21 February 2019
EMA/138662/2019
Veterinary Medicines Division

Committee for Medicinal Products for Veterinary Use

CVMP assessment report for Forceris (EMEA/V/C/004329/0000)

International non-proprietary name: toltrazuril / iron(iii) ion

Assessment report as adopted by the CVMP with all information of a commercially confidential nature deleted.

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Introduction

The applicant CEVA SANTE ANIMALE submitted on 19 June 2017 an application for a marketing authorisation to the European Medicines Agency (The Agency) for Forceris, through the centralised procedure under Article 3(2)(a) of Regulation (EC) No 726/2004 (optional scope).

The eligibility to the centralised procedure was agreed upon by the CVMP on 6 November 2015 as Forceris contains a new fixed combination of active substances (toltrazuril and iron) which was not authorised as a veterinary medicinal product in the Union on the date of entry into force of Regulation (EC) No 726/2004.

The active substances in Forceris are toltrazuril, an antiprotozoal substance, and iron (as gleptoferron), an anti-anaemic substance. The target species is piglets (pigs).

Forceris is a suspension for injection for piglets containing 30 mg/ml toltrazuril and 133.4 mg/ml iron (as gleptoferron). The product is available in multi-dose translucent plastic vials and in three different pack sizes, containing 100 ml, 250 ml or 500 ml.

The applicant applied for the following indications:

"In piglets:

- For the reduction in oocyst excretion and the prevention of clinical signs of coccidiosis in farms with a confirmed history of coccidiosis caused by *Cystoisospora suis*.
- For the prevention of iron deficiency anaemia."

The rapporteur appointed is Cristina Muñoz Madero and the co-rapporteur is Sylvie Louet.

The dossier has been submitted in line with the requirements for submissions under Article 13b of Directive 2001/82/EC – a fixed combination application.

On 21 February 2019, the CVMP adopted an opinion and CVMP assessment report.

On 23 April 2019, the European Commission adopted a Commission Decision granting the marketing authorisation for Forceris.

Scientific advice

The applicant received scientific advice from the CVMP on 10 July 2014 (EMA/CVMP/SAWP/285834/2014) with a clarification on 6 November 2014 (EMA/CVMP/SAWP/604365/2014). The scientific advice pertained to efficacy aspects of the dossier. The recommendations in the scientific advice have been followed.

The tolerance studies carried out by the company do not exactly meet the requirements established in VICH GL43. However, the deviations were supported by the CVMP in the scientific advice provided and are highlighted in the respective section.

MUMS/limited market status

Not applicable.

Part 1 - Administrative particulars

Detailed description of the pharmacovigilance system

The applicant has provided a detailed description of the pharmacovigilance system (dated December 2015) which fulfils the requirements of Directive 2001/82/EC. Based on the information provided, the applicant has the services of a qualified person responsible for pharmacovigilance and the necessary means for the notification of any adverse reaction occurring either in the Community or in a third country.

Manufacturing authorisations and inspection status

Manufacture of the dosage form, packaging and batch control take place within the EU.

Batch release takes place in Ceva Santé Animale, Libourne (France). The site has a manufacturing authorisation issued by the Agence Nationale du Médicament Vétérinaire - Agence Nationale de Sécurité Sanitaire de l'alimentation, de l'environnement et du travail (ANMV–ANSES).

GMP certifications, which confirm the date of the last inspection and show that all involved sites are authorised for the manufacture and/or batch release of such veterinary dosage forms, have been provided.

GMP declaration for each active substance manufacturing site was provided from the Qualified Person at the EU batch release site. The declarations were based on an on-site audit by the manufacturing site responsible for batch release.

Overall conclusions on administrative particulars

The detailed description of the pharmacovigilance system was considered in line with legal requirements.

The GMP status of all the active substance and finished product manufacturing sites has been satisfactorily established and are in line with legal requirements.

Part 2 - Quality

Composition

The finished product is presented as an aqueous suspension for injection containing 133.4 mg of iron (as gleptoferron 20% Fe) and 30 mg of toltrazuril per ml as the active substances.

Other ingredients are phenol, silica, povidone, sodium chloride, docusate sodium, simethicone emulsion and water for injections.

The product is available in multi-dose plastic vials containing 100 ml, 250 ml or 500 ml, as described in section 6.5 of the SPC.

Containers

The primary packaging for this product is translucent multi-layered plastic (polypropylene/ethylene vinyl alcohol/polypropylene) vials closed with type I bromobutyl grey stoppers coated with a

fluoropolymer film, and then sealed with aluminium closures (with plastic flip caps). The pack sizes of 100 ml, 250 ml and 500 ml have been justified.

The vials are constructed of a multi-layered plastic material designed to ensure suitable protection of the product and are composed of (inside to outside) polypropylene (PP), adhesive, ethylene vinyl alcohol (EVOH), adhesive, polypropylene (PP).

The stoppers are type I bromobutyl grey stoppers coated with a fluoropolymer film and they comply with the relevant European Pharmacopoeia (Ph. Eur.) monograph 3.2.9. (Rubber closures for containers for aqueous parenteral preparations, for powders and for freeze-dried powders). The vials are sealed with aluminium closures (with plastic flip caps).

Extraction and interaction studies have been carried out to assess the compatibility of the primary (plastic) packaging with the product. The studies were done according to the methodology described in Ph. Eur. 3.1.6 (Polypropylene for containers and closures for parenteral preparations and ophthalmic preparations) and the Guideline on plastic immediate packaging materials (EMEA/CVMP/205/04).

The plastic vials and the rubber stoppers are both sterilised by gamma-irradiation, and validation data in accordance with ISO 11137 have been provided which are suitable.

The choice of the container-closure system and the sterilisation dose range has been validated by stability data and is satisfactory for the intended use of the product.

Statements that the rubber stoppers comply with the relevant Ph. Eur. and EU requirements are included. The description of the secondary packaging materials of the finished product is also provided.

Development pharmaceutics

The objective of the development pharmaceutics was to develop a multi-dose parenteral product containing a combination of the two well-established active substances, iron (as gleptoferron 20% Fe) and toltrazuril, for administration to piglets by the intramuscular route.

The recommended dose to be administered is 200 mg of iron and 45 mg of toltrazuril per piglet, corresponding to 1.5 ml of the suspension for injection per animal in one single injection to be administered 24 to 96 hours after birth.

The active ingredient, iron (as gleptoferron 20% Fe), is frequently used in veterinary medicines in the form of a solution for injection for piglets.

The active ingredient toltrazuril is a frequently used molecule in veterinary medicines in the form of an oral suspension for piglets. (It is also authorised for other species.) The proposed formulation is a suspension for injection formulated with toltrazuril.

The physico-chemical characteristics liable to affect the bioavailability of toltrazuril are its particle size and crystalline form.

The excipients utilised within the formulation are well established and all are listed within the Ph. Eur. or in the USP. The list of excipients is included in section 6.1 of the SPC.

The concentration of the phenol preservative is justified by the antimicrobial preservative efficacy test data provided and also by data from preliminary stability studies.

The proposed manufacturing method is justified according to the Annex to the Note for guidance: Development pharmaceutics for veterinary medicinal products: Decision trees for the selection of sterilisation methods (EMEA/CVMP/065/99) and is acceptable.

The methods of sterilisation have been justified and are acceptable.

The packaging, the in-use stability and the antimicrobial preservation allow for the product to be presented in a multi-dose vial. The maximum number of possible punctures of the rubber closure has been studied and is reflected in the SPC accordingly.

Method of manufacture

The manufacturing process consists of preparation of the aqueous phase (which involves the dissolution in water for injections of sodium chloride, docusate sodium, povidone and the additional phenol required), addition and homogenisation of the gleptoferron 20% Fe to the aqueous phase addition of the toltrazuril and remaining excipients (simethicone emulsion and silica colloidal anhydrous sterile), filling, stoppering and crimping.

Information is provided regarding the sterilisation processes used and manufacturing parameters are well-defined and justified.

The process could be considered to be a complex non-standard manufacturing process. A range of commercial batch sizes have been proposed. The validation of the proposed manufacturing method has been satisfactory conducted on three full scale batches.

The sterilisation (by gamma-irradiation) of the primary packaging materials (vials and rubber closures) is described as part of the manufacturing process. The development and validation of the sterilisation of the vials is described.

The manufacturing process and in-process controls have been described in detail, and the suitability of the holding time proposed is confirmed with regard to GMP.

Control of starting materials

Active substances

Iron (as gleptoferron 20% Fe)

Gleptoferron is regarded as the active substance and it contains iron as the active moiety.

Gleptoferron is solely manufactured as an intermediate product (gleptoferron 20% Fe) which contains the excipients water and phenol (in addition to the gleptoferron). There is no clear distinction between the manufacture of the active substance and that of the intermediate product.

All the scientific documentation related to the preparation and controls of the active substance is discussed as a part of the manufacturing process of the intermediate product.

Toltrazuril

The chemical name of toltrazuril is 1-methyl-3-[3-methyl-4-[4-(trifluoromethyl)thio]phenoxy]-phenyl] 1,3,5-triazine-2,4,6-(1H,3H,5H)-trione.

Toltrazuril is a crystalline powder, soluble in acetone, sparingly soluble in acetonitrile, and practically insoluble in water.

Toltrazuril has a non-chiral molecular structure.

Polymorphism has been observed for this active substance.

The physico-chemical characteristics liable to affect the bioavailability of toltrazuril are its particle size

and crystalline form. Identification of the crystalline form produced is included in the specifications of the active substance. Studies have been performed to confirm the absence of any impact of manufacturing steps on the physico-chemical properties or degradation product levels of the toltrazuril.

The information on the toltrazuril active substance is provided in accordance with the Active Substance Master File (ASMF) procedure. Two active substance suppliers have each provided an ASMF. There is no monograph for toltrazuril in the Ph. Eur. and an in-house monograph has been defined for each of the manufacturers.

Specifications which are common for each of the 2 manufacturers have been defined for testing this active substance before its use in the medicinal product, except regarding the residual solvents, which are dependent of the manufacturing process of each manufacturer. The analytical methods included in the in-house specifications for each manufacturer will be used to control the toltrazuril active substance.

The toltrazuril specifications for both manufacturers include tests for identity (IR, melting point), appearance of solution, water content (Ph. Eur.), sulphated ash (Ph. Eur.), heavy metals, assay (HPLC), and impurities (HPLC). Specific parameters have been defined by the medicinal product manufacturer to control suitability of the active substance for use in the preparation of the finished product. Particle size and microbiological quality are controlled according to the specification defined in the manufacturer's in-house monographs.

Information regarding test methods, including updated descriptions and validation data, is provided for tests which are additional to the Ph. Eur. methods. Batch data from testing performed by the finished product manufacturer from both sources have been provided. Information regarding reference standards used by the finished product manufacturer is also provided.

The characterisation of the active substance is in accordance with the Guideline on the chemistry of new active substances (CPMP/QWP/130/96 Rev. 1). Described potential and actual impurities were discussed with regards to their origin and characterised. The in-process controls do include a suitable control of impurities.

Detailed information on the manufacture of the active substance has been provided in the restricted parts of the ASMFs. The active substance is synthesised using well-defined starting materials. The quality of water used in the last steps of the synthesis is confirmed to be suitable in both manufacturers. Defined potential and actual impurities were discussed with regards to their origin and characterised.

Stability data on batches of active substance from the proposed manufacturers stored in the intended commercial package under long term conditions at 25 °C/60% RH and for up to 6 months under accelerated conditions at 40 °C/75% RH, according to the VICH guidelines, were provided. Photostability testing, in accordance with VICH GL5, was also performed. Also, results from stress conditions, under high temperature, high humidity, light irradiation, and under acid, alkaline and oxidising conditions were provided.

Parameters controlled during the stability studies are those susceptible to change during storage and likely to influence quality, safety and/or efficacy. Regarding the results of the data available, no significant change or trend is observed in any of the tested parameters.

