10 October 2024
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Veterinary Medicines Division

Committee for Veterinary Medicinal Products (CVMP)

CVMP assessment report for BRAVECTO TriUNO (EMEA/V/C/006311/0000)

INN: Fluralaner / Moxidectin / Pyrantel

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

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Introduction

The applicant Intervet International B.V. submitted on 1 September 2023 an application for a marketing authorisation to the European Medicines Agency (The Agency) for Bravecto TriUNO, through the centralised procedure under Article 42(4) of Regulation (EU) 2019/6 (optional scope).

The eligibility to the centralised procedure was agreed upon by the CVMP on 22 March 2023 as no other marketing authorisation has been granted for the veterinary medicinal product within the Union.

At the time of submission, the applicant applied for the following indications:

For dogs with, or at risk from, mixed parasitic infestations by ticks or fleas, gastrointestinal nematodes, lungworm and/or heartworm. The veterinary medicinal product is only indicated when use against ticks or fleas and one or more of the other target parasites is indicated at the same time.

For the treatment of tick and flea infestations in dogs providing immediate and persistent flea (Ctenocephalides felis and C. canis) and tick (Dermacentor reticulatus, Ixodes hexagonus, I. ricinus, and Rhipicephalus sanguineus) killing activity for 1 month.

The veterinary medicinal product can be used as part of a treatment strategy for the control of flea allergy dermatitis (FAD).

For reduction of the risk of infection with Babesia canis via transmission by D. reticulatus for 1 month. The effect is indirect due to the product's activity against the vector.

For reduction of the risk of infection with Dipylidium caninum via transmission by C. felis for 1 month. The effect is indirect due to the product's activity against the vector.

Treatment of infections with gastrointestinal nematodes of the following species: roundworms (immature adult (L5) and adult stages of Toxocara canis, and adult stages of Toxascaris leonina) and hookworms (L4, immature adult (L5) and adult stages of Ancylostoma caninum and adult stages of Uncinaria stenocephala).

Prevention of heartworm disease (caused by Dirofilaria immitis).

Prevention of angiostrongylosis (by reduction of the level of infection with immature adult (L5) and adult stages of Angiostrongylus vasorum).

The active substances of Bravecto TriUNO are fluralaner, moxidectin and pyrantel. Fluralaner is an ectoparasiticide belonging to the isoxazoline group, with inhibitory activity on GABA- and glutamate-gated chloride channel located in nervous system of invertebrates, preventing the postsynaptic uptake of chloride ions by GABA- and glutamate-gated ion channels and thus resulting in depolarization, paralysis and death of the target parasite. Moxidectin belongs to the milbemycin group of macrocyclic lactones and has parasiticidal activity against nematodes. It interferes with neuromuscular transmission of the glutamate-gated chloride channels and of GABA-gated channels, thus leading to the opening of the chloride channels on the postsynaptic junction to allow the inflow of chloride ions which results in flaccid paralysis and eventual death of parasites exposed to the drug. Pyrantel is a nicotinic acetylcholine (ACh) channel receptor (nAChR) agonist; following receptor binding, the channel opens to allow the influx of cations resulting in a depolarization and excitatory effects on nematode muscle, ultimately leading to spastic paralysis of the worm and death. The target species is dogs.

Bravecto TriUNO chewable tablets is presented in 6 different strengths: 25 mg/0.0625 mg/12.5 mg,

50 mg/0.125 mg/25 mg, 100 mg/0.25 mg/50 mg, 200 mg/0.5 mg/100 mg, 400 mg/1 mg/200 mg and 600 mg/1.5 mg/300 mg of fluralaner/moxidectin/pyrantel (as embonate) respectively. Each strength will be available in packs containing 1, 3 or 6 chewable tablets.

The rapporteur appointed is Rory Breathnach and the co-rapporteur is Andrea Christina Golombiewski.

The dossier has been submitted in line with the requirements for submissions under Article 20 of Regulation (EU) 2019/6 – a combination veterinary medicinal product application.

On 10 October 2024, the CVMP adopted an opinion and CVMP assessment report.

On 22 November 2024, the European Commission adopted a Commission Decision granting the marketing authorisation for Bravecto TriUNO.

Scientific advice

The applicant received scientific advice from the CVMP on user safety and efficacy studies.

The approach taken is consistent with the scientific advice provided to the applicant on the combination fluralaner-moxidectin-pyrantel and its safety profile.

The approach taken in the *in vitro* investigations of geographical differences in susceptibility of ectoparasites to fluralaner is consistent with the scientific advice provided to the applicant.

The approach taken with regards diagnostic methods replacing necropsy in dose confirmation studies for nematodes, presentation of US field study data for *Dirofilaria immitis*, the conduct of a dose confirmation program against ticks limited to the tick species known to be the least sensitive to fluralaner (i.e., *Rhipicephalus sanguineus*), the suitability of conducting the dose confirmation laboratory program against *Rhipicephalus sanguineus* using parasite isolates from the US showing representativeness of US isolates for the European field situation, the suitability of restricting the clinical program to the dose determination/confirmation laboratory program for *Angiostrongylus vasorum* with the aim to reduce the use of live animals for scientific purposes and the suitability of conducting the dose confirmation laboratory program against *C. felis* using parasite isolates from the US showing representativeness of US isolates for the European field situation is consistent with the scientific advice provided to the applicant.

Part 1 - Administrative particulars

Summary of the Pharmacovigilance System Master File

The applicant has provided a summary of the pharmacovigilance system master file which fulfils the requirements of Article 23 of Commission Implementing Regulation (EU) 2021/1281. 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.

Manufacturing authorisations and inspection status

Active substances

Fluralaner

Manufacture and quality control testing of the active substance fluralaner and its intermediate take place outside the EEA. A GMP declaration for the active substance manufacturing sites involved was provided from the Qualified Person (QP) at the EU batch release site. The declaration was based on an onsite audit by the MIAH or a corporate representative of the MIAH.

Moxidectin

Manufacture of the active substance moxidectin takes place outside the EEA. A GMP declaration for the active substance manufacturing site was provided from the Qualified Person (QP) at the EU batch release site. The declaration was based on an onsite audit by the MIAH or a corporate representative of the MIAH.

Pyrantel embonate

Manufacture of the active substance pyrantel embonate takes place outside the EEA. A GMP declaration for the active substance manufacturing site was provided from the Qualified Person (QP) at the EU batch release site. The declaration was based on an onsite audit by the MIAH or a corporate representative of the MIAH.

Finished product

Batch release of the finished product takes place at Intervet GesmbH, Vienna, AT. The site has a manufacturing authorisation issued on 3rd June 2022 by the competent authority of Austria. GMP certification, which confirms the date of the last inspection and shows that the site is authorised for activity indicated above, has been provided.

Overall conclusions on administrative particulars

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

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

Part 2 - Quality

Composition

The finished product is presented as a chewable tablet containing the active substances fluralaner (12.50% w/w), pyrantel (6.25% w/w) and moxidectin (0.03125% w/w). The product contains the active substance pyrantel in the form of pyrantel embonate. The tablets are manufactured in six different sizes for the treatment of dogs in six different body weight ranges between 1.27 kg and 60 kg; for dogs above 60 kg, appropriate combinations of chewable tablets should be used. The tablets are light pink to light brown coloured, mottled, round shaped tablets.

Other ingredients are:

Cellulose microcrystalline, croscarmellose sodium, pigment blend brown, sodium laurilsulfate,

hypromellose, butylhydroxytoluene, poloxamer, magnesium aluminometa silicate, and magnesium carbonate light, , pork liver flavour, croscarmellose sodium, pigment blend brown (as a colourant), colloidal anhydrous silica and magnesium stearate.

The product is presented in aluminium-aluminium foil blisters, in a carton box. Each box contains one, three or six chewable tablets.

Containers and closure system

The primary packaging is an aluminium foil blister sealed with PET aluminium foil lid stock. The material complies with the relevant EU requirements. Certificates of analysis for the aluminium blister foil and the peel-open lidding foil are provided and demonstrate compliance with the specifications.

The choice of an Alu-Alu blister as the container closure system has been validated by stability data. The choice of the container closure system is adequate for the intended use of the product.

Product development

The application for 'Bravecto TriUNO chewable tablets for dogs' has been submitted as a combination veterinary medicinal product application in accordance with Article 20 of Regulation (EU) 2019/6. The tablets are a fixed combination chewable tablet of three active substances; fluralaner, moxidectin and pyrantel (as pyrantel embonate).

The applicant has presented a substantial pharmaceutical development report. The objective of formulation development was to develop a palatable, stable, chewable tablet formulation for once per month administration to dogs for the treatment and prevention of flea and tick infestations, prevention of heartworm disease and treatment and control of gastrointestinal nematode infections.

During the development much consideration was given to the uniformity and stability of moxidectin given its low concentration in the product. The applicant also wished to develop a formulation where all tablet sizes can be manufactured from a common blend.

All excipients are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur. Standards with the exception of the colourant (pigment blend brown) and the flavour (pork liver flavour) for which an in-house quality standard is in place. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 2 of the SPC.

The proposed process design involves the preparation of two separate granulations which are then mixed. The extra-granular excipients which include the flavour ingredient are mixed with the blended granules. The final blend is lubricated and compressed into tablets.

The final tablet formulation includes 0.1% w/w butylhydroxytoluene as an antioxidant within the granule containing moxidectin. Its inclusion as an excipient can be accepted, it is widely used in veterinary pharmaceuticals and the final concentration in the formulation does not pose any risk to the animal or the user.

The development report describes palatability studies which support the use of irradiated pork liver flavour as the palatant in the formulation. This is the same flavour as is used in the authorised 'Bravecto chewable tablets'.

The 'lead formulation' for the chewable tablets is described in the development report and is the same as the proposed finished product formulation.

The potential for active substance polymorphic form changes during manufacture of the finished

product, particularly during the wet granulation steps, was considered during the development work. Data is presented which demonstrates that no change in polymorphic form occurs and hence there is no risk to product quality from potential change in polymorphic form during manufacture.

The product development report addresses the experimental testing conditions for the dissolution test. The test has been adequately described and the choice of dissolution medium, and paddle speeds have been justified. Data to demonstrate the discriminatory power of the method has been provided.

The description of the tablets as chewable has been satisfactorily justified by the applicant.

Description of the manufacturing method

The manufacturing process of the tablets involves manufacture of 2 granules followed by mixing of both and compression. The dossier includes a manufacturing formula for a blend of the commercial batch size. All 6 tablet strengths are compressed from a common blend.

The manufacturing process is considered to be well described in the dossier. The dossier included a stepwise narrative description of the manufacturing process and a manufacturing process flow chart. Factorisation for active substance assay content is carried out for all three actives. No overages are detailed.

In process controls (IPCs) are detailed. The dossier also describes 'critical process parameters' (CPPs) for this manufacturing process with specified value or ranges which have the potential to impact finished product CQAs.

The calculation of the shelf-life of the finished product is in accordance with the "Note for Guidance on Start of the Shelf-life of the Finished dosage form" (EMA/CVMP/453/01).

Process evaluation has been carried out on three full-scale registration batches. The data provided for process evaluation of the three full-scale GMP registration batches are considered to fulfil the requirement to provide process validation data in the dossier.

A process validation scheme for validation of tablet manufacture at the commercial batch size has been provided.

Overall, finished product batch data presented in the dossier demonstrates that the manufacturing process is capable of producing a finished product of the intended quality in a reproducible manner.

Control of starting materials

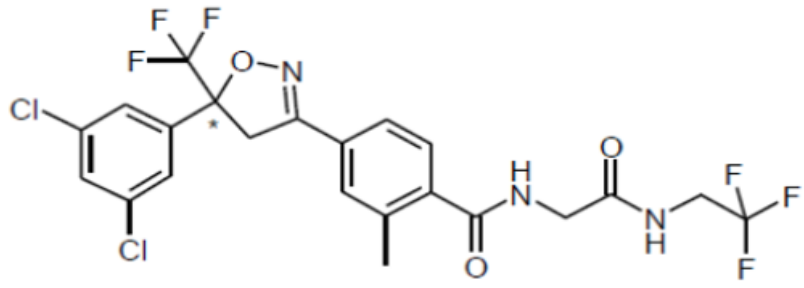
Active substances

The finished product includes three active substances; fluralaner, moxidectin for veterinary use (Ph. Eur.) and pyrantel embonate (Ph. Eur.).

Fluralaner

The chemical name of fluralaner is (±)-4-[5-(3,5-dichlorophenyl)-5-(trifluoromethyl)-4,5-dihydroisoxazol-3-yl]-2-methyl-N-[2-oxo-2-(2,2,2-trifluoroethylamino)ethyl]benzamide (racemic mixture of R and S enantiomer).

Fluralaner has the following structure:



(racemic mixture)

Fluralaner is a non-hygroscopic white to pale yellow solid with a melting point of 176.1 °C. It is insoluble in water and hexane, slightly soluble in toluene, freely soluble in acetone and ethyl acetate, soluble in methanol and acetonitrile and very soluble in *N,N*-dimethylacetamide. Fluralaner exhibits polymorphism. Three polymorphic forms of fluralaner have been described. One of the polymorphic forms of fluralaner was selected for development as no polymorphic changes were observed in the solid state in stability studies using that form. The IR identification test on the specification simultaneously controls the polymorphic form. The dossier notes that the fluralaner proposed for use in the candidate product is the same active substance material as is used in authorised 'Bravecto' products from this MAH.

For the drug substance, the ASMF Procedure is used. Information regarding fluralaner reference standards has been presented.

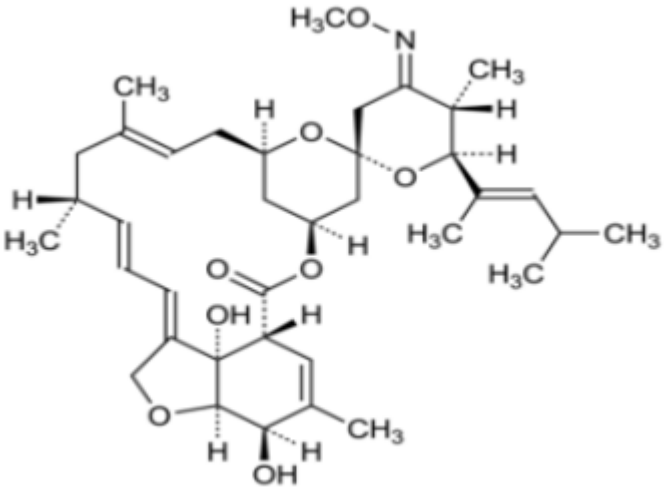
Satisfactory batch analysis data for 3 batches of fluralaner have been provided. The results are within the specifications and consistent from batch to batch.

Stability data has been presented for the active substance and the proposed re-test period can be accepted.

Moxidectin for veterinary use Ph. Eur.

The chemical name of moxidectin is (13a*S*,14*R*,17*R*,17a*R*,2'*R*,4'*E*,42*R*,44*S*,5'*S*,6*E*,6'*S*,9*R*,10*E*,12*E*)-13a,17-Dihydroxy-4'-(methoxyimino)-16,5',7,9-tetramethyl-6'-[(2*E*)-4-methylpent-2-en-1-yl]-13a,14,17,17atetrahydro-12*H*-3-oxa-1(4,3)-[1]benzofurana-4(4,2)-oxanaspiro[cyclododecaphane-6,10,12(13)-trien-46,2'-oxan]-2-one ((6*R*,23*E*,25*S*)-5-*O*-demethyl-28-deoxy-6,28-epoxy-23-(methoxyimino)-25-[(2*E*)-4-methylpent-2-en-1-yl]milbemycin B).

Moxidectin has the following structure:



Moxidectin is a white or pale-yellow, amorphous powder with a melting range of 145 °C to 154 °C that is slightly hygroscopic. It is practically insoluble in water, very soluble in ethanol (96%) and slightly soluble in hexane. The physical form of moxidectin is described as mostly amorphous. This parameter is not critical as the active substance is dissolved during the manufacturing process of the finished product.

There is a monograph for moxidectin for veterinary use in the Ph. Eur. The manufacturer of the active substance has been granted a Certificate of Suitability of the European Pharmacopoeia (CEP) for moxidectin for veterinary use, a copy of which has been provided within the application. The relevant information has been assessed by the EDQM before issuing the Certificate of Suitability. The control tests were carried out to comply with the specifications and test methods of the Ph. Eur. monograph.

The dosage form site specification for moxidectin for veterinary use is in line with the current Ph. Eur. monograph with additional tests for antioxidant and residual solvents. These specifications are consistent with those listed on the CEP. The CEP for moxidectin includes a retest period for the substance of 3 years if stored in a polyethylene bag, in an aluminium bag, placed in an aluminium container.

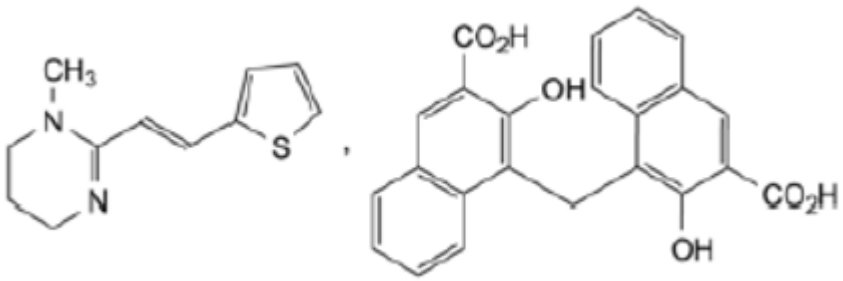
Information regarding moxidectin and butylhydroxytoluene primary standards have been presented. The dossier does not indicate that secondary (working) standards have been established.

Batch analysis data on 2 batches of the active substance have been provided by both the active substance and finished product manufacturers. The results are within the specifications and consistent from batch to batch.

Pyrantel embonate Ph. Eur.

The chemical name of pyrantel embonate is 1-Methyl-2-[(E)-2-(thiophen-2-yl)eth-1-en-1-yl]-1,4,5,6-tetrahydropyrimidine hydrogen 4,4’-methylenebis(3-hydroxynaphthalene-2-carboxylate).

Pyrantel embonate has the following structure:



Pyrantel embonate is an odourless, light yellow to tan crystalline powder with a melting range of 262 °C to 266 °C that is slightly hygroscopic. It is soluble in dimethyl sulfoxide, slightly soluble in dimethylformamide, practically insoluble in water and in methanol. The substance does not exhibit polymorphism.

There is a monograph for pyrantel embonate in the Ph. Eur. The manufacturer of the active substance has been granted a Certificate of Suitability of the European Pharmacopoeia (CEP) for pyrantel embonate, a copy of which has been provided within the application. The relevant information has been assessed by the EDQM before issuing the Certificate of Suitability. The control tests were carried out to comply with the specifications and test methods of the Ph. Eur. monograph.

The CEP for pyrantel embonate includes a retest period for the substance of 3 years if stored in double polyethylene bags placed in fibre drums.

Information regarding pyrantel embonate primary standards have been presented. The dossier does not indicate that secondary (working) standards have been established.

Batch analysis data on 3 batches of the active substance have been provided by both the active substance and finished product manufacturers. The results confirm compliance with the specification including particle size distribution and are consistent from batch to batch.

Excipients

With the exception of the colourant and the flavour (pork liver flavour), all excipients are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur. monographs. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 2 of the SPC.

