98/8 Doc IIIA section No.	6.1.3 / 01	Acute toxicity – Inhalation	Official use only
91/414 Annex Point addressed	II 5.2.3 / 01	Acute toxicity - inhalation	

Title:	MK 936 tech: Single Exposure (Nose-only) Toxicity Study in the Rat	
Lab Report Number:	No. 1992/1-D6144 (Syngenta No. MK936/0810),	
Authors:	[redacted]	
Test Substance:	MK 936 tech., batches № [redacted] (purity [redacted]), [redacted] (purity [redacted]) and [redacted] (purity [redacted])	
Species:	Rat	
Guidelines:	92/69/EEC B.2, OECD 403, OPPTS 870.1300; Deviations: none	
Date of Report:	07.12.2001	
Published:	No	
GLP:	Yes	

Characteristics

Reference/notifier	:	[redacted] (2001a)	Exposure	:	4 h (nose only)
Type of study	:	Acute inhalation toxicity study	Dose	:	0.21, 1.78 and 5.25 mg/L (MMAD 4.2, 3.7 and 2.7 resp., GSD 3.8, 4.9 and 3.4 resp.)
Year of execution	:	2001	Vehicle	:	-
Test substance	:	Abamectin technical (purity [redacted] and [redacted]	GLP statement	:	yes
Route	:	inhalation	Guideline	:	In accordance with OECD 403
Species	:	Rat (Crl:Han Wist)	Acceptability	:	acceptable
Group size	:	5/sex/dose	LC50 rats	:	< 0.21 mg/L

Study design

The study is in accordance with OECD 403.

Results

Mortality: There was 100% mortality in all dose groups. Death or moribund sacrifice occured during exposure or within 2 h for the 5.25 mg/L animals, within 5 h for the 1.78 mg/L animals and by the day following exposure for the 0.21 mg/L animals.

Symptoms of toxicity: tremors, rigid tail and prostrate body position were observed in all animals, whereas signs observed in some animals included ataxia, cyanosis, subdued behaviour, piloerection, noisy respiration, coloured tears, staining of the eye and fur, squinting, wet fur, vocalisation on handling and tail flicking.

Body weight: Body weight analysis was not appropriate due to the early termination of the rats.

Pathology: dark, darkened or red areas in the lungs were observed in all dose groups, inflated lungs and firmness along the length of the tail was noted for all 5.25 mg/L animals.

Acceptability

The study is considered acceptable.

Conclusions

The acute 4-hour inhalatory LC_{50} in rats is <0.21 mg/L.

Reliability Indicator	1	
Data Protection Claim	Yes	

98/8 Doc IIIA section No.	6.1.3 / 02	Acute toxicity – Inhalation	Official use only
91/414 Annex Point addressed	II 5.2.3 / 02	Acute toxicity - inhalation	

Title:	Abamectin: 4-Hour acute inhalation Toxicity Study in the Rat.	
Lab Report Number:	Not applicable	
Authors:	[redacted]	
Test Substance:	MK 936 tech., batches № [redacted] (purity [redacted]), [redacted] (purity 8[redacted]) and [redacted] (purity [redacted])	
Species:	Rat	
Guidelines:	92/69/EEC B.2, OECD 403, OPPTS 870.1300; Deviations: none	
Date of Report:	2003	
Published:	No	
GLP:	Yes	

Characteristics

Reference/notifier	:	[redacted] (2003)	Exposure	:	4 h (nose only)
Type of study	:	Acute inhalation toxicity study	Dose	:	m + f: 0.051 mg/L (MMAD 2.11, 2.29; GSD 1.69, 1.83); f: 0.034 mg/L (MMAD 2.80, 2.57; GSD 1.73, 1.70)
Year of execution	:	2003	Vehicle	:	-
Test substance	:	Abamectin technical (purity [redacted])	GLP statement	:	yes
Route	:	inhalatoir	Guideline	:	In accordance with OECD 403
Species	:	Rat (Alp:APfSD)	Acceptability	:	acceptable
Group size	:	5/sex (0.05 mg/L) and 5f (0.03 mg/L)	LC50 rats	:	>0.051 mg/L (m); >0.034 mg/L and <0.051 mg/L (f)

Study design

The study is in accordance with OECD 403, with the following deviation: there are only 2 concentrations tested, and exposure to 0.03 mg/L was performed with 5 females only, pathology was not performed.

Results

Mortality: In the 0.05 mg/L group, one female was found dead and 2 females were killed on day 2 due to the severity of the clinical signs on day 2.

Symptoms of toxicity: reduced splay reflex, prostrate and tip toe gait, shaking, comatose, increased response to touch, reduced stability, decreased visual placing response, abnormal respiratory noise, increased breathing depth were observed in animals of the 0.051 mg/L group. In the 0.034 mg/L group, abnormal respiratory noise was observed staining of the oral and nasal cavities and eye discharge, wet fur, hunched posture, piloerection and chromodacryorrhea. Full recovery was apparent by day 4 for surving females and for males by day 15.

Body weight: normal

Pathology: not performed

Acceptability

The study is considered acceptable.

Conclusions

The acute 4-hour inhalatory LC_{50} in rats is >0.051 mg/L for males and between 0.034 mg/L and 0.051 mg/L for females.

SYNGENTA CONCLUSION

Conclusion:	The inhalation LC_{50} of Abamectin technical in rats was >0.051 mg/l for males and >0.034 mg/l for females. According to Council Directive 67/548/EEC, Abamectin technical should be classified as R26 'Very toxic by inhalation'. Comment: Abamectin technical has a very low vapour pressure of less than 3.7×10^{-6} Pa at 25°C (trigger value of the European Community according to guideline 91/414/EEC is 1×10^{-2} Pa). Therefore it is unlikely that inhalation exposure occurs because of evaporation of	

	Abamectin. Furthermore, the physico-chemical characteristics of Abamectin, a product obtained by fermentation as a solid "wet cake", do not indicate an inhalation hazard. Therefore, there is no risk associated with inhalation exposure and in compliance with 91/414/EEC the herewith presented acute inhalation toxicity study is considered inappropriate for acute hazard classification.	

Reliability Indicator	1	
Data Protection Claim	Yes	

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	*5 November 2007; updated January 2009*
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion. Discuss if deviating from view of*

	rapporteur member state
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

98/8 Doc IIIA 6.1.4 / 01 section No.	Acute toxicity – Skin and eye irritation	Official use only
91/414 Annex II Point addressed 5.2.4 / 01	Acute toxicity - skin irritation	

Title:	MK-936 primary skin irritation in rabbits	
Lab Report Number:	No. TT 81-2941	
Authors:	[redacted]	
Test Substance:	Technical grade Abamectin (batch no. [redacted], purity of batch not reported but specified elsewhere as [redacted] by HPLC)	
Species:	Rabbit	
Guidelines:	In house method that deviates from OECD guideline no. 404 (July 1992) and 92/69/EEC, B.4 in the following respects: Six rabbits, each exposed on intact and abraded skin, were used since these are regulatory requirements of EPA-FIFRA. Adjacent untreated skin sites not used as reference. Test article was applied as a dry powder, without wetting. Exposure period was 24 hours. Skin sites were examined immediately after patch removal and then daily for 14 days. Grading system for dermal reactions not specified.	
Date of Report:	2 July 1981	
Published:	No	
GLP:	Yes	

Characteristics

Reference/notifier	:	[redacted] (1981b)	Exposure	:	24 h
Type of study	:	Skin irritation study	Dose	:	500 mg
Year of execution	:	1981	Vehicle	:	-
Test substance	:	Abamectin technical (purity [redacted]	GLP statement	:	yes
Route	:	dermal	Guideline	:	-
Species	:	rabbit	Acceptability	:	acceptable
Group size	:	3/sex	Effect	:	Not irritating to skin

Note: this study evaluated the dermal irritancy of abamectin technical, an abamectin formulation and its vehicle. This summary refers only to the data on abamectin technical.

Study design

The method used deviates from OECD 404 in the following respects: 3/sex rabbits were exposed on both intact and abraded skin, untreated skin sites were not used as a reference, the test substance was applied as a dry powder without wetting, the exposure period was 24 h, the skin sites were examined immediately after patch removal and then daily for 14 days, grading system for dermal reactions was not specified, no individual data were presented, no data on body weight were presented.