The stability results indicate that the toltrazuril manufactured by both of the two suppliers is sufficiently stable. The stability results justify the proposed retest periods of the active substance in the declared container-closure system.

Excipients

All the excipients are well known pharmaceutical ingredients and their quality is compliant with their respective current Ph. Eur. or USP monographs. The list of excipients is included in section 6.1 of the SPC.

Specific measures concerning the prevention of the transmission of animal spongiform encephalopathies

The product does not contain any materials derived from human or animal origin.

Valid TSE declarations from the manufacturers of the finished product have been provided.

Control tests during production

Iron (as gleptoferron 20% Fe)

Iron (as gleptoferron 20% Fe) could not be classified as an active ingredient product only as it contains the excipients water and phenol in addition to the gleptoferron.

The gleptoferron complex is a complex of iron (III) hydroxide oxide dextranglucoheptanoate hydrate. Although its complete structure is not available, the dossier enclosed an acceptable bibliographical proposal for the iron-dextran complex structure (London, E., J. of Pharmaceutical Sciences, vol. 95, nº 7, July 2004).

The molecular formula of gleptoferron is x FeH_2O.n $C_6H_{10}O_5$. $C_7H_{14}O_8$. y H_2O.

Iron and the polysaccharide dextran are the most important raw materials in gleptoferron.

There is no clear distinction between the manufacture of the active substance (gleptoferron) and that of the intermediate product (gleptoferron 20% Fe), as the manufacture takes place in an aqueous environment from the start. The documentation belonging to parts II.C and II.D of the dossier therefore cannot be clearly separated. The complete documentation is provided to justify the quality of both the active substance (gleptoferron) and the intermediate (gleptoferron 20% Fe).

The glucose units and the α-linkages are responsible for the chirality of gleptoferron. The resulting value of the specific optical rotation of the dextran chains is not measurable because of the strong inherent colour of the iron (III) hydroxide oxide dextranglucoheptanoate hydrate complex at a wavelength of 589.3 nm.

Regarding its physico-chemical characteristics liable to affect bioavailability, because gleptoferron forms a colloidal solution in the finished product polymorphism is not relevant.

There is no monograph for gleptoferron in the Ph. Eur. so an in-house specification has therefore been defined. This includes tests for identity (iron, dextran), purity (chloride), pH, non-volatile residue, relative density, kinematic viscosity content of iron (atomic absorption spectometry) and content of dextran (total organic carbon). All the methods are from the Ph. Eur.

The specification proposed for the intermediate, gleptoferron 20% Fe, is adequate and the test procedures are described in detail. The methods are all those of the Ph. Eur. and consequently their validation is not required.

Adequate in-process controls are applied during the synthesis/complexation/intermediate manufacture. The specifications and control methods for the intermediate products, starting materials and reagents have been presented.

Detailed information on the manufacture of the gleptoferron has been provided. The active substance is synthesised using well-defined starting materials. The starting material specifications are acceptable. Potential and actual impurities were discussed with regards to their origin and have been adequately characterised.

Detailed information on the manufacture of the intermediate, gleptoferron 20% Fe, has been provided.

Stability data from batches of the intermediate, gleptoferron 20% Fe were stored in the intended commercial package under long term conditions at 25 °C/60% RH for up to 6 months under accelerated conditions at 40 °C/75% RH according to the VICH guidelines were provided. Photostability testing was performed in accordance with VICH GL5.

The holding time proposed for the intermediate, gleptoferron 20% Fe, has been adequately justified.

Control tests on the finished product

The finished product specification is acceptable and includes the following parameters relevant to the dosage form: appearance, re-dispersibility, relative density, filling volume, identification of iron, identification of iron, identification of toltrazuril, identification of phenol, pH, identification of dextran, pH, dissolution, assay of toltrazuril, assay of phenol, total iron, dextran, toltrazuril degradation products, iron degradation products, and sterility.

The release and shelf life finished product specifications differ only in a couple of limits. Firstly, the assay for phenol content, in which wider limits are justified and used in the shelf life specification, and secondly, there are wider limits in the shelf life specification for the iron degradation product, free iron. The limit for content of free iron at release is as per the one previously justified for the active substance/intermediate and is considered to be acceptable and, the shelf life limit was justified by appropriate toxicological data.

Regarding toltrazuril degradation products, the stability data provided show no significant changes or trends on storage and therefore the limits in the release and shelf life specifications are the same and in accordance with the EMA Q&A on setting specifications for impurities in veterinary medicinal products.
(http://www.ema.europa.eu/ema/index.jsp?curl=pages/regulation/q_and_a/q_and_a_detail_000072.jsp&mid=WC0b01ac058002c2b0#section7).

The control methods are well described and appropriately validated in accordance with VICH guidelines.

Batch analysis results are provided for three industrial size batches and these confirm the consistency of the manufacturing process and its ability to manufacture to the intended product specification.

Stability

Stability data were provided from three batches of the finished product stored under long term conditions for 24 months at 25 °C/60% RH and for up to 6 months under accelerated conditions at 40 °C/75% RH, according to VICH GL3 (Stability testing of new veterinary drug substances and medicinal products). The batches of product were identical to those proposed for marketing and were packed in the commercial primary packaging. The batches used in the study were manufactured with toltrazuril sourced from each of the two suppliers.

Samples were tested for appearance, relative density, toltrazuril content, phenol content, total iron, toltrazuril degradation products and iron degradation products, and pH. Sterility testing was carried out

at the start then after 6 months in the accelerated study and after 36 months in the long term study. Dissolution, efficacy of antimicrobial preservation (challenge test), and packaging tests (appearance, evaluation of plastic additives and IR spectra) were studied during the development studies.

The analytical procedures used are stability indicating and have been appropriately validated.

In addition, two batches of the product were exposed to light as defined in VICH GL5 (Stability testing: photostability testing of new veterinary drug substances and medicinal products). The results showed that the medicinal product is stable in the primary package (translucent multi-layer plastic vials).

No significant changes were observed in any of the parameters tested.

Additional information has been submitted to demonstrate that the medicinal product can withstand low relative humidity conditions.

In-use stability studies have been conducted on samples from a pilot scale batch, at the beginning and at the end of shelf life, stored for 4 weeks (28 days) after the vial had been first broached. The suspension was proven to be stable in freeze-thaw studies.

Finally, the following commitment is provided in the dossier:

- The on-going stability study will be continued with all the three batches stored at 25 °C/60% RH for up to 36 months.

The proposed shelf life of 3 years, without any special storage conditions, is acceptable.

Overall conclusions on quality

The finished product is a fixed combination product containing 133.4 mg of iron (as gleptoferron 20% Fe) and 30 mg of toltrazuril per ml as the active substances which is presented as an aqueous suspension for injection.

Other ingredients in the formulation are phenol (antimicrobial preservative), silica (colloidal anhydrous) and povidone (suspending agents), sodium chloride, docusate sodium and simethicone emulsion (surfactant and antifoaming agents) and water for injections.

The product is presented in translucent multi-layered plastic (polypropylene/ethylene vinyl alcohol/polypropylene) vials containing 100 ml, 250 ml or 500 ml, closed with type I coated bromobutyl stoppers coated with a fluoropolymer film, and then sealed with aluminium closures (with plastic flip caps), as described in section 6.5 of the SPC. The plastic vials and the rubber stoppers are both sterilised by gamma-irradiation. The choice of the container-closure system and the sterilisation dose range used, have been validated by stability data and are adequate for the intended use of the product.

The manufacturing process and in-process controls have been described in detail, and the suitability of the holding times are confirmed by means of a GMP inspection.

The manufacturing process for the finished product could be considered to be a non-standard manufacturing process. The choice of sterilisation process is justified according to the Annex to the Note for guidance: Development pharmaceutics for veterinary medicinal products: Decision trees for the selection of sterilisation methods (EMEA/CVMP/065/99) and is acceptable.

Iron (as gleptoferron 20% Fe) could not be classified as an active ingredient as it also contains the excipients water and phenol in addition to the active substance. Complete scientific documentation related to the quality of the active substance preparation (gleptoferron) and the intermediate

(gleptoferron 20% Fe) has been provided and is discussed together.

The information on the active substance, toltrazuril, is provided in accordance with the ASMF procedure. Two different sources of toltrazuril have been used, each has a different ASMF. Detailed information on the manufacture of the active substance has been provided in the restricted parts of the two ASMFs. Information on the manufacture and controls of the active substance from each of the two different sources have been presented and are satisfactory.

The stability results indicate that the toltrazuril manufactured by both of the two suppliers is sufficiently stable.

The excipients are well known pharmaceutical ingredients and their quality is compliant with their respective current Ph. Eur. or USP monographs.

The finished product specification is acceptable and includes parameters relevant to the dosage form. Batch analysis results are provided for three industrial size batches and these confirm the consistency of the manufacturing process and its ability to manufacture to the intended product specification.

The proposed shelf life and storage conditions of the finished product, 3 years without any special storage conditions, are acceptable.

In general, comprehensive and clear information has been provided in the dossier to support the authorisation of this medicinal product, and current regulations and guidelines have been taken into account.

Part 3 – Safety

The active substances of Forceris are toltrazuril, an antiprotozoal substance, and iron (as gleptoferron), an anti-anaemic substance, and these are a new fixed combination of active substances not authorised for a veterinary medicinal product in the EU before. A full safety file in accordance with Article 13b of Directive 2001/82/EC - a fixed combination application, has been provided.

Safety documentation

A comprehensive data package with recently performed studies and data from current literature has been presented by the applicant. The pharmacological and toxicological properties of toltrazuril and iron have been characterized and described in the dossier based on available literature and laboratory studies performed by the applicant.

Pharmacodynamics

See part 4.

Pharmacokinetics

See part 4.

Toxicological studies

Toltrazuril: The active substance toltrazuril was previously assessed by the CVMP in the context of the establishment of MRLs. The key findings of the toxicity studies evaluated (see European Public MRL Assessment Report (EPMAR) (EMA/MRL/314/97-FINAL)) are summarised below.

Iron: Although iron dextran and iron glucoheptonate both have "No MRL required" classifications in Regulation 37/2010, EPMARs are not available for these substances. Published literature references have been provided to describe the toxicity of iron. The key findings of the toxicity studies are summarised below.

Toltrazuril and iron have low acute toxicity potential. The LD_{50} of toltrazuril by oral route in rat is 2000 mg/kg bw. The LD_{50} of iron dextran is 1000 mg/kg bw oral in mouse, 450 mg/kg bw IM in mouse, and 690 mg/kg bw IM in rabbit.

In repeat dose toxicity studies with toltrazuril an oral NOEL of 1 mg/kg bw/day from a 90-day study in the rat (the most sensitive species) was determined based on a decrease of the weight gain and the daily food intake. Iron accumulates in the liver, the target organ, when it is in excess after repeated administrations of high doses.

With respect to reproductive toxicity, toltrazuril was administered orally in rats during two generations. The number of still-born pups increased in all treated groups but the statistical significance was only observed for the highest dose and for the first generation at the lowest dose. The LOEL was set at the lowest dose of 0.3 mg/kg bw/day due to these equivocal results. In studies of developmental toxicity, a NOEL of 3 mg/kg bw/day (maternotoxicity) and 10 mg/kg bw/day (embryotoxicity) for rat, and 0.5 mg/kg bw/day for rabbit were established. No data are available on iron.

Toltrazuril did not show genotoxic activity. No data are available for iron. In carcinogenic studies with toltrazuril, carcinogenic findings were considered species-specific, as rodents are known to be more sensitive to hormonal changes than humans. NOELs of 3 mg/kg bw/day and 1 mg/kg bw/day were established for neoplastic tumours and pre-neoplastic lesions for rat, respectively. Iron dextran is anticipated to be a human carcinogen by the IARC and was classified in group 2B (possibly carcinogenic to humans). However, the carcinogenic effects are considered to be associated with high local concentrations, such as arise following injection, but are not considered relevant to the human consumer, who would be exposed by the oral route.