The dossier indicates that the excipients butylhydroxytoluene and microcrystalline cellulose are to be controlled in line with Ph. Eur. monographs and for additional parameters. The particle size distribution is a functionality-related characteristic of the excipient microcrystalline cellulose and is controlled with specific limits. All of the other compendial product excipients are controlled in line with Ph. Eur. monograph specifications only. No additional tests for relevant functionality-related characteristics have been proposed for the excipients colloidal anhydrous silica, magnesium stearate and croscarmellose sodium, as per respective Ph. Eur. monographs however, acceptable justification has been provided concerning this.

The non-pharmacopoeial excipients used in this product are the colourant and the flavour (pork liver flavour). The dossier includes the qualitative and quantitative composition of the colourant and a

satisfactory control specification. The flavouring ingredient pork liver flavour is the same as that used in the authorised 'Bravecto Chewable Tablets'. The dossier provides details of the qualitative composition of this excipient. Its quantitative composition has not been included in the dossier as the excipient has been used in the same target species and route of administration for a long time. A control specification for pork liver flavour has been provided which includes an IR test for identification. The dossier includes a general description of the manufacturing method of the excipient pork liver flavour and a process flow-chart.

Sample certificates of analysis for each of the product excipients are included in the dossier. The colourant has been declared to comply with Directive 2009/35/EC and Commission Regulation (EU) No. 231/2012, in line with the requirements of Annex II to Regulation 2019/6.

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

The product constituents pigment blend brown, moxidectin and pork liver flavour are derived from material of human or animal origin but do not present any BSE/TSE risk. Pigment blend brown contains lactose monohydrate of bovine origin which is produced from milk obtained from healthy animals in the same condition as those used to collect milk for human consumption. In the manufacturing process of moxidectin, lactose and casein enzymatic hydrolysate are used as a raw material and reagent, respectively. These materials are declared to be of 'other animal origin' and therefore do not present a TSE risk. Pork liver flavour contains enzyme-hydrolysed pork liver. As pigs are not a BSE/TSE-relevant species, there is no risk of BSE/TSE transmission to the target animal.

Control tests on the finished product

The specifications proposed at release are appropriate to control the quality of the finished product.

The finished product specification includes tests for appearance, active substances identification, uniformity of dosage units, active substances assay, degradation products of each active substance, butylhydroxytoluene content, residual solvent content, water content, dissolution and microbial quality. Tests for average tablet hardness and average tablet weight are included on the release specification.

An elemental impurities risk assessment has been conducted which found that the product complies with ICH Q3D requirements for elemental impurities. A summary report of the elemental impurities risk management is included in the dossier, in line with the requirements of the 'Reflection paper on risk management requirements for elemental impurities in veterinary medicinal products' EMA/CVMP/QWP/15364/2018.

The analytical methods used for assay, related substances, content uniformity and dissolution have been adequately described and in the main, are appropriately validated in accordance with the VICH GL1: *Validation of analytical procedures: definition and terminology* and VICH GL2: *Validation of analytical procedures: methodology*.

Validation is also presented for the method for residual solvent content, for water determination and for the microbial enumeration test.

Information regarding the reference standards for each of the active substances and for butylhydroxytoluene has been presented.

Batch analysis results are provided for batches tested for according to the proposed release specification and all results met the proposed specification.

Stability

The specification parameters proposed at the end of shelf-life are appropriate to control the quality of the finished product.

The batches were placed on stability under VICH long term (30 °C/65%RH) and accelerated (40 °C/75%RH) storage conditions, with batches from the first campaign also placed on stability at 25 °C/60%RH. The parameters monitored in the stability studies are description, active substances assay, related substances (from the 3 actives), butylhydroxytoluene content, water, dissolution and microbial limits. The shelf-life proposed for all tablet sizes is 2 years with the storage precaution 'Store in the original package in order to protect from light'. The proposed 2-year shelf-life is considered acceptable.

The dossier includes a photostability study and a temperature cycling study. The photostability studies were carried out in accordance with VICH GL5, Option 2 and demonstrate that under direct exposure photolytic stress conditions, there is a reduction in butylhydroxytoluene assay content. The blister packaging was demonstrated to protect the tablets and prevents any such loss in butylhydroxytoluene content. This data indicates that the finished product is light sensitive and as such, a warning has been included in Section 5.3 of the SPC i.e. 'Store in the original package in order to protect from light'. The data from the temperature cycling study demonstrates that product quality is not adversely impacted by temperature excursions.

Overall conclusions on quality

The finished product is presented as a chewable tablet containing the active substances fluralaner (12.50% w/w), pyrantel (6.25% w/w) and moxidectin (0.03125% w/w). The product contains the active substance pyrantel in the form of pyrantel embonate. The tablets are manufactured in six different sizes for the treatment of dogs in six different body weight ranges between 1.27 kg and 60 kg. The tablets are light pink to light brown coloured, mottled, round shaped tablets.

The product is presented in aluminium-aluminium foil blisters, in a carton box. Each box contains one, three or six chewable tablets.

The objective of formulation development was to develop a palatable, stable, chewable tablet formulation for once per month administration to dogs. All tablet sizes are to be compressed from a common blend. The main challenges in formulation development are uniformity and stability of moxidectin given its low concentration in the product (0.03125% w/w) and its sensitivity to degradation.

The development report describes various formulation optimisation studies.

The manufacturing process was developed using technical batches and then using batches of pilot scale. A scale-up program was then established at the proposed finished product manufacturing site. The data indicates that the final blend has satisfactory uniformity with respect to all three active substances and has comparable particle size distribution between different batches, a blend which has acceptable flowability and the compressed tablets have acceptable content uniformity with respect to each active substance.

The product development report addresses the experimental testing conditions for the dissolution test.

The tablets are described as chewable and this has been satisfactorily justified in the dossier.

Process evaluation has been carried out on three full-scale GMP registration batches and data has

been presented. Information on the control of starting materials has been provided. The active substances moxidectin and pyrantel embonate are monographed in the Ph. Eur. and are to be sourced for this product from suppliers who are holders of Ph. Eur. Certificates of Suitability. The control specifications for these active substances include the test of the Ph. Eur. monographs with additional specifications. In the case of moxidectin, additional tests for the antioxidant and residual solvents are included on the dosage form manufacturer's specification. In the case of pyrantel embonate, the dosage form manufacturer's specification includes an additional test for particle size distribution. The active substance fluralaner is non-compendial material. The supporting data for the active substance is provided in the form of an ASMF. The version of the ASMF supplied with this application has already been approved in relation to authorised Bravecto formulations. The dosage form manufacturer has provided a specification for fluralaner.

With the exception of the colourant (pigment blend brown) and the flavour (pork liver flavour), all excipients are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur. monographs. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 2 of the SPC.

Satisfactory data and specifications have been provided for the non-compendial excipients pigment blend brown and pork liver flavour.

The dossier includes the required supporting data for the finished product container closure system including sample IR spectra for the blister foils. The choice of Alu-Alu blisters for this dosage form is considered to be appropriate.

Finished product specifications for release and shelf-life have been provided. The finished product specification at time of release controls those parameters appropriate for the dosage form. The analytical methods are well described and have been validated in line with VICH guidelines. Data to demonstrate that the test methods for active substance assay and related substances determinations are stability indicating, has been provided. Batch data for pilot scale batches and full-scale batches has been provided. All results are within the specifications as proposed for release and are comparable between batches. The finished product shelf-life specification is the same as that proposed for release however, tests for uniformity of dosage units and ethanol have been removed. Differences between the release and shelf-life specification have been appropriately justified.

In terms of dosage form stability, the data presented meets the proposed shelf-life specifications.

The 3 full-scale batches were placed on stability under VICH long term (30 °C/65%RH) and accelerated (40 °C/75%RH) storage conditions. A shelf-life of 2 years for the veterinary medicinal product as packaged for sale with the storage precaution 'Store in the original package in order to protect from light'. The shelf-life is considered acceptable.

The dossier includes a photostability study and a temperature cycling. It is concluded that the product is photosensitive and its quality is not adversely impacted by temperature excursions.

Information on the development, manufacture and control of the active substance and the 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.

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SPC. Physicochemical aspects relevant to the performance of the product have been investigated and are controlled in a satisfactory way.

Part 3 – Safety documentation

Bravecto TriUNO is a new fixed combination for dogs containing fluralaner, moxidectin and pyrantel embonate indicated for the treatment of mixed parasitic infestations by ticks or fleas, gastrointestinal nematodes, lungworm and/or heartworm. Fluralaner is an acaricide and insecticide belonging to the isoxazoline group. Moxidectin belongs to the milbemycin group of macrocyclic lactones and has parasiticidal activity against a range of internal and external parasites including various nematode species, while pyrantel embonate is an anthelmintic of the tetrahydropyrimidine class.

A full safety file in accordance with Article 8 of Regulation (EU) 2019/6 has been provided. The studies provided for fluralaner have previously been evaluated by CVMP with the original marketing authorisation applications for the veterinary medicinal products "Bravecto chewable tablets for dogs" and "Bravecto spot-on solution for dogs/cats" as well as with the application for the establishment of Maximum Residue Limits (MRL) for fluralaner in chickens (i.e. EMEA/V/C/002526, EMEA/V/C/2526/X/0005 and EMEA/V/MRL/004380/FULL/0001).

Moxidectin and pyrantel are well-established substances that have been widely and safely used in veterinary medicine for more than 10 years. Reference to published 'MRL summary report' data concerning the toxicity of moxidectin and pyrantel as evaluated by the Joint FAO/WHO Expert Committee on Food Additives (JECFA, 1995) and/or CVMP has been made. In addition, the studies provided for moxidectin were assessed by CVMP in the context of the authorisation of "Bravecto Plus spot-on solution for cats" (EMEA/V/C/004440). Additionally, new proprietary subacute and sub-chronic dermal repeat-dose toxicity studies with moxidectin in rats were conducted by the applicant and are described in more detail in the respective section.

Safety tests

Pharmacology

Pharmacodynamics

See part 4.

Pharmacokinetics

See part 4.

Note: No pharmacokinetic interaction was observed between the three active ingredients; pharmacokinetic values for fluralaner, moxidectin and pyrantel, when given in combination, are comparable to those obtained when the active substances are administered separately.

Toxicology

The active substance fluralaner was previously assessed by the CVMP in the context of the establishment of MRLs (for chickens) and authorisation of veterinary medicinal products (Bravecto tablets (EMEA/V/C/002526) and spot-on solutions (EMEA/V/C/2526/X/0005), Exzolt (EMEA/V/C/004344) and Bravecto Plus spot-on (EMEA/V/C/004440)). The active substance moxidectin has a well-established use and has also been previously assessed by CVMP in the context of an MRL evaluation (EMEA, 1997), as well as by JECFA. In addition, the active substance moxidectin was assessed by CVMP in the context of the authorisation of Bravecto Plus spot-on solution for cats (EMEA/V/C/004440). The active substance pyrantel embonate was previously

assessed by the CVMP in the context of the establishment of MRLs (EMEA/MRL/491/98-Final). Key findings of the toxicity studies previously evaluated by the CVMP are summarised below:

Single-dose toxicity

Fluralaner

An acute oral GLP toxicity study using the active substance was performed in rats in accordance with OECD Test Guideline 423. All animals survived until the end of the study period. No adverse effects were observed in this study, except for slightly ruffled fur in all animals. An LD_{50} of > 2000 mg/kg bw could be derived from this study.

An acute dermal toxicity study using the active substance (limit test: 2000 mg/kg bw) was performed in rats in accordance with OECD Test Guideline 402. No adverse effects were observed in this study, except for some local effects in some of the animals (erythema, scaling and scabs). An LD_{50} of > 2000 mg/kg bw could be derived from this study.

In conclusion, fluralaner is of low acute oral and dermal toxicity (LD_{50} > 2000 mg/kg bw; limit dose).

Moxidectin

An acute oral toxicity study with moxidectin resulted in an LD_{50} of 106 mg/kg bw for rats and 84 mg/kg bw for mice (JECFA, 1995).

An acute dermal toxicity study in rabbits with moxidectin resulted in an LD_{50} of > 2000 mg/kg bw (JECFA, 1995).

Pyrantel embonate

The acute oral toxicity of pyrantel embonate is low with LD_{50} values in mouse, rat and dog >2000 mg/kg bw (EMEA/MRL/491/98-Final).

Repeat-dose toxicity

Fluralaner

The effects of repeated doses of fluralaner have been investigated in repeat dose toxicity studies. The presented studies were previously assessed by CVMP during the registration procedures of Bravecto tablets/spot-on, Exzolt and Bravecto Plus spot-on.

Oral

Repeat dose oral toxicity was studied in rats (studies with durations of 2, 4 and 13 weeks) and dogs (studies with durations of 4, 13 and 52 weeks).

In 2-week and 4-week toxicity studies, rats were given fluralaner by oral gavage at doses of 0, 30, 60, and 600 mg/kg bw per day. The main target organ in the repeat dose toxicity studies was the liver, which is the main organ for elimination of fluralaner. Effects (increased organ weight, hepatocellular fatty change, effects in related blood parameters) were observed at all dose levels, though considered mild at the lower doses. Decreased thymus and increased adrenal weight was observed at the highest dose. A no-observed adverse effect level (NOAEL) of 60 mg/kg bw per day was established by CVMP.

A 13-week oral toxicity study in rats, when fluralaner was administered at 0, 20, 40 and 400 mg/kg bw per day, confirmed the effects on liver. In addition, at the dose of 400 mg/kg bw per day effects on thymus and adrenal weight and microscopic changes in lung and thymus were observed. As the effects were mild at lower doses, a NOAEL of 40 mg/kg bw per day was

established by CVMP.

Two 4-week toxicity studies in dogs were provided with respective oral dose (by capsule) levels of 0, 100, 250, 750 mg/kg bw per day and 0, 20, 40, 100 mg/kg bw per day. Reductions in cholesterol, phospholipid and triglyceride levels were observed at all dose levels, in both sexes and at different time points. Although no histopathological changes of the liver were observed, it could not be concluded that the observed effects should be considered non-adverse. A lowest-observed-adverse-effect level (LOAEL) of 20 mg/kg bw per day was therefore established by CVMP.

In a 13-week oral toxicity study, dogs were given fluralaner by capsule at doses of 0, 2, 4 and 8 mg/kg bw per day. Reductions in cholesterol and phospholipids were observed at 4 and 8 mg/kg bw per day in both males and females. In addition, reductions of triglycerides concentration were observed at 4 and 8 mg/kg bw per day in males. Based on this study, the CVMP concluded on a no-observed effect level (NOEL) of 2 mg/kg bw per day over 13 weeks.

Similar results were observed in the 52-week study in dogs, when dosed 0, 1, 2, or 4 mg/kg bw per day. Reductions in cholesterol and phospholipids were observed in males at 2 and 4 mg/kg bw per day, and in females at 4 mg/kg bw per day. Reduction of triglycerides concentration were observed at 2 mg/kg bw per day in males and in females at 4 mg/kg bw per day and the derived NOEL was set at 1 mg/kg bw per day.

Dermal

The potential subacute effects of fluralaner were investigated in one 2-week (dose range finding) and two 4-week dermal (6 hour semi-occlusive) toxicity studies in rats. From the first of those 4-week studies, fluralaner was dosed at 0, 100, 200 or 1000 mg/kg bw per day, no NOAEL could be established. Treatment related effects were observed at all doses and included: fatty change in the liver, effects on serum liver enzymes, triglyceride, albumin and globulin and moderately increased liver weights. In addition, at all dose levels spleen weights were increased in males, though not correlated with histopathological findings. A further 4-week study was conducted using the doses 0, 25, 50 or 100 mg fluralaner/kg bw per day. At 100 mg/kg bw per day, microvesicular fatty change, periportal or diffuse, was observed in the liver of three males and three females. However, there was no other indicator of liver injury. No effects were observed at the other doses. Taking into account all three studies (the effects are considered to be mild and comparable to the effects observed in the oral studies) and taking account of the oral NOAEL from the rat studies (it is not conceivable that the dermal NOAEL will be lower than the oral NOAEL), CVMP decided that a NOAEL of 100 mg/kg bw per day was appropriate when considering repeated dose toxicity. The studies were performed using fluralaner in a carboxymethyl-cellulose aqueous solution.

A 90-day dermal toxicity study was performed in rat, administered doses of 0, 25, 50 or 500 mg/kg bw per day. At 500 mg/kg bw/day liver effects were observed, which were similar to the effects observed in the 2-, 4- and 13- week oral studies as well as the 2- and 4-week dermal studies. At the highest dose also alveolar histiocytosis was observed in females, in some animals accompanied by (multi)focal interstitial lobular inflammation and intra alveolar amorphous material. Therefore, a dermal NOAEL of 50 mg/kg bw per day was derived from this study by CVMP.

The potential systemic effects following subchronic and chronic exposure (oral and dermal) have been comprehensively investigated in the rat. The studies conducted meet with guideline requirements (GLP and relevant OECD Test Guidelines). The liver appears to be the most sensitive organ for effects (increased organ weight, hepato-cellular fatty change, effects in related blood parameters). These effects were observed at dose levels above 20 mg/kg bw/day in the oral studies and above 100 mg/kg bw/day in the dermal studies.

Subchronic and chronic (up to 52 weeks) effects following oral exposure in dogs were also comprehensively investigated. Reductions in cholesterol, phospholipids and triglycerides were consistently observed at dose levels above 1 mg/kg bw/day in the oral studies, although no histopathological changes of the liver were reported. It can be concluded that the dog is more sensitive to the effects of fluralaner than the rat and that systemic exposure of fluralaner is greater in dogs than in rats.

Moxidectin

Study summaries from the published literature have been provided reviewing the effects of repeated doses of moxidectin on laboratory animals. The oral studies with moxidectin have previously been evaluated by JECFA (1995) and by CVMP during the registration procedure of Bravecto Plus spot-on. Two new repeated dose dermal toxicity studies for moxidectin have been provided.

Oral

A 28-day mouse oral (dietary) toxicity study with moxidectin resulted in a NOEL of 6.9 mg/kg bw per day, based on tremors, hypersensitivity to touch and urine-stained fur at the next higher levels. No NOEL could be derived in a 28-day rat oral (dietary) toxicity study with moxidectin, as hypersensitivity to touch was observed at the lowest dose level of 12 mg/kg bw per day.

A 13-week rat oral (dietary) toxicity study with moxidectin resulted in a NOEL of 3.9 mg/kg bw per day, based on hypersensitivity to touch, depressed bodyweight and increased adrenals weights in females and increased testes weights as observed at the next level of 7.9 mg/kg bw per day.

A 4-week dog oral (dietary) toxicity study with moxidectin resulted in a NOEL of 0.5 mg/kg bw per day, mainly based on effects on the nervous system observed at the next higher dose level.

A 13-week dog oral (dietary) toxicity study with moxidectin resulted in a NOEL of 0.3 mg/kg bw per day, based on dose-dependent reductions in absolute body weights and food consumption at the next level of 0.9 mg/kg bw. It is noted that this NOEL was selected by JECFA and CVMP for establishing the ADI (JECFA, 1995; EMEA/MRL/139/96-final).

A 52-week dog oral (dietary) toxicity study with moxidectin resulted in a NOEL of 1.15 mg/kg bw per day, the highest dose-level tested.

Dermal

The potential subacute effects of moxidectin were investigated in one 4-week dermal and one 13-week dermal (6 hour semi-occlusive) toxicity studies in rats.