Results

No skin irritation was observed in any of the rabbits.
One animal died on day 8, without signs of toxicity (cause of death unknown). Loss of appetite occurred after 2 days, and 5 animals showed slight body weight loss at 7 and 14 days.

Acceptability

The study is considered acceptable.

Conclusions

Abamectin does not need to be classified as irritating to skin.

Reliability Indicator	1	
Data Protection Claim	Yes	

98/8 Doc IIIA 6.1.4 / 02 section No.	Acute toxicity – Skin and eye irritation	Official use only
91/414 Annex II Point addressed 5.2.5 / 01	Acute toxicity - eye irritation	

Title:	Primary eye irritation/corrosion study of Abamectin technical in rabbits	
Lab Report Number:	No. 91104843	
Authors:	[redacted]	
Test Substance:	Abamectin technical ([redacted], batch no. [redacted], purity [redacted] [redacted] avermectin B1a and [redacted] avermectin B1b])	
Species:	Rabbit	
Guidelines:	OECD Guideline 405 "Acute Eye Irritation/Corrosion" Deviations: 6 rabbits instead of 3 were used; the eyes of 3 rabbits remaining unwashed (regulatory requirement in US) and the eyes of 3 animals were washed 30 seconds after instillation of test article.	
Date of Report:	8 February 2000	
Published:	No	
GLP:	Yes	

Characteristics

Reference/notifier	:	[redacted] (2000)	Exposure	:	Single instillation in conjunctival sac
Type of study	:	Eye irritation study	Dose[1]	:	37 mg (0.1 ml)
Year of execution	:	1999	Vehicle	:	-
Test substance	:	Abamectin technical (purity [redacted])	GLP statement	:	yes
Route	:	ocular	Guideline	:	In accordance with OECD 405

Species	:	rabbit	Acceptability	:	acceptable
Group size	:	2m/4f	Effect	:	Not irritating to eyes

1: the test material was described as a white powder. No information is available on the vehicle used, and no justification is given for a dose of 37 mg.

Study design

The study is in accordance with OECD 405, with the following deviations: only a dose of 37 mg was used in this study, and no information is given on the vehicle used.

Both eyes of 3 animals were flushed with water for one minute starting 30 seconds after instillation of the test substance. The eyes of the other 3 animals remained unwashed.

Results

The results are summarized in Table below.

Individual eye irritation score in unwashed eyes

	1 h	24 h	48 h	72 h
Cornea/opacity	0, 0, 0	0, 0, 0	0, 0, 0	0, 0, 0
iris	0, 0, 0	0, 0, 0	0, 0, 0	0, 0, 0
Conjunctivae redness	1, 1, 1	0, 0, 0	0, 0, 0	0, 0, 0
Conjunctivae chemosis	0, 1, 0	0, 0, 0	0, 0, 0	0, 0, 0
Conjunctivae discharge	0, 0, 0	0, 0, 0	0, 0, 0	0, 0, 0

Acceptability

The study is considered acceptable.

Conclusions

Abamectin does not need to be classified as irritating to eyes.

Reliability Indicator	1	
Data Protection Claim	Yes	

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
Date	**EVALUATION BY RAPPORTEUR MEMBER STATE** *5 November 2007; updated January 2009*

Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
	[redacted]
	[redacted]
Remarks	

	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion. Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

98/8 Doc IIIA section No. 6.1.5 / 01	Acute toxicity – Skin sensitisation	Official use only
91/414 Annex Point addressed	II 5.2.6 / 01	Acute toxicity - skin sensitisation	

Title:	MK 936 tech. - Skin Sensitisation Study in the Guinea pig	
Lab Report Number:	No. 20002053	
Authors:	[redacted]	
Test Substance:	Test material: MK 936 tech.; Batch No. [redacted], purity [redacted].	
Species:	Guinea Pigs	
Guidelines:	96/54/EEC Method B6; OECD 406; OPPTS 870.2600 and J-MAFF	
Date of Report:	26.02.2001	
Published:	No	
GLP:	Yes	

Characteristics

Reference/notifier	:	[redacted] (2001b)	Exposure	:	Intradermal and topical induction, topical challenge
Type of study	:	Skin sensitization study (GPMT)	Dose[2]	:	5% intradermal injection, 55% topical induction, 40% challenge
Year of execution	:	2000	Vehicle	:	Arachis oil
Test substance	:	Abamectin technical (purity [redacted])	GLP statement	:	yes
Route	:	dermal	Guideline	:	In accordance with OECD 406
Species	:	Guinea pig	Acceptability	:	acceptable
Group size[1]	:	19 test animals (f), 10 controls (f)	Effect	:	Not sensitizing

1one test animal was sacrificed on day 1 before treatment.

2 test concentrations were based on the results from screening tests.

Study design

The study is in accordance with OECD 406.

Results

After 48 h, slight erythema was observed in 7 test animals, but also at the vehicle site in 4 of these animals. Slight erythema at the test site was observed in one control animal after 24 h, and in 3 animals after 48 h. In the control group slight reactions were observed at the vehicle site in one animal after 24 h and 48 h. Desquamation was observed in both test and control animals at test site and vehicle site.

Acceptability

The study is considered acceptable.

Conclusions

Under the conditions of the GPMT test, abamectin was not sensitizing and does not need to be classified as sensitizing.

Reliability Indicator	1	
Data Protection Claim	Yes	

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	*5 November 2007; updated January 2009*
Materials and Methods	[redacted]
Results and discussion	
Conclusion	
Reliability	
Acceptability	
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion. Discuss if deviating from view of rapporteur member state*

Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Summary of acute toxicity

Abamectin is acutely toxic. From the range of data available, the most modern guideline acceptable study design resulted in a median lethal dose estimation of 221 mg/kg bw for the rat (combined sexes). On basis of this result the compound should be classified as R22 'Harmful if swallowed' according to Commission Directive 93/21/EEC. The acute toxic dose via the topical route was investigated in rats and rabbits. The LD50 was greater than the limit dose of 2000 mg/kg bw and therefore no classification is relevant for dermal toxicity of Abamectin.

The test substance was tested in two inhalation studies confirming the LC50 value to lie between 0.034 and 0.051 mg/L, therefore requiring classification as R26 Very toxic by inhalation.

Indications of dermal or ocular irritation were transient and graded as mild. Because the mean values of the readings were below the thresholds defined in the Directive 93/21/EEC, no classification for dermal or ocular irritancy is required.

In a Maximisation test according to Magnusson and Kligman no sensitisation effects were detected and therefore no classification is necessary.

In an antidote investigation in dogs, Ipecac administered within 15 minutes of ingestion of 8 mg/kg abamectin technical prevented coma and death and reduced the incidence and/or severity and/or duration of typical clinical signs of abamectin intoxication, (mydriasis, ataxia, tremors and convulsions). However, administration of charcoal and ipecac more than 15 minutes after abamectin technical ingestion were ineffective in reducing abamectin-induced toxicity.

Summary from abamectin PPP DAR. This is included for information.

The results of the acute toxicity studies, the irritation studies and the sensitization study, which are suitable for evaluation in the context of Annex II are presented in tables below.