Tolerance in the target species of animal

See part 4.

Studies of other effects

Two studies evaluating the dermal and ocular irritancy of the veterinary medicinal product are reported.

Dermal irritation potential of Forceris was evaluated in an in vivo study in New Zealand White rabbits. The product was found to be non-irritant to the skin.

Eye irritation potential of Forceris was evaluated in an in vivo study in New Zealand White rabbits. The test item is a slight irritant for the eye.

Iron dextran is known to be a sensitizer in humans. Toltrazuril is not considered to be sensitising according to the information provided. The performance of a sensitization study with the final formulation is considered not necessary. Further useful information is not expected from a new study in animals.

Excipients

Phenol, sodium chloride, docusate sodium, simethicone emulsion, silica colloidal and povidone are common excipients in injectable formula. The available toxicological data on the excipients indicate that a toxicological reference value ($BMDL_{05}$ 98.7 mg/kg bw/day) is allocated for the developmental toxicity of phenol (EFSA CEF Panel, 2013). In addition, there might be local irritation and/or granulomatous reactions related to simethicone and silica after accidental self-injection. These data have been taken into account in the user safety.

User safety

A user safety risk assessment has been conducted in accordance with CVMP guideline EMEA/CVMP/543/03-Rev.1.

The main potential routes of accidental contact with the product have been considered and it was concluded that the most likely are those of dermal and/or eye exposure and accidental self-injection.

The dermal irritation study confirmed that the product does not cause effects in the test animals used. However, the eye irritation study demonstrated that Forceris is a slight irritant to the eye in rabbits. A corresponding precautionary statement is proposed on the product information.

The qualitative risk characterisation also demonstrates that local risk might be expected after self-injection. The quantitative risk characterization shows that the MOEs for dermal and accidental injection exposure to toltrazuril are not acceptable for both systemic and developmental toxicity. Calculated MOEs vary from 1.2 to 4 considering the NOEL = 1 mg/kg bw/d (3-month oral study) and LOEL =0.3 mg/kg bw/d for developmental toxicity.

Based on this outcome the following warnings and measures are considered appropriate:

> "People with known hypersensitivity to iron gleptoferron or any of the excipients should avoid contact with the veterinary medicinal product.
>
> Exposure to the veterinary medicinal product may cause eye irritation or adverse effects to the skin. Avoid skin and eye contact with the product. In case of accidental exposure to the skin or eyes, wash the affected area with water.
>
> Accidental self-injection may cause local reactions such as irritation, granulomas, or severe anaphylactic reactions in sensitive people. Care should be taken to avoid accidental self-injection. In case of accidental self-injection, seek medical advice immediately and show the package leaflet or the label to the physician.
>
> This product may be harmful for the unborn child. Pregnant women and women intending to conceive should avoid contact with the veterinary medicinal product, especially accidental self-injection.
>
> Wash hands after use."

Based on the above risk assessment the CVMP concluded that the product does not pose an unacceptable risk to the user when used in accordance with the SPC

Environmental risk assessment

A Phase I environmental risk assessment (ERA) was provided according to the CVMP/VICH guidelines. The Predicted Environmental Concentration for soil was calculated for toltrazuril in accordance with

VICH guideline GL6 and the CVMP guideline on the Environmental Impact Assessment for Veterinary Medicinal Products in support of the VICH guidelines GL6 and GL38 (EMEA/CVMP/ERA/418282/2005-Rev.1). As these guidelines do not give details on the calculation of PEC soil for piglets, the applicant has justified the use of the number of piglets raised per place and year.

Exposure assessment	Value (µg/l – µg/kg)
PEC soil	9 µg/kg

The environmental risk assessment can stop in Phase I and no Phase II assessment is required because:

- Iron is a natural substance, the use of which will not alter the concentration or distribution of the substance in the environment.
- The initial predicted environmental concentration of toltrazuril in soil (PECsoil, initial = 9 µg/kg) is less than 100 µg/kg.

Given the potential of leaching to groundwaters of the main metabolite of toltrazuril (ponazuril) the possibility of a tailored phase II assessment was discussed. However, ponazuril was considered in a previous assessment by CVMP, under an Article 35 referral on toltrazuril, and the conclusion of that assessment was that the concentrations that may occur in groundwater do not pose a risk.

Conclusions on the environmental risk assessment

An ERA was provided according to the CVMP/VICH guidelines. Based on the data provided the ERA can stop at Phase I. Forceris is not expected to pose a risk for the environment when used according to the SPC.

Residues documentation

MRLs

The MRL status of the constituents of Forceris is as follows:

Pharmacologi-cally active substance	Marker residue	Animal species	MRL	Target tissues	Other provisions	Therapeutic classification
Toltrazuril	Toltrazuril sulfone	All mammalian food producing species	100 µg/kg 150 µg/kg 500 µg/kg 250 µg/kg	Muscle Fat Liver Kidney	For porcine species the fat MRL relates to 'skin and fat in natural proportions'. Not for use in animals from which milk is produced for human consumption. Not for use in	Antiparasitic agents/ Agents acting against protozoa
		Poultry	100 µg/kg 200 µg/kg 600 µg/kg 400 µg/kg	Muscle Skin and fat Liver Kidney		

Pharmacologi-cally active substance	Marker residue	Animal species	MRL	Target tissues	Other provisions	Therapeutic classification
					animals from which eggs are produced for human consumption.	
Iron (as gleptoferron)	The "No MRL required" classification for iron dextran and iron glucoheptonate is considered to apply to gleptoferron as gleptoferron is expected to release iron dextran and iron glucoheptonate.					

The excipients listed in section 6.1 of the SPC are either allowed substances for which table 1 of the annex to Commission Regulation (EU) No 37/2010 indicates that no MRLs are required or are considered as not falling within the scope of Regulation (EC) No 470/2009 when used as in this product.

Residue studies

Pharmacokinetics

Data from three studies in the target species show the absorption profile of the product following the administration of the recommended single dose. The following parameters were derived from these studies: toltrazuril (C_{max}: 7 mg/l; T_{max}: 6 days; $T_{1/2}$: 3.5 days), toltrazuril sulfone (C_{max}: 9 mg/l; T_{max}: 13 days; $T_{1/2}$: 3.7 days); iron (C_{max}: 645 µg/ml; T_{max}: 0.5 days; $T_{1/2}$: 21 days). Bibliographic data about distribution, metabolism and excretion were provided, showing species-specific properties for both substances.

Depletion of residues

Two GLP compliant tissue depletion studies for piglets were provided.

The first study was designed as a marker residue study to determine toltrazuril sulfone concentrations in muscle, liver, kidney skin/fat and the injection site of the target animals and to derive withdrawal periods. The target animals were treated with Forceris at the dose and by the route of administration intended for marketing. A sufficient number of animals (4 animals/group) and slaughter time points (5 points: 56, 70, 84, 98 and 112 days post-treatment) were investigated.

Piglets (0.9–2.18 kg bw) were treated by a single intramuscular administration with 50 mg/kg bw of toltrazuril and 222 mg/kg bw of iron. The maximum recommended volume (1.5 ml) was administered. Tissue residues were determined up to 112 days post dose using a validated HPLC-UV method (LOQ: 50 µg/kg for muscle, 125 µg/kg for kidney, 250 µg/kg for liver and 75 µg/kg for fat/skin). All values of toltrazuril sulfone residues at all times and in all tissues are well below the MRLs for all animals. At the first slaughter time, 56 days, all animals have residues below the LOQ in each tissue.

In order to complete the information on the depletion of residues at slaughter time points earlier than 56 days, as well as to confirm that the marker residue at the injection site is toltrazuril sulfone, a second residue study in piglets was submitted.

The concentration of toltrazuril sulfone and toltrazuril were analysed in core and surrounding injection sites, whereas toltrazuril sulfone was analysed in loin muscle, kidney, liver and skin/fat tissues. Piglets

(1.068–2.343 kg bw) were treated by intramuscular route at the higher proposed dose of 50 mg toltrazuril and 222 mg iron/kg bw. Five animals/group and 4 slaughter time points post-administration were studied (28, 42, 56 and 70 days). Tissue residues were determined using a validated HPLC-UV method. The marker residue, toltrazuril sulfone, as well as the parent compound, toltrazuril, were analysed at the core and surrounding injection site. The residues of toltrazuril sulfone at 56 days after the treatment were below the MRL in all animals. It is confirmed that toltrazuril sulfone is the marker residue at the injection site since residues of toltrazuril are below the LOQ at early sample points. Taking into account all tissues, toltrazuril sulfone residues are below the corresponding MRL in loin muscle, liver and kidney at 56 days post-treatment, and only one of five animals had residues above the MRL in skin/fat tissue at this time, remaining below the LOQ at 70 days post-treatment in all tissues.

Analytical method

Toltrazuril sulfone and toltrazuril were analysed by a properly validated HPLC-UV method. The following parameters were studied: linearity, specificity, selectivity, recovery, LOD, LOQ, accuracy, precision, carry-over effect and stability of solutions.

Withdrawal periods

Iron was not assayed. However, consideration is given to the low level of toxicity of this compound in comparison to toltrazuril, and the "No MRL required" status for this active substance. It is considered appropriate that the elimination of toltrazuril will determine the withdrawal period for piglet meat and offal.

With respect to toltrazuril, based on data of the second residue study provided (slaughter time points: 28, 42, 56 and 70 days post-treatment), the limiting tissue as regards residue depletion is the skin/fat. The conclusion on the withdrawal period determination has been based on the following facts:

- When applying the statistical method in accordance with current guidelines, the assumption for homogeneity of variance (Cochran-test, $0.01<p<0.05$ and Bartlett-test, $0.05>p>0.025$) was not met for skin/fat. Consequently the statistical method could not be used and the alternative method was therefore used instead.

- A withdrawal period of 70 days without the addition of a safety span is considered acceptable. This was based on several considerations. It was noted that at the previous timepoint (some 14 days earlier, that is, at day 56) only one data point was above the MRL and only for fat/skin. Also, at 70 days all animals had toltrazuril residues below the LOQ in all tissues.

For these reasons, a withdrawal period of 70 days, derived using the alternative method, can be considered as safe for the consumer.

Overall conclusions on the safety and residues documentation

Pharmacology:

Toltrazuril is a triazinone derivative, active against all intracellular development stages of coccidia, by entering through the host cell membrane and cytoplasm.

Iron is an essential micronutrient having a major role in the transport of oxygen via haemoglobin and myoglobin and in redox reactions as cofactor.

After intramuscular administration toltrazuril maximal concentration is reached 6 days after administration (T_{max} ranging from 4 to 7 days). Toltrazuril is mostly transformed to toltrazuril sulfone and eliminated slowly with a half-life of 3.5 days. The principal route of excretion is in faeces.

After intramuscular injection, iron is absorbed rapidly from the injection site into the capillaries and the lymphatic system. Since iron is recycled in the body little of the absorbed iron is excreted. Very small losses occur in faeces, sweat and urine.

Toxicology:

The toxicity of the active substances, toltrazuril and iron, has been mainly documented providing some scientific reports.

Toltrazuril has a low acute toxicity. After 3 month oral treatment the lowest value of NOEL in rat is 1 mg/kg bw/day. A NOEL of 0.5 mg/kg bw for developmental toxicity was established in rabbits after oral administration (the most sensitive species). Toltrazuril did not show any mutagenic activity in the regulatory battery of tests. Toltrazuril is not used in human medicine.

