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GUIDELINES

FOR THE REGISTRATION OF VETERINARY ECTOPARASTICIDES IN THE EAST AFRICAN COMMUNITY

Draft agreed by Technical Working Group	
Draft released for consultation by representatives of East African region regulatory agencies	09/10/2023
End of consultation period	08/03/2024
EAC code	PSS/1/1/21/.....
Enters into force	

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ACRONYMS

AA	Annual Average
ADI	Acceptable Daily Intake
ADME	Absorption, Distribution, Metabolism and Excretion
AOEL	Acceptable Operator Exposure Levels
AP	Acidification Potential
AI	Active substance s
ARFD	Acute Reference Dose
AS	Active Substance
AUC	Area Under the Curve
BCF	bio-concentration factor
BCOP	Bovine Corneal Opacity and Permeability
CA	Chemical Abstracts
CAS	Chemical Abstracts Service
CIPAC	Collaborative International Pesticides Analytical Council
COA	Certificate of Analysis
EC	European Commission
EC	Emulsified Concentrate
ECX	Effective Concentration
EFT	Efficacy Field Trial
EG	Emulsified Granules
EP	Eutrophication Potential
EQS	Environmental Quality Standards
FIFO	First in First Out
FISH	Fluorescence In-Situ Hybridisation
GC	Gas Chromatography
GC MS	Gas Chromatography Mass Spectrometry
GLP	Good Laboratory Practice

GMP	Good Manufacturing Practices
GWP	Global Warming Potential
HET-CAM	Hen's Egg Test - Chorio-Allantoic Membrane
HPLC	High-Performance Liquid Chromatography
HPLC MS	High-Performance Liquid Chromatography Mass Spectrometry
HQ	Hazard Quotient
HR	Highest Residue
ICE	Isolated Chicken Eye
IGR	Insect growth regulator
ILV	Independent Laboratory Validation
IR	Infrared
IRE	Isolated Rabbit Eye Test
ISO	International Organization for Standardization
IUPAC	International Union of Pure and Applied Chemistry
JMPS	Joint Meeting on Pesticide Specifications
LD50	The amount of a material, given all at once, which causes the death of 50% (one half) of a group of test animals
LLC	Lowest Lethal Concentration
LLNA	Local Lymph Node Assay
LOD	Limit of Detection
LOQ	Limit of Quantification
MAC	Maximum Acceptable Concentration
MRL	Maximum Residue Levels
MS	Mass Spectra
NMR	Nuclear Magnetic Resonance
NOAEL	No Observed Adverse Effect Level
NOEC	No Observed Effect Concentration
NOEL	No Observed Effect Level
OD	Oil-based Suspension Concentrates

OECD	Organization for Economic Co-operation and Development
OPD	Ozone Depleting Potential
PEC	Predicted Environmental Concentration
PECA	Predicted environmental concentration in air
PECGW	Predicted environmental concentration in groundwater
PECS	Predicted environmental concentration in soil
PECSED	Predicted environmental concentration in sediment
PECSW	Predicted environmental concentration in surface water
POCP	Photochemical Ozone Creation Potential
POP	Persistent Organic Pollutant
PSUR	Periodic Safety Update Reports
QA	Quality Assurance
QSPR	Quantitative Structure Property Relationship
RQ	Risk Quotient
STMR	Supervised Trials Median Residue
STP	Sewage Treatment Plant
TER	Toxic Exposure Ratio
TGAI/S:	Technical grade of the Active substance /substance
TK	Technical Concentrate
UV/VIS	Ultraviolet/Visible

DEFINITIONS

In these guidelines, unless the context otherwise requires:

Active Substance(s): the ingredient(s) of the product that provides action against the ectoparasites.

Applicant: the party (manufacturer, Formulator, Distributer, importer, or their representative) that makes an application for registration of an ectoparasiticide to the responsible authority.

Ambient Temperature: common, prevailing, and uncontrolled atmospheric conditions in a room or place.

Antidote: medicine given to counteract poison.

Batch: Means the result of the manufacturing process and represents one homogeneous amount of a formulation after release. Often, other descriptors such as lot may be used instead of a batch. An Expiry Date is set at batch level.

Calibration: (of application equipment) measuring and adjusting the output and working rate of application equipment, so as to achieve precision and accuracy of the measurement.

CIPAC Methods: analytical and physical test methods published by Collaborative International Pesticides Analytical Council.

Concentrates: the form in which veterinary ectoparasiticides products are usually sold, mostly requiring dilution before use. 'Emulsifiable' concentrates are liquids which form emulsions upon dilution; and 'suspension' concentrates form suspensions.

Co-formulant: also referred to as "inert" or "formulation inert". A co-formulant is a substance or a mixture other that the active substance(s), added in the formulation during the formulation process in order to dilute and/or to bring a desirable property to the final formulated ectoparasiticide.

Container Closure System: refers to all components intended to seal and protect the ectoparasiticides product

Commercial) Sales Pack: container in which an ectoparasiticide is placed on the market.

Competent Authority: refers to the competent agency/institution responsible for Ectoparasiticides registration and monitoring of ectoparasiticides within the respective country, as set out in the *Introduction* of these guidelines and as may be updated from time-to-time.

Dates: format and details of dates as defined by country-specific regulations.

Dilution: the addition of (usually) water to reduce the concentration of a veterinary ectoparasiticides before use.

Dose Rate: the amount of veterinary ectoparasiticides recommended to be used on (usually) the animal body.

Ectoparasiticides: means any substance, or mixture of substances of chemical or biological nature intended for repelling, killing or controlling ectoparasites such as ticks, mites, lice, fleas, tsetse flies, biting and nuisance flies, and the substance being applied directly to the animal.

Expiry Date: date beyond which a batch of a product should not be used without further appropriate re-examination and the manufacturer cannot guarantee its safety and efficacy. The date is calculated and based on the starting date of a batch. The Expiry Date is always related to a particular batch of a veterinary ectoparasiticides.

Equivalence: the determination of the similarity of the impurity and toxicological profile, as well as of the physical and chemical properties, presented by supposedly similar technical material originating from different manufacturers, in order to assess whether they present similar levels of risk

FAO/WHO Tolerance Limits: tolerance limits established by FAO / WHO for the Active substance content as outlined in the FAO/WHO manual, considering analytical and sampling errors and the manufacturing variance. The Active substance content of a ECTOPARASITICIDEs must stay within these tolerances during its entire Shelf Life.

Formulated Product: any formulation containing one or more Active substances which is effective for the purpose claimed and for the envisaged mode of application.

Formulation: the combination of various ingredients designed to render the product useful and effective for the purpose claimed and for the envisaged mode of application.

or

term to describe an unpacked preparation with defined composition containing Active substance (s) and co-formulant(s).

Good Laboratory Practice (GLP): a quality system concerned with the organizational process and the conditions under which non-clinical health and environmental safety studies are planned, performed, monitored, recorded, archived, and reported.

Guideline: a document that aims to streamline processes according to a set routine.

In-process Control: checks performed during production in order to monitor and, if necessary, to adjust the process to ensure that the product conforms to its specifications.

Label a written, printed, or graphic matter on, or attached to, the ectoparasiticide container or the immediate container thereof and to the outside container or wrapper.

Leaflet: a written material that is supplied in the form of a detachable or separate information(s), booklet(s) or similar, rather than attached permanently to the container.

Maximum Residue Limit (MRL): the maximum concentration of a residue that is legally tolerated or recognized as acceptable in a food product obtained from an animal that has received a veterinary medicine.

Manufacture: Means formulating, production, quality control, release and storage of veterinary ectoparasiticides products, and the related controls.

Manufacturer: a corporation or other entity in the public or private sector (including an individual) engaged in the business or function (whether directly or through an agent or entity controlled by or under contract with it) of manufacturing an ectoparasiticide Active substance or preparing its formulation or product.

Or

a company that carries out operations such as production, packaging, repackaging, labeling, and re-labelling of veterinary ectoparasiticides.

Or

means a person who is responsible for formulating, manufacturing, processing, labelling and packaging the ectoparasiticide, and/or may sell the products under their own name, or under a trade mark, design, trade name or other name or mark owned or controlled by the person, or for assigning to it a purpose, whether those tasks are performed by that person or on their behalf

Marketing Authorization: refers to a procedure for approval of an ectoparasiticide product for marketing after it has undergone a process of evaluation to determine the safety, efficacy and quality of the product and the appropriateness of the product information.

Product License: registration certificate) - means a legal document issued by the competent authority that establishes the detailed composition and formulation of the product and the CIPAC Methods or other recognized specifications of its ingredients and of the final product itself, and includes details of packaging, labelling and shelf-life.

A Marketing Authorization holder (MAH): is the person or company who is licensed to distribute, sell and commercialize a medical product.

No Significant Change: physical, chemical and technical properties of a ectoparasiticide stay within the registration of the product, e.g. Active substance content stays within FAO tolerances, and the product stays fit for use.

No significant Weight Change: Measures used to evaluate the suitability and tightness of a packaging containing the ectoparasiticide upon storage.

Packaging: refers to the science, art and technology of enclosing or protecting products for distribution, storage, sale, and use.

Partner State - refers to the East African Community partner states.

Ecotoxicology - is the study of the effects of toxic chemicals on biological organisms, especially at the population, community, ecosystem, and biosphere levels.

Or

Ecotoxicology is a multidisciplinary field, which integrates toxicology and ecology.

Ectoparasite: any species, strain or biotype of arthropods, animal or pathogenic agent injurious to animal and animal materials or environments, and includes vectors which transmit parasites or pathogens of human and animal disease and animals causing public health nuisance

Pictogram: a graphical presentation that may include a symbol plus other graphic elements, such as a border, background pattern or colour that is intended to convey specific information.

Precautionary Statement: a phrase (and or/pictogram) that describes recommended measures that should be taken to minimize or prevent adverse effects resulting from exposure to an ectoparasiticide, or improper storage, transportation or handling of an ectoparasiticide.

Product: refers to a formulated product (Ectoparasiticides) in the form such as emulsifiable concentrate, powder, tablets, granules oil etc in which it is packaged and sold.

Process Validation: the documented evidence that the process, operated within established parameters, can perform effectively and reproducibly to produce an ectoparasiticides product meeting its predetermined specifications and quality attributes.

Production: all operations involved in the preparation of a veterinary ectoparasiticides product, from receipt of materials, through processing, packaging and repackaging, labelling and re-labelling, to completion of the finished product.

Personal Protective Equipment: clothing and equipment selected or designed to protect the wearer against contamination by veterinary ectoparasiticides products. Means proper attire to protect the health of any person handling ectoparasiticides such as a facemask, goggles, aspirators, rubber gloves, plastic or rubber aprons, rubber boots, overalls and caps in order to protect the wearer against contamination by veterinary ectoparasiticide products; to be worn, as recommended, when handling, mixing, and applying ectoparasiticide product

Registration Dossier: the set of data/information about a product that is submitted by applicants, in a structured manner, in support of their application for registration.

Regulatory Authority: the government agency or agencies responsible for regulating ectoparasiticides and more generally for implementing ectoparasiticide legislation.

Release Date: the date from which the supplier guarantees a shelf-life of at least 2 years, unless stated otherwise, under actual conditions of storage in the area where the technical grade active substance or formulated product is to be marketed.

Risk: the probability and severity of an adverse health or environmental effect occurring as a function of a hazard and the likelihood and the extent of exposure to an ectoparasiticide.

Signal word: a word used to indicate the relative level of severity of hazard and alert the reader to a potential hazard on the label.

Symbol: graphical element intended to succinctly convey information related to the classification of the ectoparasiticide hazard.

Toxicity: a physiological or biological property which determines the capacity of an ectoparasiticide to do harm or produce injury to a living organism by other than mechanical means.

No Seepage: No seepage can be defined as no leakage of product through the container walls, no stickiness at the outside of the package, tight closure and pack, no significant ballooning or panelling, no significant weight change.

Shelf Life: period during which the product remains suitable for use and complies with its specifications when stored in the unopened original sales pack under conditions recommended by the manufacturer. The Shelf Life is established for a formulated product in general (including its packaging) and applies to all its manufactured batches.

Manufacturing Date: Date as used in different guidance documents, e.g. FAO / WHO Manual (1). Refers to a date on which a batch becomes the finished product as it is described by its specifications. Products normally have a shelf life of two years from the starting date.

Release Date: Release date has been defined as the date from which the supplier guarantees a shelf-life of storage in the area where the technical grade active substance or formulated product is to be marketed.

Specification: a list of detailed requirements with which the products or materials used or obtained during manufacturing must conform. They serve as a basis for quality evaluation.

Technical Grade of the Active Substance: A chemical material containing an Active substance which contains no ingredient, other than one used for manufacture or purification of the Active substance and which is produced on a commercial or pilot-plant production scale (whether it is ever held for sale). Technical materials are deemed to have a purity of not less than 95%

Veterinary Ectoparasiticide: is an antiparasitic product used in the treatment of external parasite infestations. Or Medicine used to kill the parasites that live on the body surface of an animal. Also referred to as formulated product in this document.

Volatile: describing chemicals which evaporate readily, even at low temperatures.

Withholding Period: in relation to the use of a veterinary ectoparasiticides, means the minimum period that needs to elapse between the last use of the product in relation to an animal and slaughtering of the animal, or the collection of milk or eggs from the animal for human or animal consumption, as the case may be in order to ensure that the product's residues fall to or below the recommended maximum limit.

INTRODUCTION

This guideline is intended for use by Applicants intending to submit veterinary ectoparasiticides for registration within the East African Community (EAC) Partner States. Applicants are guided on the arrangement of information to be included in the ectoparasiticides registration dossier for the registration of formulated ectoparasiticides products.

OBJECTIVES

The objective is to provide guidance to Applicants seeking Marketing Authorisations for their Veterinary ectoparasiticides in the East Africa Community Partner States in generating of appropriate data and the arrangement of information to be included in the registration dossiers. It also describes the scientific data and information that may be needed to allow governments and regulator to evaluate veterinary ectoparasiticides for the purpose of their registration as well as to facilitate the interpretation and application of the EAC legislation concerning the sale and supply of Veterinary ectoparasiticides Products within the East African Community. For registration of veterinary ectoparasiticides in a group of countries; it harmonizes the data requirements for ectoparasiticide registration. It also describes the data required to support the safety, quality, and efficacy of the veterinary ectoparasiticides.

SCOPE

This guideline covers the requirements for registration of veterinary ectoparasiticides used in the control of ectoparasites such as ticks, mites, lice, fleas, tsetse flies, biting and nuisance flies etc. in domestic animal species (poultry, ruminants, equines, canines, felines, camelids and porcine). It will also form a basis of which East African competent Authorities will evaluate the registration dossiers of the products. The guideline is arranged in eight sections as follows:

Section 1: Administrative Information.

Section 2: Details of technical grade/Active substance.

Section 3: Particulars of the formulated product

Section 4: Safety studies

Section 5: Metabolism and Residues Data

Section 6: Fate and Behaviour in the Environment

Section 7: Efficacy Studies

Section 8: Exposure data and information

GENERAL INFORMATION

The data required to support an application for registration should cover all relevant aspects of the product, from manufacture to use and ultimate disposal. The following sections explain the categories of data and information that are required: information on the proposed application; data to identify the product (identity, composition, analysis); data to assess risks to humans and the environment including data of effects on non-target species; and data to assess the efficacy of the product. This information will help Competent authorities to determine whether and when these data may be applicable for the Veterinary ectoparasiticides regulatory decisions.

a) This guideline shall apply to registration of veterinary ectoparasiticides used in the control of ectoparasites in domesticated animal.

b) All applications and accompanying data to be submitted to the EAC Partner States regulatory agencies shall be typewritten or computer printed in ENGLISH and if an applicant wishes to submit material in another language, they must submit a notarised /certified English translation.

c) The Applicant shall be a person who desires to engage in the manufacture, import, export, distribution, and sales of the veterinary ectoparasiticides and such products should satisfy the eligibility requirements to be submitted together with the application.

d) The applicant shall prepare and present the product dossier information according to the requirements and format as stipulated in this guideline.

e) This guideline is not legally binding but a guidance to information documents to be submitted to the regulators. It represents the harmonised view of the EAC Partner States.

f) Each complete application must contain a complete index to the various appendices and each page of the application dossier must be numbered.

g) The appropriate application fee shall accompany each complete application form, as per the national Competent authority /agency fees' structure/schedule in each East African Country. Applicants are advised to contact the national authority through their Local Technical Representative (LTR) for their current fee structure/schedule. Subsequent applications to amend any part of the application shall be accompanied by additional fees for the required variation and the guideline on submission of amendment application shall be observed.

h) The application should be submitted in accordance to participating Partner State pathway either hard and/or soft copies (CD-ROM or External Driver) addressed to the competent authority or through online submission portal.

i) The PDF documents should be in Optical Character Recognition, selectable and searchable.

j) A separate application is required for each product of different strength and formulation. A veterinary ectoparasiticides shall be considered different from another ectoparasiticide, if:

i. the Active substance of that ectoparasiticide is different.
ii. the trade name or trademark of that ectoparasiticide is different.
iii. the active substances with different concentrations.
iv. the ectoparasiticide with different dosage form.
v. the ectoparasiticide with different excipients.
vi. the ectoparasiticide is manufactured by a different manufacturer.

k) All Veterinary ectoparasiticides should be registered before use within the region.

l) No veterinary ectoparasiticides shall, be registered before use within the region.

m) Technical assessment of the product should be done concurrently with GMP. When it passes the quality and safety assessment then EFT can be done

Type of Veterinary Ectoparasiticide

There is a wide range of veterinary ectoparasiticides formulation and many more under development which includes.

This guideline will provide guidance for registration of conventional veterinary ectoparasiticides products while for other types of ectoparasiticides, applicants are requested to refer to relevant guidelines.

Product Samples to be Submit for Registration

A minimum of three samples of the formulated product for the smallest commercial pack size should be submitted to all participating Partner State where application is to be submitted and shall be in the form in which it shall appear on the market. For other pack sizes, the applicant is requested to provide the product mock-ups.

Authorised Technical Representative

Authorised Local Technical Representative shall be a body corporate (company), licensed to handle Veterinary Ectoparasiticides Products, shall be the applicant's local technical representative in country where application is submitted with legal authorisation to take full responsibility for the product on behalf of the applicant, and will be answerable to the competent authority registering the product.

This body corporate shall be called the Local Technical Representative (LTR). A copy of the legal authority/power of attorney given to the representative or agent by the sponsor shall be presented to the regulator. Such a body may be:

a) a Distributor
b) a retail outlet
c) a registered local branch/office of the applicant, in which an authorised person is employed.

EAC MRP Veterinary Ectoparasiticides National Competent Authorities

Country	Address
Burundi:	Ministry of the Environment, Agriculture and Livestock BP 161 Gitega, Burundi. BP 161 Gitega, Burundi. Website: www.minagrie.gov.bu
Democratic Republic of Congo	18-20, Av.Liberation(ex 24Novembre), Commune de la Gombe/ Kinshasa/RDC Ref. Immeuble SNDE
Kenya	Veterinary Medicines Directorate P. O. Box 66171-00800 NAIROBI, KENYA. Email: info@vmd.go.ke
Rwanda	Rwanda Food and Drugs Authority P.O. Box 1948 Kigali, Rwanda Nyarutarama Plaza Rwanda KG 9 Avenue, Kigali Email: info@rwandafda.gov.rw Website: https://rwandafda.gov.rw/
South Sudan	Ministry of Livestock and Fisheries P.O. Box 293 Juba. South Sudan http://mar.gov.sd/.
Uganda	The Secretary to the Authority, National Drug Authority, Plot 46 – 48 Lumumba Avenue P.O. Box, 23096, Kampala, UGANDA Website: www.nda.or.ug And The National Council of Science and Technology-, Plot 6, Kimera Road, Ntinda P.O.Box 6884, Kampala Uganda Email: info@uncst.go.ug Website: www.uncst.go.ug
United Republic of Tanzania; Tanzania Mainland	Tanzania Veterinary Laboratory Agency Veterinary Complex 131 Nelson Mandela Road P.O. Box 9254 Dar Es Salam

	Tanzania Email: info@tvla.go.tz Website: https://www.tvla.go.tz/
Zanzibar	Zanzibar Food and Drug Agency P.O.BOX 3595 Changu Road Mombasa Area, Zanzibar, Tanzania Email: info@zfda.go.tz

SECTION 1.0 ADMINISTRATIVE INFORMATION

This section should include the completed and signed Mutual Recognition Procedure Application Form, Summary of Product Characteristics, Primary container label, Secondary container text (carton) Packaging leaflet and user Information leaflet

1.1 Registration in Other Countries

If the veterinary ectoparasiticides has been registered in the country of manufacture, the conditions of registration and copy of the registration certificate/marketing authorisation shall be provided.

A copy of the manufacturing license of the manufacturer shall be provided.

If a product is not registered in country of manufacture, provide a free sale certificate from a competent National Authority in the country of manufacture.

1.2 Compliance to the Current Good Manufacturing Practices (cGMP)

Veterinary ectoparasiticides (ectoparasiticides) intended for registration in the region must fulfil all the usual requirements of approval process. The product must be manufactured in a Good Manufacturing Practise (GMP) compliant Facility or Quality Management System (QMS) ISO standard equivalent to GMP. The applicant is to provide current Good Manufacturing Practise cGMP certificate from a competent authority of manufacturing country. It is also mandatory for the manufacturing plant to be inspected by the EAC GMP joint inspection team. The Veterinary ectoparasiticides manufacturing facilities must comply with GMP requirements before the product is granted a marketing authorization. The applicants should refer to the EAC MRP GMP guidelines.

1.3 Pre-registration Requirement for Efficacy Field Trials in the Regional Environment

Proof of efficacy field trial conducted under regional conditions in selected EAC Partner State is a mandatory pre-registration requirement in the EAC region.

Regional efficacy data must be generated from trials conducted within the East African countries. Additional data from trials in other countries may be submitted as supporting data in a registration application, to support safety, quality, efficacy, and any other label claims. Where no capacity exists in EAC region to conduct specific ectoparasite/application method, then the

registration applicant must consult with the competent authority for ectoparasiticide registration on conducting a trial outside of EAC, or whether the specified trial/ectoparasite/application method claim can be waived for registration.

The applicant will bear all the costs of the trial. The duration of the study shall cover the label claim period and should be conducted in the worst-case scenario; dry/hot and wet seasons encountered in the region. The field efficacy studies shall be conducted in two sites translating to two countries using the agreed trial site selection criteria. The Concerned Countries and References countries should provide guidance to applicants to select appropriate Partner States to conduct each EFT per application to encourage inclusivity. This should be supported by invitro laboratory efficacy studies.

Duration of validity of EFT reports & relevant supporting information should remain in force until post-market surveillance report(s) demonstrate the need for new study. Annual periodical reports by the regulator in liaison with license holder should be the source of supporting information.

Where the pre-registration field efficacy trials are waived especially for the generic's equivalents already in the market, then invitro studies using local parasite samples must be done, with full payment of regulatory fees.

Laboratory efficacy studies should be conducted when resistance is suspected as per the post-market surveillance reports from regulators or where the pre-registration field efficacy trials are waived especially for the generic's equivalents already in the market.

FAO-recommended bioassays for susceptibility/resistance should be conducted to confirm label claims on Active Substance concentration before EFT commences.

Field trial sites (including dip-stability study sites) shall be selected as outlined in the study protocol in the EFT guideline. For the minimum requirements for the format of EFT protocol; refer to VICH GCLP Format.

The Contract Research Organizations shall be selected based on the availability of sufficient infrastructures, storage and handling facility for trial products, qualified Principal Investigator, and personnel. Regulators shall publish list of such CROs.

1.4 The pre-requisite for conduct of EFTs are:

i. When a product has new active substance(s)
ii. the active substances with different concentrations
iii. the ectoparasiticide with different dosage form. When new dosage.
iv. the ectoparasiticide with different excipients.

Applicants are advised to refer and use the guideline on Pre-registration Efficacy Field Trials in the EAC for details on the EAC Website using the link: http://www.eac.int/documents/category/livestock

SECTION 2.0 PARTICULARS OF THE TECHNICAL GRADE/ACTIVE SUBSTANCE

2.1 Introduction

Every active substance is evaluated for safety and efficacy before it reaches the market in a product. This evaluation consists of an assessment of the risk to humans, animals, and the environment and includes an assessment of the residues in food. The assessment is conducted for a representative product containing the active substance.

Applicants are required to submit data on the specification of the active substance, such as data regarding the identity of any impurities and/or additives, including by-products of synthesis, the isomeric composition, and the method of manufacture and the manufacturing source. This information affects the toxicity profile of the substance and thereby the risk assessment.

It is recognised that no chemical substance is 100% pure. It contains by-products that are produced during manufacturing, and impurities carried over from the raw materials used for manufacturing. The WHO hazard classification based on the latest UN GHS directive (Globally Harmonized System of Classification and Labelling of Chemicals), recommends the technical grade of the Active substance s for use in ectoparasiticides manufacturing. Technical grade usually means a purity of ≥95%, i.e., they contain ≤5% impurities. The minimum acceptable grade for all topical ectoparasiticides/veterinary ectoparasiticides products (sprays, dips, etc) is the technical grade.

Regulators are concerned about impurities in ectoparasiticides, particularly impurities which are present in the technical grade of the active substance such as those substances carried over from a starting material, or from an intermediate, and impurities formed through side reactions between the active substance and any other component of the product or packaging of the product or by degradation of the active substance.

2.2 Identification of the Active Substance and Composition

Provide the identity and composition of the technical material; to determine the quality of the ectoparasiticides submitted for registration; to identify impurities of toxicological or ecotoxicological concern or any other relevant impurity; and to identify other hazards. Provide a copy of all technical specifications, material safety data sheets, or a certificate of analysis.

Provide the following details of the identity of each active Substance:

- the common name
- the International Union of Pure and Applied Chemistry (IUPAC) chemical name.
- Chemical Abstracts Service (CAS) registry number
- the manufacturer's code number(s) and/or synonyms
- the chemical structure.

2.2.1 Common Name

The applicant should provide the International Organization of Standardization (ISO) common name, or proposed ISO common name, and where relevant, other proposed or accepted common names (synonyms), including the name (title) of the nomenclature authority concerned.

2.2.2 IUPAC Chemical Name

Provide the full chemical name, in accordance with both the IUPAC and the Chemical Abstracts (CA) nomenclature.

2.2.3 Chemical Abstracts Service Registry Number (CAS)

If available, Provide the CAS number of the active substance. If the CAS number has not been allocated, indicate that the number is not yet allocated.

2.2.4 Manufacturer's Code Number(s) and/or Synonyms

The manufacturer or laboratory code numbers and/or synonyms should be provided, as applicable.

2.2.5 Chemical Structure

2.2.5.1 Molecular and Structural Formulae and Molecular Mass

The applicant should provide the Chemical group, molecular formula, molecular mass, and structural formula of the active substance. For active substances that are salts or hydrates, provide the molecular mass of the free base or anhydrous form. For polymeric compounds this should include weight average (M_w), number average, molecular weight (M_n) and molecular weight distribution.

