CLH report

Proposal for Harmonised Classification and Labelling

Based on Regulation (EC) No 1272/2008 (CLP Regulation), Annex VI, Part 2

International Chemical Identification:

Emamectin benzoate (ISO); (4’’R)-4’’-deoxy-4’’-(methylamino)avermectin B1 benzoate

EC Number: -

CAS Number: **155569-91-8 (formerly 13751274-4 and 179607-18-2)**

Index Number: -

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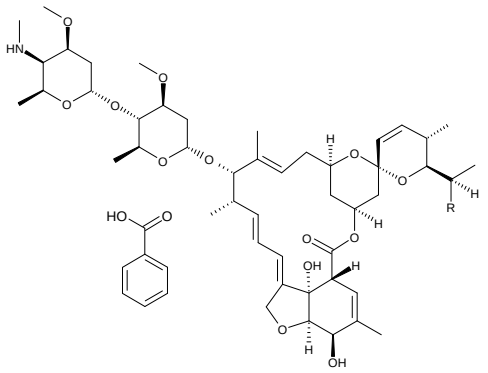
1 IDENTITY OF THE SUBSTANCE

1.1 Name and other identifiers of the substance

Table 1: Substance identity and information related to molecular and structural formula of the substance

Name(s) in the IUPAC nomenclature or other international chemical name(s)	Emamectin B1a benzoate: *(10E,14E,16E,22Z)-(1R,4S,5'S,6S,6'R,8R,12S,13S,20R,21R,24S)-6'-[(S)-sec-butyl]-21,24-dihydroxy-5',11,13,22-tetramethyl-2-oxo-(3,7,19-trioxatetracyclo[15.6.1.14,8.020,24]pentacosa-10,14,16,22-tetraene)-6-spiro-2'-(5',6'-dihydro-2'H-pyran)-12-yl 2,6-dideoxy-3-O-methyl-4-O-(2,4,6-trideoxy-3-O-methyl-4-methylamino-α-L-lyxo-hexapyranosyl)-α-L-arabino-hexapyranoside benzoate* Emamectin B1b benzoate: *(10E,14E,16E,22Z)-(1R,4S,5'S,6S,6'R,8R,12S,13S,20R,21R,24S)-21,24-dihydroxy-6'-isopropyl-5',11,13,22-tetramethyl-2-oxo-(3,7,19-trioxatetracyclo[15.6.1.14,8.020,24]pentacosa-10,14,16,22-tetraene)-6-spiro-2'-(5',6'-dihydro-2'H-pyran)-12-yl 2,6-dideoxy-3-O-methyl-4-O-(2,4,6-trideoxy-3-O-methyl-4-methylamino-α-L-lyxo-hexapyranosyl)-α-L-arabino-hexapyranoside benzoate*
Other names (usual name, trade name, abbreviation)	Emamectin benzoate: (4’’R)-4’’-deoxy-4’’-(methylamino) avermectin B1 benzoate or 4’’-deoxy-4’’-(methylamino)-(4’’R)-avermectin B1 benzoate Emamectin B1a benzoate: (4"R)-5-O-demethyl-4"-deoxy-4"-(methylamino)avermectin A1a benzoate or 5-O-demethyl-4”-deoxy-4”-(methylamino)-(4”R)-avermectin A1a benzoate Emamectin B1b benzoate: (4"R)-5-O-demethyl-25-de(1-methylpropyl)-4"-deoxy-4"-(methylamino)-25-(1-methylethyl)-avermectin A1a benzoate or 5-O-demethyl-25-de(1-methylpropyl)-4”-deoxy-4”-(methylamino)-25-(1-methylethyl)-(4”R)-avermectinA1a benzoate
ISO common name (if available and appropriate)	Emamectin benzoate (ISO status not mentioned) *Emamectin benzoate consists of emamectin B1a benzoate and emamectin B1b benzoate.*
EC number (if available and appropriate)	Not available
EC name (if available and appropriate)	Not available
CAS number (if available)	emamectin benzoate: 155569-91-8 formerly 137512-

	74-4 and 179607-18-2) emamectin B1a benzoate: 138511-97-4 emamectin B1b benzoate: 138511-98-5
Other identity code (if available)	Emamectin benzoate, anhydrous form CIPAC No.: 791.412
Molecular formula	Emamectin B1a benzoate: C56H81NO15 or C49H75NO13.C7H6O2 Emamectin B1b benzoate: C55H79NO15 or C48H73NO13.C7H6O2 Emamectin benzoate exists as the anhydrous and various hydrated forms having different crystal morphologies. The amount of water is non-stoichiometric, however in the hemihydrate the amount of water is more or less fixed
Structural formula	 R = CH_2CH_3 for emamectin B1a benzoate R = CH_3 for emamectin B1b benzoate
SMILES notation (if available)	Not available
Molecular weight or molecular weight range	emamectin B1a benzoate: 1008.3 emamectin B1b benzoate: 994.2 emamectin benzoate hemihydrate: 1016
Information on optical activity and typical ratio of (stereo) isomers (if applicable and appropriate)	Emamectin benzoate consists of emamectin B1a benzoate and emamectin B1b benzoate
Description of the manufacturing process and identity of the source (for UVCB substances only)	Not relevant
Degree of purity (%) (if relevant for the entry in Annex VI)	min 950 g/kg as emamectin benzoate anhydrous (min. 920 g/kg emamectin B1a benzoate and max. 50 g/kg emamectin B1b benzoate)

1.2 Composition of the substance

Table 2: Constituents (non-confidential information)

Constituent (Name and numerical identifier)	Concentration range (% w/w minimum and maximum in multi-constituent substances)	Current CLH in Annex VI Table 3.1 (CLP)	Current self-classification and labelling (CLP)
emamectin benzoate CAS no.: 155569-91-8	95-100%	Not available	

Table 3: Impurities (non-confidential information) if relevant for the classification of the substance

Impurity (Name and numerical identifier)	Concentration range (% w/w minimum and maximum)	Current CLH in Annex VI Table 3.1 (CLP)	Current self-classification and labelling (CLP)	The impurity contributes to the classification and labelling
No relevant impurities (of toxicological, ecotoxicological and/or environmental concern) in the active substance as manufactured that contribute to the classification of the substance				

Table 4: Additives (non-confidential information) if relevant for the classification of the substance

Additive (Name and numerical identifier)	Function	Concentration range (% w/w minimum and maximum)	Current CLH in Annex VI Table 3.1 (CLP)	Current self-classification and labelling (CLP)	The additive contributes to the classification and labelling
No additives					

Table 5: Test substances (non-confidential information)

Identification of test substance	Purity	Impurities and additives (identity, %, classification if available)	Other information	The study(ies) in which the test substance is used
emamectin benzoate CAS no.: 155569-91-8	95-100%	Not available	-	Emamectin benzoate in the anhydrous form was tested in studies related to physiochemical properties, evaluation of health hazards and the evaluation of environmental hazards unless otherwise stated.
emamectin B1a benzoate CAS no.: 138511-97-4	92-100%	Not available	-	Emamectin benzoate in the anhydrous form was tested in studies related to physiochemical properties, evaluation of health hazards and the evaluation of environmental hazards unless otherwise stated.
emamectin B1b benzoate	0-5%	Not available	-	Emamectin benzoate in the anhydrous form was tested in studies related to

Identification of test substance	**Purity**	**Impurities and additives (identity, %, classification if available)**	**Other information**	**The study(ies) in which the test substance is used**
CAS no.: 138511-98-5				physiochemical properties, evaluation of health hazards and the evaluation of environmental hazards unless otherwise stated.

It should be noted that emamectin is the name as it is placed in the list of accepted active ingredients (see Regulation (EC) No. 828/2013 and Review report SANCO / 11454/2013 rev 2 dated 16 July 2013). Regulation (EC) No. 828/2013 states that this is the benzoate variant and also in the DAR of emamectin the following is stated *'the specification of the purity of technical emamectin is benzoate: minus 950 g / kg (=min. 900 g / kg emamectin B1a benzoate and max. 70 g / kg emamectin B1b benzoate).*’ The studies included in the DAR are performed with emamectin-benzoate. Thus, the substance as referred to in the current CLH report and the placement in Annex I refer to the same substance. Emamectin benzoate is the substance as produced and is the variant used in the formulations available on the market based on this active substance.

2 PROPOSED HARMONISED CLASSIFICATION AND LABELLING

2.1 Proposed harmonised classification and labelling according to the CLP criteria

Table 6: Proposed harmonised classification and labelling for emamectin benzoate (ISO) according to the CLP criteria

	Index No	International Chemical Identification	EC No	CAS No	Classification		Labelling			Specific Conc. Limits, M-factors	Notes
					Hazard Class and Category Code(s)	Hazard statement Code(s)	Pictogram, Signal Word Code(s)	Hazard statement Code(s)	Suppl. Hazard statement Code(s)		
Current Annex VI entry	No current Annex VI entry										
Dossier submitters proposal	TBD	emamectin benzoate (ISO); (4’’R)-4’’-deoxy-4’’-(methylamino) avermectin B1 benzoate	-	155569-91-8	Acute Tox. 3 Acute Tox. 3 Acute Tox. 3 STOT RE 1 Eye Dam. 1 Aquatic Acute 1 Aquatic Chronic 1	H331 H311 H301 H372 (nervous system) H318 H400 H410	GHS05 GHS06 GHS08 GHS09 Dgr	H331 H311 H301 H372 (nervous system) H318 H410		inhalation: ATE = 0.663 mg/l dermal: ATE = 500 mg/kg bw oral: ATE = 60 mg/kg bw M=10000 M=1000	
Resulting Annex VI entry if agreed by RAC and COM	TBD	emamectin benzoate (ISO); (4’’R)-4’’-deoxy-4’’-(methylamino) avermectin B1 benzoate	-	155569-91-8	Acute Tox. 3 Acute Tox. 3 Acute Tox. 3 STOT RE 1 Eye Dam. 1 Aquatic Acute 1 Aquatic Chronic 1	H331 H311 H301 H372 (nervous system) H318 H400 H410	GHS05 GHS06 GHS08 GHS09 Dgr	H331 H311 H301 H372 (nervous system) H318 H410		inhalation: ATE = 0.663 mg/l dermal: ATE = 500 mg/kg bw oral: ATE = 60 mg/kg bw M=10000 M=1000	

Table 7: Reason for not proposing harmonised classification and status under public consultation

Hazard class	Reason for no classification	Within the scope of public consultation
Explosives	data conclusive but not sufficient for classification	Yes
Flammable gases (including chemically unstable gases)	hazard class not applicable	No
Oxidising gases	hazard class not applicable	No
Gases under pressure	hazard class not applicable	No
Flammable liquids	hazard class not applicable	No
Flammable solids	data conclusive but not sufficient for classification	Yes
Self-reactive substances	data conclusive but not sufficient for classification	yes
Pyrophoric liquids	hazard class not applicable	No
Pyrophoric solids	data conclusive but not sufficient for classification	Yes
Self-heating substances	data conclusive but not sufficient for classification	Yes
Substances which in contact with water emit flammable gases	data conclusive but not sufficient for classification	Yes
Oxidising liquids	hazard class not applicable	No
Oxidising solids	data conclusive but not sufficient for classification	Yes
Organic peroxides	hazard class not applicbale	No
Corrosive to metals	Hazard class not applicable	No
Acute toxicity via oral route	harmonised classification proposed	Yes
Acute toxicity via dermal route	harmonised classification proposed	Yes
Acute toxicity via inhalation route	harmonised classification proposed	Yes
Skin corrosion/irritation	data conclusive but not sufficient for classification	Yes
Serious eye damage/eye irritation	harmonised classification proposed	Yes
Respiratory sensitisation	data lacking	No
Skin sensitisation	data conclusive but not sufficient for classification	Yes
Germ cell mutagenicity	data conclusive but not sufficient for classification	Yes
Carcinogenicity	data conclusive but not sufficient for classification	Yes
Reproductive toxicity	data conclusive but not sufficient for classification	Yes
Specific target organ toxicity-single exposure	data conclusive but not sufficient for classification	Yes
Specific target organ toxicity-repeated exposure	harmonised classification proposed	Yes
Aspiration hazard	hazard class not applicable	No
Hazardous to the aquatic environment	harmonised classification proposed	Yes
Hazardous to the ozone layer	data lacking	No

3 HISTORY OF THE PREVIOUS CLASSIFICATION AND LABELLING

Emacetin benzoate has not previously been assessed for harmonised classification by RAC or TC C&L.

Emamectin benzoate is not registered under REACH (September 2017).

Emamectin (and not emamectin benzoate) has been evaluated within the context of Regulation EC 1107/2009. According to the data presented in the DAR (2011), the classification of emamectin is: H301 Toxic if swallowed; H311Toxic in contact with skin; H331 Toxic if inhaled; H319 Causes serious eye irritation; H372 (“Causes damage to the nervous system through prolonged or repeated exposure”); H400 Very toxic to aquatic life; H410 Very toxic to aquatic life with long lasting effects. It is noted that the DAR of emamectin includes studies with emamectin benzoate, therefore a similar classification should be required for emamectin benzoate. However, it should be noted that initially, the classification in the DAR was based on 67/548/EEC. According to 67/548/EEC R41 “risk of serious damage to eyes” was assigned, corresponding to H318 Causes serious eye damage under Regulation 1272/2008 instead of H319.

The conclusions on the peer review of the pesticide risk assessment of emamectin was published as an EFSA scientific report (2012;10(11):2955). The classification was unchanged. The DAR can be requested via: http://dar.efsa.europa.eu/dar-web/provision. EFSAs peer review is available via the EFSA website (https://www.efsa.europa.eu/en/efsajournal/pub/2955).

4 JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL

There is no requirement for justification that action is needed at Community level.

5 IDENTIFIED USES

Emamectin benzoate is used as an insecticide (larvicide). Emamectin benzoate displays strong stomach action and some contact activity on leaf feeding lepidopterous larvae. Following application insects cease feeding within hours preventing subsequent crop damage. Death occurs within 4 days maximum. Emamectin benzoate has translaminar properties and degrades rapidly on the leaf surface. Emamectin benzoate is not systemic (not translocated within plants), but forms a reservoir of active ingredient (and degradates) on or within the leaf. This provides residual activity against foliar-feeding insects.

6 DATA SOURCES

This CLH report is compiled based on the data on emamectin that was submitted and evaluated in the DAR (2011).

7 PHYSICOCHEMICAL PROPERTIES

Emamectin exists in various forms: as emamectin, as emamectin benzoate salt (MK244) and as emamectin hydrochloride (MK243). In addition various hydration forms exist for the emamectin benzoate salt. Unless stated otherwise, the following data relate to the variant emamectin benzoate in the anhydrous form.

Due to the difficulties to purify emamectin benzoate, a technical substance with a high purity was used for the determination of the physico-chemical properties. Also the technical material contains a little amount of water, the material was named emamectin benzoate hemihydrate by the notifier. The amount of water in this material was determined and found to be fixed.

No data were submitted for the emamectin B1a benzoate and emamectin B1b benzoate variants, but these data are not an EU requirement.

With regard to physical and chemical hazards, emamectin itself should normally be tested to be able to conclude on its hazard profile. Only data on the benzoate variant is available, which is the substance as manufactured (minimum purity 950g/kg, anhydrous emamectin benzoate). Considering the structure of emamectin, which is structurally strongly related to abamectin (methylamino is replaced by a hydroxyl moiety), which is not classified with regard to physical and chemical hazards, it is probable that the substance does not require classification. Although the physical and chemical properties of a salt variant can be quite different, the size and structure of the molecule indicate it will not be flammable, self-heating, oxidising or explosive. In addition, salt variants, usually are less likely to be flammable as they are generally less volatile. Extrapolation of the benzoate salt data is therefore considered acceptable.

The emamectin benzoate used was the hemihydrate unless otherwise mentioned.

Table 8: Summary of physicochemical properties

Property	**Value**	**Reference**	**Comment (e.g. measured or estimated)**
Physical state at 20°C and 101,3 kPa	solid (powder) at 25 °C	Das, 2000 (MK244/0216)	Visual inspection
Melting/freezing point	160.5°C (98.1%)	McCauley, 1992 (MK244/0047) and Geoffroy, 2007 (L07-668)	Measured (EEC A1; OECD 102; differential scanning calorimetry (DSC))
Boiling point	Decomposition before boiling	McCauley, 1992 (MK244/0047) and Geoffroy, 2007 (L07-668)	Measured (EEC A1/A2; OECD 103; differential scanning calorimetry (DSC))
Relative density	At 23.3 ± 0.1 °C: mean D^{23}_4 = 1.20 ± 0.03 (n=3; 1s)	McCauley, 1992 (MK244/0047)	Measured (EEC A3; OECD 109 pycnometer method with hexane as displacement solvent)
Vapour pressure	4 x 10-6 Pa at 21°C (97.8%)	McCauley, 1992 (MK244/0047)	Measured (EEC A4; OECD 104 gas saturation method using nitrogen gas, C18 adsorbent and HPLC-UV analysis)
Surface tension	At 20 °C (96.5%): 48.8 mN/m (90 % saturated solution)	Angly, 2000 (MK244/0223)	Measured (EEC A5; OECD 115 Wilhelmy plate method)
Water solubility	0.32 – 0.024 – 0.0001 g/L at pH 5, 7, 9	Martin, 2000 (MK244/0219)	Calculation
Partition coefficient n-octanol/water	Log Kow at 23 □C (97.8%): pH 5: 3.0 pH 7: 5.0 pH 9: 5.9 Although this was determined for/as emamectin benzoate these values are also applicable to emamectin, although the results at pH 5 may be influenced by the solubility of benzoic acid	McCauley, 1992 (MK244/0047)	Measured (EEC A8; OECD 107 shake flask method)
Flash point	Not required	-	-

Property	Value	Reference	Comment (e.g. measured or estimated)
Flammability	not highly flammable	Angly, 2000a (MK244/0220)	Measured (EEC A10)
Explosive properties	No explosive properties	Angly, 2000c (MK244/0221)	Measured (EEC A14 steel sleeve test falling hammer friction apparatus)
Self-ignition temperature	395 °C	Angly, 2000b (MK244/0222)	Measured (EEC A16)
Oxidising properties	no oxidising properties	Angly, 2000 (MK244/0223)	Measured (EEC A17)
Granulometry	No data avialable	-	-
Stability in organic solvents and identity of relevant degradation products	No data available	-	-
Dissociation constant	At 23.3-23.7 °C in water (97.8%) For emamectin benzoate two dissociation constants are found: a pKa of 7.7 for the epi-methylamino part of the emamectin ion ((R2-NH2+; conjugated acid) and a pKb of 9.8 for the benzoate ion (conjugated base), which corresponds to a pKa of 4.2 for benzoic acid. The pKa constants describe the following equations: benzoic acid + H2O ↔ benzoate- + H3O+ (pKa = 4.2) R2-NH2+ + H2O ↔ R2-NH + H3O+ (pKa = 7.7) The benzoic acid form is predominantly present at pH <2.2, the benzoate form is predominantly present at pH >6.2, while both species are present at in between values. The R2-NH2 + form of emamectin is predominantly present at pH < 5.6, the R2-NH form of emamectin is predominantly present at pH > 9.6, while both species are present at in between values.	McCauley, 1992 (MK244/0047) and Hörmann, 2000 (MK244/0200)	Measured (OECD 112)
Viscosity	Not applicable since emamectin benzoate is a solid	-	-

8 EVALUATION OF PHYSICAL HAZARDS

Emamectin exists in various forms: as emamectin, as emamectin benzoate salt (MK244) and as emamectin hydrochloride (MK243). In addition various hydration forms exist for the emamectin benzoate salt. Unless stated otherwise, the following data relate to the variant emamectin benzoate in the anhydrous form.

8.1 Explosives

Table 9: Summary table of studies on explosive properties

Method	Results	Remarks	Reference
EEC A.14	No explosive properties	Technical grade a.i. (96.5%)	Study report MK244/0221

8.1.1 Short summary and overall relevance of the information provided on explosive properties

One study in accordance with EEC A.14 was provided on explosive properties. Based on the study outcome it was concluded that emamectin benzoate is not explosive.

8.1.2 Comparison with the CLP criteria

Emamectin benzoate does not contain any chemical groups associated with explosive properties as given in section 2.1.4.2 of the CLP Guidance. Moreover, data from test method EEC A.14 indicate that emamectin benzoate has no explosive properties. Therefore no classification is required.

8.1.3 Conclusion on classification and labelling for explosive properties

No classification is proposed.

8.2 Flammable gases (including chemically unstable gases)

Hazard class not applicable (emamectin benzoate is not a gas).

8.2.1 Short summary and overall relevance of the provided information on flammable gases (including chemically unstable gases)

Not relevant.

8.2.2 Comparison with the CLP criteria

Not relevant.

8.2.3 Conclusion on classification and labelling for flammable gases

Hazard class not applicable.

8.3 Oxidising gases

Hazard class not applicable (emamectin benzoate is not a gas).

8.3.1 Short summary and overall relevance of the provided information on oxidising gases

Not relevant.

8.3.2 Comparison with the CLP criteria

Not relevant.

8.3.3 Conclusion on classification and labelling for oxidising gases

Hazard class not applicable.

8.4 Gases under pressure

Hazard class not applicable (emamectin benzoate is not a gas).

8.4.1 Short summary and overall relevance of the provided information on gases under pressure

Not relevant.

8.4.2 Comparison with the CLP criteria

Not relevant.

8.4.3 Conclusion on classification and labelling for gases under pressure

Hazard class not applicable.

8.5 Flammable liquids

Hazard class not applicable (emamectin benzoate is not a liquid).

8.5.1 Short summary and overall relevance of the provided information on flammable liquids

Not relevant.

8.5.2 Comparison with the CLP criteria

Not relevant.

8.5.3 Conclusion on classification and labelling for flammable liquids

Hazard class not applicable.

8.6 Flammable solids

Table 10: Summary table of studies on flammable solids

Method	Results	Remarks	Reference
EEC A.10	not highly flammable	Technical grade a.i. 96.5%	Angly, 2000a (MK244/0220)

8.6.1 Short summary and overall relevance of the provided information on flammable solids

One study in accordance with EEC A.10 was provided on flammable solids. Based on this study it was concluded that emamectin benzoate is not highly flammable.

8.6.2 Comparison with the CLP criteria

The test indicated that the smouldering fire does not spread out fast enough along the 200 mm powder train and therefore the test material should not be classified as a flammable solid under CLP.

8.6.3 Conclusion on classification and labelling for flammable solids

No classification is proposed.

8.7 Self-reactive substances

Data lacking.

8.7.1 Short summary and overall relevance of the provided information on self-reactive substances

Not relevant.

8.7.2 Comparison with the CLP criteria

Data on self-reactivity is lacking. However, emamectin benzoate does not contain any chemical groups associated with explosive or self-reactive properties (i.e. the presence of the N-oxides) as laid down in section 2.8.4.2 of the CLP guidance. Therefore, emamectin benzoate is not considered self-reactive.

8.7.3 Conclusion on classification and labelling for self-reactive substances

No classification proposed.

8.8 Pyrophoric liquids

Hazard class not applicable (emamectin benzoate is not a liquid).

8.8.1 Short summary and overall relevance of the provided information on pyrophoric liquids

Not relevant.

8.8.2 Comparison with the CLP criteria

Not relevant.

8.8.3 Conclusion on classification and labelling for pyrophoric liquids

Hazard class not applicable.

8.9 Pyrophoric solids

Data lacking.

8.9.1 Short summary and overall relevance of the provided information on pyrophoric solids

Data on pyrophoric solids is lacking.

8.9.2 Comparison with the CLP criteria

Data on pyrophoric solids is lacking. Emamectin benzoate has been handled in air within all studies available in the dossier and there are no reports of self-ignition (see references in all sections).

8.9.3 Conclusion on classification and labelling for pyrophoric solids

No classification is proposed.

8.10 Self-heating substances

Table 11: Summary table of studies on self-heating substances

Method	Results	Remarks	Reference
EEC A.16	Self-ignition at 395°C	Technical grade a.i. 96.5%	Angly H., 2000b (MK244/0222)

8.10.1 Short summary and overall relevance of the provided information on self-heating substances

The data available indicate that self-ignition starts at 395°C.

8.10.2 Comparison with the CLP criteria

The data available indicate that the onset temperature for self-heating of emamectin benzoate is >140 °C and that no classification is thus warranted.

8.10.3 Conclusion on classification and labelling for self-heating substances

No classification is proposed.

8.11 Substances which in contact with water emit flammable gases

Data lacking.

8.11.1 Short summary and overall relevance of the provided information on substances which in contact with water emit flammable gases

No specific data derived in accordance with the recommended test method in CLP has been provided. However, emamectin benzoate has been handled in water within many of the studies available in the dossier and there are no reports of violent reaction and emission of gas.

8.11.2 Comparison with the CLP criteria

Based on experience in handling of emamectin benzoate, it is not considered a substance which in contact with water emit flammable gases.

8.11.3 Conclusion on classification and labelling for substances which in contact with water emit flammable gases

No classification is proposed.

8.12 Oxidising liquids

Hazard class not applicable (emamectin benzoate is not a liquid).

8.12.1 Short summary and overall relevance of the provided information on oxidising liquids

Not relevant.

8.12.2 Comparison with the CLP criteria

Not relevant.

8.12.3 Conclusion on classification and labelling for oxidising liquids

Hazard class not applicable.

8.13 Oxidising solids

Table 12: Summary table of studies on oxidising solids

Method	Results	Remarks	Reference
EEC A.17	no oxidizing properties	Technical grade a.i. 96.5%	Angly, 2000 (MK244/0223)

8.13.1 Short summary and overall relevance of the provided information on oxidising solids

One study in accordance with EEC A.14 was provided on oxidising solids. Based on the study outcome it was concluded that emamectin benzoate is not oxidising.

8.13.2 Comparison with the CLP criteria

According to the CLP criteria the mean burning time in the 4:1 or 1:1 sample-to-cellulose ratio (by mass) should be determined. However, the test was perfomered with different ratios and a conclusion on the need for a classification under CLP can thus not been made. Nevertheless, because the maximum burning rate of the test item is lower than the maximum burning rate of the reference, emamectin benzoate is not considered an oxidizing substance.

8.13.3 Conclusion on classification and labelling for oxidising solids

No classification is proposed.

8.14 Organic peroxides

Hazard class not applicable (emamectin benzoate is not an organic peroxide).

8.14.1 Short summary and overall relevance of the provided information on organic peroxides

Not relevant.

8.14.2 Comparison with the CLP criteria

Not relevant.

8.14.3 Conclusion on classification and labelling for organic peroxides

Hazard class not applicable.

8.15 Corrosive to metals

Not applicable.

8.15.1 Short summary and overall relevance of the provided information on the hazard class corrosive to metals

Data lacking.

8.15.2 Comparison with the CLP criteria

Data lacking.

8.15.3 Conclusion on classification and labelling for corrosive to metals

No classification is proposed.

9 TOXICOKINETICS (ABSORPTION, METABOLISM, DISTRIBUTION AND ELIMINATION)

Emamectin was originally developed as the hydrochloride salt MK 243 (L-656,748-010V), but was subsequently changed to MK 244, the benzoate salt (L-656,748-038W), and benzoate hydrate (L-656,748-052S) because of superior storage and handling characteristics. There is no complete toxicity data set available for each salt. For the present evaluation, the available toxicity data are a composition of studies performed with abovementioned three emamectin salts, since the notifier considers these three emamectin salts as toxicologically bioequivalent. This was considered acceptable by the DS. Moreover, in the EFSA conclusion (EFSA Journal 2012;10(11):2955) the following is stated: *Toxicological studies were performed with different emamectin salts: hydrochloride salt, the benzoate salt and the benzoate hydrate salt. The three salts are considered toxicologically equivalent.*

The studies reported, with the exception of those employing molar concentrations, were performed using dose levels calculated as ‘base compound’ to account for differences in the molecular weights of the salts. The factors applied are indicated in the original study reports, and were generally 1.04 (hydrochloride), 1.14 (benzoate) and 1.16 (benzoate hydrate). In the DAR all endpoints were expressed as ‘base compounds’ and in some cases also as ‘emamectin benzoate’. However, since the the substance as referred to in the current CLH report is emamectin benzoate, all endpoints were expressed as emamectin benzoate. The ratio of emamectin to emamectin benzoate is 1.14.

The mammalian toxicity studies of emamectin benzoate were assessed in the Draft Assessment Report (2011), addenda and Proposed Decision of the Netherlands prepared in the context of the approval (Reg. (EU) No. 828/2013), under Reg. (EC) 1107/2009. Studies considered valid in the DAR (reliability score of 1 or 2) have been included in this report and were considered for classification purposes. The study summaries as presented in the DAR are included in Annex 1. All studies were carried out under GLP unless indicated otherwise. The non-GLP studies were range-finding studies or mechanistic studies. Other than the mechanistic studies all studies reported in this section were carried out in accordance with OECD guidelines. Minor deviations were noted in some cases but these did not affect the overall reliability of the studies. The deviations are included in the summaries were relevant.

Table 13: Summary table of toxicokinetic studies

Method	Results	Remarks	Reference
Absorption, distribution, metabolism and excretion Oral administration, rat (No guideline)	Absorption: Oral abs >20%. Distribution: Radiolabel was observed in all organs/tissues sampled, irrespective of dose level or regime.Highest levels observed in glandular tissues (Harderian gland, pituitary, adrenals, (para)thyroid, sublingual gland), spleen, lungs and liver. Metabolism: After oral administration of MAB1a, one metabolite, AB1a, was identified. Generally, of the % radioactivity in the organs/tissues 60-90% was present as parent compound and 5-26% as metabolite. Excretion: Plasma, 78-100% of the radioactivity was MAB1a and 5-18% AB1a, for both sexes. Plasma half lives were 27.3 h and 19.5 h for males and females, respectively. Urine, the major portion of radioactivity was associated with very polar material, and only small amounts of radioactivity (0.05%) were associated with MAB1a and AB1a. In urine of both sexes, more parent compound than metabolite was observed, but compared to males, females excreted more metabolite. Faeces, mainly parent compound MAB1a was observed (66-95%), while metabolite AB1a was present for 2-22% (both sexes). Bile, 68% (m) and 49% (f) was excreted as MAB1a at the lower dose, whereas AB1a accounted for 9% and 14% in males and females, respectively.	-	B.6.1.1, STUDY 1
Absorption, distribution, metabolism and excretion Oral and intravenous application, rat (guideline comparable to OECD 417)	Absorption: Oral abs 55% (m), 74% (f). Distribution: Organs with highest level of radioactivity were spleen, lungs, GI-tract, kidney and liver, with higher tissue residue levels in males compared to females. Compared to single low oral exposure, radioactivity levels in organs/tissue were supra-proportional after single high oral exposure. Metabolism: In faeces and tissues, one metabolite, AB1a was identified (metabolites were not profiled in urine, due to low levels of radioactivity in urine), and accounted for 0.04-2.2% of the doses on days 1, 3 and 7 after exposure. Also small amounts of polar metabolites were detected in faeces and tissues. As in faeces, also the majority of tissue radioactivity was parent MAB1a, and the only significant metabolite was identified as AB1a. The % radioactivity identified as AB1a in liver, kidney, muscle and fat was 4-23% (all treatments). Excretion: Plasma elimination half live after low oral dosing were ~28 h and ~68 h for males and ~14 h and ~76 h for females. Plasma	-	B.6.1.1, STUDY 2

Method	Results	Remarks	Reference
	elimination half lives after low i.v. dosing were ~28 h and ~72 h for males and ~18 h and ~80 h for females. Faeces (>94% within 96h), and only minor amounts (0.1-0.3%) excreted in urine or observed in tissues (0.07-1.6%).		
Disposition Intravenous, rat (Guideline OECD 417)	The major route of elimination was excretion in faeces, acounting for 36-38% and 44% of the dose after 72h in male rats and the female rat, respectively. Biliary excretion accounted for 11-17% in male rats and 6% in the female rat and urinary excretion accounted for 1.4-1.5% in male rats and 0.6% in the female rat. In the carcass, 34-37% and 37% of the dose was observed in male rats and the female rat, respectively.	-	B.6.1.1, STUDY 3

9.1 Short summary and overall relevance of the provided toxicokinetic information on the proposed classification(s)

Two rat ADME studies and one rat disposition study with emamectin B1a were available.

In the ADME studies the benzoate salt was administered. In these studies it was shown that emamectin B1a is (partly) taken up by the gastro-intestinal tract and mainly eliminated via intestinal secretion and subsequent excretion in faeces. This is consistent with the role of p-glycoprotein as a known drug efflux transporter of avermectins. After single oral administration of 0.5 mg/kg bw, bioavailability was about 55% in male rats and slightly higher (but <74%) in female rats. Plasma elimination half lives in males were about 27h to 28h and 68h for the rapid and slow elimination phase, respectively. In females, plasma elimination half lives were about 14h to 20h and 76h for these phases, respectively. Comparable elimination half lives were observed after single i.v. administration of 0.5 mg/kg bw. Compared to single low dose oral administration, single high dose (20 mg/kg bw) oral administration showed a dose-proportional increase of Cmax whereas AUC values were supra-proportional to dose. Organs with the highest levels of radioactivity were glandular tissue, spleen, lungs, GI-tract, kidney and liver, with generally higher tissue residue levels in males compared to females (after single low and high dose oral exposure as well as after repeated low dose oral exposure). Compared to single low oral exposure, radioactivity levels in organs/tissue were supra-proportional after single high oral exposure. The majority of orally administered radioactivity was excreted in faeces (90% or more over 168h), with less than 3% excreted in bile and only 0.1-0.3% in urine. Emamectin is not metabolized to a substantial extent. In faeces, bile and tissues, one metabolite was identified, AB1a, accounting for 2-22% (faeces) and 4-26% (organs) of the radioactivity.

In the rat disposition study, administration of a single intravenous dose of 0.5 mg/kg bw [14C]-emamectin B1a to rats resulted in similar overall excretion rates for male and female rats (approximately 50% over 72h) although a sex difference was observed in the route of excretion with males having more urinary (1.4-1.5%) and biliary (11-17%) excretion than the female rat (0.6% and 6% in urine and bile, respectively).

Since it cannot be excluded that the extent of metabolism can be influenced by different salts of emamectin, and no data for emamectin-HCl and emamectin-benzoate hydrate were provided, the amounts of metabolite are restricted to emamectin-benzoate exposure.