In a 4-week dermal toxicity study in rats, moxidectin was administered at dose levels of 0, 10, 70 or 500 mg/kg bw/day. Treatment related effects were observed at 70 mg/kg bw/day and included underactivity and effects on sensory reactivity and motor activity. A NOAEL of 10 mg/kg bw/day was derived based on these studies.

In a 13-week dermal toxicity study in rats, moxidectin was administered at dose levels of 0, 2, 20 or 160 mg/kg bw/day. A NOAEL could not be established since histopathological findings identified retinal atrophy at all dose levels and unsteady gait in one low dose animal, resulting in a dermal LOAEL of 2 mg/kg bw.

Pyrantel embonate

The repeat dose oral toxicity studies with pyrantel were previously assessed by CVMP during the MRL procedure for pyrantel embonate (EMEA/MRL/491/98-Final).

Oral

In a 13-week toxicity study in rats, rats were given pyrantel tartrate at doses equivalent to 0.012, 0.12, 1.2 and 12 mg base/kg bw/day. Although variations in the biochemical parameters were seen (CO_2 content and alkaline phosphatase), these were without toxicological significance and no adverse effects were reported up to 12 mg/kg bw/day pyrantel base administered as its tartrate salt.

Rats were given tartrate salt by oral route at doses equivalent to 0, 3, 30 and 115 mg base/kg bw/day for up to 93 weeks. A NOEL of 3 mg/kg bw/day was established based on reductions in bodyweight gain, changes in haematology values indicative of anaemia and changes in some organ weights.

In a 13-week dog oral toxicity study, dogs received capsules containing pyrantel embonate at doses equivalent to 35, 105 and 210 mg pyrantel base/kg bw, five days per week. An oral NOEL of 35 mg/kg bw/day was established based on increased serum aspartate aminotransferase and serum alanine aminotransferase values at higher dose levels.

In a 13-week toxicity study pyrantel tartrate was administered orally to dogs (4 animals/sex) in capsules at doses equivalent to 0.012, 0.12, 1.2 and 12 mg pyrantel base/kg bw/day. Diarrhoea was reported in the high dose group. No changes attributable to the drug treatment were reported for doses up to 12 mg/kg bw/day pyrantel base administered as its tartrate salt.

In a 2-year oral (dietary) toxicity study, dogs received pyrantel tartrate at doses equivalent to 0, 3, 15, 30 mg base/kg bw/day, 5 days per week. A NOEL of 3 mg/kg bw/day based on increased liver weights and serum alanine aminotransferase values at higher doses was derived.

Dermal

No dermal studies investigating the repeated administration of pyrantel embonate were conducted.

Tolerance in the target species

See Part 4.

Reproductive toxicity, including developmental toxicity

The presented studies for fluralaner were previously assessed by CVMP during the registration procedures of Bravecto tablets/spot-on, Exzolt and Bravecto Plus spot-on. Similarly, the studies for moxidectin have previously been evaluated by CVMP during the registration procedure of Bravecto Plus spot-on. The studies with pyrantel were previously assessed by CVMP during the MRL procedure for pyrantel embonate (EMEA/MRL/491/98-Final).

Study of the effect on reproduction

Fluralaner

In the one-generation study, rats were given fluralaner at a dose level of 0, 50, 100 or 500 mg/kg bw per day. Liver, thymus, lung and adrenals appear to be affected in parents at the lowest dose of 50 mg/kg bw per day, resulting in a LOAEL of 50 mg/kg bw per day. The effects are consistent with the adverse effects observed in the repeated dose studies. The reproduction NOEL was set at 100 mg/kg bw per day, based on reduced litter size due to reduced implantation rate and increased post-implantation loss at the higher dose of 500 mg/kg bw per day. Statistically significant reductions in thymus weight, and lymphoid atrophy in the thymus was observed in pups at all doses, showing a clear dose response and resulting in a LOAEL of 50 mg/kg bw per day as

concluded by CVMP.

In the two-generation study, rats were given fluralaner at a dose level of 0, 8, 50 or 500 mg/kg bw per day. In the parental and/or first generation (F1) generation, peribronchial inflammatory lesions in the lungs and increased hypertrophy of the adrenal cortex and atrophy/involution of the thymus were observed at all dose levels, resulting in a LOAEL of 8 mg/kg bw per day for parental toxicity, though the effects are considered marginal at the lowest dose. The reproduction NOEL was set at 50 mg/kg bw per day, based on higher post-implantation, post-natal and breeding loss at the higher dose of 500 mg/kg bw per day. The pup NOEL was set by CVMP at 50 mg/kg bw per day based on reduced body weight, clinical signs, pathological findings, and delayed physical and sexual development at 500 mg/kg bw per day.

Moxidectin

In a one-generation rat study with moxidectin, no parental effects were observed at dose levels up to and including 3.9 mg/kg bw per day. A pup NOEL of 0.4 mg/kg bw per day was derived based on reduced pup weight and reduced survival rate in the F1b pups.

In a three-generation rat toxicity study with moxidectin, significant reductions in pup survival indices were observed during day 0-21 for the F1a litters, and during day 0-4 for the F2a litters at the highest dose level tested (0.83 mg/kg bw per day). A pup NOEL of 0.41 mg/kg bw per day could be derived. Slight reductions in weight were observed in males during the pre-mating (F2), mating and post-mating (F1 and F2), resulting in a parental NOEL of 0.41 mg/kg bw per day. No factual reproduction effects were observed.

Pyrantel embonate

In a reproduction and lactation study in rats, animals were administered pyrantel embonate at dietary doses equivalent to 0, 9, and 90 mg pyrantel base/kg bw/day from Day 14 prior to mating, throughout pregnancy, and until all pups had been weaned. There were no significant differences between treated rats and control rats on fertility, gestation, viability or lactation indices up to the highest dose of 90 mg/kg bw/day (EMEA/MRL/491/98-Final).

Study of developmental toxicity

Fluralaner

Oral

Developmental toxicity was studied in the rat at doses (oral gavage) of 0, 100, 300 or 1000 mg fluralaner/kg bw per day. Food consumption was significantly reduced in the two higher dose groups; in the highest dose group, body weight and body weight gain were also reduced. In the foetuses of rats in the two highest dose groups, a higher incidence of dilated renal pelvis/ureter and supernumerary ribs were observed at both foetus and litter level. The NOEL for toxicity to maternal and foetal organisms was set to 100 mg/kg bw per day by CVMP.

Developmental toxicity was studied in rabbit at doses (oral gavage) of 0, 50, 250 or 1000 mg fluralaner/ kg bw/day. The NOAEL for maternal toxicity was 50 mg/kg bw per day, based on reduction in food consumption at 250 mg/kg bw per day. No NOAEL for foetal toxicity could be established, the LOAEL was 50 mg/kg bw per day based on adverse embryo-foetal developmental effects observed at the lowest dose of 50 mg/kg bw per day. A complementary prenatal developmental toxicity study using lower oral doses of 10, 25 and 250 mg/kg bw per day was therefore conducted. Fatty changes of the liver and the related changes in blood biochemistry were observed at all doses in dams, however considered mild at the lowest dose level, resulting in a

NOAEL of 10 mg/kg bw per day. Based on the increase in fusions in cervical vertebra 2 at 25 mg/kg bw per day, the developmental NOEL was set to 10 mg/kg bw per day by CVMP.

Dermal

Developmental toxicity was studied in rabbits administered fluralaner suspended in 0.5% (w/v) carboxymethylcellulose aqueous solution containing 0.1% (v/v) polysorbate 80 at doses of 0, 50, 100 and 1000 mg/kg bw per day. A maternal dermal NOAEL of 1000 mg/kg bw per day was set (the highest dose tested in the pivotal study). However, it was noted that liver (blood biochemistry), which appeared the most sensitive target organ, and the basis for the maternal NOAEL in the rabbit oral study, was not investigated in the dermal study. Based on adverse effects observed at 1000 mg/kg bw per day including external and visceral abnormalities and skeletal abnormalities such as fusion of cervical vertebra 2 and sternebrae, decreased ossification of the humerus and femur of fore- and hindlimbs, the NOEL for foetal toxicity was set to 100 mg/kg bw per day by CVMP.

Moxidectin

The potential effects of moxidectin on development have been investigated in oral prenatal developmental toxicity studies, one in rats and one in rabbits and were previously assessed by CVMP.

A rat prenatal developmental toxicity study with moxidectin resulted in an oral NOEL of 2.5 mg/kg bw per day, based increases in the total number of foetuses with abnormalities (increased incidences of cleft palate and wavy or incompletely ossified ribs) at 5, 10 and 12 mg/kg bw per day. It is noted that these effects were observed in the presence of maternal toxicity (reduced bw and food consumption).

In a rabbit developmental toxicity study with moxidectin, no embryo-foetal developmental effects were observed (highest dose tested: 10 mg/kg bw per day orally). A maternal NOEL of 1 mg/kg bw per day was derived, based on reduced body weight gain.

Pyrantel embonate

No evidence of teratogenicity, foetotoxicity or maternal toxicity was observed in rats after oral administration of pyrantel embonate at daily oral doses equivalent to 0, 9, and 90 mg pyrantel base/kg bw/day. No statistical and no dose-related incidence of malformations was observed when compared to the control group (EMEA/MRL/491/98-Final).

Pyrantel embonate was administered to rabbits at doses equivalent to 0, 9, and 90 mg pyrantel base/kg bw/day from GD 7 to 17. An increase in the incidence of resorptions (8.5% and 12.6% versus 2.5% for controls was observed. No conclusions could be drawn with regard to a NOEL for maternal toxicity. No adverse effects of toxicological significance were reported for foetuses up to the highest dose of 90 mg/kg bw/day (EMEA/MRL/491/98-Final).

Genotoxicity

Fluralaner

The potential genotoxic effects of fluralaner have been investigated in three *in vitro* tests (Ames-test, mouse lymphoma thymidine kinase locus assay, chromosomal aberration test in human lymphocytes *in vitro*) and one *in vivo* test (micronucleus assay in bone marrow cells of the mouse) on genotoxicity. The results of all four tests were negative. It was previously concluded by CVMP and JECFA that fluralaner does not have genotoxic potential.

Moxidectin

The potential mutagenicity of moxidectin has been investigated in four *in vitro* tests and one *in vivo* test. The studies have previously been evaluated by JECFA (1995) and during the registration procedure of Bravecto Plus spot-on. Moxidectin was reported to be non-mutagenic in the Ames test, did not induce mutations in a bacterial/microsome or a forward mutation assay, did not induce unscheduled DNA synthesis and was negative in a chromosome aberration test. Moxidectin is considered not to be genotoxic, as previously concluded by both CVMP and JECFA.

Pyrantel embonate

Genotoxicity studies with pyrantel were previously assessed by CVMP during the MRL procedure for pyrantel embonate (EMEA/MRL/491/98-Final). As pyrantel has the same metabolic pathway as its related analogue morantel leading to the same major metabolites, reference to the CVMP MRL assessment of morantel tartrate and citrate has also been made.

Pyrantel tartrate gave negative results in the Salmonella microsomal assay (TA98, TA100, TA1535, TA1537, TA1538) in the absence and presence of metabolic activation at concentrations of 0.75 to 7500 µg/mL.

One *in vitro* mouse lymphoma assay conducted at concentrations in the range of 390 to 2,205 µg morantel base/mL, given as the tartrate salt, and an *in vivo* mouse micronucleus test with morantel citrate administered orally at doses of 2.8, 25.5 and 50 mg morantel base/kg bw were regarded as adequate for the assessment of genotoxicity by EMEA (2005). Results showed morantel to be non-mutagenic.

CVMP concluded that pyrantel does not have mutagenic potential.

Carcinogenicity

Fluralaner

Studies on fluralaner for carcinogenic potential were not submitted. This is justified by the negative results in all genotoxicity assays and the absence of pre-neoplastic lesions in repeated dose toxicity studies (tested up to 365 days, i.e. there is no evidence for a carcinogenic potential of fluralaner, at the relevant exposure levels).

Moxidectin

The studies with moxidectin have previously been evaluated by JECFA (1995) and by CVMP during the registration procedure of Bravecto Plus spot-on and in the context of the establishment of maximum residue limits for moxidectin (EMEA, 1997).

In a chronic dietary toxicity and oncogenicity study moxidectin was administered to mice at doses equivalent to 0, 2.5, 5.1 or 12 mg/kg bw/day for 2 years. After 9 weeks on the study, the highest dose was reduced to 7.9 mg/kg bw/day because of increased mortality in this group. For rats, moxidectin was administered during a chronic dietary toxicity and oncogenicity study for 2 years at doses equivalent to 0, 0.8, 3.2 or 9.8 mg/kg bw/day. The highest dose was reduced to 5.1 mg/kg bw/day after 8 weeks because of increased mortality in this group. No increased incidence of any tumour type was observed in either study. CVMP concluded that moxidectin did not show carcinogenic potential.

Pyrantel embonate

Carcinogenicity studies were not conducted with pyrantel embonate. CVMP concluded in the context of the establishment of maximum residue limits for pyrantel embonate that the available long-term

feeding studies with pyrantel tartrate in rats (93 weeks) and dogs (2 years) were not acceptable to assess the carcinogenic potential of pyrantel. However, based on the absence of mutagenicity, confirmatory carcinogenicity studies are not considered necessary.

Other requirements

Skin irritation

Fluralaner

The presented study was assessed by the CVMP during the registration procedure of Bravecto tablets and spot-on and Bravecto Plus spot-on. An acute dermal irritation/corrosion study using the active substance fluralaner was performed in rabbits in accordance with OECD guideline 404. No signs of skin irritation or corrosion occurred during the evaluation period. The CVMP concluded that fluralaner is non-irritating to the rabbit skin.

Moxidectin

This study has been assessed by JECFA (1995) and reviewed by the CVMP during the registration procedure of Bravecto Plus spot-on. A dermal irritation study in rabbits showed mild signs of skin irritation up to 72 hours. The CVMP concluded that moxidectin may be slightly irritating to the skin.

Pyrantel embonate

No dermal irritation studies conducted with pyrantel embonate were provided. It is reported that pyrantel embonate can cause skin irritation (ECHA, 2022).

Eye irritation

Fluralaner

The presented study was assessed during the registration procedure of Bravecto tablets and spot-on and Bravecto Plus spot-on. An eye irritation study using the active substance was performed in rabbits in accordance with OECD guideline 405. Slight reddening of both the conjunctiva and the sclera and slight ocular discharge were observed in all three animals at the 1-hour observation. These effects were reversible and were no longer evident 24 hours after treatment. No other effects were observed. The CVMP concluded that fluralaner was non-irritating to the rabbit eye.

Moxidectin

This study has been assessed by JECFA (1995) and reviewed by the CVMP during the registration procedure of Bravecto Plus spot-on. An eye irritation study in rabbits demonstrated moderate signs of eye irritation, though effects were no longer present at 48h and 72h after treatment. The CVMP concluded that moxidectin may be slightly irritating to the eye.

Pyrantel embonate

No eye irritation studies conducted with pyrantel embonate were provided. It is reported that pyrantel embonate can cause eye irritation (ECHA, 2022).

Sensitisation and other effects

Fluralaner

The presented study was assessed by the CVMP during the registration procedure of Bravecto tablets and spot-on and Bravecto Plus spot-on. A skin sensitisation test using the active substance fluralaner was performed in guinea pigs in accordance with OECD guideline 406. The CVMP concluded that fluralaner did not have sensitising potential when tested in the guinea pig

maximisation test of Magnusson and Kligman.

Thymus atrophy was observed in several studies but was mostly associated with high doses and/or not accompanied by significant or consistent adverse effects on other organs of the immune system or haematology. The repeated dose toxicity studies were considered sufficient to cover potential effects on the immune system.

Moxidectin

This study has been assessed by JECFA (1995) and reviewed by the CVMP during the registration procedure of Bravecto Plus spot-on. A skin sensitisation study in guinea pigs was conducted with no evidence of skin sensitisation. The CVMP concluded that moxidectin is non-sensitising to the skin.

Pyrantel embonate

It is reported that pyrantel embonate can cause skin sensitisation (ECHA, 2022).

Neurotoxicity

Fluralaner

No effects on the nervous system have been reported for fluralaner in the toxicity tests provided. Absence of additional neurotoxicity studies is therefore justified.

Moxidectin

No specific studies on the neurotoxicity of moxidectin were provided. In the repeat dose toxicity studies presented, neurological signs (tremors) were observed in mice administered moxidectin at 7.9 mg/kg bw/day for two years and in rats transient hypersensitivity to touch was observed at 7.9 mg/kg bw/day in a 13-week dietary study. The NOEL for rats was 3.9 mg/kg bw/day. In dogs administered moxidectin in the diet for 90 days neurological signs were observed at 1.6 mg/kg bw/day, with no neurological signs in the 0.9 mg/kg bw/day group.

Pyrantel embonate

No effects on the nervous system have been observed for pyrantel embonate in the toxicity tests provided. Absence of additional neurotoxicity studies is therefore justified.

Observations in humans

Fluralaner:

Fluralaner has been developed exclusively for veterinary use. No study data are available on health effects of fluralaner in humans. Isoxazoline anti-parasitics in general are currently not used in human medicine. However, post-authorisation safety data have shown that sensitivity reactions are observed in humans when exposed to fluralaner-containing products (e.g., Bravecto tablets and spot on), even though studies to investigate the skin sensitisation potential of fluralaner and other Bravecto products (tablets and spot on) were negative. Indeed, based on the post-authorisation safety data, the product information for Bravecto Spot-on and Bravecto Tablets has been updated with the user safety warning: "*Hypersensitivity reactions in humans have been reported*". A similar warning is included for this product.

Moxidectin:

Based on data from the published literature provided by the applicant, it appears that moxidectin is well tolerated by human subjects when administered at doses in the range of 3 – 36 mg/kg bw. Moxidectin is generally safe and well tolerated, with a slightly higher incidence of transient, mild, and

moderate central nervous system AEs as the dose increased compared to the placebo. No severe adverse effects were noted, with the main findings being mild-moderate transient events such as headache.

Pyrantel:

Pyrantel has been used in human medicine for more than 40 years. It is normally administered orally at doses up to 20 mg pyrantel embonate/kg bw/day for 1 to 3 days. When overdosed, the reported adverse effects are disturbances of the gastrointestinal tract, central nervous system effects, and skin reactions as well as elevation of serum aspartate aminotransferase and alanine aminotransferase in a small number (1.8%) of patients. These observations reflect the results of animal studies as described in the 1998 CVMP Pyrantel Embonate Summary Report (EMEA/MRL/491/98-FINAL). Data from the ECHA Classification & Labelling inventory regarding the local tolerance of pyrantel embonate indicate that pyrantel embonate is irritating to the skin, causes serious eye irritation, and may cause an allergic skin reaction. An appropriate warning is included in the product information.

Excipients

The applicant has provided information on the common use and safety profile of the individual excipients in the final formulation of the VMP, as intended for marketing.

Based on the information presented, noting the reported current and historical safe use, it can be considered that the excipients of the VMP are unlikely to have a potential for adverse systemic effects. It is accepted that they will not pose a concern to the user and that any risk to the user due to exposure to the final product will be determined by the active substances (both in terms of potential for systemic toxicity and local effects - potentially irritating to the skin and eyes and potentially sensitizing to skin).

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).

