7 September 2023
EMA/426807/2023
Veterinary Medicines Division

Committee for Veterinary Medicinal Products (CVMP)

CVMP assessment report for Oxmax (EMEA/V/C/005132/0000)

INN: hemoglobin betafumaril (bovine)

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

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Introduction

The applicant New Alpha Innovation Biopharmaceutical Ireland Limited (Previously: New A Innovation Biopharmaceutical (Ireland) Limited) submitted on 7 March 2019 an application for a marketing authorisation to the European Medicines Agency (The Agency) for Oxmax, 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 14 September 2018 as Oxmax contains a new active substance (hemoglobin betafumaril (bovine)), 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.

At the time of submission, the applicant applied for the following indication: For the management of canine haemorrhagic shock by increasing tissue oxygenation and achieving a comparable 24 hour survivability with blood.

The active substance of Oxmax is hemoglobin betafumaril (bovine), a blood substitute acting as an oxygen-carrier. The target species are dogs. The product is a solution for administration by intravenous infusion and is presented in packs of 2 infusion bags, each containing 100 ml.

The applicant is registered as an SME pursuant to the definition set out in Commission Recommendation 2003/361/EC.

The rapporteur appointed is Rory Breathnach and the co-rapporteur is Anna Wachnik-Święcicka.

The dossier has been submitted in line with the requirements for submissions under Article 12(3) of Directive 2001/82/EC (full application).

On 7 September 2023, the CVMP adopted an opinion and CVMP assessment report.

On 19 October 2023, the European Commission adopted a Commission Decision granting the marketing authorisation for Oxmax.

Scientific advice

The applicant received four scientific advices (and several clarification/follow-up reports) from the CVMP pertaining to quality, safety and clinical development issues of the dossier.

In general, the applicant followed the scientific advice; more details are provided in the relevant sections of this report.

MUMS/limited market status

The applicant requested classification of this application as MUMS/limited market by the CVMP, and the Committee confirmed that, where appropriate, the data requirements in the relevant CVMP guideline(s) on minor use minor species (MUMS) data requirements would be applied when assessing the application. MUMS/limited market status was granted as the proposed indication was considered to represent a limited market in dogs.

Part 1 - Administrative particulars

Detailed description of the pharmacovigilance system

At time of submission of the marketing authorisation application, the applicant provided a detailed description of the pharmacovigilance system (Version 0.1, dated 4th March 2019).

Given that Regulation (EU) 2019/6 came into effect when the application was in the clock-stop phase, the applicant provided a summary of the pharmacovigilance system master file which fulfils the requirements of Article 23 of Commission Implementing Regulation (EU) 2021/1281, which was acceptable. Based on the information provided the applicant has in place a pharmacovigilance system master file (PSMF), has the services of a qualified person responsible for pharmacovigilance, and has the necessary means to fulfil the tasks and responsibilities required by Regulation (EU) 2019/6. The active substance contained in the veterinary medicinal product is considered a new active substance as it has not yet been authorised in a veterinary medicinal product in the European Union. In the pharmacovigilance database, all the results and outcomes of the signal management process, including a conclusion on the benefit-risk balance, should be reported annually.

Manufacturing authorisations and inspection status

Manufacture of the active substance (hemoglobin betafumaril (bovine)) takes place at manufacturing sites outside the EEA. Appropriate QP declarations for the active substance manufacturing sites was provided by the Qualified Person (QP) at the EU batch release sites that the active substance has been manufactured in line with GMP requirements.

Manufacture of the finished product takes place at manufacturing sites outside the EEA. Good Manufacturing Practice (GMP) certification, which confirms the date of the last inspection and shows that the sites are authorised for the manufacture of such veterinary dosage form, has only been provided for one site as the GMP certificate was not available for the second site. The second site was recently inspected, and the EU based inspectorate confirmed that the site complies with the GMP requirements. A post authorisation measure is in place to submit the certificate when available.

Batch release within the EU takes place at Klifovet GmbH, Munich (Germany), which holds a valid manufacturing authorisation.

Overall conclusions on administrative particulars

The summary of the pharmacovigilance system master file is considered to be in line with legal requirements.

The GMP status of both the active substance and finished product manufacturing site has been satisfactorily established and is in line with legal requirements.

A post authorisation measure is in place to provide the GMP certificate for the second site, when available.

Part 2 - Quality

Composition

Oxmax is a dark purple solution for infusion for dogs consisting of the hemoglobin betafumaril (bovine) (INN) as the active substance at a concentration of 65 mg/ml. Excipients of the Oxmax formulation are acetylcysteine, also referred to as N-acetyl-L-cysteine (NAC) and Ringer's acetate

solution (consisting of sodium chloride, potassium chloride, calcium chloride dihydrate, sodium acetate trihydrate, sodium hydroxide, and acetic acid, glacial) and water for injections (WFI) (vehicle) as described in section 6.1 of SPC.

The product is available in a multi-layered plastic intravenous (IV) infusion bag containing 100 ml of the infusion solution, overwrapped with aluminium foil pouch with a twist off port, as described in section 6.5 of the SPC.

Containers

The formulated and stabilised hemoglobin betafumaril (bovine) solution for infusion is filled aseptically into sterile IV bags (100 ml per bag). The IV bag consists of a multi-layered laminated (film) with a twist off port. The film material of the infusion bag meets the standard of European Pharmacopoeia (Ph. Eur.). As haemoglobin (Hb) is prone to oxidation the primary packaging (IV infusion bag) is protected by a secondary sealed aluminium overwrap pouch. The stability data of several batches showed the percentage of Oxy-Hb and Met-Hb in the finished product remained constant and within the required acceptance criteria for up to 2 years.

The immediate packaging inner layer (in contact with the product) meets the Ph. Eur. 3.1.7 requirements. Appropriate suppliers' certificates of analysis (CoAs) are provided and in-house testing is described and considered acceptable.

The sterilisation method of the infusion bag complies with Ph. Eur. 5.1.1.

The suitability of the twist off cap to prevent oxygen intrusion and contamination of the bag is described and supported with stability data of several batches where levels of Oxy-Hb and Met-Hb met the required specifications over time, indicating no effect of oxygen levels. A container closure integrity test (CCIT) was also conducted with the IV bag with twist off cap, it demonstrated that the container closure system of the IV bag (100ml) provided a sterile barrier against microbial contamination.

Development pharmaceutics

Oxmax consists of hemoglobin betafumaril (bovine) (INN) at a concentration of 65 mg/ml in a solution for infusion for dogs.

Ringer's acetate solution is used to confer isotonic and isosmotic properties to the formulation, and the electrolytes conferred are controlled throughout the process for optimal intravenous use in dogs. The final formulation also includes an antioxidant. All excipients used in the formulation are compliant with Ph. Eur. standards. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 6.1 of the SPC.

The formulation used for the pivotal dose confirmation study is the same as that intended for marketing.

Method of manufacture

The manufacture is a multi-step process consisting of filtration steps before aseptic filling of the IV bags. A satisfactory flowchart of the manufacturing process is provided. The applicant adequately describes the process.

The in-process controls are adequate for this manufacturing process.

Acceptable sterile filter validation studies were conducted using scaled down processing conditions on the filters used. The applicant has provided process validation results for several batches. The

data for these batches support process validation requirements.

In line with the guideline on process validation for finished products (EMA/CHMP/CVMP/QWP/BWP/70278/2012-Rev1,Corr.1) full scale process validation will be required from each site. Oxmax was given MUMS classification and therefore process validation post authorisation from manufacturing sites will be acceptable. The results of the final product process validation of three consecutive full-scale batches manufactured at each manufacturing sites, will be available for scrutiny during GMP inspections post authorisation in accordance with the guideline on quality data requirements for veterinary medicinal products intended for MUMS. The competent authority(ies) must be informed if problems are encountered on validation of the process at the full scale, together with the proposed actions' as stated in the guideline on quality data requirements for veterinary medicinal products intended for MUMS (EMA/CVMP/QWP/128710/2004-Rev.1).

Control of starting materials

Active substance

The active substance hemoglobin betafumaril (bovine) contained in the veterinary medicinal product is considered a new active substance as it is novel and has not yet been authorised in a veterinary medicinal product in the European Union.

Hemoglobin betafumaril (bovine) is a modified and stabilised bovine haemoglobin consisting of two alpha-beta chains (alpha 1-beta 1, and alpha 2-beta 2). The structural formula is provided. The molecular mass of active substance is determined.

The applicant has presented adequate in-process testing to determine whether the quality of the active substance is suitable for further processing of Oxmax.

Structure elucidation is performed on the finished product as the active substance is not isolated from the continuous process. Methods for structure elucidation were provided. The elucidation of the primary and secondary structure of hemoglobin betafumaril (bovine) has been provided. The secondary structure of purified haemoglobin solution (PHS), the starting material, was also determined. Results from the batches tested showed that the secondary structure of Oxmax was in line with the bovine Hb reference from the Protein CD Data Bank and using the CAPITO tool (a matching based prediction where the query spectrum is matched against protein reference curves in the database) the applicant also concluded that that cross-linking in Oxmax did not greatly alter the secondary structure of the haemoglobin. The results from ESI-MS of trypsin-digested peptides from Oxmax showed the primary structure to have up to 96% sequence homology with alpha and beta subunits of bovine haemoglobin. The modification of the active substance, hemoglobin betafumaril (bovine) is adequately described.

Physicochemical characterisation of Oxmax included molecular weight, isoform pattern and Pi, native gel electrophoresis, UV-visible spectrophotometry, viscosity and colloid osmotic pressure (COP). The results demonstrated a high degree of consistency of product characteristics and consistent results across the tested production batches.

The biological activity was determined.

The active substance preparation is performed in two distinct stages and is satisfactorily described: manufacture of the starting material (PHS) from bovine blood and then manufacture of the active substance from the PHS. Validation of the manufacturing process for the starting material OC-PHS was provided for four consecutive batches manufactured at the proposed manufacturing scale. The validation report is provided in Part 2.C1-13. The process validation for OC-PHS is satisfactorily

described and from the results provided the validation batches appear consistent and below specification. The endotoxin levels are consistent across the batches at ≤0.03 EU/ml. The endotoxin limit is proposed at 0.1 EU/ml for the starting material and is considered acceptable.