10 EVALUATION OF HEALTH HAZARDS

Acute toxicity

10.1 Acute toxicity - oral route

Table 14: Summary table of animal studies on acute oral toxicity

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels, duration of exposure	Value LD_{50}[1]	Reference
Acute toxicity, up & down procedure (Guideline OECD 425)	Rat; Sprague Dawley albino, 1-3 females /dose	Emamectin technical, benzoate salt Batch: SNA6B019 Purity: 96.2% (MK244)	Single dose, gavage 20.8, 66, 208, and 658 mg/kg bw* *Dose levels are expressed as base compound (factor 1.14)	237 mg/kg bw* conf. interval: 69-709 mg/kg bw Mortality: one rat tested at 208 mg/kg and one rat at 658 mg/kg died.	B.6.2.1.1, STUDY 1
Acute toxicity (guideline in accordance with OECD 401)	Rat Crl:CD(SD) BR strain, 5 /sex/dose	L656,748-010V003 Hydrochloride salt Purity: 96.9%* * 92.8% L-656,748 B1a and 4.1% B1b; 0.76% (w/w) propylgallate added as an antioxidant.	Single dose, gavage 44.4, 66.6, 100, 150, and 225 mg/kg bw** ** dose levels are expressed as base compound (factor 1.04).	100 mg/kg bw (m) 87 mg/kg bw (f) Mortality: Males: 1, 3, 5 and 5 males exposed to, respectively, 66.6, 100, 150 and 225 mg/kg bw died and for females the mortality observed at these dose levels was 2, 4, 5 and 5.	B.6.2.1.1, STUDY 2
Acute toxicity (Guideline in accordance with OECD 401)	Rat Crl:CD(SD) BR strain, 5 /sex/dose	MK 0244 L656,748-052S lot #5 Benzoate hydrate salt Purity: 97.8%	Single dose, gavage 32, 41.6, 54.1, 70.3, and 91.4 mg/kg bw* *dose levels are expressed as base compound (factor 1.16).	72 mg/kg bw (m) 87 mg/kg bw (f) 80 mg/kg bw (combined) Mortality: 2, 3 and 4 males dosed, respectively, 54.1, 70.3 and 91.4 mg/kg bw died. For females the mortality was 1 and 5 at doses of 70.3 and 91.4 mg/kg bw, respectively.	B.6.2.1.1, STUDY 3
Acute toxicity bioequivalence study (no guideline, but resembles OECD 401)	Rat Crl:CD(SD) BR strain, 5 females/group	MK 0244 L656,748-038W lot #2 (benzoate-methyl t-butyletherate solvate; purity 96.4 %) and L656,748-052S lot #2 (Benzoate hydrate salt; purity 99.1%)	Single dose, gavage 40, 60, 90 and 135 mg/kg bw* * all dose levels are expressed as base compound: factor 1.14 for 038W and factor 1.16 for 052S.	038W: 60 mg/kg bw (f) 052S: 66 mg/kg bw (f) Mortality: 038W: 4, 5 and 5 animals died after exposre to 60, 90 and 135 mg/kg bw, respectively. 052S: 3, 5 and 5 animals died at dose levels of 60, 90 and 135 mg/kg bw, respectively.	B.6.2.1.1, STUDY 4
Acute toxicity study (no guideline, but	Rat Crl:CD(SD) BR strain, 5 females/group	MK 0244 L656,748-038W lot #2 (benzoate-methyl	Single dose, gavage 40, 68, 116, 196,	038W: 101 mg/kg bw (f) 052S:	B.6.2.1.1, STUDY 5

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels, duration of exposure	Value LD_{50}[1]	Reference
resembles OECD 401)		t-butyletherate solvate; purity unknown) and L656,748-052S lot #1 (Benzoate hydrate salt; purity unknown)	and 334 mg/kg bw* * all dose levels are expressed as base compound: factor 1.14 for 038W and factor 1.16 for 052S.	101 mg/kg bw (f) Mortality: 038W: 1, 4, 5 and 5 animals died at dose levels of 68, 116, 196 and 334 mg/kg bw, respectively. 052S: 5 animals per group died in the highest dose groups tested (116, 196 and 334 mg/kg)	
Acute toxicity (guideline in accordance with OECD 401)	Mouse Crl:CF-1 BR strain, 5 /sex/dose	L656,748-010V003 Hydrochloride salt Purity: 96.9% * * 92.8% L-656,748 B1a and 4.1% B1b; 0.76% (w/w) propylgallate added as an antioxidant.	Single dose, gavage 20, 30, 45, 67.5 and 101.2 mg/kg bw ** **dose levels are expressed as base compound (factor 1.04)	25 mg/kg bw (m) 35 mg/kg bw (f) Mortality: 2, 4, 5, 5 and 5 animals died at dose levels of 20, 30, 45, 67.5 and 101.2 mg/kg bw, respectively. At the same dose levels, 3, 0, 4, 4 and 5 females died.	B.6.2.1.1, STUDY 6
Acute toxicity study (no guideline, but resembles OECD 401)	Mice Crl:CD-1(ICR) BR strain, 5 females/group	MK 0244 L656,748-038W lot #2 (benzoate-methyl t-butyletherate solvate; purity unknown) and L656,748-052S lot #1 (Benzoate hydrate salt; purity unknown)	Single dose, gavage 5, 10, 20, 40, and 80 mg/kg bw* (1st study) 80, 144, 259, and 466 mg/kg bw * (2nd study) * all dose levels are expressed as base compound: factor 1.14 for 038W and factor 1.16 for 052S	038W: 137 mg/kg bw (f) 052S: 122 mg/kg bw (f) Mortality: 038W: 4, 5 and 5 animals died after exposure to 144, 259 and 466 mg/kg bw, respectively. 052S: 1, 4, 5 and 5 animals died exposed to 80, 144, 259 and 466 mg/kg bw, respectively.	B.6.2.1.1, STUDY 7
Acute toxicity study (no guideline, but resembles OECD 401)	Mice Crl:CD-1(ICR) BR strain, 5/sex/dose, except high dose group 5 f only	MK 0244 L656,748-052S lot #2 Benzoate hydrate salt; Purity: 97.6%	Single dose, gavage 70, 120, 192, and 307 mg/kg bw * * dose levels are expressed as base compound (factor 1.16)	153 mg/kg bw (m) 178 mg/kg bw (f) 165 mg/kg bw (combined) Mortality: 1, 0 and 5 animals died at doselevels of 70, 120 and 192 mg/kg bw. At 192 and 307 mg/kg bw 4 and 5 females, repectively, died.	B.6.2.1.1, STUDY 8
Acute toxicity study	Mice Crl:CD-1(ICR) BR strain, 5 females/group	MK 0244 L656,748-038W lot #2	Single dose, gavage	038W: 188 mg/kg bw (f)	B.6.2.1.1, STUDY 9

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels, duration of exposure	Value LD_{50}[1]	Reference
(no guideline, but resembles OECD 401)		(benzoate-methyl t-butyletherate solvate; purity 96.4%) or L656,748-052S lot #2 (Benzoate hydrate salt; purity 99.1%)	60, 90, 135, and 202 mg/kg bw* * all dose levels are expressed as base compound: factor 1.14 for 038W and factor 1.16 for 052S	052S: 161 mg/kg bw (f) Mortality: 038W: 5 animals died at the highest dose tested. 052S: 2 and 5 animals died at levels of 135 and 202 mg/kg/ bw.	
Acute neurotoxicity (no guideline)	Rat Crl:CD(SD) Br strain, 10 /sex/dose	MK-0243: L-656,748-010V003 hydrochloride salt, purity: 96.9%	Single dose; oral, 0, 27.4, 54.8, and 82.2 mg/kg bw* *dose levels are expressed as base compound (factor 1.04).	76 mg/kg bw (m) 80 mg/kg bw (f) Mortality: 2 and 8 males died at 54.8 and 82.2 mg/kg bw, respectively. At the same dose levels, 2 and 7 females, respectively, died.	B.6.7.1, STUDY 1
Acute neurotoxicity (no guideline)	Rat Crl:CD(SD) Br strain, 10 /sex/dose	MK-0243: L-656,748-038W002 benzoate salt Purity: 94.2%	Single dose; oral, 0, 0.5, 2.5, 5.0, 10, 25 mg/kg bw* *dose levels are expressed as base compound (factor 1.14).	> 29 mg/kg bw Mortality: at 0.5 and 2.5 mg/kg bw one female died. No animals died at the higher dose levels tested.	B.6.7.1, STUDY 2

[1] *Studies were performed expressing the active substance as base compound in the original study reports, but all LD_{50} values were recalculated and expressed as emamectin benzoate (ratio base compound to emamectin benzoate 1.14)*

Table 15: Summary table of human data on acute oral toxicity

No data available.

Table 16: Summary table of other studies relevant for acute oral toxicity

No data available.

10.1.1 Short summary and overall relevance of the provided information on acute oral toxicity

Emamectin benzoate was tested for acute toxicity as three different salts (benzoate, benzoate hydrate and hydrochloride). The salts were found to be of similar acute toxicity. Emamectin benzoate is toxic to mouse and rat by oral administration, with the rat more sensitive than the CD-1 mouse, but less sensitive than the CF-1 mouse. The sensitivity of the CF-1 mouse to emamectin benzoate was also observed in the neurotoxicity tests. Characteristic signs of acute emamectin benzoate toxicity in mice and rats are tremors, ataxia, brady-pnoea and decreased activity.

The LD50 values for the rat ranged between 72 and 100 mg/kg bw for male rats and between 60 and 101 mg/kg bw for female rats, although in a more recent study, performed according to the up & down procedure, the LD50 in female rats was 237 mg/kg bw.

10.1.2 Comparison with the CLP criteria

The rat LD_{50} of emamectin benzoate was found to range between 60 and 237 mg/kg bw/day (EFSA Journal 2012;10(11):2955). It should be noted that in the summary of the notifier it is mentioned that "*All studies prior to 2006 were conducted on test substance sourced from Merck. Additional studies were conducted in 2006 on test substance sourced from Nantong in China.*" (remark: these additional studies are performed with 'benzoate salt'). However, the DS consideres that it is highly unlikely that differences in the impurity profile are the cause of the differences in acute toxicity. Therefore, all acute toxicity studies were included for classification. According to Regulation No. (EC) 1272/2008 a substance should be classified as acute toxic category 3 if the LD50 is within the limits 50 < ATE ≤ 300 mg/kg bw.

The lowest rat LD50 of 60 mg/kg bw is suggested as ATE for acute oral toxicity. It should be noted that also a mice study is available with a LD50 < 50 mg/kg bw which would result in classification in a different category. However, P-glycoprotein deficient animals including CF-1 mouse are more sensitive to emamectin benzoate, studies performed with non-functional P-glycoprotein are considered not relevant for human and the studies performed with rats are therefore considerd to be more representative for classification.

10.1.3 Conclusion on classification and labelling for acute oral toxicity

According to the CLP Regulation (Annex I, Part 3, Table 3.1.1 and 3.1.2 Acute toxicity: Category 3, 50 < ATE ≤ 300 mg/kg bw, LD50/ATE = 60 mg/kg bw) classification with acute oral toxicity category 3, toxic if swallowed (H301) is proposed for emamectin benzoate.

It is proposed to assign an ATE of 60 mg/kg bw for acute oral toxicity.

10.2 Acute toxicity - dermal route

Table 17: Summary table of animal studies on acute dermal toxicity

Method, guideline, deviations if any	**Species, strain, sex, no/group**	**Test substance,**	**Dose levels duration of exposure**	**Value LD_{50}[1]**	**Reference**
Acute toxicity (guideline OECD 402)	Rat; Sprague Dawley albino, 5 /sex/dose	Emamectin technical, benzoate salt Batch: SNA6B019 Purity: 96.2% (MK244)	Single dose, 24 h 877 mg/kg bw (f), and/or 1754 mg/kg bw (m/f)* * Dose levels are expressed as base compound (factor 1.14) (when expressed as emamectin salt, the dose levels are 1000 and 2000 mg/kg bw)	>2000 mg/kg bw (m/f)* Mortality: Two females of the high dose group were euthanized for humane reasons on day 2. They showed mouth discharge and were in moribund condition.	B.6.2.1.2, STUDY 1
Acute toxicity study (no guideline)	Rat Crl:CD(SD) BR strain, 5 /sex/dose	MK 0244 L656,748-052S lot #5 (Benzoate hydrate salt; purity 96.4%)	Single dose for 24h 2000 mg/kg bw* *dose levels are expressed as base compound (factor 1.16)	> 2280 mg/kg bw Mortality: One male rat died on day 7. This death was considered to be due to oral ingestion since the rat was found	B.6.2.1.2, STUDY 2

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels duration of exposure	Value LD_{50}[1]	Reference
				without the occlusive dressing on the morning of day 2.	
Acute toxicity (guideline OECD 402)	Rat; CRL:(WI)BR Wistar, 5 /sex/dose	Emamectin technical, benzoate salt Batch: SNA6A015 Purity: 96.6% (MK244G)	Single dose, 24 h 439 mg/kg bw, 877 mg/kg bw, and 1754 mg/kg bw* *Dose levels are expressed as base compound (factor 1.14) (when expressed as emamectin salt, the dose levels are 500, 1000 and 2000 mg/kg bw)	Between 500 and 1000 mg/kg bw for males and 1892 mg/kg bw for females Mortality: 1, 3 and 1 males died at dose levels of 439, 877 and 1754 mg/kg bw, respectively. 3 females died at the highest dose level tested.	B.6.2.1.2, STUDY 3
Acute neurotoxicity	Rabbit NZW, 5 f/group	MK-0243: L-656,748-038W, Lot 2 Purity: 94.2%	Single dose; dermal (24h), 500, 1000, and 2000 mg/kg bw* * no factor was mentioned in the study report; Other studies with this batch used a conversion factor of 1.14, so it may be assumed that this was used for this study.	> 2000 mg/kg bw* Mortality: none.	B.6.7.1, STUDY 3

[1] Studies were performed expressing the active substance as base compound in the original study reports, but all LD_{50} values were recalculated and expressed as emamectin benzoate (ratio base compound to emamectin benzoate 1.14)

Table 18: Summary table of human data on acute dermal toxicity

No data available.

Table 19: Summary table of other studies relevant for acute dermal toxicity

No data available.

10.2.1 Short summary and overall relevance of the provided information on acute dermal toxicity

Emamectin benzoate was tested for acute dermal toxicity in three different studies performed with rat.

One study was performed with benzoate hydrate salt (B.6.2.1.2, STUDY 2). This study is not a guideline study although it resembles OECD 402. The following deviations from OECD guideline 402 were defined as follows: the test substance was applied as neat compound on the test site which

was moistened with 0.5 mL saline and covered with an occluded dressing. Collars were placed on the animals during the whole study. Based on this study, the dermal LD50 for emamectin benzoate for male and female rats is > 2280 mg/kg bw.

Two studies are available in which the new source of emamectin benzoate was used. One guideline study performed in accordance with OECD 402 is available in which male and female Sprague Dawley rats were exposed to benzoate hydrate salt via the skin (B.6.2.1.2, STUDY 1). Based on this study, the dermal LD50 for emamectin benzoate for male and female rats is > 2000 mg/kg bw.

Another study performed with the new source and resembles OECD guideline 402 (B.6.2.1.2, STUDY 3). The study deviates from the protocol since the test substance, a white powder, was applied as neat compound on the test site while a solid compound should be moistened sufficiently to ensure good contact with the skin. Despite this deviation, the study is considered acceptable since the test substance has apparently been in good contact with the skin based on the study outcome. Based on the results of this study, the acute dermal LD50 for emamectin benzoate salt is between 500 and 1000 mg/kg bw for males and 1892 mg/kg bw for females, expressed as emamectin benzoate.

Based on an acute neurotoxicity study, the LD50 in rabbits was found to be > 2000 mg/kg bw (B.6.7.1, STUDY 3).

10.2.2 Comparison with the CLP criteria

For male rat, the lowest LD50 was found to range between 500 and 1000 mg/kg bw. According to Regulation No. (EC) 1272/2008 a substance should be classified as acute toxic category 3 if the LD50 is within the limits 200 < ATE ≤1000. For rabbit the the LD50 was found to be > 2000 mg/kg bw and is thus outside the limits for classification as acute toxic category 3 and classification was therefore done based on the results obstained in the rat study representing lower LD50 values. Emamectin benzoate should thus be classified as harmful for acute dermal toxicity.

The LD_{50} of 500 mg/kg bw is suggested as ATE for acute dermal toxicity. It is noted that the lowest LD_{50} is a range (i.e. 500-1000 mg/kg bw, male rat) and not an exact number. This introduces uncertainty when establishing the ATE. It is also noted that the converted acute toxicity estimate corresponding to this experimentally obtained LD_{50}-range would be 300 mg/kg bw (according to CLP Regulation, Annex I, Part 3, Table 3.1.2) which is even lower than the lower level of the LD_{50}-range. Therefore, the Dossier Submitter considers the value of 500 mg/kg bw more appropriate as ATE for acute dermal toxicity.

10.2.3 Conclusion on classification and labelling for acute dermal toxicity

According to the CLP Regulation (Annex I, Part 3, Table 3.1.1 and 3.1.2 Acute toxicity: Category 3, 200 < ATE ≤ 1000 mg/kg bw, LD50/ATE = 500 mg/kg bw) classification with acute dermal toxicity category 3, toxic in contact with skin (H311) is proposed for emamectin benzoate.

It is proposed to assign an ATE of 500 mg/kg bw for acute dermal toxicity.

10.3 Acute toxicity - inhalation route

Table 20: Summary table of animal studies on acute inhalation toxicity

Method, guideline, deviations if any	**Species, strain, sex, no/group**	**Test substance, , form and particle size (MMAD)**	**Dose levels, duration of exposure**	**Value LC_{50}**	**Reference**
Acute toxicity study (guideline OECD	Rat HsdRccHan:WIST strain, 5 m and/or f/group*	MK 0244 G SNA6B019/milled (emamactin benzoate	4 h; nose only 0, 0.239, 0.506, 1.049, 1.981	M: between 1.049 and 1.981 mg/L F: 0.663 mg/L	B.6.2.1.3, STUDY 1

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance, , form and particle size (MMAD)	Dose levels, duration of exposure	Value LC_{50}	Reference
403)	*in the low dose group (0.25 mg/L) only females were tested; in the mid dose group (1.0 mg/L) only males were tested.	technical; purity 96.2%) Inhalation (aerosol) MMAD: 2.7, 3.7, 3.8, and 2.9 resp.; GSD: 1.9, 2.6, 1.7, and 1.6 resp.	mg/L (achieved conc.)** ** target dose levels were 0.25, 0.5, 1.0, and 2.0 mg/L	Mortality: 4 males died after exposure to 1.981 mg/L. 3 and 5 females died at dose levels of 0.506 and 1.981 mg/L, respectively.	
Acute toxicity study (no guidance, but resembles OECD 403)	Rat Crl:CD(SD) BR strain, 5 /sex/dose	MK 0244 L656,748-052S 006 (Benzoate salt; purity 96.9%) Inhalation (aerosol) MMAD:1.2, 4.1, 3.7, and 4.3 resp.; GSD: 3.0, 2.3, 2.0, and 2.1 resp.	4 h; nose only 0, 0.24, 0.44, 2.12, 4.44 mg/L (achieved conc.)* *the achieved concentrations (and also target and nominal concentrations) are expressed as benzoate salt	Between 2.12 and 4.44 mg/L (m/f) Mortality: 3 and 5 males died at dose levels of 2.12 and 4.44 mg/L, respectively. At dose levels of 2.12 and 4.44 mg/L 1 and 3 females died, respectively.	B.6.2.1.3, STUDY 2
Acute toxicity study (no guidance, but resembles OECD 403)	Rat Crl:CD(SD) BR strain, 5/ sex/ dose	MK 0244 L656,748-052S 006 (Benzoate salt; purity 96.9%) MMAD: 3.2, 2.9, and 3.5 resp.; GSD: 2.0, 2.2, and 2.0 resp.	4 h; nose only 0, 0.01, 0.05, 0.1 mg/L (achieved conc.)* * the achieved concentrations were equivalent to the target concentrations. The nominal concentrations were 0, 0.02, 0.08, and 0.17 mg/L. The concentrations are expressed as benzoate salt.	LC50 not determined. The study is considered acceptable as follow-up for evaluation of the effects in the nerve tissue. Mortality: none.	B.6.2.1.3, STUDY 3

Table 21: Summary table of human data on acute inhalation toxicity

No data available.

Table 22: Summary table of other studies relevant for acute inhalation toxicity

No data available.

10.3.1 Short summary and overall relevance of the provided information on acute inhalation toxicity

One study is available in which the new source of emamectin benzoate was tested (B.6.2.1.3, STUDY 1). The study design was based on OECD 403, but also on the 'up and down' procedure. The first exposure concentration tested was 2.0 mg/L and was based on previous studies (see below). Subsequent exposure concentrations were selected in a stepwise manner. The test article is a white powder which was milled by the sponsor to reduce the particle size to a respirable range. The acute 4-hour inhalation LC50 for emamectin benzoate salt in rats could be calculated for females and was 0.663 mg/L, expressed as a base compound. The LC50 for males is between 1.049 and 1.981 mg/L when expressed as emamectin benzoate.

In the older inhalation studies special attention has been given to possible effects on brain, sciatic nerve and the spinal cord. In a study resembling OECD 403 the acute 4-hour inhalation LC50 for emamectin benzoate hydrate in rats (m/f combined) is between 2.12 and 4.44 mg/L when expressed as emamectin benzoate (B.6.2.1.3, STUDY 2). Post mortem findings of neuronal vacuolar degeneration in the brain and spinal cord and/or sciatic nerve degeneration were evident in some animals from all groups, and thus a no effect level for these changes could not be established. A follow up study was performed for evaluation of the effects in the nerve tissue. A NOAEL for neuronal degeneration in brain and nerve degeneration in sciatic nerve and/or spinal cord was established at 0.1 mg/L.

10.3.2 Comparison with the CLP criteria

The lowest LC_{50} was 0.663 mg/L in females and varied between 1.049 and 1.981 mg/L in males when expressed as a base compound. According to Regulation No. (EC) 1272/2008 a substance should be classified as acute toxic category 3 if the LD50 is within the limits 0.5 < ATE ≤1.0 mg/L (dusts and mists). Emamectin benzoate should thus be classified as acute inhalation toxicity, catergory 3.

The LC_{50} of 0.663 mg/l is suggested as ATE for acute inhalation toxicity

10.3.3 Conclusion on classification and labelling for acute inhalation toxicity

According to the CLP Regulation (Annex I, Part 3, Table 3.1.1 and 3.1.2 Acute toxicity: Category 3, 0.5 < ATE ≤ 1.0 mg/L, LC50/ATE = 0.663 mg/l) classification with acute inhalation toxicity category 3, toxic if inhaled (H331) is proposed for emamectin benzoate.

It is proposed to assign an ATE of 0.663 mg/l for acute inhalation toxicity.

10.4 Skin corrosion/irritation

Table 23: Summary table of animal studies on skin corrosion/irritation

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels duration of exposure	Results -Observations and time point of onset -Mean scores/animal -Reversibility	Reference
skin irritation study (Guideline OECD 404)	Rabbit NZW, 1 m; 2f	Emamectin technical, benzoate salt Batch: SNA6B019 Purity: 96.2% (MK244)	4 h semi-occlusive, 500 mg	Observations made at 1, 24, 48 and 72h Mean scores at 24h, 48h and 72h: Erythema: 0.7; 0.3; 0 Oedema: 0 Reversibility: all effects were reversed at 72h	B.6.2.2.1, STUDY 1
skin	Rabbit	Emamectin	4 h semi-	Observations made at 30 to 60 minutes and	B.6.2.2.1,

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels duration of exposure	Results -Observations and time point of onset -Mean scores/animal -Reversibility	Reference
irritation study (in accordance with OECD 404)	NZW, 3 /sex	technical, benzoate salt Batch: SNA6B019 Purity: 96.2% (MK244)	occlusive, 500 mg	24, 48, 72, 144 and 168 hours after patch removal Mean scores: Erythema: 0 Oedema: 0 Reversibility: not applicable	STUDY 2

Table 24: Summary table of human data on skin corrosion/irritation

No data available.

Table 25: Summary table of other studies relevant for skin corrosion/irritation

No data available.

10.4.1 Short summary and overall relevance of the provided information on skin corrosion/irritation

Emamectin benzoate causes only slight erythema when applied to the skin, which is reversed at 72 h (B.6.2.2.1, STUDY 1 and B.6.2.2.1, STUDY 2).

10.4.2 Comparison with the CLP criteria

Emamection was found to cause slight erythema when applied to the skin. According to Regulation No. (EC) 1272/2008 a substance should be classified as skin irritant if:

(1) Mean score of ≥ 2,3 - ≤ 4,0 for erythema/eschar or for oedema in at least 2 of 3 tested animals from gradings at 24, 48 and 72 hours after patch removal or, if reactions are delayed, from grades on 3 consecutive days after the onset of skin reactions; or

(2) Inflammation that persists to the end of the observation period normally 14 days in at least 2 animals, particularly taking into account alopecia (limited area), hyperkeratosis, hyperplasia, and scaling; or

(3) In some cases where there is pronounced variability of response among animals, with very definite positive effects related to chemical exposure in a single animal but less than the criteria above.

Emamectin benzoate does not fulfil the criteria for skin irritation as the scores for erythema and oedema were below 2.3 in all animals at all time points and no signs of inflammation were observed.

10.4.3 Conclusion on classification and labelling for skin corrosion/irritation

No classification is proposed for emamectin benzoate.

10.5 Serious eye damage/eye irritation

Table 26: Summary table of animal studies on serious eye damage/eye irritation

Method, guideline,	Species, strain,	Test substance,	Dose levels duration of	Results - Observations and time point of onset	Reference

deviations if any	**sex, no/group**		**exposure**	**- Mean scores/animal - Reversibility**	
eye irritation study (guideline OECD 405)	Rabbit NZW, 2 m; 1f	Emamectin technical, benzoate salt Batch: SNA6B019 Purity: 96.2% (MK244)	Single instillation in conjunctival sac, 0.1 mL (weighing 60 mg)	Observations at 1h, 24h, 48h, 72h, day 4 and day 7 Mean scores per animal at 24h, 48h and 72h: Corneal opacity: 0.0; 0.3; 1.0 Corneal area: 4.0; 3.0; 1.3 Iris: 0.7; 1.0; 1.0 Conj. Redness: 1.3; 2.0; 2.0 Conj. Chemosis: 0.3; 1.3; 1.3 Conj. Discharge: 1.0; 1.0; 2.0 Reversibility: reversible within the 7 day observation period	B.6.2.2.2, STUDY 1
eye irritation study (in accordance with OECD 405)	Rabbit NZW, 3 /sex	MK 0243 L656,748 (4”-deoxy-4-epi-methylamino avermectine B1 benzoate); purity 96.2%)* *Purity of substance: 91.1% B1a and 5.1% B1b.	Single instillation in conjunctival sac, 0.1 mL (weighing 28 mg)	Observations at 1h, 24h, 48h, 72h and daily up to day 14 Mean scores: Corneal opacity: 0.0; -; -; -; 0; 0.3 Iris: 1.0; -; -; -; 1.0; 0.3 Conj. Redness: 3.0; 3.0; 3.0; -; 3.0; 1.3 Conj. Chemosis: 2.7; 4.0; 4.0; 4.0; 2.3; 0.3 Conj. Discharge: 3.0; 3.0; 2.3; 3.0; 2.3; 0.0 - indicates that the sign could not be read at one or more time points due to chemosis. Reversibility: irritation was not reversible within the 14 day observation period	B.6.2.2.2, STUDY 2

Table 27: Summary table of human data on serious eye damage/eye irritation

No data available.

Table 28: Summary table of other studies relevant for serious eye damage/eye irritation

No data available.

10.5.1 Short summary and overall relevance of the provided information on serious eye damage/eye irritation

Two eye irritation studies are available. One study was performed using the new source of emamectin benzoate (emamectin benzoate salt). The study is in accordance with OECD 405 (B.6.2.2.2, STUDY 1). Prior to each instillation, two drops of an ocular anaesthetic (tetracaine hydrochloride ophthalmic solution 0.5%) were placed into both the treated and control eye of each animal. Then the test substance was instilled in the test eye. Ocular changes were graded according to Draize. The eyes were examined up to 7 days after instillation. Signs of irritation were observed in the eyes of the rabbits. The mean scores for conjunctival redness and chemosis over the period 24 – 72 h are not greater than 2.5 and 2.0, respectively. Damage to the iris with score 1 was observed in all 3 animals up to 48 hours and still seen in 2 animals at 72 h. Mean scores for conjunctival redness was 2 in 2 out of 3 animals. All signs of irritation (including effects on iris and cornea) were reversible within the 7 day observation period of the study.

In the other available study, performed in accordance with OECD 405, the mean scores for conjunctival redness and chemosis over the period 24 – 72 h are greater than 2.5 and 2.0, respectively (B.6.2.2.2, STUDY 2). Furthermore, animals were sacrificed after 72 hours because of severe signs of irritation since chemosis and redness became severe, and white mucoid discharge was observed. The chemosis observed in these animals hampered reading of other effects on the eyes including opacity. Moreover, the study authors noted a complete lack of blink response in one animal observed at 24 and 48 hours. The remaining animals also showed severe redness but moderate chemosis and two animals showed a white mucoid discharge. Irritation was not reversible for the remaining animals within the 14 day observation period of the study. Unfortunately, the study stopped at 14 days and it is therefore unknown whether the effects are reversed after 21 days.

10.5.2 Comparison with the CLP criteria

According to Regulation No. (EC) 1272/2008 a single hazard category (Category 1) is adopted for substances that have potential to seriously damage the eyes. For such substances the following criteria apply:

A substance that produces:

(a) in at least one animal effects on the cornea, iris or conjunctiva that are not expected to reverse or have not fully reversed within an observation period of normally 21 days; and/or

(b) in at least 2 of 3 tested animals, a positive response of:

(i) corneal opacity ≥ 3 and/or

(ii) iritis > 1,5

calculated as the mean scores following grading at 24, 48 and 72 hours after

installation of the test material.

According to Regulation No. (EC) 1272/2008 a single hazard category (Category 2) is adopted for substances that have potential for eye irritation. For such substances the following criteria apply:

Substances that produce in at least in 2 of 3 tested animals, a positive response of:

(a) corneal opacity ≥ 1 and/or

(b) iritis ≥ 1, and/or

(c) conjunctival redness ≥ 2 and/or

(d) conjunctival oedema (chemosis) ≥ 2

calculated as the mean scores following grading at 24, 48 and 72 hours after installation of the test material, and which fully reverses within an observation period of 21 days

Eye irritation studies show that emamectin benzoate is an irritating compound. In fact, emamectin benzoate was found to cause effects to the eyes (iritis, conjunctival redness, conjunctival chemosis and conjunctival discharge) with Draize scores falling into the criteria of Cat 2. However, the effects are not reversible within the 14-day period of observation. Although not examined in the second study, considering the severity of the effects (that lead to euthanasia of 3 animals), it is not expected that they will have reversed at 21 days and therefore emamectin benzoate should be classified as severely irritating.

10.5.3 Conclusion on classification and labelling for serious eye damage/eye irritation

Classification with eye irritation category 1, causes serious eye damage (H318) is proposed for emamectin benzoate.

10.6 Respiratory sensitisation

No data available.

10.6.1 Short summary and overall relevance of the provided information on respiratory sensitisation

No data available.

10.6.2 Comparison with the CLP criteria

No data available.

10.6.3 Conclusion on classification and labelling for respiratory sensitisation

No classification is proposed due to the lack of data.

10.7 Skin sensitisation

Table 29: Summary table of animal studies on skin sensitisation

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels duration of exposure	Results	Reference
Skin sensitization study (GPMT) (in accordance with OECD 406)	Guinea pig (Hartley albino), 11 f (test group); 10 f (control)	MK 0244 L656,748-038W 002 (benzoate-methyl t-butyletherate solvate) purity > 95%	5% intradermal injection, 7.5% topical induction, 1.25% challenge test concentrations were based on the results from screening tests: 5% intradermal was well tolerated but a 15% epicutaneous application evoked tremors in 2 out of 5 animals. In a 4-day exploratory irritation study, a concentration of 1.25% was not irritating, whereas a concentration of 2.5% was slightly irritating. A factor 1.14 was used in preparing the doses.	Not sensitizing	B.6.2.2.3, STUDY 1
Skin sensitization study (LLNA) (OECD 429)	Mouse CBA/Ca/Ola/Hsd strain, 4 f/group	Emamectin benzoate technical Batch: SNA6B019 Purity: 96.2% (MK244G)	topical on dorsal surface of the ears, 0.5, 1, and 2.5% w/v MK244G test concentrations were based on the results from screening tests in which animals were exposed to 3 repeat topical exposures of: 0.1, 0.5, 1, 2.5, and 5% w/v MK244G. Only the 5% w/v dose group showed signs of systemic toxicity.	Not sensitizing	B.6.2.2.3, STUDY 2

Table 30: Summary table of human data on skin sensitisation

No data available.

Table 31: Summary table of other studies relevant for skin sensitisation

No data available.

10.7.1 Short summary and overall relevance of the provided information on skin sensitisation

Two skin sensitisation studies were performed. In the guinea pig maximisation test (GPMT), performed in accordance with OECD 406, the old source of emamectin benzoate was tested (B.6.2.2.3, STUDY 1). Only after 48 h, slight erythema was observed in 2 test animals, but also in 3 control animals. Following re-challenge in these animals, only after 48 h slight erythema was observed in 1 test animal and in 1 control animal. No reactions were observed at the vehicle sites of any of the animals.

During histopathological examination, most animals of the control and treated group showed (very) slight acanthosis. This lesion was considered to be the result of non specific irritation due to occlusive application of the vehicle petrolatum.

Under the conditions of the GPMT test, emamectin benzoate was not sensitizing.

In the local lymph note assay (LLNA), performed according to OECD 429, animals were exposed to 25µl of a 0.5, 1 or 2.5 % w/v preparation of the test substance applied to the dorsal surface of each ear (B.6.2.2.3, STUDY 2). The procedure was repeated daily for 3 consecutive days. In this assay the new source of emamectin benzoate was tested.

There was no effect on body weight. The stimulation index (SI) was 1.3, 1.1 and 2.1 for the low, mid, and high dose groups, respectively. Since the stimulation index was below 3, the test is considered negative. The positive control group was positive (SI of 7.2) at the highest test concentration only.

Under the conditions of the LLNA test, emamectin benzoate technical is considered not to be a skin sensitiser.