The structural formula should include (where relevant) stereochemical properties of the active substance; for example, geometric isomerism (*cis/trans, E/Z*), the number of chiral centres and the configuration at each centre. Where possible, the structural formula should be provided diagrammatically with all possible or known stereochemistry.

Table 1: The Chemical Structure

	Spectroscopic Data	**Interpretation /Results**
^{1}H and ^{13}C nuclear magnetic resonance (NMR) spectra		
Mass spectrum (MS)		
Infrared (IR) spectra		
^{19}F and ^{31}P spectral data, where relevant		
Discussion of the synthetic route as evidence of structure		
elemental analysis with theoretical values		
Discussion on ultraviolet (UV) characteristics including pH dependence shifts.		
Any other related information used to confirm the structure (for example, X-ray diffraction).		

2.2.5.2 Elucidation of Structure and Other Characterization of Structure

The applicant should provide confirmation of the chemical structure of the active substance and impurities. The elucidation of structure based on the spectroscopic data should be provided, along with their interpretation, and all other appropriate physical and chemical test results. Such a process requires the application of the correct methods coupled with a skilled and highly knowledgeable approach like mass spectrometry techniques.

2.2.6 Physical and Chemical Properties

All physico-chemical characteristics of technical grade of active substance are required. The manufacturer should provide the relevant physical and chemical properties of the active substance and/or manufacturing concentrate should be provided for development of specification for technical grade Active substance and formulations. Provide the DT_{50} of the Active

substance, with mention of temperature and pH parameters employed during the determination of Photolysis. Provide the DT_{50} of the active substance (in days). Where relevant, provide method/test.

The information should include, as appropriate and reported in a tabular form.

The purity of the test substance used to generate the physical and chemical properties should be stated.

Provide a brief description of the methods used to generate the data. Where the method used is described in a scientifically recognised publication or manual—for example, those by the Organization for Economic Cooperation and Development (OECD), the Collaborative International Pesticide Analytical Council (CIPAC), or the American Society for Testing Materials (ASTM)—a reference to the relevant publication will suffice.

Provide the physical properties such as solubility in water and vapour pressure from tests conducted at ambient temperature (20–25˚C). However, if data is available at a different temperature, these may be provided. The temperature at which these tests were conducted, or other relevant test conditions, should be stated and justified.

For innovate products, provide the Patent and exclusivity status of the Active substance and formulation.

Provide the Flammability. Flash point Explosive properties, oxidizing properties, Absorption spectra –UV/Visible, infrared, NMR, MS. Reactivity towards container material properties including the analytic method used for identification. Reference to the Collaborative International ectoparasiticides Analytical Council (CIPAC) guidelines. The guidelines promote the international agreement on methods for the analysis and physio-chemical test methods for active substance in formulations.

2.2.6.1 Melting Point and Boiling Point

The melting point or where appropriate the freezing or solidification point of purified active substance shall be determined and reported. Measurements shall be taken up to 360 °C.

The boiling point of purified active substance shall be determined and reported. Measurements shall be taken up to 360 °C.

Where melting point or boiling point cannot be determined because of decomposition or sublimation, the temperature at which decomposition or sublimation occurs shall be reported.

2.2.6.2 Vapour Pressure Volatility

The vapour pressure of purified active substance at 20 °C or25 °C shall be reported. Where vapour pressure is less than 10– 5 Pa at 20 °C the vapour pressure at 20 °C or 25 °C shall be estimated by a vapour pressure curve with measurements at higher temperatures.

In the case of active substances which are solids or liquids, volatility (Henry's law constant) of purified active substance shall be determined or calculated from its water solubility and vapour pressure and be reported (in Pa × m3 × mol–1).

2.2.6.3 Appearance (physical state, colour)

Provide a description of both the colour, if any, and the physical state of both the active substance as manufactured and purified active substance.

Spectra (UV/VIS, IR, NMR, MS), molar extinction at relevant wavelengths, optical purity. The following spectra, including a table of signal characteristics needed for interpretation, shall be determined, and reported: ultraviolet/visible (UV/VIS), infrared (IR), nuclear magnetic resonance (NMR) and mass spectra (MS) of purified active substance.

Molar extinction at relevant wavelengths shall be determined and reported (ε in L × mol–1 × cm–1). Relevant wavelengths include all maxima in the UV/visible absorption spectrum, as well as the wavelength range of 290- 700 nm.

In the case of active substances which are resolved optical isomers, the optical purity shall be measured and reported.

Where necessary for the identification of the impurities considered to be of toxicological, ecotoxicological or environmental significance, the UV/visible absorption spectra IR, NMR and MS spectra, shall be determine and reported.

2.2.6.4 Solubility in Water

The water solubility of purified active substances under atmospheric pressure shall be determined and a value reported for 20 °C. These water solubility determinations shall be made in the neutral range (that is to say in distilled water in equilibrium with atmospheric carbon dioxide). If the pKa is between 2 and 12, water solubility shall also be determined in the acidic range (pH 4 to 5) and in the alkaline range (pH 9 to 10). Where the stability of the active substance in aqueous media is such that water solubility cannot be determined, a justification based on test data shall be provided.

2.2.6.5 Solubility in Organic Solvents

The solubility of the active substances as manufactured or purified active substance in the following organic solvents at15to25°C shall be determined and reported if less than250g/L; the temperature applied shall be specified. Results shall be reported as g/L.

(a) Aliphatic hydrocarbon: preferably heptane

(b) Aromatic hydrocarbon: preferably toluene

(c) Halogenated hydrocarbon: preferably dichloromethane

(d) Alcohol: preferably methanol or isopropyl alcohol

(e) Ketone: preferably acetone

(f) Ester: preferably ethyl acetate.

If for a particular active substance, one or more of those solvents is unsuitable (for example reacts with test material), alternative solvents may be used instead. In such cases, choices of solvents shall be justified in terms of their structure and polarity.

2.2.6.6 Partition Co-efficient N-octanol/Water

The n-octanol/water partition coefficient (Kow or log Pow) of purified active substance and of all components of the residue definition for risk assessment shall be determined and reported for 20 °C or 25 °C. The effect of pH (4to 10) shall be investigated when the active substance has a pKa value between 2 and 12.

2.2.6.7 Dissociation in Water

Where dissociation in water occurs, the dissociation constants (pKa values) of the purified active substance shall be determined and reported for 20 °C. The identity of the dissociated species formed, based on theoretical considerations, shall be reported. If the active substance is a salt the pKa value of the non-dissociated form of the active substance shall be given.

2.2.6.8 Flammability and Self-Heating

The flammability and self-heating of active substances as manufactured shall be determined and reported. A theoretical estimation based on structure shall be accepted if it meets the criteria set out in Appendix 6 of the United Nations' Recommendations on the Transport of Dangerous Goods Manual of Tests and Criteria (1). In justified cases, data for purified active substance may be used.

2.2.6.9 Flash Point

The flash point of active substances as manufactured with a melting point below 40 °C shall be determined and reported. In justified cases, data for purified active substance may be used.

2.2.6.10 Explosive Properties

The explosive properties of active substances as manufactured shall be determined and reported. A theoretical estimation based on structure shall be accepted if it meets the criteria set out in Appendix 6 of the United Nations 'Recommendations on the Transport of Dangerous Goods, Manual of Tests and Criteria'. In justified cases, data for purified active substance may be used.

2.2.6.11 Surface Tension

The surface tension of purified active substance shall be determined and reported.

2.2.6.12 Oxidizing Properties

The oxidising properties of active substances as manufactured, shall be determined and reported. A theoretical estimation based on structure shall be accepted if it meets the criteria set out in Appendix 6 of the United Nations 'Recommendations on the Transport of Dangerous Goods Manual of Tests and Criteria'. In justified cases data for purified active substance may be used.

2.2.6.13 Other Studies

Supplementary studies necessary for the classification of the active substance by hazard shall be carried out.

Table 2*:* Supplementary studies for classification of the Active Substance

Parameters	**Specification**
A general description, for example, appearance, colour, odour, physical state	
The stereochemical properties of the molecule, for example geometric isomerism, number of chiral centres and configuration at each centre	
If a new active substance contains one or more chiral centres, a specification of whether the active substance is a pure enantiomer, racemate or fixed combination of non-enantiomeric isomers	
If a new active substance contains geometric isomers (cis/trans, E/Z), a specification of whether the active substance is a pure geometric isomer, or a fixed combination of geometric isomers	
If the active substance is optically active, a specific optical rotation measurement with limits	
The melting point or range (for solids)	
The boiling point or range (for liquids)	
If the melting and/or boiling point cannot be determined because of decomposition or sublimation, the temperature at which decomposition or sublimation occurs.	

Parameters	**Specification**
The condensation point (for gases)	
The refractive index (for liquids)	
The density or specific gravity (for liquids)	
The UV absorption maxima and molar absorptivity	
The pH and pKa values	
Vapour pressure (preferably in temperature range 20-25°C)	
Henry's Law Constant	
Solubility in water (preferably in temperature range 20-25°C) expressed as g/L or mg/L, in the neutral range, acidic range (pH 4 to 6) and in the alkaline range (pH 8 to 10)	
The solubility in various organic solvents expressed as g/L or mg/L	
partition coefficient between water and an appropriate non-miscible solvent (n-octanol/water partition coefficient (Kow))	
The hydrolysis in aqueous solution under acid, neutral and basic conditions	
The dissociation characteristics including acid/base dissociation constants (when appropriate).	
The flash point (where the melting point is below 40˚C)	
The flammability including auto-flammability.	
Explosive properties	

Parameters	Specification
Photochemical properties	
Oxidising properties	
The auto-ignition temperature	
Corrosion characteristics	
Absorption spectra -ultraviolet, visible, and infra-red.	
The dangerous goods classification as per the WHO/FAO Dangerous Goods Code.	

2.2.7 Stability Data

Provide the Stability status of Active substance in water, hydrolysis rate, effect of light, identity of breakdown products, Stability in organic solvents used in Formulation and Stability in air, effect of light, identity of breakdown products. This should be clearly indicated in a tabular form. The applicant should provide the results of stability studies (long-term and/or accelerated conditions) conducted on at least one batch of the active substance.

Stability studies should establish the inherent stability of the molecule; the degradation pathway and the identity of any degradation products, especially toxicologically significant impurities formed during storage.

For storage stability studies, samples of the active substance should be stored for a minimum of 14 days at 54˚C and/or, for long term testing, for up to two years at room temperature (30˚C or under normal EAC regional warehouse conditions). If the active substance is unstable at 54˚C and 30˚C, it may be stored at lower temperatures of 0–5˚C and 2–8˚C respectively.

The content of the active substance and, if appropriate, any toxicologically significant impurities, should be monitored initially, at sufficient sampling intervals and at the end of the stability study.

The submission should include:

- the actual stability results (that is, raw data)
- details of the analytical methods employed for the determination of the active substance.

- details of any toxicologically significant impurities and degradation products
- a summary of the results and conclusion.

Provide the effect of metal or metal ions on the stability of the active substance if there is a likelihood that the active Substance will come into contact with metals or metal ions during its storage or use.

Guidelines suitable for the generation of such data should be followed (please see guideline on Generation of storage stability data for agricultural chemical products). Testing should be undertaken in the proposed commercial packaging or in smaller packages of the same construction and materials.

2.2.8 Method of Manufacture of the Active Substance

2.2.8.1 Manufacturer and Manufacturing Site

Provide the name and business address of the manufacturing plant of the active substance and the street address in which the active substance is manufactured. If a toll or contract manufacturer is involved, provide their name, and business address of the manufacturing plant of the active substance and the street address in which the active substance is manufactured. The name and address of the producer of the ingredient (if different from the applicant)

2.2.8.2 Description of the Manufacturing Process

Provide an accurate and detailed description of the manufacturing process and process controls and include the following information:

- an introductory paragraph detailing the number of chemical steps, whether the process is a batch or continuous process, and significant purification steps (Synthesis pathways).
- a flow diagram of the synthetic processes that outlines the sequence of all the steps.
- a reaction scheme that outlines the sequence of all the chemical reactions and includes
- molecular formulae
    - chemical structure of starting materials, intermediates, reagents, and catalysts
    - the final products formed
    - key reaction conditions
    - amounts used or formed
    - yields obtained

    - the relative amounts of each starting material and their order of addition.

- the reaction conditions (temperature, pressure, pH, reaction times and addition rate etc.)
- the duration and yield of each step of the process
- information on intermediates that are isolated and purified.
- a description of any purification procedures for the active Substance, including procedures to recover starting materials, intermediates, or the final product.
- if a manufacturing concentrate is produced, details of the final concentration of the active Substance present, methods used to confirm the concentration, and details of the diluents and/or any additives used.

Describe the synthetic process in sufficient detail to enable the regulator to assess the potential presence of impurities and impurities of toxicological significance. Specification of purity of the active substance should be expressed in g/kg. Providing all critical points and critical point controls to determine whether the manufacturing process is likely to result in any impurities or manufacturing by-products of toxicological of concern.

2.2.8.3 Quality Control

Provide the following information to ensure the quality of the active substance:

- specifications or purity for all starting materials, reagents, catalysts, and key intermediate products
- the measures used to monitor and assess the performance of an ongoing manufacturing operation, such as the analysis to determine the concentration of a reactant or product to check the completion of a reaction (for example, gas chromatography or high-performance liquid chromatography)
- tests and acceptance criteria (with a justification that includes experimental data) performed at critical steps of the manufacturing process to ensure that the process is controlled.
- representative data relating to in-process quality control.

2.2.8.4 Impurities

The applicant should identify and report on the impurities that are or may be present in the active substance at levels of greater than or equal to 0.1 per cent—note that toxicologically significant impurities at any level must be identified, characterised, and quantified.

Information provided with respect to impurities should include structural formulae and, if possible, a scheme for the formation of the impurity, followed by a text discussion of its formation.

Potential sources of impurities or related substances include:

- impurities in the starting materials, from incomplete or side reactions, or isomerisation
- residual solvents, reagents, and immediate precursors.
- trace elements arising from the use of catalysts or other sources.
- the degradation of the active substance that may occur after manufacture.
- the amount of water or moisture present
- the amount of solvent left after the final purification.

2.2.8.5 Impurities of Toxicological Significance

If there is potential for the formation of toxicologically significant impurities or by-products this must be declared and quantified. Provide details of the conditions leading to their formation and the steps taken to control the formation of toxicologically significant impurities.

2.2.9 Declaration of Composition

Provide a comprehensive Declaration of Composition (DoC) for the active Substance —Table below provides an example. The DoC should be signed and dated by the person responsible for it and include the following information:

- the minimum purity of the active Substance (in grams per kilogram [g/kg] or grams per litre [g/L], as appropriate on a dry-weight basis) as well as the ratio of the content of isomers or diastereoisomers (where relevant)
- the maximum content of all impurities present in quantities of 0.1 per cent or more including water
- any toxicologically significant impurities present at any level (including less than 0.1 per cent)
- if the active substance is a manufacturing concentrate, the minimum concentration of the active substance in the manufacturing concentrate as well as the minimum purity of the active Substance and the maximum content of all impurities on a dry-weight (diluent or additive free) basis, and the content of diluents and/or any additives in g/kg or g/L
- the chemical names, with company code numbers (where applicable), Chemical Abstracts Service (CAS) registry numbers (where they exist),

empirical formulae, molecular weights and structural formulae for all identified impurities.

All impurities present at or above the 0.1 per cent level should be identified and reported. If identification of an impurity is not feasible, a summary of laboratory studies demonstrating the unsuccessful effort should be included in the application.

Specify the minimum purity of the active substance and the maximum content of each impurity based on the previously and recently manufactured batch analysis results from the manufacturer. It's recommended that analysis of at least five batches manufactured within the last five years should be provided, as this offers some statistical certainty on the reliability of the data and demonstrates that the manufacturer is currently capable of producing the active substance.

Table 3: Recommended Format for a Declaration of Composition

<table>
<tr><td colspan="5">Declaration of Composition</td></tr>
<tr><td colspan="5">[Letterhead of the manufacturer including name and address]</td></tr>
<tr><td colspan="5">[Common name of the active substance]</td></tr>
<tr><td colspan="5">[Name and address of the applicant]</td></tr>
<tr><td rowspan="2">Compound (chemical name)</td><td rowspan="2">CAS number</td><td colspan="2">Limits (g/kg or g/L)</td><td rowspan="2">Type

A = active
I = impurity
T = toxicologically significant impurity</td></tr>
<tr><td>Upper limit</td><td>Lower limit</td></tr>
<tr><td></td><td></td><td></td><td></td><td></td></tr>
<tr><td></td><td></td><td></td><td></td><td></td></tr>
<tr><td></td><td></td><td></td><td></td><td></td></tr>
<tr><td colspan="3">[Name and title of responsible person]</td><td colspan="2">[Signature of responsible person and date]</td></tr>
</table>

2.2.10 Batch Analysis Data

The applicant should provide batch analysis results (analysed within the last five years) for at least five commercial-scale production batches of the active substance to demonstrate routine compliance with the DoC and to demonstrate that the manufacturer is in control of the process.

If data on commercial-scale batches are not available, provide batch analyses for pilot-scale batches manufactured using the same process as intended for commercial-scale batches. Data from Laboratory-scale batches should not be included as they do not demonstrate the capability of full-scale manufacture.

The results should include:

- batch size
- batch number
- date of manufacture
- date of analysis
- results of the analytical determination for the content of the active substance and each impurity present at a concentration of 0.1 per cent or more using specific methods—actual numerical results should be provided rather than vague statements such as 'within limits' or 'conforms'.
- content of toxicologically significant impurities (present at any level)
- information on the analytical methods used to generate the data and the validation of these methods.
- where applicable, chromatograms of the batches showing separation of impurities. Chromatograms should be clearly labelled with
    - batch numbers
    - peak identity
    - peak integration data
    - a software-generated table with retention time and peak area of associated peaks
- a copy of all raw data used to generate the results.

To determine impurities in the active substance, reference standards should be prepared for each of the identified impurities, particularly those known to be toxic, and the concentration of impurities should be measured against their own reference standards.

It is acceptable to use the active substance of known purity as an external standard to estimate the levels of impurities (diluted to the appropriate concentration), provided the response factors of those impurities are sufficiently close (90 per cent or more) to that of the active substance. In cases where the response factor is not close, it may still be acceptable to use the

active substance provided a correction factor is applied providing the rationale on how a correction factor is used.

The sum of the quantitative level of the active substance and impurities is often referred to as the mass balance. Mass balance is an important parameter in the batch analysis to ensure that all major impurities have been detected. The mass balance need not add up to exactly 100 per cent, because of the analytical error associated with each analytical procedure; however, it is expected to be in the range of 98–102 per cent.

2.2.11 Analytical Methods

Analytical profile of batches for the technical material, relevant impurities and metabolites, are to be provided, including the full details of the test methods used for determining the active substance, all impurities at or above 0.1 per cent and toxicologically significant impurities (even when present at less than 0.1 per cent) in the active substance.
The following information should be included in a written analytical method:

- a copy of the actual laboratory method. If this laboratory method is not in English, please include an English version.
- the principle of the method
- the method summary
- more than two analytical methods should be employed preferably HPLC and GC
- among other methods of analysis shall include method which regulator can access
- sample preparation techniques
- equipment or reagents (for example, for chromatographic methods, details of the column include column name, manufacturer, packing material and dimensions)
- Eluent (including gradients, where applicable)
- column temperature
- detector and retention times of all components
- purity of reference standard(s), source, and batch number of reference standard(s)
- where chromatographic techniques are used
    - relevant chromatograms (blank, standard, and sample) including retention times.
    - peak-assignment and peak-integration data

    - original printouts from the chromatographic system which include retention times, peak areas, and peak-height tables.

- worked examples of all calculations.

2.2.12 Validation Data

Provide validation data for the method(s) used to assay the active Substance and impurities. Address the following parameters, where appropriate:

- selectivity or specificity
- linearity
- precision
- recovery (accuracy)
- limit of detection (LOD) for impurities
- limit of quantification (LOQ) for impurities.

Note that LOD and LOQ are not required for the quantification of the active substance, only the determination and quantification of the impurities.

2.2.13 Analytical Reference Standards

Application for approval of new active Substance be accompanied by samples to the Local recognised laboratory for each participating country as detailed below:

- 1 gram of the analytical reference standard of each pure active substance, or where the active substance is a mixture of major isomers which can be separated, 1 gram of each isomer.
- 100 grams of active substance as manufactured by the active substance manufacturer.
- 10 milligrams of analytical reference standards for all toxicologically significant impurities present in the active substance.
- 100 milligrams analytical standard for all metabolites identified and for which a maximum residue limit applies.

Provide justification to the regulator for providing less than 1 gram of analytical reference standard and/or less than 100 grams of active substance as manufactured. The justification will be considered on their merits.

Storage instructions and information on the recommended shelf life of the analytical reference standard and active substance are required, especially if degradation is likely to occur under transport or storage.

Samples should be accompanied by a letter stating:

- the reason for submitting the samples including the application and active substance numbers.
- the purity of each of the materials supplied with a separate Certificate of Analysis for each.
- the storage instructions
- the acute oral and dermal toxicities of the materials, or the appropriate safety data sheet.

Care should be taken to ensure that samples are properly packed. Samples that arrive leaking or otherwise damaged will be destroyed and replacement samples will be requested. Samples should be provided to the regulator recognised laboratories before approval of a new active substance. When standards are supplied to these laboratories, documentation to this effect should be forwarded to the participating countries regulators for confirmation purposes.

From time to time, the regulators may request that active-substance approval holders provide replacement material to maintain our inventory of reference materials.

2.2.14 Biochemical Properties of the Active Substance

2.2.14.1 Use of the Active Substance

The information provided shall describe the intended purpose for which the formulated ectoparasiticides containing the active substance are used or are intended to be used and the dose and manner of their use or proposed use.

2.2.14.2 Function

State the intended functions of active substance (e.g., Acaricide or other function).

2.2.14.3 Effects on Ectoparasites

The nature of the effects on ectoparasites shall be stated (e.g. Contact action, stomach action, inhalation action, desiccant, reproductive inhibitor, or others).

2.2.14.4 Mode of Action

To the extent that it has been elucidated, a statement shall be provided as to the mode of action of the active substance in terms, where relevant, of the biochemical and physiological mechanisms and biochemical pathways involved. Where available, the results of relevant experimental studies shall be reported.

Where it is known that to exert its intended effect, the active substance must be converted to a metabolite or breakdown product following application or use of Veterinary ectoparasiticides Product containing it, the following

information shall be provided for active metabolite or breakdown products: (a) chemical name in accordance with IUPAC and CAS nomenclature.

2.2.14.5 Occurrence or Development of Resistance to Active Substances

Provide Information on the occurrence or possible development of resistance to the active substance and appropriate management strategies.

2.2.14.6 Methods and Precautions of Handling, Storage, Transport, or Fire

Methods and precautions concerning handling, storage, transport, or fire.

A safety data sheet shall be provided for all active substances.

The studies, data and information submitted, together with other relevant studies, data, and information, shall both specify and justify the methods and precautions to be followed in the event of fire. The possible products of combustion in the event of fire shall be estimated, based on the chemical structure and the chemical and physical properties of the active substance.

2.2.14.7 Procedures for Destruction or Decontamination:

In many cases the preferred or sole means to safely dispose of active substances, contaminated materials, or contaminated packaging is through controlled incineration in a licensed incinerator.

Other methods to dispose of the active substance, contaminated packaging, and contaminated materials, where proposed, shall be fully described. Data shall be provided for such methods, to establish their effectiveness and safety.

2.2.14.8 Emergency Measures in Case of an Accident

Procedures for the decontamination of water and soil in case of an accident shall be provided. The studies, data and information submitted, together with other relevant studies, data, and information, shall demonstrate the suitability of measures proposed for use in emergency situations.

2.2.15 Packaging

The packaging, or storage or shipping containers must be appropriate for the characteristics of the active substance.

A description of the packaging materials used for the active substance and information regarding the corrosive effect, if any, of the active substance on the packaging materials should be provided. This information is not required if the active substance is formulated into a product at the site of manufacture.

2.3 SAFETY IMPACT ON HUMAN AND ANIMAL (MAMMALIAN TOXICOLOGICAL DATA

2.3.1 Toxicology and Metabolic Studies of the Active Substance

Include an executive summary discussing toxicology and metabolic studies or provide copies of the individual summaries from each study relating to these studies. Studies on Absorption, Distribution, Metabolism and Excretion in Mammals should include.

Absorption, distribution, metabolism, and excretion after exposure by oral route Absorption, distribution, metabolism, and excretion after exposure by other routes

2.3.2 Absorption, Distribution, Metabolism, and Excretion (Toxicokinetic, Pharmacokinetics)

Provide studies examining the absorption, distribution, metabolism, and elimination of active Substance in appropriate laboratory animals. The route of administration for these studies should be carefully considered and consider routes of likely exposure to the active substance in question.

Basic toxicokinetic (TK) parameters determined from these studies should also provide information on the potential for accumulation of the test substance in tissues and/or organs and the potential for induction of biotransformation because of exposure to the test substance.

TK data can be used to assess the adequacy and relevance of the extrapolation of animal toxicity data (particularly chronic toxicity and/or carcinogenicity data) to human hazard and/or risk assessment. Additionally, toxicokinetic studies may provide information useful for addressing issues of dose setting (linearity aspects), route of administration effects, bioavailability (especially with respect to risk assessment issues such as high dose animal to low dose human exposure and route to route extrapolation), and issues related to study design. Specific TK data can be used to develop a physiologically based toxicokinetic (PBTK) model.

There are important uses for metabolite/TK data such as suggesting possible toxicities and modes of action and their relation to dose level and route of exposure. In addition, metabolism data can provide information useful for assessing the toxicological significance of exposures to exogenously produced metabolites of the test substance.

2.3.3 Metabolism and Toxicokinetic Studies in Laboratory Animals

Studies are required on the fate of the active substance in experimental animals. The regulators would normally expect the following information on the active substance from applicants:

2.3.3.1 The degree and rate of absorption after oral administration in at least one mammalian species (an investigation of the extent of absorption after dermal application is desirable; the vehicle chosen for the dermal study should closely resemble that proposed for the product)

2.3.3.2 The extent and rate of distribution and storage in the tissue of animals, or any bioaccumulation that may occur.

2.3.3.3 Biotransformation in animals, including the rate and degree of such biotransformation, together with a description of any metabolites produced.

2.3.3.4 The mode and extent of excretion or elimination of the parent compound and/or its degradation products in animals, including the rate at which such excretion occurs.

2.3.4 Metabolism and Pharmacokinetic Studies in Target Animals

The applicant should conduct metabolism studies using a radiolabelled active substance with, optionally, a pharmacokinetic component, on animal species representative of those likely to come into contact with the material, such as animals directly treated or grazing or fed treated crops or crop commodities. Studies are usually conducted using goats or cows (to represent the ruminants) and chickens. A monogastric animal (for example, a pig) study is not normally required because data are available from studies with rats. However, if metabolism in the rat is different from that in the ruminant and chicken, a pig study will be required.

