters", "same reagents") were used. These methods were requested by the Committee and have been provided.

Data up to 18 months at 25°C/60% RH, up to 12 months at 30°C/60% RH and up to 6 months at 40°C/75% RH were provided in the dossier, demonstrating that all batches comply with the proposed

specification. A retest period of 24 months has been claimed. When additional information becomes available, the retest period may be extended.

Additional testing results up to 24 months at 25°C/60% RH have been provided and a retest period of 36 months has been claimed. At present, the Applicant has committed to retain a retest period of 24 months.

Purposeful degradation of tulathromycin drug substance has been examined under a variety of stress conditions and the major degradation products have been identified.

The dossier contains a detailed description of the experiments and results, but no raw data have been provided. Chromatograms of samples before/after degradation were supplemented at the request of the Committee. Known degradation products have been identified. HPLC/MS analysis has been conducted to acquire structural information on unknown compounds.

Photostability studies according to VICH conditions indicate that the drug substance should be stored in light protective containers such as the proposed commercial packaging.

Supporting stability studies:
Additional stability studies were performed on tulathromycin drug substance stored up to 6 months in open dish containers at 40°C/75% RH. These samples showed no significant differences to samples stored in the proposed commercial packaging with respect to appearance, water content, assay, CP-614,159 levels, Total Unspecified Impurities and Total Impurities.

A stability protocol for the first three commercial batches of tulathromycin drug substance has also been provided. The only planned process change for the upcoming commercial batches is an increase in batch size.

Stability data of 6 batches of tulathromycin demonstrate that storage does not have an impact on isomer ratio of tulathromycin active substance. Additionally, it is evident that equilibration of the isomers in tulathromycin injectable solution depends on the conditions in solution during the manufacturing process rather than on the isomer ratio in the solid state active substance.

II.F.2 Stability Tests on the Finished Product

2.1 Product Specification and Routine Tests for shelf life:

The specifications for drug product apply to release testing of the drug product and also include specific requirements to be met at the end of shelf life. A lower limit of 95 % of active ingredient content at time of release was justified. . The Applicant agreed to tighten the shelf-life limits for active substance content to 92 to 105% of label claim. This limit is supported by the additional stability data provided.

2.2 Stability Tests

Stability studies are described for three compounded lots of tulathromycin injectable solution filled into 20, 100 and 500 ml Type I, flint glass vials. The storage conditions being studied are 30 °C/60% RH (intermediate) and 25 °C/60%RH (long term) for 24 months. The accelerated storage condition studied was 40°C /75% RH for 9 months. Vials are stored in both the upright and side-stored orientation, those latter vials addressing for any potential stopper/product interactions. VICH light challenge and thermal cycling studies have also been conducted.

Whereas in the original submission, data through 9 months storage time had been provided, now additional testing results up to 18 months under long term conditions are available.

Quality criteria tested include appearance, particulate contamination (visible particles), tulathromycin assay and isomer ratio, degradation products, content of monothioglycerol, pH, sterility, and antimicrobial effectiveness.

No significant change in appearance, particulate contamination, content of tulathromycin, isomer ratio and pH was observed. Loss in monothioglycerol content and some increase in degradation products was observed. Acceptable antimicrobial effectiveness and sterility of the finished product have been demonstrated. All stability samples to date met the proposed specifications.

Photostability testing: When exposed to UV light (200 watt hours / m^2) followed by fluorescent light (achieved exposure: 1.2 million lux hours) no loss in active ingredient content nor increase of impurity levels have been observed.

With the submission of long time data for up to 18 months, the Applicant has claimed a shelf life period of 36 months. A limited extrapolation of the real time data from the long term storage condition beyond the observed range to extend the shelf-life can be undertaken at approval time, if justified. No justification for the extension of the shelf-life has been presented. Therefore, it is only possible to grant a shelf-life of 18 months.

Batches manufactured at Amboise facility should be placed on accelerated and long-term stability studies and sterility and antimicrobial effectiveness should be monitored.

2.3 In-use Stability Tests

Two in-use programmes have been planned. The first addresses in-use stability testing of fresh product samples at the initial stability time point. The second programme involves in-use stability evaluation of product stored at 30°C/60% RH at intervals throughout product shelf life. An in-use stability programme and a maximum puncture programme with evaluation throughout product shelf life are proposed and results will be reported as they become available.

Results from the study at the initial stability time point show that a in-use shelf life of 4 weeks is justified. Results of a second in-use stability study and maximum puncture study were submitted including results of the fragmentation and self-sealing test.

The integrity of the closures following 100 punctures per closure has been investigated and been found to be acceptable, particularly when the warnings in section 5.8 of the SPC are considered. However, the range of bodyweights of pigs likely to be treated is 8 to 40 kg. This corresponds to dose volumes of 0.2 to 1.0 ml. If the 100 ml vial is considered, in theory up to 500 piglet doses may be available. The potential number of doses is further magnified for the 250 ml and 500 ml pack sizes. Therefore, the 250 ml and 500 ml pack sizes are restricted for use in cattle only.

II G AND H: DATA RELATED TO THE ENVIRONMENTAL RISK ASSESSMENT FOR PRODUCT CONTAINING OR CONSISTING OF GENETICALLY MODIFIED ORGANISMS (GMOS)

Not applicable.

II Q. OTHER INFORMATION

TSE risk assessment: A TSE statement has been provided confirming that Draxxin Solution for Injection does not contain any substance of animal origin and is in compliance with the Annex to Commission Directive 1999/104/EEC.

OVERALL CONCLUSION ON PART II

In general the dossier takes into account current rules and guidelines. Therefore Part II was found acceptable taking into account the commitment of the Applicant to fulfill certain provisions, which are listed below.

The Applicant committed to the following points at the time of the opinion:

1. To provide validation data (prior to commercial distribution) on the bulk solution compounding process on three consecutive batches, and should validate the filling process on each vial size (q.v. Process Validation Scheme for Tulathromycin Injectable Solution 100 mg/ml). The Applicant should include in the Process Validation Scheme a proposal of the timeframe and the maximum hold time between sterile filtration and commencing filling.

2. For the proposed medicinal product's batch size it should be verified that the scale up does not affect the reproducibility of the process, i.e. that the process described in the dossier is capable of providing the medicinal product with adequate quality. The Applicant should confirm in writing when any out of specification results should occur (with proposed action).

3. For tulathromycin, active substance limits for assay and total impurities should be revised taking into account the batch analysis data of the next campaign of tulathromycin production. If the results are consistent with those values attained during the November/December 2000 campaign, the specification should be tightened appropriately. The Applicant should state within 3 months when the next production campaign is likely to be and should indicate when they will be able to review and if appropriate vary the defined limits.

4. Revision 02 of the relevant Certificate of Suitability for erythromycin was issued 03/02 and the up-to-date Certificate of Suitability should be submitted within 3 months.

5. To confirm that both manufacturers of CP-60,273 will use the same synthetic route, the same quality control procedures, the same specifications and the same supplier of the erythromycin A (Abbott Laboratories). Otherwise, the necessary detailed information should be provided within 3 months.

6. To provide information on Class II solvents especially on methanol at step 1 of the synthesis of the starting material.

7. For future scale up of the commercial batch size of tulathromycin active substance it should be verified that the change does not affect the reproducibility of the process, i.e. that the process described in the dossier is capable of providing tulathromycin active substance with adequate quality.