Bravecto TriUNO chewable tablets for dogs are supplied in an aluminium foil/blister package, with 1, 3 or 6 tablets per blister card. It is a hard, non-divisible, flavoured chewable tablet. The maximum strength tablet contains 600 mg fluralaner/1.5 mg moxidectin/300 mg pyrantel. The product is to be administered up to once monthly. The safety and efficacy data provided suggest that the combination of active substances does not present additional or increased toxicity effects compared to the sole treatment with any of the three individual active substances.

The main potential routes of exposure are considered to be dermal contact by pet owners/veterinary professionals during administration of the product to dogs and accidental oral ingestion by children.

Regarding adults, it can be accepted that the intended use and type of formulation means that the extent of user exposure to the active substances is likely to be very low. The MOE for adults (dermal and oral routes), exceeds 100 for each of the three active substances. For moxidectin, the MOE for pregnant women exceeds 1000. Taking into account the low risk of exposure, standard hygiene measures are considered to be adequate in minimising dermal exposure from handling the tablets and any hand-to-mouth or hand-to-eye transfer that might occur.

Since dust generation (potentially resulting in eye irritation) and high dose or prolonged user contact (potentially resulting in skin irritation and sensitization) with components of the VMP are

not expected at application based on the product properties, local effects for users by contact with this solid product are unlikely but should not be ruled out. However, hypersensitivity reactions were observed in humans for Bravecto products based on post-authorisation safety data, even though studies evaluating the potential for skin sensitisation for fluralaner or Bravecto (tablets and spot on) were negative. It is noted that the product information for these products have been updated with the user safety warning 'Hypersensitivity reactions in humans have been reported'. Furthermore, the applicant has cited information regarding the local tolerance of pyrantel embonate from the ECHA (2022) Classification & Labelling inventory: pyrantel embonate is irritating to the skin, causes serious eye irritation, and may cause an allergic skin reaction.

Despite the low risk, appropriate conservative advice to maintain elementary personal hygiene before, during and after use of the product is proposed for inclusion in the product information.

In relation to accidental oral ingestion by children, a 12.5 kg child accidentally ingesting the largest tablet strength would be exposed to 48 mg/kg fluralaner, 0.12 mg/kg moxidectin and 24 mg/kg pyrantel. For this scenario, the applicant has estimated margin of exposures of 0.21/4.2/0.5.

It is agreed that the exposure scenario with the greatest risk is the accidental ingestion of the largest tablet by a child. As the estimated MOE for oral exposure in a child for each active substance is less than 100, appropriate risk control options are required. The risk will be mitigated, in part, by the inclusion of appropriate warnings and safety measures on the SPC and package leaflet. In addition, the applicant has demonstrated that the primary packaging is child resistant, in accordance with ISO 14375:2018. The CVMP finds this an acceptable approach to mitigating the risk of accidental ingestion by a child.

Based on pharmacovigilance data reporting hypersensitivity reactions to Bravecto, and the known potential for sensitivity reactions to pyrantel embonate, the product literature includes the following statement: "Persons with known hypersensitivity to any of the active substances and/or excipients should avoid contact with the veterinary medicinal product." In addition, noting that the margin of exposure for children following accidental oral ingestion is below 100 for all active substances, and as adverse events have been observed in some humans administered moxidectin or pyrantel, additional information relating to the risk from accidental ingestion has been included in the product literature.

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-Corr.1).

Phase I:

The environmental risk assessment can stop in at Question 3 of Phase I and no Phase II assessment is required, as the veterinary medicinal product will only be used in non-food producing species.

However, the active ingredient moxidectin is classified as a PBT substance. In accordance with the *Question and answer document in support of the guideline on the assessment of persistent, bioaccumulative and toxic (PBT) or very persistent and very bioaccumulative (vPvB) substances in veterinary medicinal products* (EMA/CVMP/ERA/52740/2012) the PBT status of moxidectin is communicated in the product literature.

Overall conclusions on the safety documentation: safety tests

Bravecto TriUNO chewable tablets for dogs is a combination product containing the active substances fluralaner, moxidectin and pyrantel (as embonate). It has been developed as flavoured chewable tablets of different proportional strengths.

Fluralaner is a member of the isoxazoline class of parasiticides which acts by inhibition of the insect GABA gated chloride channels. Moxidectin belongs to the milbemycin group of macrocyclic lactones and has parasiticidal activity against a range of internal and external parasites. Pyrantel belongs to the group of tetrahydropyrimidines anthelmintics, which targets the nicotinic acetylcholine receptors (nAChRs) on nematode somatic muscle cells.

Pharmacodynamics and pharmacokinetics are addressed in Part 4. The data presented are considered adequate to characterise the toxicity profile of the active substances. The main toxicological findings can be summarised as follows:

Fluralaner:

- The potential systemic effects following sub-chronic and chronic exposure (oral and dermal) have been comprehensively investigated in the rat. The studies conducted meet guideline requirements (GLP and relevant OECD Test Guidelines). The liver appears to be the most sensitive organ for effects (increased organ weight, hepato-cellular fatty change, effects in related blood parameters). These effects were observed at dose levels above 20 mg/kg in the oral studies and above 100 mg/kg in the dermal studies. Sub-chronic and chronic (up to 52 weeks) effects following oral exposure in dogs was also comprehensively investigated. Reductions in cholesterol, phospholipids and triglycerides were consistently observed, although no histopathological changes of the liver were reported. It can be concluded that the dog is more sensitive to the effects of fluralaner than the rat and that systemic exposure of fluralaner is greater in dogs than in rats.
- The oral NOAEL for maternal toxicity and NOEL for embryo/foetal development were determined to be 10 mg/kg bw/day, as derived from the rabbit developmental toxicity study. In the developmental toxicity study the oral NOEL for maternal and foetal organisms in rats was set to 100 mg/kg bw per day by CVMP.
- Studies on carcinogenicity were not conducted for fluralaner. This is justified by the negative results in all genotoxicity tests, and further by the absence of any pre-neoplastic lesions in the multiple repeat-dose studies of up to chronic duration and at a wide range of dose levels.
- Fluralaner was non-irritating in an ocular irritation study and non-irritating in a dermal irritation study. It is not considered a sensitiser based on results of a maximization test conducted in albino guinea pigs. However, hypersensitivity reactions to fluralaner have been reported in periodic safety assessments for other Bravecto products. No effects on the nervous system in humans have been reported for fluralaner in the toxicity tests provided.

Moxidectin:

- In repeat dose toxicity studies, an oral NOEL of 0.3 mg/kg bw/day was established in a dog 90 -day toxicity study based on dose dependent reductions in absolute body weights and food consumption identified in dogs. Nervous system effects were observed at higher doses in mice, rats and dogs. In a 4-week dermal toxicity study conducted in rats a dermal NOAEL of 10 mg/kg bw/day could be derived, based on effects on sensory reactivity and motor activity at 70 mg/kg bw/day.

- In the reproduction studies, a NOEL of 0.4 mg/kg bw/day was concluded based on reduced pup survival at doses of moxidectin above 0.4 mg/kg bw/day in rats. In teratogenicity studies in rats and rabbits, the NOEL for maternotoxicity was 5 mg/kg bw/day in rats and 1 mg/kg bw/day in rabbits. Foetal alterations such as cleft palate, micrognathia, not ossified or incomplete ossified ribs were reported for doses higher than 2.5 mg/kg bw/day in rats. No effects on foetal development were observed in rabbits. The NOEL for embryotoxic effects was 2.5 mg/kg bw/day in the rat and more than 10 mg/kg bw/day in the rabbit.
- Moxidectin is not considered to be of mutagenic concern. Carcinogenicity studies in rats and mice did not show the potential for carcinogenicity.
- Moxidectin may be slightly irritating to the eyes and skin. While acknowledging that only limited information was available in the JECFA monograph for moxidectin, the CVMP concluded previously that moxidectin is non-sensitising to the skin.

Pyrantel embonate:

- In dogs orally administered pyrantel embonate in a 13-week repeat dose toxicity study, a NOEL of 35 mg/kg bw/day was established based on increased serum aspartate aminotransferase and serum alanine aminotransferase values at higher dose levels.
- Reproduction toxicity was only studied in laboratory animals and not in the target species. Data in rats indicate that pyrantel embonate is not a developmental or reproductive toxicant.
- Pyrantel is not considered to be mutagenic. Carcinogenicity studies have not been performed and are not required.
- It is reported that pyrantel embonate can cause eye and skin irritation as well as skin sensitisation. In the battery of studies on fluralaner and moxidectin and in the publicly available information on pyrantel embonate, a potential risk for users by irritation to the skin and eyes and immunotoxicity to the skin was identified after possible dermal exposure to the VMP, mainly based on pyrantel embonate. No effects on the nervous system have been observed for pyrantel embonate in the toxicity tests provided.

The proposed excipients are generally considered to be of low toxicity and/or present at low concentrations. Therefore, the systemic and local toxicity of this product will likely be determined by its active substances, fluralaner, moxidectin and pyrantel (as embonate).

A user safety assessment in line with the relevant CVMP guidance has been presented. The worst-case scenario for user safety is ingestion of a tablet by a child, with an estimated margin of exposure (MOE) of 0.21, 4.2, and 0.5 for fluralaner, moxidectin and pyrantel embonate, respectively. The risk identified will, in part, be mitigated by the inclusion of appropriate safety advice/warning statements in the SPC and package leaflet. Additionally, given the potential risk to children by accidental ingestion, the applicant has demonstrated that the packaging is child resistant in accordance with ISO 14375:2018. In general, appropriate warnings for the user have been included in the product literature. However, based on pharmacovigilance data reporting hypersensitivity reactions to the Bravecto range of products, and the known potential for sensitivity reactions to pyrantel embonate, the potential risk of hypersensitivity reactions to the active substances and/or excipients is communicated in the product information.

An appropriate environmental risk assessment was provided. Based on the data provided the ERA can stop at Phase I, as none of the Phase II criteria are met. However, as the active ingredient moxidectin is classified as a PBT substance, this is communicated in the product information.

Part 4 – Efficacy

Pre-clinical studies

Bravecto TriUNO is a new fixed combination VMP for dogs containing fluralaner, moxidectin and pyrantel indicated for the treatment of mixed parasitic infestations by ticks or fleas, gastrointestinal nematodes, lungworm and/or heartworm. Fluralaner is an acaricide and insecticide belonging to the isoxazoline group. Moxidectin belongs to the milbemycin group of macrocyclic lactones and has parasiticidal activity against a range of parasites including various nematode species, while pyrantel is an anthelmintic of the tetrahydropyrimidine class, with efficacy against gastrointestinal parasites.

The proposed dose is 10-20 mg/kg of fluralaner, 0.025-0.05 mg/kg of moxidectin and 5-10 mg/kg of pyrantel, and the product may be administered as frequently as one treatment per month, year-round.

The dossier submitted in support of efficacy contains both proprietary studies, and references to published literature. The application also makes reference to the dossier / studies and previous assessment of the CVMP in the context of the marketing authorisation application for 'Bravecto chewable tablets for dogs'.

Pharmacology

Pharmacodynamics

The applicant has provided a comprehensive summary of the pharmacodynamics of the active substances fluralaner, moxidectin and pyrantel based on published literature. The main pharmacodynamic characteristics may be summarised as follows.

Fluralaner belongs to the isoxazolines class and has parasiticidal activity against ectoparasites but is ineffective against nematodes. Moxidectin is a semisynthetic derivative of nemadectin and belongs to the milbemycin group of macrocyclic lactones. It has parasiticidal activity against a range of internal and external parasites (including mites, larval and adult stages of nematodes, lungworm and heartworm). Pyrantel is a pyrimidine-derivative anthelmintic with parasiticidal activity against gastrointestinal parasites.

Fluralaner and moxidectin have common receptor targets, the ligand-gated chloride channel receptors GABA-R and glutamate-R. Fluralaner is a potent inhibitor of GABA-R, and when bound prevents binding of endogenous GABA in the arthropod CNS, causing paralysis and death. Fluralaner also inhibits glutamate-R, albeit less potently. Moxidectin is a positive modulator of glutamate-R, and also demonstrates (lesser) activity on GABA-R. Binding of moxidectin to its target receptors also results in insect paralysis and death. Pyrantel is a selective agonist of the nematode nicotinic acetylcholine receptor (nAChR), and binding to this receptor causes a depolarising neuromuscular blockade resulting in paralysis and death of the parasite.

Glutamate-Rs are exclusively expressed in invertebrates, whereas GABA-Rs and nAChRs are expressed in both invertebrate and mammalian tissues. Although invertebrate GABA-Rs are significantly more sensitive to macrocyclic lactones, including moxidectin, than mammalian GABA-Rs, binding of moxidectin to mammalian GABA-R can result in neurotoxicity. Similar high affinity of fluralaner for invertebrate GABA-Rs (as compared to mammalian) is reported, however fluralaner is not reported to have the same neurotoxicological potential as that of moxidectin. P-glycoprotein (p-gp)-mediated efflux at the blood-brain barrier limits exposure of the mammalian CNS to moxidectin, however in sensitive sub-populations with non-functioning p-gp such as dogs that are homozygous for the MDR1 (-/-) mutation, the threshold for signs of neurotoxicity is lower than in other animals.

Limited oral bioavailability of pyrantel along with high specificity for nematode nAChRs reduces the potential for secondary (adverse) pharmacodynamic effects associated with pyrantel.

In respect of the potential for interaction between the active substances in the VMP impacting efficacy or target animal safety, the following points can be concluded:

- nAChRs are sufficiently distinct (in terms of specificity for ions, structure and physiological function) from GABA-R and glutamate-R for interaction between pyrantel and fluralaner or moxidectin to be excluded.
- Taking into account the different interactions of fluralaner and moxidectin on GABA-R and glutamate-R (inhibition and positive modulation respectively), and relevant *in vitro* data, it is unlikely that fluralaner shares the macrocyclic lactone binding site on these receptors.
- Laboratory studies as previously assessed by CVMP in the context of the MAA for Bravecto) have demonstrated a lack of efficacy of fluralaner on nematodes.
- The selectivity of fluralaner and moxidectin for mammalian GABA-R is low which is supported by safety study data in target animals including ivermectin-sensitive Collies. Pyrantel also demonstrates high affinity for nematode nAChRs, and is poorly absorbed from the gastrointestinal tract, thereby limiting its potential to adversely affect target animal safety.

Despite a theoretical potential for interaction between fluralaner, moxidectin and the general risk for interaction due to co-medication with pyrantel, the results of *in vivo* studies performed using the VMP provided no evidence of any clinically relevant interference of the combination regarding efficacy, pharmacokinetics, and safety.

The CVMP considers the information included in section 4.2 'Pharmacodynamics' of the SPC to have been suitably supported by the information provided, noting also that in respect of fluralaner, this information is largely consistent with that already approved by the CVMP for the SPC of the authorised VMP 'Bravecto chewable tablets for dogs'.

Further detail (from proprietary studies) is also provided in respect of the mechanism of action of fluralaner, which reflects the relatively recent use of this particular active substance in veterinary medicinal products, as compared to moxidectin and pyrantel which are considered by the applicant to be in well-established use.

The applicant has presented three *in vitro* studies concerning the efficacy of fluralaner against isolates of the flea species *Ctenocephalides felis* from three different geographical regions.

The fluralaner binding sites in the arthropod GABA-R and glutamate-R have been predicted based on published scientific literature, and the relevant coding sequences for these fluralaner binding sites were compared in *Ctenocephalides felis* isolates from three distinct geographic regions (USA, EU and Australia). For both GABA-R (the main pharmacological target of fluralaner) and glutamate-R (a less significant pharmacological target), it was concluded based on the results of these studies that the sequences of the predicted fluralaner binding sites did not differ between the three isolates.

Acknowledging that differences in susceptibility to fluralaner may not only be determined by on-target genetic differences, the applicant also performed an *in vitro* functional comparison on three *C. felis* isolates from distinct geographical regions. The insecticidal activity of fluralaner was evaluated by means of a flea-contact-test. The number of dead, damaged and live fleas were counted after 48h continuous exposure to fluralaner. Predicted lethal and effective concentration values were calculated. It was concluded that *in vitro* susceptibility of *C. felis* to fluralaner did not differ based on the geographical origin of the *C. felis* isolate.

Taking the totality of the data presented into account, including *in vivo* laboratory and field efficacy data assessed in previous applications, the CVMP accepts that based on the three *Ctenocephalides felis* isolates evaluated (from the USA, the EU and Australia), there is currently no strong evidence to suggest that susceptibility of *C. felis* to fluralaner will differ based on geographical origin.

The applicant has presented three *in vitro* studies concerning the efficacy of fluralaner against isolates of the tick species *Rhipicephalus sanguineus* from two geographical regions (the EU and the US).

The fluralaner binding sites in the arthropod GABA-R and glutamate-R have been predicted based on published scientific literature, and the relevant coding sequences for these fluralaner binding sites were compared in *R. sanguineus* isolates from two distinct geographic regions (USA and EU). For both GABA-R (the main pharmacological target of fluralaner) and glutamate-R (a less significant pharmacological target), it was concluded based on the results of these studies that the sequences of the predicted fluralaner binding sites did not differ between the isolates.

Acknowledging that differences in susceptibility to fluralaner may not only be determined by on-target genetic differences, the applicant also performed an *in vitro* functional comparison on two *R. sanguineus* isolates (from the US and EU). The insecticidal activity of fluralaner was evaluated by means of immersion for 5 minutes in fluralaner at a range of concentrations. The number of dead, damaged and live ticks were counted after 48h incubation post-exposure. Predicted effective concentration values (EC50, EC90) were also presented.

The results of this study support similar *in vitro* susceptibility to fluralaner in isolates from the EU and the US and based on the results of the *in vivo* laboratory studies presented and assessed in the context of the original MAA for Bravecto, comparable efficacy up to 58 days is reported for tick isolates from the EU and the US. Noting also the reported similarity of the predicted fluralaner binding sites in the GABA and glutamate receptors of two isolates of *R. sanguineus* (from the EU and the US), it is considered that there is currently no strong evidence to suggest that susceptibility of *R. sanguineus* to fluralaner will differ based on geographical origin.

In summary, the applicant has presented a clear and comprehensive summary of the pharmacodynamic properties of the active substances in the VMP that is based on peer-reviewed published literature. Sufficient information was presented in order to conclude on the mechanism of the main (adverse) secondary pharmacodynamic effects in the target animal species. The potential for interaction between the three active substances that could adversely affect the efficacy or target animal safety of the VMP was suitably evaluated. Further results from proprietary studies concerning the mechanism of action of fluralaner have also been provided.

Pharmacokinetics

The applicant has presented the results of three *in vivo* pharmacokinetic studies (one pivotal and two pilot), and pharmacokinetic data were also generated in the pivotal target animal safety study.

In a GLP-compliant pivotal pharmacokinetic study the pharmacokinetics (based on plasma concentrations) of the three active substances (fluralaner, moxidectin and pyrantel) in the target animal species (Beagle dogs) were characterised when administered in a combined oral tablet, and separately.

The test articles (based on the VMP) were administered at the following dose rates: 0.5X, 1X and 2X the maximum recommended clinical dose (that is, 20/0.5/10 mg/kg of fluralaner/moxidectin/pyrantel). Chewable tablets containing the individual active substances were also administered at a dose equivalent to 1X the maximum recommended clinical dose for each active substance. Fluralaner and moxidectin were administered to one group via intravenous bolus at 5 +

0.5 mg/kg bw of fluralaner and moxidectin, respectively. Blood samples were obtained from animals post-administration of the test product for a suitable time period to define relevant pharmacokinetic parameters.