Acute toxicity studies

Test substance	LD_{50}/LC_{50} (mg/kg bw or mg/L)	Species	Route	Notifier	Reference
Abamectin technical (vehicle sesame oil)	m: 8.7 f: 12.8	Rat / CD	Oral	Syngenta	[redacted] (1981c)
Abamectin technical (vehicle methylcellulose/water)	m: 232 f: 214	Rat / CD	Oral	Syngenta	[redacted] (2001)
Abamectin technical (vehicle saline)	>330	Rat / CD	dermal	Syngenta	[redacted] (1985a)
Abamectin technical (vehicle saline)	>2000	Rabbit	dermal	Syngenta	[redacted] (1984g)
Abamectin technical	<0.21	Rat/Crl:Han Wist	inhalatory	Syngenta	[redacted] (2001)
Abamectin technical	m: >0.051 f: >0.034 and <0.051	Rat/Alpk:APfSD	inhalatory	Syngenta	[redacted] (2003)

Studies on metabolites

Study/species dose levels	Test article	NOAEL (mg/kg bw)	LOAEL (mg/kg bw)	Major effects	Reference
Acute oral toxicity; CF-1 strain mice	Polar metabolite Non-polar metabolite(8,9-Z-isomer)	- LD50 > 48		Reduced activity and bradypnea.	[redacted] (1984f)
Acute oral toxicity; CF-1 strain mouse	8,9-Z isomer	LD50 >80mg/kg		Decreased activity, bradypnea, ataxia, ptosis and death.	[redacted] (1986c)
Exploratory acute oral toxicity; CF-1 & CD-1 mice	8,9-Z isomer	LD50 = 217mg/kg in CD-1 females LD50 = >10 and <20mg/kg in CF-1 males	50 mg/kg in CD-1 females <10 mg/kg in CF-1 males	Decreased activity, bradypnea, tremors, urine staining, hunched posture and death. Decreased activity, bradypnea, ptosis, tremors, urine staining and death.	[redacted] (1996)

Other acute toxicity studies

Study/ species dose levels	NOAEL (mg/kg bw/day)	LOAEL (mg/kg bw/day)	Effects at LOEL	Reference
Acute oral toxicity; Pregnant / non-pregnant CF-1 mice	LD50 non-pregnant mice: 15.0 mg/kg bw LD50 pregnant mice: 11.8 mg/kg bw	< 5 < 5	Death, loss of righting reflex, bradypnea Death, tremors, bradypnea, clonic convulsions	[redacted] (1986h)

Acute oral toxicity; Pregnant / non-pregnant CF-1 mice	LD50 non-pregnant mice: >20 and <40 mg/kg bw LD50 pregnant mice: 19 mg/kg bw	< 5 5	Deaths, tremors, bradypnea Deaths, tremors, bradypnea	[redacted] (1986h)
Exploratory acute oral toxicity; CF-1 mice of known genotype for P-glycoprotein	LD50 (+/+ genotype female mice): 28 mg/kg bw LD50 (+/- genotype female mice): 14 mg/kg bw	< 10 < 10	Tremors, bradypnea, decreased activity. Tremors, bradypnea, decreased activity, weight loss during first week	[redacted] (1997)
Exploratory oral non-specific antidote study; Dog; 8 mg/kg bw MK-0936 + 30 ml ipecac (15min) or 3 g charcoal (30min)	Results: Ipecac administered within 15 minutes of abamectin ingestion prevented coma and death, and reduced incidence and/or severity of mydriasis, ataxia, tremors and convulsion Charcoal, or ipecac administered more than 15 minutes after ingestion, were ineffective in reducing abamectin-induced toxicity			[redacted] (1984h)

Skin and eye irritation studies

Test substance	Effect	Species	Route	Notifier	Reference
Abamectin technical	Not irritating to skin	Rabbit / NZW	Dermal	Syngenta	[redacted] (1981a)
Abamectin technical	Not irritating to eyes	Rabbit / NZW	Ocular	Syngenta	[redacted] (2000)

Skin sensitization studies

Test substance	Effect	Species	Route	Notifier	Reference
Abamectin technical (vehicle Arachis oil)	negative	Guinea pig / Hartley	Intradermal and topical	Syngenta	[redacted] (2001)

Abamectin technical is very toxic to the rat by oral administration in sesame oil (LD50 values 8.7 and 12.8 mg/kg in males and females, respectively). A subsequent study with an aqueous vehicle showed that abamectin was significantly less toxic orally with this vehicle (LD50 values 232 and 214 mg/kg in males and females, respectively).

Abamectin technical is very toxic by the inhalation route (LC50>0.051 mg/L in males and LC50 lies between 0.034 and 0.051mg/L in females).

Topical application of abamectin resulted in the rabbit in a 24hr LD_{50} value >2000mg/kg and in the rat in a 24hr LD_{50} value >330mg/kg (highest dose tested).

Characteristic signs of abamectin toxicity, tremors and ataxia, occur in rats after a single low oral dose and in rats and rabbits after a single very high dermal dose. Abamectin technical is non-irritant to skin and eyes and is not a skin sensitizer.

Based on the acute oral LD50 value and acute inhalation LC50 observed in the rat, abamectin technical needs to be classified as (R28) "very toxic if swallowed" and (R26) "very toxic by inhalation", according

to the criteria mentioned in Annex VI of Directive 2004/73/EC.

The polar photodegradate of abamectin exhibits a very low order of acute oral toxicity to CF-1 mice since deaths do not occur at dose levels up to 5000 mg/kg. However, since the polar metabolite was not identified, and purity and stability were not determined, the results of this study are less valuable.
The acute oral toxicity of the 8,9-Z isomer of avermectin B1a in CF-1 mice has been determined as > 80 mg/kg in both sexes, but has also been reported as between 10 and 20 mg/kg in male CF-1 mice. Death and clinical signs of intoxication, ataxia and bradypnea, occur at oral dose levels as low as 5 mg/kg bw of 8,9-Z isomer in CF-1 mice. In contrast, no deaths occur in CD-1 female mice at acute oral dose levels of 162 mg/kg bw of 8,9-Z isomer, but death occurs in 100% of animals at 290 mg/kg bw giving a calculated acute oral LD50 value of 8,9-Z isomer in CD-1 strain female mice of 217 mg/kg bw. Since the CF-1 mouse exhibits typical clinical signs of neurotoxicity, it is probable that the increased susceptibility of CF-1 mice is related to the accessibility of the 8,9-Z isomer to the target organ, and hence to the presence or absence of P-glycoprotein expression.

In two studies with pregnant and non-pregnant CF-1 mice, singly orally exposed to abamectin technical at day 10, 11 or 12 of gestation, it was shown that the LD50's in pregnant animals were slightly, not statistically significantly lower (LD50 = 19 mg/kg bw and LD50 = 11.8 mg/kg bw in study 1 and 2, respectively) compared to the LD50's in non-pregnant mice (LD50 = between 20 and 40 mg/kg bw and LD50 = 15 mg/kg bw in study 1 and 2, respectively). Typical clinical signs of neurotoxicity (tremors, clonic convulsion and bradypnea) occured in both pregnant and non-pregnant animals.

In a study with female CF-1 strain mice, heterozygous (+/-) or homozygous positive (+/+) for the mdr1 gene (which codes for P-glycoprotein expression), the LD50 for abamectin in homozygous positive (+/+) female mice was 28 mg/kg bw, whereas the LD50 in heterozygous female mice was 14 mg/kg bw.

In an antidote study with dogs it was demonstrated that 30 ml ipecac administered within 15 minutes of abamectin ingestion (8 mg/kg bw) prevented coma and death and reduced the incidence and/or severity of mydriasis, ataxia, tremors and convulsion. However, 3g charcoal or ipecac administered more than 15 minutes after ingestion, were ineffective in reducing abamectin-induced toxicity.

Evaluation by Competent Authorities
Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
EVALUATION BY RAPPORTEUR MEMBER STATE

Date	*5 November 2007; updated January 2009*
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	[redacted]
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion. Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*

Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

2. TOXICOKINETICS :

98/8 Doc IIIA 6.2/ 01 section No.	Metabolism studies in mammals. Basic toxicokinetics, including a dermal absorption study	**Official use only**
91/414 Annex II **Point addressed 5.1 / 01**		

Title:	Distribution and clearance of avermectin B1a in rats	
Lab Report Number:	ARM-1	
Authors:	[redacted] (1984)	
Test Substance:	avermectin B1a	
Species:	Rats	
Guidelines:	No guideline at time of conduct Deviations from Council Directive 87/302/EEC: Three animals/group Animals not observed for clinical signs of toxicity Biliary cannulation not performed, therefore degree of absorption cannot be calculated	
Date of Report:	1 February 1984.	
Published:	No	
GLP:	No	

STUDY 1

Characteristics

Reference	:	[redacted] (1984).	Exposure	:	Single low and high dose and repeated low dose
Type of study	:	Distribution and clearance of avermectin B_{1a}	Doses	:	0.138 (f), 0.142 (m), 1.38 (f), 1.42 (m) mg/kg bw
Year of execution	:	1984	Vehicle	:	Sesame oil
Test substance	:	avermectin B_{1a}-5-^{3}H, avermectin B1a-3,7,11,13,23-^{14}C and unlabelled avermectin B_{1a}	GLP statement	:	no
Route	:	Oral (gavage)	Guideline	:	-
Species	:	Rat (CRCD)	Acceptability	:	acceptable
Group size	:	3/sex/dose/time point			

Study design

Rats were treated orally, by gavage, with avermectin B1a-5-^{3}H, avermectin B1a-3,7,11,13,23-^{14}C or unlabelled avermectin B1a dissolved in sesame oil, according to the tabulated schedule shown below. A further 9 rats of each sex, treated with sesame oil alone, acted as controls. Sub-groups of 3 males and 3 females within each group were caged together, by sex, for the collection of urine and faeces. Urine and faeces were collected at 24-hour intervals until sacrifice (see also study 5).