10.7.2 Comparison with the CLP criteria

According to Regulation No. (EC) 1272/2008 a single hazard category (Category 1) is adopted for substances that have potential for skin sensitisation. For such substances the following criteria apply:

Category 1:	*Substances shall be classified as skin sensitisers (Category 1) where data are not sufficient for sub-categorisation in accordance with the following criteria: (a) if there is evidence in humans that the substance can lead to sensitisation by skin contact in a substantial number of persons; or (b) if there are positive results from an appropriate animal test (see specific criteria in paragraph 3.4.2.2.4.1).*
Sub-category 1A:	*Substances showing a high frequency of occurrence in humans and/or a high potency in animals can be presumed to have the potential to produce significant sensitisation in humans. Severity of reaction may also be considered.*
Sub-category 1B:	*Substances showing a low to moderate frequency of occurrence in humans and/or a low to moderate potency in animals can be presumed to have the potential to produce sensitisation in humans. Severity of reaction may also be considered.*

For emamectin benzoate no human data and comparison with CLP critiria can therefore only be done based on animal data. The following CLP criteria apply:

Animal test results for sub-category 1A:

Assay Criteria Local lymph node assay	*EC3 value ≤ 2 % Guinea pig maximisation test ≥ 30 % responding at ≤ 0,1 % intradermal induction dose or ≥ 60 % responding at > 0,1 % to ≤ 1 % intradermal induction dose*
Buehler assay	*≥ 15 % responding at ≤ 0,2 % topical induction dose or ≥ 60 % responding at > 0,2 % to ≤ 20 % topical induction dose*

Animal test results for sub-category 1B

Assay Criteria Local lymph node assay	*EC3 value > 2 % Guinea pig maximisation test ≥ 30 % to < 60 % responding at > 0,1 % to ≤ 1 % intradermal induction dose or ≥ 30 % responding at > 1 % intradermal induction dose*
Buehler assay	*≥ 15 % to < 60 % responding at > 0,2 % to ≤ 20 % topical induction dose or ≥ 15 % responding at > 20 % topical induction dose*

Under the conditions of the GPMT test, emamectin benzoate was not sensitizing. Moreover, based on the LLNA assay, a stimulation index below 3 was obtained indicating that the test is considered negative. Under the conditions of the LLNA test, emamectin benzoate technical is considered not to be a skin sensitiser and classification is not required.

10.7.3 Conclusion on classification and labelling for skin sensitisation

Emamectin benzoate does not need to be classified for skin sensitization.

10.8 Germ cell mutagenicity

Table 32: Summary table of mutagenicity/genotoxicity tests in vitro

Method, guideline, deviations if any	**Test substance,**	**Relevant information about the study including rationale for dose selection (as applicable)**	**Observations**	**Reference**
Ames test Point mutation, gene mutation (Study design resembles OECD 471)	emamectin HCl salt (MK-0243), purity 92.8%	Organism/ strain: *S. typh.* (TA 97a, TA 98, TA 100, TA 1535) *E. coli* (WP2, WP2 uvrA, WP2 UvrA pKK101) Concentration tested: A high dose of 953 µg/plate was used in this assay since previous experience with emamectin compounds indicated that this is the approximate level where bacterial toxicity was seen.	Results without activation: - Results with activation: -	B.6.4.1, STUDY 1

Method, guideline, deviations if any	Test substance,	Relevant information about the study including rationale for dose selection (as applicable)	Observations	Reference
		Positive controls: salmonella tester strains and *E. coli* strains WP2 uvrA and WP2 uvrA pKM101: 2-aminoanthracene *E. coli* WP2: hydrazine sulfate.		
Chromosome aberrations (Test design resembles OECD 476)	MK-0244, batch L-656,748-010V003, HCl salt, purity 96.9%	Organism/ strain: Chines hamster V79 Concentration tested: 0, 0.005- 0.06 mM with S9, 0, 0.001-0.01 mM without S9 Positive controls: 3-methylcholanthrene for incubations with metabolic activation and methylnitrosourea for incubations without metabolic activation Exposure duration: 3h	Results without activation: - Results with activation: -	B.6.4.1, STUDY 2
Chromosome aberrations (Study design resembles OECD 473)	MK-0244, batch L-656,748-052S002, purity avermectine B1a 92.5%, purity avermectine B1b 5.3%.	Organism/ strain: Chines hamster oveary cells (CHO-WBL) Concentration tested: 0, 1, 2, 3, 4, 5, 6, 7, 8, 9 and 10 µM Positive controls: cyclophosphamide and mitomycin for incubations with and without metabolic activation, respectively Exposure duration: 3h with subsequent reincubation for 17h.	Results without activation: - Results with activation: -	B.6.4.1, STUDY 3
DNA strand breaks (no official guideline)	MK-0243, batch L-656,748-010V003, HCl salt, purity 92.8%, Note: In another study (STUDY 1) by the same study author the same batch was used. In that study purity was reported as: avermectine B1a 92.8%, avermectine B1b 4.1%	Organism/ strain: rat hepatocytes in an in vitro alkaline elution/rat hepatocyte assay Concentration tested: 0.003- 0.02 mM Positive control: Aflatoxin B1 Exposure duration: 3h.	No DNA strand breaks	B.6.4.1, STUDY 4

Table 33: Summary table of mutagenicity/genotoxicity tests in mammalian somatic or germ cells in vivo

Method, guideline, deviations if any	Test substance,	Relevant information about the study (as applicable)	Observations	Reference
chromosome aberration (EPA data requirement subdivision F, Series 84-2. Design resembles OECD 475)	MK-0244, batch L-656,748-052S002, purity 95.9%	Organism/ strain: Mouse (Crl :CD-1 (ICR) BR, 5 males/ dose Concentration tested: single oral exposure 0, 8, 26, 80 mg/kg bw (expressed as free base) Analysis at 6, 12 and 48 h after treatment Positive control: Mitomycin G	no chromosome aberrations	B.6.4.2, STUDY 1

Table 34: Summary table of human data relevant for germ cell mutagenicity

No data available.

10.8.1 Short summary and overall relevance of the provided information on germ cell mutagenicity

Several in vitro studies were performed. Based on a bacterial mutagenesis assay,of which the study design resembles OECD 471, emamectin HCl salt (MK-0243), purity 92.8% did not result in a two-fold or greater increase in the number of revertant colonies and a dose-related increase in number of revertant colonies was also not observed (B.6.4.1, STUDY 1). Under the test conditions, emamectin HCl salt (MK-0243) did not induce point mutations in *S. typhimurium* or *E. coli*.

Another in vitro test is available which was performed to determine the potential of emamectin HCl salt to be mutagenic in V-79 Chinese hamster lung fibroblasts (B.6.4.1, STUDY 2). The test design resembles OECD 476. Emamectin HCl salt did not induce an increased resistance to 6-thioguanine under the test condition in the absence or presence of metabolic activation. A positive dose resonse relation was lacking. Under the test conditions, emamectin HCL salt (MK-0243) did not induce gene mutations in mammalian cells.

Another study was performed to determine if emamectin benzoate hydrate salt has the potential to cause chromosome aberrations in Chinese hamster ovary (CHO) cells (study design resembles OECD 473) (B.6.4.1, STUDY 3). No statistically significant increased incidence in chromosome aberrations was observed for dose levels up to 7 µM (with S-9) or 6 µM (without S-9). Moreover, a dose-related response was not observed. Under the test conditions, emamectin benzoate hydrate salt (MK-0244) did not induce chromosome aberrations in mammalian cells (Chinese hamster ovary cells) up to doses that caused marked growth reduction.

The potential of MK-0243 (emamectin HCl salt) to induce single- and double-strand DNA breaks and cytotoxicity was tested in rat hepatocytes in an *in vitro* alkaline elution/rat hepatocyte assay (B.6.4.1, STUDY 4). For this test no official guideline is available and the study is therefore considered supplementary. Under the test conditions emamectin HCl salt (MK-0243) did not induce DNA strand breaks in primary rat hepatocytes.

In an in vivo study (B.6.4.2, STUDY 1), male mice received by gavage a single dose of emamectin benzoate hydrate in 0.5% aqueous methylcellulose in doses of 0, 8, 26 or 80 mg/kg bw. The study design resembles OECD 475. Under the test conditions, emamectin benzoate hydrate did not induce chromosome aberrations in mouse bone marrow cells. Clinical signs (tremors, erect tails, decreased activity, ptosis, bradypnoea and hypothermia) were observed in the high dose group throughout the study. One animal in the high dose group died. In the mid-dose group all animals had ptosis 6h after administration. At 24 and 48h occasionally ptosis and tremors were observed in this group. No clinical signs were observed in the low dose group. No statistically significant increase in chromosome aberrations was observed in the emamectin benzoate hydrate-treated mice. The positive control, mitomycin G, induced highly significant increases in chromosome aberrations.

10.8.2 Comparison with the CLP criteria

According to Regulation EC No 1272/2008 (CLP), Table 3.5.2.2, classification in Category 1 mutagen is based on:

Substances known to induce heritable mutations or to be regarded as if they induce heritable mutations in the germ cells of humans. Substances known to induce heritable mutations in the germ cells of humans.

The classification in Category 1A is based on positive evidence from human epidemiological studies. Substances to be regarded as if they induce heritable mutations in the germ cells of humans.

The classification in Category 1B is based on: – positive result(s) from in vivo heritable germ cell mutagenicity tests in mammals; or – positive result(s) from in vivo somatic cell mutagenicity tests in mammals, in combination with some evidence that the substance has potential to cause mutations to germ cells. It is possible to derive this supporting evidence from mutagenicity/genotoxicity tests in germ cells in vivo, or by demonstrating the ability of the substance or its metabolite(s) to interact with the genetic material of germ cells; or – positive results from tests showing mutagenic effects in the germ cells of humans, without demonstration of transmission to progeny; for example, an increase in the frequency of aneuploidy in sperm cells of exposed people.

Classification in Category 2 mutagen is based on:

- *Positive evidence obtained from experiments in mammals and/or in some cases from in vitro experiments, obtained from:*
    - *Somatic cell mutagenicity tests in vivo, in mammals; or*
    - *Other in vivo somatic cell genotoxicity tests which are supported by positive results from in vitro mutagenicity assays*

Based on the results of an adequate range of in vitro studies (gene mutation tests with bacterial and mammalian cells, a chromosome aberration test with mammalian cells, and a test for DNA strand breaks with primary rat hepatocytes) and an in vivo study (a chromosome aberration test in male mice), it is concluded that emamectin benzoate does not fulfill the criteria for classification for germ cell mutagenicity.

10.8.3 Conclusion on classification and labelling for germ cell mutagenicity

No classification is proposed.

10.9 Carcinogenicity

Table 35: Summary table of animal studies on carcinogenicity

Method, guideline, deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results[1]	Reference
104 weeks, oral, rat oral Rat, Sprague-Dawley Crl:CD(SD)BR Test substance: 75/sex/dose Controls: 130/sex (no guideline, but comparable to OECD 453)	MK-0244 (L-656,748-052S, technical; purity 95.9% at initiation, 97.4 to 98.6% weeks 10, 41, 60, 82 and 105)[a] 0, 0, 0.25, 1.0, and 5.0[c]/ 2.5[d] mg/kg bw/day a: the stability of the test compound was not reported (but was determined to be satisfactory in this study by the notifier). b: the dose of emamectin benzoate hydrate salt were calculated as base compound by using a factor of 1.15 (based on the stoichiometry of water in the MK-0244 crystal structure) (note of the notifier) c: f: weeks 1-9 and m: weeks 1-5 d: f: weeks 10-104 and m: weeks 6-104	NOAELcarcinogenicity: 5.8/2.9 mg/kg bw/day (highest dose tested) Neoplastic findings: no treatment related effect	B.6.5.1, STUDY 1
79 weeks, oral, mouse Oral, Mouse, Crl:CD-1, 50/sex/group (no guideline, but comparable to OECD 451)	MK-0244 (L-656,748-052S (Lot #2); purity 97.%) m: 0, 0, 0.5, 2.5 and 12.5/7.5/5 mg/kg bw/day f: 0, 0, 0.5, 2.5 and 12.5/7.5 mg/kg bw/day since the compound was provided as benzoate salt and to account for the stoichiometry of water in the MK-0244 crystal structure doses of the compound were calculated as base compound by using a factor of 1.15 (except week 1 in which 1.16 was used)	NOAEL carcinogenicity: 14.4/8.6/5.8 (m), 14.4/8.6 (f) (highest dose tested) Neoplastic findings: no treatment related effect	B.6.5.1, STUDY 2

[1] *Studies were performed expressing the active substance as base compound in the original study reports, but all endpoints were recalculated and expressed as emamectin benzoate (ratio base compound to emamectin benzoate 1.15)*

Table 36: Summary table of human data on carcinogenicity

No data available.

Table 37: Summary table of other studies relevant for carcinogenicity

No data available.

10.9.1 Short summary and overall relevance of the provided information on carcinogenicity

A long-term oral toxicity/carcinogenicity study with rats (0, 0.25, 1.0 and 5.0/2.5 mg/kg bw/day during 104 weeks) and an oral carcinogenicity study with mice (0, 0.5, 2.5 and 12.5/7.5/5.0 (m) or 12.5/7.5 (f) mg/kg bw/day during 79 weeks) were performed (B.6.5.1, STUDY 1 and B.6.5.1, STUDY 2). Both species were administered emamectin benzoate hydrate via the diet.

In the study with rats (B.6.5.1, STUDY 1), several parameters were changed in high dose animals, the most prominent effect being vacuolar degeneration of neurons in brain and spinal cord and effects on bodyweight gain. Based on effects observed on female blood triglyceride levels and on male relative weights of kidney and liver at and above 1.2 mg/kg bw/day, the NOAEL in this study is 0.29 mg/kg bw/day. In this study with rats, no substance-related increase in tumours was observed.

Based on increased mortality, marked decreased weight gain, clinical signs of neurotoxicity (tremors), increased incidence of skin lesions, changes in haematological parameters and increased relative organ weights observed in high dose mice, the NOAEL in the mice study is 2.9 mg/kg bw/day, expressed as emamectin benzoate (B.6.5.1, STUDY 2). No treatment-related increase in tumour incidence was observed in mice.

Overall, the NOAEL for long term oral exposure to emamectin benzoate is 0.29 mg/kg bw/day. There was no evidence of carcinogenicity in either the rat or the mouse at any of the dose levels employed. In addition, no increases in pre-neoplastic changes were observed.

10.9.2 Comparison with the CLP criteria

According to Regulation EC No 1272/2008 (CLP), Table 3.6.1, classification for carcinogens is based on:

CATEGORY 1: Known or presumed human carcinogens A substance is classified in Category 1 for carcinogenicity on the basis of epidemiological and/or animal data. A substance may be further distinguished as:

- *Category 1A, known to have carcinogenic potential for humans, classification is largely based on human evidence, or*
- *Category 1B, presumed to have carcinogenic potential for humans, classification is largely based on animal evidence.*

The classification in Category 1A and 1B is based on strength of evidence together with additional considerations (see section 3.6.2.2). Such evidence may be derived from:

- *human studies that establish a causal relationship between human exposure to a substance and the development of cancer (known human carcinogen); or*
- *animal experiments for which there is sufficient (1) evidence to demonstrate animal carcinogenicity (presumed human carcinogen).*

In addition, on a case-by-case basis, scientific judgement may warrant a decision of presumed human carcinogenicity derived from studies showing limited evidence of carcinogenicity in humans together with limited evidence of carcinogenicity in experimental animals.

CATEGORY 2: Suspected human carcinogens The placing of a substance in Category 2 is done on the basis of evidence obtained from human and/or animal studies, but which is not sufficiently convincing to place the substance in Category 1A or 1B, based on strength of evidence together with additional considerations (see section 3.6.2.2). Such evidence may be derived either from limited(1) evidence of carcinogenicity in human studies or from limited evidence of carcinogenicity in animal studies.

For emamectin benzoate, there was no evidence of carcinogenicity in either the rat or the mouse at any of the dose levels employed in the available studies.

10.9.3 Conclusion on classification and labelling for carcinogenicity

No classification is proposed.

10.10 Reproductive toxicity

10.10.1 Adverse effects on sexual function and fertility

Table 38: Summary table of animal studies on adverse effects on sexual function and fertility

Method, guideline, deviations if any, species, strain, sex, no/group	**Test substance, dose levels duration of exposure**	**Results[1]**	**Reference**
Range-finding study Rat, Sprague-Dawley Crl:CD(SD) Br strain 12 mated females/dose (no guideline)	Orally, by gavage or in the diet from GD 0 to LD 21 Gavage: 0, 0.1, 0.7 and 5.0 mg/kg bw/day a Diet, 0, 1, 7 and 50 ppm, approximately equal to 0, 0.1, 0.7 and 5.0 mg/kg bw/day	<u>Critical effects:</u> Treatment-related maternal toxicity (body weight loss during lactation period, decreased food consumption) was observed in the high-dose gavage- and diet-treated groups. In the offspring of the high-dose gavage- and diet-treated groups toxicity was evidenced by clinical signs (tremors) and reduced body weight gains, as well as (gavage only) increased mortality between LD8-15. In pups of the high-dose diet-treated group a reduced brain weight and neuronal degeneration in the brain and spinal cord were additionally observed. Observations: At the highest does levels tested (gavage and dietary treatment group) body weight loss was observed up to 78% difference in body weight gain compared to the control on lactation day 8 and decreased food consumption up to 42% compared to the control at lactation day 12 was observed. Clinical signs (tremors) and reduced body weight gains (up tot 62% compared to the control at lactation day 14) were observed in the offspring in the high dose group (gavage and deietary treatment group). In the gavage treatment group, an increased mortality was observed following exposure to 5.0 mg/kg bw/day. Tremors were observed at LD11-12 in about one-third of the pups in the gavage treatment group (5.0 mg/kg bw/day). In addition, a few pups were pale, cold, weak and/or breathing shallow. In the dietary treatment group a reduction in post implantation survival was mainly due to 1 female with 100% post implantation loss. This was considered not treatment-related, since post implantation survival in the other high-dose females was comparable to controls. At LD11-12 tremors were observed in all pups of the high-dose group. In addition a few pups in the high dose groups were pale, cold, weak and/or breathing shallow. In 12/16 examined pups neurons within the pons, and spinal gray matter were swollen. The swollen neurons had an increased amount of eosinophilic cytoplasm, central chromatolysis and the nucleus was displayed to the periphery of the cell body.	B.6.6.1, STUDY 1
Dietary 2-generation	0, 0.1, 0.6 and 3.6/1.8 mg/kg bw/day. a,b	<u>NOAEL:</u> Parental: 0.68 mg/kg bw/day	B.6.6.1, STUDY 2

Method, guideline, deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results[1]	Reference
study of reproductive toxicity (Resembles OECD 416) Rat, Sprague-Dawley Crl:CD(SD) Br strain, 33/sex/dose	a Expressed as base (factor 1.15) b The high-dose level was lowered to 1.8 mg/kg bw/day for F0 and F1A females on GD0.	fertility: 0.68 mg/kg bw/day development: 0.68 mg/kg bw/day <u>LOAEL:</u> Parental: 2.1 mg/kg bw/day fertility: 2.1mg/kg bw/day development: 2.1mg/kg bw/day <u>Critical effects:</u> Parental: Reduced bw gain (males), food consumption in females during premating, reduced food consumption during lactation (females), neuronal degeneration in brain, spinal cord and (at 4.1 mg/kg bw in males expressed as emamectin benzoate) sciatic nerve. Development: reduced bw gain during lactation Fertility: Reduced fecundity Observations: At the highest dose tested a decrease in body weight gain in F0 males during/after 2nd mating (21%) was observed. In females several effects were observed including an increased body weight gain (16%) and food consumption in females during premating (13%). In the brains and spinal cords of animals of the high dose group very slight to slight degeneration of neurons was observed (brains 29/33 and 23/33 in males and females, respectively, and in spinal cord this was 31/33 and 5/33 for males and females). In the males of the high dose group a few animals (4/33) had very slight degeneration of the sciatic nerve. A slight decrease in fecundity index (pregnant females/mated females) was observed in all treatment groups during the first mating. These values were within the historical control range. No effect on fecundity was observed in the low-and mid-dose groups during the production of the F1b and F2 generation. Therefore the reduced fecundity observed in the low and mid-dose animals in the first mating was considered not related to treatment. In the second mating a decreased fecundity was observed in the high-dose F0 animals producing the F1 generation. Analysis of the affected sex indicated that approximately 20% of the high-dose females failed to produce a pregnancy after mating occurred. Since a reduced fecundity was also observed in the high-dose group producing the F2 generation, it is considered that the observed reduced fecundity at the high dose is treatment-related.	

[1] Studies were performed expressing the active substance as base compound in the original study reports, but all endpoints were recalculated and expressed as emamectin benzoate (ratio base compound to emamectin benzoate 1.14)

Table 39: Summary table of human data on adverse effects on sexual function and fertility

No data available.

Table 40: Summary table of other studies relevant for toxicity on sexual function and fertility

No data available.

10.10.2 Short summary and overall relevance of the provided information on adverse effects on sexual function and fertility

Prior to a 2-generation study a range finding study was performed (B.6.6.1, STUDY 1). In this range finding test groups of 12 mated female rats received the test substance (emamectin benzoate salt), either by gavage or in the diet from gestational day (GD) 0 to lactational day (LD) 21. Treatment-related maternal toxicity (body weight loss and decreased food consumption during the lactation period only) was observed in the high-dose gavage- and diet-treated groups. In the offspring of the high-dose gavage- and diet-treated groups toxicity was evidenced by clinical signs (tremors) and reduced body weight gains, as well as (gavage only) increased mortality between LD8-15. In pups of the high-dose diet-treated group a reduced brain weight and neuronal degeneration in the brain and spinal cord were additionally observed.

In the 2-generation study of reproductive toxicity, resembling OECD 416, emamectin benzoate hydrate was administered via the diet to rats at doses of 0, 0.1, 0.6 and 3.6/1.8 mg/kg bw/day (B.6.6.1, STUDY 2). The NOAEL for parental toxicity was 0.68 mg/kg bw/day, expressed as emamectin benzoate, on the basis of a reduced body weight gain in F0 males during/after 2nd mating, a reduced food consumption and reduced body weight gain in females during lactation, and neuronal degeneration in the brain, spinal cord and (4.1 mg/kg bw/day males only expressed as emamectin benzoate) sciatic nerve, observed at 4.1/2.1 mg/kg bw/day, expressed as emamectin benzoate.

The NOAEL for reproductive toxicity was 0.68 mg/kg bw/day on the basis of a reduced fecundity.

The lower fecundity (and fertility) values across all treated groups in the first mating was not reproduced in the following two matings, except for effects observed in the highest dose group, suggesting that it was not treatment related. Moreover, the values were found to be within the historical control range. In addition there was no clear dose response at the first two matings. However, a dose related decreased fecundity was observed in the high-dose group producing the F2 generation. The reduced fecundity and fertility at the high dose level is considered to be treatment related, and to be a secondary consequence of neurotoxicity to the male leading to ineffective copulation.

10.10.3 Comparison with the CLP criteria

According to Regulation EC No 1272/2008 (CLP), Table 3.7.2.2, classification as for effects on fertility is based on:

Category 1A:

Known human reproductive toxicant

Category 1B:

Presumed human reproductive toxicant largely based on data from animal studies

- *clear evidence of an adverse effect on sexual function and fertility in the absence of other toxic effects, or*
- *the adverse effect on reproduction is considered not to be a secondary non-specific consequence of other toxic effects*

Category 2:

Suspected human reproductive toxicant

- *some evidence from humans or experimental animals, possibly supplemented with other information, of an adverse effect on sexual function and fertility and*
- *where the evidence is not sufficiently convincing to place the substance in Category 1 (deficiencies in the study).*
- *the adverse effect on reproduction is considered not to be a secondary non-specific consequence of the other toxic effects*

According to the CLP criteria classification as Repr. 1A is based on human data. No human data is available for emamectin benzoate and therefore, classification as Repr 1A is not justified.

The reproductive effects observed included reduced fecundity. Reduced fecundity is only observed at a high dose. Although a clear dose response relationship was absent during first and second mating producing F1 animals, a steep dose response relationship was observed at the highest dose in animals producing the F2 generation. At this dose, mating behaviour is considered to be influenced by parental effects not directly related to reproduction (e.g. neurotoxicity), and therefore the effects on mating behaviour may not warrant classification. In fact, based on the available (neurotoxicity) studies performed with emamectin benzoate it can be concluded that most characteristic for the toxicity of emamectin benzoate is the clinical and histopathological evidence of neurotoxicity, with tremors and neuronal degeneration in brain and spinal cord observed in the majority of toxicity studies which also showed a very steep dose-response curve for these effects.

The criteria for classification for cat. 1B and cat 2 are not met. Emamectin benzoate was not found to have a direct effect on fertility and it is therefore considered not necessary to classify emamectin benzoate for this endpoint.

10.10.4 Adverse effects on development

Table 41: Summary table of animal studies on adverse effects on development

Method, guideline, deviations if any, species, strain, sex, no/group	**Test substance, dose levels duration of exposure**	**Results[1]**	**Reference**
Oral developmental toxicity study (no guideline) Rat (CD), 25 mated females/dose	MK-0243: L-656,748-038W002 benzoate salt, Purity: 94.2% orally by gavage; days 6-19 of gestation, 0, 2, 4, and 8 mg/kg bw per day* * a factor 1.14 was used to calculated the dosages as base compound	<u>NOAEL:</u> Matern: 2.28 mg/kg bw/day Dev: 2.28 mg/kg bw/day <u>LOAEL:</u> Matern: 4.56 mg/kg bw/day Dev: 4.56 mg/kg bw/day <u>Critical effects:</u> Maternal: decreased bw gain; at high dose also clin.	B.6.6.2, STUDY 1

Method, guideline, deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results[1]	Reference
		signs of neurotox Development: incomplete ossification	
Range finding -Oral developmental toxicity study (no guideline) Rabbit NZW, 10 pregnant females/dose**	MK 0243; L656,748-038W002, Benzoate salt, purity 96.2% orally by gavage; days 6-18 of gestation, 0, 2, 4, 6 and 8 mg/kg bw per day* * a factor 1.14 was used to calculated the dosages as base compound ** one female in the 4 mg/kg group was misdosed on GD 6, removed from the study and replaced by another female.	<u>NOAEL:</u> Matern: 4.56 Dev: 9.12 <u>LOAEL:</u> Matern: 6.84 Dev: >9.12 <u>Critical effects:</u> Maternal: decreased bw gain and food consumption; Development: no embryo/foetotox.	B.6.6.2, STUDY2a
Oral developmental toxicity study Rabbit NZW, 18 pregnant females/dose	MK 0243; L656,748-038W002 Benzoate salt, purity 94.2% orally by gavage; days 6-18 of gestation, 0, 1.5, 3, and 6 mg/kg bw per day* * a factor 1.14 was used to calculated the dosages as base compound	<u>NOAEL:</u> Matern: 3.42 mg/kg bw/day Foeto: 6.84 mg/kg bw/day <u>LOAEL:</u> Matern: 6.84 mg/kg bw/day Foeto: >6.84 mg/kg bw/day <u>Critical effects:</u> Maternal: decreased bw gain; mydriasis; decreased pupillary reaction Development: no embryo/foetotox.	B.6.6.2, STUDY 2
Developmental neurotox. Study Rat (CD) Oral range-finding reproduction study in female rats Rat Crl:CD(SD) Br strain 25 f/dose (no guideline, but in accordance with OECD 426)	MK-0244: L-656,748-052S002 Purity: >97% Orally by gavage, GD6-LD20 0, 0.1, 0.6, and 3.6/2.5* mg/kg bw** * between gestation day 17 and 20 the high dose level of 3.6 mg/kg bw per day was reduced to 2.5 mg/kg bw per day due to the appearance of pup tremors in the 3.6 mg/kg dose group of a concurrent 2-generation reproduction study ** all dose levels are expressed as base compound (factor 1.15)	<u>NOAEL:</u> maternal: 2.85 mg/kg bw/day development: 0.68 mg/kg bw/day <u>LOAEL:</u> maternal: >2.85 mg/kg bw/day development: 2.85 mg/kg bw/day <u>Critical effects:</u> maternal: - development: clinical signs of neurotox; growth retardation; neurobehavioural effects	B.6.7.3, STUDY 1
Range-finding	Orally, by gavage or in the diet	<u>Critical effects:</u> Treatment-related maternal toxicity	B.6.6.1,

Method, guideline, deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results[1]	Reference
study Rat, Sprague-Dawley Crl:CD(SD) Br strain 12 mated females/dose (no guideline)	from GD 0 to LD 21 Gavage: 0, 0.1, 0.7 and 5.0 mg/kg bw/day a Diet, 0, 1, 7 and 50 ppm, approximately equal to 0, 0.1, 0.7 and 5.0 mg/kg bw/day a	(body weight loss, decreased food consumption) was observed in the high-dose gavage- and diet-treated groups. In the offspring of the high-dose gavage- and diet-treated groups toxicity was evidenced by clinical signs (tremors) and reduced body weight gains, as well as (gavage only) increased mortality between LD8-15. In pups of the high-dose diet-treated group a reduced brain weight and neuronal degeneration in the brain and spinal cord were additionally observed.	STUDY 1
Dietary 2-generation study of reproductive toxicity (Resembles OECD 416) Rat, Sprague-Dawley Crl:CD(SD) Br strain, 33/sex/dose	0, 0.1, 0.6 and 3.6/1.8 mg/kg bw/day. a,b a Expressed as base (factor 1.15) b The high-dose level was lowered to 1.8 mg/kg bw/day for F0 and F1A females on GD0.	<u>NOAEL:</u> Parental: 0.68 mg/kg bw/day fertility: 0.68 mg/kg bw/day development: 0.68 mg/kg bw/day <u>LOAEL:</u> Parental: 2.05mg/kg bw/day fertility: 2.05 mg/kg bw/day development: 2.05 mg/kg bw/day <u>Critical effects:</u> Parental: Reduced bw gain (males), increased body weight gain and food consumption in females during premating, reduced food consumption during lactation (females), neuronal degeneration in brain, spinal cord and (at 3.6 mg/kg bw in males) sciatic nerve. Development: Clinical signs (tremors, hindlimb splay), reduced bw gain during lactation Fertility: Reduced fecundity	B.6.6.1, STUDY 2

[1] Studies were performed expressing the active substance as base compound in the original study reports, but all endpoints were recalculated and expressed as emamectin benzoate (ratio base compound to emamectin benzoate 1.14)

Table 42: Summary table of human data on adverse effects on development

No data available.

Table 43: Summary table of other studies relevant for developmental toxicity

No data available.

10.10.5 Short summary and overall relevance of the provided information on adverse effects on development

The reduced number of pups per litter observed in the oral range finding study in rat (B.6.6.1, STUDY 1) at the high dose level is unlikely to be treatment related, as there was no clear dose response, and

the effect was not reproduced across the next two matings, suggesting that this apparent reduction at the high dose in the F1a generation probably reflects normal variation in this parameter.

In an oral developmental study in rats (B.6.6.2, STUDY 1), the NOAEL for maternal toxicity is established at 2.28 mg/kg bw per day, based on decreases in body weight gain in the mid and high dose group and clinical signs of neurotoxicity in the high dose group. The NOAEL for embryo/foetotoxicity is established at 2.28 mg/kg bw per day, expressed as emamectin benzoate, based on dose-related increases in the number of foetuses with incomplete ossification (19/344, 18/315, 27/312 and 46/332 at 0, 2, 4 and 8 mg/kg bw/day, respectively) and the number of sites with incomplete ossification (20/344, 21/315, 32/312 and 78/332 at 0, 2, 4 and 8 mg/kg bw/day, respectively). Additionally, a slight decrease in foetal weight, an increase in the number of resorptions, and an increase in skeletal variations (wavy rib and supernumary ribs) were observed in the high dose group administered emamectin benzoate salt. There was no evidence of a teratogenic effect in the rat. The incomplete ossification was in some cases significant in only the high dose group. The is possibly related to decreased fetal weight and might not be a specific effect of emamectin benzoate salt on skeletal maturation. However, it should be noted that the observed decrease in fetal body weight was only minor and not significantly different from control animals. Although the effect on body weight gain in mid dose females appears to be rather mild, a clear decrease in body weight gain was observed from GD 14 onwards. The fetal effects might be correlated to the BW effects in the dams. Overall the incomplete ossification is considered correlated to decreased body weight gain in fetuses and most likely dams, is only slight in the mid dose.

In a range finding oral developmental study in rabbits (B.6.6.2, STUDY2a), a body weight loss and reduced food consumption were observed at 6.84 and 9.12 mg/kg bw/d, expressed as emamectin benzoate, and clinical signs (including tremors in 1 female) were observed in high dose females administered emamectin benzoate salt. No maternal effects were observed at 4.56 mg/kg bw/d. In 2 fetuses in the high dose cleft palate and/or hydrocephaly was observed. These fetuses were from 2 separate litters, and from females with the greatest body weight loss and/or tremors. The effects were therefore considered to be secondary to maternal toxicity.

In an oral developmental study in rabbits (B.6.6.2, STUDY 2), a decrease in body weight gain, mydriasis and decreased pupillary reaction were observed in the high dose group, but no clinical signs of neurotoxicity. The NOAEL for maternal toxicity is established at 3 mg/kg bw per day. There was no evidence of a teratogenic effect in the rabbit, and no embryo and/or foetotoxic effects were observed at any dose level. Thus, the NOAEL for embryo/foetotoxicity is established at 6 mg/kg bw per day, the highest dose tested.