Validated HPLC-MS/MS methodology was used to measure concentrations of the following analytes in plasma samples: R-fluralaner, S-fluralaner, moxidectin and pyrantel. Total fluralaner is reported and is the sum of the enantiomers S-fluralaner and R-fluralaner. Pharmacokinetic parameters based on plasma concentrations of analytes were estimated using a non-compartmental approach.

All test articles were well-tolerated in all animals. Administration of the combined active substances (in a chewable tablet) resulted in systemic exposure to all active substances and substantial differences in parameters were not observed when the same dose was administered as part of a combination, or as a single active substance.

Bioavailability for fluralaner ranged from 48.3 – 73.3% over the dosages tested and was similar whether 20 mg/kg bw fluralaner was administered alone (50.6%) or as part of a combination product (48.3%). Bioavailability for moxidectin ranged from 54.5 – 88.9% over the dosages tested and was higher when 0.5 mg/kg bw moxidectin was administered part of a combination product (73.2%) as compared to alone (60.2%). Pyrantel is poorly absorbed following oral administration.

Noting that this fixed combination product does not concern new active substances, and the pharmacokinetics of the active substances concerned have been previously documented, it is considered appropriate that the focus of this pivotal pharmacokinetic study was the investigation of whether administration of the combination product has any impact on the pharmacokinetic profile of each active substance compared to when the active substance is administered alone. It is accepted that the results of this study support the contention that administration of the combination of fluralaner, moxidectin and pyrantel in a combined chewable tablet does not impact on the pharmacokinetics of any of the individual active substances.

The results of two non-GLP-compliant pilot pharmacokinetic studies in which plasma concentrations of the active substances following administration of a combined tablet were compared to those following administration of fluralaner alone, and another authorised combination product were also provided, however these studies did not evaluate the final formulation of the VMP to be marketed and are considered preliminary in nature.

Taking into account the pharmacokinetic data generated in the studies submitted with this dossier and supplemented with publicly available information (concerning metabolism and excretion of active substances primarily), the pharmacokinetics of the active substances fluralaner, moxidectin and pyrantel have been summarised in the SPC.

The SPC text is supported by the data provided and was accepted by CVMP.
It is noted that based on the results of the pivotal TAS study accumulation of fluralaner and moxidectin occurred over the course of the study (during which the VMP was administered to puppies 7 times at 30-day intervals). Considering the proposed dosing regimen that allows for potentially unlimited monthly treatment, the information that accumulation occurs for fluralaner and moxidectin after repeated monthly administration has been included in SPC section 4.3, with cross-reference to sections 3.5 and 3.10 for further detail concerning the available safety data. For fluralaner, when administered at 1X RTD, steady state conditions were not reached after the 6^{th} dose; for moxidectin administered at 1X RTD, steady state conditions were achieved after approximately 5 doses.

A statement concerning the effect of prandial state on the extent of absorption of fluralaner is consistent with information approved in the SPC for Bravecto chewable tablets for dogs. Although the effect of prandial state *per se* was not investigated in the studies presented with this dossier, given

the lack of evidence for any interaction between the active substances, it is considered likely that a similar effect of prandial state on the absorption of fluralaner will be applicable for the VMP.

It is concluded that the applicant has presented a suitably thorough account of the pharmacokinetics of the active substances fluralaner, moxidectin and pyrantel. Furthermore, it is accepted that there are no data to suggest that interaction(s) between the active substances will occur and impact upon pharmacokinetics in the target animal species. The information as included in section 4.3 of the SPC is supported by the data presented.

Justification of the fixed combination

The applicant has provided a detailed summary of the 'Justification of the combination of fluralaner, moxidectin and pyrantel in 12.5% (w/w) fluralaner + 0.03% (w/w) moxidectin + 6.25% (w/w) pyrantel chewable tablet'.

The combination is considered justified based on the broadening of the spectrum of activity, the current favourable resistance situation of the three parasiticides, and general therapeutic advantages, such as enhancement of treatment practicability and compliance. Based on both pharmacological and clinical endpoints, no interaction is to be expected between fluralaner, moxidectin and pyrantel, when administered as a chewable tablet to dogs, at the clinical dose range of 10-20 mg/kg fluralaner, 0.025-0.05 mg/kg moxidectin and 5-10 mg/kg pyrantel.

The combination of fluralaner, moxidectin and pyrantel in the VMP is considered to have been suitably justified in line with the requirements of guideline EMA/CVMP/83804/2005-Rev.1.

Development of resistance and related risks in animals

A review of both published and proprietary data concerning the development of resistance to the three active substances fluralaner, moxidectin and pyrantel has been provided.

Due to the exclusive preponderant use of fluralaner in companion animal veterinary medicinal products, and relatively recent initial authorisation of the active substance (in 2014), there are limited data in the public domain concerning resistance to fluralaner. It has been demonstrated in *in vitro* studies that resistance to dieldrin in arthropod species (conferred by the single point mutation A302S in the GABA-R subunit Rdl) and also in the cat flea species, *Ctenocephalides felis* is not associated with cross-resistance to fluralaner. Furthermore, the predicted fluralaner binding sites in 3 *C. felis* isolates (obtained from the EU, Australia and the US) are reported as being the same in respect of their genetic (and protein) sequences, and *in vitro* fluralaner susceptibility. Fluralaner is also reported to be effective against a fipronil tolerant strain of *C. felis.*

Concerning moxidectin, there are recent reports of multiple anthelmintic drug (including moxidectin) resistance in hookworms (*Ancylostoma caninum*) in the US. There are also reports of treatment failure of macrocyclic lactones for *Dirofilaria immitis*, although it is also reported that heartworm treatment failures are also frequently associated with non-compliance. Resistance of *D. immitis* to ivermectin and moxidectin has been reported in published laboratory studies. However, in two published laboratory studies comparing multiple macrocyclic lactones, only animals treated with moxidectin were reported to be 100% free of adult *D. immitis*. Mechanisms of resistance to macrocyclic lactones remain incompletely understood. In support of efficacy of the VMP for treatment of *D. immitis*, the applicant conducted analysis of adult *Dirofilaria immitis* worms collected at necropsy in two dose confirmation studies (both conducted in the US). The results of the study demonstrated that the strains collected from one study were considered macrocyclic lactone resistant, whilst the strains collected from the other study were considered macrocyclic lactone susceptible. A further study was conducted to determine the genotypic susceptibility/resistance status

of field isolates collected at random across Europe. The samples exhibited a similar genotypic profile to that of susceptible isolates, which suggests that macrocyclic lactone resistance in *D. immitis* is not prevalent in Europe currently.

Pyrantel resistance is not considered to have clinical significance in companion animals in Europe. A high level of pyrantel resistance in *Ancylostoma caninum* has been reported in a study in dogs in Australia, and multi-drug (including pyrantel) resistant *A. caninum* has also been reported in the US.

The applicant has presented a reasonably comprehensive and current summary of knowledge concerning development of resistance to the active substances fluralaner, moxidectin and pyrantel. Although limited information concerning the mechanisms of resistance has been provided, it is acknowledged that for fluralaner, such information is not available and in the case of moxidectin, it is also acknowledged that there is incomplete understanding of the mechanisms of resistance to macrocyclic lactones. Concerning the risk of development of resistance in animals, it is considered notable that reports of resistance to both moxidectin (*Ancylostoma caninum* and *Dirofilaria immitis*) in the US, and to pyrantel (*A. caninum*) in the US and Australia have been published recently.

The applicant has included text in the SPC for the purposes of mitigating against the risk of resistance development arising from use of the VMP. The text is consistent with the recommended text for these sections as included in the Guideline on the summary of product characteristics for antiparasitic veterinary medicinal products (EMA/CVMP/EQP/170208/2005-Rev.1 Corr.).

Dose determination and confirmation

Dose justification

Bravecto TriUNO is for oral administration at a dose rate of 10-20 mg/kg of fluralaner, 0.025-0.05 mg/kg of moxidectin and 5-10 mg/kg of pyrantel. A dosing table has been provided in the product information. The VMP is for monthly administration on a year-round basis.

The dose rate and dosing interval for Bravecto TriUNO was established based on the findings of dose determination studies conducted in the target species, published literature and extrapolation from already authorised indications for the active substances. This is considered acceptable.

Dose determination studies

Fluralaner

In support of dose determination data for the active substance fluralaner, the applicant has made reference to previous applications for Bravecto chewable tablets and Bravecto Spot-on, with data having been provided with those applications, demonstrative that *Amblyomma americanum* is the US tick species which is the least susceptible to fluralaner at all assessment timepoints. However, given that *A. americanum* is not prevalent in the EU a speed of kill study against the EU prevalent tick species *D. reticulatus* has been presented with this application, the results of which are summarised below.

The study was conducted outside of the EU using an EU tick strain. Therefore, it is reasonable to consider the tick isolate used as representative of the current field situation in the EU.

With regards sample size and study animals, these can be considered largely adequate in terms of both guideline requirements and representativeness of the target population. With regards housing, it is noted that animals were individually housed for the duration of the study, which is neither in accordance with Directive 2010/63/EU or guideline EMEA/CVMP/EWP/005/2000 Rev.4, which recommends that housing of animals in groups should be considered, where feasible/appropriate.

With regards the test articles, it is noted that neither of those evaluated comprised the final formulation, one being a combination product, whilst the other contained solely fluralaner. Despite these deficiencies, it can be accepted that fluralaner is the active substance with acaricidal activity against ticks. Furthermore, and as discussed under the pharmacokinetics section, it can be accepted that there are no interactions between the active substances included in the formulation and given that similar observations are expected for the fluralaner combination, it can be accepted that the findings derived from this study can be extrapolated to fluralaner alone or fluralaner in combination with moxidectin and pyrantel.

Approximately 50 unfed adult *D. reticulatus* ticks (25 male: 25 female) were applied to each study animal, which is in accordance with guideline EMEA/CVMP/EWP/005/2000 Rev.3. The infestation method was well characterised. Following infestation, at each tick-count timepoint all control animals were determined to be adequately infested. Efficacy calculations were conducted in line with Abbott's formula using arithmetic mean data. For the fluralaner combinations, an improved level of efficacy was observed for the 10 and 20 mg fluralaner/kg bw dose rates over the 5 mg fluralaner/kg bw dose rate, with an adequate level of efficacy (≥90%) observed at 12 hours on SD0 and at 24-36 hours at all timepoints thereafter (up to Day 30). Similar results were observed for the mono-active formulation with administration of 10 mg fluralaner/kg bw demonstrating an adequate level of acaricidal efficacy (≥90%) at 12 hours on SD0 and at 24 hours at all timepoints thereafter.

The applicant has additionally provided the results of dose determination studies conducted with the American tick species, *A. americanum*: the studies demonstrate the 10 mg fluralaner/kg bw dose as the most effective for the longest duration (38 days).

With regards the tick species selected for the provision of dose determination data in support of this application, although efficacy for *D. reticulatus* was evaluated, it is noted that the relevant European dose limiting tick species is *R. sanguineus*. Indeed, from the data provided, a reduced duration of efficacy was clearly observed for *R. sanguineus* compared to the other tick species, as reflected in the approved product indications for Bravecto chewable tablets and Bravecto Spot-on with a shorter claim for efficacy for *R. sanguineus* compared to the other tick species (8 vs 12 weeks, respectively).

That said, a number of dose confirmation studies have been conducted in the dose-limiting species, *R. sanguineus* and the applicant has provided bridging data to allow for extrapolation of those data to the tick species, *D. reticulatus*, *I. ricinus* and *I. hexagonus*.

Moxidectin

In support of dose determination for moxidectin, the applicant has referred to the well-established use for this active substance. In support of this claim, the following is noted:

- Moxidectin has been authorised as a VMP for the prevention of heartworm disease for in excess of 10 years;
- The proposed dose for the product for the prevention of heartworm disease (0.025-0.05 mg moxidectin/kg bw) is in line with the dose of other VMPs containing moxidectin which have been approved for that indication;
- A large amount of information on moxidectin is in the public domain, providing some assurance of an acceptable level of safety.

The applicant additionally provided the results of a dose determination study conducted for moxidectin with *A. vasorum*. This study was conducted largely in line with the relevant guidelines: VICH GL9, VICH GL7, and VICH GL19. Infection was induced with European isolates. The inoculate

volume was 200 infective *A. vasorum* larvae and whilst the relevant guidelines do not specify a number of infective forms required to achieve an adequate infection with *A. vasorum,* given that all of the control animals were considered to be adequately infected at necropsy within a range of 32-137 worms, it can be accepted that the inoculate volume was suitable.

This study included four treatment groups, one negative control group and three groups administered the VMP at doses of 0.5XRTD (Group 1), 1XRTD (Group 2) and 2XRTD (Group 3). The VMP was confirmed as the final formulation. The VMP was administered in accordance with the SPC, 31 days post-inoculation (which would allow for evaluation of efficacy against the L5 larval stage and adult stages).

The primary efficacy parameter was adult worm count at necropsy with each of the test article groups compared to the control group. Additional efficacy parameters included faecal larvae counts, respiratory parameters as markers of the disease and serology for the assessment of *A. vasorum* antigen and antibody. For the primary efficacy parameter, animals were euthanized for necropsy at SDs 33, 34 and 35, with pulmonary tissue processed for the collection of *A. vasorum worms.* Efficacy calculations were conducted using Abbott's formula based on geometric mean data. For all doses of moxidectin, the percentage efficacy met guideline requirements (>90%) however it was observed to increase with increasing dose with 92.2% efficacy observed for Group 1 (moxidectin at 0.0125 mg/kg), 99% for Group 2 (moxidectin at 0.025 mg/kg) and 100% for Group 3 (moxidectin at 0.05 mg/kg). When efficacy calculations were conducted using arithmetic mean data, >90% efficacy was similarly observed (90.1% for Group 1, 98.7% for Group 2 and 100% for Group 3). A statistically significant difference was observed at all timepoints.

The findings derived from the secondary efficacy parameters supported the efficacy findings of the primary endpoint. For faecal larval counts, efficacy was >90% at all dose rates (range 99-100%), for respiratory parameters, improvements in respiratory rates were observed in the higher dose groups whilst the lowest dose group remained unchanged, and the untreated control increased. Similarly for serology, changes in *A. vasorum* antibody and antigen levels across the groups over the course of the study were indicative of an adequate infection rate, but also of product efficacy with antigen levels reduced in the treated groups compared to the control reflective of product efficacy for the prevention of larvae development into adult worms.

Based on the results of the study, an acceptable level of efficacy (>90%) against *A. vasorum* infection was observed at all doses of moxidectin; however given the dose effect observed, the applicant's decision to progress to dose confirmation with the 0.025 mg/kg dose is considered reasonable. In light of both the data derived from the dose determination study for *A. vasorum* and the justification provided for the claim of well-established use for moxidectin, the dose determination requirements for moxidectin can be considered satisfied.

Pyrantel

In order to exempt from the requirement to provide dose determination data for pyrantel, the applicant makes reference to the well-established use for this active substance, noting the following:

- Pyrantel has been authorised as a VMP for oral administration to dogs within the community for in excess of 10 years;
- Pyrantel-containing products are approved for the treatment of ascarids (*T. canis, T. leonina* (adult and late immature forms) and hookworms (*U. stenocephala, A. caninum* (adults)). The doses approved for other products are in line with that proposed for the formulation (5-10 mg pyrantel/kg bw);

- A large amount of information on pyrantel is in the public domain, providing some assurance of an acceptable level of safety.

In light of the above, the omission of specific dose determination data with this application can be accepted as pyrantel when administered at a dose rate of 5-10 mg/kg to dogs, can be accepted as well-established.

Dose confirmation studies

Ticks

In support of the indications for *Dermacentor reticulatus, Ixodes hexagonus, Ixodes ricinus,* and *Rhipicephalus sanguineus*, the applicant has presented four dose confirmation studies designed to investigate efficacy against the dose limiting tick species, *R. sanguineus*. In all cases the formulation of the VMP administered was confirmed as the final formulation for marketing.

Infestations were with recent US and Brazilian isolates, however, as discussed under the pharmacodynamics section of the scientific overview, there is currently no evidence to suggest that susceptibility of *R. sanguineus* to fluralaner will differ based on geographical origin and therefore, the results derived from these studies can be extrapolated to the European situation.

Concerning housing, it is noted that the animals were individually housed for the study duration, which is not in line with EMEA/CVMP/EWP/005/2000 Rev.4 nor Directive 2010/63/EU, and for some of the studies, animals were selected at a very young age (7-8 weeks). With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses close to the minimum evaluated (10.15-19.2 mg fluralaner/kg bw, 0.025-0.048 mg moxidectin/kg bw and 5.1-9.615 mg pyrantel/kg bw).

In all studies, the infestation methods were well characterized and considered appropriate and adequate tick infestations were observed for the control animals at all timepoints. For each study, approximately 50 unfed adult *R. sanguineus* ticks in a ratio of 50:50 male to female ticks were applied to each study animal, which is in accordance with guideline EMEA/CVMP/EWP/005/2000 Rev.3. Following infestation, at each tick-count timepoint an adequate number of control animals (minimum 6) were determined to be adequately infested.

According to guideline EMEA/CVMP/EWP/005/2000-Rev.4, when claiming immediate efficacy, efficacy testing at day 0 up to 48 hours may be acceptable whilst for short-term persistent efficacy (up to 4 weeks), infestation should be conducted on a weekly basis with efficacy testing at 48 hours post-challenge. A persistent killing effect against *R. sanguineus* for 30 days is mentioned in the product SPC and in these studies, tick counts were conducted 48 hours post-treatment or infestation for the entire duration of efficacy (study days 2, 7, 14, 21 and 31).

Efficacy calculations were conducted in line with Abbott's formula using arithmetic mean data. Efficacy based on live tick count was considered to be the primary efficacy parameter with a percentage reduction (compared to control) of ≥99.5% at all timepoints up until study day 31 for three of the four studies presented. A statistically significant difference was observed at all timepoints. For the treatment claim, dead tick count was evaluated and percentage reduction (compared to control) was 100% at all timepoints, with a statistically significant difference observed at all timepoints except SD 31.

For one study, efficacy was 85.3% at SD 31. However, given that an adequate level of efficacy (≥90%) was observed for all other dose confirmation studies up until SD 30 (which is the duration of efficacy for the product) and an adequate level of efficacy was concluded in the clinical field trial data presented, the indication for the control and treatment of *R. sanguineus* ticks for up to 30 days

can be considered supported.

With regards adverse events, it is noted that vomiting and diarrhoea episodes were observed in a number of animals across these studies, however, the CVMP notes that emesis and diarrhoea have been included as examples of 'digestive tract disorders' under SPC section 3.6 as a common AE.

In conclusion, the above studies can be considered supportive of efficacy against *R. sanguineus* with persistent effect for up to 30 days post-treatment. Given that this tick species can be considered the dose-limiting EU tick species, extrapolation of these data to the other tick species (*D. reticulatus, I. ricinus and I. hexagonus)* is considered acceptable.

Fleas

In support of the indications for *C. felis* and *C. canis*, the applicant has presented five dose confirmation studies which were well designed and largely compliant with the relevant guidance documents. In all cases the formulation of the VMP administered was confirmed as the final formulation for marketing.