The aims of livestock animal metabolism studies are:

2.3.4.1 To provide an estimate of the total terminal residues in the edible animal commodity.

2.3.4.2 To identify the major components of the total terminal residues.

2.3.4.3 To indicate the distribution and nature of residues in muscle, fat, milk, eggs, liver, and kidney, to identify target tissues and determine whether the residues are fat-soluble.

2.3.4.4 To show the efficiency of extraction procedures for various components of the residues.

2.3.4.5 To identify bound residues

2.3.4.6 To assist in determining residue definitions for enforcement and risk assessment.

Measuring the mode and extent of excretion or elimination of the parent compound and/or its degradation products in livestock to identify any potential for bioaccumulation is optional.

Further information is available at the OECD guidelines for the testing of chemicals website and the Food and Agriculture Organization's JMPR Guidance and related documents website; Joint FAO/WHO Expert Committee on Food Additives (JECFA).

An investigation of the extent of dermal absorption of the active substance or product is desirable for risk assessment. In the absence of dermal absorption data, a default value of 100% substance applied to the skin is assumed as a worst-case value. Applicants can refine the dermal absorption value by exploring further sources of information to estimate dermal absorption.

For dermal absorption studies provided in support of an application, the tested formulation should be identical to, or closely resemble, the product under consideration. The adequacy of this similarity will be determined on a case-by-case basis. Tested concentrations should represent expected human exposure concentrations; for example, the concentration of chemical in the product and

the proposed end-use concentration(s) should be tested. Submission of *in vitro* dermal absorption studies using (1) rat and (2) human skin in conjunction with (3) an *in vivo* rat dermal study (six pack') is recommended to enable likely human dermal absorption to be estimated. The adequacy of dermal absorption data that does not follow the six-pack approach will be assessed on a case-by-case basis and may be of reduced value for risk assessment purposes.

Further guidance on conducting and interpreting acute toxicity study can be found in the annex of this guideline.

2.3.5 Toxicity Studies for Technical Active Substance

The toxicological assessment of ectoparasiticides active substances involves the independent evaluation of data that inform on their effects on animal and human health. These data are used to derive conclusions on the toxicity of the ectoparasiticides substance which are used to make formal regulatory recommendations. These data are submitted by chemical companies to the regulators to support the registration process, which should include metabolism and kinetic investigations, single and repeated dose toxicity studies ranging from acute to lifetime *via* oral, dermal or inhalation routes, tests on mutagenicity and reproductive (fertility and developmental) effects and other specific investigative studies.

2.3.6 Acute Toxicity Studies

Acute toxicity studies examine the adverse effects arising from administration of a single oral dose or a single dermal or inhalation exposure of a substance over a specified period or multiple doses given within 24 hours.

To allow assessment of the acute toxicology of a substance, studies in animals should examine the most likely routes and forms of exposure in humans and animal.

Acute oral toxicity studies should be performed in at least one mammalian species. Rats are the preferred rodent species for oral studies unless a species more representative of human toxicity is known. Additionally, provide acute dermal and inhalation studies in at least one species. For skin and eye irritation studies, rabbits are an acceptable species, but alternatives from adopted OECD guidelines for the testing of chemicals (or other recognised guidelines) to the usual *in vivo* test may be suitable. *In vivo* eye irritation tests may not be appropriate in certain circumstances. If an eye irritation study waiver is requested, provide a valid scientific argument as to why these studies should not be included. For example, if the results from a skin irritation study or validated *in vitro* study demonstrated corrosivity or severe irritation, it is acceptable not to test the product in an eye irritation study, as it is presumed that the product will be corrosive to the eye. Similarly, products with pH extremes of 2 or less, or 11.5 or more are considered corrosive to the eye, unless the acid or alkaline reserve (buffering capacity) of the product suggests otherwise.

A skin sensitisation study is performed to test for possible hypersensitivity reactions to the substance. Guinea pigs are normally used for sensitisation studies.

For a veterinary ectoparasiticides product, submit a 'six-pack' of acute toxicological data. This consists of the following studies on the product:

2.3.7 Acute Oral in Rats: where acute toxicity data on the formulation is available, it would be used to determine whether the value of 1500 milligram per kilogram body weight is not appropriate and may be increased.

Whether one or two swallows (approximately 10 gram or 10 millilitre) of the product presents an acutely toxic dose to an infant or small child.

2.3.7.1 Acute Dermal in Rats: A veterinary ectoparasiticides product should not be acutely toxic at dermal doses up to 2000 milligram per kilogram body weight**.**

2.3.7.2 Acute Inhalation in Rats: A veterinary ectoparasiticides product should not be acutely toxic at inhalational concentrations up to 2000 milligram per cubic metre (four-hour exposure) for a gas, 20 milligram per litre (four-hour exposure) for a vapour and 5 milligram per litre (four-hour exposure) for dusts and mists.

2.3.7.3 Eye Irritation in Rabbits: The irritancy to eyes of veterinary ectoparasiticides products should be low. The formulation and application methods of a product will be taken into consideration on a case-by-case basis. Provide relevant information regarding any risk mitigation measures available for the proposed product**.**

2.3.7.4 Skin Irritation in Rabbits; The irritancy to skin of veterinary ectoparasiticides products should be low. The formulation and application methods of a product will be taken into consideration on a case-by-case basis. Provide relevant information regarding any risk mitigation measures available for the proposed product.

2.3.7.5 Skin Sensitisation Study (In Guinea Pigs)

If such data are not available, provide valid scientific argument as to why you have not submitted data. In certain circumstances, a toxicological evaluation of the product may be conducted by taking the known toxicological properties of the active Substances and excipients in the formulation and extrapolating these to estimate the acute toxicity of the product. We recommend that you adequately address the reasoning for not providing toxicity studies.

2.3.7.6 Repeated Exposure

Provide data to demonstrate that veterinary ectoparasiticides products are of low risk on repeated use and will therefore not induce irreversible toxicity effects.

2.3.8 Short-term Toxicity Studies (Repeat-Dose Studies for 21- Or 28-Days Dermal Toxicity (In Rats)

Short-term toxicity studies involve multiple administration of a substance for periods of less than 90 days. Such studies provide information on the possible health hazards likely to arise from repeated exposures over a limited period.

For classes of chemicals that cause cholinesterase inhibition, short-term oral (gavage) studies in animals, incorporating frequent monitoring of cholinesterase levels, are desirable.

2.3.9 Sub-chronic Toxicity Studies (90 Days to Less Than 12 Months)

Sub-chronic toxicity studies are performed to assess possible effects observed in short-term repeated exposure and as preliminary dose range-finding studies before chronic studies are started. They should demonstrate a range of activity, from the no-observed-effect level through to a toxic effect level. Often this range can be encompassed in a single study using one control and three test groups a non-rodent species. Dogs are the commonly used non-rodent species. Rabbits are not considered an acceptable non-rodent species unless available data suggest that they are more relevant for the prediction of health effects in humans.

2.3.10 Long-term (Chronic) Toxicity Studies (12 Months or Longer)

2.3.10.1 Chronic Toxicity Studies

Provide long-term (chronic) studies to assess long-term toxic effects (chronic toxicity) that may not be demonstrable in sub-chronic studies (for example, from cumulative toxicity).

Chronic toxicity studies normally consist of long-term, continuous, daily oral administration of the test compound to two species. The use of both a rodent and non-rodent species is desirable to provide an adequate assessment of interspecies variation. Rats and dogs are the preferred species. If you do not provide long-term studies in both a rodent and non-rodent species, we will consider other information you have provided, such as the findings from the sub-chronic studies in rodent and non-rodent species in relation to differences in species sensitivity and target organ toxicity (taking into consideration potential dose-spacing issues). The absence of chronic toxicity studies in both a rodent and non-rodent species may be considered a significant data omission or may require an additional safety factor to be implemented to account for observed differences in species sensitivity.

In chronic toxicity studies, it is desirable to have a dose–response relationship as well as a no-observed-effect level. To this end, normally one control and at least three test groups should be used. The highest dosage should induce a recognisable toxic response without eliciting excessive lethality. At least one dosage level should result in no observed toxic effects. Where a no-observed-effect level is not achieved and the study is identified as the key study for risk assessment purposes and/or establishing an acceptable daily intake value, an additional safety factor may be implemented to account for the uncertainty regarding a lower limit of toxicity.

2.3.10.2 Carcinogenicity Studies

Carcinogenicity studies are normally performed in two species. Such studies should be regarded as relevant whenever biologically significant residues of the compound or its metabolites occur, or when human exposure to the compound results from the normal use pattern of the compound.

Carcinogenicity testing may not be relevant for an active Substance that is not intended for food-producing use, has a restricted use pattern, and is not an *in vivo* somatic cell genotoxicant, and where findings in available systemic toxicity data do not raise concerns for carcinogenicity (for example, absence of pre-neoplastic lesions).

Carcinogenicity studies involve administration of the test material, usually in the feed, throughout the major portion of the life span of the species. An adequate number of animals should be included at each dose level to enable suitable statistical evaluation of the results (that is, most of the animals should survive for the duration of the study). It is recommended that rodent species such as rats and mice be used. The use of non-rodent species may be considered when available data suggest that they are more relevant for the prediction of health effects in humans.

You should present historical data describing the normal occurrence of a finding in the particular species and strain of animal in the testing laboratory for the route of administration tested. This assists in deciding whether or not a tumour or lesion is compound related. The submission of historical control data not from the testing laboratory, and/or not by the route of administration that the test used, may be of reduced or no regulatory value.

Where a tumour is considered to be of low relevance to humans, provide a supporting scientific argument, based on mechanistic data identifying the mode of action and using a weight-of-evidence approach for the identified mode of action to human health based on the Bradford Hill criteria.

The regulatory value of scientific arguments that tumour findings were of low relevance to humans, will be determined on their merits and reliability.

2.3.10.3 Combined Chronic Toxicity and Carcinogenicity Studies

Oncogenicity study (not less than 24 months for rats and 18months for mouse. This study can be combined with chronic feeding study, if appropriate). A combined chronic toxicity and carcinogenicity study may provide information on the possible chronic and carcinogenic effects likely to arise for a period lasting up to the entire life span of the species. However, careful design is suggested because information for each objective may differ.

You should present historical data describing the normal occurrence of a finding in the particular species and strain of animal in the testing laboratory for the route of administration tested. This assists in deciding whether or not a tumour or lesion is compound related. The submission of historical control data not from the testing laboratory, and/or not by the route of administration that the test used, may be of reduced or no regulatory value.

Where a tumour is considered to be of low relevance to humans, provide a supporting scientific argument, based on mechanistic data identifying the mode of action and using a weight-of-evidence approach for the identified mode of action to human health based on the Bradford Hill criteria.

The regulatory value of scientific arguments that tumour findings were of low relevance to humans, will be determined on their merits and reliability.

2.3.11 Reproductive Studies (2 Generations of Rodents and One Litter)

Reproductive studies involve the administration of a substance over one or more generations (multi-generation studies) to provide information on the effects of the substance on male and female reproductive systems, including gonadal function, the oestrus cycle, mating behaviour, conception, gestation, parturition, lactation, and weaning, and the growth and development of the offspring.

Such studies may also provide information about the effects of the test substance on neonatal morbidity, mortality, and preliminary data on prenatal and postnatal developmental toxicity, and serve as a guide for subsequent tests. These studies should be conducted with at least three dose groups and a concurrent control group, and would normally be conducted using rodents, preferably rats.

If other species are used, justification should be given, and the test parameters should be modified as appropriate.

2.3.12 Developmental Toxicity Studies.

Developmental studies involve administration of a substance to pregnant animals over a specified period of gestation (organogenesis) to provide information on prenatal exposure on the pregnant test animal and on the developing foetus, and may include assessment of maternal effects as well as death, structural anomalies and abnormalities, or altered growth in the foetus. Functional deficits, although an important part of development, are generally assessed in reproduction and developmental neurotoxicity studies.

Developmental toxicity studies should be performed in a rodent and non-rodent species. Rats are the preferred rodent species and rabbits are the preferred non-rodent species. Provide justification if another species is used.

2.3.13 Genotoxicity Studies

It is now known that some substances can cause changes to the genetic material. These changes may involve a single gene, or whole chromosomes (structural and/or numerical), and damage to deoxyribonucleic acid (DNA) via effects such as unscheduled DNA synthesis, DNA strand breaks, DNA adduct formation or mitotic recombination. A set of well-validated tests able to detect different classes of genetic toxicants will demonstrate the potential of a compound to induce genetic damage in humans. Tests (i) and (ii) described below should be conducted in the first instance:

2.3.13.1 a test designed to demonstrate the induction of point mutations (base-pair substitution and frameshift) in a microbial assay (for

example, salmonella reverse mutation test), with and without the use of appropriate metabolic activation systems.

2.3.13.2 a test designed to demonstrate the production of chromosome damage in an *in vitro* mammalian cell assay (for example, Chinese hamster ovary assay), with and without the use of appropriate metabolic activation systems.

2.3.13.3 An *in vivo* test is also recommended.

2.3.13.4 If (i) or (ii) are positive, two of three tests described below under (iii), (iv) and (v) should be carried out in rodents (rats or mice) in order to characterise the genotoxic potential *in vivo* in somatic cells:

2.3.13.5 a test designed to demonstrate the production of cytogenetic damage (for example, micronuclei) in the bone marrow or other proliferative cells of intact animals

2.3.13.6 a test designed to demonstrate genotoxic damage, involving other than cytogenetic endpoints (for example, unscheduled DNA synthesis or P32-post-labelling adduct formation) and preferably in a suspect or known target tissue for the substance

2.3.13.7 a test designed to demonstrate mutations in transgenic rats or mice that have transgenes containing reported genes for the detection of various types of mutations in somatic tissues.

2.3.13.8 If (iii), (iv) or (v) are positive, a test described below under (vi), (vii) or (viii) should be carried out in rodents (rats, mice or Chinese hamsters) in order to better characterise the genotoxic potential *in vivo* in germ cells:

2.3.13.9 a test designed to demonstrate a dominant lethal event in a germ cell that does not cause dysfunction of the gamete, but which is lethal to the fertilised egg or developing embryo.

2.3.13.10 a test designated to demonstrate the production of chromosome aberrations in spermatogonial cells.

2.3.13.11 a test designed to demonstrate mutations in transgenic rats or mice that have transgenes containing reporter genes for the detection of various types of mutations through the germline.

2.3.14 Teratogenicity Study (2 Species, One Rodent and One Non-Rodent)

This study for developmental toxicity testing is designed to provide general information concerning the effects of prenatal exposure on the pregnant test animal and on the developing organism; this may include assessment of maternal effects as well as death, structural abnormalities, or altered growth in the foetus. Functional deficits, although an important part of development, are not a part of this Guideline. They may be tested for in a separate study or as an adjunct to this study using the Guideline for developmental

neurotoxicity. Reference should be made to OECD guideline for the testing of chemicals.

2.3.15Neurotoxicity Studies

A neurotoxic effect is an adverse change in the structure or function of the nervous system (central or peripheral) that results from exposure to a substance. A neurotoxic effect may arise in offspring from exposure of the mother during pregnancy and lactation. Adverse changes may result from single or repeat exposure to a substance.

Tests should be designed to detect or characterise major neurobehavioral and neuropathological effects in test animals. While behavioural effects—even in the absence of morphological changes—can reflect an adverse impact on the organism, not all behavioural changes are specific to the nervous system. Therefore, any changes observed should be evaluated in conjunction with correlative histopathological, haematological, or biochemical data as well as data on other types of systemic toxicity. A developmental and delayed neurotoxicity study should be considered based on all the available information. A developmental neurotoxicity study should be conducted when neurotoxicity is observed in acute or repeat dose studies. Delayed neurotoxicity studies (acute and repeat dose) should be conducted if the substance is an organophosphorus compound.

Relevant testing (acute, sub-chronic, developmental, and delayed neurotoxicity) should be conducted in accordance with appropriate test guidelines.

2.3.16 Acute Delayed Neurotoxicity in Hens (For Organophosphates and Carbamates)

The objective of this delayed neurotoxicity study is to assess the potential of certain classes of substances to cause delayed neurotoxicity or delayed polyneuropathy following acute exposure. The delayed neurotoxicity study following acute exposure is recommended if the test material is an organophosphorus substance, which includes uncharged organophosphorus esters; thioesters or anhydrides of organophosphoric, organophosphonic, or organophosphoramidic acids; or of related phosphorothioic, phosponothioic, or phosphorothioamidic acids; or is structurally related to other substances that may cause the delayed neurotoxicity sometimes seen in this class of chemicals.

The delayed neurotoxicity study is conducted with the adult domestic hen using the technical grade.

2.3.17 Repeated Dose 28 Days Oral Delayed Neurotoxicity in Hens (Organophosphates and Carbamates if Triggered by Findings of Acute Delayed Neurotoxicity).

The objective of the 28-day delayed neurotoxicity study is to assess the potential of certain classes of substances to cause delayed neurotoxicity or delayed polyneuropathy following repeated exposure. The 28-day delayed neurotoxicity study is recommended if results of acute neurotoxicity study

indicate significant statistical or biological effects or if other available data indicate the potential for this type of delayed neurotoxicity, as determined by the responsible authority.

The 28-day delayed neurotoxicity study is conducted with the adult domestic hen using technical grade Active substance.

2.3.18 Additional Studies

2.3.18.1 Toxicity Studies of Metabolites and Impurities (At Least One Species)

Although it is recognised that toxicity studies usually examine the toxicity of the active substance, impurities or metabolites may contribute significantly to the toxicity of the compound. In general, studies employing the active substance provide an overall estimate of toxicity of the parent compound and its metabolites. However, where metabolites produced in target animals are significantly different from those produced in laboratory animals, provide toxicity studies on those metabolites. Submitted data should allow an assessment of what metabolites should be included in the residue definition for risk assessment purposes.

All impurities with concentrations of one gram per kilogram or greater (or those impurities with concentrations of less than one gram per kilogram that are toxicologically significant) in the active substance should be identified and, where necessary, subjected to appropriate toxicological studies or a scientific justification.

If you will be providing toxicological studies, provide at least one acute oral and one *in vitro* genotoxicity study (as outlined under the heading '*Genotoxicity studies'*. These studies should be conducted on the listed impurities and metabolites and if these are demonstrated to be more hazardous than the active substance, they may be identified as toxicologically relevant for risk assessment purposes.

Toxicity studies on metabolites are done on a case-by-case basis, considering the amount of metabolite and the chemical structure of the metabolite compared to the parent structure.

2.3.18.2 Other Adverse Effects

Individual compounds that show specific toxicological effects (for example, immunotoxicity, neurotoxicity) during normal repeat dose testing should be further investigated using appropriate tests for the abnormalities induced to enable definitive hazard characterisation to be established. Similarly, new compounds that belong to chemical classes known to produce particular toxicological effects should also be tested appropriately, for example, delayed neurotoxicity with organophosphorus insecticides. In the absence of such information, the toxicity profile of a compound may be deemed incomplete. The regulatory impact of an incomplete toxicity profile will be determined on a case-by-case basis.

2.3.18.3 Toxicity of Mixtures

Where two or more active Substance s are formulated together in a novel combination (that is, a hazard profile has not been previously established), toxicity studies should be performed with the formulated product to investigate the possibility of synergism or potentiation. In the absence of data, provide a scientific argument so that a determination of the regulatory value of the argument can be considered. When undertaking toxicological studies, acute toxicity studies are usually sufficient for this purpose (that is, acute oral, dermal, and inhalational toxicity studies, skin and eye irritation studies, and a skin sensitisation study).

Where synergism or potentiation is found, further studies or scientific argument may be necessary to clarify its toxicological significance.

2.3.18.4 Mechanistic Studies and Mode of Action

Mechanistic studies may be undertaken to supplement data obtained from standard studies conducted in accordance with the adopted OECD guidelines for the testing of chemicals, or other recognised test guidelines, so as to explain the process involved in, or responsible for, an observed toxicological finding. Together, the data may identify the overall mode of action by which a substance produces its toxicological effect, from a subcellular level through to histopathological changes.

When proposing that the observed toxicological effect is of low relevance to humans, provide a scientific argument based on the available data, identifying the mode of action and using a weight-of-evidence approach for the relevance of the identified mode of action to human health based on the Bradford Hill criteria.

2.3.19 Immunotoxicity

2.3.19.1 Introduction

An immunotoxic effect is an adverse effect on the components and/or function of the immune system from exposure to a substance resulting from either direct or indirect actions reflecting either permanent or reversible toxicity.

While OECD test guidelines for short-term, sub-chronic and chronic toxicity studies may provide data to give an indication of immunological effects, there is no specific OECD test guideline to determine functional immunotoxicity. If such studies provide an indication of an immunological effect, you should consider further testing to investigate immunotoxicity using appropriate tests. Reference should be made to US Environmental Protection Agency that has a functional immunotoxicity test guideline designed to evaluate the immunosuppressive potential of a substance (OPPTS 870.7800).

2.3.19.2 Endocrine Disrupting Properties

The active substance that may have endocrine disrupting properties, additional information or specific studies should be provided including the mode or mechanism of action. Sufficient evidence for relevant adverse effects should also be provided.

The applicants can make reference to EPA, ECHA and EFSA, 2016 guidance documents.

2.3.20 Human Toxicological Data (Such as Industrial Exposure Data, Accidental Data, or Volunteer Data).

Provide all available information relating to human experience with the substance. The information may arise because of voluntary intake, occupational exposure during the manufacture of the substance, worker exposure during use, or reports of accidental poisoning on humans by direct observations, epidemiological studies, diagnosis of poisoning, proposed treatment, and expected effects of poisoning. A brief description of effects to humans using the product should be described and, where appropriate, precautions during preparation and use of the product should be proposed.

Include studies relating to occupational and/or worker exposure in Occupational health and safety section of your application.

These data and information should be used to define and state the symptoms of poisoning and the effectiveness of first aid and therapeutic measures. Such data and information shall include reports of any studies investigating antidote pharmacology or safety pharmacology. Where relevant, the effectiveness of potential antagonists to poisoning shall be investigated and reported.

A statement should be provided on the effects of human exposure and extrapolations made with respect to target organs. A clear communication should be provided on the reversibility of adverse effects. Such data may be generated following accidental, occupational exposure or incidents of intentional self-poisoning, and shall be reported if available.

2.3.21 Acceptable Daily Intake for Humans (Toxicological)(ADI)

The acceptable daily intake for humans is the level of intake of a substance that can be ingested daily over an entire lifetime without appreciable risk to health based on the available information at the time of evaluation. It is expressed in milligrams of the substance per kilogram of body weight per day (mg/kg bw/day).

For this purpose, 'without appreciable risk' means that adverse effects are unlikely to result even after a lifetime of exposure. The acceptable daily intake is intended to give a guide to the maximum amount of a substance that can be ingested daily in the food without appreciable risk to the consumer. Accordingly, the figure is derived as far as possible from feeding studies in animals.

2.3.22 Acute Reference Dose (Toxicological)

The acute reference dose of a substance is an estimate of the amount of a substance in food and/or drinking water, expressed in milligrams of substance per kilogram of body weight (mg/kg bw), that can be ingested over a short period of time, usually in one meal or during one day, without appreciable health risk to the consumer, on the basis of all known facts at the time of the evaluation. For some substances, an acute reference dose may not be

necessary because the substance is not considered to cause appreciable acute risk after a single dose or exposure (that is, 24 hours or less).

2.4 ECO-TOXICOLOGY FOR NEW MOLECULES ONLY

Provide either an executive summary or individual summaries of studies on the behaviour of the Veterinary ectoparasiticide Product in the environment.

2.4.1 Acute Toxicity to Birds and Mammals

Birds (2 species): Provide details of at least one land and one water bird, LD50 in mg product/kg bird. Weight and the NOAEL. Furthermore, provide information on the effect on reproduction.

Acute toxicity to birds and mammals where possible, the test should provide for birds and mammals, the LD50 values, the lethal threshold dose, time courses of response and recovery and the no observed effect dose (NOED) for lethality and must include relevant gross pathological findings. Study design should be optimised for the achievement of an LD50 rather than for any secondary endpoint.

2.4.2 Birds

For the acute oral toxicity studies of an active substance a quail species (Japanese quail, Coturnix coturnix japonica or bobwhite quail, Colinus virginianus) should be used. The highest dose used in tests need not normally exceed 2000 mg/kg body weight. Due to issues of regurgitation, it is recommended not to use the mallard duck. Where regurgitation or emesis occurs at doses used for risk assessment, additional information is required to complete the risk assessment. The amount of regurgitated material should be assessed for determination of the ingested dose. In the absence of this information, the lowest overall no observed effect level (NOEL) should be used for risk assessment purposes. Where more than one study has been submitted, it is recommended to use the study where no regurgitation has occurred. If, however, mortalities appear in the study in which regurgitation has occurred (at dose levels at or around the LD50value for the non-regurgitation study), then it is proposed to use the NOEL from the study where regurgitation has occurred.

2.4.3 Mammals

The following acute oral test methods with mammals are available (LD50 mg/kg bw):

> *OECD Test 420 (OECD, 2001a): Acute oral toxicity – fixed dose procedure*
> *OECD Test 423 (OECD, 2001b): Acute oral toxicity – acute toxic class method OECD Test 425 (OECD, 2006c): Acute oral toxicity – up-and-down procedure 22* Organisation for Economic Co-operation and Development Risk assessment for birds & mammals.

The fine details of the above studies vary but the underlying principles are the same. Animals (normally rats) are dosed once by oral gavage and observed for 14 days. Observations include body weight, clinical signs, death, and necropsy findings. A limit dose of 2000 mg/kg bw or 5000 mg/kg bw (depending on

study) should not be exceeded. The fixed dose procedure and the acute toxic class method are range estimators and are useful for mammalian wildlife risk assessment only in cases where they can be used as a limit test (e.g., > 2000 mg/kg bw), or to provide a conservative surrogate for the LD50 (i.e. lowest value of range).

2.4.4 Standard Toxicity Tests with Aquatic Organisms

The following toxicity tests should be submitted for every substance even when it is not expected that preparations containing it could reach surface water following the proposed conditions of use: acute toxicity to fish (Oncorhynchus mykiss), acute toxicity for Daphnia species (preferably for Daphnia magna) and effects on the growth for a green alga. A limit test at 100 mg substance/L may be performed when the results of a range finding test indicate that no effects are to be expected. To minimise fish testing, a threshold approach to an acute fish test should be considered (OECD, 2010). An acute fish limit test should be conducted at 100 mg substance/L or at an appropriate concentration selected from aquatic endpoints following consideration of the threshold exposure. Other considerations for setting the limit in a limit test could be compound properties (e.g. water solubility, or needs for RA. When mortality is detected in the fish limit test, an acute fish dose–response toxicity study should be performed to determine an LC50 for use in RA. The endpoints required for toxicity tests in the revised data requirements 'for the standard tests and the other requested additional types of tests are EC10, EC20 and EC50, together with their 95 % confidence intervals (or an explanation if they cannot be estimated) and corresponding NOEC values. How these should be determined is explained below

2.4.5 Fish

Fish (2 species): Provide details on at least two species studied, LC50 (in mg of product / litre of water) and the NOAEL. Furthermore, provide information on the effect on reproduction. Indicate the bio-concentration factor (BCF) on the active substance in tissues.