A developmental neurotoxicity study was conducted with rats given emamectin benzoate hydrate by gavage at doses of 0, 0.1, 0.6 or 3.6/2.5 mg/kg bw/day (B.6.7.3, STUDY 1). No deaths or abortions occurred and there were no treatment-related clinical signs of toxicity in F0 females at any dose level, but weight gain during gestation was significantly ($p < 0.05$) elevated by 11 and 15% in the 0.6 and 4.1/2.85 mg/kg bw per day groups, respectively. This increase in body weight gain is not considered adverse, given the absence of any other effect in these females. Reproductive performance, as assessed by implantation rate, live litters, duration of gestation, post-implantation survival and pup viability at birth, was unaffected at all dose levels. Further, no effects were found on the external morphology of F1 pups and their sex ratio and pre-weaning survival. In pups of the high dose group, intermittent head tremors progressing to whole body tremors and hind limb extension progressing to hind limb splay were observed in all pups, starting between day 6 and day 10 of lactation and persisting into the post-weaning period. Pre-weaning and post-weaning pup weights and weight gain were reduced in both sexes at this dose level. The decreases in F1 pups preweaning were first apparent on day 11 in both females and males (14% and 10% below control, respectively) and became progressively worse with time (to 42% and 40% below control, respectively on day 21). In F1 pups postweaning treatment-related decrease in average postweaning body weight gain in the highest dose tested were 18% and 17% below the control in females and males, respectively. A delay of 3.6 - 3.7 days in preputial separation and vaginal opening, also observed in pups of the high dose group are considered to be associated with this reduced weight gain. Test substance-related behavioural effects were observed in pups of the high dose group, but

not at lower dose levels. Increased motor activity, occurring on day 13, is interpreted by the study authors as an expression of stereotypical behavioural movement consistent with the observed tremors. On day 17, motor activity was reduced by 30 – 41% but was not significantly affected on day 21. An effect on the auditory startle response was apparent on day 22, the amplitude of response was reduced by 74% and the interval between stimulus and peak response increased by 22 – 33%. Effects on motor activity in females and the auditory startle response in both sexes persisted into the post-weaning period. Learning and short- and long-term retention, measured by a passive avoidance technique, was unaffected by test substance at all dose levels. The effects on brain weights seen in both 11 and 60 day old pups of the high dose group were considered to be secondary to the observed growth retardation. No test substance-related histopathological and histomorphometric effects occurred in the brains of either the 11 or 60 day old F1 pups. Further, no test substance-related gross or microscopic alterations occurred in central and peripheral nervous tissues or skeletal muscle in 60 day old F1 pups from the high dose group.The NOAEL for developmental neurotoxicity was established as 0.68 mg/kg bw per day, expressed as emamectin benzoate, based on the occurrence of clinical evidence of neurotoxicity, growth retardation and alterations of neurobehavioural function in the F1 progeny of females administered emamectin benzoate hydrate at 2.85 mg/kg bw per day during the period of gestation (day 6) through lactation (day 20). In the pups, no histopathological evidence of neurotoxicity was observed. In the absence of evidence of toxic effects in dams, the NOAEL for maternal toxicity was 2.85 mg/kg bw/day, expressed as emamectin benzoate, the highest dose tested.

The NOAEL for developmental toxicity was 0.68 mg/kg bw/day on the basis of reduced body weight gain during the lactation period.

10.10.6 Comparison with the CLP criteria

Annex I (3.7.2.4.2) of the CLP criteria states the following: ‘*Based on pragmatic observation, maternal toxicity may, depending on severity, influence development via non-specific secondary mechanisms, producing effects such as depressed foetal weight, retarded ossification, and possibly resorptions and certain malformations in some strains of certain species. However, the limited number of studies which have investigated the relationship between developmental effects and general maternal toxicity have failed to demonstrate a consistent, reproducible relationship across species. Developmental effects which occur even in the presence of maternal toxicity are considered to be evidence of developmental toxicity, unless it can be unequivocally demonstrated on a case-by-case basis that the developmental effects are secondary to maternal toxicity. Moreover, classification shall be considered where there is a significant toxic effect in the offspring, e.g. irreversible effects such as structural malformations, embryo/foetal lethality, significant post-natal functional deficiencies.’*

According to Regulation EC No 1272/2008 (CLP), Table 3.7.2.2, classification as for effects on development is based on:

Category 1A:

Known human reproductive toxicant

Category 1B:

Presumed human reproductive toxicant largely based on data from animal studies

- *clear evidence of an adverse effect on development in the absence of other toxic effects, or*
- *the adverse effect on development is considered not to be a secondary non-specific consequence of other toxic effects*

Category 2:

Suspected human reproductive toxicant

- *some evidence from humans or experimental animals, possibly supplemented with other information, of an adverse effect on development and*
- *the evidence is not sufficiently convincing to place the substance in Category 1 (deficiencies in the study).*
- *the adverse effect on development is considered not to be a secondary non-specific consequence of the other toxic effects*

According to the CLP criteria classification as Repr. 1A is based on human data. No human data is available for emamectin benzoate and therefore, classification as Repr 1A is not justified.

In the developmental toxicity studies, there was no strong evidence of teratogenicity. In rats, the incomplete ossification is possibly correlated to decreased body weight gain in fetuses and most likely dams. In the mid-group dams a slightly decreased body weight gain was observed from GD 14 onwards. At the high dose level, a remarkable reduction of body weight was observed in the dams (33%). The fetal effects observed in the high dose group might thus be correlated to the BW effects in the dams. According to the CLP Criteria (section 3.7.2.2.1.1) ‘*Classification is not necessarily the outcome in the case of minor developmental changes, when there is only a small reduction in foetal/pup body weight or retardation of ossification when seen in association with maternal toxicity.*’ Classification is not warranted in such cases. However, considering that there is no strong correlation between the incomplete ossification observed in the mid dose groups and the decreased body weight gains in the dams, a primary effect on ossification related to treatment cannot be completely ruled out. Nevertheless, since this was the only developmental effect observed and considering that ossification was only delayed without causing any permanent damage, classification is not warranted even when the effect on ossification would be primary caused by emamectin benzoate exposure.

In rabbits, in 2 fetuses in the high dose cleft palate and/or hydrocephaly was observed in a range-finding study. These fetuses were from 2 separate litters, and from females with the greatest body weight loss and/or tremors. The effects were therefore considered to be secondary to maternal toxicity. In the developmental rabbit study, there was no evidence of a teratogenic effect in the rabbit, and no embryo and/or foetotoxic effects were observed at any dose level.In the developmental rabbit study, a slight increase in foetal weight seen in the mid dose group was not considered treatment-related since there was no dose relation.

In the developemental neurotoxicity study in rat also an effect on pup weight was observed as pre-weaning and post-weaning pup weights and weight gain were reduced in both sexes at the high dose level. Moreover, a slight decrease in post-weaning weight gain was also observed at the lower doses tested but this was not significantly different compared to the control. These findings were observed in the presence of maternal toxicity and not considered sufficient for classification.

It is considered not necessary to classify emamectin benzoate for developmental toxicity.

10.10.7 Adverse effects on or via lactation

According to Regulation EC No 1272/2008 (CLP), Table 3.7.2.2.2, classification for lactation effects is based on:

(a) human evidence indicating a hazard to babies during the lactation period; and/or

(b) results of one or two generation studies in animals which provide clear evidence of adverse effect in the offspring due to transfer in the milk or adverse effect on the quality of the milk; and/or (

c) absorption, metabolism, distribution and excretion studies that indicate the likelihood that the substance is present in potentially toxic levels in breast milk.

The 2-generation study and developmental toxicity studies did not report any adverse findings occurring via lactation.

10.10.8 Conclusion on classification and labelling for reproductive toxicity

No classification is proposed.

10.11 Specific target organ toxicity-single exposure

Table 44: Summary table of animal studies on STOT SE

Method, guideline, deviations if any	Species, strain, sex, no/group, test substance, dose levels, duration of exposure	Results[1]	Reference
Acute oral toxicity, up & down procedure (Guideline OECD 425)	Rat; Sprague Dawley albino, 1-3 females/dose Emamectin technical, benzoate salt Batch: SNA6B019 Purity: 96.2% (MK244) Single dose, gavage 20.8, 66, 208, and 658 mg/kg bw* *Dose levels are expressed as base compound (factor 1.14)	Mortality: In the up and down procedure the first rat tested at 237 mg/kg died, the 2nd survived and the 3rd died. Also the rat dosed with 750 mg/kg died. Symptoms of toxicity: Signs of toxicity were found in the 208 and 750 mg/kg dose groups: hypoactivity, tremors, soft faeces and/or reduced faecal volume, diarrhoea, ano-genital staining. Body weight: During the first week the surviving animal at 237 mg/kg dose level lost weight, but gained weight over the remaining week of the study. Animals of the lower dose groups gained weight over the total period of the study. Pathology: Discolouration of intestines was found in the decedents of the 237 and 750 mg/kg dose groups.	B.6.2.1.1, STUDY 1
Acute oral toxicity (guideline in accordance with OECD 401)	Rat Crl:CD(SD) BR strain, 5/sex/dose L656,748-010V003 Hydrochloride salt Purity: 96.9%* * 92.8% L-656,748 B1a and 4.1% B1b; 0.76% (w/w) propylgallate added as an antioxidant. Single dose, gavage 44.4, 66.6, 100, 150, and 225 mg/kg bw** ** dose levels are expressed as base compound (factor 1.04).	Mortality: Death occurred from day 1-5 in the high dose group and from day 4 -12 in all other groups. Rats were moribund with loss of righting reflex before death. Symptoms of toxicity: Clinical signs were similar for both sexes. At all dose levels mucous-like stools were noted 2-4 hours after dosing. Ataxia and whole body tremors occurred in all animals of all dose groups within 4 hours after exposure in the high dose groups (171 and 257 mg/kg) and starting after 2 days in the low dose groups and persisted sometimes several days. Bradypnoea was observed in some animals in the low dose groups and nearly all animals of the high dose groups. Irritability, salivation and lacrimation were seen at most dose levels from the 2nd to the 3rd or 4th day. And also decreased activity was observed in several animals. Body weight: Unaffected in low dose group. At higher dose levels (76 mg/kg and above) animals showed weight loss or slower weight gain during first week. Weight gain resumed in second week. Pathology: No treatment-related gross findings.	B.6.2.1.1, STUDY 2
Acute oral toxicity	Rat Crl:CD(SD) BR strain, 5/sex/dose	Mortality: Death occurred from day 3-6. **Dose group** | **36** | **47** | **62** | **80** | **104**	B.6.2.1.1, STUDY 3

<table>
<tr><th>Method, guideline, deviations if any</th><th>Species, strain, sex, no/group, test substance, dose levels, duration of exposure</th><th>Results[1]</th><th>Reference</th></tr>
<tr>
<td>(Guideline in accordance with OECD 401)</td>
<td>MK 0244
L656,748-052S lot #5
Benzoate hydrate salt
Purity: 97.8%

Single dose, gavage

32, 41.6, 54.1, 70.3, and 91.4 mg/kg bw*

*dose levels are expressed as base compound (factor 1.16).</td>
<td>
<table>
<tr><th>(mg/kg bw)*</th><th></th><th></th><th></th><th></th><th></th></tr>
<tr><td>Males
Females</td><td>0
0</td><td>0
0</td><td>2
0</td><td>3
1</td><td>4
5</td></tr>
</table>
*dose levels are expressed as emamectin benzoate

<u>Symptoms of toxicity:</u> Clinical signs were similar for both sexes. Tremors occurred in all animals of all dose groups. Bradypnoea, ptosis and decreased activity were observed in the 47 mg/kg dose group and higher, and lateral recumbency, urine staining and red discharge of nose and eyes were observed at the two highest dose levels.

<u>Body weight:</u> Compared to the low dose group, a dose dependent decrease in body weight gain was observed in all other groups in the first week after dosing. Body weight losses were not observed.

<u>Pathology:</u> No treatment-related gross findings.
</td>
<td></td>
</tr>
<tr>
<td>Acute oral toxicity bioequivalence study

(no guideline)</td>
<td>Rat Crl:CD(SD) BR strain, 5 females/group

MK 0244
L656,748-038W lot #2
(benzoate-methyl t-butyletherate solvate; purity 96.4 %)
and
L656,748-052S lot #2
(Benzoate hydrate salt; purity 99.1%)

Single dose, gavage

40, 60, 90 and 135 mg/kg bw*

* all dose levels are expressed as base compound: factor 1.14 for 038W and factor 1.16 for 052S.</td>
<td>
<u>Mortality:</u> Death occurred from day 2-6.
<table>
<tr><th>Dose group (mg/kg bw)</th><th>46</th><th>68</th><th>103</th><th>154</th></tr>
<tr><td>038W
052S</td><td>0
0</td><td>4
3</td><td>5
5</td><td>5
5</td></tr>
</table>
*dose levels are expressed as emamectin benzoate

The incidence of mortality for both solvates showed no significant difference (calculated by the study authors conform the Mantel Haenszel procedure: J. Natl. Cancer Inst. 22: 719-748, 1959).

<u>Symptoms of toxicity:</u> Clinical signs were similar for both solvates. Most effects were seen at dose levels of 68 mg/kg and higher: bradypnoea, decreased activity, ptosis, salivation, laying on one side, blood-like staining of eyes, nose or mouth, urine staining, and loss of righting reflex. Tremors and ataxia were observed already in the 46 mg/kg dose group. Some of the effects like bradypnoea, loss of righting reflex and ptosis started for 038W at a lower dose level than for 052S, whereas decreased activity was seen at a lower dose level for 052S.

<u>Body weight:</u> A slight decrease in body weight gain was observed in the 68 mg/kg bw dose groups in the first week after dosing compared to the low dose group. Body weight losses were not observed (only based on 1 surviving animal for 038W, and 2 surviving animals for 052S).

<u>Pathology:</u> No treatment-related gross findings.
</td>
<td>B.6.2.1.1, STUDY 4</td>
</tr>
<tr>
<td>Acute oral toxicity study

(no guideline, but resembles OECD 401)</td>
<td>Rat Crl:CD(SD) BR strain, 5 females/group
MK 0244
L656,748-038W lot #2
(benzoate-methyl t-butyletherate solvate; purity unknown)</td>
<td>
<u>Mortality:</u> Death occurred within 24 h after dosing up to day 6.
<table>
<tr><th>Dose group (mg/kg bw)</th><th>46</th><th>78</th><th>132</th><th>223</th><th>381</th></tr>
<tr><td>038W
052S</td><td>0
0</td><td>1
0</td><td>4
5</td><td>5
5</td><td>5
5</td></tr>
</table>
*dose levels are expressed as emamectin benzoate
</td>
<td>B.6.2.1.1, STUDY 5</td>
</tr>
</table>

<table>
<tr><th>Method, guideline, deviations if any</th><th>Species, strain, sex, no/group, test substance, dose levels, duration of exposure</th><th>Results[1]</th><th>Reference</th></tr>
<tr><td></td><td>and
L656,748-052S lot #1 (Benzoate hydrate salt; purity unknown)

Single dose, gavage

40, 68, 116, 196, and 334 mg/kg bw*

* all dose levels are expressed as base compound: factor 1.14 for 038W and factor 1.16 for 052S.</td><td><u>Symptoms of toxicity:</u> Clinical signs were reported to be similar for both salts and occurred at all dose levels within 2-5 h after dosing. Effects observed were: tremors, ataxia, and decreased activity. Further, irritability, bradypnoea and salivation were observed from 78 mg/kg bw (irritability already seen at 46 mg/kg bw for compound 038) and lacrimation and loss of righting reflex were observed at 1332 mg/kg bw and higher. Surviving animals appeared normal by day 8 or 9.

<u>Body weight:</u> It was reported that no apparent effects on body weight were seen.

<u>Pathology:</u> Not performed.</td><td></td></tr>
<tr><td>Acute oral toxicity

(guideline in accordance with OECD 401)</td><td>Mouse Crl:CF-1 BR strain, 5 /sex/dose
L656,748-010V003 Hydrochloride salt
Purity: 96.9% *

* 92.8% L-656,748 B1a and 4.1% B1b; 0.76% (w/w) propylgallate added as an antioxidant.

Single dose, gavage

20, 30, 45, 67.5 and 101.2 mg/kg bw **

**dose levels are expressed as base compound (factor 1.04)</td><td><u>Mortality:</u> Death occurred from 33 minutes to 7 days and was preceded by bradypnoea and loss of righting reflex.
<table><tr><th>Dose group mg/kg bw</th><th>23</th><th>34</th><th>51</th><th>77</th><th>115</th></tr><tr><td>Males
Females</td><td>2
3</td><td>4
0</td><td>5
4</td><td>5
4</td><td>5
5</td></tr></table>*dose levels are expressed as emamectin benzoate

<u>Symptoms of toxicity:</u> Clinical signs were similar for both sexes. Ataxia and whole body tremors occurred in all animals of all dose groups within 2 hours after exposure and persisted several days.

<u>Body weight:</u> Generally unaffected.

<u>Pathology:</u> No data.</td><td>B.6.2.1.1, STUDY 6</td></tr>
<tr><td>Acute oral toxicity study

(no guideline, but resembles OECD 401)</td><td>Mice Crl:CD-1(ICR) BR strain, 5 females/group
MK 0244
L656,748-038W lot #2 (benzoate-methyl t-butyletherate solvate; purity unknown)
and
L656,748-052S lot #1 (Benzoate hydrate salt; purity unknown)

Single dose, gavage

5, 10, 20, 40, and 80 mg/kg bw* (1st study)

80, 144, 259, and 466 mg/kg bw * (2nd study)

* all dose levels are expressed as base</td><td><u>Mortality:</u> No mortality occurred in the first study up to and including 91 mg/kg bw. The mortality in the second study is presented in the table. Death occurred within 24 h after dosing up to day 7 in mice treated with 052S and up to day 10 when treated with 038W.
<table><tr><th>Dose group (mg/kg bw)</th><th>91</th><th>164</th><th>295</th><th>531</th></tr><tr><td>038W
052S</td><td>0
1</td><td>4
4</td><td>5
5</td><td>5
5</td></tr></table>
<u>Symptoms of toxicity:</u> No clinical signs were observed in mice dosed up to 46 mg/kg bw. Clinical signs were similar for both salts. Tremors, ataxia, bradypnoea and decreased activity were seen from day 2 of dosing at 91 and 164 mg/kg bw, and already after 2 hours at higher dose levels. Other effects at these higher dose levels were ptosis and loss of righting. Surviving animals appeared normal by day 7.

<u>Body weight:</u> Marked body weight losses were reported for mice treated with 038W at 295 and 531 mg/kg bw and still alive at day 7. Surviving mice given 164 mg/kg bw showed</td><td>B.6.2.1.1, STUDY 7</td></tr>
</table>

<table>
<tr><th>Method, guideline, deviations if any</th><th>Species, strain, sex, no/group, test substance, dose levels, duration of exposure</th><th>Results[1]</th><th>Reference</th></tr>
<tr><td></td><td>compound: factor 1.14 for 038W and factor 1.16 for 052S</td><td>slight body weight losses during the first week, but recovered most of the weight loss by day 14.

<u>Pathology:</u> Not performed.</td><td></td></tr>
<tr><td>Acute oral toxicity study

(no guideline, but in accordance with OECD 401)</td><td>Mice Crl:CD-1(ICR) BR strain, 5/sex/dose, except high dose group 5 f only

MK 0244
L656,748-052S lot #2
Benzoate hydrate salt;
Purity: 97.6%

Single dose, gavage

70, 120, 192, and 307 mg/kg bw *

* dose levels are expressed as base compound (factor 1.16)</td><td><u>Mortality:</u> Death occurred in the high dose group on day 2 (in a single animal on day 5). At 219 mg/kg bw mortality occurred between days 2 and 10.
<table><tr><th>Dose group (mg/kg bw)</th><th>80</th><th>137</th><th>219</th><th>350</th></tr><tr><td>Males
Females</td><td>1
0</td><td>0
0</td><td>5
4</td><td>-
5</td></tr></table>
<u>Symptoms of toxicity:</u> Tremors were observed in all animals during the first week and in surviving animals of the 120 mg/kg dose group and higher during the second week. Other signs of toxicity were ataxia, bradypnoea, ptosis, decreased activity, and lateral recumbancy. Unkempt coat was observed in some animals of the 137 and 219 mg/kg dose group.

<u>Body weight:</u> Marked body weight losses during the first week were found for the 219 mg/kg dose group. In the 137 mg/kg bw dose group, body weight losses of about 11% were observed during the first week, but mice regained most of the weight loss by day 14. Slight decreases in bodyweight were observed in several animals of the low dose group.

<u>Pathology:</u> No treatment-related gross findings.</td><td>B.6.2.1.1, STUDY 8</td></tr>
<tr><td>Acute oral toxicity study

(no guideline, but resembles OECD 401)</td><td>Mice Crl:CD-1(ICR) BR strain, 5 females/group
MK 0244
L656,748-038W lot #2 (benzoate-methyl t-butyletherate solvate; purity 96.4%)
or
L656,748-052S lot #2 (Benzoate hydrate salt; purity 99.1%)

Single dose, gavage

60, 90, 135, and 202 mg/kg bw*

* all dose levels are expressed as base compound: factor 1.14 for 038W and factor 1.16 for 052S</td><td><u>Mortality:</u> Deaths occurred between day 2 and 12 for compound 038W, and between day 1 and 4 for compound 052S.
<table><tr><th>Dose group (mg/kg bw)</th><th>68</th><th>103</th><th>154</th><th>230</th></tr><tr><td>038W
052S</td><td>0
0</td><td>0
0</td><td>0
2</td><td>5
5</td></tr></table>
The incidence of mortality for both solvates showed no significant difference (calculated by the study authors conform the Mantel Haenszel procedure: J. Natl. Cancer Inst. 22: 719-748, 1959).

<u>Symptoms of toxicity:</u> Clinical signs were observed from the 103 mg/kg dose group onwards in both exposed groups and were observed only during the first week, or up to mortality. Clinical signs in these groups were tremors, decreased activity, ataxia, and ptosis. Additional signs of toxicity observed in the two high dose groups were bradypnoea and loss of righting reflex.

<u>Body weight:</u> Marked body weight losses over the first week were found for the two animals of the 038W-high dose group that died in week 2. In the 154 mg/kg bw dose groups, expressed as emamectin benzoate, slight weight</td><td>B.6.2.1.1, STUDY 9</td></tr>
</table>

Method, guideline, deviations if any	Species, strain, sex, no/group, test substance, dose levels, duration of exposure	Results[1]	Reference
		loss was found in the 052S group and moderate weight loss (17%) in the 038W group. Very slight weight losses were found in most animals of the 103 mg/kg dose groups, expressed as emamectin benzoate. All surviving animals regained (most of) the weight loss during week 2. Pathology: No treatment-related gross findings.	
Acute dermal toxicity (guideline OECD 402)	Rat; Sprague Dawley albino, 5 /sex/dose Emamectin technical, benzoate salt Batch: SNA6B019 Purity: 96.2% (MK244) Single dose, 24 h 877 mg/kg bw (f), and/or 1754 mg/kg bw (m/f)* * Dose levels are expressed as base compound (factor 1.14) (when expressed as emamectin salt, the dose levels are 1000 and 2000 mg/kg bw)	Mortality: Two females of the high dose group were euthanized for humane reasons on day 2. They showed mouth discharge and were in moribund condition. Symptoms of toxicity: Clinical signs were similar for both sexes. All rats at both dose levels showed hypoactivity, tremors and/or ataxia. The females of the 1000 mg/kg dose group, expressed as emamectin benzoate, also showed irregular respiration. All rats recovered by day 11 or 12. Body weight: During the first week animals lost weight. However, all but 2 females of the high dose group gained weight over the remaining week of the study. Pathology: In two females of the high dose group the lungs were found extremely red. Further, no gross abnormalities in any of the animals (m/f).	B.6.2.1.2, STUDY 1
Acute dermal toxicity study (no guideline)	Rat Crl:CD(SD) BR strain, 5 /sex/dose MK 0244 L656,748-052S lot #5 (Benzoate hydrate salt; purity 96.4%) Single dose for 24h 2000 mg/kg bw* *dose levels are expressed as base compound (factor 1.16)	Mortality: One male rat died on day 7. This death was considered to be due to oral ingestion since the rat was found without the occlusive dressing on the morning of day 2. According to the study authors it is likely that this rat ingested a lethal dose since the oral lethal dose is about 91 mg/kg bw. Symptoms of toxicity: Prior to death this animal was observed with tremors, bradypnoea and decreased activity. Tremors were also seen in three of the remaining four males from days 4 or 7, up to day 10, and in one of five females from days 7 to 10. No signs of dermal irritation were seen. Body weight: A decrease in bodyweight (about 15 %) was observed in only one male and female over the first week. Both animals regained some of the weight loss by day 14. Pathology: No treatment-related gross findings	B.6.2.1.2, STUDY 2
Acute dermal toxicity (guideline	Rat; CRL:(WI)BR Wistar, 5 /sex/dose Emamectin technical, benzoate salt	Mortality: **Dose group (mg/kg** | **500** | **1000** | **2000**	B.6.2.1.2, STUDY 3

<table>
<tr><th>Method, guideline, deviations if any</th><th>Species, strain, sex, no/group, test substance, dose levels, duration of exposure</th><th>Results[1]</th><th>Reference</th></tr>
<tr><td>OECD 402)</td><td>Batch: SNA6A015
Purity: 96.6% (MK244G)
Single dose, 24 h
439 mg/kg bw, 877 mg/kg bw, and 1754 mg/kg bw*
*Dose levels are expressed as base compound (factor 1.14) (when expressed as emamectin salt, the dose levels are 500, 1000 and 2000 mg/kg bw)</td><td>
<table>
<tr><th>bw)</th><th></th><th></th><th></th></tr>
<tr><td>Males
Females</td><td>1
0</td><td>3
0</td><td>1
3</td></tr>
</table>
<u>Symptoms of toxicity</u>: In all dose groups clinical signs were observed 2 days after the treatment. These included vocalization, irritability, tremors, tonic convulsion, piloerection, decreased activity, hunched back, discharge coloured, nose, area around eyes and incoordination. Additionally, prone position, dyspnoea and laying on the side were noted in some animals dosed at 1000 mg/kg and 2000 mg/kg.
<u>Body</u> weight: The majority of surviving animals showed marked bodyweight loss during the first week of the observation period.
<u>Pathology:</u> No gross abnormalities in any of the animals (m/f). No treatment related skin irritation was observed in any animal throughout the study.</td><td></td></tr>
<tr><td>Acute toxicity study
(guideline OECD 403)</td><td>Rat HsdRccHan:WIST strain, 5 /sex/dose *
*in the low dose group (0.25 mg/L) only females were tested; in the mid dose group (1.0 mg/L) only males were tested.
MK 0244 G
SNA6B019/milled
(emamactin benzoate technical; purity 96.2%)
Inhalation (aerosol)
MMAD: 2.7, 3.7, 3.8, and 2.9 resp.; GSD: 1.9, 2.6, 1.7, and 1.6 resp.
4 h; nose only
0, 0.239, 0.506, 1.049, 1.981 mg/L
(achieved conc.)**
** target dose levels were 0.25, 0.5, 1.0, and 2.0 mg/L</td><td><u>Mortality:</u>
<table>
<tr><th>Exposure concentration (mg/L)</th><th>0.239</th><th>0.506</th><th>1.049</th><th>1.981</th></tr>
<tr><td>Males
Females</td><td>n.a.
0</td><td>0
3</td><td>0
n.a.</td><td>4
5</td></tr>
</table>
n.a.: not applicable
<u>Symptoms of toxicity:</u> During exposure all animals of all exposure groups showed salivation, wet fur, associated with restraint and test substance staining around the snout. Reduced response to sound was observed in animals of the 0.506 mg/L dose group and above.
After exposure animals showed next to the symptoms mentioned above, decreased activity, hunched posture, piloerection, reduced response to sound, reduced righting reflex, and shaking for nearly all animals of the high dose group, some males of the 1.049 mg/L dose group, and several females of the 0.506 mg/L dose group. Male animals recovered within 9 days and females within 11 days.
<u>Body weight:</u> During the first week surviving males of the high dose group, several males of the 1.049 mg/L dose group, and the surviving females of the 0.506 mg/L dose group showed weight loss, but gained weight again in the second week.
<u>Pathology:</u> Only stained nostrils in the nasal cavity in one male of the high dose group was considered treatment-related. Other findings were not related to treatment.</td><td>B.6.2.1.3, STUDY 1</td></tr>
<tr><td>Acute toxicity study</td><td>Rat Crl:CD(SD) BR strain, 5 /sex/dose</td><td><u>Mortality:</u> All animals of the high dose group died and 1f and 3m of the 2.12 mg/L dose group. <u>Symptoms of toxicity:</u> Hypersalivation, trembling, lying on one side, ataxia, hypersensitivity to touch and gasping were noted in a few animals of the two low dose groups and in nearly all</td><td>B.6.2.1.3, STUDY 2</td></tr>
</table>

Method, guideline, deviations if any	Species, strain, sex, no/group, test substance, dose levels, duration of exposure	Results[1]	Reference
(no guidance)	MK 0244 L656,748-052S 006 (Benzoate salt; purity 96.9%) Inhalation (aerosol) MMAD:1.2, 4.1, 3.7, and 4.3 resp.; GSD: 3.0, 2.3, 2.0, and 2.1 resp. 4 h; nose only 0, 0.24, 0.44, 2.12, 4.44 mg/L (achieved conc.)* *the achieved concentrations (and also target and nominal concentrations) are expressed as benzoate salt	animals of the two high dose groups. These signs were observed between days 2 and 7. Body weight: There were dose-related transient losses in the bodyweights of the majority of animals exposed to the test substance. In general, animals in the 0.24 and 0.44 mg/L groups had a slight decrease in bodyweight (less than 10%) between days 1 and 2, with the majority subsequently gaining bodyweight. In the two high dose groups, bodyweight losses were generally more severe. Between days 4 and 7, there were increases in the bodyweights of surviving animals in the 2.12 mg/L group. Pathology: No treatment-related gross findings.	
Acute toxicity study (no guideline)	Rat Crl:CD(SD) BR strain, 5 /sex/dose MK 0244 L656,748-052S 006 (Benzoate salt; purity 96.9%) Inhalation (aerosol) MMAD: 3.2, 2.9, and 3.5 resp.; GSD: 2.0, 2.2, and 2.0 resp. 4 h; nose only 0, 0.01, 0.05, 0.1 mg/L (achieved conc.)* *the achieved concentrations were equivalent to the target concentrations. The nominal concentrations were 0, 0.02, 0.08, and 0.17 mg/L. The concentrations are expressed as benzoate salt	Mortality: No mortality. Symptoms of toxicity: Increased activity was observed in some animals of the low dose group and in all animals of the higher dose groups between day 2 and 4 of observation. Body weight: No effects. Pathology: No treatment-related gross findings, and no microscopic changes in brain, spinal cord and sciatic nerve.	B.6.2.1.3, STUDY 3
Acute	MK-0243: L-656,748-	NOAEL: < 31 mg/kg bw/day	B.6.7.1,

Method, guideline, deviations if any	Species, strain, sex, no/group, test substance, dose levels, duration of exposure	Results[1]	Reference
neurotoxicity (no guideline)	010V003 hydrochloride salt, purity: 96.9% Single dose; oral, 0, 27.4, 54.8, and 82.2 mg/kg bw* Rat Crl:CD(SD) Br strain, 10 /sex/dose * dose levels are expressed as base compound (factor 1.04).	LOAEL: 31 mg/kg bw/day Critical effects: clin. signs of neurotox; reduced bw gain; histopathological lesions in brain, spinal cord and sciatic nerve	STUDY 1
Acute neurotoxicity (no guideline)	MK-0243: L-656,748-038W002 benzoate salt Purity: 94.2% Single dose; oral, 0, 0.5, 2.5, 5.0, 10, 25 mg/kg bw* Rat Crl:CD(SD) Br strain, 10 /sex/dose * dose levels are expressed as base compound (factor 1.14).	NOAEL: 5.7 mg/kg bw/day LOAEL: 11.4 mg/kg bw/day Critical effects: clin. signs of neurotox	B.6.7.1, STUDY 2
Acute neurotoxicity	MK-0243: L-656,748-038W, Lot 2 Purity: 94.2% Single dose; dermal (24h), 500, 1000, and 2000 mg/kg bw* Rabbit NZW, 5 f/group * no factor was mentioned in the study report; Other studies with this batch used a conversion factor of 1.14, so it may be assumed that this was used for this study. However, this remains undetermined.	NOAEL: < 500 mg/kg bw/day LOAEL: 500 mg/kg bw/day Critical effects: clin. signs of neurotox; histopathological lesions in brain, spinal cord and sciatic nerve, body weight loss	B.6.7.1, STUDY 3

[1] *Studies were performed expressing the active substance as base compound in the original study reports, but all endpoints were recalculated and expressed as emamectin benzoate (ratio base compound to emamectin benzoate 1.14)*

Table 45: Summary table of human data on STOT SE

No data available.

Table 46: Summary table of other studies relevant for STOT SE

No data available.

10.11.1 Short summary and overall relevance of the provided information on specific target organ toxicity – single exposure

Emamectin benzoate was tested for acute toxicity as three different salts (benzoate, benzoate hydrate and hydrochloride). The salts were found to be of similar acute toxicity. Emamectin benzoate is toxic to mouse and rat by oral administration, with the rat more sensitive than the CD-1 mouse, but less sensitive than the CF-1 mouse. The sensitivity of the CF-1 mouse to emamectin benzoate was also observed in the neurotoxicity tests. Characteristic signs of acute emamectin benzoate toxicity in mice and rats are tremors, ataxia, bradypnoea and decreased activity.

From two single dose oral neurotoxicity studies in rats it can be concluded that mortality occurred at 62.5 mg/kg bw and higher. A NOAEL was established at 5.7 mg/kg bw, based on clinical signs of neurotoxicity. Microscopic lesions in brain, spinal cord and sciatic nerve were observed from 29 mg/kg bw, expressed as emamectin benzoate. In a single dose dermal study in rabbits, one rabbit exposed for 24 hours at 2000 mg/kg was sacrificed due to severe signs of neurotoxicity. A single dose of 500 mg/kg bw applied to the skin during 4 h only, already induced clinical signs of neurotoxicity and substance-related morphological changes in spinal cord and peripheral nerves. The incidence and/or severity of the effects increased at higher dose levels and/or longer exposure duration, when also lesions in brain were observed.

10.11.2 Comparison with the CLP criteria

Clinical (*e.g.* tremors, ataxia and decreased activity) evidence of neurotoxicity occurring at dose levels below 300 mg/kg bw/day were found in the acute toxicity studies. This conclusion is strengthened by the results from acute neurotoxicity studies (oral and dermal). According to the guidance value ranges for single-dose exposures laid down in the CLP criteria (Annex I 3.8.2.1.9.3), this effect should be classified as category 1.

Table 3.8.1, defines specific target organ toxicity single exposure, cat. 1 as follows:

‘Substances that have produced significant toxicity in humans or that, on the basis of evidence from studies in experimental animals, can be presumed to have the potential to produce significant toxicity in humans following single exposure Substances are classified in Category 1 for specific target organ toxicity (single exposure) on the basis of: a. reliable and good quality evidence from human cases or epidemiological studies; or b. observations from appropriate studies in experimental animals in which significant and/or severe toxic effects of relevance to human health were produced at generally low exposure concentrations. Guidance dose/concentration values are provided below (see 3.8.2.1.9) to be used as part of weight-of-evidence evaluation.’