Iron has a low acute toxicity. Chronic repeated high doses of iron lead to an accumulation of iron in liver. High dose of iron dextran may cause increased incidences of teratogenicity and embryotoxicity in laboratory animals, but no effect was observed after single intramuscular dose below 125 mg/kg bw. Iron is used in human medicine for treatment of iron deficiency and by parenteral route a dose of 1.7 to 3.3 mg/kg bw. Symptoms of overdose are associated with gastrointestinal effects, and serious hypersensitivity reactions including life threatening and fatal anaphylactic/anaphylactoid reactions have been reported in patients receiving parenteral iron dextran.

Forceris can be considered as non-irritant to skin whereas it was considered slightly irritant for the eyes, based on two studies with rabbits.

The data presented are considered adequate to characterise the toxicity profile of the actives substances and product.

User safety:

A user safety assessment in line with the relevant guidance was provided. The qualitative risk characterisation demonstrates that local risk might be expected after eye exposure and self-injection. The quantitative risk characterisation shows that the MOEs for dermal and self-injection exposure to toltrazuril are below acceptable levels for both systemic and developmental toxicity. Based on this outcome appropriate warnings and measures have been included in the product literature. The product does not pose an unacceptable risk to the user when used in accordance with the SPC.

Environmental risk assessment:

An appropriate environmental risk assessment was provided. The product is not expected to pose a risk for the environment when used according to the SPC.

Residues:

In a non-radiolabelled residue depletion study in piglets, toltrazuril sulfone concentrations were below the MRL in all tissues (including the injection site), except skin/fat tissue at 56 days after-treatment. The limiting tissue as regards residue depletion is the skin/fat. A withdrawal period of 70 days (the first time when all the residues are below the MRL) is considered acceptable.

Part 4 – Efficacy

Justification of the fixed combination

Forceris is a fixed combination containing toltrazuril and iron (as gleptoferron) in a suspension for intramuscular injection in piglets weighing from 0.9 to 3 kg on day 1 to day 3 after birth *i.e.* 24 to 96 hours after birth, at a fixed dose of 45 mg toltrazuril and 200 mg iron per piglet (i.e. 1.5 ml of product per piglet). The product is indicated for the concomitant prevention of iron deficiency anaemia and prevention of clinical signs of coccidiosis (diarrhoea) as well as reduction in oocyst excretion, in piglets in farms with a confirmed history of coccidiosis caused by *Cystoisospora suis* (*C. suis*). Toltrazuril (Baycox 5%) and iron as gleptoferron (Gleptosil) are already authorised for this use separately as an oral suspension at a dose of 20 mg toltrazuril/kg bw in piglets between 3 and 5 days of life and as an intramuscular solution at a dose of 200 mg iron/piglet in piglets within the first 3 days of life, respectively.

The present application is based on iron's well-established use, public documentation on both active substances and original *in vivo* studies performed in neonatal piglets with the Forceris formulation intended to be marketed (2 dose confirmation studies and the pivotal field study), as well as *in vivo* studies performed to confirm the new proposed route of administration for toltrazuril (intramuscular route) and establish the fixed dose of toltrazuril by this route.

A satisfactory justification for the combination product, in accordance with the CVMP Guideline on pharmaceutical fixed combination products (EMEA/CVMP/83804/2005), was provided on some aspects of the use of this fixed combination versus combined use of single substances:

- A clinical benefit, since almost 100% of fast-growing piglets need parenteral iron supplementation and coccidiosis is a widespread disease. In farms where piglets are at risk of developing coccidiosis, these animals also need to be treated with iron in their first days of life;
- A practical benefit for the breeder, i.e. an injection of iron associated with an oral administration of toltrazuril will be replaced by a single injection of the new fixed combination product;
- The new formulation will also improve animal welfare since neonatal piglets are manipulated very often during the neonatal period (navel cord care, teeth clipping, tail docking, ear notching for identification, castration of males and cross-fostering). Thus, this new product reduces one management procedure of piglets.

Moreover, the non-inferiority of the fixed combination product (final formulation) with an iron mono-product was demonstrated in the EU pivotal field study. The efficacy of the fixed combination product against coccidiosis was also shown in this study and was compared to the efficacy of the control toltrazuril mono-product (Baycox) in a well-conducted experimental study (see description of the studies thereafter).

Pharmacodynamics

Forceris is a fixed combination containing toltrazuril and iron (as gleptoferron) in a suspension for intramuscular injection in piglets. Toltrazuril (Baycox 5%) and iron as gleptoferron (Gleptosil) are already authorised for this use separately as an oral suspension and as an intramuscular solution, respectively.

Toltrazuril and iron are well-known active ingredients and therefore the pharmacodynamics of both substances has been documented based on the available literature. Thus, toltrazuril, a symmetrical

triazinone, has been reported to be effective against coccidia. It is a broad-spectrum anticoccidial substance, chemically unrelated to all other known anticoccidials. On the other hand, iron is an essential element for the function of all body cells, playing a critical role in cell-cycle regulation, electron transport in the respiratory chain, DNA synthesis and other metabolic reactions. In the respiratory chain located in mitochondria, the change of valence of iron in cytochromes and cytochromes oxidase results in an electron transport and establishes a proton gradient through the inner membrane of the mitochondria, a prerequisite for the synthesis of adenosine triphosphate (ATP), necessary for endergonic processes.

Development of resistance

The applicant presented a bibliographical search where no resistance to toltrazuril in pigs was reported for *C. suis* in the field; however, a recent case of resistance was observed in a Dutch pig farm. Also, resistance to toltrazuril is well-known for poultry coccidiosis. Because of the long-term use of toltrazuril and the absence of alternative treatments, resistance to toltrazuril in pig farms could become a problem as in poultry. Therefore, this new combination product must be used only when the indication is well justified. Nevertheless, there is no indication in the scientific literature of a sudden increase in the spread and severity of toltrazuril resistance in the past two years. Moreover, preventive measures against anticoccidial resistance such as the need for hygienic measures have been appropriately addressed in section 4.4 of the SPC.

Pharmacokinetics

The Guideline on pharmaceutical fixed combination products (EMEA/CVMP/83804/2005) clearly states the necessity to provide preclinical data (pharmacokinetic and/or pharmacodynamic) for the combination product to demonstrate its mode of action, investigate possible interactions or clearly establish that interactions do not occur. The applicant has performed pharmacokinetic studies in piglets of 2 days of life, which corresponds to the mean age at which animals would be treated. Pharmacokinetic profiles are not expected to differ significantly in piglets of 1, 2 or 3 days of life. Thus, this approach follows the Guidelines for the conduct of pharmacokinetic studies in target animal species (EMEA/CVMP/133/99-FINAL), section 3.1, and the pharmacokinetic studies are representative of neonates between 1 and 3 days of life. Moreover, this experimental design for pharmacokinetic studies was previously agreed in the scientific advice provided by the CVMP (EMA/CVMP/SAWP/285834/2014).

The applicant has carried out a series of GLP-compliant studies to support the pharmacokinetics of Forceris, taking into account that it is a fixed combination product. In all these studies, the methodology used is considered appropriate for pharmacokinetic studies. Thus, the product was administered and blood specimens were collected many times from Day -1 until Day 55 post-treatment. Each animal was observed at least once daily and any abnormal finding was recorded. Toltrazuril, toltrazuril sulfone and total iron determination was performed using validated methods. Finally, a non-compartmental analysis was performed to determine the following pharmacokinetic parameters for iron, toltrazuril and toltrazuril sulfone: C_{max}, t_{max}, V_z, AUC_{0-t}, $AUC_{0-\infty}$, $t_{1/2}$. Dose normalised parameters, C_{max}/dose and AUC/dose were also calculated. Baycox and Gleptosil were used as control products.

An original pilot PK study was performed in order to evaluate the impact of the dose (mg/kg bw or mg/animal) after intramuscular administration of two Forceris formulations in 2 day-old piglets. The main results can be summarized as follows:

- the administration of toltrazuril by intramuscular route is safe for young piglets at doses up to 52.6 mg/kg bw;
- the administration of toltrazuril by intramuscular route delayed its absorption by about 2 days compared to oral route;
- the bioavailability of toltrazuril was increased by about 33% when administered by intramuscular route at the dose of 20 mg/kg bw compared to oral route;
- after intramuscular administration of toltrazuril, the kinetic profile of its major metabolite, toltrazuril sulfone, also clinically active on all stages of the endogenous parasite stages, was close to the profile observed after oral administration, taking into account the time of administration (Baycox was administered 3 days later than Forceris) and suggested a clinical efficacy at least comparable to oral administration of toltrazuril;
- administration of 200 mg iron/piglet is the required dose to avoid anaemia in piglets as it has been described extensively in the literature.

According to the Guideline on pharmaceutical fixed combination products (EMEA/CVMP/83804/2005), it is necessary to investigate any possible interaction between active ingredients (toltrazuril and iron as gleptoferron in this case) or clearly establish that interactions do not occur. A three-way study would normally be necessary to establish the absence of interaction between toltrazuril and iron, with one group receiving the fixed combination product and two groups receiving one of the active ingredients. Taking into account the information available, a group not being administered iron is not possible considering that piglets, in all cases, need to receive iron in the first days of life to avoid anaemia; thus, a toltrazuril-alone group would not be feasible from an ethical point of view. Moreover, the plasma kinetics should be followed for several weeks since toltrazuril and its active metabolite, toltrazuril sulfone, have a long plasmatic half-life (3-4 days); however, iron administration cannot be delayed for weeks in piglets. As a conclusion, the absence of a group treated with toltrazuril only is scientifically sound and this was acknowledged in the scientific advice provided by the CVMP.

An original GLP interaction study was performed in order to determine the pharmacokinetic profile of iron administered intramuscularly in young piglets, alone (as Gleptosil) or in association with toltrazuril (as the Forceris final formulation intended for marketing).

As recommended in the scientific advice provided, this study was conducted on 2-day-old piglets, using the methodology previously described for pharmacokinetic studies. Following a single intramuscular injection of 200 mg of iron per animal, total iron plasma concentrations increased rapidly to reach a peak 12 h after administration, both for the control item (Gleptosil) and the test item (Forceris). Then, iron plasma concentrations decreased more or less rapidly to reach a constant baseline level around 1 µg/ml. This baseline concentration was maintained until Day 55 (last blood sampling time). Following the single intramuscular administration of iron, it appeared that the mean C_{max} and AUC of iron were significantly decreased by about 40% and 70%, respectively, when administered in association with toltrazuril (Forceris) compared to alone administration as control item (Gleptosil).

These results indicated that the rate and extent of iron absorption were significantly decreased when iron was administered as the test item (Forceris). Thus, it seemed that this combination with toltrazuril could negatively affect the bioavailability of iron. A possible explanation for this could be that the distribution of iron into the extravascular compartment was more rapid when iron was administered as the test item (Forceris) compared to the control item (Gleptosil). Further investigations were considered necessary to explain this phenomenon and to evaluate the impact of iron supplementation on efficacy.

Regarding toltrazuril, it appeared that after a single intramuscular administration of 45 mg per animal, its absorption was slow, maximal plasma concentration being observed 6 days after administration. Then, plasma concentrations decreased slowly and toltrazuril was slowly eliminated with a mean terminal half-life of about 3.5 days (*i.e.* about 84 h). Toltrazuril sulfone, the main metabolite of toltrazuril, appeared in plasma between 1 and 2 days after injection, and the maximal concentration was observed 13 days after injection. Toltrazuril sulfone was eliminated more slowly, with a mean terminal half-life of about 3.7 days (*i.e.* about 89 h).

In summary, this study showed that the bioavailability of iron was significantly decreased when it was administered in association with toltrazuril as the Forceris formulation, compared to alone administration as Gleptosil, following a single intramuscular injection in young piglets. Unfortunately, no determination of blood parameters was carried out in order to investigate if this apparently negative pharmacokinetic effect had an impact on blood parameters. Thus, at that stage of the development of Forceris, it seemed that the combination could negatively modify the pharmacokinetic profile of iron and, as a consequence, it could affect the efficacy of this combination in regards to prevention of anaemia in piglets.