In all cases, infestations were with US, Australian or Brazilian isolates of *C. felis*. As discussed under the pharmacodynamics section of the scientific overview, there would appear to be no evidence to suggest that susceptibility of *C. felis* to fluralaner will differ based on geographical origin. In all studies, the infestation methods were well characterized and considered appropriate and adequate flea infestations were observed for the control animals at all timepoints. In one study, animals were offered food and water once midway through a 24-hour period. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses close to the minimum of the recommended dose range (10.0-15.625 mg fluralaner, 0.025-0.039 mg moxidectin and 5.00-7.81 mg pyrantel per kg bw).

For four of the studies, approximately 100 newly emerged and unfed adult *C. felis* fleas were applied to each study animal. Following infestation, at each flea-count timepoint an adequate number of control animals (minimum 6) were determined to be adequately infested in line with EMEA/CVMP/EWP/005/2000 Rev.4. For two of the four studies, flea counts were conducted in general 24 hours post-treatment or infestation to cover the whole period of efficacy (study days 1, 7, 14, 21 and 30 or 2, 5, 9, 16, 23, 30, 37 and 44) with exception of the first observation time point in one study, which was 48h after infestation. Efficacy calculations were conducted in line with Abbott's formula using arithmetic mean data. Against existing infestations evaluated at 24 hours post- treatment, percentage reduction (compared to control) was 100% for both arithmetic and geometric means data and therefore above threshold. Against new infestations, percentage reductions were ≥ 99.8% at 24 hours following infestation up to study day 30. A statistically significant difference was observed at all timepoints.

One study evaluated efficacy against existing infestations at 12 hours after treatment and for new infestations at 12 hours post-infestation. For existing infections, percentage reduction (compared to control) was 100% for both arithmetic and geometric mean data. Against new infestations, percentage reductions at 12 hours, based on arithmetic means, were 99.9%, 85.9%, 73.2% and 56.2% on Days 7, 14, 21 and 30 respectively. Efficacy based on geometric means data was 99.9%, 88.1%, 77.7% and 66.0% on Days 7, 14, 21 and 30 respectively. A statistically significant difference was observed at all timepoints.

Another study evaluated efficacy at only a single timepoint, that is 4 hours post-treatment with 72.2% efficacy observed for arithmetic mean data and 80% for geometric means data. Whilst this study provides evidence of some efficacy against pre-existing *C. felis* infestations, the efficacy observed is not acceptable in accordance with guideline recommendations (≥95%) to support a

claim for efficacy at 4 hours.

In the study conducted in Australia, between 50 and 100 adult fleas were applied to each study animal, and adequate infestation was confirmed. Flea counts were conducted 48 hours after infestation at each timepoint (until Day 37). While counting fleas at 48h post infestation represents a deviation from guidance for determination of immediate and persistent efficacy against fleas, it is noted that 100% efficacy was reported at all timepoints.

In conclusion, the above studies can be considered supportive of the indication for immediate and persistent flea killing activity against *C. felis*, given that ≥99.8% efficacy was observed at 24 hours post-infestation up to study day 30, with >95% efficacy also observed at 12 hours post-treatment and 12 hours post-infestation on study day 7.

Fluralaner has previously been demonstrated to have similar potency for both *C. felis* and *C. canis*, therefore it is considered acceptable to extrapolate efficacy data for *C. felis* to *C. canis*.

Nematodes

Ancylostoma caninum

Adult claim

In support of the indication for adult *A. caninum*, the applicant has presented three dose confirmation studies which were well designed and largely compliant with the relevant guidance documents. The formulation evaluated was the final formulation for marketing. All of the studies involved induced infections, with one of the experimental isolates originating from naturally occurring infections in Europe in 2018, with the others originating from the US in 2022. This is considered acceptable in line with VICH GL19 which specifies that 'If both studies are conducted using experimentally infected animals, then parasites must have originated from naturally occurring infections from different geographical regions no older than 10 years prior to use for inducing infection'. The inoculate volumes were 250-300 *A. caninum* L3 larvae, which is in line with VICH GL19.

The studies were negatively controlled. The study animals and sample sizes can be accepted in line with guideline specification and as representative of the target population. However, the study animals were individually housed for the study duration, which is not in line with EMEA/CVMP/EWP/005/2000 Rev.4. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses close to the minimum of the recommended dose range (dose rates ranged from 10-16.13 mg fluralaner, 0.025-0.040 mg moxidectin and 5.0-8.06 mg pyrantel per kg bw). Treatment for adult *A. caninum* was administered 25 days post-inoculation, which is consistent with VICH GL19 which specifies that the recommended time of treatment after infection is >21 days.

For two of the studies the primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, as observed following necropsy (SD 7). Adequate infections were observed in at least 6 control animals in each study. For these studies, the percentage reduction for the formulation compared to control was calculated as 99.3% using geometric mean data and 99.1% using arithmetic mean data for one study and, for the other study, 85.8% efficacy was observed when calculations were conducted using geometric mean data.

The third dose confirmation study was a non-terminal study: the primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, evaluated via faecal samples collected on SDs 12-16, following administration of the VMP on Day 0 and administration of an authorised dewormer on Day 12. For the primary efficacy parameter, effectiveness calculations conducted using Abbott's formula determined a 100% efficacy both via geometric and arithmetic

means. Secondary efficacy parameters included fecal egg counts conducted over SDs 10-12 (100% efficacy observed), ELISA testing (all animals in the control group testing positive between SDs 10-12 and all animals in the VMP group testing negative over the same time period) and PCR testing (9 control animals testing positive between SDs 10-12 and 10 animals in the VMP group testing negative over the same period).

Whilst an inadequate level of efficacy was observed for one dose confirmation study (85.8%), a claim for efficacy for adult *A. caninum* can be accepted given the totality of the data provided: including, the findings of the other dose confirmation studies (≥ 99.3% efficacy observed), the fact that efficacy against *A. caninum* has been investigated in field studies and, for pyrantel, a well-established use claim can be accepted for the target species, the dose rate and the indication for adult *A. caninum*.

L4 and L5 larval claims

In support of the indication for the L4 and L5 larval stages of *A. caninum*, the applicant has provided four dose confirmation studies which were well designed and largely compliant with the relevant guidance documents. The formulation evaluated was the final formulation for marketing. All studies involved induced infections with one of the experimental isolates originating from naturally occurring infections in Europe less than 10 years prior to study commencement, and the others originating from the US in 2019 and 2021. This is considered in line with VICH GL19 which specifies that 'If both studies are conducted using experimentally infected animals, then parasites must have originated from naturally occurring infections from different geographical regions no older than 10 years prior to use for inducing infection'. The inoculate volumes were 200-300 *A. caninum* L3 larvae, which is in line with VICH GL19.

The studies were negatively controlled. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses within the recommended dose range (dose rates ranged from 10.00-19.231 mg fluralaner, 0.025-0.048 mg moxidectin and 5.00-9.62 mg pyrantel per kg bw). For the L4 claim, treatments were administered 7 days post-inoculation, which is acceptable in accordance with VICH GL19. For the L5 claim, treatments were administered 11 days post-inoculation. While no recommendations are provided in VICH GL19 on the recommended time of treatment after infection for L5 evaluation, day 11 can be accepted as reasonable timing of treatment given that the pre-patent period is stated to be 15-26 days and the recommended treatment after infection is for L4 6-8 days and for adults >21 days, respectively according to GL CVMP/VICH/835/99-FINAL.

The primary efficacy parameter was the reduction in worm burden for the L4 and L5 larval stages of *A. caninum* in the VMP group compared to control, as observed at necropsy on days 7 and 11 post-treatment, respectively. All animals in the control groups could be considered adequately infected based on total worm count at necropsy. When efficacy calculations for the primary efficacy parameter were conducted using Abbott's formula and geometric mean data, the percentage reduction for the formulation compared to control was calculated as ≥93.1% for both the L4 and L5 larval stages. In another study the primary endpoint was the reduction in *A. caninum* worm burden in the VMP treated group compared to the control group between SD 23-26, following administration of the VMP on Day 7 (L4) or Day 11 (L5) and administration of an authorised anthelmintic on Day 23. Whilst a non-terminal study design is not typically considered appropriate for a dose confirmation study, it is noted that the applicant sought advice from the CVMP regarding the acceptability of this approach. CVMP agreed with the proposed study design considering that potential false negative results of diagnostic methods like FECR test and counting of expelled worms in faeces, due to intermittent egg shedding, low egg shedding levels and undetectable prepatent stages, are compensated by the use of repeated molecular methods (PCR, ELISA) and analysis of FEC from at least 3 consecutive days. CVMP advised that the criterion to decide on sufficient efficacy (>90%) should be clearly pre-specified and that

applicability of the study design for the assessment of other stages or nematodes was demonstrated by the applicant. Given that these conditions have been fulfilled, the study design can be considered appropriate.

Given that timing of treatment appears appropriate for targeting L4 and L5 stages, the CVMP can accept that an adequate level of efficacy was observed to support the indications for L4 and L5 *A. caninum*.

Toxocara canis

Adult claim

In support of the indication for adult *T. canis*, the applicant has presented three dose confirmation studies which were well designed and largely compliant with the relevant guidance documents. The formulation evaluated was the final formulation for marketing. Two of the studies involved induced infections with isolates originating from the US, whilst the third study involved naturally infected animals in Europe. For the induced infection studies, inoculate volumes were 250 *T. canis* L3 larvae, which is in line with VICH GL19.

The studies were negatively controlled. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses close to the minimum of the recommended dose range (dose rates ranged from 10.135-14.423 mg fluralaner, 0.025-0.036 mg moxidectin and 5.068-7.212 mg pyrantel per kg bw). For the studies in which infections were induced, treatment was administered 50 days post-inoculation, which is consistent with VICH GL19.

For the two induced infection studies, the primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, as observed subsequent to necropsy on study day 7. For one of these studies, adequate infections were observed in 9 control animals and when efficacy calculations for the primary efficacy parameter were conducted using Abbott's formula and geometric mean data, the percentage reduction for the formulation compared to control was calculated as 92.6%. However, for the other study an inadequate level of infection (only four of ten control animals were adequately infected) was observed and therefore efficacy calculations could not be conducted.

The third study was a non-terminal study involving naturally infected animals. In this study, the primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, evaluated via faecal samples collected on study days 10-12, following administration of the VMP on Day 0 and administration of an authorised dewormer on study day 12. For the primary efficacy parameter, effectiveness calculations conducted using Abbott's formula determined a 97.49% efficacy via geometric means, with 99.29% efficacy calculated via arithmetic means. For the secondary efficacy parameters, 99.32% efficacy was observed for faecal egg count and a statistically significant difference observed with regards the number of control animals testing negative (44.1%) for roundworms on ELISA compared to the number of VMP-treated animals testing negative (88.2%).

Given that adequate levels of efficacy (>90%) were observed in two dose confirmation studies, one involving induced infection and the other naturally infected animals, the dose confirmation data presented can be considered adequate to support the indication for adult *T. canis.*

L5 larval claim

For the L5 larval claim for *T. canis*, the applicant has presented three dose confirmation studies, which were well designed and largely compliant with the relevant guidance documents. The formulation evaluated was the final formulation for marketing.

One study involved an induced infection, with isolates originating from the USA in 2016; the inoculate volume was 250 *T. canis* L3 larvae which is in line with VICH GL19. The study was negatively

controlled. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses within the recommended dose range (dose rates ranged from 10.4167-17.8571 mg fluralaner, 0.026-0.0446 mg moxidectin and 5.21-8.93 mg pyrantel per kg bw). Treatment was administered 24 days post-inoculation, and whilst no guidance is provided in VICH GL19, given the pre-patent period, timing of treatment is considered acceptable in terms of evaluating efficacy for L5 *T. canis*. The primary efficacy parameter was the reduction in worm burden in the VMP group compared to control evaluated at necropsy on day 26 after treatment. Effectiveness calculations conducted using Abbotts formula determined 94.8% efficacy via geometric means. Given that the threshold for efficacy (90%) was reached, this study can be considered to support the claimed indication for L5 *T. canis*.

Another study involved an induced infection, with isolates originating from Italy in 2016; the inoculate volume was 300 *T. canis* L3 larvae. The study was negatively controlled. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses close to the minimum of the recommended dose range (dose rates ranged from 10.64-15.15 mg fluralaner/kg bw, 0.027-0.039 mg moxidectin/kg bw and 5.32-7.58 mg pyrantel/kg bw). Treatment was administered 21 days post-inoculation. Timing of treatment is considered acceptable in terms of evaluating efficacy for L5 *T. canis*. The primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, evaluated via faecal samples collected on study days 56-60 following administration of the VMP on Day 0 and administration of an authorised dewormer on study day 56. Effectiveness calculations conducted using Abbott's formula determined 79% efficacy via geometric means. Given that the threshold for efficacy (90%) was not reached, this study cannot be considered to support the claimed indication for L5 *T. canis*.

For the third study, which involved naturally infected animals, the primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, evaluated via faecal samples collected on study days 10-12, following administration of the VMP on Day 0 and administration of an authorised dewormer on study day 12. For the primary efficacy parameter, effectiveness calculations conducted using Abbotts formula determined a 96.29% efficacy; however, it is noted that only 2 of the control animals were observed to be adequately infected with L5 *T. canis* larvae and therefore this study cannot be considered supportive of the claimed indication for L5 *T. canis.*

Given that >90% efficacy was only confirmed in one of three dose confirmation studies, the data presented are not considered adequate to support an indication for L5 *T. canis.* The applicant has withdrawn the indication from the product information as a result.

Toxascaris leonina

The applicant has presented one non-terminal, dose confirmation study in support of the indication for *T. leonina*. The study was conducted in line with GCP and the relevant guidelines, VICH GL9, VICH GL7 and VICH GL19. The infection was induced with isolates collected in Europe, and therefore can be considered representative of the current EU situation. The inoculate volume was 1500-2000 *T. leonina* infective larvated eggs which is acceptable in line with VICH GL19. The study was negatively controlled, with the VMP group administered the final formulation of the VMP. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses close to the minimum of the recommended dose range (dose rates ranged from 10.00-12.38 mg fluralaner, 0.025-0.031 mg moxidectin and 5.00-6.19 mg pyrantel per kg bw). Treatment was administered 77 days post-inoculation, which is acceptable given that VICH GL19 specifies a 70 day interval.

The primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, evaluated via faecal samples collected on SDs 91-93, following administration of the VMP on

Day 0 and administration of an authorised dewormer on Day 91. Given that 8 animals in the control group were considered adequately infected with a mean worm count of 12.9, infection can be considered adequate. Effectiveness calculations conducted using Abbott's formula determined 99.4% efficacy via geometric means and 99.6% efficacy via arithmetic means. Secondary efficacy parameters included faecal egg count reduction (100% efficacy observed), ELISA testing (9/10 animals in the control group and 1/10 animals in the VMP group testing positive between SD 88-90) and PCR testing (all of the control animals testing positive for *T. leonina* DNA and 1/10 animals in the VMP group testing positive between SD 88-90).

The results of this study demonstrate an acceptable level of efficacy (≥90%) against adult *T. leonina* with an adequately infected control group and a statistically significant difference observed and therefore, can be considered supportive of the indication for adult *T. leonina.*

Whilst only a single dose confirmation study has been provided in support of the indication for adult *T. leonina*, the CVMP notes that: it was a well conducted study in accordance with guideline recommendations; efficacy is also supported by the results of a pivotal field trial conducted within the EU; and, for pyrantel, a well-established use claim can be accepted for the target species, the dose rate and the indication for adult *T. leonina.* Given the totality of the data available, the CVMP considers the indication for adult *T. leonina* adequately supported.

Uncinaria stenocephala

The applicant has presented two non-terminal, dose confirmation studies in support of the indication for *U. stenocephala*.

Both studies were conducted in line with GCP and the relevant guidelines, VICH GL9, VICH GL7 and VICH GL19. In both cases, infections were induced with recently collected EU isolates and therefore the infections can be considered representative of the current EU situation. The inoculate volumes were 1250 third stage *U. stenocephala* larvae which is acceptable in line with VICH GL19. Both studies were negatively controlled, with the VMP group administered the confirmed final formulation of the VMP. With regards housing, as for studies described earlier in this AR, the animals were individually housed for the study duration, which is not in line with EMEA/CVMP/EWP/005/2000 Rev.4.

With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses evaluated close to the minimum of the recommended dose range (dose rates ranged from 10.14-11.72 mg fluralaner, 0.025-0.029 mg moxidectin and 5.07-5.86 mg pyrantel/kg bw). The first treatments were administered 25 days post-inoculation, which is consistent with VICH GL19 specification.

The primary efficacy parameter was the reduction in worm burden in the VMP group compared to control, evaluated via faecal samples collected on SDs 12-15, following administration of the VMP on Day 0 and administration of an authorised dewormer on Day 12. In accordance with guideline specification, at least 6 control animals in each group were determined to be adequately infected. Effectiveness calculations conducted using Abbotts formula, determined a ≥99.8% efficacy using both geometric and arithmetic mean data. Efficacy is further supported by the secondary efficacy parameters including fecal egg counts conducted over study days 10-11 or 10-12 (with a ≥99.8% efficacy observed), ELISA testing (9-10 animals in the control group testing positive between SD 10-12 and 9-10 animals in the VMP group testing negative over the same time period) and PCR testing (at least 7 control animals testing positive between SDs 10-12 and at least 7 VMP animals testing negative over the same period).

The results of these studies demonstrate an acceptable level of efficacy (≥99.8%) against adult

U. stenocephala, and the applicant has been provided suitable justification for the omission of a terminal dose confirmation study from the efficacy data provided in support of the indication for treatment of *U. stenocephala.*

Dirofilaria immitis

The applicant has presented two dose confirmation studies in support of the indication for *D. immitis.*

Both studies were conducted in line with GCP and the relevant guidelines, VICH GL9, VICH GL7 and VICH GL19. Although both studies were induced with US isolates of *D. immitis*, the applicant conducted an *in vitro* analysis to compare the genotypic susceptibility/resistance status of the US isolates used for the induction of infection in the dose confirmation studies presented with isolates collected at random across Europe and consequently it was concluded that macrocyclic lactone resistance in *D. immitis* is not prevalent in Europe currently and the data derived from the dose confirmation studies presented can be extrapolated to support efficacy against European isolates.

Both studies were negatively controlled. With regards treatment administration, the methods and dose rates were in accordance with the SPC, with doses evaluated close to the minimum of the recommended dose range (ranging from 10.0-13.158 mg fluralaner, 0.025-0.031 mg moxidectin and 5.0-6.579 mg pyrantel per kg bw). The first treatments were administered 30 days post-inoculation, consistent with VICH GL19 specification and depending on study group treatments were repeated at thirty-day intervals thereafter.

For both studies, the primary efficacy parameter was the number of adult *D. immitis* worms recovered at necropsy compared to the control. Necropsy was conducted on study days 118 and 122 and given the lifecycle of the parasite (with reported molting to fifth stage larvae within 50-70 days of infection, migration to the pulmonary artery within 75-120 days post-infection), this is considered acceptable. Serology for heartworm antigen and microfilaria was also conducted prior to enrolment and again on study days 89 and 90. Given that positive results for canine antigen and circulating microfilaria would not be expected until at least 5 and 6 months post-infection respectively, this timing was considered appropriate.

At necropsy all animals in the control groups for both studies were considered adequately infected. Effectiveness calculations conducted using Abbott's formula and geometric mean data, determined efficacy of ≥96.6%, whilst for arithmetic means, ≥96% efficacy was observed.

Given the acceptable levels of efficacy observed (≥96.6%) against *D. immitis* infections, the studies can be considered supportive of the indication.