However, it should be noted that Regulation EC No 1272/2008 (CLP), section 3.8.1 states that:

“Acute toxicity refers to lethality and STOT-SE to non-lethal effects. However, care should be taken not to assign both classes for the same toxic effect, essentially giving a “double classification”, even where the criteria for both classes are fulfilled. In such case the most appropriate class should be assigned.“

It should be noted that the classification for acute toxicity (H301, H311 and H331) is already considered for emamectin benzoate. Since it is already proposed to classify emamectin benzoate for acute toxicity on the basis of the LD_{50} studies, no additional classification of emamectin benzoate for Specific Target Organ Toxicity-Single Exposure (STOT-SE) is necessary.

10.11.3 Conclusion on classification and labelling for STOT SE

No classification is proposed.

10.12 Specific target organ toxicity-repeated exposure

Table 47: Summary table of animal studies on STOT RE

Method, guideline, deviations if any, species, strain, sex, no/group	Test substance, route of exposure, dose levels, duration of exposure	Results[1]	Reference
13 weeks, oral, rat Crl:CD[SD] BR, 20/sex/group (OECD 408)	L-656,748-010V003; purity 92.8% B1a and 4.1% B1b 0, 0.5, 2.5 or 12.5/8/5 mg/kg bw/day	NOAEL: 0.57 mg/kg bw/day LOAEL: 2.85 mg/kg bw/day Critical effects: Brain lesions in males Observations: Brain leasions were found in 2/20 males dosed 2.5 mg/kg bw/day. In the 12.5 mg/kg bw dose group, tremors (20/20 in male and female), a marked decrease in weight gain (36% in males and 31% in females), decreases in blood glucose (36% in males and 37% in females) and numbers of leucocytes (23% in males), lymphocytes (24% in males), monocytes (40% in males and 32% in females) and and segmented neutrophils (11% in males and 32% in females) were observed. In the high dose group morphological changes in the brain (15/20 in males and 16/20 in females), spinal cord (neuron vacuolation 20/20 in males and 18/20 in females and degeneration 10/20 in males and 5/20 in females), optic (0/20 in males and 1/20 in females) and sciatic nerves 17/20 in males and 18/20 in females), and bone (17/20 in males and 9/20 in females) and skeletal muscle (20/20 in males and females) were observed.	B.6.3.3, STUDY 1
13-week toxicity study in mice range-finding study for carcinogenicity study Oral Mouse (Crl:CD-1 (ICR) BR strain 15/sex/dose	Repeated by diet, 13 weeks 0, 0.5, 1.5/10, 4.5 and 15 mg/kg bw/day a: a factor of 1.14 was used for calculation of the desired concentrations since MK-0243 was provided as a benzoate salt.	a maximum level of approximately 12.5 mg/kg bw/day expressed as base compound was chosen for a subsequent carcinogenicity study in mice with MK-0243 Critical effects: One male and one female in the highest dose group died. Neither clinical signs nor histopathological changes were observed in these animals. In the highest dose group, reduced body weight gain was observed in both males (-41%) and females (-21%). Decreased plasma glucose values in males and females of the highest dose group and in males of the 5.1 mg/kg bw/day group, expressed as emamectin benzoate. The values of the 1.5/10 mg/kg bw/day group expressed as base compound are considered not representative for 13 weeks exposure to a defined dose level. In males of the highest dose group, a 16% increase of relative (to bw) liver weight was observed.	B.6.3.3, STUDY 2

52 weeks, oral, rat Rat (Crl:CD(SD) BR), 20/sex/dose (no guideline)	MK-0244 (L656,748-052S002; purity 92.5% B1a, 5.3% B1b) Repeated by diet, 52 weeks m: 0, 0.1, 1.0 and 2.5 mg/kg bw/day f: 0, 0.1, 1.0 and 5.0/2.5 mg/kg bw/day	NOAEL: 1.1 mg/kg bw/day LOAEL: 2.85 (m), 5.7/2.85 (f) mg/kg bw/day Critical effects: clinical signs of neurotoxicity (f) and neuronal degeneration in brain and spinal cord; Increased number of males with lower arousal (open field) and increased plasma levels of triglycerides Observations: Clinical signs of neurotoxicity (9/20) and neuronal degeneration in brain (19/20) and spinal cord (2/20) were observed in high dose female rats (5.0/2.5 mg/kg bw/day). At this high dose level, males showed increased plasma triglyceride levels (224% compared to the control), together with a small increase in overall bodyweight gain (7%), and a shift to lower arousal than normal (1/10); females showed increased plasma glucose levels (18%), decrease in grip strength of the forelimb (70% compared to the control in week 14 only) and an increase in centrilobular vacuolation in the liver (6/20).	B.6.3.3, STUDY 3
14 weeks, oral, dog Beagle dog, 4/sex/dose (guideline according to OECD 409	L656,748-010V003 hydrochloride salt; purity 96.6%) Repeated by gavage, 14 weeks Days1-14/15: 0, 0.5, 1.0 and 1.5 mg/kg bw/day Days 14/15-91/92: 0, 0.25, 0.5 and 1.0 mg/kg bw/day	NOAEL: 0.29 mg/kg bw/day LOAEL: 0.6 mg/kg bw/day Critical effects: Brain white matter multifocal degeneration, spinal cord multifocal degeneration, skeletal muscle atropy Observations: Clinical signs of neurotoxicity (tremors 3/4 in males and females, mydriasis 1/4 in males and females, ptyalism 1/4 in males, recumbency 2/4 in males and females and ataxia 3/4 in females), decreased food consumption (42-70% in males and females) and body weight loss (47% difference in body weight gain compared to the control) at a dose level of 1.5 mg/kg bw/day (highest dose group) after approximately two weeks. Morpholocical Microscopic changes were observed in the brain (3/4 in males and females), spinal cord (4/4 in males and females), peripheral (4/4 and 3/4 in males and females) and optic nerves (2/4 and 3/4 in males and females), skeletal muscle (3/4 and 4/4 in males and females), thymus (1/4 and 2/4 in males and females) and bone marrow (1/4 and 2/4 in males and females) of dogs in the highest dose group. The observed slight to severe decrease in the number of erythropoietic cells and thymus atrophy were confined to the animals that were killed during the study (1/4 males and 2/4 females). Reduction of the dose from 1.5 to 1.0 mg/kg bw/day resulted in less severe signs of neurotoxicity since clinical signs were absent. At the 1.0 mg/kg bw/dose level, white matter degeneration in the brain (3/4 in males and 1/4 in females), degeneration of the spinal cord (1/4 in males) and atrophy of skeletal muscle (1/4 in males and females) was observed.	B.6.3.3, STUDY 4

52 weeks, oral, dog Oral, Beagle dog (OECD 452)	MK-0244 (L656,748-038W002, benzoate salt; purity >97%) Repeated by gavage, 52 weeks 0, 0.25, 0.5 and 1.0 mg/kg bw/day additional group of 0.75 mg/kg bw/day	NOAEL: 0.29 mg/kg bw/day LOAEL: 0.6 mg/kg bw/day Critical effects: Clinical signs of neurotoxicity and histological changes in central and peripheral nervous system and in muscle fibre Observations: Clinical signs of neurotoxicity (fine tremors 4/4 in males and females, mydriasis 3/4 in males and 4/4 in females, decreased motor activity 2/4 in males and 4/4 in females, stiffness hindlegs, difficult to get up, ataxia, hyperreaction to touch) and histological changes in central and peripheral nervous system and muscle fibre were observed at 1.0 mg/kg bw/day, together with decreased neutrophil number in blood. The histological changes included brain axonal degeneration (4/4 in males and females), brain neuron focal degeneration (1/4 in males and 2/4 in females), spinal cord axonal degeneration (4/4 in males and females) , nerve axonal degeneration (4/4 in males and females), eye retina cellular degeneration (3/4 in males and females) and eye optic nerve axonal degeneration (4/4 in males and 3/4 in females). In the 1.0 mg/kg bw/day group all animals showed signs of severe toxicity, and were killed after 19 doses. At 0.75 mg/kg bw also clinical signs of neurotoxicity were observed (fine tremors 4/4 in males and 3/4 in females, mydriasis 1/4 in males and females, stiffness hindlegs, difficult to get up 2/4 in females, ataxia 1/4 in females, hyperreaction to touch 1/4 in females). Skeletal muscle degeneration (1/4 in males and 3/4 in females), brain axonal degeneration (4/4 in males and 3/4 in females), brain neuron focal degeneration (2/4 in males), spinal cord axonal degeneration (2/4 in males and 4/4 in females), nerve axonal degeneration (4/4 in males and females), eye retina cellular degeneration (2/4 in males and 1/4 in females) and eye optic nerve axonal degeneration (3/4 in males and 1/4 in females) were observed. At 0.5 mg/kg bw fine tremors (1/4) and difficulty to get up (1/4) in females was observed. Histoilogical changes included skeletal muscle degeneration (1/4 in females), brain axonal degeneration (1/4 in males and 2/4 in females), spinal cord axonal degeneration (2/4 in females) and nerve axonal degeneration (3/4 in males and 1/4 in females).	B.6.3.3, STUDY 5

104 weeks, oral, rat Rat, Sprague-Dawley Crl:CD(SD)BR Test substance: 75/sex/dose Controls: 130/sex (no guideline)	MK-0244 (L-656,748-052S, technical; purity 95.9%) Rat, Sprague-Dawley Crl:CD(SD)BR Test substance: 75/sex/dose Controls: 130/sex	NOAEL: 0.29 mg/kg bw/day LOAEL: 1. 140 mg/kg bw/day Critical effects: Blood triglyceride level in females and relative weights of kidney and liver in males. In high dose animals several parameters were changed, the most prominent effect being vacuolar degeneration of neurons in brain and spinal cord and effects on bodyweight gain. Observations: In high dose animals (5.0/2.5 mg/kg bw), the most prominent effect being vacuolar degeneration of neurons in brain (27/28 in males and 12/23 in females) and spinal cord (15/28 in males and 1/23 in females) and an decrease on bodyweight gain (24% diffenrence in weight gain in males). In females, triglyceride levels were increased (568%). At 1.0 mg/kg bw, effects were observed on female blood triglyceride levels (228% increase compared to the control) and on male relative weights of kidney (27%) and liver (27%).	B.6.5.1, STUDY 1
79 weeks, oral, mouse Mouse, Crl:CD-1 50/sex/dose (no guideline, but comparable to OECD 451))	MK-0244 (L-656,748-052S (Lot #2); purity 97.%) Repeated by diet, 79 weeks m: 0, 0, 0.5, 2.5 and 12.5/7.5/5 mg/kg bw/day f: 0, 0, 0.5, 2.5 and 12.5/7.5 mg/kg bw/day	NOAEL: 2.9 mg/kg bw/day LOAEL: 14.3/ 8.6/ 5.0 (m), 14.3/ 8.6 (f) mg/kg bw/day Critical effects: Increased mortality, marked decreased weight gain, clinical signs of neurotoxicity, changes in haematological parameters and microscopical changes (degeneration sciatic nerve) Observations: Increased mortality, marked decreased weight gain (26% in males and 24% in females), clinical signs of neurotoxicity including tremor (3/50 in males), vocalization (5/50 in males), fine fasciculating tremor of forequarter/ forelimbs (35/50 in males and 45/50 in females) and skin lesions (49% and 6% in controle males and females, repectively, and 84% in males and 74% in females in the high dose group), changes in haematological parameters and increased relative organ weights (kidney 31% and liver 13% in females) observed in high dose mice.	B.6.5.1, STUDY 2

Range-finding study Rat, Sprague-Dawley Crl:CD(SD) Br strain 12 mated females/dose (no guideline)	Orally, by gavage or in the diet from GD 0 to LD 21 Gavage: 0, 0.1, 0.7 and 5.0 mg/kg bw/day a Diet, 0, 1, 7 and 50 ppm, approximately equal to 0, 0.1, 0.7 and 5.0 mg/kg bw/day a	<u>Critical effects:</u> Treatment-related maternal toxicity (body weight loss, decreased food consumption) was observed in the high-dose gavage- and diet-treated groups. In the offspring of the high-dose gavage- and diet-treated groups toxicity was evidenced by clinical signs (tremors) and reduced body weight gains, as well as (gavage only) increased mortality between LD8-15. In pups of the high-dose diet-treated group a reduced brain weight and neuronal degeneration in the brain and spinal cord were additionally observed. Observations: At the highest does levels tested (gavage and dietary treatment group) body weight loss was observed up to 78% difference in body weight gain compared to the control on lactation day 8 and decreased food consumption up to 42% compared to the control at lactation day 12 was observed. Clinical signs (tremors) and reduced body weight gains (up tot 62% compared to the control at lactation day 14) were observed in the offspring in the high dose group (gavage and deietary treatment group). In the gavage treatement group, an increased mortality was observed following exposure to 5.0 mg/kg bw. Tremors were observed at LD11-12 in about one-third of the pups in the gavage treatment group (5.0 mg/kg bw/day). In addition, a few pups were pale, cold, weak and/or breathing shallow. In the dietary treatment group a reduction in post implantation survival was mainly due to 1 female with 100% post implantation loss. This was considered not treatment-related, since post implantation survival in the other high-dose females was comparable to controls. At LD11-12 tremors were observed in all pups of the high-dose group. In addition a few pups in the high dose groups were pale, cold, weak and/or breathing shallow. In 12/16 examined pups neurons within the pons, and spinal gray matter were swollen. The swollen neurons had an increased amount of eosinophilic cytoplasm, central chromatolysis and the nucleus was displayed to the periphery of the cell body.	B.6.6.1, STUDY 1

Dietary 2-generation study of reproductive toxicity (Resembles OECD 416) Rat, Sprague-Dawley Crl:CD(SD) Br strain, 33/sex/dose	MK-0244: L-656,748-052S002 benzoate hydrate salt Purity: >96% by HPLC 0, 0.1, 0.6 and 3.6/1.8 mg/kg bw/day. a,b a Expressed as base (factor 1.15) b The high-dose level was lowered to 1.8 mg/kg bw/day for F0 and F1A females on GD0.	<u>NOAEL:</u> Parental: 0.68 mg/kg bw/day Developmental: 0.68 mg/kg bw/day Offspring: 0.68 mg/kg bw/day <u>LOAEL:</u> Parental: 2.1 mg/kg bw/day Developmental: 2.1 mg/kg bw/day Fertility: 2.1 mg/kg bw/day <u>Critical effects:</u> Reduced bw gain (males), increased body weight gain and food consumption in females during premating, reduced food consumption during lactation (females), neuronal degeneration in brain, spinal cord and (at 3.6 mg/kg bw in males, expressed as emamectin benzoate) sciatic nerve. Clinical signs (tremors, hindlimb splay), reduced bw gain during lactation Reduced fecundity Observations: At the highest dose tested a decrease in body weight gain in F0 males during/after 2nd mating (21%) was observed. In females several effects were observed including an increased body weight gain (16%) and food consumption in females during premating (13%). In the brains and spinal cords of animals of the high dose group very slight to slight degeneration of neurons was observed (brains 29/33 and 23/33 in males and females, respectively, and in spinal cord this was 31/33 and 5/33 for males and females). In the males of the high dose group a few animals (4/33) had very slight degeneration of the sciatic nerve. A slight decrease in fecundity index (pregnant females/mated females) was observed in all treatment groups during the first mating. In the second mating again a decreased fecundity was observed in the high-dose animals. Analysis of the affected sex indicated that approximately 20% of the high-dose females failed to produce a pregnancy after mating occurred. Since a reduced fecundity was also observed in the high-dose group producing the F2 generation, it is considered that the observed reduced fecundity at the high dose is treatment-related.	B.6.6.1, STUDY 2

Oral developmental toxicity study Rat Crl:CD(SD) Br strain 25 mated females/dose (no guideline)	MK-0243: L-656,748-038W002 benzoate salt, Purity: 94.2% orally by gavage; days 6-19 of gestation, 0, 2, 4, and 8 mg/kg bw per day* * a factor 1.14 was used to calculated the dosages as base compound	<u>NOAEL:</u> Maternal: 2.3 mg/kg bw/day Developmental: 2.3 mg/kg bw/day <u>LOAEL:</u> Maternal: 4.6 mg/kg bw/day Developmental: 4.6 mg/kg bw/day <u>Critical effects:</u> Maternal: decreased bw gain; at high dose also clin. signs of neurotox Developmental: incomplete ossification Observations: Maternal toxicity was observed mainly in the high dose group with clinical signs of neurotoxicity (tremors in 15/25 animals) and a decrease in body weight gain (33%). A slight decrease in body weight gain was also observed in the mid dose group (5.2%). At 2 mg/kg bw per day, a dose-related increases in the number of foetuses with incomplete ossification (19/344, 18/315, 27/312 and 46/332 at 0, 2, 4 and 8 mg/kg bw/day, respectively) and in the number of sites with incomplete ossification (20/344, 21/315, 32/312 and 78/332 at 0, 2, 4 and 8 mg/kg bw/day, respectively) was observed. Additionally, in the high dose group a slight decrease in foetal weight (4% in males and 6% in females), an increase in the number of resorptions (400% compared to the control), and an increase in skeletal variations (wavy rib; 0 in control and 5 in high dose group, and supernumary ribs; 36 in control and 73 in high dose group) were observed, of which the dams showed excessive toxicity (reduced bw gain of 33%). There was no evidence of a direct and irreversible teratogenic effect in the rat.	B.6.6.2, STUDY 1

Range finding - Oral developmental toxicity study (no guideline) Rabbit NZW, 51 pregnant females/dose?** ** one female in the 4 mg/kg group was misdosed on GD 6, removed from the study and replaced by another female.	MK 0243; L656,748-038W002, Benzoate salt, purity 96.2% orally by gavage; days 6-18 of gestation, 0, 2, 4, 6 and 8 mg/kg bw per day* * a factor 1.14 was used to calculated the dosages as base compound	NOAEL: Maternal: 4.6 Developmental: 9.1 LOAEL: Maternal: 6.8 Developmental: >9.1 Critical effects: Maternal: decreased bw gain and food consumption; Developmental: no embryo/foetotox. Observations: Treatment-related maternal clinical effects were observed in the high dose group and consisted of tremors (1 female, d19-28), soft and/or small feces (3 females) and no urine on one day in 1 of these females. Moreover, decreased body weight gain and food consumption was observed. Food consumption was decreased in the 6 and 8 mg/kg/d groups on GD 16 and 19 (-11 and -45%), and on GD 22 in the high dose group (-13%). In 2 fetuses from separate litters at 8 mg/kg bw/d, malformations were observed; one fetus had cleft palate and hydrocephaly, the other fetus had hydrocephaly. These were considered to be incidental or secondary to maternal toxicity since these affected fetuses were from dams which showed the greatest body weight losses during the dosing period and/or had tremors.	B.6.6.2, STUDY2a
Oral developmental toxicity study Rabbit NZW, 18 pregnant females/dose	MK 0243; L656,748-038W002 Benzoate salt, purity 94.2% orally by gavage; days 6-18 of gestation, 0, 1.5, 3, and 6 mg/kg bw per day* * a factor 1.14 was used to calculated the dosages as base compound	NOAEL: Maternal: 3.4 mg/kg bw/day Developmental: 6.8 mg/kg bw/day LOAEL: Maternal: 6.8 mg/kg bw/day Developmental: >6.8 mg/kg bw/day Critical effects: Maternal: decreased bw gain; mydriasis; decreased pupillary reaction Developmental: no embryo/foetotox. Observations: Clinical signs of neurotoxicity were not observed in this study. The only clinical signs of maternal toxicity were mydriasis (9/18) and decreased pupillary reaction (16/18) in the high dose group and a decrease in body weight gain (29% difference compared to the control). There was no evidence of a teratogenic effect in the rabbit, and no embryo and/or foetotoxic effects were observed at any dose level.	B.6.6.2, STUDY 2

Sub-acute neurotoxicity Oral, via diet Mouse Crl:CF-1 Br strain 10 /sex/dose (no guideline)	MK-0243: L-656,748-038W002 Purity: 96.9% [a] 16 days 0, 0.05, 0.10, 0.30, and 0.90 mg/kg bw per day* a L-656,748-038W002: 96.9% epi-methylamino avermectin (B1a + B1b’) and 92.1% epi-amino avermectin (B1a) * no factor is mentioned in the study report;Other studies with this batch used a conversion factor of 1.14, so it may be assumed that this was used for this study. However, this remains undetermined.	NOAEL: 0.1 mg/kg bw per day LOAEL: 0.3 mg/kg bw/day Critical effects: Clinical signs of neurotoxicity were observed at 0.3 and 0.9 mg/kg bw per day, and four mice in each of these dose groups were killed in moribund condition. No clinical signs of neurotoxicity or reduced body weight gain were observed in CF-1 mice dosed up to and including 0.1 mg/kg bw per day. Observations: In the high dose, 4 animals were sacrificed due to severe whole body tremor (one male), decreased motor activity, slow righting reflex, and slow, irregular breathing (one female) or due to tremors and became moribund (two females). In the mid dose group, mild tremors were observed in 6 animals, 4 of which became moribund. In the mid dose group mean weight gain decreased (18% difference compared to the control group), caused by a severe weight loss of 9.8 g observed in one animal showing tremors.	B.6.7.2, STUDY 1
Neurotoxicity study, 2 weeks; oral Oral, via diet Mouse Crl:CD-1(ICR) Br strain 10 /sex/dose	MK-0243: L-656,748-010V003 Hydrochloride salt Purity: 96.1% a 2 weeks 0, 0.2, 0.6, 1.2, and 2.0 mg/kg bw per day* a L-656,748-010V003: 96.1% B1a + B1b (with 0.76% propylgallate added as an oxidant) * dose levels are expressed as base compound (factor 1.04).	NOAEL: 2.3 mg/kg bw/day LOAEL: > 2.3 mg/kg bw/day Critical effects: - Observations: Body weight gain was increased in males in the 0.6 and 1.2 mg/kg groups (30% and 70%, respectively), but not in males at higher or lower dose levels or in females at any dose level. No test substance-related gross or microscopic lesions were apparent in any of the tissues examined from the high dose group. However, one high dose male had a slight, cellular infiltrate in the meninges of the brain. This probably indicates an asymptomic viral infection, occassionally seen in control mice in the laboratory in question.	B.6.7.2, STUDY 2
Neurotoxicity study, 5 weeks; oral (dog) Oral, gavage Beagle dog, 1-3/sex/dose (no guideline)	MK-0243: L-656,748-010V003 Hydrochloride salt Purity: 96.9% [a] 5 weeks 0, 0.5, and 1.5 mg/kg bw per day* a L-656,748-010V003: purity 96.6% with 0.76% propylgallate added as an oxidant * dose levels are expressed as base compound (factor 1.04).	NOAEL: 0.6 mg/kg bw/day LOAEL: 1.7 mg/kg bw/day Critical effects: clin. signs of neurotox; weight loss; histomorphological alterations in central and peripheral nerve tissue Observations: Occurrence of weight loss, reduced food consumption, clinical signs of neurotoxicity and histomorphological alterations in central and peripheral nerve tissues were observed in all dogs dosed at 1.5 mg/kg bw per day. No effects were observed in dogs dosed at 0.5 mg/kg bw per day.	B.6.7.2, STUDY 3

14 weeks; oral (Rat (CD)) Oral, via diet Rat Crl:CD(SD) Br strain 10/sex/dose	MK-0244: L-656,748-052S002 Purity: 95.9% 14 weeks 0, 0.25, 1.0 and 5.0 mg/kg bw per day* * all dose levels are expressed as base compound (factor 1.15).	<u>NOAEL:</u> 1.1 mg/kg bw/day <u>LOAEL:</u> 5.7 mg/kg bw/day <u>Critical effects:</u> clin. signs of neurotox (incl. FOB test); decreased bw gain in males; histopathological lesions in brain, spinal cord and sciatic nerve Observations: At a dose level of 5 mg/kg per day both clinical and histopathological evidence of neurotoxicity in both male and female rats (mild tremors (8/10 males), effects observed in the FOB, decreases in bodyweight gain (25%) in male rats, neuronal vacuolation in the brain (6/6 males and females) and spinal cord (6/6 males and females), degeneration of nerve fibres in the spinal cord (6/6 in males and 4/6 in females) and sciatic nerve (6/6 males and 1/6 females), and skeletal muscle atrophy in 3/7 high dose males) with, in general, more pronounced effects observed in males. No clinical or histopathological evidence of neurotoxicity was observed at lower dose levels tested in this study (0.25 and 1 mg/kg per day).	B.6.7.2, STUDY 4
Developmental neurotox. Study Rat (CD) Oral range-finding reproduction study in female rats Rat Crl:CD(SD) Br strain 25 f/dose? (no guideline)	MK-0244: L-656,748-052S002 Purity: >97% MK-0244: L-656,748-052S002 Purity: >97%	<u>NOAEL:</u> mat: 2.85 mg/kg bw/day prog: 0.68 mg/kg bw/day <u>LOAEL:</u> mat: >2.85 mg/kg bw/day prog: 2.85 mg/kg bw/day <u>Critical effects:</u> Mat: - Prog: clinical signs of neurotox; growth retardation; neurobehavioural effects Observations: Clinical signs pre-weaning included intermittent head tremors (10/25), intermittent body tremors (23/25), whole body tremors (25/25), hind limb extension (25/25) and hind limb splay (25/25). Post-weaning these effects were seen in much less animals. Moreover, growth retardation (decrease of 18% and 17% body weight gain in males and females, respectively) and alterations of neurobehavioural function in the F1 progeny of females administered 2.85 mg/kg bw/day.	B.6.7.3, STUDY 1

[1] Studies were performed expressing the active substance as base compound in the original study reports, but all endpoints were recalculated and expressed as emamectin benzoate (ratio base compound to emamectin benzoate 1.14)

Table 48: Summary table of human data on STOT RE

No data available.

Table 49: Summary table of other studies relevant for STOT RE

No data available.

10.12.1 Short summary and overall relevance of the provided information on specific target organ toxicity – repeated exposure

Subacute and subchronic toxicity

There were no acceptable subacute studies. Data on semichronic oral exposure of rats (13- and 52-week dietary studies) and of dogs (14- and 52-week gavage studies) were available.

In rats, administration of emamectin-HCl at a dose levels of 0, 0.5, 2.5 and 12.5/8/5 mg/kg bw/day, expressed as base compound in the study report, during 13 weeks resulted in tremors and a marked decrease in weight gain in rats of the highest dose group, together with reductions in blood glucose and in numbers of leucocytes, lymphocytes, monocytes and segmented neutrophils (B.6.3.3, STUDY 1). Reduction of the dose level to 8.0 and then to 5.0 mg/kg bw/day resulted in some improvement, although the signs of toxicity persisted in most of the affected animals. The observed signs of neurotoxicity in rats of the highest dose group were accompanied by morphological changes in the brain, spinal cord, optic and sciatic nerves, and bone and skeletal muscle. Although animals at the mid dose (2.5 mg/kg bw/day) did not display signs of neurotoxicity, 2 males had similar lesions in the brain as animals in the high dose group. The NOAEL in this study was 0.6 mg/kg bw/day, expressed as emamectin benzoate.

Exposure of rats during 52 weeks to emamectin-benzoate hydrate at dose levels of 0, 0.1, 1.0 and 2.5 (m) or 5/2.5 (f), expressed as the base compound, resulted in clinical signs of neurotoxicity (f) and neuronal degeneration in brain and spinal cord in high dose rats (B.6.3.3, STUDY 3). At this high dose level, males showed increased plasma triglyceride levels, together with a small increase in overall bodyweight gain, and a shift to lower arousal than normal; females showed increased plasma glucose levels, decrease in grip strength of the forelimb (in week 14 only) and an increase in centrilobular vacuolation in the liver. Based on these effects in the high dose group, the NOAEL in this study is 1.1 mg/kg bw/day, expressed as emamectin benzoate.

Administration of emamectin-HCl at dose levels of 0, 0.5, 1.0, 1.5 (days 1-14) and 0, 0.25, 0.5, 1.0, expressed as base compound, (days 15-91) during 14 weeks to dogs resulted in clinical signs of (neuro)toxicity (such as tremors, mydriasis, ataxia), decreased food consumption and body weight loss at a dose level of 1.5 mg/kg bw/day (highest dose group) after approximately two weeks (B.6.3.3, STUDY 4). Reduction of the dose from 1.5 to 1.0 mg/kg bw/day resulted in less severe signs of neurotoxicity. Morphological lesions in the brain, spinal cord, and skeletal muscle were observed at the mid and high dose, resulting in a NOAEL of 0.29 mg/kg bw/day for this study, expressed as emamectin benzoate.

Following oral administration of emamectin-benzoate to dogs for 52 weeks to dose levels of 0, 0.25, 0.5, 0.75 and 1.0 mg/kg bw/day, clinical signs of (neuro)toxicity (such as tremors, mydriasis, decreased motor activity) and histological changes in central and peripheral nervous system and muscle fibre were observed at and above 0.5 mg/kg bw/day, together with decreased neutrophil numbers in blood (B.6.3.3, STUDY 5). Sub-chronic exposure of dogs to emamectin-HCl or emamectin-benzoate resulted in a NOAEL of 0.29 mg/kg bw/day, expressed as emamectin benzoate.

Overall, a NOAEL for subchronic exposure is 0.29 mg/kg bw/day.

Long term toxicity

A long-term oral toxicity/carcinogenicity study with rats (0, 0.25, 1.0 and 5.0/2.5 mg/kg bw/day during 104 weeks) and an oral carcinogenicity study with mice (0, 0.5, 2.5 and 12.5/7.5/5.0 (m) or 12.5/7.5 (f) mg/kg bw/day during 79 weeks) were performed (B.6.5.1, STUDY 1 and B.6.5.1, STUDY 2). Both species were administered emamectin benzoate hydrate via the diet.

In the study with rats (B.6.5.1, STUDY 1), several parameters were changed in high dose animals, the most prominent effect being vacuolar degeneration of neurons in brain and spinal cord and effects on bodyweight gain. Based on effects observed on female blood triglyceride levels and on male relative weights of kidney and liver at and above 1.0 mg/kg bw/day, the NOAEL in this study is 0.29 mg/kg bw/day, expressed as emamectin benzoate. In this study with rats, no substance-related increase in tumours was observed.

Based on increased mortality, marked decreased weight gain, clinical signs of neurotoxicity (tremors), increased incidence of skin lesions, changes in haematological parameters and increased relative organ weights observed in high dose mice, the NOAEL in this study is 2.85 mg/kg bw/day, expressed as emamectin benzoate (B.6.5.1, STUDY 2). No treatment-related increase in tumour incidence was observed in mice.

Overall, the NOAEL for long term oral exposure to emamectin benzoate is 0.29 mg/kg bw/day. There was no evidence of carcinogenicity in either the rat or the mouse at any of the dose levels employed.

Reproduction toxicology

In a 2-generation study of reproductive toxicity, emamectin benzoate hydrate was administered via the diet to rats at doses of 0, 0.1, 0.6 and 3.6/1.8 mg/kg bw/day (B.6.6.1, STUDY 2). The NOAEL for parental toxicity was 0.68 mg/kg bw/day, expressed as emamectin benzoate, on the basis of a reduced body weight gain in F0 males during/after 2nd mating, an increased bw gain and food consumption in females during premating, a reduced food consumption in females during lactation, and neuronal degeneration in the brain, spinal cord and (3.6 mg/kg bw/day males only) sciatic nerve, observed at 3.6/1.8 mg/kg bw/day.

The NOAEL for offspring toxicity was 0.68 mg/kg bw/day on the basis of clinical signs (tremors, hind limb splay) and reduced body weight gain during the lactation period.

Teratogenicity

In an oral developmental study in rats given emamectin benzoate salt by gavage at doses of 0, 2, 4 or 8 mg/kg bw/day, the NOAEL for maternal toxicity is established at 2 mg/kg bw per day, based on decreases in body weight gain in the mid and high dose group and clinical signs of neurotoxicity in the high dose group (B.6.6.2, STUDY 1). In a second oral developmental study, rabbits were administered emamectin benzoate by gavage at doses of 0, 1.5, 3 or 6 mg/kg bw/day. A decrease in body weight gain, mydriasis and decreased pupillary reaction were observed in the high dose group, but no clinical signs of neurotoxicity.

Neurotoxicity

Acute and semi-chronic oral neurotoxicity studies were performed with rats (B.6.7.1, STUDY 1; B.6.7.1, STUDY 2; B.6.7.2, STUDY 4; B.6.7.3, STUDY 1), and sub-acute oral neurotoxicity studies with mice and dogs (B.6.7.2, STUDY 1; B.6.7.2, STUDY 2 and B.6.7.2, STUDY 3). All species were vulnerable to the neurotoxic effects of the compound. The most remarkable effects observed were clinical signs of neurotoxicity like tremors and ataxia, and microscopic lesions (white matter and/or neuronal degeneration) in brain, spinal cord and sciatic nerve. The CF-1 mouse was the most vulnerable species, with a NOAEL of 0.11 mg/kg bw per day (16 day exposure study), followed by dogs, rats and CD-1 mice.

From two single dose oral studies in rats it can be concluded that mortality occurred at 54.8 mg/kg bw and higher. A NOAEL was established at 5.7 mg/kg bw, expressed as emamectin benzoate, based on clinical signs of neurotoxicity. Microscopic lesions in brain, spinal cord and sciatic nerve were observed from 29 mg/kg bw, expressed as emamectin benzoate.

In a sub-acute oral exposure study (16 days; emamectin benzoate) in the CF-1 mouse doses of 0.3 and 0.9 mg/kg bw per day caused clinical signs of neurotoxicity (at 0.9 mg/kg bw/day already after the first day of dosing) and mortality (some animals were killed in moribound condition), but no microscopic lesions (B.6.7.2, STUDY 1). The NOAEL for CF-1 mice was 0.11 mg/kg bw per day. In contrast, in a sub-acute oral exposure study (2 weeks; emamectin HCL) in the CD-1 mouse (B.6.7.2, STUDY 2), no clinical signs of neurotoxicity, nor any other effects, were observed at doses up to and including 2.28 mg/kg bw per day, expressed as emamectin benzoate, the highest dose tested.

In a sub-acute oral exposure study (5 weeks; emamectin HCl) in dogs, occurrence of weight loss, reduced food consumption, clinical signs of neurotoxicity (observed from the second week of dosing) and histomorphological alterations in central and peripheral nerve tissues were observed at 1.5 mg/kg bw per day (B.6.7.2, STUDY 3). No effects were observed in dogs dosed at 0.6 mg/kg bw per day, expressed as emamectin benzoate.