In order to thoroughly investigate the apparently faster iron distribution observed after Forceris administration compared to the control product Gleptosil, an additional original GLP interaction study was performed, focusing on iron tissue distribution and haematology parameters.

The Forceris formulation used in this study was the final formulation intended for marketing. As recommended in the scientific advice received, this study was conducted on 2-day-old piglets. After a single intramuscular administration of 200 mg of iron per animal to young piglets, plasma concentrations of iron increased rapidly to reach a peak between 6 and 12 h after dosing, regardless of the administered formulation. Then, plasma concentrations decreased more or less rapidly depending on formulation. When comparing the different formulations, it appeared that a significant formulation effect was elicited on the extent of iron absorption. For the Forceris formulation, the extent of iron absorption was significantly decreased by about 50% compared to the Gleptosil formulation. Moreover, toltrazuril did not seem to modify significantly the rate and extent of iron absorption, regardless of the administered formulations. However, although a significant formulation effect was elicited on the pharmacokinetics of iron, no difference could be clearly established between treatments for haemoglobin, haematocrit and red blood cells levels. However, the mean values of blood parameters seemed to reach the baseline level slightly more rapidly after Forceris administration compared to other treatments and especially Gleptosil (*i.e.* between D3-D4 with Forceris *vs.* D5-D6 with Gleptosil for haemoglobin and haematocrit and between D6-D7 with Forceris *vs.* D7-D10 with Gleptosil for red blood cells).

It can be concluded from this study that, despite a significant formulation effect on the pharmacokinetics of iron, no pharmacodynamic effect was elicited for haemoglobin, haematocrit and RBC profiles. This effect is probably mainly due to a rapid distribution of iron to the tissues, leading to a rapid synthesis of haemoglobin. Moreover, a higher C_{max} with Forceris compared to the control product Gleptosil was not observed. Consequently, the Forceris formulation did not release more free-iron than Gleptosil.

In order to explain the differences observed in previous studies with regard to iron pharmacokinetic profile with Forceris formulation, another original PK study was performed to evaluate the distribution of iron at injection site and in physiological iron storage organs, *i.e.* liver and spleen, following a single intramuscular administration of iron alone (Gleptosil and Forceris formulation containing only iron) or in association with toltrazuril (Forceris final formulation intended for marketing) in young piglets. From this study, the following conclusions could be drawn:

- iron is not trapped at the injection site core after administration of Forceris. This point is relevant even if the animals are beginning the rearing period when this medicinal product is administered;
- toltrazuril impacts neither iron absorption, nor iron distribution into the tissues. Thus, there is no pharmacokinetic interaction between the two active ingredients of this fixed combination product;
- iron seems to be distributed quicker in liver and spleen, the usual iron storage organs, after administration of Forceris formulation and this confirms the results obtained in study on iron blood levels. Thus, there is an interaction between iron and Forceris formulation that it is not linked with the presence of toltrazuril.

This interaction resulting in the increase of the concentration of iron in organs (during the first kinetic times) after the administration of Forceris may predispose piglets to a higher rate of bacterial infections versus the administration of iron alone. However, this effect was not observed when the product was administered 1 or 2 times the recommended dose, as demonstrated in tolerance studies (see below).

In conclusion, the applicant has provided robust information that allows characterising precisely the pharmacokinetics of toltrazuril and iron (the two active ingredients of Forceris) not only in plasma, but also in tissues in the case of iron. In particular, information has been provided on absorption and distribution of the two active substances in combination. However, no information has been submitted about metabolism and excretion of these active ingredients. The CVMP considers that it is not necessary for the applicant to provide new data since this information is available in the public domain for both active ingredients.

In summary, the information provided by the applicant on the pharmacokinetics of Forceris is in line with the requirements and is considered acceptable.

Dose justification

Given that no interaction between the active substances has been demonstrated (see above), no dose justification/determination studies for iron were carried out by the applicant. The choice of the fixed dose of 200 mg iron/piglet by intramuscular route was based on the well-established use of iron by this route for the prevention of iron deficiency anaemia in neonatal piglets (i.e. piglets in the first 3 to 4 days of age), similarly to the proposed target population of Forceris.

Concerning toltrazuril dose, the applicant explored the efficacy of 20 and 40 mg/kg bw doses administered by intramuscular route for the prevention of clinical signs of coccidiosis in neonatal piglets in an experimental infection study. In addition, dose titration and dose confirmation studies have been performed (see thereafter) to justify the proposed recommended dose of toltrazuril.

In the infection study, a total of 37 healthy piglets were selected after completion of farrowing (SD -2) and included in the study. All the piglets were injected with iron (Gleptosil, 1 ml/piglet, equivalent to 200 mg iron per piglet). At the age of two days (SD0), the piglets were randomly allocated to 4 treatment groups: 20 mg toltrazuril/kg bw intramuscularly (n=9), 40 mg toltrazuril/kg bw intramuscularly (n=11), 20 mg toltrazuril/kg bw orally (n=9) and negative control (n=8).

Three days after treatment (SD3), the piglets were orally challenged with a well-known and characterised strain of *Cystoisospora suis*. Safety, clinical and parasitological parameters were followed for 26 days post-treatment.

The treatments (oral and injection) were well tolerated. No general or local safety issue was observed immediately and after administration of the treatments.

McMaster countable oocyst excretion was observed in all piglets of the negative control group and in none of the piglets from the treatment groups (100% reduction in oocyst excretion). The average faecal score increased above 2 in the control group from Day 5 to Day 12 post-infection while faecal consistency was unchanged in all experimentally infected and treated piglets. The incidence of diarrhoea was 100% in control group with an average duration of 7.0 days whereas diarrhoea was fully prevented in treated groups.

This proof of concept study confirmed the efficacy of toltrazuril administered either intramuscularly or orally, at the same dose for both routes (*i.e.* 20 mg/kg bw), as well as the safety of toltrazuril administered by intramuscular route at a higher dose (i.e. 40 mg/kg bw). The minimum effective dose of 15 mg/kg bw of toltrazuril was thereafter determined in two dose finding studies.

Dose determination / finding studies

The dose of 15 mg/kg bw of toltrazuril to be administered by intramuscular route was chosen for the subsequent dose determination studies, considering the increase in the bioavailability of toltrazuril by about 33% when administered by intramuscular route compared to oral route (see PK section above).

Two laboratory dose determination studies for toltrazuril were conducted in the same facilities by the same investigators and using similar methods. Piglets from a conventional pig farm were randomly allocated to a treatment group. Control animals received 200 mg iron (Gleptosil) as the only treatment. Each piglet received orally a single dose of approximately 1,500 sporulated oocysts of *C. suis* suspended in 2 ml of water at 3 days of life. The strain of *C. suis* used in this challenge was isolated from a farm without history of anticoccidial treatment in March 2005 and had been passaged regularly (every 2 to 6 months) through commercial piglets without anticoccidial treatment. The experimental infection model successfully induced a coccidiosis outbreak in the negative control group of each study. The primary efficacy criterion was the number of oocysts per gram of faeces (OPG) and more precisely the area under the OPG-time curve (AUC) over the major period of oocysts excretion. Oocyst counts were performed using a modified McMaster method. Secondary efficacy criteria consisted of faecal consistency (score of 1 to 4), occurrence of diarrhoea, average daily weight gain percentage of piglets showing diarrhoea and mean duration (days) of oocyst excretion. Product safety was assessed as a secondary objective of these studies. Quantitative variables were statistically compared between treatment groups using the Kruskal-Wallis non-parametric procedure. In case of significance at the $p<0.05$ threshold, the groups were compared pairwise using the Mann-Whitney U-test with Bonferroni adjustments for multiple comparisons. This methodology is suitable to carry out these studies.

The first dose determination study explored the efficacy of several doses of toltrazuril (0, 7.5, 15 or 30 mg/kg bw) when administered once by intramuscular route in neonatal piglets in the prevention of *C. suis* infection.

This study was a parallel group, mono-centric, double blinded, randomised, four-armed, GCP experimental design. Forty-eight piglets (12 per each dose of toltrazuril tested) were treated at 2 days after birth and orally challenged at 3 days after birth with a single dose of approx. 1,500 sporulated oocysts of *C. suis* (see above). The product tested contained toltrazuril and gleptoferron, in a formulation close to the final one. Considering that the dose of iron for the prevention of anaemia in pigs is 200 mg per piglet and depending on the administered volume of the test product, an additional injection of iron was performed to complete the dose until 200 mg of iron per piglet.

All negative control piglets excreted oocysts after infection (5 days of excretion on average), this

confirming the success of the experimental infection model.

No statistically significant difference was detected between the 7.5 mg/kg bw, 15 mg/kg bw and 30 mg/kg bw doses on oocyst excretion and faecal score. However, no piglet in the 15 mg/kg bw and 30 mg/kg bw groups excreted oocysts, compared to the ones in the 7.5 mg/kg bw group. From the results of this first study, it could be concluded that the minimum effective dose lied between 7.5 mg/kg bw and 15 mg/kg bw. Accordingly, the applicant decided to use a 10 mg/kg bw dose of toltrazuril in the following study.

In this second study, the efficacy of a 10 mg toltrazuril/kg bw dose when administered once by intramuscular route in neonatal piglets was evaluated for the prevention of *C. suis* infection. Another objective of the study was to investigate a more flexible time window than 2 days of age for product administration (*i.e*. from 1 to 3 days of age). This study was a parallel group, mono-centric, double blinded, randomised, three-armed, GCP experimental design. A negative control group (infected and non-treated) was compared to groups treated with 10 mg toltrazuril/kg bw once by intramuscular route at 1 and 3 days after birth (i.e. on study day (SD) 1 or 3). Study day 0 (SD 0) was the day of birth. In each group, 13-14 animals were included. The product tested contained toltrazuril and gleptoferron, in a formulation close to the final one.

An additional dose of gleptoferron was injected as necessary. Experimental infection was performed on study day 3 (i.e. 3 days after birth), similarly to the previous study. At both 1 and 3 days of age, a dose of 10 mg toltrazuril/kg bw administered once by intramuscular route in piglets showed no safety issues and proved to be effective in preventing oocyst excretion and clinical signs (diarrhoea), compared to negative control. No statistically significant differences were detected between the treated groups on oocyst excretion and faecal score. However, the mean OPG count, the mean duration of excretion and the percentage of piglets excreting oocysts were higher when piglets were treated on 1st day of life than on the 3rd day of life. These results allow proposing a larger timing for Forceris administration, increased from a 1-day period (2 days after birth, i.e. 48 to 72 hours of life) to a 3-day period (1 to 3 days after birth, i.e. 24 to 96 hours of life). However, the 10 mg toltrazuril/kg bw dose seemed to show a better efficacy when the product was administered on the 3rd day of life than on the 1st day of life. This difference in efficacy was attributed to the toltrazuril dose (10 mg/kg bw) and the applicant inferred that a dose of 15 mg of toltrazuril per kg bw by intramuscular route would provide the same efficacy level over the treatment window, i.e. 24 to 96 hours of life.

Consequently, further to the results of both studies, a dose of 15 mg/kg bw toltrazuril was considered by the applicant more appropriate to be proposed as the minimum recommended dose. Concerning these two studies, the applicant has provided information to justify that the formulation used was similar to the final one.

According to the information provided, a dose of 15 mg/kg bw is justified as the minimum recommended dose of toltrazuril for Forceris.

Dose confirmation studies

No dose confirmation studies for iron were carried out by the applicant. This is justified based on the well-established use of gleptoferron in prevention of iron deficiency anaemia in newborn piglets. Two dose confirmation studies conducted under laboratory conditions were provided by the applicant with regard to toltrazuril.