8. To submit the results of the stability studies carried out with the first 3 commercial batches of the medicinal product, when available. The shelf-life specifications should be tightened in accordance with the results obtained. Besides, between the tests to be performed, the antimicrobial effectiveness should be included to be carried out at the end of this study.

9. For tulathromycin, a 24 months retest period is supported by the data submitted by the Applicant (and not 36 month as suggested by the Applicant). Therefore, the Applicant should commit to restrict the retest interval of the active substance to 24 months.

III SAFETY AND RESIDUE DOCUMENTATION

INTRODUCTION

Tulathromycin, is a semi-synthetic macrolide triamilide antimicrobial intended for the treatment and prevention of bacterial respiratory disease in non-lactating cattle and pigs.

The active substance tulathromycin is an equilibrated mixture of two structural isomers with an isomer ratio of 9:1. Pharmacology and toxicology studies have been submitted and reviewed in the application for the establishment of MRLs in cattle and swine. An ADI of 0.011 mg/kg bw was established based on microbiological endpoints. Tulathromycin is currently included in Annex III of Council Regulation (EEC) 2377/90.

III.A. SAFETY TESTING

III.A.2. PHARMACOLOGICAL STUDIES

III.A 2.1. Pharmacodynamics

See section IV.A.1.1.

III.A.2.2. Pharmacokinetics

See section IV.A.1.2.

III.A.3. TOXICOLOGICAL STUDIES

All toxicological studies were submitted with the MRL dossier.

III A.3.1. Single dose toxicity

Four studies were provided in dogs and rats investigating toxicity of a single dose of tulathromycin after oral or intravenous administration.

Oral toxicity of tulathromycin is low and the adverse effects (increase in transaminases, vomiting, diarrhoea) are reversible. In rats, the oral LD_{50} was estimated with >2000 mg/kg bw and a NOEL of 300 mg/kg was identified in the oral toxicity study in rats.

Intravenous toxicity of tulathromycin was considered intermediate/high. Adverse effects observed in rats were transient ataxia/decreased activity and in dogs, diarrhoea, erythema and apnoea (1 dog receiving 33.8 mg/kg). The type and possible mechanism of toxicity after intravenous application remained unclear. In rats, mortality was observed in doses of 33.8 mg/kg. A NOEL of 11.3 mg/kg was identified in the acute intravenous toxicity study in rats.

The acute dermal toxicity in rabbits (24 h exposure to 2000 mg/kg) showed no mortality, but slight oedema, slight skin desquamation, decreased food consumption and decrease defecation.

III. A 3.2. Repeated dose toxicity

Oral repeated toxicity studies were performed in rats (10 days, 1 month, 3 months) and dogs (1 month, 3 months and 1 year).

The main adverse reactions observed were an increase in serum liver enzymes (ALT, AST and SHD) without histopathological changes of the liver or evidence of enzyme induction. Furthermore, sporadic

gastrointestinal disturbances (vomiting, diarrhoea), decreased serum proteins, increase kidney weight and ophthalmological changes in dogs and minimal elevation in monocyte and eosinophil counts were observed.

Conclusions (single and repeat dose):
Tulathromycin is of low oral toxicity but toxicity following single intravenous administration was considerably higher. Due to the low bioavailability of the compound after oral administration, the oral toxicity studies were only partially suitable for the target animal safety assessment of this product.

III A 3.3 Tolerance in the target species

See Part IV 1.B.

III A 3.4 Reproductive toxicity, including teratogenicity

Reproduction studies were conducted with rats and developmental toxicity studies were performed in rats and rabbits.

No teratogenic effects were observed. Adverse effects on maternal reproductive parameters or foetal/neonatal development could only be induced with high oral overdoses. Thus, it is concluded that tulathromycin could be used in pregnant cows or sows.

III A.3.5. Mutagenicity

Tulathromycin was examined for its mutagenic activity employing four *Salmonella* indicator strains and one strain of *E.coli* in the presence and absence of metabolic activation. No indication of a mutagenic effect could be found. In the mammalian gene mutation test, tulathromycin was examined for the induction of gene mutations at the HPRT locus in CHO cells both, directly and in the presence of a rat liver activation system. No indication of a mutagenic effect could be found.

Tulathromycin was tested for its potential to induce chromosome aberrations in human lymphocyte cultures. No indication of a clastogenic effect of the compound could be found, neither in the presence or absence of metabolic activation. In the mouse micronucleus test no increase in the frequency of micronuclei in the polychromatic erythrocytes was found in rats at any time or at any dose level. No change in the ratio of polychromatic/normochromatic erythrocytes was observed indicating no bone marrow depression. The clastogenic activity was tested *in vitro* at the thymidine kinase locus in the mouse lymphoma L5178Y cell line. None of the treatments induced a mutant frequency above the minimum criteria for a positive response both with and without metabolic activation.

Conclusion: The genotoxic potential of tulathromycin was evaluated in a number of *in vitro* and *in vivo* genetic toxicology assays. The results of the genetic toxicology assays indicate that tulathromycin is not genotoxic.

III A 3.6. Carcinogenicity

Studies on carcinogenicity were not provided. However, due to the absence of a chemical structural relationship to known carcinogens, the negative results of genotoxic assays, the absence of degenerative or proliferative lesions in subchronic toxicity studies and the lack of a carcinogenic potential of other macrolide antibiotics it is assumed that tulathromycin is devoid of a carcinogenic risk.

III.A.4. Studies of other effects

III A.4.1. Immunological and irritation studies

The Applicant provided studies on immunological effects and irritation studies (skin, eye). The results showed that tulathromycin proved to be non-irritating and non-corrosive on rabbit skin.

However, the compound was found to be a severe ocular irritant in rabbits. Also, sensitisation potential was observed in the guinea pig maximization test (GPMT) and tulathromycin was therefore considered to be a potential skin sensitiser. An appropriate warning for the user has been included in the product literature for Draxxin.

Tulathromycin was developed solely for veterinary use. Therefore, to date, no human use data are available. However, tulathromycin has many similar effects (gastrointestinal such as vomiting, loose stools and hepatic such as slight increases in the liver enzymes ALT, AST, SDH) to other macrolide antimicrobials used in human medicine.

III A.4.2 Microbiological studies (studies on human gut flora and organisms used in food processing)

In vitro studies show that tulathromycin exposure to *Fusobacterium* and *Bifidobacterium* strains did not inhibit their growth under conditions simulating those of the human gastrointestinal tract.

The Applicant provided data indicating that the majority of the field isolates did not contain known macrolide resistance genes that may be inducible and that tulathromycin was a relatively poor inducer compared with erythromycin for the inducible macrolides, lincosamides and streptogramin B (MLS_B). Furthermore, the frequencies of the random-mutations vary depending on the bacterial species and the target site involved. The frequencies detected are comparable to those observed for spontaneous rates of resistance to fluoroquinolones. The spontaneous rates of resistance in these studies were not high and the frequency of macrolide resistance gene transfer in the presence of tulathromycin is comparable to frequencies observed when erythromycin or tilmicosin are the selecting agents.

III. A. 4.3 Studies on metabolites, impurities, other substances and formulation

The hepatotoxic potential of macrolides has been associated with the formation of nitrosoalkane metabolites, which can inhibit drug metabolism in the liver by complex formation and inactivation of microsomal drug oxidising enzymes. However, the only nitroso metabolite of tulathromycin was a very minor metabolite in all matrices investigated.