Acute toxicity to fish A 96-h test on rainbow trout (Oncorhynchus mykiss) shall always be carried out for active substance, even when it is not expected that the compound will end up in the surface water. In that case it will be used for classification and labelling. Consideration should be given to allow the reduction of animal testing.

Chronic toxicity to fish Circumstances in which required A long-term or chronic toxicity study on fish is required for all active substance. where exposure of surface water is likely and the substance is deemed to be stable in water, that is, there is less than 90 % loss of the original substance over 24 h via hydrolysis at all relevant pH values (hydrolysis DegT90 > 24 h). An early-life stage study is always required in these circumstances; however, if a full fish life cycle study has been generated, an early-life stage study is not required. The fish early-life stage test should determine effects on development, growth, survival and behaviour, and details of observed effects on fish early-life stages. A full fish life-cycle test may be required depending upon the persistence and bioaccumulative potential of the substance.

OECD Test No. 223: Avian Acute Oral Toxicity Test
OECD test No. 236: Fish Embryo Acute Toxicity (FET) Test

2.4.6 Daphnia: LC50 mg/l

The applicant should provide data using studies using Daphnia as a representative invertebrate. Acute toxicity data and chronic data should be provided if there is continued or repeated exposure due to frequent application. Chronic data are therefore required for compounds that are applied more than once, or for those whose dissipation rate (DT50) in water is greater than or equal to 2 days. It should be noted that short-term exposure may lead to sublethal effects which are not covered by acute toxicity testing. If there are such concerns in special cases further evaluations might be provided.

Test No. 211: Daphnia magna Reproduction Test

2.4.7 Algae: LC50 mg/l

A test with green algae is required in all cases. The second species should be from a group other than green algae, such as diatoms or the blue-green algae. Comparisons between the endpoints growth rate and biomass have been made and concluded that biomass - or cell number – is usually the most sensitive endpoint. Nevertheless, both biomass and growth rate should be reported. As there is no clear evidence available to indicate which is the most relevant endpoint for the field situation the lower figure should be used in the risk assessment. Toxicity values should be based on the period of exponential growth.

Standard toxicity tests with algae A 72- to 96-h test should always be carried out on one green alga (such as Pseudokirchneriella subcapitata, synonym Selenastrum capricornutum). The algal test (OECD guideline 201) is a short-term test, although it provides chronic endpoints.

Test No. 201: Freshwater Alga and Cyanobacteria, Growth Inhibition Test

2.4.8 Bees: LC50 µg/Bee

Data requirements and testing Acute toxicity to bees. If honeybees are likely to be exposed to the active substance both acute oral and contact toxicity tests must be conducted as the toxicity by one route of exposure cannot be predicted from the other. Where there is only one relevant route of exposure (e.g. oral exposure in the case of soil application), testing can be restricted to this exposure route. The test result should be presented as µg active substances/bee or g formulation/bee. If there are problems with solubility of the active substance, then the test should be conducted with a representative formulation.

tests should be conducted according to EPPO 170, or OECD 213 and OECD 214 guidelines.

2.4.9 Soil Organisms

2.4.9.1 Earthworms: LC50 mg/kg

Testing is always required where contamination of the soil is possible.

Calculate predicted environmental exposure (PEC) soil values using the realistic worst-case best fit $DT_{50/90}$ values. This should be the longest non-normalised $DT_{50/90}$ values derived from field dissipation studies. In the absence of field data, use normalised laboratory data (to 20°C and pF2 using the Arrhenius and Walker converter from tools. The risks arising specifically from the active substance(s) in the formulated product and from the formulation must be assessed. These assessments use toxicity data on the active substance and/or on the formulation; exposure estimates are based on the proposed use. The soil risk assessment uses a soil predicted environmental exposure (PEC) calculated by Environmental Fate. Acute effects on earthworms testing are always required where contamination of the soil is possible.

2.4.9.2 Effects on Soil Nitrogen Transformation

A test shall provide sufficient data to evaluate the impact of active substances on soil microbial activity, in terms of nitrogen transformation.

2.4.10 Effects on Other Terrestrial Organisms (Flora and Fauna)

Any available data on the effects of the product on other terrestrial organisms shall be submitted.

2.4.11 Effects on Biological Methods for Sewage Treatment

A test shall provide an indication as to the potential of the active substance on biological sewage treatment systems.

2.4.12 Monitoring Data

Available monitoring data concerning adverse effects of the active substance to non-target organisms shall be reported.

EFSA Guidance on tiered risk assessment for plant protection products for aquatic organisms in edge-of-field surface waters.

EFSA Risk Assessment for Birds and Mammals

SECTION 3.0 PARTICULARS OF THE FORMULATED PRODUCT (ECTOPARASITICIDES)

The EAC veterinary medicinal products regulators require that a product including veterinary ectoparasiticides be registered before distribution, sale or use in East Africa, regardless of whether the product is formulated in the region or overseas.

The formulation and manufacturing data should be submitted to support the registration of a veterinary ectoparasiticides. This guideline is applicable to products that are prepared by chemical synthesis. Details about information to support applications to register products that are not conventional ectoparasiticides will be provided in a separate guideline.

3.1 Veterinary Ectoparasiticides (formulated product)

3.1.1 Trade Name, Brand Name or Trademark of the Product

The distinguishing product name is the name by which the product is (or is to be) known, registered, labelled, advertised, and sold. It should be unique to a particular product and include such descriptive words or phrases as are needed to distinguish the product from other products and to identify the purpose for which it is to be used.

3.1.2 Applicant, Formulator, and Formulation Facility Details

Applicant name, company address, contact name, telephone number and e-mail address. The applicant should provide the name, physical address, and address of all formulating facilities—including the GPS coordinates—involved in any step of the manufacture of the product. Provide names of all co-formulants in the product and indicate whether they influence the toxicity of the product. This includes toll or contract manufacturers and subcontractors involved in packaging and labelling, and testing, up to and including release for supply, stating the relationship with the marketing authorization holder.

Information on whether the applicant is to import or manufacture (including repacking, formulating, and manufacturing from raw materials) the ectoparasiticide product. If the application is for manufacturing in the region, the applicant should give the location of the manufacturing plant.

3.1.3 Type of Formulation

The applicant should indicate the type of the formulation to be registered (for example emulsifiable concentrate, granule, dustable powder or pour-on etc). If a description of the formulation type does not appropriately describe the proposed formulation type, refer to the FAO/WHO guidelines (JMPS 2010) to provide a formulation type.

3.1.4 Purpose of the Product and Target Animal Species

Brief description of the method and site of application of the veterinary ectoparasiticides. The application rate per animal treated and concentration of Active substance in the material as applied (for example, if the product is

diluted before application). Application and mixing instructions, including method of application, type of equipment used, application techniques and rates and amount per animal.

Number, frequency, and timing of applications (e.g., per month, per week) and proposed duration of protection.

Proposed instructions on how to use the product, including in a manner that protects human health and the environment (e.g., buffer zones; personal protective equipment such as gloves, coveralls, respirator; prohibited application areas such as aquatic areas and around homes; and prohibited tank mixes or incompatibility with other products).

Procedures for cleaning application equipment, if relevant to the proposed use

3.1.5 Classification of Active Substance by Hazard

The active Substance (s) proposed for use in the veterinary ectoparasiticides must be an approved Active Substance (s). Product registration holders must ensure the active substance used in the veterinary ectoparasiticides complies with the FAO and WHO specifications. Reference should also be made to Stockholm Convection list of active substances.

The following information about the active substance should be provided:

3.1.5.1 the name of the manufacturer and the street address of the manufacturing plant(s), and the approval number, where available

3.1.5.2 the batch analysis data for the batch of the active substance (analysed within the last two years) to be used in product formulation. The batch should be analysed for active substance content and impurities and analysed for any toxicologically significant impurity(ies) at any level.

3.1.5.3 full details of the analytical methods used for the determination of the active substance, any isomer and impurities should be provided.

3.1.5.4 appropriate validation data for the analytical methods used for the determination of the active Substance s and relevant impurities and toxicologically significant impurity(ies) should be provided for in-house methods.

3.1.6 Quantitative and Qualitative Information on the Composition of Formulation

Applicant shall provide a statement of the formula identifying each active substance, and inactive substances and, in certain cases, impurities that may be present in the product. The product composition data is used to determine whether the product contains any ingredient in an amount which may cause unreasonable adverse effects to the animal, human, and environment.

In addition to identifying the substances in the product, the applicant must also provide certified limits for the substances listed in the statement of

formula. Upper and lower certified limits must be established for each active substance and each intentionally added inactive substance. For some types of products, an upper (maximum) certified limit must be established for certain impurities that will be present in the product at any time while it is in market.

An applicant is required to submit a copy of available technical specifications by which the supplier of a starting material describes its composition, properties, and/or toxicity as well as any other information available to them concerning the composition and properties of the starting material. Applicants may not be required to analyse starting materials, but to submit information on purity of starting materials.

The formulation composition describes the qualitative and quantitative formulation of the product. Provide a full description of the formulation of the product to be marketed or supplied for the purposes covered by the application including:

- the common or chemical names of the substance and their identification or composition (International Organization for Standardization [ISO]), International Union of Pure and Applied Chemistry (IUPAC) name. In some cases, safety data sheets (SDS) may be submitted in lieu of the manufacturer's CoA. For proprietary non-active Substance s, specifications from the supplier should be provided.

- the Chemical Abstracts Service (CAS) Registry Number (if available)

- the concentration of all active and non-active Substance s in the formulation

- reference to the quality standards

- the purpose of the substance s in the formulation (for example, whether it is the active substance, a surfactant, an emulsifier, or a filler).

Based on the actual purity or potency of the active substance and all non-active Substance s, adjust the formulation composition by calculating the concentration of substance (s). For example, if the theoretical concentration of an active substance in a batch of product (that is, the label claim) is 275 grams per litre (g/L), and the purity of the technical active substance being used is 950 grams per kilogram (g/kg) (95 per cent weight per weight), then the factorised concentration of the technical active Substance to be added would be:

275 g/L÷ (950/1000) =289.5 g/L

Concentrations of technical active substance s, together with the stated nominal concentration of active substance (and any overage), adjuvants, and inert Substance s should be expressed in g/L for liquid formulations and g/kg for solid formulations. If these units are not appropriate for a particular formulation, the applicant should propose suitable units (for example, a biological unit).

Table 4: **Example of an Adequate Formulation Composition Statement**.

Substance name	**CAS Registry Number**	**Substance Standard**	**Concentration (g/L)**	**Purpose in formulation**
(a) Active Substance /s				
Active substance				
(b) Other Substance (s)				
Non active 1		MS		
Non active 2		MS		
Non active 3		MS		
TOTAL (w/v)				
Specific gravity (for liquid products) at 25 °C	N/A	N/A		N/A

MS = manufacturer's specification

3.1.7 Product Registration Status in Other Countries and WHO Restriction

Provide information on product authorizations in other countries, refusal of registration or cancellation of registration (including reasons). Provide any existing FAO and WHO assessments.

3.1.8 Risk Management Statement

3.1.8.1 Provide a statement about any risk for the development of resistance in the controlled ectoparasite and propose resistance prevention methods.

3.1.8.2. Withholding periods for animal products must be provided. Disposal procedures, detailed actions in the event of an accident during transport, storage or use and decontamination procedures

for use in the event of accidental spillage or fire.

3.1.8.3 Information on antidotes, if any, and medical treatment in the case of accidental exposure; names of co-formulants that may influence the toxicity of the product.

3.1.8.5 Proposed hazard classification, labelling and safety phrases and symbols.

3.1.8.6 Proposed complete, commercial label, packaging sizes, and materials and specimens of proposed packaging.

3.1.8.7 A statement about any risk arising from the recommended methods of application and precautions and handling procedures, to minimize those risks (e.g., precautionary statements of the *Globally harmonized system of classification and labelling of chemicals* [*8*])

3.2 Quality, Manufacturer & Control

3.2.1 Quantitative and Qualitative Particulars of the Product

3.2.1.1 Introduction

Veterinary Ectoparasiticides Product manufacturing involves the industrial synthesis of molecules produced or modified to provide compounds (active substances). Manufacturing goes through several steps and all steps, and particularly the reactions, may generate air emissions, effluents, and waste / by-products which should be appropriately controlled for safety in the manufacturing plant.

Veterinary Ectoparasiticides Product formulation process involves mixing, blending, or diluting one or more active substances and inert ingredients to obtain a product used for additional processing or as a final product; The formulations can be gas formulations (aerosols and fumigants), liquid formulations and solid formulations.

The finished Veterinary Ectoparasiticides Products are packed in container closure systems designed from materials that can effectively contain the ectoparasiticide to optimize its handling and application and reduce health or environmental risks to humans or ecosystems by preventing spillage.

The facilities where Veterinary Ectoparasiticides Product are manufactured, formulated, and packaged should employ current Good Manufacturing Practice (cGMP) procedures to ensure quality product.

3.2.1.2 Method of Manufacture

The following information must be provided for each process resulting in a separately isolated substance:

3.2.1.2.1 A general characterization of the process (e.g., whether it is batch or continuous) and quantity of product produced (lbs or kg) per theoretical batch size (or per unit time, if continuous).

3.2.1.2.2 Describe the key stages of manufacture detailing the precautions taken to ensure that the method produces a final product of consistent quality. Include quantitative descriptions of all the substances used during manufacture and indicate the steps at which they are added.

3.2.1.2.3 Provide a flow chart showing the intended chemical reactions occurring during each step of the process, and of the major unit operations, including separation steps. In addition, a flow chart showing the various stages of manufacture including, blending, and filling. Show the stages where sampling is taken for Quality Control tests, naming the tests performed at each stage should be provided.

And/or A detailed description of the manufacturing processes and in-process control for critical points. Inclusion of an executed batch manufacturing record for a commercial batch is recommended.

3.2.1.2.4 The identities of the reactants, solvents, and catalysts used to produce the product, their quantities in kilograms, and the order in which they are added.

3.2.1.2.5 Provide a description of the equipment used that may influence the composition of the substance produced, reaction conditions and details of the chemical reactions intended to occur.

3.2.1.2.6 A description of the conditions (e.g., temperature, pressure, pH, humidity) that are controlled during each step of the process to affect the composition of the substance produced, and the limits that are maintained.

3.2.1.2.7 A description of the purification steps (including those used to recover or recycle starting materials, intermediates, or the substance produced).

3.2.1.2.8 A description of the procedures used to assure consistent composition of the substance produced, e.g., calibration of equipment, sampling regimens, analytical methods, and other quality control measures.

3.2.2 Formulation Process and Quality Control

The applicant should provide a description of the method of formulation of the product and the sequence of operations, plus an indication of the typical size of the production-scale batch.

Full details of the quality control procedures used by the formulator to ensure batch-to-batch reproducibility of the product should also be provided. Information should include details of the control checks performed at various stages of the manufacture, processing, and packaging of the product. The description of the in-process testing should include the specifications and tests for pivotal and key or critical intermediates. A description of the process to deal with a product (out of specification) that does not comply to release specifications should be provided.

3.2.3 Cleaning of Validation Equipment

The applicant should provide a description of the method and specifications for cleaning of equipment between different batches that contain different active Substance s, non-active Substance s, or formulation.

3.2.4 Physical and Chemical Properties of the Formulated Product

The following data on the physical and chemical properties of the product should be provided.

Table 5: Physical properties of the Formulated product

Parameter	Standard	observation
Provide full visual description of the product including colour, odour, physical state, and other relevant features.		
Acidity, alkalinity, and pH value		
Relative density and bulk density (for solids)		
Density or specific gravity (liquids)		
Viscosity and surface tension (for liquids)		
Delivery form and characteristics of the formulation the product is presented, e.g., solution, suspension, spray, pour-on, wettability and persistent foam)		
flash point		
Flammability and self-heating.		
Explosive properties		
Oxidising properties		
corrosive hazard		
dangerous goods classification), if applicable.		

3.2.4.1 Chemical Properties of the Formulated Product

Table 6

Parameter	Standard	observation
Wettability		
Persistent foaming		
Suspensibility, spontaneity of dispersion and dispersion stability		
Degree of dissolution and dilution stability		
Particle size distribution, dust content, attrition and hardness and integrity.		
Emulsifiability, re-emulsifiability, emulsion stability.		
Flowability, pourability and dustability.		
Physical and chemical compatibility with other products including ECTOPARASITICIDEs with which its use is to be authorized.		

3.2.5 Potential for the Formation of Impurities of Toxicological Concern

Monitoring and reporting of toxicologically significant impurities present in the active substance and non-active Substance s in the product or formed during the formulation of the product and storage or by migration of packaging material into the product should be continuous.

3.2.6 Product Specifications

Provide product specifications, with suitable upper and lower limits for the active substance and the toxicologically significant impurities, and the relevant physical characteristics of the product.

The specification limits should consider the use of any overages in the formulation.

Specifications should address the test parameters for the particular formulation types, as outlined in the storage stability section.

Table 7 is an example of a suitable format for product specifications.

Table 7: Product Specifications

Test	Product Specification	Method
Active Substance		
Appearance		
pH		
Emulsion characteristics		
Persistent foam		
Density		

3.2.7 Batch Records of formulated product.

The product registration holders and/or the active substance supplier are required by regulator as a condition for product registration to maintain analysis records for each batch of the active substance used in their registered products. During GMP audit the active substance batch records held by veterinary ectoparasiticides registration holders may be verified.

3.2.8 Content of Active Substance —Allowable Variations

The difference between the stated label amount or concentration and the actual content of the active Substance s should not exceed the statutory limits for chemical products. The allowable active substance is specified in the FAO specifications (Table 8).

Table 8: Statutory Allowable Variations in the Declared Content of an Active Substance in Ectoparasiticide Products

Test	Product Specification
Up to 25	±15% of the declared content for 'homogeneous' formulations (EC, SC, SL, etc.) or ±25% for 'heterogeneous' formulations (GR, WG, etc.)
Above 25 up to 100	±10% of the declared content
Above 100 up to 250	±6% of the declared content
Above 250 up to 500	±5% of the declared content
Above 500	±25 g/kg or g/L

EC = emulsifiable concentrate; SC = aqueous suspension concentrate; SL = soluble concentrate; GR = granule; WG = water-dispersible granule

Note: In each range the upper limit is included. (Sourced from JMPS, 2010) Comments for Table 3

The allowable variations (tolerances) refer to the average analytical result obtained and take into account manufacturing, sampling, and analytical variations, except where an overage is required. Positive deviations from the upper limits given in the table may be utilised if the formulation is manufactured with an overage to compensate for degradation in storage.

3.2.9 Batch Analysis Data

Provide a detailed protocol on final product tests performed on each batch, including the batch release specification. The analyses must be designed to measure the amount of Active substance present in the product and to identify and quantify (if present) any impurity associated with an Active substance which is expected (based the theoretical discussion) to constitute 0.1 percent or more of the product.

Provide results from three production batches of the product to demonstrate compliance to the product specifications. The data should address all parameters listed for the product specifications.

Note: The initial results from stability trials may be sufficient.

3.3 Validation of Analytical Methods

Validation data for both accuracy and precision should be submitted for all methods. Applicants should provide information on the analysis of data gathered throughout the design and manufacturing of the product to confirm consistency of the production process in delivering quality products (e.g., batch to batch consistency providing for summary of the results of tests on five consecutive batches of veterinary ectoparasiticides. Product must be provided to support the application for registration of the product to demonstrate batch to batch consistency). These batches may be pilot or production batches. Full details of the results of the batch tests demonstrating conformity with the specifications should be included.

Provide validation data for the methods used for the determination of active substance and, where appropriate, relevant impurities.

3.3.1 Analytical Methods

Provide a full description of the analytical methods used for testing the product including details of equipment, materials and conditions used. The methods of analysis should be appropriate for the type of active substance and the formulation matrix of the product. Provide the following information to demonstrate the suitability of the analytical methods used to generate data for product registration: Competent authorities require applicants to provide samples of a certified analytical standard of active substance, the technical material used in the formulated product andanalytical standards of relevant metabolites and all other components included in all monitoring residue definitions. samples of reference substances for the relevant impurities.

- full details of the analytical methods used for determining the active Substance and, where appropriate, relevant toxicologically significant impurities in formulated veterinary ectoparasiticides during stability testing.
- a description and purity of the reference standards
- where chromatographic techniques are used, representative chromatograms of the
    - blank
    - reference standard
    - product sample
- chromatograms labelled with
    - batch number
    - peak identity
    - peak integration data
    - X and Y axis labels
- worked examples of the calculations.

The applicability of existing CIPAC methods shall be assessed and reported. In case of use of a CIPAC method, further validation data shall not be required, but example chromatograms shall be submitted, where available.

3.3.1.1 Regulatory Analytical Methods

The analytical methods for ectoparasiticides active Substance s and formulated chemical products described in the following documents are recognised as the regulatory methods:

- Handbooks of the Collaborative International Pesticide Analytical Council (CIPAC).
- The Association of Official Analytical Chemists' (AOAC International) Manual for agricultural active Substance s and agricultural chemical products.

The use of regulatory methods (if one is available) is recommended by the East Africa Veterinary ectoparasiticides and Veterinary Medicines Authorities.

It is recommended that, where available, you use the analytical methods described in the above documents for a particular active substance or formulated product.

The analytical methods described in these documents for a particular active Substance or formulation are regarded as validated and do not require revalidation. However, the applicant must verify the suitability of the method to be used under actual conditions of use (that is, selectivity and accuracy should be demonstrated for the published method when applied to the relevant sample matrix and laboratory conditions) and provide the data.

3.3.1.2 Alternative Analytical Methods

An alternative analytical method is an analytical method proposed by the registration holder for use instead of the regulatory analytical method. The applicant may use an alternative analytical method in place of a regulatory method if it is validated in accordance with this guideline.

3.3.2 Parameters to be Addressed.

- Selectivity or specificity
- Linearity
- Precision
- Recovery (accuracy)
- Limit of detection (LOD) and limit of quantification (LOQ) for relevant impurities and all toxicologically significant impurities.

Note: If the regulator has assessed the analytical methods in a previous application, you may refer to the data provided in that application. However, if the formulations are not identical, provide specificity and recovery (accuracy)

data to demonstrate that the analytical method is appropriate for use on the new formulation.

The objective of validation of an analytical method is to demonstrate that the method, when correctly applied, produces results that are fit for purpose. These guidelines describe the procedures to be carried out to validate the analytical method. The guidelines are included as part of the Part 2— Chemistry and manufacture dossier for an application for approval of an active Substance and registration of an agricultural chemical product, including those used in storage stability studies. They are not intended to apply to analytical methods for residue analysis, or to analytical methods for biological and biotechnological products. Approaches other than those set forth in this guideline may be acceptable, provided they are supported by adequate scientific justification.

Non-chromatographic analytical methods (e.g. titration methods) are not typically expected to comply with this guideline. However, we expect that users of non-chromatographic methods will provide some form of validation in order to satisfy us that the method is fit for purpose.

3.3.3 Data to Support an Analytical Method

The following information should generally be included to support the adequacy of the analytical method:

- Method description – this section should contain a full description of the analytical method. The description should include details of all-important operational parameters, such as details of sample preparation and reagents preparation (including method of extraction of the active Substance from the product), and details of the reference standards. Provide documentation confirming the purity of the reference materials.

- Validation data and all relevant data collected during validation, including:
    - copies of chromatograms that are clearly labelled with peak-identity and peak-integration data as well as X and Y axes with relevant scales.
    - nuclear magnetic resonance spectra, clearly showing chemical shifts and coupling constants.
    - formulae
    - examples of calculations used for calculating validation characteristics.

3.3.4 Parameters for Method Validation

To be fit for the intended purpose the method needs to meet certain validation characteristics. Typical validation characteristics that should be considered are:

- Selectivity (specificity)
- Linearity

- Range
- Accuracy
- Precision
- Limit of detection (LOD) and
- Limit of quantification (LOQ).

3.3.4.1 Selectivity (Specificity)

The selectivity of a method refers to the extent to which the method can determine particular analyte(s) in a complex mixture without interference from other components in the mixture. The terms selectivity and specificity have often been used interchangeably. The term selectivity generally refers to a method that provides responses for a number of chemical entities that may or may not be distinguished from each other, while the term specificity refers to a method that produces a response for a single analyte only. Since very few analytical methods respond to only one analyte, the use of the term selectivity is more appropriate than specificity.

The selectivity of the analytical method should be demonstrated by providing data to show that the analyte chromatographic peak is absent from interference peaks with regard to degradation products, synthetic impurities and the matrix (that is, excipients present in the formulated product at their expected levels). Such data include a peak homogeneity test or peak purity test (for example, diode array, or mass spectrometry) that shows the analyte chromatographic peak is not attributable to more than one component.

3.3.4.2 Linearity

The linearity is the ability of the analytical method to produce test results that are proportional to the concentration (amount) of analyte in samples within a given concentration range, either directly or by means of a well-defined mathematical transformation. Linearity should be determined by using duplicate determinations at 3 or more concentrations, or a single determination at 6 or more concentrations that span 80% to 120% of the expected nominal concentration.

The linearity of a method should be established by visual inspection of a plot of analytical response as a function of analyte concentration. If there is a linear relationship, test results should be evaluated by appropriate statistical methods (for example, by calculation of the regression line by the method of least squares). In some cases, the test data may need to be subjected to a mathematical transformation prior to regression analysis.

Your report(s) should include the:

- equation of the calibration line
- slope of the line
- intercept
- correlation coefficient (r).

The slope should demonstrate a clear correlation between response and analyte concentrations. The test results should not show a significant deviation from calculated results by the calibration equation – indicated by the correlation coefficient, r – greater than 0.99 over the range (80% to 120%). If r is less than 0.99, an explanation on how accurate calibration is to be maintained must be provided. In cases where a non-linear response is deliberately used, an explanation should be provided.

3.3.4.3 Range

The specified range is normally derived from the linearity studies. The range of an analytical method is the interval between the upper and lower concentration (amounts) of analyte in the sample for which it has been demonstrated that the analytical method has suitable levels of precision, accuracy, and linearity.

The following minimum specified ranges should be considered for the:

- assay of the active substance of an ectoparasiticides product, at least 80% to 120% of the nominal concentration
- determination of an impurity, at least from the specification level to 120% of the specification level.

3.3.4.4 Accuracy

The accuracy of an analytical method is defined as the degree to which the determined value of analyte in a sample corresponds to the true value. Accuracy may be measured in different ways and the method should be appropriate to the matrix. The accuracy of an analytical method may be determined in any of the following ways:

- In analysing a sample of known concentration and comparing the measured value to the 'true' value – the use a well-characterised sample (for example, a reference standard) should be made.
- The placebo (product matrix) recovery method, or ‘spiking’ – whereby a known amount of reference standard is added to a placebo sample (that is, a sample that contains all other ingredients except the active Substance [s]), the resulting mixture is assayed, and the results obtained are compared with the expected result.
- The standard addition method – whereby a sample is assayed, a known amount of reference standard is then added, and the sample is again assayed; the difference between the results of the 2 assays is then compared with the added amount.