Storage conditions and transport of the bulk OC-PHS to the manufacturing sites are adequately described.

The starting material is manufactured from bovine whole blood that is obtained from cattle born and bred in New Zealand from controlled herds and collected according to New Zealand national standards for slaughter and blood collection. Viral screening of the whole blood is performed. A risk assessment on viral safety for material of animal origin, in line with Ph. Eur. 5.1.7 requirements, and an updated herd management plan are provided.

The specification and routine text methods proposed are acceptable for the active substance, hemoglobin betafumaril (bovine). The contents of total Hb (t-Hb), oxy-Hb and metHb of active substance is identified.

The impurities were defined and the corresponding acceptance limits was set in the Oxmax finished product release specifications.

Two virus removal process steps are satisfactorily validated in line with the Note for guidance on virus validation studies (CPMP/BWP/268/95).

Excipients

Routine tests for all the excipients are to Ph. Eur. standards and acceptable CoAs are provided.

All excipients are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur. standards. There are no novel excipients used in the finished product formulation. 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 purified haemoglobin solution (PHS) which is used as the starting material for the preparation of the active substance (hemoglobin betafumaril (bovine)) is extracted from adult bovine whole blood. All bovines used as source animals are of New Zealand origin. New Zealand is classified as having a negligible BSE risk in accordance with Chapter 11.4 of the Terrestrial Code (WHOA). Sufficient controls are included in the PHS production process to allow traceability of the blood used for its production. Valid TSE certificates of suitability for PHS were provided. The TSE risk is considered negligible.

Control tests on the finished product

The finished product tests include appearance, total Hb, met-Hb, oxy-Hb, molecular weight distribution (DMW), pH, particulate contamination, sterility, endotoxins, and osmolality. The test to measure the oxygen-carrying capability is a preferred test method for release, and stability. The Co-oximetry test provides information about the 'potency' or 'functionality' of the product and also the level of the product-related impurity which is important from a safety perspective.

The appearance testing is adequately described.

The quantitation of the finished product as total Hb was determined and the corresponding validation protocol and reports for are provided. The results were within the specifications set. Additionally results from the release testing of finished product placed on stability study demonstrated consistent results for haemoglobin within the set specifications at release and up to 24 months of storage.

Satisfactory conditions for the test method to determine the purity are described. The limits of impurities have been established, based on the data provided for the batches used in the safety studies and consistency batches.

The pH testing was adequately described with calibrated equipment. The particulate content of the finished product is performed according to the Ph. Eur. 2.9.19. The osmolality of finished product tested is determined at both sites in line with Ph. Eur. 2.3.35.

HPLC is used to measure acetylcysteine in the finished product, the specification set is <0.22%. An acceptable test protocol and validation have been provided with appropriate standards.

The endotoxin level of finished product is determined and in line with Ph. Eur Eur. 2.6.14. A suitable protocol for the validation of the method is provided and an acceptable CoA, for the reference material used within the ranges proposed, is presented. The proposed endotoxin release limit for Oxmax is ≤0.25 EU/ml which was justified by the applicant as meeting the requirements of the Ph. Eur. 5.1.10 (Guidelines for using the test for Bacterial Endotoxins), the Ph. Eur. (0169) acceptance criteria for water for injections in bulk is <0.25 EU/ml and the maximum daily dose for Oxmax is 10 ml/kg and the calculated endotoxin limit (Ph. Eur. 5.1.10) is 0.5 EU/ml.

Sterility testing is performed and verified in line with Ph. Eur. 2.6.1.

Batch analysis data for Oxmax batches shows that batches manufactured met the required release specifications and therefore show batch to batch consistency within each site and comparability between sites. CoAs for each of the batches were provided in line with the Guideline on process validation for finished products (EMA/CHMP/CVMP/QWP/BWP/70278/2012-Rev1). The applicant provided a detailed process validation protocol and confirmed that the results of the validation of the manufacturing process study will be available for GMP inspectors and any out of specification results will be reported to the authorities, together with proposed actions in accordance with the guideline on quality data requirements for veterinary medicinal products intended for MUMS (EMA/CVMP/QWP/128710/2004-Rev.1) post-authorisation.

Stability

The applicant has provided stability data for several batches of PHS which is produced in New Zealand. A shelf life for PHS bulk stored at 2-8 °C was accepted and this was supported by acceptable data for batches tested.

A number of batches have undergone the container-closure integrity testing (CCIT) and are also tested for sterility after CCIT; reports for the CCIT testing are provided.

The integrity of the primary container (infusion bag) is confirmed by sterility testing of samples of the solution aseptically removed from the bag and testing them in accordance with the current Ph. Eur. 2.6.1.

Data from production scale batches have been presented to 36 months storage at 2-8° C. All results met the required specifications at each time point to 36 months. Additionally, the applicant provided data from an accelerated stability study for these batches of finished product up to 12 months stored at 25°C ±2°C / 60% ±5%RH. The results of all tests complied with the acceptance limits, although levels of non-stabilised tetramer (dimer) increased notably after 6 months storage at accelerated conditions. No changes in DMW distribution were detected when stored at refrigerated conditions.

Three consecutive batches of Oxmax manufactured at one of the manufacturing sites and packaged in the final packaging proposed for the commercial product have been placed on the ongoing stability

program. An interim report with the stability data up to 24 months of storage at 2°C – 8°C in the proposed packaging is provided. Results of all the tests were within the specifications up to 24 months.

Real time stability data for Oxmax batches manufactured at the other manufacturing site have been provided up to 30 months of storage at 2°C – 8°C. All the results were within the specifications, without any trends for deviation.

An aluminium foil overwrap is included as part of the packaging, and it is recommended that the product is protected from light and this is acceptable.

There is no in-use shelf life for this product although the product is administered over a 24 hour period. The SPC section 6.3 refers to '*Use immediately and do not store after opening'*, which is acceptable.

The proposed shelf-life of the packaged finished product for 2 years kept refrigerated (2°C – 8°C) is supported by the data provided and is acceptable in accordance with MUMS requirements.

A CVMP scientific advice (EMA/CVMP/SAWP/6360/2015) stated that the stability data from one of the manufacturing sites can be used to support the shelf life of batches from the additional site if acceptable comparative data between the sites is provided. The data provided from the manufacturing sites is considered sufficient to support the proposed shelf-life of 2 years.

Overall conclusions on quality

The composition of Oxmax in regard to the active substance and excipients has been adequately described. Sufficient details of the container and closure system have been provided. The immediate packaging inner layer meets Ph. Eur. 3.1.7 requirements, and the intravenous bag is appropriately sterilised which complies with Ph. Eur. 5.1.1.

Overall, the development of Oxmax has been adequately described. The final formulation is based on the literature and development data. Ringer's acetate solution is used to confer isotonic and isosmotic properties and acetylcysteine is used as an anti-oxidant and reducing agent. All excipients used in the formulation are compliant with Ph. Eur. standards.

Manufacture of the finished product takes place at manufacturing sites outside the EEA. Manufacturing Practice (GMP) certification, which confirms the date of the last inspection and shows that the sites are authorised for the manufacture of such veterinary dosage form, has been provided for the manufacturing sites.

In general, the manufacturing process for Oxmax has been adequately described. Process validation protocols are provided. The results of the final product process validation of three consecutive full-scale batches if relevant, will be available for scrutiny during GMP inspections post-authorisation in accordance with the guideline on quality data requirements for veterinary medicinal products intended for MUMS.

The virus validation of the process performed in line with Note for guidance on virus validation studies (CPMP/BWP/268/95) is considered acceptable.

The manufacturing process from starting material (PHS) to Oxmax finished product is a continuous process, the bulk active substance is not isolated from the process.

The applicant has presented adequate in-process testing to determine whether the quality of the active substance is suitable for further processing of Oxmax.

Characterisation of Oxmax is satisfactorily described. Acceptable details of the elucidation of the primary and secondary structure are provided. The modification of the hemoglobin betafumaril

(bovine) is adequately described. Physicochemical characterisation of Oxmax is satisfactorily described and test methods are validated.

The biological activity of Oxmax was determined and validation of the test has been provided. The potency of the finished product is comparable between finished product manufactured at different manufacturing sites.

The active substance (hemoglobin betafumaril (bovine)) preparation performed in two distinct stages is satisfactorily described. The process validation for the active substance is described for 2 pilot batches at one of the manufacturing sites and the data is acceptable for this MUMS application. The endotoxin levels are consistent across the active substance batches.

The starting material is stated as purified haemoglobin solution (PHS) manufactured from bovine whole blood that is obtained from cattle born and bred in New Zealand. Viral screening of the whole blood is performed. A satisfactory risk assessment on viral safety for material of animal origin in line with Ph. Eur. 5.1.7 is provided. CoA for a batch of PHS manufactured from the New Zealand site is provided.

All manufacturing sites have adopted the same specifications and routine test methods for active substance hemoglobin betafumaril (bovine). An acceptable validation of the potency test was performed on the finished product instead of the active substance.

Potential product and process impurities and tests to identify them were adequately described.

The PHS used as the starting material for the active substance is extracted from adult bovine whole blood. All bovines used as source animals are of New Zealand origin. New Zealand is classified as having a negligible BSE risk in accordance with Chapter 11.4 of the Terrestrial Code (OIE). TSE certificates of suitability for PHS was provided. The TSE risk is considered negligible.

The tests listed in the finished product specifications were considered appropriate. The analytical methods were described, and validated in accordance with VICH requirements.

Batch analyses data for several Oxmax batches shows that batches manufactured at all manufacturing sites met the required release specifications and therefore show batch to batch consistency within each site and comparability between the sites. The applicant initiated a finished product manufacture process validation study, the results are to be available to GMP inspectors for scrutiny post-authorisation during a site inspection at each site. Any problems encountered during the process validation are to be reported to the authorities in accordance with MUMS guidance.

Data of several Oxmax batches at production scale manufactured at each of the manufacturing sites (stored for either 24 or 30 months at 2°C – 8°C) support the proposed shelf-life of the packaged finished product for 2 years kept refrigerated (2°C – 8°C).

Part 3 – Safety

The active substance hemoglobin betafumaril (bovine) of Oxmax is a new active substance not authorised for a veterinary medicinal product in the EU before. Oxmax is a solution for infusion containing 65 mg/ml hemoglobin betafumaril (bovine) and is intended for administration to dogs.