In the final drug formulation, two degradation products are controlled, for which a maximum limit is established. These products are also metabolites identified in cattle, swine, rat and dog.

The CVMP concluded that the qualification of impurities/ degradation products is considered adequate and in agreement with the recommendations of the relevant guidelines.

III.A.5. User Safety

Tulathromycin is considered a skin sensitiser and ocular irritant therefore, the following warning has been included in the SPC / product literature:

- Tulathromycin is irritating to eyes. If accidental eye exposure occurs, flush the eyes immediately with clean water.
- Tulathromycin may cause sensitisation by skin contact. If accidental skin exposure occurs, wash the skin immediately with soap and water.

- Wash hands after use.

The low oral bioavailability and the low protein binding of tulathromycin are considered to decrease the risk associated with accidental oral intake and no further warnings with regard to the user safety are considered necessary.

III.A.6. Ecotoxicity

A Phase I assessment provided by the Applicant demonstrated satisfactorily that the correct use of tulathromycin in accordance with the SPC will not result in levels of drugs residues that are hazardous to the environment. The predicted environmental concentration of tulathromycin for use in cattle and pigs, based on worst case assumptions, was calculated to be 27.8 µg/kg (pigs) and 42.8 µg/kg (cattle) and, therefore, does not exceed the threshold concentration of 100 µg a.i./kg soil. In accordance with the CVMP VICH Topic GL6 (Ecotoxicity Phase I) Guideline on Environmental Impact Assessment (EIAs) for Veterinary Medicinal Products – Phase I (CVMP/VICH/592/98-final) a Phase II environmental risk assessment is not required. Data on the fate and/or effects of tulathromycin in the aquatic and terrestrial compartments confirmed that there is no risk for the environment when Draxxin is used according to the labelling.

III.B. RESIDUE DOCUMENTATION

III. B.1 INTRODUCTION

Tulathromycin has been evaluated during the MRL application. All studies described in the residue part were conducted according to GLP regulations. The formulation used in the residue studies was similar or identical to the intended commercial formulation and consisted of the equilibrated mixture of both isomers. In the radiometric residue studies, tulathromycin was labelled with ^{14}C .

III.B.2. RESIDUE STUDIES

III B.2.1. Pharmacokinetics

See section III A 2.2

III B.2.2. Depletion of residues

Two comprehensive GLP-compliant residue depletion studies (one radiometric, one non-radiometric study) were submitted for each of the two target species. In the radiometric studies, total residues as well as the unchanged drug and the marker residue were analysed in all edible tissues of swine and cattle. Sample analysis was carried out by liquid scintillation counting (LSC) for total residues, and by HPLC/MS/MS for the unchanged drug CP-472,295(e) and for the marker residue (i.e. the acid digest common fragment CP-60,300 generated by tissue digestion in 2N HCl and expressed as tulathromycin equivalents). The method for CP-60,300 has already been accepted by the CVMP as routine analytical method (on the condition that the specificity relative to other macrolides can be demonstrated). All studies have already been assessed in the MRL procedure.

The results of the residue depletion studies revealed that in pigs, highest total residue concentrations were found in kidneys, followed by injection site and liver and then by muscle and skin/fat. In cattle, the order was liver and injection site, followed by kidneys and then by muscle and fat.

In pigs, parent tulathromycin accounted for approximately 96%, 102%, 96%, 103% and 18% of total residues in liver, kidney, muscle, injection site and skin/fat, respectively, across all time points. Analysis of swine tissues for marker residue yielded results comparable to those observed for the

parent compound (0.94 for liver, 0.83 for kidney, 0.89 for injection site, 0.86 for muscle and 0.28 for skin/fat).

In cattle, parent tulathromycin accounted for approximately 40%, 62%, 71%, 77% and 25% of total residues in liver, kidney, muscle, injection site and fat, respectively, across all time points. Analysis of cattle tissues for marker residue yielded results slightly higher than those observed for the parent compound (0.61 for liver, 0.78 for kidney, 0.91 for injection site, 0.79 for muscle and 0.46 for fat).
In the radiometric residue depletion study in pigs using the therapeutic dose of 2.5 mg/kg (intramuscular), average marker residue concentrations in liver were 2540, 1320, 538 and 192 µg/kg on days 4, 12, 24 and 36 after treatment. At the same time points average marker residue concentrations were 5340, 2030, 698 and 220 µg/kg in kidney; 557, 115, 44 and 12 µg/kg in muscle; and 182, 44, 13 and 4 µg/kg in skin/fat. At injection sites the average marker residue concentrations were 4140, 2140, 1300 and 680 µg/kg on days 4, 12, 24 and 36 after treatment.

In the radiometric residue depletion study in ruminant calves using the therapeutic dose of 2.5 mg/kg (subcutaneous), average marker residue concentrations in liver were 3600, 7700, 4100, 2900, 2300 and 700 µg/kg on days 0.5, 5, 15, 25, 36 and 48 after treatment. At the same time points average marker residue concentrations were 5000, 5600, 2300, 1100, 530 and 190 µg/kg in kidney; 1350, 820, 180, 45, 28 and 6 µg/kg in muscle; and 310, 300, 100, 47, 16 and 5 µg/kg in fat. At injection sites the average marker residue concentrations were 170 000, 10000, 5200, 2200, 1800 and 600 µg/kg on days 0.5, 5, 15, 25, 36 and 48 after treatment.

In the non-radiometric residue depletion study in ruminant calves using the therapeutic dose of 2.5 mg/kg (subcutaneous), average marker residue concentrations in liver were 5600, 3900, 3200, 2400, 1200 and 650 µg/kg on days 5, 12, 18, 25, 36 and 48 after treatment. At the same time points average marker residue concentrations were 4600, 2500, 1300, 700, 400 and 210 µg/kg in kidney; 550, 170, 89, 50, 19 and 9 µg/kg in muscle; and 260, 130, 100, 42, 21 and 8 µg/kg in fat. At injection sites the average marker residue concentrations were 5100, 3200, 2300, 800, 900 and 500 µg/kg on days 5, 12, 18, 25, 36 and 48 after treatment.

In the non-radiometric residue depletion study in pigs, a drawback was the inadequate sampling of injection site tissues. Since sampling was not in line with CVMP guidance III/5933/94, the data of this study were not taken into account when the withdrawal periods were calculated.

III B 2.3 MRLs and ADI

Commission Regulation (EC) No.1937/2002 inserted tulathromycin in Annex III of Council Regulation (EEC) No 2377/90 for cattle and pigs in accordance with the following table:

Pharmacologically active substance(s)	Marker residue	Animal Species	MRLs	Target Tissues	Other Provisions
Tulathromycin	(2R,3S,4R,5R,8R,10R,11R,12S,13S,14R)-2-ethyl-3,4,10,13-tetrahydroxy-3,5,8,10,12,14-hexamethyl-11-[[3,4,6-trideoxy-3-(dimethylamino)-β-D-xylo-hexopyranosyl]oxy]-1-oxa-6-azacyclopentadecan-15-one*, expressed as tulathromycin equivalents	Bovine	100 µg/kg 3000 µg/kg 3000 µg/kg	Fat Liver Kidney	Not for use in animals producing milk for human consumption Provisonal MRLs expire on 1.7.2004
		Porcine	100 µg/kg 3000 µg/kg 3000 µg/kg	Skin + fat Liver Kidney	Provisonal MRLs expire on 1.7.2004

* also known as CP-60,300, the hydrolysis product of tulathromycin and selected metabolites

The excipients (propylene glycol, monothioglycerol, citric acid and hydrochloric acid) are included in Annex II of Council Regulation (EEC) No 2377/90.