Dietary administration of emamectin benzoate hydrate to rats during 14 weeks at a dose level of 5 mg/kg per day produced both clinical and histopathological evidence of neurotoxicity in both male and female rats (mild tremors, effects observed in the FOB, decreases in bodyweight gain in male rats, neuronal vacuolation in the brain and spinal cord, degeneration of nerve fibres in the spinal cord and sciatic nerve, and skeletal muscle atrophy in some high dose males) with, in general, more pronounced effects observed in males (B.6.7.2, STUDY 4). The first clinical signs of neurotoxicity were observed in week 7. No clinical or histopathological evidence of neurotoxicity was observed at lower levels tested in this study (0.25 and 1.0 mg/kg bw/day). The NOAEL for neurotoxicity was 1.1 mg/kg bw per day, expressed as emamectin benzoate.

A developmental neurotoxicity study was conducted with rats given emamectin benzoate hydrate by gavage at doses of 0, 0.1, 0.6 or 3.6/2.5 mg/kg bw/day (B.6.7.3, STUDY 1). The NOAEL for developmental neurotoxicity was established as 0.68 mg/kg bw per day, expressed as emamectin benzoate, based on the occurrence of clinical evidence of neurotoxicity, growth retardation and alterations of neurobehavioural function in the F1 progeny of females administered emamectin at 2.5 mg/kg bw per day during the period of gestation (day 6) through lactation (day 20). In the pups, no histopathological evidence of neurotoxicity was observed. In the absence of evidence of toxic effects in dams, the NOAEL for maternal toxicity was 2.85 mg/kg bw/day, the highest dose tested.

In a single dose dermal study in rabbits, one rabbit exposed for 24 hours at 2000 mg/kg was sacrificed due to severe signs of neurotoxicity (B.6.2.1.2, STUDY 1). A single dose of 500 mg/kg bw applied to the skin during 4 h only, already induced clinical signs of neurotoxicity and substance-related morphological changes in spinal cord and peripheral nerves. The incidence and/or severity of the effects increased at higher dose levels and/or longer exposure duration, when also lesions in brain were observed. Skin irritation was also observed in several animals, but there was no dose relation.

Relevance of p-glycoprotein polymorphism

It is likely that the entire series of avermectins share a common mode of action. The avermectins increase membrane permeability to chloride ions and act as GABA agonists. The severity of neurological effects will depend largely on the concentrations of avermectins attained in the target organs/tissues of the CNS and to a lesser extent the PNS.

It has been demonstrated previously (see DAR and specifically the final addendum for abamectin (Febr. 2008)) that increased sensitivity for avermectin toxicity is related to reduced P-glycoprotein expression by the mdr-1 gene. This was demonstrated for CF-1 mice and neonatal rats. CF-1 mice have reduced P-glycoprotein expression and increased sensitivity for avermectin toxicity compared to CD-1 mice. From the few studies available with emamectin benzoate tested in CF-1 mice, CF-1 mice indeed seemed more sensitive than CD-1 mice.

In rats, expression of P-glycoprotein in the brain develops to adult levels during the first 20 days after birth, and the expression of P-glycoprotein in the jejenum does not start before postnatal day 8. Due to neonatal rats having limited P-glycoprotein expression until 20 days after birth they have an increased susceptibility for emamectin benzoate toxicity. Since this susceptible period with limited P-glycoprotein expression after birth is not present in humans, effects observed in neonatal rats during lactation, as also observed after emamectin benzoate administration, are considered not relevant for human risk assessment.

The role of p-glycoprotein polymorphism in abamectin (and thus also emamectin benzoate) toxicity has been discussed in PRAPeR 39 (10-13 December 2007). Based on a recent publication, the experts agreed to the conclusions with regard to the relevance of P-glycoprotein polymorphism in mice as presented by the RMS in the abamectin addendum (Febr. 2008), and that therefore the

studies with the sensitive CF-1 mouse are not relevant for human risk assessment. They furthermore agreed that the findings in young rats are not relevant for human risk assessment.

The notifier recently conducted a single dose oral kinetic study in wild type (+/+) and p-glycoprotein mutant (-/-) CF-1 mice to investigate the concentrations of radiolabelled ivermectin, abamectin and emamectin benzoate in brain and plasma (see B.6.8.2). The results showed that brain concentrations of emamectin benzoate were about 150-fold higher in p-glycoprotein mutant (-/-) mice compared with (+/+) mice, and that the differences observed were comparable to those seen for both abamectin and ivermectin. These results provide good indirect evidence that emamectin benzoate, as expected, has similar p-glycoprotein substrate specificity to that of ivermectin and abamectin. The conclusions on abamectin with regard to the relevance of the sensitive CF-1 mouse are therefore also applicable to emamectin benzoate.

Conclusion

Most characteristic for the toxicity of emamectin benzoate is the clinical and histopathological evidence of neurotoxicity, with tremors and neuronal degeneration in brain and spinal cord observed in the majority of toxicity studies which also showed a very steep dose-response curve for these effects.

At higher dose levels, effects on bodyweight (gain) were observed, as well as effects on levels of triglycerides and/or glucose in blood. These findings were not consistent between males and females, nor between studies, but may indicate slight perturbations to lipid and energy metabolism. Based on the emamectin benzoate mechanism (GABA agonist), interference with the energy metabolism can be expected, but it is clear from the dossier that effects on energy metabolism are not the critical effect of emamectin benzoate.

The relevance for findings in animal studies for human risk assessment is also taken into account.

It has been demonstrated previously (see DAR and specifically the final addendum for abamectin (Febr. 2008)) that increased sensitivity for avermectin toxicity is related to reduced P-glycoprotein expression by the mdr-1 gene. This was demonstrated for CF-1 mice and neonatal rats. CF-1 mice have reduced P-glycoprotein expression and increased sensitivity for avermectin toxicity compared to CD-1 mice.

From the few studies available with emamectin benzoate tested in CF-1 mice, CF-1 mice indeed seemed more sensitive than CD-1 mice.

The role of p-glycoprotein polymorphism in abamectin (and thus also emamectin benzoate) toxicity has been discussed in PRAPeR 39 (10-13 December 2007). Based on a recent publication, the experts agreed to the conclusions with regard to the relevance of P-glycoprotein polymorphism in mice as presented by the RMS in the abamectin addendum (Febr. 2008), and that therefore the studies with the sensitive CF-1 mouse are not relevant for human risk assessment. They furthermore agreed that the findings in young rats are not relevant for human risk assessment. These conclusions are also applicable to emamectin benzoate.

Table 66: Extrapolation of equivalent effective dose for toxicity studies of greater or lesser duration than 90 days

Study reference	Effective dose (mg/kg/d)	Length of exposure	Equivalent guidance values	Classification supported by the study
B.6.3.3, STUDY 1	LOAEL: 2.85 mg/kg bw/day Critical effects: Brain lesions in males observed during (histo)pathology at study termination	90-days, oral	≤ 10 mg/kg bw/day (cat. 1) ≤ 100 mg/kg bw/day (cat. 2)	Yes (cat. 1)

Study reference	Effective dose (mg/kg/d)	Length of exposure	Equivalent guidance values	Classification supported by the study
B.6.3.3, STUDY 3	LOAEL: 2.85 (m), 5.7/2.85 (f) mg/kg bw/day Critical effects: clinical signs of neurotoxicity (f) (from week 3) and neuronal degeneration in brain and spinal cord (observed during (histo)pathology at study termination); Increased number of males with lower arousal (open field, observed from week 14) and increased plasma levels of triglycerides	1 year, oral	≤ 2.5 mg/kg bw/day (cat. 1), calculated according to Haber's rule ≤ 25 mg/kg bw/day (cat. 2), calculated according to Haber's rule	Yes (cat. 1)
B.6.3.3, STUDY 4	LOAEL: 0.6 mg/kg bw/day Critical effects: Brain white matter multifocal degeneration, spinal cord multifocal degeneration, skeletal muscle atropy (observed during (histo)pathology at study termination) Clinical signs of neurotoxicity occurred after approximately two weeks of dosing.	14 weeks, oral	≤ 10 mg/kg bw/day (cat. 1) ≤ 100 mg/kg bw/day (cat. 2)	Yes (cat. 1)
B.6.3.3, STUDY 5	LOAEL: 0.6 mg/kg bw/day Critical effects: Clinical signs of neurotoxicity (apparent from week 2) and histological changes in central and peripheral nervous system and in muscle fibre (observed during (histo)pathology at study termination)	1 year, oral	≤ 2.5 mg/kg bw/day (cat. 1), calculated according to Haber's rule ≤ 25 mg/kg bw/day (cat. 2), calculated according to Haber's rule	Yes (cat. 1)
B.6.5.1, STUDY 1	LOAEL: 1.1 mg/kg bw/day Critical effects: Blood triglyceride level in females (most prominent at week 52, 79 and 105) and relative weights of kidney and liver in males (observed during necropsy at study termination). In high dose animals several parameters were changed, the most prominent effect being vacuolar degeneration of neurons in brain and spinal cord (observed during terminal microscopic examination) and effects on bodyweight gain (observed after the first weeks of the study).	2-year	≤ 1.25 mg/kg bw/day (cat. 1), calculated according to Haber's rule ≤ 12.5 mg/kg bw/day (cat. 2), calculated according to Haber's rule	Yes (cat. 2)
B.6.5.1, STUDY 2	LOAEL: 14.3/ 8.6/ 5.7 (m), 14.3/ 8.6 (f) mg/kg bw/day Critical effects: Increased mortality (week 3-11 and after week 51), marked decreased weight gain (during the first year of the study), clinical signs of neurotoxicity (from week 5 on), changes in haematological parameters and microscopical changes (degeneration sciatic nerve)	1.5 year, oral	≤ 1.6 mg/kg bw/day (cat. 1), calculated according to Haber's rule ≤ 16.0 mg/kg bw/day (cat. 2), calculated according to Haber's rule	Yes (cat. 2)
B.6.6.1,	LOAEL:		≤ 30 mg/kg bw/day (cat. 1)	Yes (cat. 1)

Study reference	Effective dose (mg/kg/d)	Length of exposure	Equivalent guidance values	Classification supported by the study
STUDY 2	Parental: 1.8 mg/kg bw/day Developmental: 1.8 mg/kg bw/day Fertility: 1.8 mg/kg bw/day Critical effects: Reduced bw gain (males), increased body weight gain and food consumption in females during premating, reduced food consumption during lactation (females), neuronal degeneration in brain, spinal cord and (at 3.6 mg/kg bw in males) sciatic nerve (observed during (histo)pathology at study termination).		≤ 300 mg/kg bw/day (cat. 2)	
B.6.6.2, STUDY 1	LOAEL: Maternal: 4.56 mg/kg bw/day Developmental: 4.56 mg/kg bw/day Critical effects: Maternal: decreased bw gain (from day 6 of gestation); at high dose also clinical signs of neurotoxicity (starting from day 10). Developmental: incomplete ossification	19 days	≤ 47 mg/kg bw/day (cat. 1), calculated according to Haber's rule ≤ 470 mg/kg bw/day (cat. 2), calculated according to Haber's rule	Yes (cat. 1)
B.6.6.2, STUDY2a	LOAEL: Maternal: 6.8 mg/kg bw/day Developmental: >9.1 mg/kg ba/day Critical effects: Maternal: decreased bw gain (days 14-19) and food consumption (day 16-22). Developmental: no embryo/foetotox.	18 days	≤ 50 mg/kg bw/day (cat. 1), calculated according to Haber's rule ≤ 500 mg/kg bw/day (cat. 2), calculated according to Haber's rule	Yes (cat. 1)
B.6.6.2, STUDY 2	LOAEL: Maternal: 6.8 mg/kg bw/day Developmental: >6.8 mg/kg bw/day Critical effects: Maternal: decreased bw gain (days 6-28); mydriasis (from day 11 and up); decreased pupillary reaction (between 11 and 23 of gestation) Developmental: no embryo/foetotox.	18 days	≤ 47 mg/kg bw/day (cat. 1), calculated according to Haber's rule ≤ 470 mg/kg bw/day (cat. 2), calculated according to Haber's rule	Yes (cat. 1)

10.12.2 Comparison with the CLP criteria

Category 1 (H372):

Substances that have produced significant toxicity in humans or that, on the basis of evidence from studies in experimental animals, can be presumed to have the potential to produce significant toxicity in humans following repeated exposure.

Substances are classified in Category 1 for target organ toxicity (repeat exposure) on the basis of:

reliable and good quality evidence from human cases or epidemiological studies; or observations from appropriate studies in experimental animals in which significant and/or severe toxic effects, of relevance to human health, were produced at generally low exposure concentrations.

Category 2 (H373)

Substances that, on the basis of evidence from studies in experimental animals, can be presumed to have the potential to be Harmful to human health following repeated exposure.
Substances are classified in Category 2 for target organ toxicity (repeat exposure) on the basis of observations from appropriate studies in experimental animals in which significant toxic effects, of relevance to human health, were produced at generally moderate exposure concentrations.

Regarding classification with Category 1 adverse findings were observed at the dose levels relevant for classification with Category 1 as presented in table 66 above. These include neurotoxicity occurring in different species (rat, mice and dog) and were observed in studies with different study durations. In the sub-acute study in dog clinical signs of neurotoxicity were observed from the second week of dosing. Also in the 1-year study in dogs, neurotoxicity was apparent from week 2. When emamectin benzoate hydrate was administered to rats during 14 weeks the first clinical signs of neurotoxicity were observed in week 7. In the 52-week toxicity study in rats, clinical signs of neurotoxicity were observed from week 3. In mice, clinical signs of toxicity were observed from week 5 on when emamectin benzoate was tested in a carcinogenicity study in mice. In the developmental toxicity study in rat, maternal toxicity was observed mainly in the high dose group with clinical signs of neurotoxicity starting from day 10. These studies thus suggest that the neurotoxic effects can be considered chronic effects. Nevertheless, based on two single dose oral studies in rats clinical signs of neurotoxicity were already observed in the first week after administration. In fact, signs of neurotoxicity were even evident from 5 hours following administration. However, the acute effects were observed following treatment with relatively high doses of emamectin benzoate (i.e.10 mg/kg bw and up) while the chronic effects occurred at lower dose levels (i.e. 0.5 mg/kg bw/day and up). The neurotoxic effects observed after single and repeated exposure are thus not caused by similar emamectin benzoate dose levels. Classification for STOT RE cat. 1 is proposed for neurotoxicity.

Most characteristic for the toxicity of emamectin benzoate is the clinical and histopathological evidence of neurotoxicity, with tremors and neuronal degeneration in brain and spinal cord observed in the majority of toxicity studies which also showed a very steep dose-response curve for these effects. At higher dose levels, also effects on levels of triglycerides and/or glucose in blood were found. These effects were found at levels that trigger STOT RE cat. 1 classification. These findings were not consistent between males and females, nor between studies, but may indicate slight perturbations to lipid and energy metabolism. Based on the emamectin benzoate mechanism (GABA agonist), interference with the energy metabolism can be expected, but it is clear from the dossier that effects on energy metabolism are not the critical effect of emamectin benzoate since they were found at higher dose levels than the dose levels causing neurotoxicity and considering the lack of consistence between studies and sexes within studies. Therefore, classification for STOT RE is not warranted for these effects.

10.12.3 Conclusion on classification and labelling for STOT RE

Proposal for classification with STOT-RE 1, H372 (“Causes damage to the nervous system through prolonged or repeated exposure”).

10.13 Aspiration hazard

No data available.

10.13.1 Short summary and overall relevance of the provided information on aspiration hazard

No data available.

10.13.2 Comparison with the CLP criteria

Not relevant.

10.13.3 Conclusion on classification and labelling for aspiration hazard

No classification proposed due to lacking data.

11 EVALUATION OF ENVIRONMENTAL HAZARDS

In the following summaries, the abbreviation for the test substance, refers to emamectin benzoate (4’’-deoxy-4’’-epimethylamino avermectin B1 benzoate, NOA 405626, MAB1) a derivative of abamectin, a naturally occurring macrocyclic lactone produced by the actinomycete *Streptomyces avermitilis*.

It should be noted that emamectin is the name as it is placed (see Regulation (EC) No. 828/2013 and Review report SANCO / 11454/2013 rev 2 dated 16 July 2013). Regulation (EC) No. 828/2013 states that this is the benzoate variant and also in the DAR of emamectin the following is stated *'the specification of the purity of technical emamectin is benzoate: minus 950 g / kg (=min. 900 g / kg emamectin B1a benzoate and max. 70 g / kg emamectin B1b benzoate).*’ The studies included in the DAR are performed with emamectin benzoate. Thus, the substance as referred to in the current CLH report and the placement in Annex I refer to the same substance. Emamectin benzoate is the substance as produced and is the variant used in the formulations available on the market based on this active substance.

The environmental hazards of emamectin benzoate were assessed in the Draft Assessment Report (2011), addenda and Proposed Decision of the Netherlands prepared in the context of the approval (Reg. (EU) No. 828/2013), under Reg. (EC) 1107/2009. Studies considered valid in the DAR (reliability score of 1 or 2) have been included in this report and were considered for classification purposes. The study summaries as presented in the DAR are included in Annex 1. All studies were carried out under GLP unless indicate otherwise. Studies were carried out in accordance with relevant test guidelines. Minor deviations were noted in some cases which have been included in the study summaries below. The deviations did not affect the overall acceptability of the studies.

11.1 Rapid degradability of organic substances

Table 50: Summary of relevant information on rapid degradability

Method	Test material	Results	Remarks	Reference
Ready biodegradability activated sludge (OECD 301 F; 92/69/EEC, L383 A, C.4-D)	MK 244 (emamectin benzoate), batch nr. FL 971780, chemical purity 96.1 %, white powder	emamectin benzoate B1a is not readily biodegradable	0% degradation after 28 days	Dietschy, A. (1999) STUDY IIA, 7.7/01
Hydrolysis (OECD 111; US-EPA Subdivision N, 540/09-82-021, section 161-1 BBA 55, I and II)	[3,7,11,13,23-14C]4”-deoxy-4”-epimethylamino avermectin B1a benzoate; batch nr. L-683,825-003E001; radiochemical purity 93.6%	no hydrolysis of MAB1a at pH 5.2, 6.2, 7 and 8 pH 9 = DT_{50} of 19.5 weeks at 20°C and 29.1 days 25 °C	-	A.C. Chukwudebe (1992) STUDY IIA, 7.5.1/01

Method	Test material	Results	Remarks	Reference
Degradation in water-sediment systems, aerobic (OECD Guideline 308)	[23 14C]-emamectin benzoate B1a, batch CDC-XV-21-1, radiochemical purity > 93.4%, appearance unknown	$DT_{50,water}$ 8.7 days for silt loam and sand $DT_{50,system}$ > 120 days	DT50,sediment could not be calculatated as there appears to be no degradation in the sediment.	Hurt, AD, Grosjean, J, Mason, G (2006) STUDY IIA 7.8.3/001 (IIA, 7.2.1.3.2/001)
Degradation in soil - Aerobic degradation (95/36/EC amending Council Directive 91/414/EEC, SETAC-EUROPE Procedures Section 1.1, OECD 307)	[14C]-NOA426007 (emamectin benzoate B1a), radiochemical purity > 98%	DT_{50} values (20 °C): sandy clay loam = 45.9 d loam = 25.2 d silty clay loam = 414 d	The DT50 value for silty clay loam is an extrapolated value; at the end of the test 76.7% of AR was still present as emamectin benzoate B1a. Visual inspection of the SFO fit and distribution of residuals showed adequate fitting results.	Hand, L.H. and Fleming, E.A. (2006) STUDY IIA, 7.1.1/001
Degradation in soil - Aerobic degradation (95/36/EC amending Council Directive 91/414/EEC, SETAC-EUROPE Procedures Section 1.1, OECD 307)	[14C]-NOA426007 (emamectin benzoate B1a), radiochemical purity > 99.9%	DT_{50} (19.6 ± 0.5 °C) silt loam = 32.4-98.1 d	A lower moisture content of the soil resulted in a higher DT_{50} value. There was no qualitative effect of the lower soil moisture content.	Jungmann V., Nicollier, G. (2006) STUDY IIA, 7.1.1/002
Degradation in soil - Aerobic degradation (Subdivision N, Section 162-1, 1982)	[23-14C]-NOA426007 (emamectin benzoate B1a), radiochemical purity 98.6 %, [23-14C]-NOA422390 (emamectin benzoate B1b), purity 98.7%	DT_{50} values (25°C): emamectin B1a 63.7 d emamectin B1b 71.6 d	DT_{50} values (25 °C): 63.7 days for emamectin B_{1a} and 71.6 days for emamectin B_{1b}, recalculated to 20 °C: 95 and 107 days, respectively.	Clark, A. (2003) STUDY IIA, 7.1.1/003
Degradation in soil - Aerobic degradation (EPA Subdivision N, Section 162-1, 1982)	emamectin benzoate B1a, radiochemical purity 99.8%	DT_{50} (25 °C) emamectin B1a sandy loam = 138.6 d	DT_{50} for emamectin benzoate B1a in sandy loam soil (aerobic, 25 °C): 138.6 days, recalculated to 20 °C: 207 days	Chukwudebe, A. (1994a) STUDY IIA, 7.1.1/004
Photochemical degradation in water (US-EPA Subdivision N, 161 – 2, 1982)	[3,7,11,13,23-14C]4"-epimethylamino-4"-deoxyavermectin B1a benzoate; batch nr. L-683,825-003E006; radiochemical purity 96.7%	DT_{50} values (25 °C): phosphate buffer with ethanol = 6.3-8.5 d phosphate buffer with acetonitrile = 31.8-64.5 d phosphate buffer with acetone = 0.5-1.0 d	Direct photolysis of MAB_{1a} in aqueous solution (with 1% acetonitrile as co-solvent) exists but is slow (half-life 32 – 65 days). Acetone or ethanol causes a faster degradation (0.5 – 8.5 days).	Ballantine, L.G. (1994) STUDY IIA, 7.6/01

Method	Test material	Results	Remarks	Reference
			The photolytic degradation route under sensitised conditions was complex with up to 15 distinguishable components after 24 hours irradiation none of which represented ≥ 10% AR. The major residues at 24 hours were parent MAB_{1a} (36.2%), 8,9-Z isomer of MAB_{1a} (6.6% AR), and P3b (6.6% AR), while the remaining components were ≤ 2.4% AR.	
Photochemical degradation in soil (US-EPA 540/9-82-021, Section 161-3 95/36/EC)	[23-14C] –emamectin benzoate B1a, batch CL-LIII-77, radiochemical purity 98.6 % [23-14C] –emamectin benzoate B1b, batch CL-LIII-80, radiochemical purity 98.7 %	DT_{50} values (25 °C): emamectin benzoate B1a = 12.2 d emamectin benzoate B1b = 20.3 d	-	Anderson, W. (2003) STUDY IIA, 7.1.3/001
Photochemical degradation in soil (EPA Subdivision N, Section 161-3, 1982)	14C- emamectin benzoate B1a , radiochemical purity 98.6 %	DT_{50} (23.2-27.8 °C) = 3.9 d	Photolytic half-life recalculated to natural conditions at 40ºN: 2.5 days	Chukwudebe (1994b) STUDY IIA, 7.1.3/002
Photochemical degradation in water (EPA Subdivision N, Section 161-2, 1982;)	[3,7,11,13,23-14C]-emamectin B1a benzoate (14C-MAB1a), L-683,825-003E003, radiochemical purity 98 %; MAB1a, L-656,748	DT_{50} values (25 ± 1 °C): phosphate buffer = 22.4 d sensitised phosphate buffer = 1.4 d natural pond water = 6.9 d	-	Mushtaq, M. (1995) STUDY IIA, 7.6/02
Photochemical degradation in water (OECD draft, August 2000; JMAFF Agchem Test Guidelines 12 Nohsan N.8147, 24.11.2000 (revised 26.06.2001); EPA Subdivision N, Section 161-2, 1982)	[23-14C]-emamectin B1a benzoate, WFH-XI-3, radiochemical purity 99 %	DT_{50} (25.3 ± 0.3 °C) = 0.9 d	No identification of metabolites as these were < 10% or AR. The experimental half-life (DT_{50}) of emamectin benzoate B1a in buffer was calculated as 0.89 days. Based on the quantum yield, the environmental half-life of emamectin benzoate due to direct photolysis at the surface of pure water was calculated at 30° N, 40° N and 50° N latitude summer conditions to be 1.32, 1.35 and 1.42 days	Phaff (2005) STUDY IIA, 7.6/03

Method	Test material	Results	Remarks	Reference
			respectively.	
Field dissipation Guideline not specified	A10324A, a 50 g/kg SG formulation, containing emamectin benzoate, 4.89%	DT_{50} = 2.3 d	-	Evans, P.G. (2006) STUDY IIA, 7.3.1/001
Field dissipation Guideline not specified	A10324A, a 50 g/kg SG formulation, containing emamectin benzoate, 4.89%	DT_{50} = 0.8 d	-	Evans, P.G. (2006) STUDY IIA, 7.3.1/002
Field dissipation Guideline not specified	A10324A, a 50 g/kg SG formulation, containing emamectin benzoate, 4.89%	DT_{50} = 0.3 d	-	Seville, A.G. (2006a) STUDY IIA, 7.3.1/003
Field dissipation Guideline not specified	A10324A, a 50 g/kg SG formulation, containing emamectin benzoate, 4.89%	DT_{50} = 0.6 d	-	Seville, A.G. (2006b) STUDY IIA, 7.3.1/004

11.1.1 Ready biodegradability

The ready biodegradability of ememactin was tested according to a manometric respirometry test performed in accordance with OECD 301 F; 92/69/EEC, L383 A, C.4-D (Dietschy, 1999). Oxygen consumption in blank control was 12 – 38 mg/L after 28 days. Biodegradation of emamectin benzoate B1a was 0 % after 28 days. Biodegradation of sodium benzoate was 79 % at day 14 and 80 % after 28 days, biodegradation in toxicity control was 60 and 63 % after 14 and 28 days. Abiotic control had 0 % degradation after 28 days. Based on the results of the manometric respirometer test it is concluded that emamectin benzoate B1a is not readily biodegradable.

11.1.2 BOD_5/COD

No data available.

11.1.3 Hydrolysis

Hydrolysis of emamectin B1a was tested at different pH and temperatures (Chukwudebe, 1992). The average percentage analysed was calculated by comparison of the amount of radioactivity injected with the theoretical value calculated from the control data. Average percent of radioactivity remaining in the solution (rest is adsorbed to the walls of the container) was 74.0 % for pH 9 to 90.8 % for pH 6. Average percentage analysed ranged from 88.3 % to 101%. No correlation between time and amount of radioactivity. Therefore, losses could not be accounted for by the formation of a volatile hydrolysis product. Average HPLC recovery 98.1 to 99.0 % of AR for the different pHs. Average percentage of the total recovered radioactivity found at the retention time of MAB1a was 75 % at pH 9 and between 90.9 and 93.5% for pHs 5.2 to 8. After 6 weeks, only MAB1a in pH 9 buffer showed significant differences between 0 time and 6-week samples.

A $DT_{50,hydrolysis}$ of 19.5 weeks was determined for pH 9 (Chukwudebe, 1992). Two products, A and B, with relative retention times to MAB1a of approximately 0.21 and 1.16 respectively, were observed to form during the hydrolysis of MAB1a in pH 9 buffer. Products were 9.1 and 9.9 % of radioactivity for A and B, respectively. At four weeks the products would be represented at 6.3 and 6.9 % of radioactivity. Since neither product A nor B represented 10% or more of the total radioactivity after 30 days in pH 9 buffer, identification was not pursued. The mass at the retention time of MAB1a was isolated from the pH 9 and was confirmed to be the test compound by NMR and MS.

In short, emamectin B1a was hydrolytically stable at environmentally relevant pH (4 - 8) and temperature (25 °C). Under basic conditions (pH 9), DT50, hydrolysis of emamectin B1a was 19.5

weeks at 25 °C, resulting in a DT50 value of 29.1 days at 20 °C. Two unidentified degradation products were formed at 9.1 and 9.9% of radioactivity applied.

11.1.4 Other convincing scientific evidence

No data available.

11.1.4.1 Field investigations and monitoring data (if relevant for C&L)

No data available.

11.1.4.2 Inherent and enhanced ready biodegradability tests

No data available.

11.1.4.3 Water, water-sediment and soil degradation data (including simulation studies)

Degradation in water-sediment systems

Two laboratory water/sediment degradation studies were supplied, one of which was not accepted for degradation rate derivation since the sediment content was too low and adherence of the test substance to the walls of the test vessels was reported (with a maximum of 41.8%).

In the other study (Hurt, Grosjean and Mason, 2006), a silt loam system and a sand system were applied with [23–14C]-emamectin benzoate B1a and incubated under aerobic conditions at 20 °C in the dark.

The following DT50-values were obtained for emamectin benzoate B1a after aerobic incubation in water/sediment systems under laboratory conditions:

- DT50,water 8.7 days for both systems, dissipation rate
- DT50,system > 120 days

The decline of concentrations in the water phase was mainly determined by a rapid initial sorption, and the DT50,water thus represents dissipation rather than degradation. The maximum level of emamectin benzoate B1a found in sediment was 71.3 and 83.0% of AR after 90 and 120 days.

Bound residues increased to 20.2 – 10.7 % of AR at the end of the study. Mineralisation was low with a maximum of 1.4 % of AR after 21 days in the silt loam system.

Observed metabolites were not major and therefore not identified. The highest formation rate was reached by metabolite 1 with a maximum of 5.8% on two subsequent time steps in the water phase of the sand system.

Aquatic dissipation in the field

The laboratory studies show that dissipation of emamectin benzoate B1a from the total system is determined by the degradation rate in sediment, which is relatively slow. One metabolite was detected in environmentally relevant concentrations in the water phase; 8a -oxo MAB1a. The photolysis product 8,9-Z-MAB1a is a major product in water however, the laboratory water/sediment studies were performed in the dark, and this metabolite was likely not present in these systems. It is not possible to estimate the relevance of this compound under field conditions, although the photolysis experiment indicates a relatively fast decay (DT50 ca. 8 days) and environmental concentrations may thus be low.

Degradation in soil

Aerobic biodegradation

Emamectin B_{1a}

The rate of degradation of emamectin benzoate B_{1a} under aerobic conditions was studied in five laboratory experiments in five different soil types. Results from one of the five studies are not considered reliable since the duration of the study was only 62 days. Resulting DT_{50}'s are summarised in the tables below (Hand and Fleming, 2006; Jungmann, Nicollier, 2006; Clark, 2003; Chukwudebe, 1994a). Values obtained in the same soil type are averaged before calculation of the overall mean.

Overview of DT_{50}-values from aerobic laboratory degradation studies with emamectin benzoate B_{1a}.

Label	Soil type	Dose [mg/kg]	[°C]	OM [%]	CEC [ ]	pH	DT_{50} [d]	DT_{50}, 20 °C geometric mean per soil [d]
^{14}C	sandy clay loam	.13	0	.8	4 .8		.9	45	45.9
^{14}C	loam	.13	0	.4	5 .4		.2	25	25.2
^{14}C	silty clay loam	.13	0	.8	1 .1		4	41	414
^{14}C	silt loam[1]	.031	0	.6	4 .1	.2	.4	39	41.8
^{14}C	silt loam[2]	.031	0	.6	4 .1		.7	53	
^{14}C	silt loam[1]	.31	0	.6	4 .1	.2	.3	32	
^{14}C	sandy loam	.015	5	.5	0 .3	.2	.7	63	95
^{14}C	sandy loam		5	.0	2 .6	.2	8.6	13	207[4]
									Geomean: 68.9

[1]at a soil moisture content of 40% MWC
[2] tested at a soil moisture content of 20% MWC, normalised to 40% MWC
[4]not used in calculation of geomean, too high concentration

Overview of DT_{50}-values from aerobic laboratory degradation studies with emamectin benzoate B_{1b}

Label	Soil type	Dose [mg/kg]	[°C]	OM [%]	CEC [ ]	pH	DT_{50} [d]	DT_{50}, 20 °C geometric mean per soil [d]
^{14}C	sandy loam	.015	5	.5	0 .3	.2	.6	71	106.8

Since the DT_{50} of emamectin benzoate B_{1b} is in the range of the DT_{50} values of emamectin benzoate B_{1a}, and it is concluded that the results derived from studies conducted with MAB_{1a} the major component of emamectin benzoate are also representative for MAB_{1b}, the resulting DT_{50} value for emamectin benzoate B_{1b} is also used in calculating the geometric mean of the DT_{50} value for emamectin benzoate B_{1a}. The resulting geometric mean DT_{50} at 20 °C is 76.7 days (range 25.2 - 414 days; n = 7).

Anaerobic biodegradation

One degradation study under anaerobic conditions was submitted. The study is considered not acceptable because anaerobic conditions were established only in the second part of the study resulting in only two data points for the anaerobic part. The DT50 value is calculated based on three data points and is therefore considered not reliable.

The laboratory data suggest that the substance will degrade rapid to moderately in water sediment systems. No major metabolites were observed in water sediment systems.

The laboratory data suggest a moderate degradation of the substance in soil. However, the results of acceptable field studies at four representative sites indicate a rapid degradation in soil. Based on these studies, the relevant metabolites were identified as 8a-OH MAB1a (max 13.8 % AR after 21 d) and N-nitroso MAB1a (max. 15.3 % AR after 28 d). However, since these metabolites are from soil degradation and were not present in water-sediment systems at >10%, they are not considered relevant for classification.

11.1.4.4 Photochemical degradation

Photo degradation in water

The photo degradation of emamectin B1a was determined in three studies (Ballantine, 1994; Mushtaq, 1995; Phaff, 2005), the first of which was conducted under artificial sunlight (Ballantine, 1994). A photosensitizer (acetone) and a radical hydrogen donor (ethanol) were also used in this study. Direct photolysis resulted in a DT50,photolysis for emamectin B1a ranging from 32-65 days under study conditions. The photolytic degradation route under sensitized conditions resulted in DT50 values ranging from 0.5-8.5 days under study conditions. For the characterisation of potential photo degradation products of MAB1a, a 24-hour acetone sensitised sample was chosen because it was the most extensively degraded of the samples. The following metabolites were identified: AB1a, 8a-OH MAB1a, 8a -oxo MAB1a, 8,9-Z-MAB1a and the di-epoxide metabolite with a maximum of 6.57% for 8,9-Z-MAB1a.