The recommended dose is 10 ml/kg bodyweight administered intravenously at a rate of up to 10 ml/kg bw/h.

The application is submitted in accordance with Article 12(3) of Directive 2001/82 EC, as amended. Oxmax is classified as MUMS and the CVMP Guideline on safety and residue data requirements for veterinary medicinal products intended for minor use or minor species (EMEA/CVMP/SWP/66781/2005-Rev.1) was applied.

Safety documentation

Pharmacodynamics

See Part 4.

Pharmacokinetics

See Part 4.

Toxicological studies

Three single dose toxicity studies and one repeat-dose toxicity study were provided.

Single dose toxicity

In accordance with the guidance in effect at time of assessing the application (CVMP Guideline on safety and residue data requirements for veterinary medicinal products intended for minor use or minor species (EMEA/CVMP/SWP/66781/2005)), single dose toxicity studies were not required for applications holding a MUMS status. Nevertheless, the applicant provided three acute toxicity studies in the rat. While the studies were not good laboratory practice (GLP) compliant, it was considered that they were of a satisfactory quality. All three studies provided similar results. The test article appeared well tolerated at doses up to 6.9 g Hb/kg bw (100 ml product/kg bw) intravenously infused for 33.3 hours. No mortality occurred and no adverse effects were evident clinically. However, the following treatment-related findings (with dose effect relationship) were observed: a temporary increase in proteinuria, appearance of red blood cells in the urine and a temporary decrease in urine potassium and chloride levels (Day 2). It is possible that the presence of Hb in samples, arising from Oxmax treatment, caused assay interference resulting in a number, or all, of these effects.

It is concluded that Oxmax is of low acute toxicity potential. While not conducted to GLP and limited in terms of animal numbers (and only male rats included), all three studies show similar results. For the target species a single administration is the most relevant scenario.

Repeat dose toxicity

The applicant conducted a pilot repeat-dose study investigating the toxicity of Oxmax following repeated administration on three occasions at 48 hour intervals in the target species (dogs). The test article was administered by intravenous infusion at approximately 3 x the recommended treatment

dose (RTD) (low dose group) and at approximately 5 x RTD (high dose group). The number of dogs was 2 per group. Anaemia was induced in the study animals (by blood withdrawal and haemodilution) prior to each administration, resulting in the clinical condition of the study animals being more representative of those animals for which the product is intended, i.e. acutely anaemic dogs.

Based on the findings of this limited study, it would appear that repeated treatment with the test product was generally well tolerated in anaemic Beagles. The principal clinical effect observed was lethargy, which was attributed to the experimentally-induced clinical condition and not considered an effect of treatment. Similarly, emesis was considered secondary to the anaemia. Clinico-pathological findings of significance included transient increased levels of bilirubin and blood in the urine of treated animals along with increased protein levels in the urine of animals administered the higher dose. No no-observed effect level (NOEL) could be established. The findings of this study were similar to the findings in the acute toxicity studies in rats.

Immunotoxicity

In agreement with the scientific advice provided by the CVMP, specific studies to investigate immunotoxicity were not provided. However, to address concerns raised in respect of the immunogenic potential in the target species, the applicant made reference to published data. Neither of the studies referenced provided data on the immunogenic potential for the target species dogs: one study investigated immunotolerance to homologous and heterologous rat haemoglobin administered to rats, whereas the other study investigated immunotolerance in humans administered bovine haemoglobin. However, it was noted from the latter study that the same authors reported upon an analogous bovine haemoglobin-based preparation authorised in the United States of America for administration to dogs following repeated administration on nine occasions to eight dogs over a 50 week period. It had been reported that IgG levels to the bovine Hb were detected in 7 out of 8 dogs with antibody levels peaking after the third administration. No information on the frequency of occurrence of possible immunogenic reactions was reported.

While the potential for a repeat dose to induce an immunological reaction has not been fully investigated, section 4.3 of the proposed SPC includes a contraindication for use in animals previously exposed to the product or other bovine haemoglobin-based oxygen carriers, to avoid a potential sensitivity-type reaction upon repeat exposure. This is considered acceptable.

Tolerance in the target species of animal

See Part 4.

Reproductive toxicity

No data were provided. According to the CVMP Guideline on the safety and residue data requirements for veterinary medicinal products intended for minor uses or minor species (EMEA/CVMP/SWP/66781/2005), data in respect of reproductive toxicity (including teratogenicity) are not required provided that the product is not indicated for use in food-producing target species and that the product is not intended for administration to animals intended for breeding.

It is noted that the proposed SPC includes the following statement: "*The safety of the veterinary medicinal product has not been established during pregnancy or lactation. Use only according to the benefit-risk assessment by the responsible veterinarian"*. This is considered acceptable.

Genotoxicity

As part of the scientific advice provided by the CVMP in respect of this application, the applicant was advised that mutagenicity testing would need to be performed in accordance with the requirements of VICH GL23 on studies to evaluate the safety of residues of veterinary drugs in human food: Genotoxicity testing. In that guideline, the following battery of three tests is recommended for use as a screen of veterinary drugs for genotoxicity:

- A test for gene mutation in bacteria.
- An *in vitro* test for chromosomal effects in mammalian cells.
- An *in vivo* test for chromosomal effects using rodent haematopoietic cells.

The mutagenic potential was investigated in two *in vitro* mutagenicity tests, a bacterial reverse mutation test and a mouse lymphoma assay. In both test systems, there was no evidence that the test item has mutagenic potential.

A justification was provided for the omission of an *in vivo* test for chromosomal effects using rodent haematopoietic cells. The justification is supported by the fact that bovine haemoglobin does not contain any structural alerts suggesting a potential for genotoxicity and the results of available studies (bacterial reverse mutation test and the *in vitro* mammalian cell gene mutation test in mouse lymphoma cells) did not show evidence of any mutagenic potential. It is acknowledged that the recommendations for testing included in VICH GL23 relate specifically to the performance of genotoxicity tests for the purpose of the evaluating the safety of residues in human food. Given the fact that the intended target species for the product is dogs (a non-food producing species), and the results of both *in vitro* studies did not show evidence of any mutagenic potential, the omission of an *in vivo* test for chromosomal effects could be accepted in this instance.

Carcinogenicity

No carcinogenicity data were provided. The absence of studies investigating the carcinogenicity of the product can be accepted based on the negative findings of the bacterial reverse mutation test and the mouse lymphoma cell gene mutation test. In addition, it is accepted that the active substance (hemoglobin betafumaril (bovine)) has no structural similarity to known carcinogens.

Studies of other effects

Interference with colorimetric assays

The applicant highlighted the fact that the presence of Oxmax in serum and urine may interfere with colorimetric assays and produce unreliable results with the magnitude of interference dependent upon the dosage of Oxmax. In order to quantify such interference, the applicant conducted a GLP study investigating the effects of Oxmax administration on clinical chemistry, haematology and urine analyses.

The results of this study suggest that the presence of haemoglobin in samples of canine serum, plasma and urine may result in a statistically significant interference in the assayed levels of a number of biochemical, haematological and urological parameters. This was also shown in further data presented in which the presence of haemoglobin interfered with the accuracy of measurement of creatinine and urea at plasma concentrations that will be expected following treatment at the recommended dose. Depending on the parameter, the interference threshold may be as low as 0.50 mg/ml. It is noted that, in different pharmacokinetic studies, plasma Hb concentrations in the range 1.5–3.0 g/dl (15–30 mg/ml) have been recorded.

Based on the findings of this study, the applicant has added appropriate information/advice in section 4.5 of the SPC in regard to the possible interference with colourimetric methods in laboratory tests (chemistry, haematology, coagulation, urinalysis). In addition, clinicians are advised (section 4.9) to obtain clinical samples (blood, urine) before the administration of Oxmax. Given that numerous parameters may be affected and that the interference will vary depending on the time since infusion, type of analyser and reagents used, the proposed SPC text is considered appropriate.

Studies on local effects

No data were provided. The absence of such data (dermal/ocular irritancy, dermal sensitivity) has not been specifically addressed by the applicant. However, in view of the physiological pH of the product, and the nature of its components, it can be accepted that no local effects are likely. It is noted that most of the excipients are simple salts and are unlikely to have irritant effects for skin and eyes. The product also contains the excipient acetylcysteine (also known as N-acetyl-L-cysteine), an amino acid derivative, which is unlikely to pose undue risks to the target species.

Given the nature of the active substance and excipients, together with the presentation/packaging and the intended method of administration (limiting potential for exposure), the absence of local effect studies is accepted.

User safety

The applicant has presented a user safety risk assessment which 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 parenteral, oral and dermal exposure.

Given that the product will be restricted to professional users and is intended for administration by intravenous infusion, it is accepted that the potential for user exposure is limited. It is considered that the packaging, the intended use (intravenous infusion) and the restriction to professional users will limit user exposure to the product. The applicant provided exposure scenarios for parenteral (accidental self-injection), oral (hand-to-mouth contact) and dermal exposure and concluded that the risk to the user was acceptable in all cases. However, in the user safety assessment, the applicant states that the exact immunogenicity potential of Oxmax in man is unknown. It is acknowledged that, as the product is derived from bovine Hb, the risk of an immunogenic reaction in humans following repeat accidental injection, although low, does exist. However, CVMP concluded that it is likely unnecessary to include advice to seek medical advice immediately in every event of accidental self-administration, but it would be prudent to seek medical advice if adverse reactions develop following accidental exposure. Furthermore, it is highly unlikely that the person administering the product would have pre-existing knowledge of hypersensitivity to hemoglobin betafumaril (bovine), and the value of including a recommendation to administer with caution in this unlikely scenario is questionable. In essence, the main risk to the user arises due to the potential for hypersensitivity reactions following repeated accidental self-injection.

The applicant also considered the risk posed to the user arising from exposure to the excipient acetylcysteine. Data are available that indicate that anaphylactoid reactions are a risk following intravenous infusion of human products containing this component as the active substance. A threshold level below which such reactions would not pose a risk was not discussed. While it is anticipated that exposure to only very small amounts of this excipient might occur following accidental self-injection, and therefore that the risk of anaphylactoid reactions would be expected to be low, on the basis that the risk cannot be excluded, information is included in the PI to inform the user accordingly.