The ADI for tulathromycin based on the microbiological effects of the substance, established by the CVMP in the MRL assessment (Summary Report EMEA/MRL/842/02-FINAL) is 660 µg/person/day. The CVMP noted that the residues at the injection site must be taken into account when withdrawal periods are set to ensure that the residues in the total food package including the injection site do not exceed the ADI.

The marker analyte is a structural fragment common to tulathromycin and cladinose ring metabolites. This common fragment is generated following acid digestion of tissues, which result in hydrolytic cleavage of the cladinose moiety from the macrocyclic ring. A validated analytical method based on LC/MS/MS for the determination of the marker residue has been accepted by the CVMP as a provisional routine analytical method, provided that specificity relative to other macrolides will be demonstrated by the Applicant and definite MRLs can be established.

III B 2.4. Withdrawal periods

The withdrawal periods were calculated based on the marker residue concentrations in the target tissues liver, kidney and fat being below their specific MRLs, and, the amount of total residues in the 500 g edible portion (including 300 g injection site instead of normal muscle) being below the ADI. The calculations were performed according to the CVMP Note for guidance (Approach towards harmonisation of withdrawal periods - EMEA/CVMP/036/95).

Based on the data provided, a withdrawal period (meat and offal) of 49 days was agreed for cattle and 33 days for pigs.

Due to the low weight of the animals involved in the residue depletion studies, only small volumes were administered in these studies. In order to take account of this, a limitation of the injection volume per injection was proposed for the use of the product. For treatment of pigs over 80 kg bw and for treatment of cattle over 300 kg, the injection volume is to be divided so that no more than 2.0 ml and 7.5 ml, respectively, are injected in one site. This has been included in section 5.8 (Posology) of the SPC.

Since no MRL has been established for milk, Draxxin should not be used in animals producing milk for human consumption. An appropriate warning has been included in sections 5.3, and 5.11 (Contraindications and Withdrawal period) of the SPC / product literature. This restriction on use should be extended to pregnant cows and heifers as well because of the long withdrawal period for cattle:
“Not permitted for use in lactating cattle producing milk for human consumption. Do not use in pregnant cows or heifers, which are intended to produce milk for human consumption, within 2 months of expected parturition.”

III.B.3. ANALYTICAL METHOD(S)

HPLC/MS/MS methods were used for quantitative analysis of the unchanged drug and a common fragment, which is the marker residue ((2R,3S,4R,5R,8R,10R,11R,12S,13S, 14R)-2-ethyl-3,4,10,13-tetrahydroxy-3,5,8,10,12,14-hexamethyl-11-[[3,4,6-trideoxy-3-(dimethylamino)-β-D-xylo-hexopyranosyl]oxy]-1-oxa-6-azacyclopentadecan-15-one, expressed as tulathromycin equivalents). The common fragment was analysed by the method provisionally accepted for routine monitoring.

III.B.3.1. Analytical methods used for analysis of the unchanged drug

The method used for analysis of the unchanged drug in pigs was validated by analysis of fortified control samples. The results were within acceptance criteria for all tissues. The exception was the injection site muscle at the lowest concentration analysed (i.e. 0.1 μg/g). However, the precision at this concentration level was well within the specified limits. The method used for analysis of the unchanged drug in cattle was validated by analysis of fortified control samples. The limits for accuracy and precision were met.

III.B.3.3.2. Routine analytical methods (analysis of the marker residue)

The description of the proposed (in the meantime provisionally accepted) routine analytical methods was provided for the acid digest common fragment CP-60,300, generated by tissue digestion in 2N HCl and expressed as tulathromycin equivalents. The methods to determine the common fragment in porcine or bovine tissues have already been accepted by the CVMP as routine analytical method (Summary Report EMEA/CVMP/942/02-FINAL), provided that the specificity relative to tilmicosin, spiramycin, josamycin and acetylisovaleryltylosin will be demonstrated by the Applicant in the MRL application.

IV EFFICACY

IV.A. PRE-CLINICAL STUDIES

IV.A.1 PHARMACOLOGY

IV.A.1.1 Pharmacodynamics

Like other macrolides, tulathromycin inhibits essential protein biosynthesis by selective binding to bacterial 50S ribosomal subunits. It acts by stimulating the dissociation of the peptidyl-tRNA from the ribosome during the translocation process. The blocking of the protein synthesis confers a bacteriostatic effect on susceptible pathogens.

Specific studies on other (secondary) effects of tulathromycin have not been provided in both the target animal species and laboratory animals. However, the following observations have been made in connection with the toxicity studies provided and publications concerning pharmacological properties of other macrolides:

- Apnoea / decreased respiratory rate (rats, dog) and death due to respiratory arrest (rats) was observed in one dog and in rats immediately after single intravenous dose of 30 mg/kg bw (dog, rats) and 100 mg/kg bodyweight (rats).

- Tulathromycin produced slight increases in liver enzymes (ALT, AST, SDH) in rats and dogs. Although no histological correlates were found in both species, the results indicate that the liver is a potential target organ of tulathromycin.

- Sporadic and mild gastrointestinal disturbances (vomiting, loose stools) were reported in the single and repeated oral toxicity studies with tulathromycin in dogs. While prokinetic activity (stimulation of gastrointestinal motility via motilin receptors) has been demonstrated for other macrolides, it was not investigated with tulathromycin.

For other members of the macrolide group, a number of specific pharmacodynamic and toxic effects have been reported in published literature such as cardiotoxicity or reversible/irreversible hearing loss after oral treatment. However, the results of the repeated dose toxicity studies in rats and dogs did not show evidence of these secondary pharmacodynamic effects or signs of adverse renal or neurotoxic effects.

IV.A.1.2. Pharmacokinetics

Data on the pharmacokinetic profile of tulathromycin in plasma and lung of pigs and cattle (pre-ruminants and ruminants) were provided in several mostly GLP compliant studies (intramuscular, intravenous, oral use). Concerning distribution, excretion and metabolism in tissues, plasma and excreta of swine and cattle, two comprehensive GLP-compliant radiometric residue depletion studies were provided (one for swine and one for cattle). Several sub-studies were derived from these two main studies. All samples, analysed in these sub-studies, were collected throughout the animal phase of the two radiometric residue depletion studies. Furthermore, pharmacokinetic data on plasma and lung concentrations after oral administration were included in the repeat dose toxicity studies in rats (1 and 3 month study) and dogs (1, 3 and 12 month study).

Absorption from the injection site following a single subcutaneous (cattle, lateral neck) or intramuscular (pigs, lateral neck) administration of 2.5 mg/kg bw of tulathromycin in the commercial formulation was relatively rapid (mean plasma T_{max} 0.25 – 2 hours) and almost complete (bioavailability greater than 80 % when compared to intravenous administration). The pharmacokinetic characteristics of tulathromycin in pigs and cattle can be described by a long plasma elimination half-life of more than 70 h.

Binding to plasma proteins was low with the bound fraction ranging from 0.32 to 0.39 (approximately 40%) in cattle and 0.38 to 0.44 (approximately 40%) in pigs.