In the second study a photo sensitizer (acetone) was also used (Mushtaq, 1995). Direct photolysis without sensitizer resulted in a DT50,photolysis for emamectin B1a of 22.4 days in phoshate buffer and 6.9 days in natural pond water both under study conditions (latitude ca. 40°N in fall). The photolytic degradation route under sensitized conditions resulted in a DT50 value of 1.4 days. The major degradate in the light exposed buffer and natural water samples was the 8,9-Z-isomer of MAB1a that was found at maximum amounts of 12.3% and 17.1%, respectively. MAB1a-10,11-14,15-diepoxide was identified also in both systems, however, at amounts not exceeding 2.8% and 3.5%, respectively. Both degradates were found also in the sensitised buffer samples with MAB1a-10,11-14,15-diepoxide occurring at a greater amount (18.3%) than the 8,9-Z-isomer of MAB1a (8.4%). Recalculation to summer condition for latitude ca. 40°N revealed DT50,photolysis of 11.5 days for buffer and 3.6 days for natural water.

In the third study simulated sunlight was used (Phaff, 2005). The experimental half-life (DT50) of emamectin benzoate B1a in buffer was calculated as 0.89 days. Based on the quantum yield, the environmental half-life of emamectin benzoate due to direct photolysis at the surface of pure water was calculated at 30° N, 40° N and 50° N latitude summer conditions to be 1.32, 1.35 and 1.42 days respectively.

No major metabolites were formed.

Photo degradation in soil

Two soil photolysis studies were submitted (Anderson, 2003 and Chukwudebe, 1994b). In one study (Anderson, 2003), DT50, photolysis values for emamectin benzoate B1a of 12.2 days and of 20.3 days

for emamectin benzoate B1b were obtained under study conditions (artificial light source at 25 °C). No recalculation to natural conditions because the light intensity in the study was not reported. Unidentified radioactivity from extraction 1 should be further addressed for this study. In the second study (artificial light, 25 ºC) photolytic half-life recalculated to natural conditions at 40ºN, of 2.5 days was derived (Chukwudebe, 1994b).

Based on the data above photodegradation is considered a significant degradation pathway in water sediment systems and in soil.

11.2 Environmental transformation of metals or inorganic metals compounds

Not relevant for this dossier.

11.2.1 Summary of data/information on environmental transformation

Not relevant for this dossier.

11.3 Environmental fate and other relevant information

Two batch soil adsorption/desorption studies containing a total of 8 different soils were submitted for the active substance. The reported K_{foc} ranged between 6666 and 278983. Based on these data, emamectin benzoate can be considered to be immobile in soil.

Emamectin benzoate has a vapour pressure of 4 E-06 Pa, and a Henry's law constant of $1.7x10^{-4}$ Pa m^3/mol. Based on this information it is considered that significant volatilisation of emamectin benzoate is unlikely to occur from soil and from a water solution. The gas phase oxidation half-life for emamectin benzoate was estimated to be < 1 hour. Should emamectin benzoate volatilise, the compound will degrade quickly.

Field dissipation studies were carried out in France, Germany, and at three locations in the US (Evans, 2006; Seville, 2006a; Seville, 2006b). All studies were performed with A10324A, a 50 g/kg SG formulation containing 4.89% emamectin benzoate. A summary of conditions and results is given below.

Summary of field dissipation studies with emamectin benzoate

Location	Soil type	Land use	Dose emamectin benzoate [g as/ha]	Month of application	monthly temperature [°C] over test period		DT_{50} [d]
					min	max	
Tiercé, F	sandy loam	sparsely covered with grass	22.5	June	-3.1	22.6	2.6
Marsillargues, F	silty clay loam	sparsely covered with grass	22.5	June	-5.2	24.5	0.8
Grisolles, F	clay loam	sparsely covered with grass	22.5	May	-9.4	23.5	0.3
Wallersdorf-See	silty clay loam	bare soil	22.5	June	-6.8	18.8	0.6

The results of the field studies confirm that emamectin benzoate is rapidly dissipated under field conditions. In the first three studies, samples were also analysed for 8,9-Z-MAB_{1a} and 8a-OH MAB_{1a}, and in the fourth study also for MFB_{1a} and N-nitroso MAB_{1a}. None of the metabolites was detected under field conditions.

11.4 Bioaccumulation

Table 51: Summary of relevant information on bioaccumulation

Method	Results	Remarks	Reference
fish, bioconcentration *Lepomis macrochirus* flow-through (US EPA 54019-82-021; ASTM E1022-84)	BCF edible tissue: 30 L/ kg BCF nonedible tissue: 102 L/ kg BCF whole fish: 82 L/ kg	BCF based on total radioactivity, transformation may have taken place and values are worst case. Residues are emamectin benzoate and its demethylated product AB1a predominantly.	STUDY IIA 8.2.6.1/01

11.4.1 Estimated bioaccumulation

No data available.

11.4.2 Measured partition coefficient and bioaccumulation test data

Bioaccumulation of emamectin benzoate B1a was studied in *Lepomis macrochirus* (STUDY IIA 8.2.6.1/01) according to US EPA 54019-82-021 and ASTM E1022-84. Bluegill sunfish were exposed to emamectin benzoate and ^{3}H-MAB1a for 28 days to measure uptake of the compound and then placed in clean water for 14 days to determine elimination rate. The experiment was performed in two phases in which the second phase mimicked the first phase and served to generate tissue samples to evaluate the identity and distribution of emamectin benzoate and its metabolites in fish.

Mortality <1% in control and treatment and no abnormal behaviour. The mean measured radioactivity concentration in the treatment chambers during the exposure phase, was equivalent to 1.2 ± 0.095 µg 3H-MAB1a/L.

For confirmation that the radioactivity present was ^{3}H-MAB1a, the test item was monitored after 28 days of exposure by means of solid phase extraction and HPLC-analysis. It was found that ^{3}H-MAB1a was stable in water under the ambient flow through conditions of the study.

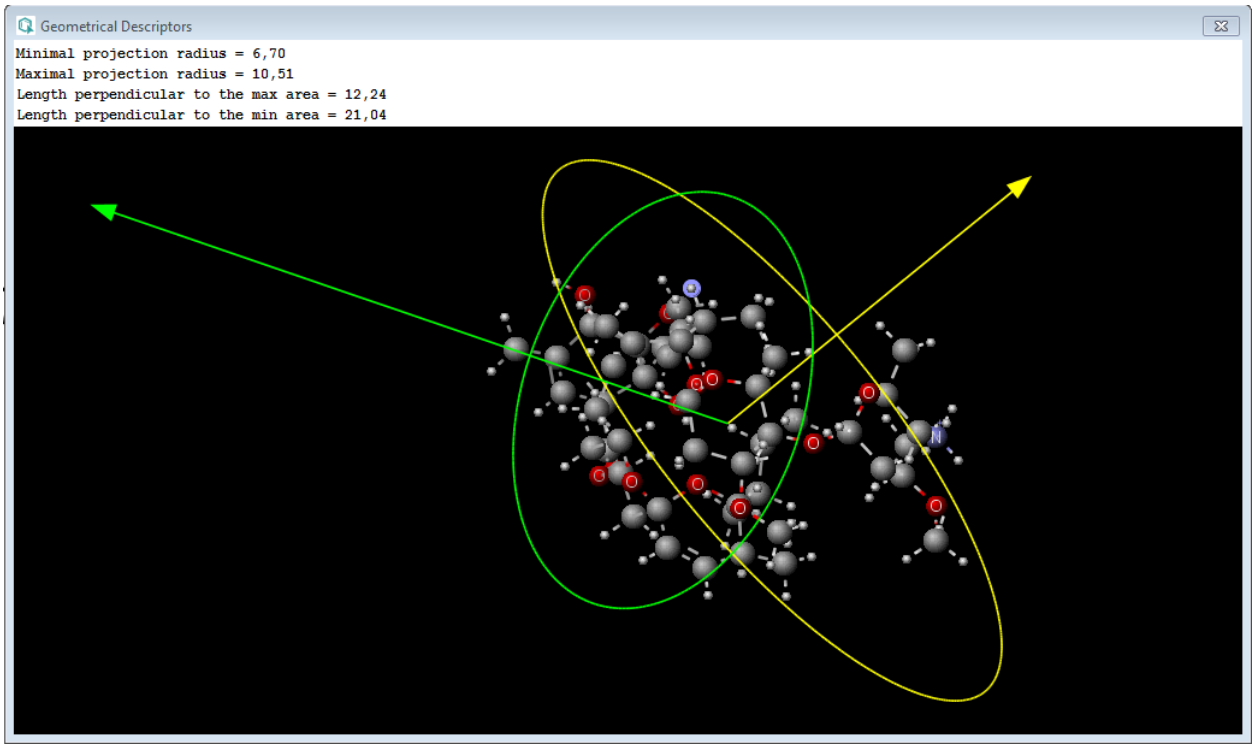
Test criteria were not completely met. According to OECD guideline 305 the test substance in fish taken at three consecutive analyses which are taken at at least two days intervals, should be within 20% of each other. Also, there should not be significant differences among the three sampling periods. However, the test concentration at day 3 was reduced to 68% (=1.1 µg/kg), while at days 0, 1 and 7 the test concentration was around 75% (=1.2 µg/kg). The nominal test concentration was 1.6 µg/kg.

Plateau levels are considered to be between 21 and 28 days by RMS. The experiment has not lasted long enough to reach three consecutive samples in the steady state. The timepoints of the analyses have not been chosen well. If one extra timepoint had been chosen in between 21 and 28 days, the plateau could have been confirmed. For the 21 – 28 days interval, the recalculated mean BCFs are 30, 102 and 82 L/kg wwt for edible tissue, nonedible tissue and whole fish, respectively. The kinetic whole fish BCF was reported to be 80 L/kg wwt, which was confirmed by the dossier submitter. It appears that the BCF values were not corrected for growth nor were they normalized to 5% lipid content. The dossier submitter could not correct nor normalize the BCF values, as data on lipid content and growth were not available. Overall, while there remains some uncertainity with regard to the bioaccumulation potential of emamectin benzoate B1a, the experimental data do suggest a low bioaccumulation potential.

The log K_{ow} for emamectin benzoate depends on pH and has been determine to be 5.9, 5.0 and 3.0 at pH 9.0, 7.0, and 5.0, respectively. The methodology has not been specified, and study details were not available, which unables the reliability assessment. Nevertheless, considering the experimentally

determined pK_a of 7.7, and the fact that the molecule is increasing neutrally charged at higher pH values, these log K_{ow} values correspond with expectations. Furthermore, QSAR estimated values are in the same range, i.e. a log K_{ow} of 2.93 for the ionic species, and a log K_{ow} of 6.17 for the neutral species (MarvinSketch logP v16.10.24). Based on the CLH criteria a value of log Kow ≥ 4 indicates a potential to bioaccumulate.

On first sight the BCF values and log K_{ow} values appear conflicting, with the BCF of 80 L/kg wwt suggesting low bioaccumulation potential and the log K_{ow} of 5.9 indicating bioaccumulation potential. However, steric hindrance can limit uptake by biota. As stated in REACH guidance R.11 a molecule with an average maximum diameter (Dmax aver) of greater than 1.7 nm plus a molecular weight of greater than 1100 may be considered as not B. Emamectin is such a large molecule with a diameter of 2.1 nM (MarvinSketch logP v16.10.24; see below), and a molecular size of 1008.3. Thus, while the log K_{ow} of the neutral molecule is above the criterion, emamectin is considered to have a low bioaccumulation potential.



11.5 Acute aquatic hazard

Table 52: Summary of relevant information on acute aquatic toxicity

Method	Species	Test material	Results	Remarks	Reference
fish, acute toxicity (freshwater) flow-through	*Oncorhynchus mykiss*	Technical MK-244 (emamectin benzoate), batch L-656,748-052 S002, purity 95.9	96-hours LC_{50} 174 µg/L, based on mean measured concentrations	actual; 68 – 123 % of nominal Water quality parameters within accepted limit and	STUDY IIA 8.2.1.1/01

ASTM E 729-88; EPA 540/9-82-024		%, appearance white powder		the study is considered acceptable.	
fish, acute toxicity (freshwater) flow-through ASTM E 729-88; EPA 540/9-82-024	*Lepomis macrochirus*	Technical MK-244 (emamectin benzoate), batch L-656,748-052 S002, purity 95.9 %, appearance white powder	96-hours LC_{50} 180 µg/L, based on mean measured concentrations	actual; 88 – 112 % of nominal Water quality parameters within accepted limit and the study is considered acceptable.	STUDY IIA 8.2.1.2/01
fish, acute toxicity (freshwater) flow-through ASTM E 729-88; EPA 540/9-82-024	*Pimephales promelas*	Technical MK-244 (emamectin benzoate), batch L-656,748-052 S005, purity 94.6 %, appearance white powder Radiolabelled MK-244, batch L-683,825-055J006, 15994-111/95-137, purity 99.3%	96-hours LC_{50} 194 µg/L, based on mean measured concentrations	actual; 69 – 87 % of nominal. test with radiolabelled emamectin MAB_{1a} Water quality parameters within accepted limit and the study is considered acceptable.	STUDY IIA 8.2.1.2/02
fish, acute toxicity (saltwater) flow-through (ASTM E 729-88; EPA 540/9-82-024)	*Cyprinodon variegatus*	Technical MK-244 (emamectin benzoate), batch L-656,748-052 S005, purity 95.9 %, appearance white powder	96-hours LC_{50} 1430 µg/L, based on mean measured concentrations	actual; 83 – 109 % of nominal Water quality parameters within accepted limit and the study is considered acceptable.	STUDY IIA 8.2.1.2/04
Daphnia, acute toxicity (freshwater) flow-through US EPA 540/9-82-024; ASTM E 729-88	*Daphnia magna*	MK-244 (emamectin benzoate), batch L-656, 748-052S002, purity 95.9%, appearance white powder	48-h EC_{50} of 1.0 µg/L, based immobility and mortality and based on mean measured concentrations	actual; 58 - 67 % of nominal Water quality parameters within accepted limit and the study is considered acceptable.	STUDY IIA 8.3.1.1/01
oyster embryo, acute toxicity (saltwater) flow-through ASTM E 729-888, EPA 540/9-82-024	*Crassostrea virginica*	Technical MK-244 (emamectin benzoate), batch L-656,748-052 S005, purity 95.9 %, appearance white powder	96-hours EC_{50} 530 µg/L, based on mean measured concentrations	actual; 76 – 120 % of nominal Water quality parameters within accepted range. The highest three test concentrations above water solubility in freshwater (0.24 mg/L at pH 7), but the solvent control was used to increase solubility. No flocculation reported for these concentrations. The study is considered acceptable.	STUDY IIA 8.3.1.1/03
mysid shrimp,	*Mysidopsis*	MK-244	96-hours LC_{50}	actual; 54 -85 % of	STUDY IIA

acute toxicity (saltwater) flow-through EPA 540/9-82-024, EPA540/9-85-010, ASTM E 729-88	*bahia*	(emamectin benzoate), batch L-656, 748-052S002, purity 95.9%, appearance white powder; 3H-MAB1a (emamectin B1a), batch L683,825-001A009; 18075-148; 93-014, radiochemical purity 97.2% (TLC, scanning) appearance clear solution	0.040 µg/L, based on mean measured concentrations	nominal. test with radiolabelled emamectin MAB_{1a} Water quality parameters within accepted limit and the study is considered acceptable.	8.3.1.1/04
Lemna, growth inhibition static-renewal US-EPA 540/9-82-020; ASTM E 1415-91	*Lemna gibba*	MK-244 (emamectin benzoate), batch nr.L656,748-052S005, purity 94.6 %, appearance-white powder and ^{3}H-MK-244 batch nr. L-683,825-005J006, purity 99.3%, appearance a clear liquid	14 –day IC_{50} > 94 µg/L, based on mean measured concentrations of fresh solutions	actual; 62 – 85% of nominal The study is considered acceptable.	STUDY IIA 8.6/01
algae, growth inhibition static US EPA 540/9-82-020 ontarget plants	*Pseudokirchneriella subcapitata*	MK-244 (emamectin benzoate), batch L-656, 748-052S002, purity 94.6 %, appearance white powder and ^{3}H-MK-244, batch L-683,825-005J006, appearance clear liquid	120-h E_rC_{50} = > 3.9 µg/L 120-h NOEC ≥ 3.9 µg a.s./L	actual, initial The pH at 0 and 120 h differed with more than 2 points in the test substance (pH 7.4 – 9.6). Only one concentration was tested and the actual concentration was too low. The study is considered acceptable.However, the experiment was not suitable to determine an accurate EC_{50}.	STUDY IIA 8.4/001
algae, growth inhibition static US EPA OPPTS 850.5400; OECD 201; EC, L383 A, Part C.3	*Pseudokirchneriella subcapitata*	MK-244 (emamectin benzoate), batch SSH2F004, purity 97.3 %, appearance white powder	96-hour E_bC_{50} = 7.2 µg/L 96-hour ErC_{50} = 7.2 µg/L NOEC <4.6 µg/L	actual; below LOQ – 55% of nominal The pH at 0 and 96h differed with more than 2 points at 3 lowest concentrations. The author considered this shift to be the result of high growth factors. Differences were less than 1 point at the 4	STUDY IIA 8.4/002

				highest concentrations. Biomass data indicated that there was an artefact as biomass seemed to increase at higher concentrations. Moreover, the growth rate did not decrease to 0. As exponential growth was still present in the control at 96 h the data can be used. The E_rC_{50} calculated by the author seems to be a mistake. The result, a 96-h E_bC_{50} of 7.2 µg/L is considered aceptable. The study is considered acceptable.	

[1] Indicate if the results are based on the measured or on the nominal concentration

11.5.1 Acute (short-term) toxicity to fish

Several acute toxicity tests in fish are available. Studies were performed using freshwater and saltwater fish. Three available studies using freshwater fish (*Oncorhynchus mykiss, Lepomis macrochirus and Pimephales promelas*) were guideline studies performed in accordance with ASTM E 729-88; EPA 540/9-82-024.

Toxicity of MK-244 to rainbow trout (*Oncorhynchus mykiss*) was tested under flow-through conditions (STUDY IIA 8.2.1.1/01). In this study, actual concentrations were 67 - 124 % of nominal at the start of the study and 62 - 142 % of nominal at the end of the study. Mean measured concentrations were 21.1, 30.4, 48.7, 132 and 246 µg/L (68 - 123 % of nominal). There were no mortality in controls and solvent controls, and at 21.1 – 48.7 µg/L, 20 % mortality at 132 µg/L, 85 % at 246 µg/L, first deaths after 96 hours. 96-hours LC50 reported as 174 µg/L (95 % CL 146 – 207 µg/L). LC50-values for the 96 h exposure period was calculated using binomial probability and the LC50 for 24, 48 and 72 hours of exposure was based on visual interpretation.

Toxicity of MK-244 to bluegill (*Lepomis macrochirus)* was tested under semi-static test conditions (STUDY IIA 8.2.1.2/01). Actual concentrations were 39 - 67 % of nominal in fresh solutions, and 75 - 98 % of nominal in old solutions. In between actual concentrations were 78 – 158 % of nominal. Mean measured concentrations were 0.056, 0.087, 0.14, 0.24 and 0.35 mg/L (88 - 112 % of nominal). Actual concentrations were considerably below nominal at the start of the experiment, but approximated nominal values at later sampling points. This was not considered to affect the outcome of the toxicity test. There was no mortality in control, solvent control, and the two lowest concentrations, 5 % mortality at 0.14 mg/L, 100 % at 0.24 and 0.35 mg/L by day 4. 96-hours LC50 reported as 0.18 mg/L (95 % CL 0.14 – 0.24 mg/L, 96-hours NOEC was 0.087 mg/L, all based on mean measured concentrations.

Toxicity of MK-244 to fathead minnow (*Pimephales promelas*) was tested under flow-through conditions (STUDY IIA 8.2.1.2/02). Actual concentrations were 74 - 88 % of nominal at start and 67 - 87 % of nominal at end. Mean measured concentrations were 27, 48, 89, 156 and 257 µg/L (69 -87 % of nominal). No mortality in controls and solvent controls, and at 27 – 89 µg/L, 5 % mortality at 156 µg/L, 100 % at 257 µg/L, first deaths after 72 hours. 96-hours LC50 reported as 194 µg/L (95 % CL 156 – 257 µg/L). LC50-values for the 96 h exposure period was calculated using binomial probability and the LC50 for 24, 48 and 72 hours of exposure was based on visual interpretation.

Toxicity of emamectin benzoate to sheepshead minnow (*Cyprinodon variegatus)* was tested under flow-through conditions in accordance to ASTM E 729-88; EPA 540/9-82-024 (STUDY IIA 8.2.1.2/04). Measured concentrations were corrected for mean procedural recovery. Actual concentrations were 85 - 109 % of nominal at start and 83 - 101 % of nominal at end. Mean

measured concentrations were 0.33, 0.50, 0.86, 1.43 and 2.63 mg/L. No mortality in controls and solvent controls, and at 0.33, 0.50 amd 0.86 mg/L, 60 % mortality at 1.43 mg/L, 95 % at 2.63 mg/L, first deaths after 72 hours. Sublethal effects (discolouration) at 0.50 mg/L. 24-hours LC50 > 2.63 mg/L, 48-hours LC50 >2.63 mg/L, 72-hours LC50 >2.63 mg/L and 96-hours 1.43 mg/L (95 % CL 1.25 – 1.67 mg/L). LC50-values for the 96 h exposure period was calculated using probit analysis and the LC50 for 24, 48 and 72 hours of exposure was based on visual interpretation.

Overall, *Oncorhynchus mykiss* is most sensitive to emamectin benzoate. The other three tested species are only slightly less sensitive. Toxicity of emamectin benzoate to the saltwater species *Cyprinus variegatus* is more than a factor of 8 higher compared to toxicity to *Oncorhynchus mykiss*.

11.5.2 Acute (short-term) toxicity to aquatic invertebrates

Two acute toxicity studies are available for *Daphnia magna*, which were performed in accordance with guideline US EPA 540/9-82-024; ASTM E 729-88 and OECD 202; EU comm. dir. 92/69/EEC, C.2; US EPA OPPTS 850.1010 for flow-through ans static test systems, respectively.

In one of the available studies (STUDY IIA 8.3.1.1/01), daphnids (< 24 h old) were exposed to emamectin benzoate for 48 h in flow-through test systems (300 mL glass beakers). Recovery at test initiation ranged from 67 to 94% of nominal and at end 63 to 81% of nominal. Mean measured concentrations were 0.30, 0.47, 0.85, 1.38 and 2.26 µg/L (corrected for mean procural recovery of 99 %). No mortality in control, solvent control and two lowest concentrations.15 % at 0.85 µg/L, 45 and 80 % mortality at 1.38 and 2.26 µg as/L. Nominal 48-hours EC_{50} reported as 1.0 µg/L (95% CL 0.84 – 1.19 µg/L).

Effects of emamectin benzoate on deposition of the eastern oyster, *Crassostrea virginica*, was tested under flow-through conditions (STUDY IIA 8.3.1.1/03). The available study was performed according to ASTM E 729-888, EPA 540/9-82-024. Measured concentrations were corrected for mean procedural recovery. Actual concentrations were 76 - 120 % of nominal at start and 84 - 110 % of nominal at end. Mean measured concentrations were 0.10, 0.15, 0.26, 0.42 and 0.77 mg/L. Shell deposition was 3.03 mm in the pooled controls. Percent inhibition was -7.3, -9.6, -29, 47 and 72% at 0.10, 0.15, 0.26, 0.42, 0.77 mg as/L. The 96-LC50 was reported as 0.53 mg as/L (95 % CL 0.35 – 1.2 mg/L).

Acute toxicity of ^{3}H-MAB_{1a} to juvenile *Mysidopsis bahia* reared at Wildlife International Ltd., was tested under flow-through conditions (STUDY IIA 8.3.1.1/04). This was done using a guideline study, performed according to EPA 540/9-82-024, EPA540/9-85-010, ASTM E 729-88. Mean measured concentrations were 7.8, 18, 26, 41 and 72 ng/L. At day 0 recovery ranged from 54 to 71 % of nominal values. At 96 h recovery ranged from 65 to 85% of nominal values. Measured concentrations at test initiation were always lower than concentrations measured later during the test. The test substance was stable during the experiment. No control mortality and no mortality occurred in solvent control and at 7.8 and 18 ng/L. 10, 45 and 100 % mortality was observed at 26, 41 and 72 ng/L. LC50 for 96 hours exposure was reported as 0.040 µg/L (95 % CL 0.035 – 0.046 µg/L), based on mean measured concentrations.

In conclusion, *Daphnia magna* is the only freshwater invertebrate species tested and most sensitive to emamectin benzoate in comparison with fish. The saltwater invertebrate *Mysidopsis bahia* was more sensitive to emamectin benzoate than *Daphnia magna*. In fact, the most sensitive acute endpoint was observed in a study on *Mysidopsis bahia*.

11.5.3 Acute (short-term) toxicity to algae or other aquatic plants

The algal growth inhibition test is a short-term test that provides both acute and chronic endpoints. However, the EC50 is treated as an acute value for classification purposes. Two studies performed with algae are available, which in accordance with the Guidance Document on Aquatic Ecotoxicology, are considered chronic. These static algae growth inhibition studies are performed according to guideline US EPA 540/9-82-020 nontarget plants and US EPA OPPTS 850.5400; OECD 201; EC, L383 A, Part C.3. In one of the studies, only one concentration was tested and the

actual concentration was too low (STUDY IIA 8.4/001). The experiment was not suitable to determine an accurate EC50. The 120-h ErC50 was determined to be > 3.9 µg a.s./L based on initial concentrations. In the second algae growth inhibition study (STUDY IIA 8.4/002) the 96-h E_bC_{50} was determined to be 7.2 µg/L (95% CL 5.4 – 9.5 µg/L) and the 96-h E_rC_{50} was 12.1 µg/L (95% CL 10.5 – 13.9 µg/L). Biomass data indicated that there was an artefact as biomass seemed to increase at higher concentrations. Moreover, the growth rate did not decrease to 0. As exponential growth was still present in the control at 96 h the data can be used.

Toxicity to aquatic plants was tested in the fresh water species *Lemna gibba*, using emamectine benzoate under static-renewal conditions by exposure through the water phase (STUDY IIA 8.6/01). Please note that the aquatic plant growth inhibition tests are normally considered as chronic tests but the EC50s are treated as acute values for classification purposes. The study performed with *lemna* gibba is a guideline study performed in accordance with US-EPA 540/9-82-020; ASTM E 1415-91. The mean concentrations of fresh solutions were 6.8 and 94 µg/L (62 and 85% of nominal) as determined by LSC. Concentrations in test chambers collected on days 1, 2, 3 and 14 were lower (49% of the mean values for the 110 µg/L solution). Plant tissue concentrations were <LOQ at day 3 and 55-78 µg/kg tissue at day 14. Doubling time of frond number in the controls was almost 3 days. Mean number of fronds after 14 days was 728 in control and 724 in solvent control, and 714 and 721 at 6.8 and 94 µg/L, respectively. Percent inhibition of frond numbers was 1.7 and 0.69%, both not significant. Mean number of plants after 14 days was 232 in control and 217 in solvent control, and 231 and 244 at 6.8 and 94 µg/L, respectively. No inhibition of number of plants. No significant differences in percentages of dead, chlorotic, or necrotic fronds. IC50 (fronds) were reported as > 94 µg/L, based on mean measured concentrations.

11.5.4 Acute (short-term) toxicity to other aquatic organisms

No data available.

11.6 Long-term aquatic hazard

Table 53: Summary of relevant information on chronic aquatic toxicity

Method	Species	Test material	Results[1]	Remarks	Reference
Daphnia, chronic toxicity flow-through US EPA 540/9-82-024; ASTM E 1193-87; US EPA 540/9-86-141	*Daphnia magna*	Technical MK-244 (emamectin benzoate), batch L-656,748-052 S-002, purity 97.5 %, appearance white powder Radiolabelled MAB1a ([^{3}H]MK-244), batch L-653,825-055J001, 15670-101-28/93-325	21-day NOEC = 0.088 µg/L	actual; 80 - 88% of nominal, radiolabelled MAB1a based on survival Water quality parameters within accepted range. Concentrations not properly chosen, mortality at three highest concentrations. The study is considered acceptable.	STUDY IIA 8.3.2/001
toxicity sediment dwelling organisms water/sediment spiked OECD 218; proposal BBA	*Chironomus riparius*	MK-244 (emamectin benzoate), batch nr. EZ910012, purity 95.6 %, appearance - white powder	29-day NOEC = 1.25 µg/kg	nominal initial in sediment phase emergence Concentrations in water phase below LOQ indicate high sorption of test compound to the sediment. The study is considered acceptable.	STUDY IIA 8.5.2/01

guideline (1995)					
fish, early life stage toxicity flow-through ASTM E1241-88, 1988 US EPA 540/9-82-024, 1982 and 540/9-86-138, 1986	*Pimephales promelas*	Technical MK-244 (emamectin benzoate), batch L-656,748-052 S005, purity 94.6 %, appearance white powder and ^{3}H-MK244, batch [3H] L-683,825-005J006, ([5-3H] epimethylaminoavermectin B_{1a} benzoate) and L-683,825-005J006, ([5-3H] epimethylaminoavermectin B_{1a} benzoate), clear liquids, substances suspended in ethanol	32-day NOEC = 12 µg/L	actual; 79 - 93 % of nominal; emamectin MAB_{1a}; based on length, wet and dry weight Water quality criteria within accepted limits and the study is considered acceptable.	STUDY IIA 8.2.4/01

[1] Indicate if the results are based on the measured or on the nominal concentration

11.6.1 Chronic toxicity to fish

Chronic toxicity was tested in the freshwater species fathead minnow (*Pimepales promelas*) in accordance to ASTM E1241-88, 1988 US EPA 540/9-82-024, 1982 and 540/9-86-138, 1986 (STUDY IIA 8.2.4/01). In this test, early life stages of fathead minnow were exposed to emamectin benzoate under flow-through conditions. The test substance was apparently binding to any particulate/organic matter in the test aquaria in spite of all precautions taken to reduce the amount of particulate matter. Hatching in the negative and solvent controls 88 and 83%. In the test treatments the hatching success varied from 80 to 86%. No significant differences with the controls. Survival was between 76 and 95 % at 3.0 to 28 µg/L At 54 µg/L a survival was 22% and significantly lower than in the controls (79 and 86% for the negative control and the solvent control, respectively). Total length, wet weight and dry weight were significantly different from pooled control group at 28 µg/L. Significance was not determined at the highest concentration because of the significant effects on survivial. Nevertheless effects had increased at the highest concentration. NOEC for length, wet and dry weight was determined to be 12 µg/L, based on mean measured concentrations.

11.6.2 Chronic toxicity to aquatic invertebrates

Chronic toxicity of emamectin benzoate to *Daphnia magna* was tested under flow-through conditions (STUDY IIA 8.3.2/001). The test performed was a guideline study, performed in accordance with US EPA 540/9-82-024; ASTM E 1193-87; US EPA 540/9-86-141. The reproduction results of the solvent control differed significantly from the negative control. Therefore the solvent control was used for comparisons among the treatment groups. Reproduction at 0.043 and 0.088 µg/L did not significantly differ from the solvent control. The NOEC was 0.088 µg/L based on daphnid survival and expressed on the basis of mean measured concentrations.

The toxicity of emamectin benzoate to daphnids is very high, compared to toxicity to other tested species. In fact, the most sensitive chronic toxicity value was observed in a study with *Daphnia magana*

11.6.3 Chronic toxicity to algae or other aquatic plants

See acute toxicity studies on algae and other aquatic plants.

11.6.4 Chronic toxicity to other aquatic organisms

In water-sediment fate studies, emamectin B1a was present in the sediment in amounts > 10% of AR and the NOEC for Daphnia was < 0.1 mg/L. Therefore, the potential risk for sediment-dwelling invertebrates was identified (STUDY IIA 8.5.2/01). This was done using a guideline study performed in accordance with guideline OECD 218; proposal BBA guideline (1995). In this study the chronic effects of emamectin benzoate (90.3% emamectin benzoate B_{1a} and 5.7% emamectin benzoate B_{1b}) on chironomid larvae were assessed in a water-sediment system under static conditions. Actual concentrations in the water were below the LOQ. In the sediment, measured concentrations of emamectin benzoate B1a were between 94 and 116% of nominal at day 0 and at day 29. Concentrations of emamectin benzoate B1b were below LOQ for all concentrations except for the highest concentration of 10 µg/L which was 100% of nominal. At test end the concentrations of emamectin B1a were between 83 and 101% of nominal. Concentrations of emamectin benzoate B1b were below LOQ for all concentrations. Mean emergence in controls was 84% and 85% in the solvent control. In the test substances the emergence (males + females) was 83%, 79%, 38%, 8% and 1% at increasing concentrations. Males and females pooled for statistical analyses. Also the two controls were pooled as these did not differ significantly. Emergence rate was significantly reduced at 2.5 µg/kg and higher. Development rate was not influenced at the tested concentrations. EC50 reported as 2.4 µg a.s./kg dwt sediment (95% C.I. 1.2 – 2.9) for emergence rate and a NOEC of 1.25 µg a.s/kg dwt sediment. For the development rate an EC50 of > 10 µg a.s./kg dwt sediment was determined for males and for males and females pooled. No EC50 was determined for females alone. The NOEC for males, for females and for males and females pooled was 10 µg as/kg dwt sediment.

11.7 Comparison with the CLP criteria

11.7.1 Acute aquatic hazard

The criteria for Category Acute 1 in line with Table 4.1.0 (a) from the Guidance on the Application of the CLP Criteria are:

96 hr LC50 (for fish)	≤ 1 mg/l and/or
48 hr EC50 (for crustacea)	≤ 1 mg/l and/or
72 or 96 hr ErC50 (for algae or other aquatic plants)	≤ 1 mg/l.

In the available studies performed with fish, the lowest LC_{50} value was found to be 0.174 mg/L and is thus lower than 1 mg/L. The toxicity to crustacea, oyster embryos, algae and aquatic plants was also below 1 mg/L. Based on the lowest EC_{50} of 0.000040 mg/L for mortality observed in *Mysidopsis bahia* (salt water), emamectin should be classified as Aquatic Acute 1; H 400, with an M-factor of 10000.

11.7.2 Long-term aquatic hazard (including bioaccumulation potential and degradation)

Emamectin is not readily biodegradable based on a guideline study performed according to OECD 301F with 0% degradation after 28 days. Emamectin was also shown to be hydrolytically stableat pHs between 5.2 and 8, while at pH 9 hydrolysis was observed with a $DT_{50,hydrolysis}$ of 19.5 weeks.