Based on the above risk assessment, it is accepted that the product is not expected to pose a risk to the user, when used in accordance with recommendations.

Environmental risk assessment

A Phase I environmental risk assessment (ERA) was provided 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).

The environmental risk assessment can stop in Phase I and no Phase II assessment is required because the veterinary medicinal product will only be used in a non-food-producing species. Oxmax is not expected to pose a risk for the environment when used according to the SPC.

Residues documentation

Not applicable.

Overall conclusions on the safety documentation

The application is submitted in accordance with Article 12(3) of Directive 2001/82 EC, as amended. Given that Oxmax is classified as MUMS, the safety data presented were in line with the data requirements outlined in the CVMP Guideline on safety and residue data requirements for veterinary medicinal products intended for minor uses or minor species (EMEA/CVMP/SWP/66781/2005 Rev. 1).

Oxmax is considered to be of low acute toxicity potential. In rat acute toxicity studies, the test product was well tolerated at doses up to 6.9 g Hb/kg bw (100 ml product/kg bw). No mortality occurred and no adverse effects were evident clinically. A number of transient and reversible effects on clinicopathological parameters were observed, possibly caused by assay interference due to the presence of haemoglobin in samples arising from Oxmax treatment.

In a pilot repeat-dose toxicity study, the test product was administered on three occasions at 48 hour intervals in anaemic Beagle dogs at approximately 3 x RTD and 5 x RTD. Based on the findings of this limited study, the test product was generally well tolerated; changes in clinical pathological parameters (clinical chemistry and urinalysis) were observed but tended to revert to baseline levels by the end of the study.

Data on reproductive toxicity were not provided. However, according to the CVMP Guideline on the safety and residue data requirements for veterinary medicinal products intended for minor uses or minor species, data in respect of reproductive toxicity (including teratogenicity) are not required for non-food-producing species that are not intended for breeding.

The applicant investigated mutagenic potential in two *in vitro* mutagenicity tests, a bacterial reverse mutation test and a mouse lymphoma assay. In both test systems, there was no evidence that the test item has mutagenic potential. However, an *in vivo* study was been provided. Given the fact that the intended target species for the product was dogs (a non-food producing species), and the results of both *in vitro* studies did not show evidence of any mutagenic potential, the omission of *an in vivo* test for chromosomal effects can be accepted.

The absence of studies investigating the carcinogenicity of the product is accepted based on the negative findings of the mutagenicity studies. In addition, it is accepted that the active substance (hemoglobin betafumaril (bovine)) has no structural similarity to known carcinogens.

An *in vitro* GLP-compliant laboratory study investigating the effect of haemoglobin on clinical chemistry, haematology and urinalysis suggests that the presence of haemoglobin in samples of canine

serum, plasma and urine may result in a statistically significant interference in the assayed levels of a number of biochemical, haematological and urological parameters. Appropriate statements reflecting this information are included in the SPC.

A user safety assessment in line with the relevant guidance document has been presented. Based on the assessment presented, the product does not pose an unacceptable risk to the user when used in accordance with the SPC.

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.

Part 4 – Efficacy

Oxmax 65 mg/ml solution for infusion for dogs is intended for administration to dogs, with the proposed indication at the time of submission of the application 'for the management of canine haemorrhagic shock by increasing tissue oxygenation and achieving a comparable 24-hour survivability with blood.' The solution contains hemoglobin betafumaril (bovine) at a concentration of 65 mg/ml, and is intended to be administered by intravenous infusion. The proposed dose is 10 ml/kg bodyweight (650 mg/kg bw) for administration at an infusion rate of up to 10 ml/kg bodyweight per hour. Hemoglobin betafumaril (bovine) is a new active substance, which has not been authorised for a veterinary medicinal product in the EU at the date of submission of the application.

The product has been granted MUMS status and the principles outlined in the CVMP Guideline for 'Efficacy and target animal safety data requirements for veterinary medicinal products intended for minor uses or minor species' (EMEA/CVMP/EWP/117899/2004-Rev.1) have been applied.

Pharmacodynamics

The pharmacodynamic effect of Oxmax is based on the oxygen-carrying capability of the active substance. The bovine-derived haemoglobin (Hb) in Oxmax is claimed by the applicant to have physical and chemical properties similar to those of canine Hb contained within red blood cells. Since the active substance is not constrained by a cellular membrane (free in plasma), the applicant originally also claimed that it can readily distribute oxygen throughout the circulation, including the smallest capillaries. However, given that it has not been specifically demonstrated that the product can readily distribute oxygen to the 'smallest capillaries', reference to the oxygen distribution capacity of the active substance was amended.

The partial pressure of oxygen in Oxmax (5 batches) resulting in a 50 percent saturation (P_{50}) of Hb ranged from 40 to 50 mmHg, which is higher than that of native bovine Hb of 27-28 mmHg and native canine Hb of 28.7-31.3 mmHg. The higher P_{50} indicates a reduced oxygen affinity of the bovine-derived Hb in Oxmax that is claimed by the applicant to correspond to an increase in oxygen delivery. However, as discussed in the following sections, it is not considered to be adequately supported that the administration of Oxmax correlates with an increase in tissue oxygenation.

The applicant claimed that there is an increase in tissue oxygen tension (TO_2) following Oxmax administration, determined by direct measurements of tissue oxygen tension in rats and in dogs. In support of the tissue oxygenating properties of Oxmax, two non-GLP compliant (pharmacokinetic) studies were submitted investigating tissue oxygenation following intravenous administration of Oxmax, one in dogs in an experimental model of acute normovolaemic anaemia, and one in healthy rats. Furthermore, the pivotal dose-determination study also undertook the evaluation of tissue oxygen tension in dogs in an experimental model of hypovolemic shock.

While the experimental dog study showed a dose-related increase in Hb in the blood of anaemic Beagle dogs following the administration of Oxmax, a corresponding dose-related increase in tissue oxygen tension was not observed, and overall it was considered that data from the study were inconclusive. In the rat study, a direct pharmacodynamic effect on tissue oxygenation was demonstrated in healthy rats following administration of a single intravenous bolus injection of 0.2 g/kg Oxmax. However, the study design was deficient in a number of aspects such that the study could not be considered as pivotal.

In the dose determination study, the results appeared to indicate an effect of treatment with Oxmax at the proposed dose of 10 ml/kg bw on tissue oxygenation. However, the clinical relevance of the observed effect is not clear from this study, that is, it cannot be assumed that the effect on tissue oxygen tension seen in this study is clinically relevant.

Based on the data provided, it is accepted that bovine-derived haemoglobin (Hb) has shown to have physical and chemical properties similar to that of native Hb contained within red blood cells. Since the active substance "hemoglobin betafumaril (bovine)" is not constrained intracellularly but is free in plasma, it can readily distribute oxygen throughout the circulation. There appears to be an effect of treatment with Oxmax on tissue oxygenation; however, the clinical relevance of this effect is not clear.

Following amendment, the text proposed to describe the pharmacodynamic effect of Oxmax in section 5.1 (pharmacodynamic properties) of the SPC was accepted.

Pharmacokinetics

Two studies have been conducted to investigate the pharmacokinetics of Oxmax in healthy Beagle dogs, one non-GLP compliant, the other one GLP-compliant. In addition, a third, non-GLP experimental study was provided in dogs using a model of acute normovolaemic anaemia. Different doses of Oxmax were administered by intravenous infusion at a rate of 10 ml/kg/h, and blood samples were taken for analysis of plasma Hb at various time points after the end of infusion.

The elimination of Oxmax in healthy dogs followed first-order kinetics. At doses between 9.6 ml/kg and 30 ml/kg bodyweight (bw) Oxmax, the C_{max} and area under the curve (AUC) were found to be dose proportional with terminal half-life ranging between 15.5 h and 17.2 h. The pharmacokinetic profile of Oxmax in dogs with acute normovolaemic anaemia was similar to that in healthy animals. C_{max} at the dose range of 15–30 ml/kg bw was 1.5–2.38 g/dl. The mean concentration of Hb at the end of the infusion period in the dose-determination study at a dose of 10 ml/kg bw was 1.1 g/dl.

The volume of distribution indicated that Oxmax following intravenous infusion was largely confined to the vascular system.

Metabolism of Oxmax was not directly investigated. It is expected that Oxmax would be metabolised and eliminated via known metabolic pathways of native Hb involving formation of bilirubin, uptake of bilirubin by the liver and subsequent biliary excretion. Renal excretion is not considered to be a likely major elimination route for Oxmax.

In one non-GLP dog study at doses of 15, 30 and 45 ml/kg bw, $T_{1/2}$ ranged from 17.5 – 42.9 h, and in another non-GLP dog study at doses of 9.6, 19.2 and 38.4 ml/kg bw, $T_{1/2}$ was 17.2, 15.5 and 36.8 h, respectively. In the third, GLP-compliant dog study, the $T_{1/2}$ of Oxmax when administered at 30 ml/kg bw was 16.1 h. The terminal half-life and the elimination time of Oxmax was set to be 17 hours and 5 days respectively.

In general terms, the pharmacokinetic data presented can be considered adequate, and the pharmacokinetics are adequately described in section 5.2 of the SPC.

Dose justification

The proposed dose of 10 ml/kg bw (i.e. 650 mg hemoglobin betafumaril (bovine)/kg bw) for Oxmax was established based on the findings of a dose determination study, investigating the effects of 1, 5 or 10 ml Oxmax/kg bw on the restoration of tissue oxygen tension in a haemorrhagic shock model. Based on the data provided in this study, it was accepted that the proposed dose of 10 ml/kg bw Oxmax would appear to be an appropriate dose for further investigation.

Dose determination / finding studies

The proposed dose for Oxmax (10 ml/kg bw) was established based on the findings of a dose determination study in the USA, investigating tissue oxygen in a haemorrhagic shock model.

In this study, controlled haemorrhage in anaesthetised dogs triggered significant physiological changes consistent with hypovolemic shock and oxygen imbalance.

Thirty Beagle dogs were randomised to five treatment groups to investigate the effects of 1, 5 or 10 ml Oxmax/kg bw on the restoration of tissue oxygen tension and the effect on surrogates of tissue oxygen debt (lactate, base excess, etc.).