Angiostrongylus vasorum

The applicant has presented two pre-clinical studies in support of the indication for *A. vasorum.* The first of these studies is discussed in more detail under dose determination.

The second study was a well-designed GCP compliant blinded randomised negatively controlled non-terminal efficacy study. The formulation (the VMP) was confirmed as the final formulation. *A. vasorum* infection was artificially induced with isolates collected in Denmark in 2012, the inoculate volume was 250 infective larvae with all the control animals considered to be adequately infected. The VMP was administered in accordance with the SPC, with treatment administered 28 days post-inoculation, to allow for evaluation of efficacy against the L5 larval stage and adult stages). Animals received 10.42 – 12.82 mg / kg fluralaner, 0.026 – 0.032 mg / kg moxidectin and 5.21 – 6.41 mg / kg pyrantel.

The primary efficacy parameter was reduction of faecal larval counts in the treated groups compared

to the control. The applicant additionally conducted serology for the detection of *A. vasorum* antigen and antibody and respiratory assessments (all of which were observed to correlate with necropsy findings in the pivotal dose determination study for moxidectin). With the inclusion of computed tomography (CT) findings, the efficacy parameters were considered appropriate both clinically and in terms of fulfilling guideline requirements. For evaluation of the primary efficacy parameter, faecal samples were collected three times weekly from Day 28 post-treatment (that is 56 days post-inoculation) and larval counts conducted. Efficacy calculations were conducted using Abbott's formula based on geometric mean data, in accordance with VICH GL7, with percentage efficacy based on geometric mean data calculated as 99.98%. Percentage efficacy based on arithmetic means was 100%.

The findings derived from the secondary efficacy parameters observed supported the efficacy findings of the primary endpoint. For respiratory parameters, increased respiratory rates and an increased incidence of abnormal findings were observed in the control group compared to the VMP group. With regards CT findings, pathologies were observed in all study groups prior to Day 0, however following treatment, improvements were observed in the VMP group progressing towards 'normal' and 'mild' scoring between SDs 54-61. Finally, for serology, lower antibody titers and antigen levels were observed in the VMP group compared to the control group, again, reflective of product efficacy for the prevention of larvae development into adult worms. With regards safety, whilst diarrhoea was observed it is noted that this has been included under SPC section 3.6 as a common AE.

In conclusion, the results of this study appear to demonstrate an acceptable level of efficacy (>90%) against *A. vasorum* infections when the product is administered as provided in the product information.

Reduction of the risk of flea- and tick-borne disease transmission

Babesia canis

Separate pre-clinical data has not been provided in support of the indication for *B. canis* transmission by *D. reticulatus*. Instead, the basis upon which the indication for *B. canis* has been justified by the applicant is that prior to transmission of *B. canis* sporozoites by *D. reticulatus*, an approximate period of feeding of 24-48 hours is required and therefore, provided the product exerts a speed of kill of <24 hours, the risk of transmission of *B. canis* will be reduced.

In support of the indication, the applicant has presented the results of a speed of kill study for the vector *D. reticulatus*, which has been discussed in more detail under the 'dose determination' section of the scientific overview. It is accepted that this speed of kill study demonstrated >90% efficacy for *D. reticulatus* at 12 hours post-treatment and at 24 hours post-infestation on study days 7, 14, 21 and 30 following administration of a 10 mg fluralaner/kg bw dose.

Regarding reduction of the risk of infection with *Babesia canis*, no separate (pre-)clinical data were submitted in the current procedure. However, pharmacodynamics of all fluralaner-containing products (tablets, spot-on or injectable) are identical and a reduction of the risk for infection with *Babesia canis* has previously been demonstrated via proprietary study data for several fluralaner-containing products. Noting that the indication for the reduction of risk of transmission of *B. canis* has previously been accepted for other fluralaner-containing products, that fluralaner's efficacy relative to the indication is against the vector *D. reticulatus*, that an approximate period of feeding of 24-48 hours is required for *D. reticulatus* prior to transmission of *B. canis* sporozoites and that >90% efficacy against *D. reticulatus* is achieved within 24 hours, the above data is considered adequate to support the indication for 'reduction of risk' for *B. canis* transmission when fluralaner is administered at a dose rate of 10 mg/kg.

Dipylidium caninum

In support of the indication for *D. caninum* transmission by *C. felis,* the applicant has provided the results of two speed of kill studies for *C. felis*.

The first speed of kill study was a well-designed and GCP compliant study with 40 dogs administered either a 10 mg fluralaner/kg bw (non-final formulation – mono-active substance) dose on study day 0 or were untreated, with all animals subsequently infested with 100 adult newly emerged unfed *C. felis* fleas on study days 1, 7, 14, 21, 28, 35, 42 and 49, and flea counts conducted at 12 and 24 hours post-infestation. Adequate infestations of the control animals were observed and for the treated animals an efficacy level ≥99.2% was observed for *C. felis* at 12 hours post-treatment up until SD 35 and 100% efficacy observed at 24 hours up until SD 35.

A second study, discussed in more detail under dose confirmation, evaluated speed of kill for *C. felis* at 12 hours, again following administration of 10 mg fluralaner/kg bw (final formulation). In that study, an acceptable level of efficacy (>95%) was observed against existing infestations of *C. felis* at 12 hours and against new infestations of *C. felis* at 12 hours up to 7 days post-treatment only (from study day 14, efficacy at 12 hours post-infestation was below threshold) and on D30 efficacy 12h p.i. was decreased to 56.2%).

The omission of a study specifically evaluating efficacy against *D. caninum* infected isolates of *C. felis* was considered justified by the applicant on the basis that the flea must be exposed to the target species' (dog) body heat for a period of 24-36 hours, to allow for development of the cestode into its infective stage, and the CVMP has previously accepted that 24-hour speed of kill data for *C. felis* can be considered adequate to support a claim for *D. caninum*, with transmission time typically occurring after this timepoint.

Further supportive of the acceptability of the indication, in spite of the omission of specific studies for *D. caninum*, is the fact that pharmacodynamics of all fluralaner-containing products (tablets, spot-on or injectable) are identical and a reduction of the risk for infection with *D. caninum* has previously been demonstrated via proprietary study data for several fluralaner-containing products. Noting that the indication for the reduction of risk of transmission of *D. caninum* has previously been accepted for other fluralaner-containing products, that fluralaner's efficacy relative to the indication is against the vector *C. felis*, that the period of time for which *C. felis* must be exposed to the host in order for *D. caninum* to reach an infective stage is 24-36 hours and that >95% efficacy against *C. felis* is achieved within 24 hours, the data provided is considered adequate to support the indication for 'reduction of risk' for *D. caninum* transmission when the formulation is administered in accordance with the SPC.

Tolerance in the target animal species

In support of tolerance of the veterinary medicinal product in the target animal species (dogs), the applicant has submitted two pivotal and one pilot (or exploratory) target animal safety studies. The target animal tolerance data recorded in the clinical studies presented with this dossier are also taken into account in the assessment of target animal safety.

In a GLP-compliant pivotal target animal safety study conducted in accordance with relevant guidance (VICH GL 43), the product was administered to 8-week-old Beagle dog puppies at 1X, 3X and 5X the high end of the RTD (that is 20 mg fluralaner / 0.05 mg moxidectin / 10 mg pyrantel per kg BW). A control group was dosed with reverse osmosis water. The dosing was performed in the fed state and

was repeated 7 times at monthly intervals. A monthly interval is consistent with the proposed recommended retreatment interval for certain parasites. No treatment-related adverse effects were observed in this pivotal target animal safety study. Two unscheduled euthanasias were performed, in both cases, the cause of morbidity was concluded to be unrelated to administration of the product. In male dogs receiving 3X and 5X the RTD, organ weight changes that were outside the concurrent control group range (higher weight for the spleen and lower weight for the pituitary) were reported, however no correlating clinical or histopathologic findings were associated with this observation in these dogs.

Tolerance of the VMP has not been tested in pregnant, lactating or breeding animals. The text in section 3.7 of the SPC reflects this information, and Bravecto TriUNO is not recommended for use in pregnant, lactating or breeding animals. Section 3.7 of the SPC contains the following statement relating to fetotoxic and teratogenic effects observed when moxidectin was administered to laboratory animals "*Laboratory studies with moxidectin in rats and mice have shown evidence of fetotoxic and teratogenic effects*". It was noted that an increase in fusions in cervical vertebra 2 at 25 mg/kg bw per day was observed in rabbits in a previous study evaluating oral fluralaner administration. However, based on the totality of information available for this active substance, and that Bravecto TriUNO is not recommended for use in pregnant or lactating animals or those intended for breeding, the CVMP agreed that a specific statement relating to safety of fluralaner in such animals was not warranted.

As the VMP contains the macrocyclic lactone moxidectin, one pilot and one pivotal target animal tolerance study were performed in ivermectin-sensitive (MDR1 (-/-)) collie dogs with a focus on assessment of neurological clinical signs. The results of the pilot study (which was non-GLP compliant and unblinded and uncontrolled) indicate that when administered at 5X RTD, the product caused mild to moderate neurological signs in this population of dogs.

In the pivotal study the product was administered to adult ivermectin sensitive (MDR1 (-/-) dogs at 1X, 3X and 5X the high end of the RTD (that is 20 mg fluralaner / 0.05 mg moxidectin / 10 mg pyrantel per kg BW). A control group was dosed with tap water. The dosing was performed in the fed state and was performed once. Mild depression was observed in animals in all treatment groups with an apparent dose-dependent effect (increased incidence, not severity, of the clinical sign with increasing dose). Emesis (at 3X and 5X) and muscle fasciculations (at 5X) were also observed within 24h of dosing. All adverse events were mild and self-limiting, and resolved without concomitant treatment. Since tolerance in a sensitive subpopulation was only investigated after single dosing, i.e. before attainment of steady state conditions, relevant information concerning use of the VMP in this susceptible sub-population has been included in sections 3.5 and 3.10 of the SPC.

In these target animal safety studies, dogs were single housed (without necessity and/or without adequate enrichment/compensation) and in cages that were smaller than stipulated in the relevant Directive 2010/63/EU, or inadequately equipped.

The following observations from the clinical data submitted with this dossier are also noted:

Both diarrhoea and vomiting were observed following administration of the VMP in the dose confirmation and clinical field trials presented, at incidence rates consistent with their inclusion as 'common' adverse events.

In the clinical field trial, 3 animals experienced convulsions/seizures (2 of which were subsequently euthanized), that were reported as serious adverse events, whilst 2 additional dogs experienced convulsions which were recorded as non-serious adverse events. It is also noted, and considered relevant, that in two other clinical field trials either households which included an animal with epilepsy, or dogs with epilepsy were excluded from these trials. It cannot therefore be ruled out that

these exclusion criteria may have resulted in an under-reporting of potential neurological adverse events in the study populations. Taking into account these points an adverse event term and frequency descriptor ('convulsion', very rare) were included in the SPC to capture this potentially serious adverse event in section 3.6.

In order to report expected adverse events and their frequency in the SPC, the applicant refers to post-marketing pharmacovigilance data concerning a fluralaner-containing VMP that is administered at the same dose as that proposed for the VMP, that has been marketed in other territories. The applicant also refers to a number of dose confirmation and field studies conducted in Australia performed with the formulation that were not used in determining efficacy for the product. Based on these data, the adverse events 'muscle tremors' and 'ataxia' are reported as occurring very rarely, and very rare is also a frequency descriptor for 'convulsions.' In reference to another fluralaner-containing VMP, an appropriate warning statement regarding the use of the product in dogs with neurological disorders (and pre-existing epilepsy) in section 3.5 of the SPC has been included. The adverse event 'decreased appetite' is also included with the frequency descriptor 'uncommon.'

Concerning the reporting of adverse events in section 3.6 of the SPC, the adverse events and frequency descriptors are considered accurate.

Suitable precautions have been included in section 3.5 of the SPC concerning target animal safety; including information concerning use in MDR1 -/- dogs. The symptoms of overdose generated in ivermectin-sensitive MDR1 -/- collie dogs are reflected in SPC section 3.10.

It is accepted that when used in accordance with the SPC, the VMP will not present an unacceptable risk to the target animal species, dogs.

Clinical trial(s)

The applicant has presented the results of nine clinical trials in support of the application, which have been summarised below.

Field studies for fleas and ticks

In support of efficacy for the flea and tick species, *C. felis, C. canis, D. reticulatus, I. hexagonus, I. ricinus* and *R. sanguineus*, the applicant has presented the results of five clinical trials, one conducted across multiple sites in the EU and the other four conducted in Brazil. The studies were largely in line with the relevant guidelines and consequently are considered acceptable to evaluate efficacy. Whilst the tick and flea strains evaluated in the Brazilian studies may not be fully representative of those circulating in the EU, based on *in vitro* data presented, it can be accepted that data derived from isolates collected in the USA and Australia could be representative of the EU strains and therefore, a similar approach is considered reasonable for this scenario. Furthermore, given that the Brazilian studies evaluate efficacy in a different geographical and climatic region than the field studies conducted in the EU and they can be considered supportive.

The pivotal EU study was conducted at multiple veterinary practices in the EU and consequently, the results can be considered reflective of expected efficacy within the European community. The study was positively controlled (sarolaner/moxidectin/pyrantel). The VMP, which was the final formulation for marketing, was administered in accordance with bodyweight and the dosing table provided in the product information.

With regards to the study animals, sample size was justified with 651 dogs encompassing a broad range of breeds, weights and ages enrolled. In accordance with guideline specification, dogs of short, moderate and long-hair length were included as well as hunting dog breeds. Dogs with epilepsy were excluded from the study (a known adverse effect of fluralaner) and a suitable warning

has been included in the SPC.

With regards tick and flea counts, an inclusion criterion was 4 ticks/5 fleas per primary dog. All four tick species proposed as indications for this product and *C. felis* were observed at enrolment. It is noted that a high incidence of *R. sanguineus*, the dose-limiting tick species, was observed. Tick/flea counts were conducted on SD 0, 7 and 31, which deviates from the aforementioned guideline that tick/flea counts should be conducted at weekly or two-weekly intervals, respectively. However, in the dose confirmation studies presented with this application and in the four other field trials conducted in Brazil, evaluation of efficacy was conducted at weekly intervals and therefore the data derived from those studies can be considered to support efficacy evaluated on a weekly basis.

Primary efficacy was based on the percentage of primary dogs observed to be free of ticks and/or fleas at SD 31, with a non-inferiority versus the control product (sarolaner/moxidectin/pyrantel) analysed. It is noted that 98.3% of dogs in the VMP group were observed to be tick free and 97.6% of dogs in the VMP group were observed to be flea-free at SD31 and calculations conducted in accordance with the statistical guideline demonstrated significant non-inferiority with the control product.

Secondary efficacy calculations (percentage reduction) were conducted using Abbott's formula with calculations conducted via geometric means. Calculations using arithmetic mean data were additionally provided. In this study, a ≥99.7% reduction in tick burden and a 99.6% reduction in flea burden was observed.

In line with guideline EMEA/CVMP/EWP/005/2000 Rev.4 and to support a claim for FAD, the applicant has evaluated the incidence of FAD in the population of animals included in this study and their response to treatment. It is noted that all animals exhibiting FAD lesions prior to treatment and which were subsequently administered the VMP demonstrated a recovery of skin lesions by Day 31. Given the efficacy observed for the treatment of flea infestations and the recovery observed in animals affected by FAD, these data can be considered supportive of the claim for FAD. With regards safety observations, it is noted that both emesis and hypersalivation were observed in animals administered the VMP, consistent with the AEs detailed under SPC section 3.6.

The results of this study demonstrate an acceptable level of efficacy for the treatment of infestations with the various tick species claimed for the product and also for *C. felis* and by extrapolation, *C. canis.* The applicant aimed to demonstrate non-inferiority with a control product authorised for the same indications for ticks and fleas, with significant non-inferiority confirmed. Consequently, this study is considered supportive of the claims for the treatment of *Dermacentor reticulatus, Ixodes hexagonus, I. ricinus, Rhipicephalus sanguineus, Ctenocephalides felis* and *C. canis* for 1 month.

Study data was provided for two additional clinical field studies conducted in Brazil for *R. sanguineus* and two for *C. felis*.

These were single-arm studies, with all animals administered the VMP (final formulation for marketing). Thirty-two to thirty-three animals were included in each study and a range of weights and ages represented. The VMP was administered in accordance with bodyweight and the dosing table provided in the product information.

For *R. sanguineus*, the primary efficacy parameter was the percentage reduction in tick count at 7, 14, 21 and 30 days post-treatment, with baseline values taken on study days -2 and 0 serving as control. Baseline tick counts were considered adequate. Effectiveness calculations conducted using Abbotts formula determined ≥90.21% efficacy up until 30 days post-treatment, with statistical significance observed. These results demonstrate an acceptable level of acaricidal efficacy (>90%) for the treatment and control of *R. sanguineus* infestations from SD 7-30, in line with the proposed

duration of efficacy for the product.

For *C. felis,* the primary efficacy parameter was the percentage reduction in flea count at 7, 14, 21 and 30 days post-treatment, with baseline values taken on study days -2 and 0 serving as control. Baseline flea counts were considered adequate. Effectiveness calculations conducted using Abbott's formula determined 100% efficacy up until 30 days post-treatment, with statistical significance observed. These results demonstrate an acceptable level of insecticidal efficacy (>95%) for the treatment of *C. felis* infestations from SD 7-30. Due to the shortcomings of the study (single-arm study without control, outside of the EU) the results provide supplementary information.

Field studies for nematodes

In support of efficacy for the gastrointestinal nematodes, *Toxocara canis, Toxascaris leonina, Ancylostoma caninum* and *Uncinaria stenocephala,* the applicant has presented the results of three clinical field trials.

A pivotal EU multi-site study was conducted. With regards to the study animals, sample size was justified with 429 dogs encompassing a broad range of breeds, weights and ages included. It is noted that dogs with epilepsy were excluded from the study (a known adverse effect of fluralaner) and an appropriate warning statement regarding the use of the product in dogs with neurological disorders (and pre-existing epilepsy) in section 3.5 of the SPC has been included.

The study was positively controlled (afoxolaner/milbemycin oxime)) and the formulation evaluated was the final formulation for marketing. The VMP was administered in accordance with bodyweight and the dosing table provided in the product information.

The primary efficacy parameter was the percentage reduction in faecal egg count for each nematode species at 14 days post-treatment, with baseline values taken between SDs -7 and 0 serving as control. At baseline, adequate infestations were observed for *T. canis, T. leonina* and hookworm. Whilst the objective of the study was also to evaluate efficacy for *A. caninum* and *U. stenocephala*, inadequate numbers of animals had positive faecal egg counts for these nematode species at baseline (hookworm eggs could be differentiated in very few dogs due to overlapping egg sizes), and therefore efficacy calculations could not be considered reliable for the respective hookworm species. However, it can be assumed that the study results are valid (due to a total of 110 hookworm-positive dogs at baseline) and therefore efficacy against hookworms can also be confirmed. Effectiveness calculations conducted using Abbott's formula with geometric mean data determined ≥99.1% efficacy for *T. canis, T. leonina* and hookworm. Efficacy calculations were also conducted using arithmetic mean data with ≥95.46% efficacy observed for *T. canis, T. leonina* and hookworm. A secondary efficacy parameter was the percentage of nematode-free dogs compared to the positive control, with significant non-inferiority observed for the formulation compared to control.