In cattle and pigs, the bioavailability in the therapeutic target tissue (lung) is extensive, with lung AUCs being 60 times greater than plasma AUCs. Peak lung levels of approximately 4.1 µg/g and 3.5 µg/g are achieved after a single dose injection of 2.5 mg/kg bw in cattle and swine, respectively. Maximum concentrations in the lung were generally observed within 24 hours following dose administration and mean lung tissue concentrations (as evidenced by AUC) were substantially greater (73.7 x in cattle, 61.4 x in pigs) than plasma concentrations. In calves, lung concentrations above 3 µg/g persisted for 144 hrs after injection. In pigs, lung concentrations were approx. 2 µg/g 72 hrs after administration. Consequently lung concentrations in cattle remained above the tulathromycin MIC_{90} values for *Mannheimia haemolytica* (2.0 µg/ml) and *Pasteurella multocida* (1.0 µg/ml) for approximately 9 and 15 days, respectively, and lung concentrations in pigs remained above the tulathromycin MIC_{90} values for *Pasteurella multocida* (2.0 µg/ml) and *Mycoplasma hyopneumoniae* (0.5 µg/ml) for approximately 5 and 15 days, respectively.

The elimination half-life in the lung tissue was exceptionally long with 6 and 8 days in pigs and cattle, respectively. Pharmacokinetic profiles in both plasma and lung tissue of preruminant calves (4 -7 weeks of age) were found to be generally similar to those observed in ruminant cattle. Dose linearity of the pharmacokinetic parameters of tulathromycin was shown for calves but was not definitely demonstrated in pigs. The observations in cattle and pigs are consistent with those obtained in the studies in dogs and rats with a high distribution into tissue relative to plasma concentrations and an exceptionally long elimination half-life in the lung. Even higher concentrations of the substance were achieved in cells of inflammation (neutrophiles, alveolar macrophages) from the target species under *in vitro* conditions.

Metabolism was investigated in cattle and pigs using the recommended therapeutic dosage of a single subcutaneous or intramuscular injection of 2.5 mg/kg tulathromycin, and in rats and dogs given the radiolabelled drug by aqueous oral gavage (15 mg/kg/day or 50 mg/kg/day for two consecutive days for dogs or rats, respectively). Radiotracer HPLC and HPLC/MS analysis of urine, faeces, bile, liver, kidney, muscle, fat, skin/fat, and injection site samples suggested that the ^{14}C residues in the matrices of pigs and cattle were predominantly the unchanged drug. Except for swine skin/fat and cattle bile, only minor quantities of metabolites were formed in all collected matrices. In swine skin/fat, one metabolite was identified as the desosamine N-oxide (about 19.7% of the radioactivity). In cattle bile, one metabolite was identified as the N-despropylation product of the cladinose moiety (about 16.3% of the radioactivity). In all other matrices, none of the metabolites exceeded 10% of the total radioactivity for the matrix. Apart from 3 metabolites found in cattle, the metabolic profiles obtained from matrices of cattle and pigs were largely similar to those shown in liver, bile, urine and faeces of dogs and rat.

Excretion of the radiolabelled ^{14}C tulathromycin in pigs and cattle was relatively slow but more or less complete. In pigs, more than 90 % of the dose was excreted within 23 day. Faeces contained about 2/3 and urine about 1/3 of the administered dose. In cattle, excretion was somewhat slower (about 70 % within 47 days) with the excreted dose being nearly equally divided between urine (40%) and faeces (32%).

IV.A.1.3 Microbiology - MIC and Breakpoint

Several studies were provided to determine the *in vitro* susceptibility of the main pathogens isolated from the respiratory tract of cattle with clinical signs of Bovine Respiratory Disease (BRD) or pigs with Swine Respiratory Disease (SRD). The isolates were part of an European MIC survey programme and from clinical studies in various EU Member States.

The EU countries selected for bacterial strain sampling represent approximately 80 % and 60 % of the overall EU cattle and pig population respectively. The geographical distribution of the bacterial strains

in each of the selected EU countries is not in every case equivalent with the major animal producing regions. The number of bacterial strains tested from cattle was considered insufficient for *Haemophilus somnus* and *Mycoplasma bovis*. The Committee decided that the indication for *Mycoplasma bovis* could not be accepted. The Applicant provided a commitment to provide a post-authorisation monitoring programme, the protocol for which needs prior approval by the CVMP, in order to determine the susceptibility (MIC values) for the claimed pathogens. *Haemophilus somnus* strains from the mediterranean region would be included in this programme.

MIC data for the pathogens isolated from **cattle** showed that for *Haemophilus somnus*, *Mannheimia (Pasteurella) haemolytica* and *Pasteurella multocida* breakpoints of ≤ 8 µg/ml for susceptibility (S) and ≥ 16 µg/ml for resistance (R) can be derived. For *Mycoplasma bovis* the applicant demonstrated by *in vitro* experiments different populations, a susceptible population with MICs ≤ 0.5 µg/ml and a resistant population with MICs ≥ 64 µg/ml. The corresponding microbiological breakpoints are ≤ 0.5 µg/ml (S) and ≥ 1 µg/ml (R).

Mycoplasma hyopneumoniae strains from **pigs** show *in vitro* MICs ≤ 0.125 µg/ml for the susceptible population and MICs ≥ 0.25 µg/ml for the resistant population. For the other claimed pathogens from pigs the MIC values show that for *Pasteurella multocida* the microbiological breakpoint for the susceptible population can be defined as ≤ 2 µg/ml and for Actinobacillus pleuropneumoniae as ≤ 16 µg/ml. The corresponding breakpoints for the resistant populations are ≥ 4 µg/ml resp. ≥ 32 µg/ml.

30% of the *M. bovis* isolates tested in the field efficacy studies showed MICs against tulathromycin higher than 64 µg/ml; furthermore, there were deficiencies noted in the studies performed on this pathogen. The CVMP considered the difficulties in the laboratory methodology (isolation, culture, MIC determination) for Mycoplasma, but taking into account the insufficient *in vitro* and clinical data provided for *M. bovis* the Committee concluded that the indication for *M. bovis* should be deleted.

In addition, the Applicant submitted data on the accumulation of tulathromycin in bovine phagocytes, porcine neutrophiles and alveolar magrophages, i.e. cells involved in respiratory infections.

Furthermore, the CVMP considered that the clinical efficacy of tulathromycin could not be derived from a simple PK (pharmacokinetic)-MIC relationship because this model would not reflect environmental conditions and intracellular uptake, which have a great impact on the *in vivo* efficacy of the substance. Therefore, based on the results of the clinical studies and these *in vitro* data, the CVMP agreed to include the proposed bacteria (cattle: *Mannheimia haemolytica, Pasteurella multocida, Haemophilus somnus,* pigs*: Actinobacillus pleuropneumoniae, Pasteurella multocida, Mycoplasma hyopneumoniae)*.