Dogs received an intravenous infusion of 20 ml/kg bw of a colloid plasma volume expander (Hextend, HEX) at the same time as either a crystalloid solution (10 ml/kg bw lactated Ringer's solution, LRS) alone (control group), or together with Oxmax, at 1 ml/kg bw (+ LRS 9 ml/kg bw) or 5 ml/kg bw (+ LRS 5 ml/kg bw), or Oxmax alone at 10 ml/kg bw (group 4). An additional group was included that did not receive the colloid plasma volume expander but was treated with 20 ml/kg bw LRS + 10 ml/kg bw Oxmax (group 5).

The total volume infused was 30 ml/kg bw for groups 1 to 4, and 90 ml/kg bw for group 5. For all groups, the duration of infusion was 60 minutes. Dogs were monitored for three hours post-initiation of infusion. A clear dose-dependent effect was observed for certain parameters evaluated; however, many of the parameters/physiological factors that were altered as a result of the hypovolaemic shock model were restored across all the treatment groups.

After the induction of haemorrhagic shock in dogs, the dose of Oxmax at 1 ml/kg bw or 5 ml/kg bw did not significantly increase mean tissue oxygen (TO_2) tension relative to controls. A significant difference in the mean TO_2 tension relative to the control group was observed in the 10 ml/kg bw Oxmax dose groups (groups 4 and 5).

- In the 10 ml/kg bw Oxmax + HEX group (group 4), a significant difference in the mean TO_2 tension was observed at 40 mins after the start of infusion and remained significantly different at all time-points to the end of the analysis at 180 mins after the start of infusion.
- In the 10 ml/kg bw Oxmax + LRS group (group 5), a significant difference in the mean TO_2 tension was observed at 80 (24.4 mmHg), 100 (25.3 mmHg), 120 (22.5 mmHg), 140 (23.3 mmHg) and 160 (15.0 mmHg) mins after the start of infusion.

There was no difference between treatment groups in the time to recovery of tissue oxygen but it was reported that there was a difference in the duration of recovery of tissue oxygen above 10 mmHg in the 5 ml/kg bw and the two 10 ml/kg bw dose groups compared to the control group (150.0 min in the 5 ml/kg + LRS group, 176.7 min in the 10 ml/kg + HEX group, 150.0 min in the 10 ml/kg + LRS group vs. 56.7 min in the control group). However, it is noted that two animals in the control group failed to reach the target value of 10 mmHg, and the '0 mins' duration of recovery for these animals reduced the mean value for the control group; however, it is noted that at the same time all 6 animals in the

10 ml/kg bw + 20 ml/kg bw LRS group reached the target value (as did all animals in the 5 ml/kg bw + LRS and 10 ml/kg bw + HEX groups).

There were no differences in the indirect indices of oxygen debt, lactate and base excess, in any Oxmax treatment group relative to the control group. There was an increase in systolic blood pressure (SBP) by the end of the infusion period relative to controls in groups 3, 4 and 5; and while a significant difference in the duration of haemodynamic recovery (SBP >90 mmHg), was observed only in the 10 ml/kg bw + HEX dose group (180 mins vs 137 mins in the negative control group), it is noted that the duration of haemodynamic recovery was numerically higher in the 10 ml/kg bw Oxmax + 20 ml/kg bw LRS group compared to the control group (173 mins vs 137 mins in the negative control group).

Based on the data provided in this study, it was accepted that the proposed dose of 10 ml/kg bw Oxmax would appear to be an appropriate dose for further investigation.

However, while a statistically significant difference (increase) in the mean TO_2 tension was observed in both of the 10 ml/kg bw Oxmax dose groups (high dose groups) relative to the control group, i.e. when administered in combination with Hextend or LRS, it is not clear whether this increase represents a true physiological benefit of treatment with clinical relevance. At 180 minutes the mean TO_2 level was 12.8 mmHg in the 10 ml/kg bw Oxmax + LRS group (and 17.5 mmHg in the 10 ml/kg bw Oxmax + Hextend group), whereas baseline (pre-shock) mean values for TO_2 for the five study groups ranged from 30.4 to 35.6 mmHg. While the applicant discussed that the TO_2 value associated with normal physiological function is >10 mmHg, it is noted that this relied on a single publication investigating critical O_2 tension in murine fibrosarcomas and the relevance of these findings to the current indication are unclear. Furthermore, the applicant claims that this study provides robust evidence showing that Oxmax increases plasma Hb, thus increasing systemic oxygen content, which leads to improvement in oxygen delivery and tissue oxygenation in dogs with acute haemorrhagic shock. However, in this study, there were no differences in systemic oxygen delivery between the individual treatment groups relative to the control group.

Given that these are the only data on which a claim for tissue oxygenation is based in the application, and that TO_2 levels are not linked with or investigated in the pivotal dose confirmation study, the applicant removed the proposed indication for "increasing tissue oxygenation".

Dose confirmation studies

Although eight laboratory studies are presented in the dossier as pre-clinical studies investigating the efficacy of the product, only one study is considered to be relevant in terms of supporting the proposed indication, the pivotal dose-confirmation study. Three of the studies do not provide any support for the claimed indication or dose of the product, and they are therefore not commented on further here, and four were exploratory studies only.

Exploratory dose confirmation studies:

A brief summary of the four exploratory studies is included in this section, followed by the assessment of the pivotal dose confirmation study. Three of the studies were conducted in China and non-GLP compliant (but stated to have been based on GLP requirements in the People's Republic of China).The first pilot study was conducted in the USA, investigating the effect of a single 10 ml/kg bw dose of Oxmax in combination with 20 ml/kg bw LRS on survival and clinical outcome after the experimental establishment of haemorrhagic shock in Beagle dogs. The primary endpoints were 24-hr survival post-resuscitation and time to death/euthanasia (the study period lasted up 25 hrs from the onset of dosing).

Healthy dogs were sedated, anesthetised and under strict aseptic conditions rendered hypotensive by controlled haemorrhage in order to trigger oxygen supply/demand imbalance, as reflected by an elevated arterial lactate LAC level (8 – 11 mmol/l), an arterial base deficit (BE < -12 mmol/l) and decreased venous oxygen saturation (SvO2 <60%).

Following the induction of haemorrhagic shock and baseline data collection, 20 Beagle dogs were randomly assigned to receive either control (LRS, group 1) or test article (Oxmax, group 2) in the setting of concomitant standard crystalloid-based volume therapy (LRS, 20 ml/kg bw). A third treatment group (a colloid plasma volume expander, group 3, n=10) was added via protocol amendment. Overall, 30 ml/kg bw of total fluid volume were administered for each animal/group at a cumulative rate of 30 ml/kg bw/h.

Following the end of the dosing period, dogs were recovered (end of anaesthesia) and monitored (clinical signs, haemodynamics, blood gases and chemistries) for up to 24 hours post-resuscitation. No additional therapy was administered over the follow-up period. Analgesia was given as necessary throughout the recovery period. At the end of the follow-up period, surviving dogs were euthanised according to AVMA guidelines.

The results demonstrated that the survival rate was 30% in group 1 animals, 60% in group 2 treated animals, and 60% in group 3 animals. The differences between groups were not statistically significant; however, the study was underpowered to detect non-adjusted categorical differences in survival. Plasma Hb concentrations increased significantly in Oxmax-treated dogs reaching mean levels of 1.67 ± 0.17 g/dL at the end of dosing.

Overall, this study can be viewed as an exploratory pilot study to compare survival time at 24 hours after completion of treatment in different groups.

The second pilot, exploratory study was conducted in China to further explore the design of the pivotal dose confirmation study.

In this study, the effect of a single 10 ml/kg bw dose of Oxmax in combination with a crystalloid solution on survival and clinical outcome within 121 hours after the experimental establishment of haemorrhagic shock in Beagle dogs was investigated, compared to a volume matched control group receiving colloids in combination with crystalloids. This was a 3-arm randomised controlled trial, in which healthy dogs were anesthetised and under strict aseptic conditions rendered hypotensive by controlled haemorrhage. Following the induction of haemorrhagic shock, dogs were randomly assigned to one of three study groups receiving either control (group E) or test article (groups F and G, these groups differed with respect to the batch of Oxmax used). Following the end of the 60 minute dosing period, dogs were monitored (clinical signs, haemodynamics, blood gases and chemistries) for up to 120 h post-resuscitation. At the end of the follow-up period, surviving dogs were euthanised. The primary efficacy endpoints included 24 and 120 hour post-resuscitation survival.

The survival rate at 24 hours and at 120 hours post-resuscitation was evaluated, and based on the absence of further deaths/euthanasia after the 24 hours post-resuscitation period, it was concluded that survival at 24 hours post-treatment would be a suitable time-point for analysis of efficacy endpoints. Overall, the survival rate of animals of the Oxmax groups was higher than the survival rate in the control group; 91.7% (11/12) compared to 66.7% (4/6), respectively.

Overall, it can be accepted that the results of this pilot, exploratory study support the evaluation of the survival rate at 24 hours after the completion of administration of test article, when administered at the proposed dose of 10 ml/kg bw concomitantly with 20 ml/kg bw LRS. However, given the limited sample size, little else can be concluded.

The third pilot, exploratory study was conducted in China to further explore the design of the pivotal dose confirmation study.

In this study, the effect of a single 10 ml/kg bw dose of Oxmax in combination with a crystalloid solution on survival and clinical outcome at 25 hours after the experimental establishment of haemorrhagic shock in anaesthetised Beagle dogs was investigated, compared to a control group receiving colloids (COP matched) in combination with a crystalloid solution. The survival rate at 24 hours was compared between groups; 5/6 dogs in the Oxmax group, vs 4/6 dogs in the control group, survived at 24 hours after the end of the treatment. Overall, given the limited sample size, little can be concluded from this study.

A fourth pilot, exploratory single-arm study conducted in China was provided to evaluate the efficacy of whole blood on 24-hour post-resuscitation survival rate in a model of haemorrhage-induced controlled oxygen imbalance in anaesthetised Beagle dogs and was conducted in order to determine the study design and sample size for the dose confirmation study.

Following the induction of haemorrhagic shock, autologous whole blood (10 ml/kg bw/h) and LRS (20 ml/kg bw/h) were administered to animals (n=10). Following the end of the 60 minute dosing period, dogs were monitored (clinical signs, haemodynamics, blood gases and chemistries) for 24 hours post-resuscitation. At the end of the follow-up period, surviving dogs were euthanised.