It was concluded that the results of this study demonstrate an acceptable level of efficacy (>90%) against *T. canis*, *T. leonina* and hookworms.

Study data was provided for two additional clinical field studies conducted in Brazil. Whilst the nematode strains evaluated in the Brazilian studies may not be representative of those circulating in the EU, the results derived from these studies can be nonetheless considered supportive of the EU field trial data presented.

These were single-arm studies, with all animals administered the VMP (final formulation for marketing). Forty animals were included in each study and a range of weights and ages represented. The VMP was administered in accordance with bodyweight and the dosing table provided in the product information.

The primary efficacy parameter was the percentage reduction in faecal egg count for each nematode species at 7- and 14-days post-treatment, with baseline values taken on study days -2 and 0 serving as control. It is noted that baseline egg counts for *Ancylostoma, Toxocara, Toxascaris* were considered adequate. Effectiveness calculations conducted using Abbott's formula determined 100% efficacy.

The results of these studies were considered to demonstrate an acceptable level of efficacy (>90%) against *Ancylostoma, Toxascaris* and *Toxocara* spp., with adequately infected control groups and statistically significant differences observed. Therefore, these studies were considered supportive of indications for the treatment of infections with the above parasites.

Field study for *Dirofilaria immitis*

In support of the indication for *D. immitis,* the applicant submitted the results of one GCP compliant clinical field trial, conducted largely in line with the relevant guidelines: VICH GL9, VICH GL7 and VICH GL19. The study was conducted across 21 veterinary practices in the USA; whilst not directly representative of the EU situation, the applicant provided *in vitro* data evaluating susceptibility/resistance patterns in *D. immitis* in Europe and, based upon that data provided, the results derived from this study can be considered reflective of the EU situation.

The study included two groups, one administered the VMP (final formulation for marketing) and the other a positive control (sarolaner/moxidectin/pyrantel). With regards to the study animals, 263 dogs encompassing a broad range of breeds, weights and ages were included and considered well-representative of the target population. The VMP was administered in accordance with bodyweight and the dosing table provided in the product information, which is considered acceptable.

In respect of efficacy results, the primary efficacy parameter was based on heartworm antigen and circulating microfilaria at Day 300, with the intention of calculating non-inferiority in the case that animals in either the control or the VMP treated groups tested positive for antigen or microfilaria. However, positive test results were not observed in either the treated or the control group and therefore statistical analysis was not conducted, and it was concluded that the test article and the control were equally efficacious. Consequently, this study was considered supportive of the conclusions derived from the two dose confirmatory studies presented for *D. immitis,* that is an adequate level of efficacy has been presented to support the indication for *D. immitis*.

With regards safety, the study was of a prolonged duration and consequently a large number of AEs were observed. Whilst some of those which were observed at high incidences in the VMP are known adverse effects of the active substances and are included under section 3.6 of the SPC, others which were observed at high incidences (dermatitis, eczema and otitis) were also observed in the control group and are unlikely associated with the product. However, 3 animals experienced convulsions/seizures (2 of which were subsequently euthanized) that were reported as serious AEs. Two other dogs experienced convulsions which were recorded as non-serious adverse events. Given that neurological signs such as seizures are known AEs of the active substance fluralaner, convulsions have been included in the SPC as an adverse event.

Other studies

Palatability

The applicant has presented the results of palatability studies conducted as part of three field studies presented with this application.

For two of these studies, the same general conditions were followed: the study animal was offered the tablet on the hand/on the floor/in an empty food bowl. After intake or refusal, palatability was

scored, largely in line with the scoring system described in the Guideline on the demonstration of palatability of veterinary medicinal products. Percentage palatability was calculated as: No of animals with a palatability score of 1/Total no. of animals in the study group x 100. A palatability score of 1 was defined by the applicant as 'voluntary uptake within 5 minutes'. Based on the study results, 82.6-89.5% of animals scored 1. For these studies, treatment was administered on a single occasion. However, these data cannot be considered fully supportive of the claim given that in the aforementioned guideline, acceptance is defined as '*voluntary full consumption within the maximum offering time (e.g. two minutes)*' and raw data demonstrative of a consumption time of less than 2 minutes for 80% of the study population was not presented. Suitable justification for deviation from the protocol described in the guideline based on species and pharmaceutical form has not been provided.

For the other study presented, the same general conditions applied as for the previous studies, however, treatments were administered on 12 occasions on a monthly basis, which is compliant with the palatability guideline specification that for products intended for administration at monthly intervals, palatability should be tested for at least two administrations per animal. When the treatment dose was taken freely, the owner recorded whether it was within 1 minute or 1 to 5 minutes and if the treatment dose was not taken freely within 5 minutes, the owner had the option of administering it in treats or food. The results of this study may be considered more robust to conclude on palatability given that it provides information on multiple administrations and time to ingestion was recorded. Based on the study results, 62.1% of animals took the tablet voluntarily within 1 minute, a further 4.4% within 5 minutes and 23.1% ingested the tablets in food or treats. Given that only 62.1% of doses were voluntarily ingested within 1 minute and an additional 4.4% within 5 minutes, the results derived do not meet the threshold to conclude palatability for the product. Whilst 23.1% of animals did consume the product voluntarily in food/treats, the guideline clearly states that palatability '*should be assessed without food to avoid any effect of palatability linked to the food composition'.*

It was proposed by the applicant that further data from 2 field studies conducted in Australia (that were not submitted in support of efficacy in the current application) are pooled with the data detailed above and considered in respect of palatability. However, the pooling of data (only from the initial dosing event) from multiple field studies conducted in the EU, Australia and the US (all of which involved administration of the VMP to more than 25 dogs, and 3 of which involved multiple treatments) is not considered an acceptable approach. It is concluded that the data provided are not adequate to support a palatability claim.

Overall conclusions on efficacy

Pharmacodynamics

The applicant has provided a comprehensive summary of the pharmacodynamics of the active substances fluralaner, moxidectin and pyrantel based on published literature. The main pharmacodynamic characteristics have been suitably described in the SPC.

Despite a theoretical potential for interaction between fluralaner, moxidectin and the general risk for interaction due to co-medication with pyrantel, the results of *in vivo* studies performed using the VMP provided no evidence of any clinically relevant interference of the combination regarding efficacy, pharmacokinetics, and safety.

Further detail (from proprietary studies) is also provided in respect of the mechanism of action of fluralaner.

Three *in vitro* studies concerning the efficacy of fluralaner against isolates of the flea species *Ctenocephalides felis* from three different geographical regions (the EU, the US and Australia) were performed. Taking these and *in vivo* laboratory and field efficacy data assessed in previous applications into account, it is accepted that there is currently no strong evidence to suggest that susceptibility of *C. felis* to fluralaner will differ based on geographical origin.

Three *in vitro* studies concerning the efficacy of fluralaner against isolates of the tick species *Rhipicephalus sanguineus* from two geographical regions (the EU and the US) were performed. Taking these and *in vivo* efficacy data assessed in previous applications into account, it is accepted that there is currently no strong evidence to suggest that susceptibility of *R. sanguineus* to fluralaner will differ based on geographical origin.

Pharmacokinetics

It is concluded that the applicant has presented a suitably thorough account of the pharmacokinetics of the active substances fluralaner, moxidectin and pyrantel. Furthermore, it is accepted that there are no data to suggest that interaction(s) between the active substances will occur and impact upon pharmacokinetics in the target animal species. The CVMP considers the information included in section 4.3 of the SPC to have been supported by the data presented.

Justification of the fixed combination

The combination of fluralaner, moxidectin and pyrantel in the VMP is considered to have been suitably justified in line with the requirements of current guidance.

Development of resistance and related risks to animals

The applicant has presented a reasonably comprehensive and current summary of knowledge concerning development of resistance to the active substances fluralaner, moxidectin and pyrantel, although limited information concerning the mechanisms of resistance has been provided. Concerning the risk of development of resistance in animals, it is considered notable that reports of resistance to both moxidectin (*Ancylostoma caninum and Dirofilaria immitis*) in the US, and to pyrantel (*A. caninum*) in the US and Australia have been published recently.

The applicant has included text in the SPC for the purposes of mitigating against the risk of resistance development arising from use of the VMP that is in accordance with current guidance. In the event that resistance to the active substances in relevant companion animal parasites in Europe is reliably reported in the future, the SPC will require updates at that time.

Dose determination

In support of dose determination for fluralaner, the applicant has provided the results of dose determination studies conducted with the American tick species, *A. americanum*, and with the EU tick species, *D. reticulatus.* The findings of these studies demonstrate 10 mg fluralaner/kg bw dose as the most effective for the longest duration (up to 30 days, the proposed between-treatment interval).

In support of dose determination for moxidectin, the applicant has provided the results of a dose determination study for *A. vasorum*. The results of the study concluded that 0.025 mg moxidectin/kg bw, was the minimum dose to be evaluated in dose confirmation studies.

In support of dose determination for pyrantel, the applicant claims that the active substance has been in well-established use for at least 10 years, efficacy is well documented and an acceptable level of safety is assured. Based on the arguments presented, the omission of specific dose determination data for pyrantel can be accepted.

Dose confirmation

The applicant has presented four dose confirmation studies in support of the indications for *Dermacentor reticulatus, Ixodes hexagonus, Ixodes ricinus and Rhipicephalus sanguineus.* All of the studies presented evaluated efficacy in *R. sanguineus.* However, the applicant has presented both proprietary and published data which demonstrate that *R. sanguineus* is the dose limiting EU tick species and therefore the dose confirmation data provided for *R. sanguineus* can be considered supportive of efficacy for *Dermacentor reticulatus, Ixodes hexagonus, I. ricinus and Rhipicephalus sanguineus.*

The applicant has presented five studies in support of the indications for *C. felis.* Efficacy against both *C. felis* and *C. canis* can be accepted based on these studies.

Nematodes

In support of the indication for *A. caninum,* the applicant has provided seven dose confirmation studies, which are considered to support the indication for L4, L5 and adult *A. caninum.* In support of the indication for *T. canis*, the applicant has provided five dose confirmation studies and whilst these are considered adequate to support efficacy for adult *T. canis*, the data provided is considered inadequate to support the L5 stage of *T. canis.* Only one dose confirmation study has been provided in support of the indication for adult *T. leonina*, however, given the clinical field data presented, bibliographic data demonstrative of efficacy for the active substance pyrantel against *T. leonina* across a broad range of geographical locations and the accepted well-established use for the active for this indication, in totality the data provided can be considered adequate to support the indication. Two dose confirmation studies have been provided in support of the indication for adult *U. stenocephala and* are considered to demonstrate efficacy against *U. stenocephala*.

Two dose confirmation studies have been provided in support of the indication for *D. immitis*, with the results observed considered adequate to support efficacy. The dose determination study for *A. vasorum* has been supplemented with a dose confirmation study for this nematode. The findings of both studies support the indication for *A. vasorum.*

Reduction of the risk of flea- and tick-borne disease transmission

In support of the indication for *B. canis*, the applicant has presented the results of a speed of kill study for the vector *D. reticulatus*, in which >90% efficacy for *D. reticulatus* was observed at 12 hours post-treatment and at 24 hours post-infestation. Given that an approximate period of feeding of 24-48 hours is required for *D. reticulatus* prior to transmission of *B. canis* sporozoites, it can be accepted that the risk of transmission of *B. canis* will be reduced following administration of the product.

In support of the indication for *D. caninum*, the applicant has presented the results of a speed of kill study for *C. felis* in which 100% efficacy was observed at 24 hours post-treatment up until SD 35 and this was supported by comparable results observed for two other dose confirmation studies presented. Noting that the period of time for which *C. felis* must be exposed to the host in order for *D. caninum* to reach an infective stage is 24-36 hours, it can be accepted that the risk of transmission of *D. caninum* will be reduced.

Tolerance in the target animal species

In support of tolerance of the veterinary medicinal product in the target animal species, dogs, the applicant has submitted two pivotal and one pilot (or exploratory) proprietary target animal safety

studies. The target animal tolerance data recorded in the clinical studies presented with this dossier are also taken into account in the assessment of target animal safety.

It is accepted that when used in accordance with the SPC that the VMP will not present an unacceptable risk to the target animal species, dogs.

Clinical trials

In support of this application, the applicant presented nine clinical field trials, five evaluating efficacy for ticks and/or fleas, three evaluating efficacy for gastrointestinal nematodes and one evaluating efficacy for heartworm. Whilst a clinical field trial for lungworm was not presented, the dose determination/confirmation data presented is considered adequate to support this claim.

Based upon the results of these studies, it can be accepted that Bravecto TriUNO is effective for dogs with, or at risk from, mixed parasitic infestations by ticks or fleas, gastrointestinal nematodes and/or heartworm, when administered orally at a dose rate of 10-20 mg/kg of fluralaner, 0.025-0.05 mg/kg of moxidectin and 5-10 mg/kg of pyrantel. It is noted that the veterinary medicinal product is only indicated when use against ticks or fleas and one or more of the other target parasites is indicated at the same time.

Other studies

Palatability

The applicant has presented the results of palatability studies conducted as part of three field studies presented with this application, however, the results presented do not meet the threshold to conclude palatability for the product and consequently the data provided are not considered adequate to support a palatability claim for Bravecto TriUNO.

Part 5 – Benefit-risk assessment

Introduction

Bravecto TriUNO is a fixed combination of three active substances: fluralaner, moxidectin and pyrantel (as embonate). The active substances are well-known.

Fluralaner is a systemically acting ectoparasiticide belonging to the isoxazoline family, whilst moxidectin, a second generation macrocyclic lactone of the milbemycin family, and pyrantel, a nicotinic acetylcholine channel receptor agonist, act against endoparasites.

The product is intended for use in dogs with, or at risk from, mixed parasitic infestations by ticks or fleas, gastrointestinal nematodes, lungworm and/or heartworm. The veterinary medicinal product is only indicated when use against ticks or fleas and one or more of the other target parasites is indicated at the same time. The proposed dose of 10-20 mg/kg of fluralaner, 0.025-0.05 mg/kg of moxidectin and 5-10 mg/kg of pyrantel administered orally at 1-month intervals has been confirmed.

The application has been submitted in accordance with Article 20 of Regulation (EU) 2019/6 (combination veterinary medicinal product application).

Benefit assessment

Direct benefit

The combination of three active substances is justified on the basis that the parasites targeted by the combination product (fleas, ticks, gastrointestinal nematodes and/or heartworm and angiostrongylosis) are commonly found in dogs in Europe and can be present simultaneously on the same animal; the three active substances (fluralaner, moxidectin and pyrantel) included in the combination product have different spectra of activity and, as a consequence, the spectrum of activity is broadened.

The applicant applied for the following indications:

"For dogs with, or at risk from, mixed parasitic infestations by ticks or fleas, gastrointestinal nematodes, lungworm and/or heartworm. The veterinary medicinal product is only indicated when use against ticks or fleas and one or more of the other target parasites is indicated at the same time.

For the treatment of tick and flea infestations in dogs providing immediate and persistent flea *(Ctenocephalides felis* and *C. canis)* and tick *(Dermacentor reticulatus, Ixodes hexagonus, I. ricinus,* and *Rhipicephalus sanguineus)* killing activity for 1 month.

The veterinary medicinal product can be used as part of a treatment strategy for the control of flea allergy dermatitis (FAD).

For reduction of the risk of infection with *Babesia canis via transmission by D. reticulatus* for 1 month. The effect is indirect due to the product's activity against the vector.

For reduction of the risk of infection with *Dipylidium caninum* via transmission by *C. felis* for 1 month. The effect is indirect due to the product's activity against the vector.

Treatment of infections with gastrointestinal nematodes of the following species: roundworms *(*immature adult (L5) and adult stages of *Toxocara canis,* and adult stages of *Toxascaris leonina)* and hookworms (L4, immature adult (L5) and adult stages of *Ancylostoma caninum* and adult stages *of Uncinaria stenocephala).*

Prevention of heartworm disease (caused by *Dirofilaria immitis).*

Prevention of angiostrongylosis (by reduction of the level of infection with immature adult (L5) and adult stages of *Angiostrongylus vasorum)."*

The CVMP concluded that the claimed efficacy against most target parasites has been adequately supported, with the exception of the L5 stage of *Toxocara canis*.

Additional benefits

The effective control of fleas on treated dogs will directly reduce the risk of infestation of other animals in contact with infested dogs. The product, being a fixed combination, facilitates dog handling by reducing the total number of tablets given.

Risk assessment

Quality

Information on the development, manufacture and control of the active substance and the 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.

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SPC. Physicochemical aspects relevant to the performance of the product have been investigated and are controlled in a satisfactory way.

Safety

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

Risks for the target animal:

Administration of Bravecto TriUNO in accordance with SPC recommendations is generally well tolerated, and adverse events are accurately captured in the SPC.

Risk for the user:

The risk to the user is considered acceptable noting in particular that the pharmaceutical form (chewable tablet) limits the potential for the user to be exposed to the active substance when removing the product from the packaging and administering the tablet to the animal. As the product can pose a risk to children accidentally ingesting a tablet, specific measures are necessary to mitigate the risk; the tablet is presented in suitable child-resistant packaging, and the product information includes a warning advising of the potential for adverse effects in case of accidental ingestion and specific instructions to remove tablets from the packaging only when required, and to store the product out of the sight and reach of children. The user safety for this product is acceptable when used as recommended and taking into account the safety advice in the SPC.

Risk for the environment:

Bravecto TriUNO is not expected to pose a risk for the environment when used according to the SPC recommendations. However, the active ingredient moxidectin is classified as a PBT substance. This information is communicated in the product literature.

Risk of anthelmintic resistance:

At present, no resistance of the claimed parasites in dogs to the individual active substances has been reported for Europe. However, there are reports of resistance development of *D. immitis* to macrocyclic lactones in dogs in the USA and pyrantel resistance in *A. caninum* in Australia and the US. Given the concerns regarding resistance development, section 3.4 of the SPC includes warnings relating to the potential for resistance emergence.

Risk management or mitigation measures

Appropriate information has been included in the SPC and other product information to inform on the potential risks of this product relevant to the target animal, user and environment and to provide advice on how to prevent or reduce these risks.

Conditions or restrictions as regards the supply or safe and effective use of the VMP concerned, including the classification (prescription status).

The veterinary medicinal product is subject to a veterinary prescription.

Evaluation of the benefit-risk balance

At the time of submission, the applicant applied for the following indication: "For dogs with, or at

risk from, mixed parasitic infestations by ticks or fleas, gastrointestinal nematodes, lungworm and/or heartworm. The veterinary medicinal product is only indicated when use against ticks or fleas and one or more of the other target parasites is indicated at the same time.

For the treatment of tick and flea infestations in dogs providing immediate and persistent flea *(Ctenocephalides felis* and *C. canis)* and tick *(Dermacentor reticulatus, Ixodes hexagonus, I. ricinus,* and *Rhipicephalus sanguineus)* killing activity for 1 month.

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Prevention of heartworm disease (caused by *Dirofilaria immitis).*

Prevention of angiostrongylosis (by reduction of the level of infection with immature adult (L5) and adult stages of *Angiostrongylus vasorum).*"

The CVMP agreed to the indications as proposed by the applicant with the exception of treatment of infection by L5 stage of *Toxocara canis.*

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.

Conclusion

Based on the data presented on quality, safety and efficacy the Committee for Veterinary Medicinal Products (CVMP) considered that the application for Bravecto TriUNO is approvable since these data satisfy the requirements for an authorisation set out in the legislation (Regulation (EU) 2019/6).

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