Table: Summary of preliminary microbiological breakpoints for tulathromycin

	MIC [µg/ml] MIC_{90}	MIC Breakpoint [µg/ml] S	R
Cattle (BRD)			
Mannheimia haemolytica	2	≤ 8	≥ 16
Pasteurella multocida	1	≤ 8	≥ 16
Haemophilus somnus	4	≤ 8	≥ 16
Mycoplasma bovis	> 64	≤ 0.5	≥ 1
Pigs (SRD)			
Actinobacillus pleuropneumoniae	16	≤ 16	≥ 32
Pasteurella multocida	2	≤ 2	≥ 4
Mycoplasma hyopneumoniae	0.125	≤ 0.25	≥ 0.5

IV.B.1 TOLERANCE IN THE TARGET ANIMAL SPECIES

The applicant has provided several studies about the tolerance of tulathromycin (4 in cattle and 2 in pigs). The design of the target animal safety studies in cattle and pigs was appropriate to examine the tolerance of calves and pigs towards the product at the recommended treatment dose as well as at elevated and repeated doses. The recommended dose, multiple doses (3, 4, 5, 6 and 10 fold overdose) and subcutaneous and intramuscular routes were used.

Localised morphological reactions to subcutaneous and intramuscular injection seem to be the predominant treatment effect from the present data in calves and pigs. Tissue lesions consisted of visible and palpable swellings in some cases accompanied by lameness, which persisted up to 4 weeks after injection in both species. These observations have been added in section 5.4 (Undesirable effects) of the SPC and the package insert.

In some calves, subcutaneous injection caused painful reactions as evidenced by clinical signs of discomfort (head shaking, jumping). This observation was not made in pigs after intramuscular injection. An appropriate warning has been added in section 5.4 (Undesirable effects) of the SPC. However, the CVMP agreed that these negative aspects are in part outweighed by the favourable administration schedule of Draxxin consisting of a single injection.

In pigs, clinical reactions to injection consisting of squeaking, restlessness, tremor, head or limb shaking were observed after injection of 3 fold, 5 fold and 10 fold overdoses and persisted up to 2 hours after injection. Lameness occurred when the test article was injected into the hind limb and persisted up to 21 days after injection at maximum.

Using a prototype formulation, test article-related histopathological changes were observed in the myocardia of 3 animals, 1 at 5 fold (12.5mg/kg) and 2 at 6 fold (15mg/kg) the commercial dose. Findings were minimal to mild and no myocardial degeneration was observed in any animal when the final product formulation was administered at either 10, 5 or 3 fold the commercial dose (2.5mg/kg). Myocardial degeneration is a macrolide-specific effect, which was not observed in pigs.

In one cow, red urine and tremor after injection of tulathromycin at the 10 fold overdose was observed. There was no explanation for this reaction.

IV.B 2 CLINICAL STUDIES

Draxxin is indicated for the treatment and prevention of Bovine Respiratory Disease (BRD) in cattle and for the treatment of Swine Respiratory Disease (SRD) in pigs.

Bacteria considered to be associated with the diseases and within the claim of the product are: *Mannheimia (Pasteurella) haemolytica*, *Pasteurella multocida and Haemophilus somnus (*BRD) and *Actinobacillus pleuropneumoniae*, *Pasteurella multocida* and *Mycoplasma hyopneumoniae (SRD)*.

IV.B 2.1 CATTLE

IV.2.1.1 CHALLENGE STUDIES

The Applicant provided two challenge studies:

- A dose-determination study (1.25, 2.5 and 5.0 mg/kg) against BRD associated with *Mannheimia (Pasteurella) haemolytica* and *Pasteurella multocida*.
- An infection model study evaluating the efficacy of 2.5 mg tulathromycin/kg bw against BRD associated with *Mycoplasma bovis.*

These studies had a number of shortcomings (choice and ranking of efficacy parameters, method of microbiological sampling, choice of statistical tables and analysis) which did not allow clear conclusions.

IV.2.1.2. FIELD STUDIES

A total of 10 controlled field trials were conducted: 5 dose determination/confirmation studies (1.25 and 2.5 mg/kg b.w), three studies to demonstrate treatment (2.5 mg/kg bw), and two studies to demonstrate prevention (2.5 mg/kg bw). The corresponding studies were conducted according to essentially similar study protocols.

Furthermore, two field studies were provided employing 2.5 mg/kg bw to demonstrate ongoing treatment and prevention success after observation periods longer than in the studies mentioned above.

IV.2.1.2.1 Dose-determination/-confirmation studies

Five field studies conducted in different EU Member States (France, Germany, UK) were provided for dose determination/confirmation.

Two doses of tulathromycin (1.25 and 2.5 mg/kg b.w.) or a positive control (tilmicosin) were administered at Day 0 subcutaneously to cattle aged 1 – 12 months. The animals in the field studies were diagnosed with BRD as characterised by the clinical signs and the presence of pathogens (*Mannheimia haemolytica, Pasteurella multocida* and *Haemophilus somnus*). Animals were observed until day 14.

Efficacy was assessed on the basis of clinical signs, i.e. reduction in pyrexia, improvement in clinical signs of BRD, mortality and successful completion of the study. Comparison of the different dosages (Day 0, Day 2 and Day 14 post-treatment) showed that a better treatment success (higher completion rate) was achieved after administration of the higher dose (2.5 mg/kg), and that this dose was equally effective as the positive control. Thus the dose of 2.5 mg/kg bw was chosen to be the commercial one for therapeutic and preventive treatment of BRD.

IV.2.1.2.2. Efficacy in the treatment of Bovine Respiratory Disease (BRD)

Efficacy in the treatment of BRD was demonstrated in four field studies conducted in different EU Member States (France, Germany, Italy and UK) and involving animals aged 3 weeks - 12 months. The studies were conducted during outbreaks of BRD confirmed by clinical signs and the presence of pathogens on D0 prior to treatment (*Mannheimia haemolytica, Pasteurella multocida, Haemophilus somnus* and/or *Mycoplasma bovis*). One study conducted in the US was not suitable mainly due to undifferentiated aetiology of the disease.

Animals were observed daily from D0 to D14 for clinical signs of BRD, including pyrexia, abnormal respiration and depression, and clinical signs of other disease including description of any localised injection site reactions. In one study, animals were observed until D62. No post-treatment microbiological samples were collected.

The animals received a single dose of 2.5 mg tulathromycin/kg b.w. (subcutaneously) or were treated with a positive control (tilmicosin or florfenicol) at Day 0.

Efficacy was assessed on the basis of clinical signs, i.e. reduction in pyrexia, improvement in clinical signs of BRD, mortality and successful completion of the study. On D0, D2 and D14, data were compared statistically using categorical analysis and analysis of variance. Safety was assessed considering clinical observations and observations at the injection site.

The results showed that the clinical signs of abnormal respiration, depression and increase of rectal temperature significantly improved. A number of animals showed reactions (e.g. pain, inflammation) at the injection site. This has been included in section 5.4 (Undesirable effects) of the SPC.

Since *Mycoplasma bovis* was not included in the dose-determination studies conducted in the field and the preclinical *in vitro* and *in vivo* data were insufficient CVMP, did not accept the claim for *Mycoplasma bovis.* However, this decision was amended after the initial authorisation (see the end of this EPAR).

The CVMP, therefore, concluded that a single injection of 2.5 mg tulathromycin/kg b.w. would be effective in the treatment of BRD where *Mannheimia haemolytica, Pasteurella multocida* and *Haemophilus somnus* were involved. Efficacy was comparable to that of approved commercial products containing tilmicosin and florfenicol.

IV.2.1.2.3. Efficacy in the prevention of Bovine Respiratory Disease (BRD)

Efficacy in the prevention of BRD was demonstrated in 3 field studies conducted in different EU Member States (France and Italy) involving animals up to 30 months of age. The studies were conducted in animals exposed to respiratory disease (BRD) during an outbreak of the disease, but without clinical signs of BRD (abnormal respiration or depression). Pathogens involved in the disease in affected animals (not included in the study) were *Mannheimia haemolytica, Pasteurella multocida, Haemophilus somnus* and/or *Mycoplasma bovis*.