Of the ten dogs for which haemorrhagic shock was successfully established (3 dogs died during shock induction), 4/10 died or were euthanised in the treatment period, while the remaining 6/10 dogs survived until the end of the study, at 24 hours post-treatment. Therefore, under the conditions of this study, the administration of autologous whole blood in combination with a crystalloid solution, resulted in a 60% survival rate at 24 hours post-treatment.

The data from this study does not have any direct impact on the evaluation of the efficacy of Oxmax; however, it is of relevance as it provides information on the anticipated 24 hour survival rate (under the same experimental model of canine haemorrhagic shock used in the subsequent pivotal dose confirmation study) following the administration of autologous whole blood, which could be considered to represent one of the best available treatments in the management of acute haemorrhagic shock in the (unlikely) clinical situation that autologous whole blood would be available for the patient.

***Pivotal dose confirmation study*:**

The pivotal dose confirmation study was designed to test the hypothesis that treatment with Oxmax (10 ml/kg bw) was non-inferior to treatment with whole blood (10 ml/kg bw, from a compatible donor, non-autologous) for the primary efficacy endpoint; survival rate at 25 hours after the onset of treatment. Both test and control articles were administered in combination with crystalloids; 20 ml/kg bw LRS, as a concomitant infusion with a duration of one hour (total volume of fluid administered; 30 ml/kg bw), i.e. low volume resuscitation, following the induction of severe haemorrhagic shock. The study was conducted in the People's Republic of China and is stated to have adhered to the principles of GLP in that region and to VICH GL9 (GCP), where possible. The applicant clarified that the deviations alluded to (i.e., reference to "where possible") relate to deviation from GLP requirements for specific instrumentation and confirmed that the study was conducted according to GCP. The fact that a small number of instruments used for measurements were not strictly GLP-compliant is not considered to have had an adverse impact on the study outcome given that the instruments were used in accordance with manufacturer's recommendations.

The study was a two-arm randomised, blinded, controlled trial, adaptive non-inferiority design. It was intended that at least 30 dogs would be included in each of the two treatment groups. Healthy dogs were anaesthetised and under strict aseptic conditions rendered hypotensive by controlled

haemorrhage in order to trigger oxygen supply/demand imbalance as reflected by an elevated arterial lactate LAC level (9 – 11 mmol/l), an arterial base deficit (BE < -12 mmol/l) and decreased venous oxygen saturation (SvO2 <60%). Following the induction of haemorrhagic shock and baseline data collection, 10 ml/kg bw Oxmax and 20 ml/kg bw LRS or 10 ml/kg bw whole blood (WB) and 20 ml/kg bw LRS were administered via intravenous infusion. Following the end of the 60 minutes dosing period, dogs were monitored (clinical signs, haemodynamics, blood gases and chemistries) for 24 hours post-resuscitation. No other resuscitative fluids/treatments were administered during the follow-up period.

The primary efficacy endpoint was survival rate at 25 hours from the onset of treatment. The primary aim of the study was to show non-inferiority of Oxmax in the treatment of acute haemorrhagic shock compared to administration of whole blood at 25 hours from the onset of treatment, with a non-inferiority margin of 15%. An interim analysis was performed at the end of stage I, i.e., after at least 30 animals in each group had completed 25 hours observation (from the onset of treatment). Secondary outcome measures included cardiovascular and respiratory system parameters, physical examination parameters, rectal temperature, Glasgow Pain score and laboratory parameters.

The results of the primary efficacy parameter demonstrated that the survival rate was marginally higher (numerically) in the Oxmax-treated group; i.e. 80% (24/30 dogs) compared to the whole blood-treated group (78.4%, 29/37), in the per protocol population. However, the statistical analysis failed to demonstrate non-inferiority of Oxmax treatment against whole blood treatment, using the pre-specified non-inferiority margin of 15%. The applicant claims that these data demonstrate comparable survival rates in both groups, notwithstanding that the statistical analysis failed to demonstrate non-inferiority of Oxmax treatment against whole blood treatment.

In general, the applicant has followed CVMP Scientific Advice regarding the design of this pivotal study in respect of several aspects. Originally a larger number of dogs were proposed to be included in the studies however the CVMP highlighted at that time that sample size calculations would likely result in very significant ethical concerns in relation to the relevant 3Rs legislation in the EU/EEA (Directive 2010/63/EU). On this point, it is noted that a total of 76 animals were included in the study., including 6 dogs that died during shock induction, resulting in a total of 70 dogs in the Intention-To-Treat population that were successfully randomised to test or control treatment.

The primary efficacy endpoint used, survival rate at 24 hours post-treatment, was in line with CVMP advice given. However, it was noted that in the context of CVMP advice given (not specifically related to this study) the low dose model employed in this study may have restricted application to real-life clinical practice where either high dose (90 ml/kg bw) volume replacement or repeated (boluses) low-dose volume replacement would often be indicated. As the low dose model was used, the indication in the SPC clearly states that a beneficial effect of treatment was demonstrated in the context of concomitant use of low dose resuscitative fluids (Lactated Ringer's solution).

Overall, following satisfactory clarification of a number of questions raised concerning blinding and conduct of the study, it was accepted that the study was conducted to an acceptable standard with extensive data generated and study data comprehensively reported.

The study report was entitled 'Interim Study Report' due to the fact that it was planned that at the end of stage I of the study (non-inferiority study), an interim analysis was to be conducted (on a blinded basis), and, if non-inferiority could not be demonstrated, the study would proceed to stage II, with recruitment of additional numbers of animals. The outcome of these analyses was that the Oxmax group could not be demonstrated as non-inferior to the whole blood group; however, neither could the group treated with whole blood be demonstrated as non-inferior to the Oxmax group. However, based upon the interim analysis, it would have been considered necessary to proceed with 61 animals per

group for stage II of the study (assuming that group A received Oxmax). Overall, it is clear that notably larger numbers of animals would need to be included to demonstrate non-inferiority of Oxmax treatment over whole blood given the survival rate in animals administered whole blood and the selected non-inferiority margin. The application was submitted with data based on the interim analysis, and the applicant did not proceed with stage II of the study on the basis that the large number of animals required would have been unethical and not in line with the 3R principles.

Furthermore, it should be noted that under the conditions of the fourth pilot exploratory study the administration of autologous whole blood in combination with LRS resulted in a 60% survival rate at 24 hours post-treatment. That value (60%) was used in estimating the required sample size for this study. However, in this pivotal dose determination study, the survival rate in the whole blood group was substantially higher at 78.4%. Therefore, this difference in anticipated survival vs actual survival in the positive control group was unforeseen by the applicant and is likely to have rendered the study sample size inadequate to demonstrate non-inferiority using the pre-specified non-inferiority margin of 15%.

From the perspective of clinical relevance of effect, the following are noted:

The primary efficacy endpoint, survival rate at 25 hours after onset of treatment is considered to be a robust, clinically relevant measure of efficacy of Oxmax. The ultimate aim of treatment in acute trauma/haemorrhagic shock situations in dogs is to rescue the animal from imminent death, and the function of a haemoglobin-based oxygen carrier is to substitute for a blood product in an emergency situation, when blood is not readily available and/or a delay is required prior to administration, e.g. in order to type blood to recipient. Therefore, survival at 25 hours after the onset of treatment compared with the gold standard, whole blood, is a highly clinically relevant indicator of effect, in particular noting that no further resuscitation fluids were administered in the follow-up period, and it suggests that efficacy has been investigated under challenging conditions in this study.

The survival rate was comparable; 1.6% higher in the Oxmax group compared to the whole blood positive control group (using the PP population). With at least 30 animals per group, under the highly controlled nature of the experiments conducted, the observed treatment effect of Oxmax would appear to be clinically relevant (estimated 80% survival rate at 24 hours), notwithstanding the fact that the sample size was inadequate to demonstrate non-inferiority (using the pre-specified non-inferiority margin of 15%).

Restoration of haemodynamic and clinical parameters in the post-resuscitation phase was comparable among the Oxmax and whole blood groups, and was faster/better for some parameters following Oxmax treatment (e.g. mean arterial blood pressure reduction of base deficit).

The applicant was requested to further discuss the extent to which Oxmax may be considered comparable to administration of whole blood in terms of 24 hour survival with reference to the statistical analyses conducted and most importantly, the clinical relevance of those findings. The applicant, whilst acknowledging the fact that non-inferiority with whole blood could not be demonstrated, justified the clinical relevance of the treatment effect to support the use of this product in a clinical setting, taking into account the acute nature of the condition for which the product is indicated. Although non-inferiority with whole blood was not proven (too few animals to achieve statistical significance for non-inferiority), the number of animals included in the test and control groups (30 vs 37) in the per protocol population could be considered sufficient to enable a conclusion on the effectiveness of treatment, taking into account the 3R principles and the MUMS status of this application, with convincing results showing similar survival rates in both groups. It is also acknowledged that comparison with whole blood as the 'gold standard' is a high bar against which to compare the efficacy of a haemoglobin-based oxygen carrier. Furthermore, the applicant

acknowledged that whole blood would be preferable to use in emergency situations, if available, but in the event that this is not readily available or compatible for immediate use (as is frequently the situation in clinical practice), Oxmax could fulfil a currently unmet clinical need.

Overall, the CVMP accepted that sufficient data are available to adequately support the clinical relevance of the findings of the study, and that a positive effect of treatment can be accepted.: similar survival rates at 24 hours post-resuscitation were observed in a test group treated with Oxmax + LRS for one hour compared to a control group that was treated with matched, non-autologous whole blood + LRS for one hour. Notwithstanding that the contribution of the administration of LRS for one hour to the survival rate cannot be evaluated alone, it can be assumed that whole blood + LRS is preferable to LRS alone, and would be expected to result in an improved clinical outcome. Therefore, by implication, Oxmax + LRS, with a similar survival rate to the whole blood + LRS group, can be considered as an enhancement to treatment than administration of LRS alone.

The CVMP considered that an acceptable 'indication for use' for Oxmax is as follows: "*For adjunct therapy in the management of canine haemorrhagic shock. A beneficial effect of treatment was demonstrated for 24 hour survival rate when Oxmax was administered concomitantly with low dose resuscitative fluids (Lactated Ringer's solution).*" The applicant updated the product information accordingly.