Recovery is expressed as the percentage of the observed result to the expected result. The accuracy of a method may vary across the range of concentrations and therefore must be determined at several different fortification concentrations. The accuracy should cover at least 3 concentrations (80%, 100% and 120% of the nominal concentration) in the expected range.

Accuracy may also be determined by comparing test results with results obtained using another validated test method.

The acceptance criteria for the accuracy of the method are based on expected recovery. This depends on the sample matrix, the sample processing procedure and on the analyte concentration. The mean percentage recovery of each of the 3 concentrations should be within the ranges listed in Table 9.

Table 9: Acceptance Limits for Determination of Mean Recovery

Active Substance or impurity content (%)	Acceptable mean recovery (%)
>10	98 to 102
1.0–10.0	90 to 110
0.1–1.0	80 to 120
<0.1	75 to 125

3.3.4.5 Precision

The precision of an analytical method expresses the closeness of agreement (degree of scatter) between a series of measurements obtained from multiple sampling of the same sample under the same prescribed conditions. Precision may be considered at three levels: repeatability, intermediate precision, and reproducibility. For example, repeatability can be obtained by a minimum of 5 independent replicate sample determinations with the same method, on identical test material, on the same equipment, by the same operator in the same laboratory within short intervals of time.

The precision of an analytical procedure is usually expressed as the per cent relative standard deviation of a series of measurements. When applicable, a suitable test for outliers (Dixon's or Grubbs Test) may be applied to the results. Clearly indicate where outliers have been discarded and attempt to explain the reason for the occurrence of individual outliers.

The levels of precision we recommend are listed in Table 10.

Table 10: ***Acceptance Limits for Determination of Precision***

Component measured in sample (%)	**Precision** **(Per cent relative standard deviation, (%RSD))**
>10.0	≤2
1.0 to 10.0	≤5
0.1 to 1.0	≤10
<0.1	≤20

3.3.4.6 Limit of Detection (LOD)

The limit of detection (LOD) of an analytical method is the lowest amount of an analyte in a sample that can be detected, but not necessarily quantified as an exact value.

The LOD may be determined by analysing a series of samples with known concentrations of analyte and by establishing the minimum concentration at which the analyte can be reliably detected. That is, the LOD answers 2 questions:

- Is the analyte present and can it be reported?
- Is the analyte not present and can that be reported?

The lowest concentration that produces a detectable peak response corresponding to the analyte should be normally measured with between 6 and 10 replicates. You should calculate the average response (X) and the standard deviation (SD). The LOD is X + (3 × SD).

3.3.4.7 Limit of Quantification (LOQ)

The limit of quantification (LOQ) is the lowest amount of the analyte in the sample that can be quantitatively determined with defined precision under the stated experimental conditions. The limit of quantification is a parameter of quantitative assays for low levels of analytes in sample matrices and is relevant particularly for the determination of impurities, degradation products and low levels of active substance in a product.

If a preliminary study to determine the approximate LOQ is undertaken, then the LOQ may be determined by measuring a reference standard solution that was estimated during the preliminary study. The solution is normally injected and analysed with between 6 and 10 replicates. Calculate the average response (X) and the relative standard deviation as a per cent (%RSD) of the results. The

%RSD should be less than 20%. If the %RSD exceeds 20%, you should prepare a new standard solution of higher concentration and repeat this procedure. The LOQ is X + (10 × SD).

3.3.4.8 Applying the Parameters of Method Validation

The extent to which a method needs to be validated depends on its application.

The tests we recommend for consideration for each of the categories of analytical methods described in the guidelines are listed in Table 11.

Table 11: Parameters of Method Validation

Type of test or test characteristics	**Assay of active Substance in technical active Substance**	**Quantitative test for toxicologically significant impurities in technical active Substance and/or agricultural chemical product**	**Assay of active Substance in an agricultural chemical product**
Specificity	Yes	Yes	Yes
Linearity	Yes	Yes	Yes
Accuracy	No	Yes	Yes
Precision	Yes	Yes	Yes
Range	May be recommended, depending on the nature of the specific test	Yes	Yes
Limit of detection	No	Yes	No
Limit of quantification	No	Yes	No

3.4 Specifications of the Packaging Material and Labelling Requirements

The effect of the formulation on the packaging material and vice versa is important and information on the compatibility of the packaging material is

required. Commercial sales packs (finished product containers closure) made of plastic materials also have an Expiry Date. *The following information shall be provided:* A general description of the container and closure system including primary (inner) and secondary (outer) packaging, and other components such as measuring cups and syringes. The chemical identity of materials for each component of the system. Detailed specifications and tests for primary (immediate) packaging components such as: glass containers, plastic containers, and paper closures.

Such specifications and tests shall be as per article 10.2.6 of the FAO/WHO International Code of Conduct on Pesticide Management, certificates of analysis shall be provided as proof that the packaging conforms to specifications.

3.4.1 Suitability of Packaging Materials

3.4.1.1 Different Types of Packaging Materials

Tests must be performed to ensure compatibility of the formulation with the packaging material. The packaging material is examined for its integrity and tightness throughout storage. A range of packaging materials is available with different levels of barrier properties. HDPE (High Density Polyethylene) is the most widely used material for water-based formulations. Products containing organic solvents may need an additional barrier to prevent loss of solvent or damage to the HDPE polymer. Typical materials are HDPE co-extruded with a second polymer like Polyamide (HDPE-PA), Ethylene vinyl alcohol (HDPE-EVOH) or inline-fluorinated HDPE (fluorinated HDPE, F-HDPE). Commonly used PET bottles (Polyethylene terephthalate) offer a good solvent barrier.

For solid formulations which are not sensitive to humidity, HDPE bottles or LDPE lined cardboard boxes can be used. For protection against humidity, multi-layer bags with aluminium layers are available. Other materials may also be available.

3.4.1.2 Testing Compatibility and Stability of Packaging Materials

Storage stability tests should be performed in the commercial sales pack or a representative thereof. Tightness of the container and the closure, absence of any seepage, absence of strong panelling or ballooning, and no significant weight change are recommended evaluation criteria.

3.4.1.3 Extrapolation of Packaging Materials

As a general principle, data are required to support the packaging material in which ectoparasiticide are packed. In certain instances, extrapolating data from one packaging material to another is possible according to other international guidelines.

3.4.1.3.1 Extrapolation for Liquid Formulations

According to international guidelines, the following extrapolations are acceptable:

For water-based formulations (e.g., Soluble Concentrates (SC), *Soluble* liquid (*SL*) *Soluble* powder (SP) extrapolation between plastic materials is possible and stability data generated for one of the materials can be used in support of any of the others (HDPE, f-HPPE, HDPE-EVOH, HDOE-PA &PET).

For organic solvent containing formulations (e.g. Emulsifiable Concentrate (EC) extrapolating stability data of the product in HDPE to higher barrier HDPE-type materials (HDPE-PA, HDPE-EVOH, f-HDPE) is acceptable without generating further stability data when stability in HDPE was demonstrated. However, stability study done in HDPE-PA, HDPE-EVOH, f-HDPE cannot be extrapolated for the others. For different plastics like PET, it is recommended as a minimum to provide data on seepage.

Extrapolation between higher barrier HDPE-type materials (HDPE-PA, HDPE-EVOH, f-HDPE) for solvent containing formulations is acceptable when stability of the product in any of these materials was demonstrated and data show that no seepage and no significant weight change occur.

3.4.1.3.2 Extrapolation for Solid Formulations

Extrapolation of stability data is possible between all types of packing materials when they are waterproof or have a waterproof lining or where the formulation is not moisture sensitive. This can be demonstrated by showing that the weight and moisture content of the packed product has not changed during storage.

3.4.2 Packaging and Label Information

Article 10.2.6 of the FAO/WHO International Code of Conduct on Pesticide Management (5) requires labels that clearly show the release date (month and year) of the batch, Expiry Date and relevant information on the storage stability of the product.

In cases where the Shelf Life is shorter than 2 years, a justification must be given, and the label must show both release date and Expiry Date.

Label statements should be made according to regional requirements. In case there are no specific rules, the release date (month and year) and relevant storage stability information of the product will be based on paragraph 10.2.6 of the FAO Code of Conduct.

Note: It is important to note that commercial sales packs made of plastic materials also have Expiry Dates. Therefore, this should also be indicated clearly.

3.4.3 Change of Formulations during Product Life Cycle

During the product life cycle, changes of the composition of a formulation may occur e.g., due to using alternative raw materials, using an alternative process, or replacing co-formulant sources which are no longer available.

If such a change occurs refer to EAC harmonised ectoparasiticides variation guideline provided on the EAC website.

3.4.4 Labelling

The applicant shall ensure that the primary (immediate) packaging of the product is labelled in ***English***. However, an ectoparasiticide is generation sale product and used by farmers, the instruction of use information shall be provided in *English, Swahili* and *French* for all Veterinary ectoparasiticides. Products that will be marketed in the East African Region.

3.4.4.1 Labelling Information

Veterinary ectoparasiticides shall be labelled with statements on hazard, pictograms, and precautionary statements. The following minimum information shall be required in English on the label of the immediate (primary) packaging, with instructions for use in additional language, Swahili, and French:

a) brand name where appropriate
b) Active substance
c) quantity of active substance per dosage unit
d) total contents of container
e) date of manufacture
f) date of expiry
g) batch number
h) storage conditions
i) name and address of manufacturer
j) instructions for use (User information insert attached)
k) Unique identifier code

3.4.4.2 User Information Leaflet

The product packaging shall include instructions for use **(in English, Swahili, and French)** The leaflet shall include the following minimum information:

a) Active substance
b) A brief description of the mechanism of action and pharmacological effects
c) Clinical Information:
i. Indications
ii. Method of application describing fully and indicating the type of equipment to be used, if any, as well as the type and volume of the diluent to be used
iii. dosage regimens
iv. contraindications
v. precautions in pregnancy, and in milking animals etc.
vi. adverse reactions including their frequency.
vii. symptoms and treatment of over dosage
viii. with holding period
d) Product Information:
i. dosage form
ii. strength
iii. excipients
iv. storage conditions

v. shelf-life
vi. pack size
vii. description of product
viii. name and address of the manufacturer

e) Use of Personal Protective Equipment by applicators is a requirement for all persons who are involved in the administration of the product.

3.5 Generation of Storage Conditions for Veterinary Ectoparasiticide

3.5.1 Generating Storage Stability Data

Storage stability data should be generated with the product stored in the proposed commercial packaging (or smaller packages of the same construction and material), under accelerated conditions, and/or real-time testing at room temperature, or under ambient warehouse conditions. The following information regarding the storage stability trials should be supplied to the EAC MRP veterinary ectoparasiticides application:

- Product specifications, with proposed limits for the content of active Substance s and physical characteristics of the product within which the properties of the product will remain during its proposed shelf life. Products may have different specified limits at batch release, and at the end of its shelf life.

- Where the concentration of toxicologically significant impurities is known to increase as the active substance degrades upon storage, the level of these impurities should be analysed at the commencement and completion of the relevant product storage stability study. The concentrations found after product storage should be in a proportionally appropriate. However, where toxicologically significant impurities are found as the result of 'carry-through' from raw materials and/or as a by-product of the manufacturing process for the active substance and do not increase on storage, then no analysis of these impurities is needed in the relevant product stability study.

- Test results generated before and after storage, including full details of the methods used to produce data for each of the test parameters listed in the shelf-life specifications.

- Data describing the stability of the packaging materials demonstrating no significant deterioration, as interaction with a product can cause brittleness, softening, and corrosion of packaging.

The data should confirm that the formulated product will remain within specification for at least 2 years when stored in its unopened, original container, away from direct sunlight, and at or above 25˚C ('normal storage conditions').

3.5.2 Pack Sizes and Pack Size Changes

As specified below in section 2, stability testing trials for new products should be conducted on product packaged in the same containers (materials and pack size) that are proposed for the marketing of the product. Smaller containers of the same material as the commercial containers may be used for stability

testing, where the size of the commercial container makes stability testing impractical.

Applicants may also wish to market their products in a smaller container later. Undertaking a stability study in a smaller container of the same material and construction would demonstrate the product pack size is fit-for-purpose for not only the current marketed product size, but for future, smaller pack sizes.

3.5.3 Larger Pack Sizes

For large pack size no additional stability data is required in primary pack size from that used in the stability study. The surface area to volume ratio of the product reduces as the primary pack size increases, and the interactions between the product and packaging become less significant.

Larger pack sizes may have implications for other assessment areas (for example, worker safety or environmental impact), and data or justification may be required to address those potential risks.

The packaging for a larger container must meet the conditions of registration for containers in terms of having sufficient strength to prevent leakage and must be of a practical size to allow users to comply with any label instructions, such as 'shake well before use'.

3.5.4 Smaller Pack Sizes

Smaller pack sizes have a greater ratio of surface area to volume than larger containers, interactions between the product and packaging are generally more significant, and stability data or scientific justification in-lieu of data may be required to support a smaller pack size.

Approval of a pack size 50% smaller (by volume, or by weight) than the pack size used to generate the stability study data will be considered by the EAC MRP regulators without provision of further stability data, provided there are no known stability problems with the active Substance s and/or formulation in question.

In addition, where a long history of use of other registered products with the same formulation type and active substance in smaller pack sizes exists (for example, emulsifiable concentrate), it may also be possible to support a smaller pack size without stability data. A scientific justification on the registration history of similar products in smaller packs could be provided in lieu of stability data and would be accepted by the EAC MRP regulators if there was sufficient evidence of stability based on registrations of other products.

3.5.5 Packaging Materials – New Product Registrations

Stability data should be generated for the product while stored in its proposed, commercial packaging. When multiple packaging materials are used for a product, or where a reference product uses a different packaging material even with an identical formulation, stability data should generally be provided for the product in each packaging material.

However, certain packaging materials are more resistant to corrosion because of product interaction, than others. Where the stability has been demonstrated for a less chemically resilient material, and it is proposed to add or change to a material known to be more resistant to chemical degradation, stability data would not be required for the more resilient packaging material.

Acceptable extrapolations from data generated for packaging materials are detailed below.

3.5.6 Liquid Formulations Acceptable Packaging Material

Table 12

Water based formulations, e.g., aqueous soluble concentrates and suspension concentrates	
Packaging used in the stability study	**Acceptable extrapolations**
Non-metal	All non-metal packaging types are supported with no further data
Organic solvent-based formulations, e.g. emulsifiable concentrates, non-aqueous soluble concentrates	
High density polyethylene (HDPE)	HDPE/EVOH, fluorinated HDPE and HDPE/polyamide packs are supported with no further stability data
HDPE/EVOH or fluorinated HDPE or HDPE/ polyamide	Full stability data from one of these 3 packaging types can be extrapolated to one of the others, or to plain HDPE, with the provision of seepage data, i.e. packaging stability data, which requires only observation of the effects on the packaging during storage, not testing against all the product specifications, and can be conducted as a real-time or accelerated study

3.5.7 Solid Formulations

Extrapolation is possible between all container types except:

- To or from packaging involving water-soluble bags; and
- From rigid to flexible containers for granular products due to potential issues with crushing of granules if containers are stacked.

3.5.8 Shelf-life Statement on the Product Label

Printed product labels should include the date of manufacture and expiry in (month and year) of a batch, and relevant information on the conditions under which the product is recommended for storage.

i) Store under normal storage conditions (15^{o}C - 30^{o}C).

Note that these recommendations must be reflected on the product samples submitted for registration.

3.5.9 In-use Shelf Life

Stability data should be provided to support the in-use-shelf life of the product after first opening of the Product's container. In support of the proposed in-use shelf-life, data from at least two different batches of ectoparasiticide should be provided. This data shall demonstrate compliance with the critical stability-indicating parameter(s) when the primary container is first opened and again at the end of the proposed in-use shelf.

3.5.10 Design of Stability Testing Trials

3.5.10.1 Size and Number of Batches

Stability testing should be conducted on laboratory-, pilot- or production-scale batches of a product. Batch sizes of less than 5 kilograms or 5 litres are normally not acceptable for use in stability testing, except when a normal production batch is this size. The formulation is to be the same as that of product submitted product for registration.

Please include the following information in a stability study:

- Product name
- Batch identity
- Batch size
- Date of manufacture
- Containers used for storage of the samples during the study (size, construction material and type of container).

3.5.10.2 Storage Conditions and Duration

The stability studies should be specified in line with the OECD and the WHO/CIPAC guidelines and applicants should refer to the protocol in annex I. Storage stability trials may include accelerated or real-time tests, or both. Real-time testing is generally conducted for a product that may be unstable at high temperatures and is generally a requirement for date-controlled products. Products containing microbial active Substance s are usually date-controlled, not generally stable at the elevated temperatures typically used in accelerated testing, and therefore real-time testing is almost always necessary.

For some formulations, studies at lower temperatures may be necessary due to the instability of the formulation at higher temperatures (this should be reflected in the recommended storage conditions). Liquid formulations should be tested at low temperatures (0 ± 2˚C or lower). This is to demonstrate that the product does not change at low temperatures, for example, specific components may crystallise or separate at low temperatures.

3.5.10.3 Accelerated Testing

Accelerated storage tests according to CIPAC MT 46.4

Stability tests at elevated temperatures are designed to increase the rate of chemical degradation or physical change in a product to obtain information on the shelf life of a product in a shorter time than a real-time study. Accelerated testing involves extrapolations from higher to lower temperatures, and from shorter to longer storage periods. For most ectoparasiticides' products, accelerated storage stability data is sufficient, without the provision of real-time storage stability data. Since formulations are complex mixtures and higher temperatures can induce a degradation pathway that may not exist at lower temperatures, there is uncertainty involved in such extrapolations, unless the formulation proves to be stable after real time storage at ambient conditions.

At the initial stage of a registration process a complete set of real time data is usually not available. In this case data from accelerated storage tests can be used to support registration. This approach is accepted in the region, however, data from real time storage tests at ambient temperature should be submitted once ready. The applicant is required to prepare and submit a post-approval stability data commitment plan.

Different storage regimes as defined in CIPAC MT 46.4 and referred to in the FAO/ WHO manual can be used to perform accelerated storage tests. The FAO/WHO pesticide specifications recommend testing of relevant product parameters before and after storage for 14 days at 54˚C so any potential changes can be detected. In some situations, an alternative time-temperature regime, such as those listed in Table may be used. For example, aerosol products are commonly tested at 40˚C for 8 weeks due to the safety issues associated with this type of formulation being exposure to temperatures above 50˚C.

Table 13: Accelerated Storage Temperatures and Duration of Stability Trials

Temperature	**Duration**	**Comments**
54˚C	14 days	Generally preferred testing regime (CIPAC MT 46.3)
50˚C	4 weeks	

45˚C	6 weeks	
40˚C	8 weeks	Commonly used for aerosol products
35˚C ±2˚C	12 weeks	
30˚C ±2˚C	18 weeks	

Note: The purpose of accelerated storage tests is to extrapolate from higher temperature data after short term storage to lower temperature data after long term storage, according to the Arrhenius equation

Products that exhibit an adequate stability profile at 40˚C to 54˚C are likely to be stable under normal storage conditions (at or above 25˚C) for at least 2 years.

Appropriate justification should be provided for use of a temperature regime other than 2 weeks at 54˚C (or 8 weeks at 40˚C for aerosols). This is particularly important for the 2 lowest temperature regimes, where summer temperatures, particularly in northern areas, commonly reach 30 to 35˚C. Part of the justification may include lower recommended storage temperatures. For spot-on and pour-on sufficient justification must be provided for temperature regime used for these infusion oil based of formulations.

It is recommended that samples of the formulation be taken before and after the Technical Monograph (MT 46.3) test. The 2 samples (time zero and 14 days for 54˚C trials) may be analysed concurrently after the test. This will reduce the analytical error of 2 separate analyses on different days, and/or by different analysts.

3.5.10.4 Real-time Testing

Data from accelerated stability studies can provide a useful indication of a product's stability, but in some cases, products may pass this test and yet still be unstable in long-term storage. In contrast to accelerated storage tests, real time testing does not rely on extrapolation and therefore is always acceptable for the declaration of a Shelf Life. Storage is usually performed at ambient temperature for at least 2 years or longer and the formulation must meet the specified limits at the end of the stated storage period.

In certain situations, stability data generated at ambient temperatures over a period of 2 years (real-time) may be more appropriate than accelerated testing. For example, where a proposed product has a tendency to cake over time or is subject to contamination as a result of bacterial or fungal growth, accelerated testing would not be suitable to demonstrate the product's stability.

Real-time testing is normally performed at, or above 25˚C, with storage for at least 2 years. Testing should at least be conducted at time zero, and at the end of the storage period. However, applicants may wish to consider further testing the product at intermediate time points (for example, after 6-, 12- and 18-months storage for a 2-year study), particularly if the period of stability of the product is in doubt. Deterioration or degradation of the product during the test period should be determined. At the end of the test period, the product should be examined for physical changes, such as phase separation or "clumping," and any changes which would interfere with the usefulness or safe handling of the product if used according to the label directions.

Storage tests can be carried out at constant temperature under controlled laboratory conditions or e.g. in a warehouse where the temperature is recorded during the test period and reported. Depending on region specific regulations, stability tests may need to be conducted at defined temperatures reflecting the respective climate zone. In the absence of such regulatory guidance, it is suggested to perform stability studies at ambient temperatures or under controlled conditions at e.g. 20°C ± 2°C or 25°C ± 2°C.

The following ranges of annual average temperatures are considered typical for different climate zones:

- moderate climate: 18 - 22°C
- hot climate: 23 - 27°C
- very hot climate: 28 - 31°C

The test substance should be quantitatively analyzed for Active substance content and changes in impurities because of degradation or packaging deterioration over the test period. Results should be reported as concentration in weight percent. The test substance and container should be observed for any physical changes at each test interval, recording all observations in the raw data. All test containers should be reweighed at each of the test intervals, prior to and after sampling, to monitor weight.

Real-time data are generally required for date-controlled ectoparasiticides.

Depending on the product's formulation type and packaging material, standardised relative humidity and light exposure conditions may also be recommended during testing.

3.5.10.5 Low Temperature Stability Testing

Cold temperatures may impact physical properties of certain formulation types e.g., due to crystallization of the Active substance (s) or separation of multi-phase systems. Liquid formulations (including capsule suspensions, emulsifiable concentrates, oil-in-water emulsions, micro-emulsions, soluble concentrates, and suspension concentrates) may be adversely affected by storage at low temperatures, resulting in crystallisation of active Substance (s), significant changes in viscosity, and/or phase separation of emulsions.

In some regions where temperatures may reach 0˚C, or lower. The liquid formulations should be tested at 0˚C ± 2˚C or lower, for 7 days. The effect of

low temperatures on stability should be determined and reported according to the Collaborative International Pesticide Analytical Council (CIPAC). For liquid products/formulations, the FAO/WHO manual requires low temperature storage tests according to CIPAC MT 39.3. After storage the product shall continue to comply with the relevant clauses listed in the FAO/WHO manual in section 4.6.1 “Stability at 0 °C”.

For capsule suspension formulations, the capsule walls may break because of repeated freezing and thawing (thus releasing the Active substance into the suspending liquid). Freeze-thaw cycling testing should be undertaken.

Note: Stability data generated at low temperatures are not required if the product label contains or includes a warning against exposure to low temperatures. However, the regulators will need to be satisfied that such a restriction is practical, and suitable justification is provided.

3.5.10.6 Test Parameters

The stability profile of ectoparasiticides is determined by monitoring a combination of chemical and physical properties on storage. Monitoring the content of the active Substance alone is insufficient to make any reliable prediction as to the stability of the product. Over prolonged storage, a product may not exhibit a decline in the concentration of the active Substance, yet the important physical properties (for example, wettability or suspensibility) may have changed as such to compromise the performance of the product. For most key formulation types, the FAO/WHO manual provides guidance on the physical parameters required to be tested before and after storage. The formulation shall comply with relevant limits to ensure that the formulation can be handled and applied without difficulties during the entire Shelf Life.

The determination of physical properties should be based on CIPAC methods or other international methods. Where company internal methods are used, this should be justified. This guideline includes test parameters for each formulation type. To adequately demonstrate product stability, all relevant parameters should be examined in a stability trial. If certain parameters are not addressed, scientific argument should be provided in lieu.

Note that these test parameters have been derived from the FAO/WHO pesticide specifications. For formulation types not listed in this guideline, it is recommended that applicants seek advice from the EAC veterinary medicinal products regulators in the form of an enquiry, for technical assessment before commencing a stability trial.

3.6 Analytical Methods and Validation Data

3.6.1 Determination of Active Substance Content and Relevant Impurities

Full details of the analytical methods used to monitor a product during stability trials should be provided, except where you have used collaboratively tested standard methods for the analysis (CIPAC, Association of Official Analytical Chemists, etc.). Compendial methods such as CIPAC methods are regarded as validated, and do not require full revalidation.

The EAC MRP initiative recommends that available analytical methods for a particular formulation, described in official and recognised publications such as CIPAC handbooks and AOAC, be used. These methods are legally recognised as regulatory methods.

For further details of the necessary degree of method validation, see the separate guideline on validation of analytical methods.

The following information should be included:

- Instrumentation
- Sample preparation
- Method of extraction of the active substance from the product
- Reference standards and reagent preparation
- Validation data
- Copies of representative chromatograms (if applicable)
- Representative calculations

Alternative analytical methods may be used in place of regulatory methods. Appropriate validation data is required for such methods. The type of validation data required is dependent on the analytical techniques used but will typically include demonstration of linearity over a suitable concentration range, specificity, precision and accuracy.

See the separate guideline on validation of analytical methods for further information.

3.6.2 Determination of Physical Properties

The results and interpretation of measurements of physicochemical properties are highly dependent on the analytical procedures used. The EAC regulators recommends that standard CIPAC, or equivalent accepted methods are used to measure the physicochemical properties of veterinary ectoparasiticides products.

Validation data are not required for CIPAC, or other, appropriate compendial method's physicochemical tests. If in-house company, or other methods are used for physicochemical property testing, a full description of the procedures should be provided, together with appropriate validation data. This may include a comparison between the officially recognised methods (CIPAC handbooks and the AOAC manual methods) and the in-house method, with comment on any differences, and the significance of any differences.

3.6.3 Test Parameters for Products

The data and/or testing parameters that the EAC regulators adopts are derived from the FAO/WHO pesticide specifications.

3.6.3.1 Active Content

For stability testing of most products, testing of the active content before and after storage using a suitable specific chemical analytical method, is required.

In some specific cases, chemical analyses of the active content before and after storage may not be required. The manufacturer /applicant is expected to provide justification in such cases.

For products demonstrating good stability, significant changes in active content should not be observed during real-time or accelerated stability studies. The Active substance content should, in general, not decline by more than 5% from the level measured initially in accelerated or real-time testing. Further information and justification should be provided if the level of degradation exceeds 5%.