Treatment and sacrifice schedule

No. animals (M + F)	Test article	Dose level x no. doses (p.o.)	Sacrifice schedule
12 + 12	Avermectin B1a-^{3}H	1.42mg/kg (M) x 1 1.38mg/kg (F) x 1	3M + 3F at 1, 2, 4 and 7 days after dose
12 + 12	Avermectin B1a-^{3}H	0.142mg/kg (M) x 1 0.138mg/kg (F) x 1	3M + 3F at 1, 2, 4 and 7 days after dose
17 + 17	Avermectin B1a Avermectin B1a-^{3}H	0.142mg/kg (M) x 15[a] 0.138mg/kg (F) x 15[a]	3M + 3F at 1, 2, 4 and 7 days after last dose
12 + 12	Avermectin B1a-^{3}H Avermectin B1a-^{14}C	1.42mg/kg (M) x 1 1.38mg/kg (F) x 1	3M + 3F at 1, 2, 4 and 7 days after dose
3 + 3	Sesame oil	0.55ml/rat (M) x 1 0.40ml/rat (F) x 1	3 M + 3F 7 days after dose
6 + 6	Sesame oil	0.55ml/rat/day (M) x 15 0.40ml/rat/day (F) x 15	3M + 3F 7 days after last dose

a - 14 daily doses of unlabelled material followed by a single dose of avermectin B1a-3H

At necropsy, bone, brain, fat, gonads, heart, kidney, liver, gastrointestinal tract, lung, blood, muscle, spleen and residual carcass were collected. The same tissues from the animals within each sub-group were combined and the total weights were determined. Samples of blood, urine and tissue were combusted, the $^{3}H_2O$ and $^{14}CO_2$ trapped, and total radioactivity levels were determined by scintillation spectrometer.

Results

The residue levels and depletion rates in the groups treated with tritiated avermectin B1a were comparable to the values obtained from rats treated with ^{14}C-labelled avermectin B1a.

The data reported for dose accountability were calculated on the basis of the total dose given to each group and are, therefore, average values from animals sacrificed 1, 2, 4 and 7 days after dosing. The total percent of dose accounted for in the analyzed samples for each group ranged from 85.1% to 95.1% for tritium and 94.5% to 94.9% for ^{14}C. Similarly, the excretion data reported were calculated on the basis of the total dose given to each group, whereas the daily elimination of the dose should have been calculated for each subgroup. The excretion data in % of dose for each subgroup sacrificed at intervals up to 7 days after dosing were recalculated by the notifier, based on the reported values of the daily elimination of dose in µg.

The total rate of excretion was similar for both sexes, and was independent of the dose level and treatment regime. Excretion was moderately fast with 80 to 101% of the dose excreted within 96 hours. The route of excretion was independent of the dose level. The dose was almost completely excreted via the faeces (86.8 to 104% of dose). Urinary excretion was low, accounting for 0.9 to 1.4% of dose in males and 0.5 to 0.8% of the dose in females.

Residue levels were greatest in all tissues 24 hours after treatment and were dose-dependent, as the residues in high dose groups were approximately ten-fold greater than those in low dose. The depletion rates were (relatively) independent of dose level, since the tissue half-lives were comparable between the dose levels. There were no substantive differences in the depletion half-lives among the tissues. Most of the calculated half-lives were within the range 1.2 ± 0.3 days, although brain and testes showed greater variability. The highest residue levels at 24 hours in tissues, excluding the GI tract, occurred in fat (2.89 - 5.30 and 0.16 - 0.32 ppm equivalents for high and low dose levels, respectively). The brain contained the lowest residues (0.07 - 0.13 and 0.01 ppm equivalents for high and low dose levels, respectively). In most instances, the kidney residue levels were higher than the levels in the liver. Pre-treatment for 14 days with unlabelled avermectin B1a at a dose level of 0.138 / 0.142mg/kg had no effect on either the residue levels or depletion rates compared to a single administration at the same dose level. The tissue residue levels of female rats were generally higher than the corresponding residue levels in male rats.

Acceptability

The purity of the unlabelled compound is not reported. Only 3 animals /sex/dose/time point were studied. Biliary canulation was not performed and expired air was not included in the study. These exclusions will have no major consequences for the interpretation of the data.
The study is considered acceptable for the overall toxicological evaluation.

Conclusion
Avermectin B1a is absorbed from the gastrointestinal tract and is distributed throughout all tissues and organs sampled, with the highest residues found in fat. It is rapidly eliminated from the body, almost exclusively in the faeces and there is no evidence for serious tissue accumulation on repeated administration. With the exception of dose-dependence for tissue residue levels and excretion by urine, the toxicokinetic profile is not influenced by sex, dose level and treatment regime.

98/8 Doc IIIA section No.	**6.2/ 02** Metabolism studies in mammals. Basic toxicokinetics, including a dermal absorption study	**Official use only**
91/414 Annex Point addressed	**II** **6.1 / 02** Distribution and clearance	

Title:	Absorption, Distribution, Depletion, and Excretion of [23-^{14}C] NOA 422601 in the Rat	
Lab Report Number:	048AM01	
Authors:	[redacted]	
Test Substance:	23-^{14}C- labelled avermectin B_{1a}	
Species:	Rat	
Guidelines:	Council Directive 94/79/EC, No. L 354/18, 51, 1994; OECD Guideline No. 417, 1984; OPPTS 870.7485, 1998; J-MAFF, Nohsan No 8147, 2000; Deviations: None	
Date of Report:	16 August 2001	
Published:	No	
GLP:	Yes	

STUDY 2

Characteristics

Reference	:	[redacted] (2001)	Exposure	:	Single low and high dose by stomach tube or single low dose i.v.
Type of study	:	Absorption, distribution, depletion and excretion of avermectin B1a	Doses	:	0.5 or 5.0 mg/kg bw
Year of execution	:	2000/2001	Vehicle	:	Polyethylene glycol 200/ethanol (3:2 v/v; 0.2 mg/ml (low dose) and 2.0 mg/ml (high dose))
Test substance	:	[23-^{14}C] avermectin B1a (purity [redacted]	GLP statement	:	yes
Route	:	Oral (stomach tube) or i.v.	Guideline	:	OECD 417
Species	:	Rat (Hanlbm: WIST and Ico:	Acceptability	:	acceptable

Group size	:	WI(IOPSAF/Han) 2, 3 or 6/dose/time point or 4/sex/dose/time point

Study design

Single oral doses of [23-^{14}C] labelled avermectin B_{1a} were administered at a low dose level (0.5 mg/kg bw) or at a high dose level (5.0 mg/kg bw) to several groups of rats. Additionally, [23-^{14}C] labelled avermectin B_{1a} was administered intravenously (low dose) to male rats. An overview of the experiments is presented in table below.