Animals, which were not exhibiting signs of BRD but remained in contact with animals diagnosed with BRD were treated at Day 0 either with a single dose of 2.5 mg tulathromycin per kg bw or a negative control (saline) were administered subcutaneously. The third study also included a positive control group (tilmicosin).
The effect in reducing the incidence of BRD was primarily assessed on the basis of the percentage of animals completing the studies in each treatment group at the end of the observation period, and the number of animals withdrawn from the study due to reasons related to BRD. Safety was assessed considering clinical observations and observations at the injection site.

The percentage of animals completing the study was significantly higher in the tulathromycin-treated group compared to the saline-treated group. Also, the number of animals withdrawn from the study for reasons related to BRD was significantly higher in the saline-treated group than in tulathromycin-treated animals.

The CVMP, therefore, concluded, that a single subcutaneous dose of 2.5 mg tulathromycin per kg bw administered at the start of a BRD outbreak, significantly reduced the percentage of animals exhibiting clinical signs of BRD during an outbreak where *Mannheimia haemolytica, Pasteurella multocida* and *Haemophilus somnus* were involved.

IV.B.2.2 PIGS

The Applicant provided 5 controlled field trials and 3 laboratory studies with experimentally induced *Mycoplasma hyopneumoniae, Pasteurella multocida* and *Actinobacillus pleuropneumoniae* infections.

Furthermore, 1 field study was provided employing 2.5 mg/kg b.w. to demonstrate treatment success after an observation period longer than in the studies mentioned above.

IV.2.2.1 Challenge studies

The Applicant provided 3 laboratory studies

- A dose-determination study (2.5 and 5.0 mg/kg) against Swine Respiratory Disease (SRD) associated with *Actinobacillus pleuropneumoniae* ,
- A dose-determination study (1.25, 2.5 and 5.0 mg/kg) against experimentally induced infection with *P. multocida*, and

- An infection model study evaluating the efficacy of 2.5 mg tulathromycin/kg bw against SRD associated with *Mycoplasma hyopneumoniae*. This study was provided to support the efficacy of tulathromycin since infections with *Mycoplasma hyopneumoniae* were only found in a limited numbers in the field studies.

Dose-determination: the study employing *Actinobacillus pleuropneumoniae* demonstrated that a single intramuscular injection of 2.5 or 5.0 mg tulathromycin/kg bw was effective in the treatment of experimentally induced *Actinobacillus pleuropneumoniae* pneumonia. The challenge study employing *Pasteurella multocida* (1.25, 2.5 and 5.0 mg tulathromycin/kg bw) demonstrated that a dose of 1.25 mg/kg was not effective, while doses of 2.5 and 5.0 mg/kg had similar efficacy to each other. Therefore, doses of 2.5 and 5.0 mg/kg of were used in the dose-determination field studies.

The third study demonstrated that tulathromycin administered as a single injection at a dosage of 2.5 mg/kg was effective in the treatment of induced infection of *Mycoplasma hyopneumoniae.*

IV.2.2.2. Field studies

IV.2.2.3. Dose-determination/-confirmation studies

Four field studies were conducted in different EU Member States (France, The Netherlands, UK)

The animals were diagnosed with SRD as characterised by clinical signs and the presence of pathogens isolated from the lungs or presence of characteristic pathological changes of the lungs of other pigs in the same herd (*Pasteurella multocida, Actinobacillus pleuropneumoniae, Mycoplasma hyopneumoniae,* and other non-target microorganisms). A single dose of tulathromycin (2.5 or 5.0 mg/kg b.w.) or a positive control (tiamulin or florfenicol) was administered intramuscularly to pigs at Day 0. Animals were observed until Day 10. Efficacy was assessed on the basis of clinical signs, i.e. reduction in pyrexia, improvement in clinical signs of SRD, mortality, average daily weight gain over the observation period and successful completion of the study.

Animals in the tulathromycin-treated group showed significantly improved clinical signs and a significantly lowered mean rectal temperature. In all groups, the mean body weight of animals completing the study increased from Day 0 to Day 10. There was significant difference in the average daily weight gain between treatments in favour of the tulathromycin-treated group.

For the variable distribution of clinical signs, no significant differences were observed between the positive control groups (tiamulin or florfenicol) compared to the tulathromycin group. However, the percentage of animals successfully completing one study on Day 10 was higher for the tulathromycin group (2.5mg/kg b.w.) compared to the positive control (tiamulin) group. Treatment with tulathromycin showed a comparable treatment success with both doses (2.5 or 5.0 mg/kg bw), and both doses were (at least) equally effective as the positive controls. Thus the lower dose (2.5 mg/kg) was chosen to be confirmed in further field studies.

No adverse drug reactions were reported in these studies.

IV.2.2.4. Efficacy in the treatment of Swine Respiratory Disease (SRD)

In addition to the dose determination/confirmation studies (see IV.2.1.2.1), 2 field studies from Germany were provided to support the efficacy of tulathromycin in the treatment of Swine Respiratory Disease. One of the studies comprised a prolonged observation period (compared to the other field studies) to demonstrate ongoing treatment success.

The animals in the field studies were diagnosed with SRD as characterised by clinical signs and presence of pathogens isolated from the lungs or presence of characteristic pathological changes of the lungs of other pigs in the same herd (*Mycoplasma hyopneumoniae*, *Pasteurella multocida,* and/or *Mycoplasma hyorhinis*). A single dose of tulathromycin (2.5 mg/kg bw) or a positive control (tiamulin

or florfenicol) was administered intramuscularly to pigs at D0. Animals were observed until D10. Efficacy was assessed on the basis of clinical signs, i.e. reduction in pyrexia, improvement in clinical signs of SRD, mortality, average daily weight gain over the observation period and successful completion of the study. Animals in the tulathromycin-treated group showed significantly improved clinical signs and a significantly lowered mean rectal temperature. In all groups, the mean body weight of animals completing the study increased. No significant differences were observed between the positive control groups (tiamulin or florfenicol) compared to the tulathromycin group.

The CVMP concluded that a single injection of 2.5 mg tulathromycin / kg b.w. would be effective) in the treatment of SRD when *Mycoplasma hyopneumoniae, Actinobacillus pleuropneumoniae* and/or *Pasteurella multocida* were positively identified. Efficacy was comparable to that of approved commercial products containing tiamulin or florfenicol.

IV.B.2.3 Conclusion on the Clinical Efficacy Part

Shortcomings of the data provided were noted, especially with regard to the methodology of microbiological sampling in cattle, the short duration of post treatment observation periods, low completion rates at the end of the observation period, lack of post-treatment microbiological samples, and lack of meaningful, unequivocal laboratory challenge studies in cattle. However, based on robust field data the CVMP agreed that the efficacy of a single dose of 2.5 mg tulathromycin/kg bw had been sufficiently demonstrated for the treatment and prevention of BRD associated with *Mannheimia (Pasteurella) haemolytica*, *Pasteurella multocida* and *Haemophilus somnus* and the efficacy in the treatment of SRD associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida* and *Mycoplasma hyopneumoniae* by the improvement of the clinical signs of the disease. Also, the efficacy of tulathromycin was demonstrated comparable to that of already approved veterinary medicinal products containing tilmicosin (BRD), tiamulin (SRD) or florfenicol (BRD and SRD).