One GCP clinical study was performed to confirm the efficacy of a dose of 15 mg toltrazuril/kg bw when administered once by intramuscular route at 1, 2 or 3 days of life in neonatal piglets for the prevention of *C. suis* infection after experimental challenge. Although Forceris is intended to be administered at a

fixed dose/animal, the minimum recommended dose was used in this dose confirmation study as it was considered more appropriate to ensure an efficacy in the heaviest piglets (3 kg of body weight). This approach was agreed by the CVMP in the scientific advice provided.

The study was conducted using the final formulation and the materials and methods were identical to those used in the dose determination studies. Four groups of 11-12 animals were included (3 groups treated with Forceris 1, 2 and 3 days after birth, respectively, and 1 negative control group receiving gleptoferron only). Experimental infection was performed on study day 3 (i.e. 3 days after birth). The experimental infection model successfully induced a coccidiosis outbreak in the negative control group, which excreted 21.215 ± 20.905 OPG (mean over the study period ± SD). Piglet birth weight was similar between groups and ranged between 937 and 2081 g. Oocyst excretion, excretion duration, faecal score, the percentage of piglets excreting oocysts and the percentage of piglets presenting diarrhoea were significantly higher in the negative control group than in the treated groups. No statistically significant difference was detected between the treated groups on these parameters. No oocyst excretion was observed in piglets treated with 15 mg/kg bw toltrazuril on the 1st (n=11) and 2nd day (n=11) of life, whereas one of the 12 piglets treated on the 3rd day of life excreted oocysts for one day (no statistically significant difference). No occurrence of diarrhoea in Forceris groups was observed, while increase in faecal score was observed for negative control group, in which 80% of piglets presented at least one day of diarrhoea and a decrease in daily body weight gain due to *C. suis* infection especially after 2 weeks of age. There was a trend for the negative control group presenting a lower average daily weight gain in comparison with the treated groups; however, the results were inconclusive on this parameter.

Overall, a single injection of Forceris at 15 mg toltrazuril/kg bw between the 1st and 3rd day of life successfully prevented oocyst excretion and clinical signs (diarrhoea) associated to porcine coccidiosis (*C. suis*).

Another non-GCP experimental study was carried out to demonstrate the efficacy of Forceris (final formulation) in preventing clinical signs of coccidiosis. Each enrolled piglet was artificially infected on the 3rd day of life (SD3) with a single dose of approximately 1,000 sporulated oocysts of a laboratory strain of *C. suis*. In this study, 3 treatment groups were tested: Forceris group (n=13), a positive control group (Baycox) (n=12) and a negative control group (n=10). Both Forceris and Baycox were administered to piglets according to their recommended dose.

The treatment was administered on SD2 (Forceris group; also iron administration for both control groups) or on SD4 (Baycox group). Several parameters, including oocyst excretion and clinical signs of coccidiosis (diarrhoea) from SD7 to SD20, were assessed and compared between groups. Individual faecal samples taken from SD7 to SD20 were examined for the presence of oocysts qualitatively (by autofluorescence) and quantitatively (by McMaster counting to determine OPG).

No animal excreted oocysts in the Forceris group, whereas in the positive control group three piglets shed countable oocysts at low amounts for 1.3 days in mean. In the negative control group, all piglets shed detectable oocysts and 90% excreted McMaster countable oocysts. The mean duration of excretion in the negative control group was 3.1 days. Diarrhoea was noticed in all groups but was greatly reduced in the Forceris and Baycox groups with 15.4% and 33.3% diseased piglets compared to 100% in the negative control group. Oocyst excretion and diarrhoea were significantly reduced by Forceris or Baycox in comparison with the negative control group.

According to the information provided through the dose determination and dose confirmation studies, a single injection of Forceris at 15 mg toltrazuril/kg bodyweight between 24 and 96 hours of age

successfully prevents oocyst excretion and clinical signs (diarrhoea) associated to porcine coccidiosis (*C. suis*).

Target animal tolerance

The tolerance of Forceris in target animals was assessed in two TAS studies and also based on the data derived from other studies.

TAS studies

The tolerance studies carried out by the applicant do not meet exactly the requirements established in the VICH GL43: Target Animal Safety for Veterinary Pharmaceutical Products. However, the deviations are considered justified and they were supported by the CVMP in the scientific advice provided:

1. The use of an iron negative control group - according to VICH GL43, "The IVPP should be evaluated by comparison to a placebo (e.g. saline) or untreated control". In this case, to use neither a placebo group nor an iron untreated group as control is totally justified from a welfare point of view, as newborn piglets need to receive exogenous iron in their 3 first days of life, otherwise they develop iron deficiency and become anaemic within 7 to 10 days of birth.
2. The range of doses included in the tolerance study - the toxicity of injectable toltrazuril was explored in a margin of safety study. Toltrazuril was administered at doses of 0, 60, 180, 300 and 420 mg/kg bw, once by intramuscular route in 2-days old piglets. Nine days after the administration, at the 2 highest dosages, 5/8 and 6/8 piglets died, respectively, vs. 0, 1, or 2/8 at 0, 60 and 180 mg/kg bw, respectively. Thus, the results led to a maximal well tolerated dose of toltrazuril in 2 day-old piglets of 180 mg/kg bw. Consequently, it is scientifically sound to limit the doses tested in TAS studies at 1X, 2X and 3X the maximum recommended dose of 50 mg/kg bw instead of 1X, 3X and 5X (as recommended by VICH GL43), considering the toxicological properties of toltrazuril.
3. The use of repeated administrations – in TAS studies, the applicant tested the tolerance of the product only after a single administration instead of repeated administration (as recommended in the VICH GL43). This is considered justified as Forceris is intended to be used as a single administration, only once during a piglet's life and at a fixed identical volume for all piglets. The SPC clearly indicates that the tolerance of the product after repeated administrations has not been assessed and the administration of the product must not be repeated.

Thus, the experimental design of the TAS studies described below is justified from the scientific point of view.

Two TAS studies have been provided by the applicant in support of this application.

An original GLP-compliant TAS study was conducted to evaluate the general and local tolerance of Forceris in newborn piglets after single intramuscular administration of 1X, 2X or 3X the highest recommended dose (HRD) of 50 mg/kg bw of toltrazuril and 222 mg/kg bw of iron, this corresponding to 1.7 ml/kg bw of Forceris. The study was conducted according to VICH GL43, with some deviations: Forceris was administered at 1X, 2X and 3X HRD and treatments were not repeated (as explained above). The animals chosen constitute a representative sample of the most sensitive population of the target animal species.

No pain at injection was observed. No local reactions and no macroscopic / microscopic changes at the injection site in 1X HRD group were noted at the end of the study. The administration of Forceris seemed to increase the prevalence of arthritis in all Forceris-treated groups (6/12, 5/12 and 7/12

animals in 1X, 2X and 3X HRD groups, respectively), although no clear dose-effect relationship was observed. Abscess formation was also noted in all Forceris-treated groups (2/12, 4/12 and 2/12 animals in 1X, 2X and 3X HRD groups, respectively), although again no clear dose-effect relationship was observed. Mortality (animals dead or found moribund) was observed mainly in the first 12 days after Forceris administration and affected all Forceris-treated groups (2/12, 3/12 and 5/12 animals in 1X, 2X and 3X HRD groups, respectively), a dose-effect relationship being observed. At necropsy and histological examination, characteristic lesions of bacterial impairment in organs or tissues or of septicaemia were observed in Forceris-treated animals, mainly in the 3X HRD group (2/12, 2/12 and 7/12 animals in 1X, 2X and 3X HRD groups, respectively). Although a litter effect could also be suspected, these bacterial infections may be related to the administration of iron as an overdose of iron could result in bacterial infections by stimulating bacterial growth and multiplication, by inhibition of the reticulo-endothelial system (overload of macrophages) and/or by direct damage of the immune cells (Svoboda *et al*., 2005). Lower erythrocyte counts were observed in the 3X HRD Forceris-treated groups from Day 14, also a lower haematocrit and haemoglobin concentration were observed in 3X HRD group compared to the control group, which may be related to bacterial infections. These findings are reflected in the SPC under section 4.10.

In summary, at 1X and 2X the HRD, an indirect effect of Forceris with an increase in animal sensitivity to bacterial infections in some piglets due to iron overload may not be excluded. Moreover, a dose-effect relationship on tolerance was observed after the administration of Forceris in newborn piglets. In this study, three times the highest recommended dose of Forceris was harmful for newborn piglets.

A second original GLP-compliant TAS study was conducted to evaluate the general and local tolerance of Forceris in newborn piglets after a single intramuscular administration of 1X, 2X or 3X the recommended dose (RD) of 45 mg/piglet of toltrazuril and 200 mg/piglet of iron, this corresponding to 1.5 ml/piglet of Forceris. The study was conducted according to VICH GL43, with some deviations: Forceris was administered at 1X, 2X and 3X the RD and the treatments were not repeated (as explained above).

No pain at injection was observed. No local reactions at injection sites in control and 1X RD group were noted. Five piglets died before the end of the study, mainly during the 14 first days after treatment. Except 1 piglet found dead on Day 2 without pre-existing clinical signs, dead or moribund piglets presented signs of arthritis before death. The main histopathological findings were suggestive for bacterial infection as cause of death. Although mortality/moribund status were recorded in the test item groups only, a link to the test item is unlikely as no relationship between the dose and mortality rate was found (2/10, 2/10 and 1/10 animals in 1X, 2X and 3X groups, respectively). It was therefore not possible to conclude whether these deaths were due to the product or to poor health conditions before treatment. An increase in the prevalence of arthritis was observed after the administration of Forceris at 3 times the recommended dose (5/10 animals). Three out of the 11 piglets with arthritis died prematurely (1 in each Forceris-treated groups) due to bacterial infection. For the other 8 piglets, the signs of arthritis were reversible, lasting mostly between 1 to 7 days. A litter effect could be suspected as 10 out of 11 piglets with arthritis came from 2 out of 5 litters. Minimal to slight periportal haemosiderosis secondary to iron overexposure was present in the liver following the administration of Forceris at 2X and 3X the recommended dose. No additional adverse effects were detected with the histopathology examination of organs.

In this study, a single intramuscular administration of Forceris at 1X or 2X the recommended dose was well tolerated in newborn piglets. An increase in arthritis prevalence cannot be excluded after intramuscular administration of Forceris at 3X the recommended dose in newborn piglets.

In summary, it is acknowledged that mortality rates and incidence of arthritis observed in the TAS studies for newborn piglets may be in the same order of magnitude as those occurring under practical conditions in commercial pig farms. However, it is unclear whether adverse effects observed in the TAS studies are attributed to common practical/environmental conditions or are product-related. It is noted that in the margin of safety study and in the first TAS study a dose-dependent effect of toltrazuril was observed for mortality. In the second TAS study a higher rate of arthritis was observed after the administration of Forceris in the 3X group compared to 1X and 2X groups. Moreover, in the first TAS study lower erythrocytes counts, haematocrit and haemoglobin concentrations were observed in the 3X HRD Forceris group compared to control. These findings are reflected in section 4.10 of the SPC.

Other studies

First dose determination study: 39 adverse events occurred in 29 piglets, including 2 piglets with arthritis (treated with 15 and 30 mg/kg bw of toltrazuril, respectively) and 1 piglet with lameness (treated with 15 mg/kg bw of toltrazuril). No arthritis/lameness was observed in the groups having received either 0 or 7.5 mg/kg bw of toltrazuril. The percentage of arthritis/lameness in the groups treated at the 2 highest doses was 13% (3/23). Four cases of death were also reported in the treated groups at 7.5, 15 or 30 mg/kg bw (i.e. 11.4% of mortality) and none in the control group.

Second dose determination study: 27 adverse events occurred in 22 piglets. No case of arthritis or lameness was reported. One case of death was reported in the IVP group but this was caused by crushing by the sow. One death also occurred in the control group.