Summary of dosing and sampling regime for each group treated with 23-14C-Avermectin B1a

Group B1	4 m 4 f	0.5 mg.kg bw p.o.	Collection of urine (at 6,12, 24, 48, 72, 96, 120, 144, 168h) and faeces (at 24, 48, 72, 96, 120, 144, 168 h), blood (at 0.5, 1, 2, 4, 8, 12, 24, 48 h), and various tissues 7 days after administration
Group D1	4 m 4 f	5.0 mg/kg bw p.o	Collection of urine (at 6,12, 24, 48, 72, 96, 120, 144, 168h), faeces (at 24, 48, 72, 96, 120, 144, 168 h), and expired air (24, 48 h), blood (at 0.5, 1, 2, 4, 8, 12, 24, 48, 72 h), and various tissues 7 days after administration
Group F1	12 m	0.5 mg.kg bw p.o.	Collection of various tissues at 6, 24, 48, and 72 hours after administration
Group F2	12 f	0.5 mg.kg bw p.o.	Collection of various tissues at 6, 24, 48, and 72 hours after administration
Group F3	12 m	5.0 mg/kg bw p.o	Collection of various tissues at 8, 24, 48, and 72 hours after administration
Group F4	12 f	5.0 mg/kg bw p.o	Collection of various tissues at 8, 24, 48, and 72 hours after administration
Group G1	6 m	0.5 mg.kg bw p.o.	Collection of urine (at 24, 48 h), bile (at 1, 2, 4, 8, 24, 32, 48 h), faeces (at 24, 48 h) after administration and gastrointestinal tract and remaining carcass after sacrifice
Group G2	6 f	0.5 mg.kg bw p.o.	Collection of urine (at 24, 48 h), bile (at 1, 2, 4, 8, 24, 32, 48 h), faeces (at 24, 48 h) after administration and gastrointestinal tract and remaining carcass after sacrifice
Group L1	4 m	0.5 mg.kg bw i.v.	Collection of urine and faeces, whole body radioluminography and radioluminography of the GI-tract at 6 and 24 hours after administration

Results

The animals of the high dose group showed toxic symptoms few hours after administration, i.e. piloerection, ataxia, convulsions, tremor, crouch position, yawn and stretched tail. After 24h animals had recovered, and only piloerection was observed. From 48 h to 168 h, no symptoms were observed.

After single oral administration, the maximum concentration in the blood at both dose levels and in both sexes was reached within 4 to 8 hours after administration. The half lives for elimination of radioactivity from blood were 15 and 18 hours in males and were 16 and 27 hours in females in the low and high dose, respectively. The area under the curve (AUC) values were similar between the sexes: 1.2 µg.h/g and 1.3 µg.h/g (low dose level) and 17 µg.h/g and 18 µg.h/g (high dose level) in males and females respectively.
The apparent extent of absorption, based on the amount renally and biliary excreted, was determined to be 3 to 5% of the dose. However, the real extent of absorption is significantly higher based on tissue residue data (50% of the dose determined in tissues at 6 and 8 h after administration) and comparison of urinary excretion following oral or i.v. administration. The apparent, calculated oral bioavailability of 0.86, as given by the study author, is based on comparison of urinary excretion within 24h after oral or i.v. administration of a low dose avermectin B_{1a} in males.
The major part of the orally administered doses were excreted via the faeces, i.e. more than 92% of the dose at both dose levels and both sexes within 7 days. Renal excretion accounted for 1.2 and 1.6% in males and for 0.5 and 1.0% in females of the low and high dose groups, respectively.
At the low dose level 5% (males) and 3% (females) of the absorbed dose were eliminated via bile and ultimately excreted with the faeces. The rate of excretion was slower in females as compared to males, with males excreting 81% and 73% of the dose within 48 h after administration and females excreting within the same time period 69% and 48% of the low and high dose, respectively.
The excretion of the absorbed test substance (high dose) via expired air accounted for only 0.01% of the dose within 48 h after administration.
After i.v. administration of the low dose in males, excretion of the absorbed dose by urine accounted for 1.0% and 34% was excreted with the faeces between 0 and 48 h.
The non-biliary excretion into the GI-tract 6 and 24 h after i.v. administration of the low dose in males was determined by radioluminography. Six hours after i.v administration, a significant concentration of radioactivity was observed in the secretory epithelium of the stomach and the intestinal wall along the GI-tract. The content of the GI-tract revealed increasing concentrations of radioactivity from the duodenum to the ileum. Twenty four hours after i.v administration the concentration in the intestinal wall had decreased, whereas the content of the GI-tract showed increasing concentrations from duodenum to the rectum.
Tissues and organs achieved the highest residue level 6 h after single oral administration at the low dose level, except in fat where the residues reached the mamimum level 24 h after single oral administration. The highest residues were found in adrenals, fat, liver and pancreas. The maximum residue levels for males and females were similar. Tissue half lives were in the range of 12 to 17 h for males and 13 to 33 hours for females. Within 7 days after administration tissue residues were low. Based on the slowest depletion rate the highest residues were determined in fat.

After administration of the high dose the residue levels were about 10 times compared to administration of the low dose. The profile of the maximum levels was comparable to that of the low level profile. The calculated tissue half lives ranged from 12 to 20 h in males and 20 to 35 h in females. The residues declined within 7 days after administration to low levels, except for fat (more than 10 times higher levels compared to other tissue residue levels).
The absorbed test substance and/or its metabolites were widely distributed within the body, and approximately 50% of the administered dose was recovered in tissues and organs 6 and 8 h after adminstration at the low and high dose respectively. The residue levels depleted to approximately 1% and 2% of the dose within 168h in males and females, respectively.
Comparison of tissue residues after i.v. administration with those in rats which were given a similar oral dose shows that the residues were similar at the corresponding time points, which indicates almost complete absorption after oral administration.

Acceptability

The study author calculated an apparent oral bioavailability of 0.86[1], based on urinary excretion within 24h after oral or i.v. administration of a low dose avermectin B_{1a} in males. Although the results of this study gives only limited information on absorption, data of other toxicokinetic studies support the asssumption and calculation of almost complete absorption of avermectin B1a after administration.
In 6 animals, there had been no infusion of artificial bile fluid for a duration of about 12 h based on an malfunction of the infusion pump 48 h after administration of the test substance. These exclusions will have no major consequences for the interpretation of the data.
The study is considered acceptable for the overall toxicological evaluation.

Conclusion

After oral administration to rats, the majority of avermectin B_{1a} was almost completely absorbed and then eliminated predominantly by non-biliary excretion into the GI-tract before excretion with the faeces. The rates and mechanisms of oral absorption and subsequent excretion were independent of the dose level and the sex, however with the high dose the depletion of tissue residues in males was more rapid than in females by approximately 2-fold.

[1] The value was obtained by comparison of the urinary excretion within 24h after oral versus intravenous administration. This results in a ratio which is equivalent to the bioavailability of abamectin. After i.v administration 0.73% of the dose is excreted via urine, orally it is 0.67%. Corrected for the administrated dose (0.67% x 0.46 mg/kg / 0.73% x 0.49 mg/kg) the oral absorption of 86% is obtained (copied from the

original study report, Hassler 2001).

98/8 Doc IIIA 6.2/ 03 section No.	**Metabolism studies in mammals. Basic toxicokinetics, including a dermal absorption study**	**Official use only**
91/414 Annex II Point addressed 6.1 / 03	**Distribution and clearance of [23-^{14}C]- labelled avermectin B_{1b}**	

Title:	Absorption, Distribution, Degradation, and Excretion of [23-^{14}C] NOA 421704 in the Rat	
Lab Report Number:	054AM01	
Authors:	[redacted] (2003a)	
Test Substance:	[23-^{14}C]- labelled avermectin B_{1b}	
Species:	Rat	
Guidelines:	Council Directive 94/79/EC, No. L 354/18, 51, 1994; OECD Guideline No. 417, 1984; OPPTS 870.7485, 1998; J-MAFF, Nohsan No 8147, 2000. Deviations: None	
Date of Report:	4 July 2003	
Published:	No	
GLP:	Yes	

STUDY 3

Characteristics

Reference	:	[redacted] (2003a)	Exposure	:	Single low and high dose by stomach tube
Type of study	:	Absorption, distribution, depletion and excretion of avermectin B1b	Doses	:	0.5 mg/kg bw or 5.0 mg/kg bw
Year of execution	:	2001	Vehicle	:	Polyethylene glycol 200/ethanol (4:2 v/v; 0.2 mg/ml (low dose) and 2.0 mg/ml (high dose))
Test substance	:	[23-^{14}C] avermectin B1b (purity [redacted])	GLP statement	:	yes
Route	:	Oral (gavage)	Guideline	:	OECD 417
Species	:	Rat Hanlbm: WIST (SPF)	Acceptability	:	acceptable
Group size	:	4/sex/dose			

Study design

Single oral doses of [23-^{14}C] labelled avermectin B1b were administered at a low dose level (0.5 mg/kg bw) or at a high dose level (5.0 mg/kg bw) to two groups of male and female rats. An overview of the experiments is presented in table below.