Target animal tolerance

The local and systemic tolerance of Oxmax was investigated in the pivotal GLP-compliant target animal safety study in a model of acute normovolaemic anaemia using the recommended route of administration (intravenous) in 32 splenectomised, but otherwise healthy Beagle dogs. The study was conducted in 2012 in the USA, and investigated under conditions mimicking the proposed conditions of use of Oxmax at doses of 30, 60 and 90 ml/kg bw (1950, 3900 and 5850 mg/kg bw hemoglobin betafumaril (bovine)), i.e. 3x, 6x and 9x the recommended treatment dose (RTD) in group sizes of 6, 6 and 10 dogs, respectively, with a repeat dose administered after an interval of four days. A control group (n=10) was included (administered 90 ml/kg bw modified Acetated Ringer's Solution). Anaemia was induced using blood withdrawal and simultaneous replacement with HESpan to induce normovolaemic anaemia during a series of collection intervals over a 1.5 – 3.5 hour period until the haematocrit measured 20 ± 1%. Animals recovered from anaesthesia for 30 – 60 mins prior to test article administration on day 1, followed by retreatment on day 4. The study was conducted at a time at which the proposed maximum dose for the product was considered to be 30 ml/kg bw, thus the doses used in this target animal safety study reflected what was to be considered 1x, 2x and 3x a previously considered maximum RTD. Although VICH GL43 recommends the use of healthy animals for such studies, the proposed indications for use are in dogs with acute haemorrhagic shock, i.e. animals that are in a critical situation for which death is imminent in the absence of resuscitative treatment. Therefore, the applicant considered that the evaluation of safety parameters in dogs that are already severely compromised would be more relevant than the evaluation of tolerance in healthy dogs.

With respect to investigating the margin of safety, the study included multiples of doses above the recommended dose of 10 ml/kg bw, and given that the infusion rate is constant (10 ml/kg bw/h), the inclusion of 3x, 6x and 9x overdoses encompassed a longer duration of use (infusion times were 3, 6 and 9 hours for the 3x, 6x and 9x dose groups, respectively).

A special warning is included in the SPC (section 4.3) against using Oxmax more than once, therefore the inclusion of a repeat dose administered four days after initial infusion represents a worst case scenario that should not occur in a clinical setting. The majority of animals were followed until two days after the repeat dose, except for two animals in the highest dose group, which were monitored for 24 days after the repeat dose to determine the recovery phase.

No deaths occurred during the study. Vomiting occurred in most animals in the 90 ml/kg bw dose group and in some animals in the 60 ml/kg bw group and was considered to be test-article related. Infusion site reactions (erythema or swelling), which were not dose-dependent, occurred in ten (out of 32) animals treated with Oxmax.

Treatment-related adverse reactions included discolouration of mucous membranes, urine and faeces in all dose groups, with the incidence and severity increasing with higher doses. At necropsy, pink to red discolouration of multiple tissues was observed. Histopathology revealed kidney lesions in all animals in Oxmax treatment groups at necropsy two days after the second infusion. These changes (minimal to mild) mainly concerned degeneration/necrosis of cortical epithelial cells and/or hyaline droplet deposition in tubular epithelial cells, and the incidence and severity was decreased by 24 days after the second infusion in the 90 ml/kg bw group.

Clinical pathology tests (chemistry, haematology, urinalysis) were affected by the presence of Oxmax in the plasma; however, these were mainly related to the experimental induction of anaemia in all groups and/or the infusion of bovine Hb in the Oxmax treatment groups. An increase in urinary volume was observed in all Oxmax groups compared to controls (and a decrease in urine specific gravity in the 30 ml/kg bw group) and these changes were considered related to the renal tubular lesions observed at necropsy two days after the second infusion. No changes on urinalysis parameters were observed at Day 16 or Day 28 in the cohort of animals included in the 90 ml/kg bw group which were included to determine the recovery phase.

In conclusion, Oxmax at doses up to 90 ml/kg bw appears to be generally well tolerated. The applicant considered that a no-observed adverse effect level (NOAEL) of 60 ml/kg bw was appropriate, although given that minimal to mild renal abnormalities were observed even in the 30 ml/kg bw Oxmax group, the CVMP considered that a NOEL cannot be established from this study. It is further noted that discolouration of mucous membranes, urine and faeces, and multiple tissues at necropsy were recorded in all dose groups. It is clearly stated in the SPC that renal function should be monitored during and after use of the veterinary medicinal product, and guidance is given concerning clinical signs of renal impairment and tools to assess renal function. The renal pathology observed in the target animal safety study at dose levels of 30, 60 and 90 ml/kg bw is included under the 'Overdose' section of the SPC. Overall, taking into account that the potential risk of renal damage is clearly stated and risk mitigation measures are included in the SPC, it can be accepted that the renal damage observed is unlikely to represent an unacceptable risk to the target species in an emergency situation at the proposed dose of 10 ml/kg bw.

In addition to the pivotal target animal safety study, the tolerance of Oxmax in dogs was also evaluated during the pharmacology, toxicology and clinical laboratory studies conducted with the product during product development.

Overall, concerning target animal tolerance, it is concluded that Oxmax administration at the recommended dose is associated with the following adverse reactions:

- Alterations of faeces; soft/loose stools, diarrhoea, discolouration of faeces, bloody faeces,
- Vomiting,
- Sneezing,
- Discolouration of mucous membranes, tissues, urine and faeces,
- Reactions at the site of infusion (redness and/or swelling).

Typically, the treatment-related adverse effects observed were mild and transient.

Overdosing or too rapid administration of Oxmax, may lead to circulatory overload with associated clinical signs. The applicant claims that the small average molecular weight of Oxmax (65 kDA) explains why circulatory overload was rarely observed in Oxmax-treated animals.

Treatment-related renal histopathology was observed in the GLP target animal safety study following two intravenous infusions of Oxmax at all doses (30, 60 and 90 ml/kg bw).

Although hypersensitivity reactions were not observed in the pivotal target animal safety study, the time interval between initial and repeat dose was too short (4 days) to adequately evaluate the potential for this type of reaction. Furthermore, even if the time interval between first and second dose had been longer, rare cases of hypersensitivity may not have manifested in a laboratory study with relatively low numbers of animals included. The applicant recommends that the product is not used in dogs previously treated with the product or other bovine Hb-based oxygen carrier, to avoid a potential sensitivity-type reaction upon repeat exposure. This proposal is considered appropriate.

It should be noted that changes in laboratory assays may be subject to interference by plasma Hb (associated with Oxmax administration) and should be interpreted with caution (see also part 3, 'Interference with colorimetric assays').

The applicant updated the description of adverse events in the SPC as follows: *'Diarrhoea, abnormal stool colouration, blood in faeces, vomiting, shivering, sneezing, injection site reddening and injection site swelling were very commonly reported. Discolouration of mucous membranes (necropsy finding), discolouration of tissues (necropsy finding) and discoloured urine were commonly reported.'*

The proposed text for inclusion in section 4.10 of the SPC (overdose) is considered acceptable.

In conclusion, based on the target animal safety data, the CVMP considered that Oxmax is generally well-tolerated in dogs up to doses of up to 90 ml/kg bw. While it is accepted that some findings throughout the studies conducted are likely related to the canine haemorrhagic shock model, the SPC and product information contain adequate warnings about the observed adverse effects and their frequency.

Clinical field trials

No field trials have been conducted for Oxmax.

The CVMP previously indicated in a scientific advice that an appropriately designed and controlled clinical field trial is strongly recommended. However, for this MUMS product, in principle, a good quality laboratory study, in which a clinically beneficial effect of treatment is confirmed and where data generated could be extrapolated to the field situation, would negate the need for a field study. The inherent difficulties in recruiting clinical cases with severe haemorrhage at the same time point in the shock/oxygen debt cycle are appreciated by the CVMP. Furthermore, the CVMP accepted that the basic outline of the shock model proposed in the applicant's protocol is well established in the scientific literature.

Therefore, in line with MUMS requirements, the lack of clinical field trials can be accepted.

Overall conclusion on efficacy

Pharmacodynamics:

Oxmax is a haemoglobin-based oxygen carrier, containing a solution of bovine-derived haemoglobin with physical and chemical properties similar to that of native haemoglobin contained within red blood

cells. Since the active substance is not constrained by a cellular membrane, the active substance can readily distribute oxygen throughout the circulation.

Pharmacokinetics:

The pharmacokinetic characteristics of Oxmax are generally well documented and have been satisfactorily evaluated in dogs. The elimination of Oxmax in healthy dogs follows first-order kinetics. Between 9.6 ml/kg and 30 ml/kg bodyweight (bw) Oxmax, the C_{max} and area under the curve (AUC) were found to be dose proportional with terminal half-life ranging between 15.5 h and 17.2 h. The pharmacokinetic profile of Oxmax in dogs with acute normovolaemic anaemia was similar to that in healthy animals. C_{max} at overdose (range of 15–30 ml/kg bw) was 1.5–2.38 g/dl. The mean concentration of Hb at the end of the infusion period in the dose-determination study at the recommended dose of 10 ml/kg bw was 1.1 g/dl. The volume of distribution indicated that Oxmax following intravenous infusion was largely confined to the vascular system. Metabolism of Oxmax was not directly investigated. It is expected that Oxmax is metabolised and eliminated via known metabolic pathways of native haemoglobin involving formation of bilirubin, uptake of bilirubin by the liver and subsequent biliary excretion. Renal excretion is not considered to be a likely major elimination route for Oxmax.

Dose confirmation:

The pivotal dose confirmation study showed a similar 25h survival in dogs treated with 10 ml/kg bodyweight Oxmax as for dogs treated with whole blood (both in combination with LRS and administered intravenously at a rate of up to 10 ml/kg bw/h) using a severe haemorrhagic shock experimental model. In addition, a number of supportive experimental studies were provided. It can be concluded that a dose of 10 ml/kg bw Oxmax in combination with LRS is an appropriate dose for confirmation of an effect of treatment on the survival rate.