First dose confirmation study: 11 adverse events occurred after treatment in 11 piglets including 2 piglets with lameness (treated with Forceris on SD1 and SD2, respectively), i.e. 6% of the treated piglets (2/34). No lameness was reported in the control group. Three piglets in the IVP group were found dead after treatment, probably crushed by the sow, and 2 piglets were found dead in the control group.

The analysis of a comprehensive dataset of 504 animals treated within the course of the pre-clinical and experimental clinical studies showed no significant dose-related increase in mortality rate until 600 mg of iron or 150 mg of toltrazuril per animal. The analysis also showed no significant increase in arthritis incidence when the toltrazuril dose was increased. In contrast, the incidence of arthritis increased with increasing doses of iron. More precisely, the incidence increased slightly between the 1X and 2X doses, from 8.5% to just above 10%, and then increased more rapidly between the 2X and 3X doses (24%).

Pivotal field trial: less piglets showed arthritis or lameness following treatment with Forceris (13/761=1.7%) than with control product Gleptosil (22/747=2.9%) but the difference was not statistically significant. Furthermore, fewer piglets were found dead following treatment with Forceris (58/761=7.6%) than with Gleptosil (79/747=10.6%).

Overall, the mortality rate in the field trial was 9.1% (137 piglets) and the major causes of deaths were crushing by the sow (26 vs. 39 piglets in the Forceris and Gleptosil groups, respectively) and starvation (16 vs. 21 piglets). The other causes of death were diarrhoea (3 piglets), weakness (7 piglets) and other (3 piglets). Twenty-two deaths were not explained (9 and 13 piglets in Forceris and control groups, respectively).These deaths are common in pig commercial units.

A higher incidence of mortality was observed in the field trial when Forceris was administered on the 1st (9.8%) and 2nd (8.3%) days of life compared to the 3rd (4.5%) day. However, the actual increase in mortality is much smaller than 5.3% (9.8-4.5%) after removing perinatal mortality, which acts as a confounding factor in the field trial.

Unlike mortality, the incidence of arthritis was not related to piglets' age (0.4%, 0% and 2.1% at 1, 2 or 3 days of age, respectively). In total, 6 piglets (0.8%) in the Forceris group presented arthritis compared to 12 (1.6%) in the control group.

Five piglets showed pain, swelling, hardening and/or erythema at the injection site, 3 in Forceris group and 2 in CP group, proving a very good local tolerance of both products.

Overall conclusion on TAS

An increase in the prevalence of arthritis was observed after single administration of the product at both the recommended dose and three times the highest recommended dose. Following a single injection of up to 3 times the highest recommended dose, an increased susceptibility for (systemic) bacterial disease was observed. A dose-dependent increase in mortality was observed after single administration of up to three times the maximum recommended dose. A reduced erythrocyte count, haematocrit and haemoglobin concentration without clinical signs was observed after day 14 following single administration at three times the highest recommended dose. These findings are reflected in the product information for Forceris.

Based on the data presented, it is considered that neither the bodyweight nor the age is a risk factor for the safety of Forceris. Toltrazuril has a minor impact on animal tolerance except at higher dosages (>180 mg/kg bw). Thus, the safety profile of Forceris is similar to that of mono preparations containing iron.

However, the CVMP considers that a sufficient margin of safety of Forceris cannot be derived from the target animal safety studies and therefore this low margin of safety has been mentioned in the SPC.

Clinical field trials

One clinical study was performed under field conditions to confirm the efficacy of Forceris at a fixed dose of 45 mg toltrazuril and 200 mg iron (as gleptoferron) per animal (1.5 ml of product/piglet) when administered once by intramuscular route at 1, 2 or 3 days after birth (approx. 24-96 hours of age) in piglets for reduction in oocyst excretion and prevention of clinical signs of coccidiosis (*C. suis*) and prevention of iron deficiency anaemia, compared to a control group (CP). This study was a parallel group, multicentre, double blinded, randomised, two-armed, GCP design. The animal phase was performed from August to December 2015 and piglets in farms located in France (2 farms), Spain (1 farm) and Germany (2 farms) were enrolled in the study. In each country, farms were selected based on recent history of *C. suis* incidence (oocyst excretion or clinical signs such as creamy yellow diarrhoea from 7-21 days of age, observed with or without preventive treatment within 2 months preceding inclusion). The inclusion of five investigating sites in the EU, which allowed assessment of the anticoccidial activity of Forceris in pig nurseries with different baseline *C. suis* infection levels and in different productive conditions, is considered scientifically justified and in compliance with the guidelines on field trials.

In this study, the placebo (control) group was represented by animals not receiving toltrazuril or any other anticoccidial substance but receiving iron (as gleptoferron) at the fixed dose of 200 mg gleptoferron/piglet (1.0 ml/piglet of Gleptosil), by intramuscular route (control product). Since iron deficiency anaemia and the need of iron in the first days of life are well documented in the literature, a placebo group (without iron treatment) was not considered necessary. Moreover, including a negative control group receiving no oral or parenteral iron supplementation would have been unethical as this would have resulted in anaemia and the death of too many animals. This point was agreed by the CVMP in the scientific advice provided to the company.

A total of 122 sows were selected and randomized. From the 1688 piglets born alive, 1545 piglets with normal health status (no diarrhoea, no impaired extremities or malformations) were included in the study at day 1 of age (SD1) and finally 1508 were treated and followed. The inclusion criteria for an animal to enter the study were considered satisfactory. However, the eligibility criteria for farm selection were imprecise and this may explain the absence of coccidiosis outbreak in 1 out of the 5 farms. However, adequate statistical power was still achieved after removing this farm (n=1081 pigs in the exposed full analysis set population).

761 piglets were treated once by intramuscular injection in the neck with Forceris on the first (SD1), second (SD2) or third (SD3) day following birth, and 747 piglets were administered an approved product containing iron as gleptoferron (Gleptosil) on SD1, after randomisation. The Forceris formulation used in this study was the final formulation. The piglets were weighed at birth (on SD0) and then were treated from SD1 (i.e. from the day following the birth day). The recommended age range for Forceris is thus 1 to 3 days or 24 to 96 hours after birth.

As Forceris is a combination of 2 distinct active ingredients (toltrazuril and iron) with 2 different effects, efficacy for each ingredient was assessed independently, i.e. prevention of anaemia for iron and prevention of coccidiosis for toltrazuril (*multiple primary variables*).

Efficacy of Forceris against iron deficiency anaemia (IDA) was assessed by testing non-inferiority of Forceris versus control product (CP) with respect to the proportion of animals presenting anaemia defined as haemoglobinaemia <80 g/l at study day 10. This is considered acceptable, as piglets' hepatic iron reserves and the sow's milk are sufficient for only 3–7 days.

Efficacy of Forceris against coccidiosis was assessed by testing its superiority versus CP in decreasing the percentage of piglets showing at least one positive oocyst count from a minimum of three faecal samples taken between SD4 and SD21 and in decreasing the percentage of piglets presenting signs of diarrhoea, i.e. a faecal score of 3 or 4 or other clinical signs between SD4 and SD21. Performing a minimum of three oocysts counts is considered acceptable for this study, which was conducted in field conditions. The actual mean number of faecal samples was 5.0 in both groups considering the FAS population. To compensate for the lack of sensitivity of the *in vitro* diagnosis method, Forceris was only considered effective against coccidiosis if it reduced the occurrence of diarrhoea in addition to oocysts excretion. This approach is justified taking into account the epidemiology of this disease.

The secondary efficacy criteria were: oocyst excretion from SD4 to SD21 (assessed by the area under the oocyst excretion curve (AUC)), mean daily body weight gain until SD14 and SD21, haemoglobinaemia levels at SD10 and SD21 and percentage of piglets with anaemia (i.e. with Hb <80 g/l) at SD21.

Four populations were defined as follows: Safety Set population (including all animals having received the study treatment); Full Analysis Set (FAS) population (excluding cases with failure to satisfy major entry criteria, failure to take the trial medication, or lack of any data post treatment); Exposed Full Analysis Set (Exposed FAS) population (corresponding to all litters exposed to coccidiosis in the FAS population); Per Protocol Set (PPS) population (including all randomised animals that received the required level of study medication and complied with the protocol).

The coccidiosis assessment was first performed on FAS population as main population and then confirmed using the Exposed FAS population. The anaemia assessment was performed on the FAS and PPS populations. Product safety was assessed using the Safety Set population. Primary and secondary efficacy criteria were analysed using adequate statistical procedures.

Results for main efficacy criteria

Percentage of piglets excreting oocysts of C. suis between SD4 and SD21: Treatment with Forceris significantly reduced the percentage of piglets excreting oocysts in both FAS (17.4% vs 51.3% for Forceris and CP respectively, p<0.001) and exposed FAS population (24.1% vs 66.5%, p<0.001). A significant farm effect was detected (p<0.0001). In fact, the percentage of piglets excreting oocysts ranged between 0.8% (for farm EC1 which was considered as not being exposed to *C. suis*) and 97.0% in the control group (for farm GC1). The results of different injection timings did not show timing effects on Forceris efficacy in the FAS population, with 18.2%, 15.8% and 18.1% of piglets presenting at least one oocyst excretion from SD4 to SD21 after injection at SD1, SD2 and SD3, respectively. In all the exposed farms, the lower bound of the 95% confidence limit for the Odds Ratio to present no oocyst excretion in the Forceris group compared to the CP group was clearly superior to 1, demonstrating the efficacy of Forceris for the prevention of oocyst excretion.

Percentage of piglets presenting signs of diarrhoea between SD4 and SD21: Treatment with Forceris significantly reduced the percentage of piglets presenting diarrhoea in both FAS (24.1% vs 40.5% for Forceris and CP respectively, p<0.001) and exposed FAS population (22.7% vs 44.4%, p<0.001). A significant farm effect was detected (p<0.0001). The results of different injection timings did not show timing effects on Forceris efficacy in the FAS population, with 24.3%, 21.7% and 26.4% of piglets presenting at least one diarrhoea day from SD4 to SD21 after injection at SD1, SD2 and SD3, respectively. In all exposed farms, the lower bound of the 95% confidence limit for the Odds Ratio to present no diarrhoea in the Forceris group compared to the CP group was clearly superior to 1, demonstrating the efficacy of Forceris for the prevention of clinical signs associated with coccidiosis. As only diarrhoea was assessed and significantly reduced, the word "diarrhoea" has been added in the indication. Other clinical signs of coccidiosis, such as dehydration, appetite decrease, tenesmus, were not assessed and therefore not included in the SPC.

Percentage of piglets presenting anaemia (Hb<80 g/l) at SD10: The percentage of piglets with anaemia at SD10 was 6.0% in Forceris group and 4.5% in CP group in FAS population (5.8% and 4.5% in PPS population, respectively). The results of different injection times did not show timing effects on Forceris efficacy, with 5%, 9.4% and 3.4% of piglets presenting anaemia after injection at SD1, SD2 and SD3, respectively. Treatment with Forceris was non-inferior to Gleptosil in the prevention of anaemia since the upper bound of the 95% confidence limits for the difference between groups was lower than 5% (4.0% and 3.80% when using the FAS and PPS populations, respectively).

Secondary efficacy criteria

The log transformed AUC of the *number of oocysts per gram of faeces* (OPG) versus time was significantly lower in the Forceris group (7.4±19.7 OPG×day) than in the control group (26.9±35.8 OPG×day, p<0.0001) when using the FAS population. Corresponding values for the FAS exposed population were 10.4±22.6 and 35.2±37.2 OPG×day, respectively (p<0.0001). The number of days during which piglets excreted oocysts was 3 times lower in the Forceris group (mean of 1.1 days) than in the CP group (mean of 3.2 days).

No statistical difference was detected between groups on *average daily weight gain* when using the FAS population. However, average gain for the exposed FAS population between SD0 and SD21 was 213.7±77.3 g in Forceris group and 200.1±72.9 g in control group (p=0.0043).