Aqueous photolysis studies of emamectin benzoate demonstrated that emamectin benzoate is rapidly photolysed. However, under environmentally relevant conditions in turbid and deeper waters this might be a limited degradation route. Aquatic simulation data are not available. Considering above, emamectin is considered not rapidly degradable for classification purposes. The log Kow for emamectin benzoate depends on pH (log Kow 5.9 - 5.0 - 3.0 at 23 °C at pH = 9.0 - 7.0 - 5.0, respectively). Based on the CLH criteria a value of log Kow ≥ 4 indicates a potential to bioaccumulate. However, emamectin is a large molecule and steric hindrace is likely to limit uptake by biota, as is evident from the experimentally determined whole fish BCF_k value of 80 L/kg. The

BCF being lower than 500, indicates that emamectin should be considered as having a low potential for bioaccumulation according to the CLP criteria.

The criteria for Category Chronic 1 and 2 in the CLP Guidance for non-rapidly degradable substances for which adequate chronic toxicity data are available are (Table 4.1.0 (b) (i)):

Category Chronic 1:

Chronic NOEC or ECx (for fish)	≤0.1 mg/l and/or
Chronic NOEC or ECx (for crustacea)	≤0.1 mg/l and/or
Chronic NOEC or ECx (for algae or other aquatic plants)	≤0.1 mg/l.

Category Chronic 2:

Chronic NOEC or ECx (for fish)	> 0.1 to ≤ 1 mg/l and/or
Chronic NOEC or ECx (for crustacea)	> 0.1 to ≤ 1 mg/l and/or
Chronic NOEC or ECx (for algae or other aquatic plants)	> 0.1 to ≤ 1 mg/l.

One long-term guideline study in fish is available in which a NOEC of 0.012 mg/L was derived. The NOEC for crustacea was found to be 0.000088 mg/L. A NOEC of 0.00125 mg/kg sediment was determined for midge larvae (*Chironomus riparius*). However, this NOEC cannot be used for classification puroposes as the NOEC was determined for exposure via the sediment compartment and not the water compartment. For algae and aquatic plants the lowest NOEC was determined to be ≥ 0.0039 mg/L. Considering the lowest chronic value of 0.000088 mg/L derived from the three trophic levels, classification for chronic toxicity is warranted as Aquatic Chronic 1; H 410, with an M-factor of 1000.

11.8 CONCLUSION ON CLASSIFICATION AND LABELLING FOR ENVIRONMENTAL HAZARDS

Acute (short-term) aquatic hazard: category Acute 1, M-factor: 10000.

Long-term aquatic hazard: category Chronic 1, M-factor: 1000.

12 EVALUATION OF ADDITIONAL HAZARDS

12.1 Hazardous to the ozone layer

No data available.

12.1.1 Short summary and overall relevance of the provided information on ozone layer hazard

Not relevant.

12.1.2 Comparison with the CLP criteria

Not relevant.

12.1.3 Conclusion on classification and labelling for hazardous to the ozone layer

Data lacking.

13 ADDITIONAL LABELLING

None.

14 REFERENCES

A reference list for the studies from the DAR is included below:

Physical and chemical properties (Annex IIA 2; Annex IIIA 2)

Annex point/ reference no.	**Author(s)**	**Year**	**Title** **Company, report no.** **Source (where different from company)** **GLP or GEP status (where relevant)** **Published or not**
IIA 2.1/01	Geoffroy, A	2007	Boiling point and melting point of emamectin benzoate Syngenta Crop Protection Münchwilen AG, Münchwilen, Switzerland, Study no: L07-000668 GLP Not published 14 June 2007
IIA 2.2/01 IIA 2.3/01 IIA 2.5/01 IIA 2.6/01 IIA 2.8/01 IIA 2.9/01	McCauley JA	1992	Determination of physical-chemical properties of MK-244 Novartis Crop Protection AG, Basel, Switzerland Merck & Co. Inc., Rahway NJ, United States, Report No PMLMK244001 GLP Not Published Syngenta File N° MK244/0047
IIA 2.4/01	Das R	2000	General physico-chemical properties of MK 244 tech. Novartis Crop Protection AG, Basel, Switzerland Novartis Crop Protection Münchwilen AG, Münchwilen, Switzerland, Report No 102780 GLP Not Published Syngenta File N° MK244/0216
IIA2.5/02	Oggenfuss P	1999	Report on spectra Novartis Crop Protection AG, Basel, Switzerland Novartis Crop Protection Münchwilen AG, Münchwilen, Switzerland, Report No 69803 GLP Not Published Syngenta File N° MK244/0175
IIA2.7/01	Kettner R	1999	Solubility in organic solvents of MK 244 Novartis Crop Protection AG, Basel, Switzerland Novartis Crop Protection Münchwilen AG, Münchwilen, Switzerland, Report No 79454 GLP Not Published Syngenta File N° MK244/0197

Annex point/ reference no.	**Author(s)**	**Year**	**Title** **Company, report no.** **Source (where different from company)** **GLP or GEP status (where relevant)** **Published or not**
IIA 2.10/01	Angly H	1999	Screening test for thermal stability and stability in air Novartis Crop Protection AG, Basel, Switzerland Institute of Safety and Security, Basel, Switzerland, Report No 1999.4085.TSA Project number 79559 GLP Not Published Syngenta File N° MK244/0198
IIA 2.11/01	Angly H	2000a	Flammability of solids Novartis Crop Protection AG, Basel, Switzerland Institute of Safety and Security, Basel, Switzerland, Report No 2000.4068.FLS Project number 102783 GLP Not Published Syngenta File N° MK244/0220
IIA 2.13/01	Angly H	2000b	Explosive properties Novartis Crop Protection AG, Basel, Switzerland Institute of Safety and Security, Basel, Switzerland, Report No 2000.4068.EXP Project number EZ910010 GLP Not Published Syngenta File N° MK244/0221
IIA 2.14/01	Martin N	2000	Surface tension Novartis Crop Protection AG, Basel, Switzerland Solvias AG, Basel, Switzerland, Report No PP-00/66T.SUR Sponsor's order no 102782 GLP Not Published Syngenta File N° MK244/0219
IIA 2.15/01	Angly H	2000c	Oxidizing properties of solids Novartis Crop Protection AG, Basel, Switzerland Institute of Safety and Security, Basel, Switzerland, Report No 2000.4068.OXP GLP Not Published Syngenta File N° MK244/0223

Toxicology and metabolism (Annex IIA 5; Annex IIIA 7)

Annex point/ reference no.	**Year**	**Title** **Company, report no.** **Source (where different from company)** **GLP or GEP status (where relevant)** **Published or not**
KIIA 5.1.1/01	1995	MK 244 - (3H)-MAB1a: Metabolism, Pharmacokinetic, profile, excretion, tissue distribution and biliary elimination in the rat GLP Not Published Syngenta File N° MK244/0102

Annex point/ reference no.	**Year**	**Title** **Company, report no.** **Source (where different from company)** **GLP or GEP status (where relevant)** **Published or not**
KIIA 5.1.1/02	1993	The Tissue Distribution, Metabolism, and Excretion of [14C]4"-Deoxy-4"-epimethylamino Avermectin B1a (MAB1a) Benzoate in Rats GLP Not Published Syngenta File N° MK244/0327
KIIA 5.1.2/01	2005a	Emamectin B1a: Disposition Study GLP, Not Published Syngenta File N° NOA426007/0002
KIIA 5.2.1.1/02	1994a	MK 244 - Acute oral toxicity study in rats GLP, Not Published Syngenta File N° MK244/0099
KIIA 5.2.1.1/03	1995	MK 244 (L-656,748-052S) - Acute oral toxicity study in rats. GLP, Not Published Syngenta File N° MK244/0100
KIIA 5.2.1.1/04	1993a	MK 244 - Fifteen-day acute oral bioequivalence study in female rats GLP, Not Published Syngenta File N° MK244/0033
KIIA 5.2.1.1/06 5.2.1.4/01	1992f	MK 244 - Acute oral and intraveous toxicity studies in mice and rats GLP, Not Published Syngenta File N° MK244/0082
KIIA 5.2.1.1/05 5.2.1.1/07	1992a	MK 244 - Exploratory acute oral toxicity in female mice and rats GLP Not Published Syngenta File N° MK244/0022
KIIA 5.2.1.1/08	1994a	MK 244 L-656,748 - Acute oral toxicity study in mice. GLP, Not Published Syngenta File N° MK244/0098
KIIA 5.2.1.1/09	1993b	MK 244 - Fifteen-day acute oral bioequivalence study in female mice GLP, Not Published Syngenta File N° MK244/0036
KIIA 5.2.1.1/01	2006a	Emamectin Technical - Acute Oral Toxicity Up and Down Procedure in Rats GLP, Not Published Syngenta File N° MK244/0623; T010796-05
KIIA 5.2.1.2/01	2006b	Emamectin Technical (MK244G) - Acute Dermal Toxicity Study in Rats GLP, Not Published Syngenta File N° MK244/0640; T010797-05
KIIA 5.2.1.2/02	1994b	MK 244 - Acute dermal toxicity study in rats GLP, Not Published Syngenta File N° MK244/0055
KIIA 5.2.1.2/03	2010	Emamectin Benzoate Technical - Acute Dermal Toxicity Study in Rats GLP, Not Published Syngenta File N° MK244/10173
KIIA 5.2.1.3/01	2006	Emamectin benzoate technical (MK244G) - 4-hour acute inhalation toxicity study in the rat GLP, Not Published Syngenta File N° MK244/0614
KIIA 5.2.1.3/02	1994a	MK 244 - An acute inhalation range-finding and toxicity study in the albino rat GLP, Not Published Syngenta File N° MK244/0039
KIIA 5.2.1.3/03	1994b	MK 244 - An acute inhalation toxicity study in the albino rat GLP, Not Published Syngenta File N° MK244/0032
KIIA 5.2.2.1/01	2006c	Emamectin Technical - Primary Skin Irritation Study in Rabbits GLP, Not Published Syngenta File N° MK244/0624; T010800-05

Annex point/ reference no.	**Year**	**Title** **Company, report no.** **Source (where different from company)** **GLP or GEP status (where relevant)** **Published or not**
KIIA 5.2.2.1/02	1992a	MK 243 - Primary dermal irritation in rabbits GLP, Not Published Syngenta File N° MK243/0009
KIIA 5.2.2.2/01	2006d	Emamectin Technical - Primary Eye Irritation Study in Rabbits GLP, Not Published Syngenta File N° MK244/0625; T010802-05
KIIA 5.2.2.2/02	1992b	MK 243 - Primary eye irritation in rabbits GLP, Not Published Syngenta File N° MK243/0008
KIIA 5.2.2.3/01	1992	MK 244 - Guinea pig sensitization test GLP, Not Published Syngenta File N° MK244/0024
KIIA 5.2.2.3/02	2006	Emamectin Benzoate Technical (MK244G) Skin Sensitisation - Local Lymph Node Assay In The Mouse GLP, Not Published Syngenta File N° MK244/0602
KIIA 5.3.1/01	1992	L-656,748: Three-week Dietary Range Finding Study in Rats TT# 88-046-0 Not GLP, Not Published Syngenta File N° MK244/0329
KIIA 5.3.2/01	1992a	MK 244 - Fourteen-week dietary toxicity study in rats GLP, Not Published Syngenta File N° MK244/0079
KIIA 5.3.2/02	1992b	MK 243 - Thirteen-week dietary toxicity study in mice GLP, Not Published Syngenta File N° MK243/0007
KIIA 5.3.2/03	1992c	MK 244 - Fifty-three week toxicity study in rats GLP, Not Published Syngenta File N° MK244/0037
KIIA 5.3.3/01	1994b	MK 244 - Fourteen-week oral toxicity study in dogs Novartis Crop Protection AG, Basel, Switzerland Merck Laboratories, Westpoint, United States, Report No TT 88-060-0 GLP, Not Published Syngenta File N° MK244/0083
KIIA 5.3.4/01	1992	MK 244 - 53-week toxicity study in dogs GLP, Not Published Syngenta File N° MK244/0030
KIIA 5.4.1/01	1992b	MK 244 - Microbial mutagenesis assay GLP, Not Published Syngenta File N° MK244/0076
KIIA 5.4.2/01	1993a	MK 244 - Assay for chromosomal aberrations *in vitro*, in Chinese hamster ovary cells GLP, Not Published Syngenta File N° MK244/0034
KIIA 5.4.3/01	1992c	MK 244 - V-79 mammalian cell mutagenisis GLP, Not Published Syngenta File N° MK244/0075
KIIA 5.4.4/01	1993b	MK 244 - Assay for chromosomal aberrations in mouse bone marrow GLP, Not Published Syngenta File N° MK244/0035
KIIA 5.4.5/01	1992d	MK 244 - *In vitro* alkaline elution/rat hepatocyte assay GLP, Not Published Syngenta File N° MK244/0074
KIIA 5.5.2/01	1994c	MK 244 - One-Hundred-five -week dietary carcingenicity toxicity study in rats GLP, Not Published Syngenta File N° MK244/0041

Annex point/ reference no.	Year	Title Company, report no. Source (where different from company) GLP or GEP status (where relevant) Published or not
KIIA 5.5.3/01	1994d	MK 244 - Seventy-Nine week dietary carcinogenicity study in mice. GLP, Not Published Syngenta File N° MK244/0028
KIIA 5.6.1/01	1992	MK 243 - Oral range-finding reproduction study in female rats GLP, Not Published Syngenta File N° MK243/0003
KIIA 5.6.1/02	1993	MK 244 - Two-generation dietary reproduction study in rats GLP, Not Published Syngenta File N° MK244/0040
KIIA 5.6.2/01	1992c	MK 243 - Oral developmental toxicity study in rats GLP, Not Published Syngenta File N° MK243/0015
KIIA 5.6.2/02	1992d	MK 243 - Oral developmental toxicity study in rabbits GLP, Not Published Syngenta File N° MK243/0014
KIIA 5.7.1/01	1992e	MK 243 - Acute oral neurotoxicity study in rats GLP, Not Published Syngenta File N° MK243/0012
KIIA 5.7.1/02	1992f	MK 243 - Acute oral neurotoxicity study in rats GLP, Not Published Syngenta File N° MK243/0010
KIIA 5.7.1/03	1992	MK 243 - Acute dermal neurotoxicity study in female rabbits GLP, Not Published Syngenta File N° MK243/0011
KIIA 5.7.2/01	1992d	MK 243 - Sixteen-day dietary neurotoxicity study in the CF-1 mouse GLP, Not Published Syngenta File N° MK243/0006
KIIA 5.7.2/02	1992e	MK 243 - Exploratory two-week dietary neurotoxicity study in mice GLP, Not Published Syngenta File N° MK243/0005
KIIA 5.7.2/03	1992g	MK 243 - Exploratory five-week neurotoxicity study in dogs GLP, Not Published Syngenta File N° MK243/0004
KIIA 5.7.2/04	1992e	MK 244 - Fourteen-week dietary neurotoxicity study in rats GLP, Not Published Syngenta File N° MK244/0023
KIIA 5.7.3/01	1993	MK 244 - Oral developmental neurotoxicity study in female rats GLP, Not Published Syngenta File N° MK244/0031
KIIA 5.10/01	1992f	MK-0243 Benzoate MTBE Solvate/MK-0243 Benzoate Monohydrate Bioequivalence Study in Dogs GLP, Not Published Syngenta File N° MK243/0018
KIIA 5.10/02	1992g	MK-0243 Bioequivalence Study of Benzoate and HCl Salts in Dogs GLP, Not Published Syngenta File N° MK243/0017
KIIA 5.10/03	2008	Abamectin, emamectin and ivermectin. Kinetic study in genotyped CF-1 mice with abamectin, emamectin & ivermectin. GLP, Not Published

Environmental fate and behaviour (Annex IIA 7; Annex IIIA 9)

Annex point/ reference number	**Author(s)**	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 7.1.1/01 KIIA 7.2.1/01	Hand, L.; Fleming, E.;	2006a	Emamectin-benzoate : Route and Rate of Degradation of NOA426007 in Three Soils, under Aerobic Laboratory Conditions, at 20°C Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T002559-04-REG 04JH008 GLP Not Published Syngenta File N° MK244/0532
KIIA 7.1.1/02 KIIA 7.2.1/02	Jungmann, V.; Nicollier, G.;	2006	Rate of Degradation of [14C]Emamectin B1A (14C-NOA 426007) in one Soil under Various Laboratory Conditions at 20 C .. Syngenta Crop Protection AG, Basel, Switzerland, Report No T000877-05 GLP Not Published Syngenta File N° MK244/0565
KIIA 7.1.1/03 KIIA 7.2.1/03	Clark, A.;	2003a	Aerobic Soil Metabolism of 14C-NOA426007 and 14C-NOA422390 Syngenta Crop Protection AG, Basel, Switzerland Syngenta Crop Protection, Inc., Greensboro, United States, Report No 1853-01 GLP Not Published Syngenta File N° MK244/0321
KIIA 7.1.1/04 KIIA 7.2.1/04	Chukwudebe, A.;	1994a	Aerobic soil metabolism of 14C-4"-Epimethylamino-4"-Deoxyavermectin B1a Benzoate (14C-MAB1a) Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No 618-244-93257 GLP Not Published Syngenta File N° MK244/0131
KIIA 7.1.1/05	Chukwudebe, A.;	1995a	Aerobic soil metabolism of 14C-4"-Epimethylamino-4"-Deoxyavermectin B1a Benzoate (14C-MAB1a): characterization of the unextractables residues in soil Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No MK-244-93257 GLP Not Published Syngenta File N° MK244/0132

Annex point/ reference number	**Author(s)**	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 7.1.3/01	Anderson, W.;	2003	Photodegradation of 14C-NOA-426007 and 14C-NOA-422390 on Soil under Artificial Light Syngenta Crop Protection AG, Basel, Switzerland Syngenta Crop Protection, Inc., Greensboro, United States, Report No 1854-01 GLP Not Published Syngenta File N° MK244/0322
KIIA 7.1.3/02	Chukwudebe, A.;	1994b	Photodegradation of 14C 4" Epimethylamino-4"-Deoxyavermectin B1a Benzoate (14C-MAB1a) on soil Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No 93845 GLP Not Published Syngenta File N° MK244/0133
KIIA 7.1.3/03	Crouch, L.S.;	1996	Assay and characterization of polar photodegradates of MK244 and 14C-MK244 Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No 93692 GLP Not Published Syngenta File N° MK244/0135
KIIA 7.2.3/01	Webb, J.; Oliver, R.;	2006	Emamectin-benzoate: Rate of Degradation of Soil Metabolite NOA438306 in Three Soils under Laboratory Conditions Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T005295-05-REG 05JH029 GLP Not Published Syngenta File N° MK244/0529
KIIA 7.2.3/02	Jungmann, V.;	2006	Rate of Degradation of NOA459720 (Metabolite of NOA426007, Emamectin) in Various Soils under Aerobic Laboratory Conditions at 20 °C .. Syngenta Crop Protection AG, Basel, Switzerland, Report No T014559-05 GLP Not Published Syngenta File N° NOA459720/0007

Annex point/ reference number	**Author(s)**	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 7.3.1/01	Evans, P.;	2006a	Emamectin Benzoate 05SG Formulation (A10324A) - Dissipation in or on Soil in Northern France 2005 - Final Report Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T000885-05-REG 05-5029 GLP Not Published Syngenta File N° MK244/0649
KIIA 7.3.1/02	Evans, P.;	2006b	Emamectin Benzoate 05SG Formulation (A10324A) - Dissipation in or on Soil in Southern France 2005 - Final Report Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T000887-05-REG 05-5030 GLP Not Published Syngenta File N° MK244/0650
KIIA 7.3.1/03	Seville, A.;	2006a	Dissipation Study with A10324A, Emamectin Benzoate 05SG Formulation, in or on Cultivated Soil in Southern France - Final Report Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T000886-05-REG 04-5013 GLP Not Published Syngenta File N° MK244/0634
KIIA 7.3.1/04	Seville, A.;	2006b	Emamectin: Residues in/on soil, Germany 2004 (Test product: A10324A) - Final Analytical Phase Report Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T000884-05-PHA2 gbg704004 GLP Not Published Syngenta File N° MK244/0633
KIIA 7.4.1/01	Wyeth, K; Ricketts, D.;	2005	14C-Emamectin Benzoate B1a (NOA426007)Adsorption/Desorption Properties in Soil Syngenta Crop Protection AG, Basel, Switzerland Syngenta, Jealott's Hill, United Kingdom, Report No RJ3566B 04JH009 GLP Not Published Syngenta File N° MK244/0381

Annex point/ reference number	**Author(s)**	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 7.4.1/02	Mushtaq, M.; Fellow, R.;	1993	Sorption and desorption of (3H)4"-Deoxy-4"-epimethylamino Avermectin B1a (MAB1a) benzoate with soils Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No ENC-5 GLP Not Published Syngenta File N° MK244/0155
KIIA 7.4.2/01	Voelkel, W.;	2005	Emamectin: Adsorption/Desorption of [23-14C]-8,9-Z Emamectin B1a Benzoate Salt, a Metabolite of Emamectin, on 3 soils Syngenta Crop Protection AG, Basel, Switzerland RCC Ltd., Itingen, Switzerland, Report No 857093 T002555-04 GLP Not Published Syngenta File N° MK244/0384
KIIA 7.4.2/02	Hand, L.; Fleming, E.;	2006b	Emamectin-benzoate : Adsorption/Desorption Properties of a Metabolite (NOA438306) in Three Soils Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T002556-04-REG GLP Not Published Syngenta File N° MK244/0495
KIIA 7.4.2/04	Indergand, P.; Nicollier, G.;	2006	Adsorption / Desorption of 14C-NOA459720 (metabolite of NOA426007, Emamectin B1a) on Gartenacker, Marsillargues and 18-Acres Soils .. Syngenta Crop Protection AG, Basel, Switzerland, Report No T014558-05 GLP Not Published Syngenta File N° NOA459720/0006
KIIA 7.4.2/05	Jungmann, V.;	Unsp	NOA415692 - Adsorption/Desorption of NOA415692 (metabolite of NOA426007, emamectin benzoate B1a) on Gartenacker, Marsillargues and 18-Acres Soils .. Syngenta Crop Protection AG, Basel, Switzerland, Report No T014558subB-05 GLP Not Published Syngenta File N° NOA415692/0002

Annex point/ reference number	**Author(s)**	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 7.4.5/01	Reynolds, J.L.;	1995	Aged column leaching of 14C-labeled 4"-Deoxy-4"-Epimethylamino Avermectin B1a (MAB1a) Benzoate in four soils Novartis Crop Protection AG, Basel, Switzerland Report No RPT00233 GLP Not Published Syngenta File N° MK244/0128
KIIA 7.4.6/01	Feely, W.F.;	1992	Soil thin-layer chromatography (TLC) of 14C-MK0244 Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No PMES ENC GLP Not Published Syngenta File N° MK244/0140
KIIA 7.5/01	Chukwudebe, A.;	1992	Hydrolysis of 4" Deoxy-4" Epimethylamino Avermectin B1a Benzoate as a function of pH at 25C (ENC-3) Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No ENC-3 GLP Not Published Syngenta File N° MK244/0141
KIIA 7.6/01	Ballantine, L.;	1994	Artificial sunlight Photolysis of 14C 4" Epimethylamino-4"-Deoxyavermectin B1a Benzoate (14C-MAB1a) in aqueous media Novartis Crop Protection AG, Basel, Switzerland Hazleton Laboratories, Madison, United States, Report No 618-244-93444 GLP Not Published Syngenta File N° MK244/0134
KIIA 7.6/02	Mushtaq, M.;	1995	Photodegradation of 14C 4" Deoxy-4" Epimethylaminoavermectin B1a (MAB1a) Benzoate in aqueous media Novartis Crop Protection AG, Basel, Switzerland Merck Research Laboratories, Three Bridges, United States, Report No ENC-6 GLP Not Published Syngenta File N° MK244/0130

Annex point/ reference number	**Author(s)**	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 7.6/03	Phaff, R.;	2005	Determination of 14C-Emamectin B1A (14C-NOA 426007) photolysis quantum yield Syngenta Crop Protection AG, Basel, Switzerland RCC Ltd., Itingen, Switzerland, Report No RCC 856664 T002561-04 GLP Not Published Syngenta File N° MK244/0382
KIIA 7.7/01	Dietschy, A.;	1999	Ready biodegradability of MK 244 (Emamectin benzoate) Novartis Crop Protection AG, Basel, Switzerland Novartis Services AG, Basel, Switzerland, Report No G 582 06 GLP Not Published Syngenta File N° MK244/0184
KIIA 7.8.3/01	Hurt, A.; Grosjean, J.; Mason, G.;	2006	Emamectin-benzoate: Aerobic Degradation of NOA426007 in Two Aquatic Sediment Systems Under Laboratory Conditions Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, Report No T000308-06-REG 04JH026 GLP Not Published Syngenta File N° MK244/0539
KIIA 7.8.3/02	Clark, A;	2003b	Aerobic Aquatic Metabolism of [23-14C]-NOA-426007 .. Syngenta Crop Protection, Inc., Greensboro, United States, Report No 52-02 GLP Not Published Syngenta File N° MK244/0320
KIIA 7.10/01	Stamm, E.;	1998	Atmospheric oxidation of MK-244 (abamectin) by hydroxyl radicals; rate estimation Novartis Crop Protection AG, Basel, Switzerland Novartis Crop Protection AG, Basel, Switzerland, Report No 98SM04 GLP Not Published Syngenta File N° MK244/0153

Ecotoxicology (Annex IIA 8; Annex IIIA 10)

Annex point/ reference number	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 8.1.1/01	1993	MK-244: a acute oral toxicity study with the Bobwhite GLP Not Published Syngenta File N° MK244/0125
KIIA 8.1.1/02	1992	MK-244: an acute oral toxicity study with the Mallard GLP Not Published Syngenta File N° MK244/0139
KIIA 8.1.2/01	1993	MK-244: a dietary LC50 study with the northern Bobwhite GLP Not Published Syngenta File N° MK244/0124
KIIA 8.1.2/02	1993	MK-244: a dietary LC50 study with the Mallard GLP Not Published Syngenta File N° MK244/0106
KIIA 8.1.4/01	1995	MK-244: a reproduction study with the northern Bobwhite Novartis Crop Protection AG, Basel, Switzerland Wildlife International Ltd., Easton, MD, United States, Report No 105-153 GLP Not Published Syngenta File N° MK244/0114
KIIA 8.1.4/02	1995	MK-244: a reproduction study with the Mallard GLP Not Published Syngenta File N° MK244/0115
KIIA 8.2.1.1/01	1993	MK-244: a 96-hour flow through acute toxicity test with the rainbow trout (*Oncorhynchus mykiss*) GLP Not Published Syngenta File N° MK244/0119
KIIA 8.2.1.2/01	1993a	MK-244: a 96-hour flow through acute toxicity test with the bluegill (*Lepomis macrochirus*) GLP Not Published Syngenta File N° MK244/0138
KIIA 8.2.1.2/02	1995a	MK-244: a 96-hour flow-through acute toxicity test with the fathead minnow (*Pimephales promelas*) GLP Not Published Syngenta File N° MK244/0107
KIIA 8.2.1.2/04	1994	MK-244: a 96-hour flow through toxicity test with the sheepshead minnow (Cyprinodon variegatus) GLP Not Published Syngenta File N° MK244/0122
KIIA 8.2.4/01	1995b	MK-244: an early life stage test with the fathead minnow (*Pimephales promelas*) GLP Not Published Syngenta File N° MK244/0110

Annex point/ reference number	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 8.2.6/01	1994a	MK-244: a bioconcentration test with the Bluegill (*Lepomis macrochirus*) GLP Not Published Syngenta File N° MK244/0120
KIIA 8.3.1.1/01	1993b	MK-244: a 48-hour flow through acute toxicity test with the cladocera (*Daphnia magna*) GLP Not Published Syngenta File N° MK244/0137
KIIA 8.3.1.1/02	2006	Emamectin benzoate metabolite (NOA438306): Acute toxicity to *Daphnia magna* in a 48-hour immobilization test Syngenta Crop Protection AG, Basel, Switzerland RCC Ltd., Itingen, Switzerland, Report No A42974 2032811 GLP Not Published Syngenta File N° NOA438306/0003
KIIA 8.3.1.1/03	1995	MK-244: a 96-hour shell deposition test with the eastern oyster (Crassostrea virginica) GLP Not Published Syngenta File N° MK244/0123
KIIA 8.3.1.1/04	1995	MK-244: a 96-hour flow through acute toxicity test with the saltwater mysid (Mysidopsis bahia) GLP Not Published Syngenta File N° MK244/0117
KIIA 8.3.2.1/01	1994b	MK-244: a flow through life-cycle toxicity test with the cladoceran (*Daphnia magna*) GLP Not Published Syngenta File N° MK244/0121
KIIA 8.3.3/01	2005	MK244 (Emamectin benzoate) 50 g/kg SG (A10324A): Effects on aquatic organisms in an outdoor microcosm GLP Not Published Syngenta File N° MK244/0425
KIIA 8.3.3/02	2006	Evaluation of the report "MK244 (Emamectin benzoate) 50 g/kg SG (A10324A): Effects on aquatic organisms in an outdoor Microcosm" Not GLP Not Published Syngenta File N° MK244/0561
KIIA 8.4/01	1995	MK-244: a 5-day toxicity test with the freshwater alga (*Selenastrum capricornutum*) GLP Not Published Syngenta File N° MK244/0109
KIIA 8.4/02	2003b	Emamectin benzoate (MK244): Toxicity to the green alga *Selenastrum capricornutum* GLP Not Published Syngenta File N° MK244/0317

Annex point/ reference number	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 8.5.2/01	2006	Emamectin benzoate (MK244 technical): Effects on the development of sediment-dwelling larvae of *Chironomus riparius* in a water-sediment system GLP Not Published Syngenta File N° MK244/0487
KIIA 8.6/01	1995	MK-244: a 14-day toxicity test with duckweed (*Lemna gibba* G3) GLP Not Published Syngenta File N° MK244/0108
KIIA 8.7/01	2000	Assessment of side-effects of MK 244 SG 5 (A-10324 A) on the bumble-bees (*Bombus terrestris* L.) in greenhouse compartments Not GLP Not Published Syngenta File N° MK244/0215
KIIA 8.7.1/01	2001	Acute toxicity of MK 244 05 SG (A-10324 A) to the honeybee *Apis mellifera* L. under laboratory conditions GLP Not Published Syngenta File N° MK244/0261
KIIA 8.7.2/01	1993	MK-244: a acute contact toxicity study with the honey bee (*Apis mellifera* L.) GLP Not Published Syngenta File N° MK244/0118
KIIA 8.7.3/01	1994	MK-244: a foliage residue toxicity study with the honey bee (*Apis mellifera* L.) GLP Not Published Syngenta File N° MK244/0116
KIIA 8.7.3/02	2004	Emamectin - A Foliage Residue Toxicity Study with the Honey Bee GLP Not Published Syngenta File N° MK244/0339
KIIA 8.8.1.1/01	2004a	MK 244 (emamectin benzoate): a rate-response laboratory test to evaluate the effects of a 50 g/kg SG formulation (A10324A) on the parasitic wasp *Aphidius rhopalosiphi* GLP Not Published Syngenta File N° MK244/0332
KIIA 8.8.1.2/01	2004a	MK 244 (emamectin benzoate): A rate-response laboratory test to evaluate the effects of a 50 g/kg SG formulation (A10324A) on the predatory mite *Typhlodromus pyri* (Acari: Phytoseiidae) GLP Not Published Syngenta File N° MK244/0352
KIIA 8.8.2.1/01	2004b	MK 244 (emamectin benzoate): a rate-response extended laboratory test to evaluate the effects of a 50 g/kg SG formulation (A10324A) on the parasitic wasp *Aphidius rhopalosiphi* GLP Not Published Syngenta File N° MK244/0338

Annex point/ reference number	**Year**	**Title** **Sponsor/Source** **Test Facility, Report No** **GLP or GEP status (where relevant) Published or Not** **Syngenta File No.**
KIIA 8.8.2.1/02	2004c	MK244 (emamectin benzoate): An aged residue extended laboratory test to determine the effects of a 50 g/kg SG formulation (A10324A) on a parasitic wasp *Aphidius rhopalsiphi*. GLP Not published Syngenta File No. MK244/0355
KIIA 8.8.2.2/01	2004b	MK 244 (emamectin benzoate): A rate-response extended laboratory test to evaluate the effects of a 50 g/kg SG formulation (A10324A) on the predatory mite *Typhlodromus pyri* (Acari: Phytoseiidae) GLP Not Published Syngenta File N° MK244/0353
KIIA 8.8.2.2/02	2005	MK244 (emamectin benzoate): An aged residue extended laboratory test to evaluate the effects of a 50 g/kg SG formulation (A10324A) on the predatory mite *Typhlodromus pyri* GLP Not Published Syngenta File N° MK244/0356
KIIA 8.8.2.4/01	2004	MK 244 (emamectin benzoate): A rate-response extended laboratory test to evaluate the effects of a 50 g/kg SG formulation (A10324A) on the predatory bug *Orius insidiosus* (Hemiptera: Anthocoridae) GLP Not Published Syngenta File N° MK244/0354
KIIA 8.8.2.4/02	2005	MK244 (emamectin benzoate): An extended laboratory test to determine the effects of a 50 g/kg SG formulation (A10324A) on the green lacewing *Chrysoperla carnea* (Neuroptera, Chrysopidae) GLP Not Published Syngenta File N° MK244/0376
KIIA 8.9.1/01	1999	Acute toxicity of MK 244 to earthworm Eisenia fetida GLP Not Published Syngenta File N° MK244/0192
KIIA 8.9.1/02	2006	NOA438306: Acute toxicity to the earthworm Eisenia fetida GLP Not Published Syngenta File N° NOA438306/0001
KIIA 8.12/01	2006	Emamectin (MK244) 0.95 SG formulation (A14605A): Herbicide profiling test to evaluate the phytotoxicity to terrestrial (non-target) higher plants Not GLP Not Published Syngenta File N° MK244/0456
KIIA 8.15/01	1999	Report on the test for activated sludge respiration inhibition of MK 244 A GLP Not Published Syngenta File N° MK244/0182
KIIA 8.16.2/01	2006	Analytical Phase for Emamectin-benzoate (MK244) in Arthropods and Vegetation GLP Not Published Syngenta File N° MK244/0563

15 ANNEXES

The study summaries from the DAR of emamectin have been included in Annex I.