Tolerance:

Oxmax is generally well-tolerated in dogs up to doses of 90 ml/kg bw.
In the target animal safety study, following two intravenous infusions of Oxmax at doses up to 9x the recommended dose, treatment-related adverse reactions included discolouration of mucous membranes, urine and faeces, with the incidence and severity increasing with higher doses. At necropsy, pink to red discolouration of multiple tissues was observed and histopathology revealed kidney lesions in all animals in Oxmax treatment groups at necropsy two days after a second infusion of the product, which were decreased by 24 days after the second infusion in the 90 ml/kg bw group. Clinical pathology tests (chemistry, haematology, urinalysis) were affected; however, these were mainly related to the experimental induction of anaemia in all groups and/or the infusion of bovine Hb in the Oxmax treatment groups. An increase in urinary volume was observed in all Oxmax groups compared to controls and these changes were considered related to the renal tubular lesions observed at necropsy. Taking into account that the potential risk of renal damage is clearly stated, and risk mitigation measures are included in the SPC, it can be accepted that the renal damage observed is unlikely to represent an unacceptable risk to the target species in an emergency situation at the proposed dose of 10 ml/kg bw.

In addition to the pivotal GLP target animal safety study, the tolerance of Oxmax in dogs was also evaluated during the pharmacology, toxicology and clinical laboratory studies conducted with the product during product development. Overall, Oxmax administration is associated with the following adverse reactions: alterations of faeces (diarrhoea, discoloured faeces, blood faeces), vomiting, shivering, sneezing and reactions at the site of infusion (redness and/or swelling), all of which were very commonly reported. Discolouration of mucous membranes, tissues and urine were commonly

reported. The potential adverse effects associated with Oxmax treatment are clearly described in the SPC.

Efficacy:

At the time of submission, the applicant applied for the following indication 'For the management of canine haemorrhagic shock by increasing tissue oxygenation and achieving a comparable 24 hour survivability with blood.' In support of the efficacy, only laboratory studies were submitted.

Increase in tissue oxygenation:

With respect to the claim for an increase in tissue oxygenation, in the dose determination study using a haemorrhagic shock model in dogs, a statistically significant difference (increase) in the mean tissue oxygen (TO_2) tension was observed, following the administration of Oxmax at 10 ml/kg bw relative to the control group.
However, while baseline (pre-shock) mean values for TO_2 for the five study groups ranged from 30.4 to 35.6 mmHg, notwithstanding that there was a statistically significant difference in TO_2, it was not clear whether this increase represents a true physiologically benefit of treatment, since at 180 minutes the mean TO_2 level was 12.8 mmHg in the 10 ml/kg bw Oxmax + LRS group (and 17.5 mMHg in the 10 ml/kg bw Oxmax + Hextend group). While the applicant has discussed that the TO_2 value associated with normal physiological function is >10 mmHg, it is noted that this relies on one publication investigating critical O_2 tension in murine fibrosarcomas and the relevance of these findings to the current indication are unclear. Furthermore, the applicant claims that this study provides robust evidence showing that Oxmax increases plasma HB, thus increasing systemic oxygen content, which leads to improvement in oxygen delivery and tissue oxygenation in dogs with acute haemorrhagic shock. However, in this study, there were no differences in systemic oxygen delivery between the individual treatment groups relative to the control group.
Given that these are the only data on which a claim for tissue oxygenation is based, and that TO_2 levels are not linked with or investigated in the pivotal dose confirmation study, this claim was not considered to have been appropriately supported and was therefore later removed from the indications.

Comparable 24 hour survival as with blood

The pivotal dose confirmation study investigated the efficacy of a single treatment of 10 ml/kg bw Oxmax in combination with 20 ml/kg bw LRS, at an overall administration rate of 30 ml/kg bw fluid over a 1 hour infusion period, in comparison to the control group that received 10 ml/kg bw recipient-matched non-autologous whole blood in combination with 20 ml/kg bw LRS, following the experimental induction of canine haemorrhagic shock. The primary efficacy endpoint was survival rate at 25 hours from the onset of treatment. The primary aim of the study was to show non-inferiority of Oxmax compared to whole blood at 25 hours from the onset of treatment. No other resuscitative fluids/treatments were administered during the follow-up period. The results demonstrated a numerically comparable 24 hour survival in the Oxmax group (24/30 dogs 80%) compared to the whole blood group (29/37 dogs, 78.4%). Non-inferiority with whole blood could not be demonstrated though (using the pre-specified non-inferiority margin of 15%). However, it is assumed by the applicant that the larger survival rate (78.4%) in the control animals than that anticipated based on a pilot study where 60% of animals survived to 24 hours post-treatment is a likely explanation for having failed to demonstrate non-inferiority given that the sample size was estimated based upon a 60% survival rate in the control group. In addition, it is accepted that strict criteria were employed to demonstrate efficacy, considering that whole blood would be the ideal/optimal treatment (in combination with crystalloid/colloid resuscitative fluid). In addition, the fact that no other treatment was administered during the follow-up period, as may be the case in clinical practice if resuscitation

strategy depends on a low dose fluid replacement, suggests that effectiveness has been investigated under challenging conditions. Therefore, although non-inferiority with whole blood could not be demonstrated in this study, the clinical relevance of the study findings was considered to have been sufficiently justified, also taking into account the MUMS nature of the application. However, the proposed claim for a comparable 24 hour survivability with whole blood was not considered to adequately reflect the study outcome, noting that non-inferiority with whole blood could not be demonstrated.

In addition, it was noted that the originally claimed indication for Oxmax made no reference to use as an adjunct to resuscitative fluid therapy. Taking into account that the dose-determination study and the pivotal dose-confirmation study have both investigated the efficacy of Oxmax in combination with the administration of resuscitative fluids (colloid or crystalloid in the dose determination study, crystalloid in new pivotal dose confirmation study), the CVMP considered it appropriate that the SPC should be amended to include a restriction that efficacy was evaluated in the setting of concomitant administration of low dose crystalloid fluids (LRS) in section 4.2, together with appropriate information in section 4.9. In summary, the CVMP considered that the indications for use that were adequately supported for Oxmax were as follows "Indicated as an adjunct therapy in the management of canine haemorrhagic shock. A beneficial effect of treatment was demonstrated for 24 hour survival rate when Oxmax was administered concomitantly with low dose resuscitative fluids (Lactated Ringer's solution)." The applicant updated the SPC accordingly.

Part 5 – Benefit-risk assessment

Introduction

Oxmax is a solution for intravenous infusion in dogs containing 65 mg/ml hemoglobin betafumaril (bovine) as the active substance. Hemoglobin betafumaril (bovine) is a blood substitute acting as oxygen-carrier, with physical and chemical properties similar to that of haemoglobin contained within red blood cells.

Oxmax is indicated as an adjunct therapy in the management of canine haemorrhagic shock. A beneficial effect of treatment was demonstrated for 24 hour survival rate when Oxmax was administered concomitantly with low dose resuscitative fluids (Lactated Ringer's solution).

The proposed dose is 10 ml/kg bodyweight (i.e. 650 mg/kg bw hemoglobin betafumaril (bovine)) administered intravenously at a rate of up to 10 ml/kg bw/h.

The dossier has been submitted in line with the requirements for submissions under Article 31 of Regulation (EC) No 726/2004 of 31 March 2004; and in accordance with Article 3(2)a, as the product contains a new active substance, which was not authorised in the Community on the date of entry into force of the Regulation.

The product has been classified as MUMS/limited market and therefore reduced data requirements apply that have been considered in the assessment.

Benefit assessment

Direct therapeutic benefit

The proposed benefit of Oxmax is its efficacy as an adjunct therapy in the management of canine haemorrhagic shock. As shown in a laboratory study using a haemorrhagic shock model in dogs, a

beneficial effect of treatment was demonstrated for 24 hour survival rate when Oxmax was administered intravenously at a dose of 10 ml/kg bodyweight at a rate of 10 ml/kg bw/h concomitantly with low dose resuscitative fluids (Lactated Ringer's solution).

Additional benefits

Oxmax increases the range of available treatment possibilities for the management of canine haemorrhagic shock.

Risk assessment

Quality:

Information on development, manufacture and control of the active substance and finished product has been presented in a satisfactory manner. The results of tests carried out indicate consistency and uniformity of important product quality characteristics, and these in turn lead to the conclusion that the product should have a satisfactory and uniform performance in clinical use.

Safety

Risks for the target animal:

The product appears to be generally well-tolerated in dogs at doses up to 90 ml/kg bw. When used in accordance with the proposed recommendations, the product is not expected to pose an unacceptable risk to the animal on the basis of the characterised adverse reactions.

Treatment-related adverse effects observed were mild and transient (diarrhoea, blood faeces, vomiting, shivering, sneezing, discolouration of mucous membranes, tissues, urine and faeces, and reactions at the site of infusion).

Treatment-related renal histopathology was observed following two intravenous infusions of Oxmax at overdoses of 30, 60 and 90 ml/kg bw.

Overdosing, or too rapid administration of Oxmax, may lead to circulatory overload with associated clinical signs. Adequate precautions are included in the SPC to warn the user of possible cardiopulmonary effects following overdose or an excessive rate of infusion.

Changes in laboratory assays may be subject to interference by plasma Hb (associated with Oxmax administration) and should be interpreted with caution; this information is included in the SPC.

Risk for the user:

The product is not expected to present an unacceptable risk to the user when used in line with the SPC recommendations.

Risk for the environment:

The product is not expected to pose a risk for the environment when used in line with the SPC recommendations.

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 and the user and to provide advice on how to prevent or reduce these risks.

Evaluation of the benefit-risk balance

At the time of submission, the applicant applied for the following indication: For the management of canine haemorrhagic shock by increasing tissue oxygenation and achieving a comparable 24 hour survivability with blood.' The product has been shown to be efficacious as an adjunct therapy in management of canine haemorrhagic shock and the CVMP agreed to the following indication: 'Indicated as an adjunct therapy in the management of canine haemorrhagic shock. A beneficial effect of treatment was demonstrated for 24 hour survival rate when Oxmax was administered concomitantly with low dose resuscitative fluids (Lactated Ringer's solution).' 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 and the environment when used as recommended. Appropriate precautionary measures have been included in the SPC and other product information.

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

Conclusion

Based on the original and complementary data presented on quality, safety and efficacy the Committee for Medicinal Products for Veterinary Use (CVMP) considers that the application for Oxmax 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.