Measures taken to demonstrate and/or justify the quality, safety and efficacy of a formulation that exhibits high levels of degradation during storage could include the following:

- Identification and quantification of degradation products.
- Inclusion of a manufacturing overage not greater than 10% of the label claim to allow for degradation.
- Registration of the product subject to a condition of inclusion of an expiry date shorter than the standard 2 years from the date of manufacture.
- Conducting efficacy studies on aged batches of product to show that acceptable levels of efficacy are retained despite the decline in active content.

3.6.3.2 Content of Relevant Impurities

For some active substance, testing of levels of toxicologically significant breakdown products, or impurities likely to accelerate the formation of toxicologically significant breakdown products before and after storage may be required.

Impurities must be measured using suitable methods supported by appropriate validation data. Where appropriate, compendial methods should be used (for example, CIPAC MT30.5 is a compendial method commonly used for determination of trace levels of water in formulations).

Levels of impurities must be within specified limits before, and after storage. Determination of suitable specified limits that can be justified from a human safety perspective can be sourced from the FAO pesticide specifications. When impurity limits of active substance standards are established for the technical active, not a formulated product, and appropriate conversion factors should be applied if setting limits on the basis of g/kg or g/L in a formulation.

3.6.3.3 Appearance and Physical State

These tests are performed visually, and are described in qualitative terms such as solid, liquid, suspension etc.

3.6.3.3.1 Colour

The following test methods are recommended:

- American Society for Testing and Materials (ASTM), *Standard method for specifying colour by the Munsell system D-1535.*
- ASTM *Standard method for specifying colour of transparent liquids (Gardner Colour Scale, D-1544).*

A visual description of colour is also acceptable.

3.6.3.3.2 Odour

This test is performed organoleptically and involves the use of descriptive terms (for example, thymol-like odour), characteristics of aromatic compounds (for example, garlic-like).

3.6.3.4 Acidity or Alkalinity and Potential of Hydrogen PH

This test is recommended for any product where acidity or alkalinity and pH are relevant parameters for the quality of the product. Where relevant (that is, when the product is to be applied as an aqueous dilution), the pH of a 1% aqueous dilution, emulsion or dispersion of the product should be determined and reported according to CIPAC method MT 75.3. A change in pH during storage can give an indication of instability of the active substance or product.

The acidity or alkalinity is determined by titration with standard acid or alkali according to CIPAC method MT 31.

3.6.3.5 Dry Sieve Test

The dry sieve test is designed to determine the particle size distribution of dustable powders and granules intended for direct application.

The following CIPAC methods apply:

- MT 59.1 dusts.
- MT 59.2 granular formulations.

3.6.3.5.1 Acceptable Limits for the Dry Sieve Test

Maximum 5% is retained on a 75 µm sieve (dustable powders).

For dustable powders, if 5% or more of the product is retained on a 75 micrometre (µm) sieve, the active content of material remaining on the sieve should be determined to demonstrate there was no separation of the active substance from the carrier.

3.6.3.6 Particle Size Distribution

You should determine the nominal size range of solid materials for direct application (for example, dustable powders and granules). These data are used to assess if an acceptable proportion of the product is within an appropriate size range.

The following methods apply:

- OECD method 110 – powders or dusts.

3.6.3.7 Dust Content

The dust content of solid preparations is determined to ensure the risk to operators is acceptable (for example, when transferring the product from the primary pack into a mixing tank), and to determine the potential for blockage of application equipment.

The following methods apply:

- CIPAC method MT 171 – granular products.
- OECD method 110 – powders or dusts.

MT 171 describes 2 methods for the determination of dustiness, but the gravimetric method is regarded as the reference method.

3.6.3.7.1 Acceptable Limits for Dust Content

If 1% by weight of the preparation has a particle size of less than 50 µm, Provide inhalation toxicity data.

3.6.3.8 Emulsifiability, Re-Emulsifiability and Emulsion Stability

For products that form emulsions, data on emulsifiability, emulsion stability and re-emulsifiability are used to determine whether a product forms and maintains a stable emulsion.

The following CIPAC methods apply:

- MT 36.1 – 5% dilution.
- MT 36.2 – 1% dilution.
- MT 36.3 – emulsion characteristics and re-emulsification properties.
- MT 173 – 0.1% to 2% dilution.

MT 36.1 is designed to be conducted over a 24-hour period. If no separation of cream or oil is observed after 2 hours, then no further testing is required. However, if separation is observed, you should perform the 24-hour test.

For a dilute emulsion, MT 173 is the preferred method. However, MT 36.1 may be used as a screening method. If no separation of a 5% dilution is seen after

2 hours, then no further testing is required. The test should be conducted in CIPAC waters A and D.

3.6.3.9 Acceptable Limits for Emulsifiability, re-Emulsifiability and Emulsion Stability

- MT 36.1 – maximum 2 mL cream, trace of oil after 30 minutes; if any separation is observed, re-emulsification should be complete after 24 hours.
- MT 173 – minimum 98% after 4 hours; maximum 102% after 4 hours.

If a product falls outside of these limits, evidence should be provided to demonstrate that the product remains homogeneous when applied with the appropriate application equipment. If more than a trace of oil separates, re-formulation of the product should be considered.

The appropriate application equipment. If more than a trace of oil separates, re-formulation of the product should be considered.

3.6.3.10 Viscosity

Viscosity is a property of fluids that describes the resistance offered to a shearing force under laminar flow conditions; for example, resistance to slow stirring, or to flow through a capillary, or narrow channel.

The kinematic viscosity of a liquid formulation for direct application (ultra-low-volume products) should be determined. For Newtonian fluid, the viscosity at any shear rate should be conducted. For non-Newtonian fluids (for example, a non-drip paint), viscosity values should be provided for at least 2 different shear rates.

The following CIPAC methods apply:

- MT 22.
- MT 114.

3.6.3.11 Flowability

The following methods apply:

- CIPAC method MT 44.
- OECD method MT 172.

3.6.3.11.1 Acceptable Limits for Flowability

The sample should flow through the sieve after a maximum of 5 liftings.

3.7. Parameters to be Tested in Stability Trials

In addition to the appearance and content of an active substance, relevant physicochemical properties should be monitored before and after storage, according to the formulation type.

Shelf-life specifications are drawn from the physical properties mentioned in the FAO/WHO pesticide specifications and are applicable to any given formulation type as specified in **annex I**.

Individual CIPAC MT test parameters are listed with each individual formulation type.

Table 14 Stability Test Parameters

Emulsifiable concentrates (EC)	
Recommended test parameters	**Relevant CIPAC method**
Appearance (physical state, colour, odour)	No CIPAC method
Active Substance content	Appropriate validated method
Acidity or alkalinity or pH	MT 31 or MT 191 or pH range (MT 75.3)
Emulsion characteristics	MT 36.1, MT 36.2, MT 36.3, MT 173 or MT 183
Persistent foam	MT 47.2
Low temperature stability	MT 39.3
Packaging stability	Observation of packaging stability

Oil-based suspension concentrates (OD)	
Recommended test parameters	**Relevant CIPAC method**
Appearance (physical state, colour, odour)	No CIPAC method
Active Substance content	Appropriate validated method

Oil-based suspension concentrates (OD)	
Acidity or alkalinity or pH	MT 31 or MT 191 or pH range (MT 75.3)
Pourability	MT 148
Dispersion stability	MT 180
Wet sieve test	MT 185
Persistent foam	MT 47.2
Low temperature stability	MT 39.3
Packaging stability	Observation of packaging stability

3.7.1 Toxicologically Significant Impurities

Where the concentration of toxicologically significant impurities is known to increase as the active substance degrades upon storage, the level of these impurities should be analysed at the commencement and completion of the relevant product storage stability study. The concentrations found after product storage should be in a proportionally appropriate. However, where toxicologically significant impurities are found as the result of 'carry-through' from raw materials and/or as a by-product of the manufacturing process for the active substance and do not increase on storage, then no analysis of these impurities is needed in the relevant product stability study.

3.8 Data on Application/Use

3.8.1 Objective of Ectoparasiticide Application

The objective of the application of veterinary ectoparasiticides is to keep the ectoparasite under control. The ectoparasite population must be kept suppressed to minimum biological activities to avoid economic loss of livestock production. The objective of ectoparasiticides application besides keeping the ectoparasite population under check should also be to avoid pollution and damage to the non-targets. The applicant must provide following information:

3.8.1.1 Field of Use

Brief statement should be provided on the field of use of the product, e.g For vector control on livestock including the specific list of all vectors proposed to

be controlled by the product, in case of more restricted registrations. It should be clearly stated on the label.

FOR USE ONLY ON ECTOPARASITICIDE TREATMENT and **POISON** in red including pictograms

3.8.1.2 Quality of Ectoparasiticide*:*

The applicant must provide specification of the product at manufacture and storage to confirm that the product maintains the label claims of ectoparasiticides.

Active substance during production and marketing of ectoparasiticide formulations (the batch numbers used for production and storage must be provided).

3.8.1.3 Number and Timing of Application

A brief description of the timing and frequency of ectoparasiticides application should be provided including sufficient justification of the proposed timings.

Consideration must be made when handling large herds/ flocks of livestock.

Manufacturer proposed duration of protection should be clearly stated with sufficient justification of the period.

3.8.1.4 Application rate and concertation of the active substance

A brief description of application of ectoparasiticides process and adherence to the following points should be provided.

i. Proper dosage should be applied evenly.
ii. The toxicant should reach the target.
iii. Proper droplet size /particles for dust should be delivered.
iv. Proper density of droplet on the target

The dosage recommendation is generally indicated per animal mg/kg. the dosage should be properly explained, and the exact quantities of the formulated ectoparasiticides should be calculated, stated, and applied. Ectoparasiticide are applied by different methods like spraying, dusting etc. Water is a common carrier of ectoparasiticides, but air or oils are also used as carriers. Selection of proper droplet is an important consideration. The manufacture should provide the specification of proposed equipment for adequate ectoparasiticides delivery.

3.8.1.5 Proposed Instructions for Use as Provided /Printed on Labels

The Governments and industry should ensure that all ectoparasiticides made available to the public are packaged and labelled in a manner which is consistent with FAO/WHO or other relevant guidelines on packaging and labelling and with appropriate national or regional regulations (Article 7.4) and all ectoparasiticides containers should be clearly labelled in line with relevant regulations. Basic instructions for use must always be available in a

locally understood language in the region as stated in the guideline on the label of each pack.

3.8.1.6 Suitability of Packaging and Closures

The applicant should provide a summary of data on storage stability confirming the stability of the formulation as packaged for sale, and the appropriateness and safety of the packaging. Including 'material safety data sheets' (MSDS) or 'safety data sheets' (SDS) on the formulated product, the components of the product and co-formulating agents or safeners and surfactants proposed for use in the product, in order, for example, to determine whether any of them are health or environmental hazards. These data can also be used after a product has been registered to ensure that the material being sold is exactly that which was approved by the responsible authority.

3.8.1.7 Procedures for Cleaning Application Equipment and Protective Clothing

A detailed user information must be provided, and the Users should be trained before handling ectoparasiticides in operating and calibrating a sprayer, cleaning nozzles, and determining the PPE and Respiratory Protective Equipment RPE required at each phase.

A section on the label should provide information to the user on how to clean up accidental spills. The exact measures to take will depend on the type of Active substance and components of formulation.

SECTION 4.0 SAFETY IMPACT ON HUMAN AND ANIMAL (MAMMALIAN TOXICOLOGICAL DATA)

4.1 Acute Toxicity Studies for Formulated Product

Acute toxicity studies examine the adverse effects arising from administration of a single oral dose or a single dermal or inhalation exposure of a substance over a specified period or multiple doses given within 24 hours.

To allow assessment of the acute toxicology of a substance, studies in animals should examine the most likely routes and forms of exposure in humans and animal.

Acute oral toxicity studies should be performed in at least one mammalian species. Rats are the preferred rodent species for oral studies unless a species more representative of human toxicity is known. You should also provide acute dermal and inhalation studies in at least one species. For skin and eye irritation studies, rabbits are an acceptable species, but alternatives from adopted OECD guidelines for the testing of chemicals (or other recognised guidelines) to the usual *in vivo* test may be suitable. *In vivo* eye irritation tests may not be appropriate in certain circumstances. If you do not believe an eye irritation study is appropriate, provide a valid scientific argument as to why these studies should not be included. For example, if the results from a skin irritation study or validated *in vitro* study demonstrated corrosivity or severe irritation, it is acceptable not to test the product in an eye irritation study, as it is presumed that the product will be corrosive to the eye. Similarly, products with pH extremes of 2 or less, or 11.5 or more are considered corrosive to the eye, unless the acid or alkaline reserve (buffering capacity) of the product suggests otherwise.

A skin sensitisation study is performed to test for possible hypersensitivity reactions to the substance. Guinea pigs are normally used for sensitisation studies. Internationally validated alternative methods, such as the murine local lymph node assay, are also acceptable.

4.2 Acute Toxicity Studies

For a veterinary ectoparasiticides product, you should submit a 'six-pack' of acute toxicological data. This consists of the following studies on the product:

4.2.1.1 Acute Oral in Rats: where acute toxicity data on the formulation is available, it would be used to determine whether the value of 1500 milligram per kilogram body weight is not appropriate and may be increased. Whether one or two swallows (approximately 10 gram or 10 millilitre) of the product presents an acutely toxic dose to an infant or small child.

4.2.1.2 Acute Dermal in Rats: A veterinary ectoparasiticides product should not be acutely toxic at dermal doses up to 2000 milligram per kilogram body weight.

4.2.1.3 Acute Inhalation in Rats: A veterinary ectoparasiticides product should not be acutely toxic at inhalational concentrations up to 2000 milligram per cubic metre (four-hour exposure) for a gas, 20 milligram per litre (four-hour exposure) for a vapour and 5 milligram per litre (four-hour exposure) for dusts and mists.

4.2.1.4 Eye Irritation in Rabbits: The irritancy to eyes of veterinary ectoparasiticides products should be low. The formulation and application methods of a product will be taken into consideration on a case-by-case basis. Provide relevant information regarding any risk mitigation measures available for the proposed product.

4.2.1.5 Skin Irritation in Rabbits: The irritancy to skin of veterinary ectoparasiticides products should be low. The formulation and application methods of a product will be taken into consideration on a case-by-case basis. Provide relevant information regarding any risk mitigation measures available for the proposed product.

4.2.1.6 Skin Sensitization Study in Guinea Pigs: If such data are not available, provide valid scientific argument as to why you have not submitted data. In certain circumstances, a toxicological evaluation of the product may be conducted by taking the known toxicological properties of the active Substance s and excipients in the formulation and extrapolating these to estimate the acute toxicity of the product. We recommend that you adequately address the reasoning for not providing toxicity studies.

4.3 Toxicological Database and Bibliography

Every application (including supplementary applications) should include a toxicological database comprising a full bibliography of all studies provided in the application. Every application (including supplementary applications) that contains toxicological data should include a list of all studies on the active substance and/or veterinary formulated product. Clearly identify studies lodged as part of the application.

For each listed study, provide the following information:

4.3.1 identity and the concentration or purity of the material tested (for example, active substance, product).

4.3.2 type of test (for example, acute oral study, two-year dietary study)

4.3.3 species and strain of animal used.

4.3.4 study laboratory and names of authors

4.3.5 study sponsor

4.3.6 good laboratory practice status (including certification where applicable)

4.3.7 title of the report, report number and date of report

4.3.8 date the study was completed

4.3.9 location in the application (volume, page number).

4.4 Related Studies

4.4.1 No-observed Adverse Effect Level (NOAEL)

The no-observed adverse effect level is the highest dose of a substance at which there is no detectable adverse alteration of morphology, functional capacity, growth, development, or lifespan of the target organism under defined conditions of exposure compared to those observed in control (untreated) animals, and which are observed or measured at higher dose levels used in the study.

The no-observed adverse effect level is expressed in milligrams of substance per kilogram of body weight per day (mg/kg bw/day) or, in a feeding study, as parts per million (ppm) in food. For feeding studies, conversion to mg/kg bw/day should be made, calculated from substance intake by measured or estimated food intake over the study period.

Where the test substance is given in feed, and problems with the stability of the test compound occur, the feed should be analysed at frequent intervals.

4.4.2 Lowest-observed Adverse Effect Level (LOAEL)

The lowest-observed adverse effect level is the lowest dose of a substance at which there is a detectable adverse alteration of morphology, functional capacity, growth, development, or lifespan of the target organism under defined conditions of exposure compared to those observed in normal (untreated) animals.

The lowest-observed-effect level is expressed in milligrams of substance per kilogram of body weight per day (mg/kg bw/day) or, in a feeding study, as parts per million (ppm) in food. For feeding studies, conversion to mg/kg bw/day should be made, and where problems with the stability of the test compound occur in feed, the feed should be analysed at frequent intervals.

4.4.3 Effects on Wool, Hides. Skins and Fleeces

Given the importance of the hides ad skin, wool industries in EAC region, it is recommended that data on wool staining or damaged hide or skin damage, or damage to animal products are collected and submitted. Demonstrate by data or valid scientific argument that chemicals applied externally to cattle, sheep and other hide- or fibre-producing animals will not cause damage to the hides, skins, fleeces, or fibres produced by those animals.

Wool scour ability data should be provided for pour-Ons, dips, shower sprays and other products containing dyes or other pigments that are applied to wool. This issue may be addressed by valid scientific argument, if appropriate.

4.4.4 Safety

Although LD50 data concerning the safety or toxicity of an insecticidal product is often helpful, LD50 values are not always the best indicator of the safety of specific insecticide formulations applied to pets or premises. Consideration must be given to the concentration of product (mg/mL), application rate (mg or g/m2 for environmental products, and mcg or mg/kg for topicals), route of exposure (dermal or oral), total dose, and the species exposed. The actual risk of exposure during treatment, after treatment, or after accidental ingestion can be assessed only after evaluation of these criteria.

4.4.5 Accidental Administration or Exposure to Non-Target Animals

Provide any evidence of possible adverse effects from accidental administration to or exposure of animals other than those for which the product is recommended. Appropriate warnings should be proposed for inclusion in the product label. This is particularly important for ectoparasiticides products where the product remains on the target species for a long period, and products used in situations where different species occupy the same area.

Because animal toxicity can be modified by formulation technology, Active substances are not the sole guide to safety assessment of a product. Most commercially available products have undergone adequate safety evaluation for regulatory approval; the label noting such approval remains the best source of information. Cats are sensitive to many insecticides, and use of these insecticides on or near cats must be done with caution. Human and environmental safety also should be considered, especially when treating premises (eg, some compounds may break down into more toxic components; older products on the shelf might have been withdrawn because of safety concerns). Generalizations should not be made, because formulations generally safe for grass application may induce skin reactions, or even fatal reactions, in sensitive individuals and certain breeds of dogs and cats.

4.4.6 Effects on Taste or Produce (Organoleptic Effects)

If data suggest that the use of a veterinary chemical product or its metabolites is likely to affect the taste of animal produce (for example, products that may affect milk), those data should be included in this part of the application and clearly identified as a separate section.

If there is any reason to believe that the use of a veterinary ectoparasiticides product (or its metabolite) could cause an off-flavour or tainting of a food product, an adequate investigation should be undertaken, supported by taste-panel tests or other organoleptic tests, to verify that no unacceptable tainting occurs. The data obtained should be included in this part of the application and clearly identified as a separate section.

4.4.7 Special Requirements

Products for topical use include shampoos, aerosols, spot-on, pour-on or dust formulations, ear tags, collars, clips, dipping or spray-race formulations, etc. While the general requirements also apply to products for topical use, it is necessary to consider interactions between treatment and regional climatic conditions during the trial. In particular, the applicant should consider the need for additional studies as follows:

- The effect of rainfall at various intervals before, during and after treatment.
- The effect of sunshine and hot weather under monitored conditions during and after treatment.
- The effect of dilution factors with dipping.
- The effect of washing and bathing during the treatment period.
- The effects of hair length and thickness of coat.
- The effect of dirtiness of animal coat and the effect of dirtying of preparations (e.g. of dipping formulations) during the treatment of groups;
- The effect of self-grooming or mutual grooming of treated animals.
- Different body sizes of target animals treated with a standard dose formulation.
- Effects on the quality of fleece or hide and impact on tanning or processing.

Ideally, side effects and adverse effects of the product should be monitored during the trial and for several days afterwards. Where secondary pharmacodynamic effects are seen, a study on the dose/effect relationship may be required.

In general, the particulars of the mode of actions and indications of a ectoparasiticides product shall be provided.

4.4.7.1 Directions for Safe Use

Safe use of domestic veterinary chemical products should not require safety equipment that is not readily available to the householder. Safety equipment other than gloves is not considered an appropriate mitigation option for users of domestic veterinary products, because users are not trained in handling hazardous substances and compliance is not expected. Domestic veterinary chemical products may not be supported for domestic use if safety equipment other than gloves is required for their safe use.

4.4.7.2 First Aid Directions

The product label affixed to the container and any associated leaflets should carry appropriate first aid directions in the event of poisoning. Veterinary chemical products should not require specific antidotes or aggressive first aid measures.

SECTION 5.0 METABOLISM AND RESIDUES DATA

5.1 Residues Arising from Direct Application to Farm Animals

Ectoparasiticides may be applied directly to farm animals for control of lice, fleas, mites, and ticks. Application methods include dips, sprays, pour-Ons and jetting. Residue trials using the required method of application, dosage and withdrawal times are needed if residues may occur in animal commodities.

The conditions of a supervised residue trial on farm animals should match the maximum conditions described on the label. If more than one application method is permitted, e.g., dip or pour-on treatments, residue data should be available for each method. The evaluation should record the highest residue occurring in individual animal tissues resulting from the approved method and dose. The highest residues will support the MRL recommendations. The evaluation should record the average milk residues each day across the treatment group and the MRL recommendation will depend on the highest of these average milk residues on a day achieved within the conditions described on the label.

The STMR concept is designed for supervised field trials on crops to obtain the typical residue value when an ectoparasiticides is used at maximum GAP. The STMR methodology is not directly applicable to a single direct-animal treatment trial. However, the idea of a typical residue value when an ectoparasiticides is used directly on animals (at maximum label conditions) is useful in long-term dietary intake estimations. For this purpose, the median of the residues in the tissues of animals slaughtered at the shortest interval after treatment (or later if residues were higher later) is taken to represent that typical value.

5.2 Storage Stability of Residues Studies

The aim of these studies is to demonstrate the time period for which stability has been shown in representative commodities from animal, by extrapolation to processed fractions derived from products of animal origin. It is then necessary for applicants to ensure that Magnitude of Residue (MOR) samples are analysed within the shortest period for which stability has been shown in representative commodities.

Storage stability studies for material from animals that have been treated with ectoparasiticides in the field, or from the spiking of control commodities with known amounts of each component of the residue definitions should be provided. Freshly spiked control samples of the stored commodities should be analysed at each of the time points when aged/stored commodities are removed from frozen storage for analysis. At least two sampling intervals (time zero and other) should be used; the sampling interval depends on the stability of the residues. Duplicate samples of every commodity at each time point for all components of the residue definitions should to be analysed.

5.3 Metabolism, Distribution, and Expression of Residues

Metabolism in Livestock studies are used to determine the qualitative and quantitative metabolism and/or degradation of the Active substance resulting from ectoparasiticides use on direct application to livestock, or premise treatment. The studies provide an estimate of total residues in the edible livestock commodities, as well as the excreta; identify the major components of the terminal residue in the edible tissues; elucidate a metabolic pathway for the ectoparasiticides in ruminants and poultry; provide evidence whether a residue should be classified as fat soluble.

Results from a ruminant (lactating goats) study should be provided for a single animal per experiments. For poultry (laying hens), the use of ten birds per experiments (or dose) is recommended. The minimum dosage used in livestock oral metabolism studies should approximate the level of exposure expected from the feeding of treated crops with the highest observed residues. Treatment should be administered orally (via a balling gun, capsule, or gavage) or by dermal application. The study includes: the excreta, milk and eggs collect (twice daily), and tissues collected.

The objective of OECD Test Guidelines for the ectoparasiticides residue chemistry is to assess ectoparasiticide exposure by identifying these residues in food or animal feedstuffs for purposes of dietary risk assessment and setting Maximum Residue Levels. They have been developed and are based on guidelines in use for many years in OECD countries and by the Food and Agriculture Organisation. Because of the unique nature of each study, the ectoparasiticides expected use, and the methods needed to elucidate the metabolic pathway for each chemical, the description of the test method cannot be as prescriptive as usually required for other OECD Test Guidelines. ectoparasiticides residue studies are complex; guidelines cannot specify all parameters in advance, but each study must be designed individually.

The following livestock animal metabolism studies results should be provided.

a) a summary of the pharmacokinetics of the ectoparasiticide product and its residues,

   i. An estimate of the total terminal residues in the edible animal commodity

   ii. studies on how long the product or its metabolites persist in animal tissues,

   iii. Identified major components of the total terminal residues.

   iv. the practical withdrawal periods that should be observed before slaughter of the animals for consumption, or consumption of other food products from live animals eg. eggs, milk,

v. State the distribution and nature of residues in muscle, fat, milk, eggs, liver, and kidney, to identify target tissues and determine whether the residues are fat-soluble.
vi. Show the efficiency of extraction procedures for various components of the residues.

vii. the analytical methods suitable for verifying the appropriateness of the withdrawal period.

viii. identified bound residues.
ix. determine residue definitions for enforcement and risk assessment.

Residue studies for ectoparasiticides to be administered to food-producing animals, the following shall be investigated and shall include:

Measuring the mode and extent of excretion or elimination of the parent compound and/or its degradation products in livestock to identify any potential for bioaccumulation.

Further information is available at the OECD Publications on Pesticide Residues website and the Food and Agriculture Organization's JMPR Guidance and related documents website

5.3.1 Nature of the Residue in Meat, Milk, Poultry, or Eggs

Metabolism in Livestock studies are needed to elucidate the absorption, and disposition of Active substance s whenever an ectoparasiticides use may lead to residues entering the human food chain. In addition, in vitro data are useful to show if the ectoparasiticides is likely to undergo hydrolysis (acid, base, or enzymatic), oxidation or reduction, photolysis, or other changes. Applicants should provide the proposed metabolic pathway, including a table with associated chemical structures and names (Chemical Abstract Service (CAS) and International Union of Pure and Applied Chemistry (IUPAC) as available). Any postulated intermediates/metabolites should also be indicated in the pathway. The capability of the analytical methods utilized in the metabolism study to determine the components of the residue, whether free, conjugated, or unextractable, should be clearly specified. Applicants should always be alerted to the possibility of new and unexpected metabolites of the ectoparasiticides, which may affect future maximum residue limit (MRL) proposals. Where the structure of a metabolite or alteration product is identical to that of another registered ectoparasiticides chemical and the information is in the public domain, the applicant should state this fact.