Summary of dosing and sampling regime for each group treated with 23-14C-Avermectin B1b

Group B1	4 m 4 f	Low dose p.o. 0.5 mg/kg bw	Collection of urine (at 6, 12, 24, 48, 72, 96, 120, 144 and 168 h) and faeces (24, 48, 72, 96, 120, 144 and 168 h), blood kinetics (0.5, 1, 2, 4, 8, 12, 24 and 48 h) and various tissues 7 days after administration
Group D1	4 m 4 f	High dose p.o. 5.0 mg/kg bw	Collection of urine (at 6, 12, 24, 48, 72, 96, 120, 144 and 168 h) and faeces (24, 48, 72, 96, 120, 144 and 168 h), blood kinetics (0.5, 1, 2, 4, 8, 12, 24 and 48 h) and various tissues 7 days after administration

Results

After single oral administration, the maximum concentration in the blood at both dose levels and in both sexes was reached within 4 to 8 hours after administration. The half lives for elimination of radioactivity from blood were 9 and 14 hours in males and were 13 and 21 hours in females in the low and high dose, respectively. The area under the curve (AUC) values were similar between the sexes: 0.72 µg.h/g and 0.86 µg.h/g (low dose level) and 9.0 µg.h/g and 11.5 µg.h/g (high dose level) in males and females respectively. Only 4 to 5% of the administered dose was determined in the urine over the experimental period of 168 h. The apparent absorption, based on the radioactivity determined in urine and tissue residues accounted for only 6 to 7% of the dose in both low and high dose animals. Comparison with a study with avermectin B1a ([redacted], study 2) indicates that the actual extent of absorption is significantly higher. Excretion by urine was lower in females compared to males.

The majority of the dose was excreted via the faeces, accounting for 93% (low dose) and 89% (high dose) in males and 91% (low dose) and 92% (high dose) in females. Total excretion in female rats was slower compared to male rats, with 94% (low dose) and 89% (high dose) excreted within 48 h in males and 82%

(low dose) and 70% (high dose) of the dose excreted by female rats within the same period.
Tissue residue levels were determined 7 days after administration. Both in the low and the high dose animals, the test substance was widely distributed in the whole body, with the highest residues found in fat. At both dose levels, females showed higher residue levels in adrenals and pancreas compared to males.
TLC radiograms of urine revealed a pattern of 11 metabolite fractions. Unchanged avermectin B1b was not detected in urine. Metabolite fraction B13 accounted for approximately 2.5% of the dose, whereas the other metabolite fractions each accounted for less than 0.5% of the dose. Metabolite fractions B1 and B7 were excreted more in urine by males compared to females. The metabolic fractions were not identified.
TLC radiograms of faeces revealed a pattern of 19 metabolite fractions. Unchanged avermectin B1b accounted for 9 to 17%. Metabolite fractions B7 and B13 each accounted for 13 to 32 % of the dose, metabolite fraction B10 accounted for 7 to 14 % of the dose, whereas the other metabolite fractions each accounted for less than 5% of the dose. Metabolite fractions B1, B6 and B7 were excreted more in faeces by males compared to females, whereas metabolite fractions B3 was excreted more by females compared to males.The metabolic fractions were not identified.
(note: in a study from [redacted] with avermectin B1a, metabolic fractions were identified, and the notifier suggests that the fractions B7, B10 and B13 probably correspond to the identified fractions A7, A10 and A13, see study 6).

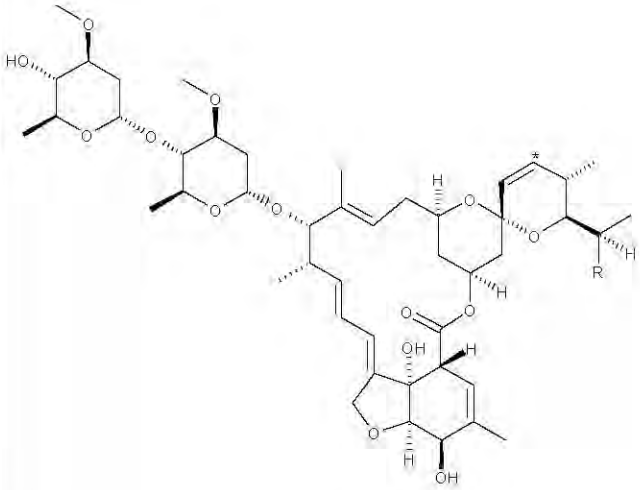
Acceptability

The study is considered acceptable for the overall toxicological evaluation.

Conclusion

After oral administration to rats, the majority of avermectin B1b was almost completely absorbed and then eliminated predominantly by non-biliary excretion into the GI-tract before excretion with the faeces. The rates and mechanisms of oral absorption and subsequent excretion were independent of the dose level and the sex, with the exception of a slower rate of excretion in females. The absorption, distribution, degradation and excretion of [23-^{14}C] avermectin B1b and [23-^{14}C] avermectin B1a are essentially similar.

98/8 Doc IIIA 6.2/ 04 section No.	**Metabolism studies in mammals. Basic toxicokinetics, including a dermal absorption study**	**Official use only**
91/414 Annex II Point addressed 5.1 / 05		

Title:	Disposition of [23-^{14}C] NOA 422601 in the Rat After Multiple Oral Administrations.	
Lab Report Number:	048AM03	
Authors:	[redacted] (2003).	
Test Substance:	[23-^{14}C] avermectin B_{1a}  * = [^{14}C] **R = –CH_2CH_3** avermectin B_{1a} (NOA 422601) Batch No: [redacted] Purity: [redacted] Specific activity.: 2480 kBq/mg (67.0 µCi/mg)	
Species:	Rat	
Guidelines:	Council Directive 94/79/EC, No. L 354/18, 51, 1994; OECD Guideline No. 417, 1984; J-MAFF, Nohsan No	

	8147, 2000; Deviations: None	
Date of Report:	14 July 2003	
Published:	No	
GLP:	Yes	

STUDY 4

Characteristics

Reference	:	[redacted] (2003)	Exposure	:	Up to 14 days
Type of study	:	Distribution and excretion of avermectin B1a	Doses	:	0.5 mg/kg bw/day
Year of execution	:	2001	Vehicle	:	Polyethylene glycol 200/ethanol (3:2 v/v; 0.2 mg/ml (low dose)
Test substance	:	avermectin B1a (purity [redacted])	GLP statement	:	yes
Route	:	Oral (gavage)	Guideline	:	OECD 417
Species	:	Rat Hanlbm: WIST (SPF)	Acceptability	:	acceptable
Group size	:	4f/time point			

Study design

Sixteen female rats were orally dosed up to 14 days with 0.5 mg/kg bw/day [23-^{14}C] avermectin B1a. Subgroups of 4 females were sacrificed on day 1, day 7, day 14 and day 20 and avermectin B1a related residues were determined in tissues and organs. Radioactivity was determined at daily intervals in blood, urine and faeces of females sacrificed on day 20.

Results

The blood concentration of avermectin B1a reached a plateau 3 days after the start of dosing. One day after the last of the 14 consecutive daily doses the blood residues declined, reaching half the maximum concentration within 19 h.

Two days after the first administration, excretion of radioactivity reached a plateau. The majority (97%) of the test substance was excreted 7 days after the last of the14 daily doses. Less than 1% of the total dose remained in the tissues and organs at the end of the experimental period. The administered test substance was mainly excreted by faeces accounting for 96% of the dose, and only 0.8% was excreted by urine.

Tissue residue levels reached a plateau after 7 days of multiple dosing for most selected organs and tissues, except fat, ovaries and spleen which showed some increase after 14 days and thyroids, which showed an almost linear increase of the residue values during the dosing period.