The Applicant committed to provide a post-authorisation monitoring programme, the protocol of which needs prior approval by the CVMP, in order to determine the susceptibility (MIC values) for the claimed pathogens in pigs and cattle.

V RISK BENEFIT ASSESSMENT

In general the quality part of the dossier takes into account current requirements and guidelines. Some of the outstanding points that have been identified were the need for validation data on the bulk solution compounding process on three consecutive batches; verification that the scale up of production at the manufacturing site in France does not affect the reproducibility of the process; revision of the limits for assay and total impurities taking into account the batch analysis data of the next campaign of tulathromycin production and the fact that the shelf-life specifications will need to be tightened in accordance with the results obtained. The Applicant has committed to provide these data as Follow Up Measures within deadlines set by CVMP.

Tulathromycin is of low oral toxicity but toxicity following single intravenous administration was considerably higher. Reproduction studies were conducted with rats and developmental toxicity studies were performed in rats and rabbits. No teratogenic effects were observed. Adverse effects on maternal reproductive parameters or foetal/neonatal development could only be induced with high oral overdoses. Thus, it was concluded that tulathromycin could be used in pregnant cows and sows. The genotoxic potential of tulathromycin was evaluated in a number of *in vitro* and *in vivo* genetic toxicology assays. The results of the genetic toxicology assays indicate that tulathromycin is not genotoxic. Studies on carcinogenicity were not provided. However, due to the absence of a chemical structural relationship to known carcinogens, the negative results of genotoxic assays, the absence of degenerative or proliferative lesions in subchronic toxicity studies and the lack of a carcinogenic potential of other macrolide antibiotics it was considered that tulathromycin is devoid of a carcinogenic risk.

Tulathromycin was found to be a severe ocular irritant in rabbits and was therefore irritating to eyes. Furthermore, tulathromycin was also considered to be a potential skin sensitiser. Therefore, appropriate user warnings have been included in the product literature for Draxxin.

Based on the data provided, a withdrawal period (meat and offal) of 49 days was agreed for cattle and 33 days for pigs. Since no MRL has been established for milk, Draxxin should not be used in animals producing milk for human consumption. An appropriate warning has been included in sections 5.3, and 5.11 (Contraindicationsand Withdrawal period) of the SPC / product literature:
"Not permitted for use in lactating cattle producing milk for human consumption. Do not use in pregnant cows or heifers, which are intended to produce milk for human consumption, within 2 months of expected parturition."

Concerning the low weight of the animals involved in the residue depletion studies, a limitation of the injection volume per injection was proposed. For treatment of pigs over 80 kg bw and for treatment of cattle over 300 kg, the injection volume is to be divided so that no more than 2.0 ml and 7.5 ml is administered per injection site, respectively. This has been included in section 5.8 (Posology) of the SPC.

Many calves and pigs exhibited injection site reactions, which persisted for about 30 days. This has been taken into account by the CVMP by including an appropriate warning in the SPC and product literature.

The CVMP considered that, based on the results of the clinical studies and *in vitro* data submitted by the Applicant, the approved claims could include the following bacteria: *Mannheimia haemolytica, Pasteurella multocida, Haemophilus somnus, (cattle) and Actinobacillus pleuropneumoniae, Pasteurella multocida, Mycoplasma hyopneumoniae (pigs)*.

The CVMP considered the difficulties in the laboratory methodology (isolation, culture, MIC determination) for Mycoplasma, but taking into account the insufficient *in vitro* and clinical data provided for *M. bovis*. concluded that the indication for *M. bovis* should be deleted. This decision was amended after the intial Marketing Authorisation following the submission of further date (see the end of this EPAR).

Shortcomings of the clinical field trial data provided were noted, especially with regard to the methodology of microbiological sampling in cattle, the short duration of post treatment observation periods, low completion rates at the end of the observation period, lack of post-treatment microbiological samples, and lack of meaningful, unequivocal laboratory challenge studies in cattle. However, the CVMP agreed that the efficacy of a single dose of 2.5 mg tulathromycin / kg b.w. was sufficiently demonstrated for the treatment and prevention of Bovine Respiratory Disease (BRD) associated with *Mannheimia (Pasteurella) haemolytica*, *Pasteurella multocida* and *Haemophilus somnus* and the efficacy in the treatment of Swine Respiratory Disease (SRD) associated with *Actinobacillus pleuropneumoniae*, *Pasteurella multocida* and *Mycoplasma hyopneumoniae* by the completion rates and improvement of the clinical signs of the disease. Furthermore, the efficacy of tulathromycin was demonstrated comparable to that of an already approved veterinary medicinal products containing tilmicosin (BRD), tiamulin (SRD) or florfenicol (BRD and SRD).

Based on the original and complementary data presented the Committee for Veterinary Medicinal Products concluded that the quality, safety and efficacy of the product were considered to be in accordance with the requirements of Council Directive 2001/82/EC.

In November 2004 the Applicant provided two additional *M. bovis* infection model studies, two MIC studies to obtain additional European *in vitro* sensitivity data on *M. bovis* and 5 additional field studies in Italy, Spain, France and Germany to evaluate the efficacy of Draxxin in the treatment and prevention of natural outbreaks of bovine respiratory disease (BRD) caused by respiratory pathogens including *M. bovis*.

Since BRD is a multi-factor induced disease complex, the difficulties to conduct feasible field studies, which allow clear conclusions in regard of *M. bovis* as the causative agent were discussed for this application. It was stressed that sites where only *M. bovis* was present, even if it was possible to find such sites in the field, would be highly unlikely and non-representative for BRD. Furthermore *M. bovis* was isolated at relatively high levels of incidence from nearly all sites. Overall it was the most prevalent pathogen present, as *M. bovis* was isolated from 32.9% of clinical cases of BRD sampled across all farms. The isolation frequency across the 4 farms ranged from 22 to 68.4%.

Both infection model studies showed that tulathromycin was effective in significantly reducing mortality rates, body temperatures and lung pathology in animals challenged with *M. bovis* with low and high MIC values. However, due to the high mortality rate, the efficacy of tulathromycin in improving or curing clinical signs of the disease (depression, abnormal respiratory signs) could not be established with statistical evidence. The CVMP considered a recovery rate for *Mycoplasma bovis* in a dimension of 10^5 cfu/ml as relatively high, although statistically significantly reduced in Draxxin treated animals when compared to saline control. The clinical field studies all dealt with mixed infections including *Pasteurella multocida*, *Mannheimia haemolytica*., *Haemophilus somnus*, and *M. bovis*. The isolation rates for *M. bovis*, *M. haemolytica, P. multocida, H. somnus* and others in phase 1 (nasopharyngeal swabs were collected from all clinical cases prior to treatment initiation) and also in phase 2 (swabs taken only from saline treated withdrawals) varied among the field studies. *M. bovis* was shown to be the most prevalent pathogen only in 1 study. In all field studies, the outbreak of bovine respiratory disease, associated with *M. bovis*, *M. haemolytica, P. multocida, H. somnus* and others was almost mild to moderate. The proportion of animals that did not fully recover after treatment was high. However, in conclusion the CVMP considered that the data submitted supported the additional indication for the treatment and prevention of bovine respiratory disease associated with *M. bovis* sensitive to tulathromycin.