The objective of these studies is to provide a qualitative approval of the absorption, translocation, and disposition of the residue. Applicants are free to include additional animals if they can justify scientifically the need. If the applicant wishes to request a waiver of the data elements associated with the livestock feeding by using the Metabolism in Livestock study, inclusion of a second animal (or group of animals in the case of poultry) treated with a realistic dose is strongly recommended. In addition, the applicant may wish

to extend the dosing period for the second animal if it is suspected that a plateau is not likely to be reached. The proposed waiver for the feeding studies would require fully adequate scientific reasoning, especially if a plateau has not been reached in milk or eggs in the metabolism study.

5.3.2 Chemical Identity

Livestock metabolism studies should be conducted using radiolabelled test compound. The applicant is to provide the identification and characterization of at least 90% of the total radioactive residue (TRR) in each edible tissue, milk, or eggs. In many cases it may not be possible to identify significant portions of the TRRs especially when low total amounts of residue are present, when incorporated into biomolecules, or when the ectoparasiticides is extensively metabolized to numerous low-level components. However, it is important for the applicants to clearly demonstrate the presence and levels of the components, and if possible, attempt to characterize them.

5.4 The Nature of the Residue in Livestock

The residues in Livestock studies are conducted to quantify levels of residues in meat, milk, eggs, and edible meat by-products following the use of an ectoparasiticide product. The situations to which such studies apply include ectoparasiticides that may be directly applied to livestock; and ectoparasiticides that are used in livestock premises. The primary purposes of the Residues in Livestock study are to provide: the basis for establishing maximum residue limits (MRLs) and for conducting dietary intake assessments for consumer safety. Separate feeding studies data should be provided for a ruminant (lactating dairy cows) and poultry (egg-laying hens). The test results should be for daily (during at least 28 days) preferably by capsule. Residues in Livestock study should comprise 3 different dose levels, 1X, 3X and 10X. Three animals per dose group (and one for the control) should be used for ruminants. For hens 9-10 animals per dose group (and 3 to 4 animals for control per study) should be provided. The study report should include daily feed consumption, bodyweights measurement, milk or egg production and analysis (after and before dosing), detailed observations (health problems…) and tissues analyses.

5.4.1 Residue Data

Both the field and laboratory phases of residue studies conducted and used to support the establishment of MRLs in food and feed commodities, must be generated in accordance with the OECD principles of good laboratory practice (GLP).

Applicants conducting GLP-compliant residue trials should refer to the OECD's publication, No.1: Principles of good laboratory practice.

For a study conducted to be GLP-compliant, it must be undertaken by a facility that is internationally accredited and within the regional GLP compliance monitoring program. Overseas studies must be conducted by

facilities covered by the relevant country's GLP compliance monitoring program.

5.4.2 Residue Studies

Data should show whether, and at what level, residues occur in edible animal tissues, milk, and eggs. The formulation to be sold in EAC region should be subjected to trials that address the maximum use rate. You may also run a concurrent trial using a rate of 1.5 to 2 times the maximum recommended application rate at one of the trial locations.

Experiments should show the rate of disappearance of residues and/or the interval that elapses before the residues substantially disappear. The study results should be sufficient to enable the setting MRLs and withholding periods correctly. There should be no extrapolation of data, but actual figures should be used. Residue studies should therefore be extensive enough to take care all probable sources of variation in residue level.

Provide residue data from livestock applications and/or argument to justify the safety of the proposed use for products intended for application on the livestock, and for which no MRL and withholding period has been established.

5.4.3 Residue Trials

You must provide full details of study procedures, including data on variables that might influence the decline of residues. Each study reported must include an abstract summarizing the methodology and the results. Each application should contain a summary of the residue application. Normally the summary should not be extended beyond a few pages.

Identifying information:

- Trial reference, volume, and page number
- Year
- Location
- Dose rate
- Number of applications and intervals
- Date
- Animal
- End-use product name
- Active Substance
- Formulation type and level of active Substance
- Recommended dose rate
- MRL proposed.
- Withholding period proposed

5.5 Livestock Housing Residue Data

Residue data are required for ectoparasiticides that are to be applied to livestock premises, regardless of whether livestock are likely to be present at the time of application. Data should be presented to show the levels of

residues in commodities from livestock and poultry normally held in such premises. Those commodities include meat, fat of meat, edible offal, eggs, and milk.

Applicants intending to register products for the control of ectoparasite species in animal housing, including sties, pens, stables, piggeries, poultry houses and feedlots. Animal housing residue studies should be provided when active substance is a volatile compound that may migrate from its off-animal application site to contact animals or may contaminate their feed. The degree of volatility of an active substance should be assessed from its vapor pressure, which is information required for the chemistry assessment of the compound. However, for compounds that are moderate or low volatility, but known from metabolism studies to accumulate in edible animal tissues, an animal housing study should be conducted under a maximum treatment regime.

The maximum treatment regime recognizes that in many cases it is not practicable to remove animals from their housing while treatment takes place. An exception would be milking sheds. The study should be conducted using the species and animal housing situation that give the most potential for animal exposure. It is essential that data are submitted to demonstrate residues resulting from a maximum treatment regime. This will enable maximum residue limits to be set high enough so that residue violations will not occur when the product is used according to the maximum label use pattern. Data from overseas studies will be an acceptable substitute for local data if the work in those studies was carried out using the same use pattern and under similar husbandry conditions.

5.6 Extrapolation from One Species to Another

Data on residue levels in one animal do not necessarily represent the residue levels that might reasonably be expected to occur in distinctly different type of animal.

The Codex classification of foods and animal feeds has been developed by the Codex Committee on pesticide Residues for international use. It provides lists of crops and raw agricultural commodities that are considered essentially similar for the purposes of recommending MRLs.

5.7 Residue Analytical Method

You must provide complete details of the analytical methods used for determining residues in the trials conducted. The methods should:

- Possess a suitable degree of specificity for the veterinary ectoparasiticide in question
- Have a limit of analytical quantification at a level considerably lower than any MRL proposed for finite residues be substantiated by adequate evidence in the form of blanks, recovery data and extraction data to show that the method is effective for determining residues in the substrates analyzed and at the levels under consideration. If the analytical method

involves an instrumental determination such as spectrophotometry, high-performance liquid chromatography or gas–liquid chromatography, specimen output charts showing blank determinations and recovery determinations should be provided to assist in evaluating the method.

It is important to relate the residue analytical procedures applied in the trial to those provided in the supporting documentation. The detailed method of analysis should be clearly identified with a distinctive reference number. The same reference number must then be specified in the section providing the relevant trial data, with an indication that this was the analytical method used to determine the residues.

Because methods can be modified over time, problems arise when, in a subsequent application, reference is simply made to previously submitted methodology. The complete method must again be described in a new application.

A method of analysis suitable for routine monitoring and for regulatory control should be submitted. In some cases, the method may be the same as the method used for determining the residues in the trials conducted for the purposes of requesting an MRL. In other cases, a separate method may be required for regulatory purposes.

5.8 Fate of Residues During Processing and Cooking

The applicant should provide any available information about the effect of processing and cooking on the level of residues at slaughter, and other relevant times, so that the likely ectoparasiticide intake in diets can be estimated. Details should be inclusive of all components of the residue definition.

5.9 Maximum Residue Limits

5.9.1 Proposed Residue Definitions and Maximum Residue Levels

Proposed residue definition should include the toxicological significance of the compounds, the amounts likely to be present, and the analytical methods proposed for post-approval control and monitoring purposes. Two different residue definitions should be provided one for enforcement purposes, based on the marker concept, and one for risk assessment purposes, considering toxicologically relevant compounds.

Analytical work in residue trials shall cover all the components of the residue definition for risk assessment. Proposed maximum residue levels (MRLs) and justification of the acceptability proposed levels. A maximum residue level should be provided for all products of animal origin. In cases where the limits are not determined a guideline level sufficient justification should be provided.

5.9.2 Definition of the Residues Relevant to MRLs

MRL is the maximum concentration of an ectoparasiticide residue (expressed as milligrams per kilogram) recommended by the Codex Alimentarius Commission to be legally permitted in or on food commodities and animal feeds.

5.9.3 Residues Relevant to Consumer Safety

Residue levels in animal commodities, e.g., meat, milk, and eggs, may arise from consumption of feed items containing residues or from direct application to a farm animal of ectoparasiticide to control ectoparasites such as ectoparasites. Methods of estimating maximum residue levels in animal commodities have been developed in recent years and their detailed explanations are given in the JMPR reports.

5.9.4 Proposed MRLs and Compliance with Existing MRLs

The limit of analytical quantification (LOQ), also sometimes referred to as the 'limit of analytical determination', is the lowest concentration of an ectoparasiticide residue in a defined matrix for which positive identification and quantitative measurement can be achieved using a specified method. The limit of analytical quantification is generally expected to be at the level of 0.05 milligrams per kilogram or less.

The method must be sufficiently sensitive to accommodate Ectoparasiticide that have low acceptable daily intakes, (for example, an active substance for which the LOQ is set at 0.002 milligrams per kilogram because the acceptable daily intake is 0.0002 milligrams per kilogram per day).

The LOQ is the lowest concentration of a compound that can be determined in a commodity with an acceptable degree of certainty. The JMPR recognizes the difficulties that may arise in regulatory laboratories analysing low levels of residues in samples of unknown origin, and so usually estimates an LOQ which is achievable under those conditions. It is this figure that is proposed as a maximum residue limit "at or about the LOQ". These limits are indicated with an asterisk (*) after the numerical value, e.g., 0.02*. This limit is often referred to as a "practical LOQ" to distinguish it from the LOQs reported in supervised trials.

However, some matrices may make it difficult to have the LOQ as low as 0.05 milligrams per kilogram therefore sufficient justification must be provided. The 1995 JMPR concluded that when residues are undetectable in a commodity an MRL based on the sum of the LOQs of the individual residue components may not be appropriate for enforcement purposes. The best option should be selected on a case-by case basis considering the relative ratio of metabolites.

5.9.5 Establishment of Maximum Residue Limits

The use pattern of an end-use product influences the level and nature of residues that will occur in food. Applications should therefore include the complete, detailed use pattern proposed for the product, including rate, timing (including interval between treatments) and number of applications, and withholding periods. You are also required to nominate MRLs for the active Substance is contained in the product. The MRLs nominated must be compatible with the proposed use pattern of the product and based on the Codex nomenclature for commodities.

Regardless of the apparent lack of toxicity of residues, human intake of agricultural chemicals should be kept to a minimum consistent with effective use. If the residue level immediately after application is unacceptably high, the use pattern will have to be re-examined or rejected. This guideline describes the general requirements for submitting residue data for the registration of veterinary Ectoparasiticide, including the establishment of maximum residue limits (MRLs) and withholding periods.

The data submitted is evaluated by the EAC Veterinary Medicines Authority. Where relevant, we may take other information into consideration, including recommendations made by other governments and internationally recognized organizations. Evaluations are subject to peer review. Public comment when a product containing a new active substance is considered for registration for the first time will be sort. MRLs may be subject to reconsideration, which could be prompted by a change in use of a chemical or by availability of new data.

With the goal of harmonizing the calculation of MRLs across the OECD, the OECD has developed a MRL Calculator. It is made up of an Excel spreadsheet which is simple to use and doesn't require extensive statistical knowledge from the user. There is one spreadsheet for single data sets and one spreadsheet for multiple data sets. A User Guide and a Statistical White Paper are available to help with the use of the Calculator.

Further information can be found on the Organization for Economic Co-operation and Development (OECD) website (under Publications on ectoparasiticide residues) and the Food and Agriculture Organization of the United Nation's website (under AGP–JMPR Guidance and related documents).

5.9.6 Nomination of Maximum Residue Limits and Withholding Periods

You should nominate a proposed complete use pattern, including withholding periods and proposed MRLs. The pertinent withholding period and residue restraints to be included on proposed labels should be selected from those specified in the relevant Agricultural Labelling Code.

When conducting trials where a finite residue occurs, and/or when a withholding period is necessary, the sampling regime should be spread across the time range within which the withholding period is expected to occur. It is

essential to sample at the recommended withholding period; otherwise, the withholding period will be set at the next longest sampling time.

For further information, refer to the guidelines, Withholding periods and Maximum residue limit proposals 'at or about the limit of analytical quantification'.

5.9.7 Maximum Residue Limits 'At or About the Limit of Analytical Quantification'

If use of the end-use product according to the proposed use pattern (which includes the withholding period) is shown not to give rise to detectable residues in food, an MRL will normally be recommended 'at or about the limit of analytical quantification'. Such use patterns (including the withholding period) must be compatible with good agricultural practice.

If the end-use product is to be used on this basis, the residue trials must show the rate of decline and ultimate reduction of residue levels to the point at which they are in fact 'at or about the limit of analytical quantification'.

Acceptable evidence may include reliable and extensive data from overseas experiments that demonstrate the absence of residues of the parent compound or its toxicologically significant metabolites following application as proposed. However, data from locally conducted trials may still be necessary.

For further advice, refer to the guideline, Maximum residue limit proposals 'at or about the limit of analytical quantification'.

5.9.8 'Finite' Maximum Residue Limits

If use of the end-use product according to the proposed use pattern (which includes the withholding period) is shown to give rise to detectable residues in food, you will need to establish a 'finite' MRL.

This will always be the case when residues are present in an animal at slaughter. Submit appropriate detailed residue data, or present sound scientific justification why such data are not necessary for specific products.

SECTION 6.0 FATE AND BEHAVIOUR IN THE ENVIRONMENT

Veterinary ectoparasiticide undergo change/degradation after application. The nature and extent of the change must be determined before appropriate methods of analysis can be developed and maximum residue limits (MRLs) for commodities can be established. The method might measure the chemical or a derivative of it and can include metabolites. In some cases, the nominal concentration of the parent compound is calculated from the measured concentration of a metabolite; in other cases, a derivative or metabolite is used as a measure of the residue.

Metabolism studies are used to assess the fate of the chemical in target animals and to assess the character of chemical residues in food-producing animals. The composition of a residue (parent and metabolites) and the target organs or food commodities in which it is present (ie muscle, fat, liver, kidney, milk, eggs and honey) should be known, so that residue-depletion trials and analytical methods deal with the relevant residue components.

- All toxicity studies shall be properly presented including the following:
- objectives
- experimental protocol including methodology and materials.
- summarised results and related statistical analysis
- discussion and conclusions
- proposed measures to minimise potential toxicity during use of the product.

6.1 Definition of the Residue Relevant to the Environment

A residue is defined as the chemical, its metabolites, and related compounds to which the MRL applies. The inclusion of specific metabolites or degradation products in the expression of a residue depends on their toxicological profile and the extent to which they occur. Metabolites that occur at levels greater than 0.1 milligram per kilogram or that comprise more than 10 per cent of the total radioactive residues should be identified and provided for evaluation. Including a brief description of the analytical methods used to determine the residue levels in the commodities used as food.

6.2 Behaviour in Environment

Applicants are required to submit the following information to allow an adequate assessment to be made about the potential environmental impact of the active substance in the formulation and related products:

- the expected volume of use
- the expected exposure, behaviour, and fate of the active Substance (s) when the veterinary ectoparasiticide is used as proposed.
- the potential harmful effects on birds, mammals, aquatic life (fish, invertebrates, algae, and higher plants), terrestrial invertebrates (honeybees and other non-target arthropods, earthworms), soil microbial processes, and non-target terrestrial plants.

This information is important in establishing whether the risk to any of these organisms posed by the proposed use of the product may be considered unacceptable or whether there are other concerns due to the behaviour of the substance in the environment.

The applicant should provide data that are relevant tailored to the nature of the proposed application and the anticipated environmental exposure pattern. Ectoparasiticide vary widely in their environmental properties and in the ways that they are introduced into the environment this should be considered while providing data.

6.2.1 Metabolism Studies (In Water and Soil for both Aerobic and Anaerobic Conditions).

The applicant should provide the aerobic and anaerobic transformation of chemicals in soil. Stating the rate of transformation of the test substance, the nature and rates of formation and decline of transformation products to which plants and soil organisms may be exposed. This study is required for veterinary ectoparasiticide because they are likely to reach the soil environment. Aerobic and anaerobic studies with one soil type are generally sufficient for the evaluation of transformation pathways however for Rates of transformation three additional soils should be used. The types of soils tested should be representative of the environmental conditions where use or release will occur. For example, chemicals that may be released in subtropical to tropical climates should be tested with Ferrasols or Nitosols (FAO system).
OECD Test No. 307: Aerobic and Anaerobic Transformation in Soil.

6.2.2 Fate and Behaviour in Air

The applicant should provide the behaviour of veterinary ectoparasiticide in air, ways of degradation, degradation products in air: Describe ways and speed of degradation in air and break down products formed. (For fumigants, spray formulations and volatile products). Route and rate of degradation in air, Transport via air, over short or long distances.

6.2.2.1 Ectoparasiticide in Air

The applicant should provide the expected contamination of air by ectoparasiticide through spray drift, volatilization of the product from animal body, soil and surface waters, and wind erosion of soil particles containing adsorbed ectoparasiticide. The fraction of ectoparasiticide emitted to ambient air should be determined and provided. For ectoparasiticides not rapidly degraded in air the levels in the atmosphere must be determined and provided.

6.2.2.2 Atmospheric Concentrations

The applicants should provide concentrations of ectoparasiticide in ambient air especially for those that persist for a longer time. Including their fate in ambient air. Their concentrations in ambient air should be measured both

from near emission regions and in distant areas. Ectoparasiticide can move through the atmosphere by volatilization in relatively warmer source regions, transport in air, and subsequent deposit in the soil and water bodies.

6.2.3 Bioaccumulation Study in Fish

The ectoparasiticide should not be used near water bodies if use of ectoparasiticide near aquatic sites is unavoidable. Adequate data must be provided for commodities from water, fish and shellfish, irrigated crops, and meat, milk, poultry, and eggs to demonstrate both the nature of the residue and the level of residues resulting from the maximum proposed use. Test procedure should characterize bioconcentration potential of substances in fish, using an aqueous (standard and minimized tests) or dietary exposure.

Test No. 305: Bioconcentration: Flow-through Fish Test

6.2.4 Fate and Behavior in Water and Sediment

Ectoparasiticide can enter shallow or deep surface waters by such routes as direct application, spray drift, run-off, drainage, waste disposal, industrial, domestic, or agricultural effluent and atmospheric deposition. The aerobic and anaerobic transformation of organic chemicals in aquatic sediment systems should be provided.

State the ways of degradation of the products in water:

Describe ways and speed of degradation of the active substance in water.

Specify the major break down products (metabolites) formed and their adsorption/desorption on sediments.

State the DT50 (days); Specify the half-life of the active substance in water.

Describe ways and speed of degradation in surface and ground water.
Test No. 308: Aerobic and Anaerobic Transformation in Aquatic Sediment Systems

6.2.4.1 Degradation and Dissipation Studies (Hydrolysis, Photolysis in Water and Soil)

The applicant should submit photo transformation of ectoparasiticides in water to determine the potential effects of solar irradiation on chemicals in surface water, considering direct photolysis only. The maximum possible losses should be estimated at 50% of the initial concentration over a 30-day period.
Test No. 316: Photo transformation of Chemicals in Water – Direct Photolysis

6.2.4.2 Fate and Behaviour in Soil

Behaviour, ways of degradation, and degradation products in soil: Indicate the degradation path of the active substance in the soil and the degradation products formed. The method described in this Test Guideline is based on soil

column chromatography in disturbed soil. Two types of experiments are performed to determine (i) the leaching potential of the test substance, and (ii) the leaching potential of transformation products in soils under controlled laboratory conditions.

At least duplicate leaching columns are packed with untreated, air-dried, and sieved soil (< 2 mm) up to a height of approximately 30 cm. Afterwards they are saturated and equilibrated with an "artificial rain" solution and allowed to drain. Then the surface of each soil column is treated with the test substance (non-volatile in water and soil) and/or with aged residues of the test substance. Artificial rain is applied to the soil columns and the leachate is collected. After the leaching process the soil is removed from the columns and is sectioned into an appropriate number of segments depending on the information required from the study. A reference substance (atrazine or monuron) should be used in the leaching experiments. For each soil segment and leachate fraction, the amounts of test substance, transformation products, non-extractables and, if included, of the reference substance should be given in % of applied initial dose.
Test No. 312: Leaching in Soil Columns.

6.2.5 Mobility Studies (Leaching and Adsorption or Desorption Studies, Volatility in Laboratory and Field)

Provide the degree of adsorption of the active substance in the soil and the degree of mobility of the metabolites in the soil. Specify the degree of mobility of the active substance in the soil hence leaching potential and possibility for ground water contamination. Determining major metabolites and describe the major metabolites fate in the soil. The DT50 (days) should be submitted; Specify the half-life of the active substance in various types of soils. If high, provide details on further studies. The veterinary ectoparasiticide may reach soil via indirect routes (e.g. via wastewater, sewage sludge, soil or air wet/dry deposition). For risk assessment of these chemicals, the applicant should provide an estimated potential for transformation in soil and for movement (leaching) into deeper soil layers and eventually into groundwater. Using soil thin-layer chromatography, soil thick-layer chromatography, soil column chromatography, and adsorption - desorption measurements. For non-ionised chemicals, the n-octanol-water partition coefficient (Pow) allows an early estimation of their adsorption and leaching potential. The method described in this Test Guideline is based on soil column chromatography in disturbed soil for definition two types of experiments are performed to determine (i) the leaching potential of the test substance, and (ii) the leaching potential of transformation products (study with aged residues) in soils under controlled laboratory conditions.

6.2.6 Veterinary Ectoparasiticide in Groundwater and Drinking Water

Ectoparasiticide applied to animals may in small quantities vertically displaced downwards from the topsoil through the soil profile and the unsaturated zone to groundwater, a process called leaching. Since

groundwater is an important source of drinking water, both environmental and human health effects may be caused by groundwater ectoparasiticide pollution. The extent to which ectoparasiticide leach to groundwater depends on many factors related to soil and ectoparasiticide properties, site conditions and management practices,

Ectoparasiticide properties also play an important role. The higher the water solubility of an ectoparasiticide, the greater its potential to dissolve in water infiltrating the soil. Ectoparasiticide for which there is only a short time to detect a 50 per cent decrease in ectoparasiticide concentration (detection time 50 per cent; DT50) may be degraded before reaching groundwater levels. Similarly, Ectoparasiticide with a high soil adsorption coefficient (Koc) are expected to be retained in topsoil layers. It should be noted, however, that ectoparasiticide with low water solubility and high Koc (and DT50) values have greater potential for particle-bound transport, i.e., adsorption to particles in infiltrating water. Therefore, the applicant should provide data on movement of the ectoparasiticide in soil. The difficult accessibility of groundwater ecosystems hampers monitoring of ectoparasiticide, which is often restricted by the availability of superficial sampling spots.

6.3 Effects on Soil Organisms

This Guideline describes procedures designed to assess bioaccumulation of chemicals in earthworms. The applicant should provide parameters which characterise the bioaccumulation of a substance include the bioaccumulation factor (BAF), the uptake rate constant (ks) and the elimination rate constant (ke). The test should consist of two phases: the uptake (exposure) phase and the elimination (post-exposure) phase. An elimination phase is always required unless uptake of the test substance during the exposure phase has been insignificant. The test organisms are exposed to the test substance during the uptake phase. The test substance is incorporated into the soil; it is recommended to use the artificial soil described in the OECD Test Guideline 207 (Earthworm, acute toxicity test). The uptake phase should be of 14 days (enchytraeids) or 21 days (earthworms) unless it is demonstrated that steady state has been reached. For the elimination phase, the worms are transferred to a soil free of test substance. The elimination phase is generally of 21 days. *Test No. 317: Bioaccumulation in Terrestrial earthworms.*

6.4 Effects on Non-Target Species

6.4.1 Effects on Terrestrial Vertebrates (Including Acute Oral Toxicity to Avian Species E.G., Pigeon, Quail, Pheasant, or Duck).

The applicant is to estimate the acute oral toxicity of test chemicals to birds and provide results for three testing options: (1) limit dose test, (2) LD50-slope test, and (3) LD50-only test. The LD50-slope and LD50-only options are sequential testing procedures. The test method selected should depend on whether a definitive median dose (LD50) and slope of the dose-response curve are both needed. Sequential testing procedures target the placement of doses and match the precision of the endpoint with the precision required. These

sequential procedures are designed to minimize the numbers of birds used. A computer programmer– Sequential designs Calculator (SEDEC) – is available to aid the placement of doses and estimate the LD50, slope and confidence limits.
Test No. 223: Avian Acute Oral Toxicity Test

6.4.2 Effects on Aquatic Species

6.4.2.1 Acute LC_{50}, 96 Hours Exposure on the Suitable Fish Species

The fish are exposed to the test chemical for a period of 96 hours, under either static, semi-static or flow-through conditions. Mortalities and visible abnormalities related to appearance and behavior are recorded. Where possible, the concentrations to kill 50% of the fish (LC50) are determined.
OECD Test No. 203: Fish, Acute Toxicity Test

6.4.2.2 Acute LC_{50}, 48 Hours Exposure on One Suitable Fish-Food Species E.G., Daphnia

The applicant is to assesses the effect of ectoparasiticide on the reproductive output of Daphnia magna Straus. Using young female Daphnia to the test substance. For semi-static tests, results from at least 10 animals at each test concentration and for flow-through tests, 40 animals divided into four groups of 10 animals at each test concentration are used. The test duration should be 21 days. The total number of living offspring produced per parent animal which does not die accidentally or inadvertently during the test and the number of living offspring produced per surviving parent animal at the end of the test are reported. The study report should also include: the daily counting of the offspring, the daily recording of the parent mortality, the weekly measurement of oxygen concentration, temperature, hardness and pH values and the determination of the concentrations of test substance. Optionally other effects can be reported, including the sex ratio of the offspring. The reproductive output of the animals exposed to the test substance is analyzed, by comparing it with that of the control in order to determine the lowest observed effect concentration (LOEC) and hence the no observed effect concentration (NOEC), and by estimating the concentration that causes an x % reduction in reproductive output by means of a regression analysis.
OECD Test No. 211: Daphnia magna Reproduction Test

6.4.3 Effects on Earthworms and Other Soil Macro- Organisms (Including Acute Toxicity on Earthworms)

This Test Guideline includes two methods: a paper contact toxicity test and an artificial soil test. The recommended specie is Eisenia foetida (Michaelsen). The initial screening test (filter paper contact test) involves exposing earthworms to test substances on moist filter paper in order to identify potentially toxic chemicals to earthworms in soil. Five or more treatment levels in a geometric series and, at least, ten replicates (one worm per vial) for each treatment should be used. Tests are done in the dark and for a period of 48 hours. The artificial soil test gives toxicity data more representative of natural

exposure of earthworms to chemicals. It involves keeping earthworms in samples of a precisely defined artificial soil. Five concentrations, in a geometric series, of the test substance have been applied. One concentration resulting in no mortality and one resulting in total mortality should be used. Four replicates for each treatment are recommended. Mortality is assessed 7 and 14 days after application.
OECD Test No. 207: Earthworm, Acute Toxicity Tests

Proposed measures to minimise the above potential risks during use of the product shall be described.