The highest residue levels were found in fat, followed by adrenals, pancreas, liver and thyroids. The

calculated half life (T1/2) for the depletion of the residual radioactivity from tissues and organs were for the selected organs and tissue, except for fat, in the range of 26 to 39 h, whereas for fat it was 58 h.
The residues in tissues and organs 7 days after the last of the 14 daily doses revealed a similar distribution pattern and were only 2 to 4 times higher compared to the tissue residues 7 days after single oral administration, except for fat, which showed a 7 times higher residue level.
The metabolite pattern of urine and faeces, investigated at day 0-1, day 6-7 and day 13-14 during the dosing period, were not influenced by multiple dosing. Approximately 40% of the administered test substance was excreted with the faeces as unchanged parent compound.

Acceptability

The study is considered acceptable for the overall toxicological evaluation.

Conclusion

In summary, the rates and routes of excretion, and the tissue distribution did not change upon multiple dosing compared to single dosing. With the exception of the thyroids (and fat, ovaries and spleen), the tissue residues reached a plateau after 7 days of dosing and for all tissues the residues declined very rapidly after the dosing period.

98/8 Doc IIIA 6.2/ 05 section No.	Metabolism studies in mammals. Basic toxicokinetics, including a dermal absorption study	Official use only
91/414 Annex II Point addressed 5.1 / 06		

Title:	The metabolism of avermectin B1a in rats	
Lab Report Number:	not specified	
Authors:	[redacted] (1986):	
Test Substance:	Selected tissue samples derived from the study "Distribution and clearance of avermectin B1a in rats" (Alvaro, R. F. *et. al.*, 1984) were analysed for parent avermectin B_{1a}.	
Species:	Rat	
Guidelines:	In accordance with Council Directive 87/302/EEC	
Date of Report:	15 May 1986	
Published:	No	
GLP:	No	

STUDY 5

Characteristics

Reference	:	[redacted] (1986)	Exposure	:	Single low and high dose and repeated low dose
Type of study	:	Metabolism of avermectin B1a	Doses	:	0.138 (f), 0.142 (m), 1.38 (f), 1.42 (m) mg/kg bw
Year of execution	:	Not specified	Vehicle	:	Sesame oil
Test substance	:	avermectin B_{1a}-5-^{3}H, avermectin B1a-3,7,11,13,23-^{14}C and unlabelled avermectin B_{1a}	GLP statement	:	no
Route	:	Oral (gavage)	Guideline	:	-
Species	:	Rat (CRCD)	Acceptability	:	acceptable
Group size	:	3/sex/dose/time point			

Study design

Selcted tissue samples derived from the study "Distribution and clearance of avermectin B1a in rats (Alvaro, R.F. et al., 1984, see B.6.1.1 study 1 from abamectin DAR) were analysed for parent avermectin B1a. In that study, rats were treated orally, by gavage, with avermectin B1a-5-^{3}H, avermectin B1a-3,7,11,13,23-^{14}C or unlabelled avermectin B1a dissolved in sesame oil, according to the tabulated schedule shown below. A further 9 rats of each sex, treated with sesame oil alone, acted as controls. Liver, kidney, muscle and fat samples were analysed by an indirect method, reverse isotope dilution assay (RIDA). The residues were separated and purified by reverse phase-HPLC radioactiovity profiling and quantified by calculation. Radioactivity in liquid samples was measured by liquid scintillation counting (LSC). Analysis and separation of metabolites was performed by HPLC analysis of the ethyl acetate extract obtained from the RIDA extraction procedure. The metabolites were identified by comparison of retention times to standards and by co-chromatography with standards. Metabolite standards were generated by performing incubations of rat liver microsomal with ^{14}C- or ^{3}H-labelled avermectin B1a (in vivo metabolites as residue levels were too low for direct isolation). Metabolite structures were identified by NMR spectrometry and FAB-Mass spectrometry. The isolated metabolites were identified as 24-hydroxymethyl-avermectin B1a (24-OHMe-B1a) and 3''-O-desmethyl-avermectin B1a (3''-DM-B1a) and were used as reference substances.

Treatment and sacrifice schedule

No. animals (M + F)	Test article	Dose level x no. doses	Sacrifice schedule
12 + 12	Avermectin B1a-^{3}H	1.42mg/kg (M) x 1 1.38mg/kg (F) x 1	3M + 3F at 1, 2, 4 and 7 days after dose
12 + 12	Avermectin B1a-^{3}H	0.142mg/kg (M) x 1 0.138mg/kg (F) x 1	3M + 3F at 1, 2, 4 and 7 days after dose
17 + 17	Avermectin B1a Avermectin B1a-^{3}H	0.142mg/kg (M) x 15[a] 0.138mg/kg (F) x 15[a]	3M + 3F at 1, 2, 4 and 7 days after last dose
12 + 12	Avermectin B1a-^{3}H Avermectin B1a-^{14}C	1.42mg/kg (M) x 1 1.38mg/kg (F) x 1	3M + 3F at 1, 2, 4 and 7 days after dose
3 + 3	Sesame oil	0.55ml/rat (M) x 1 0.40ml/rat (F) x 1	3 M + 3F 7 days after dose
6 + 6	Sesame oil	0.55ml/rat/day (M) x 15 0.40ml/rat/day (F) x 15	3M + 3F 7 days after last dose

a - 14 daily doses of unlabelled material followed by a single dose of avermectin B1a-3H

Results

HPLC profiles of tissue extracts from rats dosed with both ^{3}H- and ^{14}C-labelled avermectin B1a were identical for both labels.

Parent avermectin B1a as a percentage of the total residue was not dependent on the dose level.

The half-lives of avermectin B1a in kidney, liver and fat in high dosed animals were lower in males (0.60 to 0.65 days) than in females (0.92 to 1.0 days), whereas muscle showed comparable half-lives of 0.97 and 0.95 days for males and females respectively. At least in the low dose animals, avermectin B1a as a percentage of the total residue was lower in male than in female tissue. At the high dose level, individual differences in the tissue residue levels were apparent between the experiments (e.g. 3030 and 1149 ppb total residue in male kidney, whereas total residue levels in liver, muscle and fat where comparable). Repeated administration of the low dose had no effect on tissue residue levels.

In addition to unchanged avermectin B1a, two metabolites were identified, 24- hydroxymethyl-avermectin B1a (24-OHMe-B1a) and 3''-O-desmethyl-avermectin B1a (3''-DM-B1a).

Quantitative results for the metabolites were reported only for some treatment and sampling regime. In liver of females single dosed with 1.4 mg/kg bw and sacrificed 2 days after treatment, 24-OHMe-B1a and 3''DM-B1a accounted for 6% and 24% of the total residue, respectively, and for 3% and 24%, respectively in the kidney. Unchanged avermectin B1a accounted for 46% in the liver and 55% in the kidney. The metabolic profile in muscle was similar to the profiles in liver and kidney, but no quantitative results were reported. The metabolic profile in fat was different to that in the other tissues.

Acceptability

The study is considered acceptable for the overall toxicological evaluation.

Conclusions

Unchanged parent avermectin B1a as a proportion of total residues, is not dose-dependent but is slightly lower in the male than in the female rat. The tissue half-lives of avermectin B1a are slightly lower in males than in females (except for muscle). Avermectin B1a and/or metabolites do not accumulate in liver, kidneys, muscle or fat on repeated administration of a low dose.