The *haemoglobinaemia* was also analysed and compared between groups at SD10 and SD21. Hb mean values were 96.9 g/l and 106.9 g/l for Forceris-treated piglets at SD10 and SD21, respectively, whereas corresponding values for CP-treated piglets were 99.3 g/l and 110.3 g/l, respectively. The observed difference was not clinically relevant because no significant difference was observed between

groups from the clinical point of view (e.g. animals showing signs of anaemia).

The *percentage of piglets with anaemia* (Hb <80 g/l) at SD21 was 1.6% in the Forceris group and 2.2% in the CP group in the FAS population; no statistical difference was detected between the groups with regard to this parameter.

Safety

Overall, a good tolerance of Forceris was confirmed in this study (see section *Target animal tolerance* above).

In conclusion, a single intramuscular dose of 1.5 ml of Forceris/animal administered at 24 to 96 hours of age proved to be effective in preventing iron deficiency anaemia in piglets compared to a control product also containing iron as gleptoferron (Gleptosil) and using a non-inferiority margin of 5%. Also, Forceris reduced oocyst excretion and prevented clinical signs (diarrhoea) associated with coccidiosis caused by *C. suis* in comparison with a negative control. The good tolerance of Forceris was also confirmed in this multicentre study.

Overall conclusion on efficacy

Justification of the fixed combination

A satisfactory justification for the combination product has been provided. Thus, the use of this fixed combination versus combined use of single substances will result in a clinical benefit for the animals treated, a practical benefit for the breeder and an improvement of animal welfare.

Pharmacodynamics

Toltrazuril and iron are well-known active ingredients; toltrazuril is a broad-spectrum anticoccidial substance while iron is an essential element for the function of all body cells, playing a critical role in cell-cycle regulation, electron transport in the respiratory chain, DNA synthesis and other metabolic reactions. The pharmacodynamics of both substances has been thoroughly reviewed by the applicant based on the literature and the information provided is considered acceptable.

Development of resistance

The applicant has presented a bibliographical search where no resistance to toltrazuril in pigs was reported for *C. suis* in the field; however, a recent case of resistance was observed in a Dutch pig farm. Therefore, this new combination product must be used only when the indication is well justified. Information on frequent and repeated use of antiprotozoals and preventive measures against anticoccidial resistance development (e.g. the need for hygienic measures) is included in the SPC.

Pharmacokinetics

The pharmacokinetic characteristics of toltrazuril and iron (as gleptoferron) in combination are generally well documented and have been satisfactorily evaluated in piglets.

The kinetic profile of toltrazuril and its major metabolite toltrazuril-sulfone was comparable after intramuscular or oral administration and this suggested a comparable clinical efficacy. The bioavailability of toltrazuril was increased by about 33% when it was administered by intramuscular route compared to oral route. Additionally, the administration of toltrazuril by intramuscular route delayed its absorption by approx. 2 days compared to the administration by oral route and this suggests that treatment period can be reduced from 3-5 to 1-3 days of life if administered by intramuscular route.

A pharmacokinetic interaction was observed following a single intramuscular administration of Forceris compared to a single intramuscular administration of iron alone in neonatal piglets. When iron was administered in the form of Forceris, both the rate and extent of iron absorption appeared decreased. This was due to a formulation effect probably leading to a faster iron distribution to the extravascular compartment. However, no pharmacodynamic effects were elicited for haemoglobin, haematocrit and red blood cells levels and therefore no negative impact on iron clinical efficacy in the target population is expected.

Dose justification

No dose justification/determination/confirmation studies for iron were carried out by the applicant. This is justified based on the well-established use of gleptoferron for the prevention of iron deficiency anaemia in neonatal piglets, as Forceris is intended to be administered in the same target population by the same route and at the same dosage as other authorised iron-containing products (200 mg iron/piglet).

The applicant confirmed the efficacy of toltrazuril administered at a dose of 20 mg/kg bw by intramuscular route in the prevention of clinical signs of coccidiosis in neonatal piglets in an experimental infection study. The minimum effective dose of 15 mg/kg bw of toltrazuril was thereafter determined in dose finding studies and confirmed in dose confirmation studies.

Dose determination/finding

Two dose determination studies for toltrazuril were provided by the applicant. According to the results obtained, it can be accepted that a dose of 15 mg/kg bw is justified as the minimum recommended dose of toltrazuril for Forceris.

Dose confirmation

According to the information provided, a dose of 15 mg/kg bw is justified as the minimum recommended dose of toltrazuril for Forceris. This dose was confirmed in two dose confirmation studies conducted under laboratory conditions, where a single administration of Forceris at 15 mg toltrazuril/kg bw between 24 and 96 hours of age successfully prevented oocyst excretion and clinical signs (diarrhoea) associated to porcine coccidiosis (*C. suis*).

Target animal tolerance

An increase in the prevalence of arthritis was observed after single administration of the product at both the recommended dose and three times the highest recommended dose. Following a single injection of up to 3 times the highest recommended dose, an increased susceptibility for (systemic) bacterial disease was observed. A dose-dependent increase in mortality was observed after single administration of up to three times the maximum recommended dose. A reduced erythrocyte count, haematocrit and haemoglobin concentration without clinical signs was observed after day 14 following single administration at three times the highest recommended dose. These findings are reflected in the product information for Forceris.

Neither the bodyweight nor the age of piglets is a risk factor for the safety of Forceris. Toltrazuril has a minor impact on animal tolerance except at higher dosages (>180 mg/kg bw). Thus, the safety profile of Forceris is similar to that of approved mono preparation containing toltrazuril or iron. A sufficient margin of safety of Forceris cannot be derived from the target animal safety studies and this low margin of safety is mentioned in the SPC.

Clinical field trials

A single intramuscular dose of 1.5 ml/animal of Forceris final formulation (corresponding to the established fixed doses of 200 mg iron and 45 mg toltrazuril per piglet) administered at 24 to 96 hours of age proved to be effective in preventing iron deficiency anaemia in piglets, reducing oocyst excretion and preventing clinical signs (diarrhoea) associated with coccidiosis caused by *Cystoisospora suis* in a GCP multicentre field study performed in 3 EU countries, in comparison with a negative control. The good tolerance of Forceris when administered as recommended under field conditions was also confirmed in this multicentre study.

Part 5 – Benefit-risk assessment

Introduction

Forceris is a suspension for injection containing a fixed combination of two well-known active substances: toltrazuril (30 mg/ml) and iron (III) as gleptoferron (133.4 mg/ml). The product is available in multi-dose translucent plastic vials of three different pack sizes, containing 100 ml, 250 ml or 500 ml.

Toltrazuril is a triazinon derivative with antiprotozoal activity, and iron (as gleptoferron) is an essential micronutrient with anti-anaemic activity.

The product is intended for use in piglets (pigs) for the concomitant prevention of iron deficiency anaemia and prevention of clinical signs of coccidiosis (diarrhoea) as well as reduction in oocyst excretion, in farms with a confirmed history of coccidiosis caused by *Cystoisospora suis*. The effective dose of 45 mg of toltrazuril and 200 mg of iron per piglet (i.e. 1.5 ml of Forceris/piglet) administered once, in a single intramuscular injection, has been confirmed.

The application has been submitted in accordance with Article 13b of Directive 2001/82/EC (fixed combination).

Benefit assessment

Direct therapeutic benefit

Toltrazuril and iron (as gleptoferron) have been authorised and marketed for use in piglets, separately, for years in the EU. The two active substances can be administered separately to the same animal, but not on the same day, using the oral and intramuscular routes, respectively. Almost 100% of fast-growing piglets need parenteral iron supplementation in the first days of life. Coccidiosis is a widespread disease, and in many farms the piglets are at risk of developing coccidiosis, and consequently these animals need to be treated with a coccidiocide in their first days of life. Forceris is a fixed combination of the two active substances which provides enhanced ease of use, and a single intramuscular injection, which offers a better management of newborn piglets. Efficacy of this new veterinary medicinal product was demonstrated to be non-inferior to that of the control products, Gleptosil in preventing iron deficiency anaemia, and Baycox for the prevention of clinical signs of coccidiosis (diarrhoea).

Additional benefits

Neonatal piglets are manipulated very often during the neonatal period (navel cord care, teeth clipping, tail docking, ear notching for identification, castration of males and cross-fostering).

Therefore, Forceris presents practical benefits for the breeder, i.e. an injection of iron plus a separate oral administration of toltrazuril (on separate days) can be replaced by a single injection of the fixed combination product.

Forceris will facilitate animal handling and improve animal welfare by reducing the number of therapeutic interventions.

Risk assessment

Quality:

Two different sources of toltrazuril have been used, each has a different ASMF. Information on the manufacture of the active substance has been provided in the restricted parts of the two ASMFs. Information on the manufacture and controls of the active substance from each of the two different sources have been presented and are satisfactory.

The product's manufacturing process, finished product specification and stability support the authorisation of this medicinal product, and current regulations and guidelines have been taken into account.

Safety:

Measures to manage the risks identified below are included in the risk management section, as described.

Risks for the target animal:

The risks for the target animals are clearly identified in the sections 4.5, 4.6 and 4.10 of the SPC. Administration of Forceris in accordance with SPC recommendations is generally well tolerated. However, the recommended dose should not be exceeded given the relatively low margin of safety for this veterinary medicinal product.

Risk for the user:

Accidental self-injection is shown to be associated with systemic and developmental toxicity effects with regards to the exposure to toltrazuril that requires appropriate warning sentences to mitigate the risks. Other harmful effects identified are related to possible anaphylactoid reactions, local irritancy or granuloma formations that are also addressed with appropriate risk management measures. Therefore, Forceris is not expected to pose a risk for the user when used according to the SPC.

Risk for the environment:

Forceris is not expected to pose a risk for the environment when used according to the SPC. Standard advice on waste disposal is included in the SPC.

Risk for the consumer:

The residue studies available allow the establishment of a safe withdrawal period of 70 days in pig meat and offal. Therefore, Forceris is not expected to pose a risk for the consumer when used in accordance with this.

Risk management or mitigation measures

Appropriate information has been included in the SPC and other product information to inform on the potential risks of this product relevant to the target animal, user and consumer and to provide advice

on how to prevent or reduce these risks.

The withdrawal period is set at 70 days for meat and offal.

User safety:

Risk management or mitigation measures are proposed according to the user risk assessment.

Evaluation of the benefit-risk balance

Based on the data presented to date, the overall benefit-risk balance is considered positive.

The product indication as initially proposed by the applicant was: "In piglets: For the reduction in oocyst excretion and the prevention of clinical signs of coccidiosis in farms with a confirmed history of coccidiosis caused by *Cystoisospora suis*. For the prevention of iron deficiency anaemia". Following evaluation of the data, the CVMP agreed to the following indication: "For the concomitant prevention of iron deficiency anaemia and prevention of clinical signs of coccidiosis (diarrhoea) as well as reduction in oocyst excretion, in piglets in farms with a confirmed history of coccidiosis caused by *Cystoisospora suis*".

Information on development, manufacture and control of the active substance and finished product has been presented and lead to the conclusion that the product should have a satisfactory and uniform performance in clinical use. It is well tolerated by the target animals and presents an acceptable risk for users, the environment and consumers, when used as recommended. Appropriate precautionary measures, including withdrawal period, have been included in the SPC and other product information.

Conclusion

Based on the original and complementary data presented on quality, safety and efficacy, the Committee for Medicinal Products for Veterinary Use (CVMP) concluded that the application for Forceris is approvable since these data satisfy the requirements for an authorisation set out in the legislation (Regulation (EC) No 726/2004 in conjunction with Directive 2001/82/EC).

The CVMP considers that the benefit-risk balance is positive and, therefore, recommends the granting of the marketing authorisation for the above mentioned medicinal product.