6.4.4 Effects on Bees and Other Arthropod Species (Including Acute Oral LD_{50}, And Contact Toxicity on Honeybees)

This is a laboratory test method, designed to assess the acute contact toxicity of ectoparasiticide and other chemicals to adult worker honeybees.

Anaesthetized adult worker honeybees are exposed to five doses in a geometric series of the test substance dissolved in appropriate carrier (in total a volume of 1 ml), by direct application to the thorax (droplets). A minimum of three replicate test groups, each of ten bees, should be dosed with each test concentration. A toxic standard (usually dimethoate) should be included in the test series. The limit test corresponds to one dose level of 100 ìg Active substance /bee. The test duration is 48h. Mortality is recorded daily during at least 48 hours and compared with control values. If the mortality rate is increasing between 24 and 48h whilst control mortality remains at an accepted level, it is appropriate to extend the duration of the test to a maximum of 96h. The results are analysed in order to calculate the LD50 at 24h and 48h and, in case the study is prolonged, at 72h and 96h.
OECD Test No. 214: Honeybees, Acute Contact Toxicity Test

7 SECTION 7.0 EFFICACY STUDIES

7.1 Background Information

Introduction

This guideline provides general requirements for the assessment of efficacy of an ectoparasitic ide preparation, containing novel or established Active substance s. It is the purpose of treatment with ectoparasiticide to eliminate or to reduce arthropod parasites or to protect animals from them, to maintain animal health and to prevent losses in production. ectoparasiticide intended for external or internal use should fulfil all minimum requirements of approval of veterinary medicinal products. Where a claim for control of infestation is made, the period it takes to achieve control and the period over which control is achieved must be demonstrated. At the end of the period as indicated by the applicant, the overall efficacy of ectoparasiticide in controlling infestations of domestic animals should be achieved as follows:

Fleas: approximately 100%

Lice: approximately 100%

Mites: approximately 100%

Sarcoptic scabiei and, if possible, more than 90% for other mange mites

Ticks: more than 90%

Diptera: 80-100% (preferably more than 90%)

Larval arthropods: 80-100% (preferably more than 90%)

Where indicated and justified, clinical parameters may be used to support the efficacy of a product. Where efficacy is less than the above no claim should be made unless the applicant can demonstrate that the degree of efficacy achieved is better than or comparable with current alternatives. All claims for efficacy of the product against species of ectoparasites must be validated. While in principle, the results of dose titration and dose confirmation trials should be acceptable irrespective of where they are carried out, the regional competent authorities may require additional clinical field trials to be undertaken where scientifically justified for the assessment of efficacy of the product, e.g., where normal husbandry or environmental conditions differ markedly from reported test conditions.

EAC region's unique environmental and geographic parameters, parasite burdens and their population dynamics, farm management practices and animal breeds has necessitated the additional need to conduct efficacy trials for products to be registered within EAC Partner States. For the conduct of efficacy trials in the region the applicants are advised to refer to EFT guideline on the EAC Livestock website.

7.1.1 Delivery Systems

Consumer convenience is an important factor in product choice, especially for flea and tick control. An array of delivery systems has historically been available: powders, aerosols, sprays, shampoos, rinses, dips, spot-Ons, mousses, injectables, oral tablets or liquids, and impregnated collars. However, the safety, efficacy, and ease of use of the newer spot-on and oral application systems is proving to be more efficient.

7.1.2 Laboratory Model Efficacy Studies

The applicant should provide laboratory model efficacy studies, where appropriate. Data from such studies may indicate the extent and type of further studies that may be submitted for the target species. Laboratory model efficacy studies may also be used to support valid scientific argument but not to replace target animal efficacy studies. Pivotal studies may include laboratory/pen trials and clinical/field trials for dose confirmation. In some instances, *in vitro* studies or pharmacological/physiological end points may be used as pivotal data. This depends on the type of application, the active Substance s, and the type of product, and will be assessed on a case-by-case basis.

7.2 Dose Determination / Dose Confirmation Studies

7.2.1 Persistent Efficacy Testing

Persistent efficacy is a measurement of the product's continued efficacy in the face of continuing challenge by the target parasite. Persistent efficacy implies the presence of an ectoparasiticide at levels that are relevant in terms of efficacy in time. If the product is claimed to be effective for a seasonal period of ectoparasite activity, the study must be conducted over the entire season. As the dosage needed for relevant protection can be different to the dosage needed for initial treatment, specific dose determination studies are necessary in relation to persistence of efficacy, unless the initial treatment dosage is used. It should be noted that products achieving complete elimination by a single treatment are likely to produce protection for a period as well. However, if both claims (treatment and prevention) are made, this should be demonstrated accordingly. Volatile active substances that are applied topically to the animal may produce a repellent effect, preventing parasites from contacting the animal. Non-volatile substances lack a repellent effect and are only effective after the parasite has come into contact with the substance or ingested the substance. These differences should be considered when designing studies for a persistent efficacy claim. A persistent efficacy is useful for two reasons:

To achieve complete parasite control in infested animals with a single treatment only. This depends on the characteristics of the active substance, the formulation and dosage used, and the animal species involved.

To protect parasite-free animals from becoming (re)infested.

7.2.2 Dose determination and dose confirmation studies,

Involving acaricides or insecticides administered by any route should be conducted under controlled conditions against natural or artificial (where feasible) infestations of the target parasite. The most usual method is to mix naturally infested with uninfected Guideline on specific efficacy requirements for ectoparasiticide in cattle.

EMA/CVMP/625/2003-Rev.1 Page 5/11 animals; infestation is obtained by transmission through contact. It is accepted that artificial infestation is more difficult to achieve, except for ticks and flies and, to a lesser extent, Psoroptes spp. Dose determination studies should be carried out using at least four groups of infested animals, unless otherwise justified, treated with the proposed dose, half the recommended dose and twice the recommended dose administered by the recommended route and an untreated/vehicle-treated control group respectively. Unless welfare issues are a significant factor, then an untreated control group should be included. Delayed treatment of the untreated control group should be considered after an appropriate time period. It is preferable that the animals should have no history of treatment with acaricide/insecticides or injectable, topical or oral endecto-parasiticides. Prior treatment with acaricide/insecticides or endectocides may be acceptable provided there is a sufficient washout period to guarantee the absence of residual efficacy from any previous treatment. It is acknowledged that naturally infested animals may carry more than one species of ectoparasite. This is acceptable provided that the parasites can be distinguished. Each treatment group should be housed in separate pens throughout the course of the studies to prevent cross infestation or cross contamination. Animals should be treated with the test product once the infestation has become established and, if appropriate (e.g. for mites), they exhibit clinical signs of infestation. To establish a claim, two dose confirmation studies should be conducted with adequately infested animals. A dose determination study can be used in place of one of the dose confirmation studies if the final formulation was used and administered under label conditions.

Guideline on specific efficacy requirements for ectoparasiticide in cattle EMA/CVMP/625/2003-Rev.1 Page 6/11 As described for dose confirmation, a persistence claim should include 2 studies, each with a nontreated and treated group of adequate size and adequately infested animals. Such studies can be a continuation of a controlled efficacy study. Following treatment, cattle should preferably be artificially challenged by placing a suitable number of live parasites directly on to the skin at predilection sites (and the areas recorded on a body map in the study protocol), or into isolation cells fixed on to the skin or coat, depending on the mode of action of the substance. Where artificial challenge is not possible, challenge by contact with cattle carrying natural infestations is acceptable. The reasons for use of this method of challenge should be adequately justified. Approximately 25-50% of the herd should be left untreated as a reservoir of infestation. Untreated animals should be examined for the presence of live parasites prior to being re-introduced to the main infested group. Before these examinations and depending on the mode

of action of the active substance, it is permissible to separate the treated animals from the untreated for a period of 1-2 days as appropriate for the proposed claim. The numbers of live parasites should be counted as described above. The duration of the post-treatment interval before the first challenge will depend upon the proposed claim and should be justified by the applicant. Subsequent challenges should be made, within the same area at weekly intervals (or more frequently if justified) post-treatment, depending on the claim. Animals should be examined for the presence of live ectoparasites and the development of any lesions after each challenge. A breakdown in residual activity is recorded when live parasites are detected and, in the case of ticks, are attached and feeding. The actual length of protection is recorded as the last date of challenge that failed to initiate an infestation. For example, in conducting a study where a challenge is made once a week, if live parasites are observed 49 days post treatment, then the period of persistent efficacy is 35 days since the breakdown could have occurred after the challenge on day 35.

7.2.3 Clinical Trials

Clinical or field trials demonstrate efficacy under real conditions. Efficacy claims should therefore be supported by trials that generate clinically relevant, statistically significant data results. Suitable clinical end points that reflect the efficacy or safety issues should be used. The studies should use the product formulation that is to be marketed and should be carried out where the disease or condition occurs under optimal rather than marginal conditions. If the pivotal clinical or field trials supporting efficacy are conducted overseas, EAC regional confirmatory field trials should also be submitted.

Clinical trials are normally carried out on identified infested herds. Preferably, herds should not have been treated with any acaricidal or insecticidal spray, pour-on, injection or drench unless a sufficient washout period has elapsed to guarantee the absence of residual efficacy from any previous treatment prior to the trial. Preferably cattle should only be infested with ectoparasites belonging to the same order, i.e. infestation with lice only and not with lice and mites. However, it is acceptable for more than one species to be present. In this case, all species should be documented, and the dose and treatment schemes should be known for each ectoparasite species. The product should cover all parasite species present or relevant at the moment of treatment and be carried out in accordance with the label recommendations of the final product. Where an untreated control group is not justified because of animal welfare reasons, a positive control using an established product may be included. It should be noted that the non-inclusion of control animals is justified in exceptional cases only, e.g. sarcoptic mange and psoroptic mange. The number of trials to be conducted and animals involved in each trial will depend on the ectoparasite species, the geographical location and local/regional situations. However, usually they should be conducted in at least 2 different geographical and climatic regions. Guideline on specific efficacy requirements for ectoparasiticide in cattle EMA/CVMP/625/2003-Rev.1 Page 7/11 The choice of sampling times should be justified e.g. in

respect to the seasonal or daily time of maximum infestation with ectoparasites, taking into account sites of predilection. Efficacy should be demonstrated in at least two different common breeds to represent the target population. For topical products (e.g. spray, pour-on), the effects of coat length and density should be considered. Climatic conditions (rainfall, relative humidity, sunshine etc.), and faecal contamination, dirtiness of the coat should be documented to assess any effect of these parameters, if relevant.

7.3 Target Animal Efficacy Studies

Demonstration and confirmation of the efficacy veterinary ectoparasiticide product three-step approach should be used in dose determination studies, dose confirmation studies and confirmatory clinical/field studies. Provide a comprehensive summary of the efficacy studies, with a brief outline of the methods and a description and interpretation of the results. Individual summaries should also be provided with each study if not already included in the study report.

The type of data that may be provided to support the safety of the proposed new product in the treated species varies depending on the toxicological hazard and the intended use of the product.

Data should indicate the margin of safety to target animals and should take into consideration factors such as age, sex, breed, condition, dose regimen, animal husbandry practices, pregnancy, nutritional status, and any other matters that could reasonably be expected to affect safety in use.

The effect of treatment on reproduction and the effect of repeat treatments should also be considered. If there are no reproductive studies, we may consider the inclusion of a standard precautionary statement on the label.

We recommend that safety studies use the formulation intended for marketing, administered by the means recommended on the product label and under the proposed conditions of use. These criteria help to satisfy us about the safety criteria relevant to target animals. If the formulation intended for marketing is not used, provide data and/or valid scientific argument to bridge differences between the formulations. You may provide information on alternative methods for administration if it is available.

Either local or overseas data on the safe use of the proposed product are acceptable.

A comprehensive summary of the safety studies should be included, with a brief outline of the methods and a description and interpretation of the results. Individual summaries should be provided with each study if they have not been included as part of the study report.

The study report shall be provided in the outlined format below.

Controlled clinical efficacy studies.

Local tolerance studies to determine the maximum tolerable dose.

Objectives (see classification above)

Identity and qualifications of key personnel involved.

Location(s) of study

Dates of study

Design:

- selection of animals (inclusion, exclusion criteria)
- animal housing and feeding
- selection of control animals
- selection of control treatment (if applicable)
- number of animals
- response variables - end points
- minimisation of bias - randomisation, blinding compliance.

Treatments given:

- identity and quality of the investigational and control products used.
- dosage used
- duration of treatment
- duration of observation periods
- any concurrent treatments and their justification
- analytical methods for determining product concentrations in body fluids, & tissues.
- analysis of results including statistical analysis.

Discussions and conclusions on efficacy and safety, including but not limited to:

- suitability for control of ectoparasites such as ticks, mites, flies, and mange
- applicability of the clinical studies to the EAC region
- adverse reactions observed and their relationship with the administered dose.

7.3.1 Margin of Safety Studies

The margin of safety is defined as the ratio between the maximum recommended dose and the minimum dose producing toxic effects. We recommend that the margin of safety be determined as closely as possible when the margin is less than 5×. Taking animal welfare principles into consideration for the study design, toxic effects should be identified and described.

7.3.2 Duration of Treatment

A safety study should also be conducted using the recommended dose rate over a period that is a multiple of the proposed duration of treatment. Different

study periods may be required, depending on the nature of the product, the target animal species and the use pattern. Products proposed for administration over a period of less than two weeks should be studied over at least three times the recommended maximum duration of use. If a product is recommended for long-term administration (more than two weeks and less than three months), you should conduct safety studies in which the drug is administered for the recommended maximum duration of use or longer (with a minimum of six weeks).

If the product is recommended for ongoing use, safety studies should be conducted for at least three months.

7.4 Individual Clinical Studies

Shall Include the Following:

In the design of efficacy studies, the following must be considered:

The kind of effect(s) exerted by the Active substance (s) (e.g. flushing out, repellent, killing, anti-feeding or detaching effect, insect growth regulating effect, larvicidal, ovicidal, adulticidal or pupicidal effect);

Occurrence and susceptibility of ectoparasites in different geographic and climatic regions.

Control of ectoparasite-related diseases if indicated;

Safety of the target animal;

Pharmacokinetic behaviour of the substance under investigation.

Data on drug resistance of ectoparasite species, where available;

Products intended for the treatment of ectoparasitic conditions may affect the environment, etc. Due regard should be given to legislation in respect of operator, consumer, and environmental safety. For fixed combination products containing two or more Active substance s, it will be necessary to assess the potential advantages in the control of ectoparasites against possible disadvantages (e.g. synergistic or additive actions; antagonism; substitution of effects; non effect (overkill), taking into account the note for guidance on Fixed Combination Products.

7.5 Ectoparasiticide for Cleansing Tick-infested Cattle

Given the importance of maintaining tick control and prevention of vector borne disease in EAC region, if products claim efficacy for cleansing tick-infested cattle then treatments according to label claim should be used to ensure that cattle are free from the ticks.

If a claim for the cleansing of tick-infested cattle is requested, the applicant should conduct multiple treatments of infested cattle according to

manufacturer direction and carry out efficacy assessments. However, examinations to support such a claim should include records of adult ticks collected/counted and detailed observations of the presence of other tick stages on the animals after they have been treated.

7.6 Target Parasite Efficacy

Because of specific formulation and drug delivery technology, certain insecticides are used in a wide variety of ectoparasite control products. Efficacy of specific compounds can vary against target species, and resistance to insecticides may develop in specific locations, especially with incorrect, prolonged, or repeated use. It cannot be assumed that ticks and fleas are controlled by the same active compounds; product labels should be carefully read. Products that contain compounds specifically active against the target parasite should be chosen, whether the concern is fleas, ticks, mites, or a combination of these parasites.

Duration of activity (ie, “knockdown” or sustained effects) can be the primary concern in product choices. Products should be evaluated based on both their immediate and residual speed of kill. A rapid residual speed of kill is critically important when attempting to manage flea allergy dermatitis and to reduce the chances of a tick transmitting a pathogen.

Modern parasiticides available for flea and tick control in companion animals provide superior parasite control, but an understanding of the life cycle of the parasites, along with the mode of action of the molecules, is also important. Often, perceived product failures are a result of massive reinfestation from the environment, incorrect product use, or unrealistic expectations.

The proposed intended use(s) of the product shall be stated. Evidence of potential benefit shall be provided. Tests which have been performed on laboratory animals and target animals regarding the efficacy of the product and the indications for which it will be used (pharmacological and clinical trials).

8. SECTION 9.0 EXPOSURE DATA AND INFORMATION

Protecting the health and safety of people from risks associated with veterinary ectoparasiticide involves both the assessment and the management of risks from the potential sources of exposure. Note that while reference is made to ectoparasiticide in this document.

The assessment of risk should be conducted in three steps:

The hazard assessment determines how inherently dangerous a chemical is to human health using primarily toxicity studies on animals, and in vitro studies (tests on tissue or cell cultures)

The exposure assessment determines the way and the extent to which the chemical users, workers or the public may be exposed to A veterinary ectoparasiticide. Human exposure data are used where available, but if not, various exposure modelling techniques should be used.

The overall risk assessment is a function of exposure and hazard that permits appropriate risk management initiatives, including label directions, to be established.

8.1 Safety Data Sheets

The Safety Data Sheets (SDSs) for the veterinary ectoparasiticide should be provided for users. SDSs provide useful information such as:

The identity of the chemical substance.

Physical and chemical characteristics.

Physical and health hazards.

Primary routes of entry.

Permissible Exposure Limits (PELs).

Carcinogenic and reproductive health status.

Precautions for safe handling and use (including PPE).

Spill response procedures.

Emergency and first aid questions.

8.2 Routes of Ectoparasiticide Exposure

The applicant should provide potential health effects that may result from exposure to veterinary ectoparasiticide. This should include properties of the specific chemical (including toxicity), the dose and concentration of the veterinary ectoparasiticide, the route of exposure, duration of exposure,

individual susceptibility, and any other effects resulting from mixtures with other chemicals.

In order to understand how ectoparasiticide hazards can affect human, it is important to first understand how chemicals can get into your body and do damage. The four main routes of entry are inhalation, ingestion, injection, and absorption through the skin and eyes.

8.2.1 Inhalation Exposure

The applicant should provide information on how Inhalation exposure of chemicals occurs. Once chemicals have entered into the respiratory tract, the chemicals can then be absorbed into the bloodstream for distribution throughout the body. Chemicals can be inhaled in the form of vapours, fumes, mists, aerosols, and fine dust. Including the expected symptoms of exposure to chemicals through inhalation like eye, nose, and throat irritation, coughing, difficulty in breathing, headache, dizziness, confusion, and collapse. If any of these symptoms are noted, the operator, bystander or worker should be removed from the area immediately, get fresh air and seek medical attention if symptoms persist.

8.2.2 Ingestion Exposure

The applicant should provide information on how to prevent Chemical exposure through ingestion. Also provide information on how to handle personnel once they ingest accidently by eating or drinking a chemical; with proper housekeeping and labelling, this is less likely to occur. A higher probability of receiving a chemical exposure can occur by way of indirect ingestion. This can occur when food or drink is brought into a chemical laboratory. The food or drink can then absorb chemical contaminants (vapours or dusts) in the air and result in a chemical exposure when the food or drink is consumed. This can also occur when food or drink is stored with chemicals, such as in a refrigerator. Ingestion can occur when a laboratory worker who handles chemicals does not wear gloves or practice good personal hygiene, such as frequent hand washing, and then leaves the laboratory to eat, drink, or smoke. In all cases, a chemical exposure can result, although the effects of chronic exposure may not manifest itself until years later.

Symptoms of chemical exposure through ingestion include metallic or other strange tastes in the mouth, stomach discomfort, vomiting, problems swallowing, and a general ill feeling. The best protection against ingestion of chemicals is to properly label all chemical containers, never consume food or drink or chew gum in laboratories, always wear PPE (such as gloves), and practice good personal hygiene, such as frequent hand washing.

8.2.3 Eye and Skin Absorption Exposure

Some chemicals can be absorbed by the eyes and skin, resulting in a chemical exposure. Most situations of this type of exposure result from a chemical spill or splash to unprotected eyes or skin. Once absorbed by these organs, the

chemical can quickly find its way into the bloodstream and cause further damage, in addition to the immediate effects that can occur to the eyes and the skin.

Symptoms of eye exposure can include itchy or burning sensations, blurred vision, discomfort, and blindness. The best way to protect yourself from chemical splashes to the eyes is to always wear safety glasses in the laboratory whenever eye hazards exist (chemicals, glassware, lasers, etc.). If you are pouring chemicals, then splash goggles are more appropriate than safety glasses. Whenever a severe splash hazard may exist, the use of a face shield, in combination with splash goggles is the best choice for protection.

Symptoms of skin exposure to chemicals include dry, whitened skin, redness, swelling, rashes, blisters, itching, chemical burns, cuts, and defatting.

9.0 REFERENCES

a) Collaborative International Pesticide Analytical Council Ltd. (CIPAC) Handbook, "MT 46 Accelerated Storage Tests by Heating," CIPAC, Hatching Green, Harpenden, Hertfordshire, England (1970).
b) Globally Harmonized System of Classification and Labelling of Chemicals [2015] http://www.unece.org/fileadmin/DAM/trans/danger/publi/ghs/ghs_rev06/English/03e_part3.pdf
c) FAO/WHO Guidelines for the Registration of Ectoparasiticide [2010]; http://www.fao.org/fileadmin/templates/agphome/documents/Pests_Ectoparasiticide/Code/Registration_2010.pdf
d) FAO. 2002. International Code of Conduct on the Distribution and Use of Ectoparasiticide.
e) Revised Version - http://www.fao.org/agriculture/crops/corethemes/theme/pests/pm/code/en
f) Australian Ectoparasiticide and Veterinary Medicines Authority (APVMA): https://apvma.gov.au/node/1060
g) Dept. of Agriculture, Forestry and Fisheries, Republic of South African GL for regulations of pesticide- Guideline of the Registration Process for Agricultural Remedies - https://www.dalrrd.gov.za/doaDev/sideMenu/ActNo36_1947/AIC/Guidelines on the Registration of agricultural Remedies in Non Crop Situations.pdfhttps://www.dalrrd.gov.za/doaDev/sideMenu/ActNo36_1947/AIC/Guidel
h) https://www.epa.gov/sites/default/files/2017-05/documents/final-signed-acc-ss-cc-memo-rev.pdf
i) https://www.epa.gov/pesticide-registration/accelerated-storage-stability-and-corrosion-characteristics-study-protocol
j) https://www.fao.org/pesticide-registration-toolkit/registration-tools/data-requirements-and-testing-guidelines/study-detail/en/c/1186780/
k) https://www.regulations.gov/document/EPA-HQ-OPPT-2009-0151-0019

Note

The application form for registration of veterinary Ectoparasiticide has been provided as a separate document: 'Application Form for ectoparasiticide registration.docx'.

Annex I: Stability Study Protocol for Accelerated

EAC veterinary regulatory agencies will use this protocol to provide the Agency with all the information it needed to decide on the storage stability of veterinary ectoparasiticide. For that reason, EAC requires the applicants to follow this protocol in generating data to fulfil the Storage Stability and Corrosion Characteristics data requirements for veterinary ectoparasiticide registration and renewal process. The study uses a 14-day test duration at elevated temperatures to determine product stability and corrosion characteristics. However, this protocol is not appropriate for all veterinary ectoparasiticide' products/materials.

Applicants must make the determination, based on their knowledge of the physical and chemical properties of their products (such as thermal properties, volatility, packaging, and whether any incidents related to product instability are known), whether their products are suitable for this study or whether the one-year study is more appropriate.

For products which 14-day study protocol is found acceptable, then storage stability testing results from this study must be submitted. If a 14-day study demonstrates product instability or is performed with a product deemed unsuitable for this protocol, then a I-year study must be provided.

Below are the details of the test protocol for some products, provide an adequate study for purposes of fulfilling the Storage Stability and Corrosion Characteristics data requirements. The applicants that conduct a study that does not follow the protocol below should provide sufficient justification for the study conducted is sufficient to support storage stability and corrosion characteristics for regulatory requirements.

The applicants are to use a qualified investigator and should sign the study report. The date and location of the studies should be clearly identified.

Test Details:

1) The test should be conducted with the product in its commercial package or in smaller packages of the same construction and materials.

2) The test shall be conducted in compliance with the Good Laboratory Practice standards (GLP).

3) The test shall be conducted at 54°C ± 2°c for 14 days.

4) The product to be used in the test must be taken from a batch that has passed quality control analysis. The Active substance concentration of the product must be the same as the label claim or meet the certified limits requirements.

5) The concentration(s) of the Active substance (s) in the product shall be determined at the beginning of the test period and after 14 days, using a validated analytical method.

6) Deterioration or degradation of the product during the test period should be determined. At the end of the test period, the product should be examined for physical changes, such as phase separation or clumping, and any changes that would interfere with the usefulness or safe handling of the product if used according to label directions.

7) The product should be quantitatively analysed for Active substance content and changes in impurities because of degradation or packaging deterioration over the test period. Results should be reported as concentration in weight percent.

8) The product and container should be observed for any physical changes at the beginning and end of the test, recording all observations in the raw data.

9) Report any corrosion of the commercial packaging (metal, plastic, or paper containers) in terms of visual observations (e.g., perforations, darkening, leaking, or rust at the seam).

   If corrosion is visually evident, a gravimetric or other evaluation of the container should be conducted.

Reporting

The report must include all information relevant to the test including the following:

1) The duration of the test and the conditions under which the test was conducted.

2) Quantitative analyses for the Active substance and impurities (if new impurities are formed) at the initiation and termination of the test.

3) Description of the physical condition of the product and container at the beginning and end of the test. Any significant variations to the weight of the container (if applicable) must be reported.

4) Details of the validated analytical method used in the test including representative chromatograms.

5) The full study and results should be submitted with the option to self-certify the data. The self-certified data must be assigned and stamped.

Additional considerations:

1) When the product passes the 14-day study period, then storage stability testing is complete.

2) If a product fails the 14-day study (i.e., product instability, degradation or deterioration occurs and/or new impurities are formed after 14 days, the full one-year, room- temperature study must be conducted. The purpose of the one-year study is to determine if an expiration date is needed for the product or if advisory statements resulting from the 14-day test are adequate.

3) If a product fails the one-year storage stability study after failing the 14-day study, an expiration date will be required in addition to an advisory label statement that limits exposure to increased temperatures. Examples of such a statement are "Avoid storage at high temperatures" and "Store in a cool, dry place."

4) If a product passes the one-year storage stability study after failing the 14-day study, an advisory label statement that limits exposure to increased temperatures will be required.

5) Bridging of the accelerated or the full one-year Storage Stability and Corrosion Characteristics data will be allowed for products that are identical or 100% repacks. For all other products, bridging is determined on a case-by-case basis specifically for products that EAC determines to be substantially similar from the product chemistry point of view, i.e., the same active and inert ingredients (differing only in amounts), the same type of formulation (e.g., emulsifiable concentrate, aerosol, wettable powder, granular, etc.) and the same type of commercial packaging.

6) The regulator may request submission of the one-year data for any product if the submitted reports are not satisfactory.

7) If an alternative method is used, it is recommended that the registrant consult with the Agency prior to adopting the test method.