98/8 Doc IIIA 6.2/ 06 section No.	Metabolism studies in mammals. Basic toxicokinetics, including a dermal absorption study	Official use only
91/414 Annex II Point addressed 5.1 / 01		

Title:	The Metabolism of [23-^{14}C] NOA 422601 in the Rat	
Lab Report Number:	048AM02	
Authors:	[redacted] (2003b)	
Test Substance:	[23-^{14}C] avermectin B_{1a} Batch [redacted] Purity: [redacted] Spec. Act.: 2480 kBq/mg (67.0 µCi/mg) (used for Group B1, F1, F2) Batch [redacted] Purity: [redacted] Spec. Act.: 2430 kBq/mg (65.7 µCi/mg) (used for Group D1) Batch [redacted] Purity: [redacted] Spec. Act.: 2486 kBq/mg (67.2 µCi/mg) (used for Group G1, G2) Batch [redacted] Purity: [redacted] Spec. Act.: 2486 kBq/mg (67.2 µCi/mg) (for Group F3, F4)	
Species:	Rat	
Guidelines:	Council Directive 94/79/EC, No. L 354/18, 51, 1994; OECD Guideline No. 417, 1984; OPPTS 870.7485, 1998; J-MAFF, Nohsan No 8147, 2000; Deviations: None	
Date of Report:	1 July 2003	

Published:	No	
GLP:	Yes	

STUDY 6

Characteristics

Reference	:	[redacted] (2003b)	Exposure	:	Single low and high dose by stomach tube
Type of study	:	Metabolism of [23-^{14}C]avermectin B1a in rat.	Doses	:	0.5 or 5.0 mg/kg bw
Year of execution	:	2000-2002	Vehicle	:	Polyethylene glycol 200/ethanol (3:2 v/v; 0.2 mg/ml (low dose) and 2.0 mg/ml (high dose))
Test substance	:	[23-^{14}C]avermectin B1a (purity [redacted]	GLP statement	:	yes
Route	:	Oral (stomach tube)	Guideline	:	OECD 417
Species	:	Rat (Hanlbm: WIST and Ico: WI(IOPSAF/Han)	Acceptability	:	acceptable
Group size	:	2, 3 or 6/dose/time point or 4/sex/dose/time point			

Based on the structures identified, the metabolism of avermectin B_{1a} in the rat proceeds predominantly via demethylation, hydroxylation, cleavage of the oleandrosyl ring and oxidation reactions. The reaction scheme for the metabolism of avermectin B_{1a} in the rat is proposed in the figure below. The metabolites identified are the results of the following reaction steps:

- demethylation at the oleandrosyl ring as in [3"DM]
- hydroxylation at different positions as in [24aOH], [27OH], [28OH] and 8aOH (NOA 448112) combined with demethylation as in [3"DM,24aOH], [3"DM,27OH] and [3"DM,4aOH]
- cleavage of the oleandrosyl ring as in [DO,3"DM,4aOH]
- oxidation of the 8a position as in 8aOxo (NOA 448111) and 4OH,8aOxo (NOA 457465)

Study design

Urine, faeces, bile, fat and muscle specimens from male and female animals derived from the study "Distribution and clearance of avermectin B1a in rats" [redacted] 2001, see B.6.1.1 study 2 from abamectin DAR) were analysed to identify and quantify individual metabolites to derive the metabolic pathway of orally administered [23-^{14}C]avermectin B1a at dose levels of 0.5 mg/kg bw or 5.0 mg/kg bw.

Summary of dosing and sampling regime for each group treated with 23-^{14}C-Avermectin B_{1a}

Group B1	4 m 4 f	0.5 mg/kg bw p.o.	Collection of urine (at 6,12, 24, 48, 72, 96, 120, 144, 168h) and faeces (at 24, 48, 72, 96, 120, 144, 168 h), blood (at 0.5, 1, 2, 4, 8, 12, 24, 48

			h), and various tissues 7 days after administration
Group D1	4 m 4 f	5.0 mg/kg bw p.o	Collection of urine (at 6,12, 24, 48, 72, 96, 120, 144, 168h), faeces (at 24, 48, 72, 96, 120, 144, 168 h), and expired air (24, 48 h), blood (at 0.5, 1, 2, 4, 8, 12, 24, 48, 72 h), and various tissues 7 days after administration
Group F1	12 m	0.5 mg/kg bw p.o.	Collection of various tissues at different time points, i.e. 6, 24, 48, and 72 hours after administration
Group F2	12 f	0.5 mg/kg bw p.o.	Collection of various tissues at different time points, i.e. 6, 24, 48, and 72 hours after administration
Group F3	12 m	5.0 mg/kg bw p.o	Collection of various tissues at different time points, i.e. 8, 24, 48, and 72 hours after administration
Group F4	12 f	5.0 mg/kg bw p.o	Collection of various tissues at different time points, i.e. 8, 24, 48, and 72 hours after administration
Group G1	6 m	0.5 mg/kg bw p.o.	Collection of urine (at 24, 48 h), bile (at 1, 2, 4, 8, 24, 32, 48 h), faeces (at 24, 48 h) after administration and gastrointestinal tract and remaining carcass after sacrifice
Group G2	6 f	0.5 mg/kg bw p.o.	Collection of urine (at 24, 48 h), bile (at 1, 2, 4, 8, 24, 32, 48 h), faeces (at 24, 48 h) after administration and gastrointestinal tract and remaining carcass after sacrifice
Group L1	4 m	0.5 mg/kg bw i.v.	Collection of urine and faeces, whole body radioluminography and radioluminography of the GI-tract at two different time points, i.e. 6 and 24 hours after administration

Urine, faeces and bile pools according to sex and animal group were prepared by mixing equal aliquots of the collected samples from each animal (0-168 h or 0-48 h for bile canulated animals). Fat and muscle pools were prepared by mixing the whole (remaining) samples from animals of subgroups at 8, 24 and 48 h. Radioactivity in all samples was measured by Liquid Scintillation Counting (LSC) and the quantitative metabolite pattern was determined by thin layer chromatography (TLC). Metabolites were isolated by solid phase extraction (SPE), HPLC and TLC and were identified by LC-MS, LC-NMR and/or cochromatochraphy with authentic reference substances.

Results

The metabolite pattern in urine, faeces and bile was complex (20 fractions), qualitatively independent of the

sex and the dose level and with some quantitative variations.

The following 11 metabolites were isolated:

Code	Fraction	Abbreviation	Metabolite Name	% of dose	
				faeces	urine
	A4	[3"DM,4aOH]	3''-O-Desmethyl, 4a-hydroxy-avermectin B_{1a}	0.5-1	
	A5	[DO,3'DM,4aOH]	Desoleandrosyl, 3'-O-desmethyl, 4a-hydroxy-avermectin B_{1a}	0.8-1.6	0-0.01
	A6	[3"DM,27OH]	3''-O-Desmethyl, 27-hydroxy-avermectin B_{1a}	2.2-6.8	0.02-0.17
	A7	[3"DM,24aOH]	3''-O-Desmethyl, 24a-hydroxy-avermectin B_{1a}	4.2-13.4	0.06-0.37
	A10	[3"DM]	3''-O-Desmethyl-avermectin B_{1a}	19.3-27.0	
	A12	[27OH]	27-Hydroxy-avermectin B_{1a}	4.6-7.6	0.17-0.41
	A13	[24aOH]	24a-Hydroxy-avermectin B_{1a}	2.8-5.7	0.09-0.15
	A14	[28OH]	28-Hydroxy-avermectin B_{1a}	0.5-1.7	0.05-0.12
NOA 448111		[8aOxo]	8a-Oxo-avermectin B_{1a}		
NOA 448112		[8aOH]	8a-Hydroxy-avermectin B_{1a}		
NOA 457465		[4OH,8aOxo]	4-Hydroxy, 8a-oxo-avermectin B_{1a}		

In faeces, avermectin B1a accounted for 25 to 45% of the dose and the individual metabolites [3"DM], [24aOH], [27aOH], [3"DM,24aOH] and [3"DM,27OH] accounted each for 3-27% of the dose. The minor metabolites [28OH], [3"DM,4aOH] and [DO,3'DM,4aOH] each accounted for 0.4-2% of the dose. The soil metabolites [8aOXO], [8aOH] and [4OH,8aOxo] were additionally detected in faeces, accounting for 0.3, 0.4 and 0.08% of the dose, respectively. Metabolite fractions A7, A12 , A13 and A14 were excreted more in faeces by males compared to females.

The major faecal components, avermectin B1a and metabolite [3"DM] were not present in urine. The metabolites [24aOH], [27aOH], [3"DM,24aOH] and [3"DM,27OH] represented the majority of the urinary

radioactivity, each accounting for 0.02-0.4% of the dose. Metabolite fractions A6, A7 and A12 were excreted more in urine by males compared to females.
In bile, the major components detected were [3"DM,24aOH] and [3"DM,27OH], [3"DM] and avermectin B1a each accounting for 0.1-1.2% of the dose. Metabolite fractions A6, A7 and A12 were excreted more by bile by males compared to females.

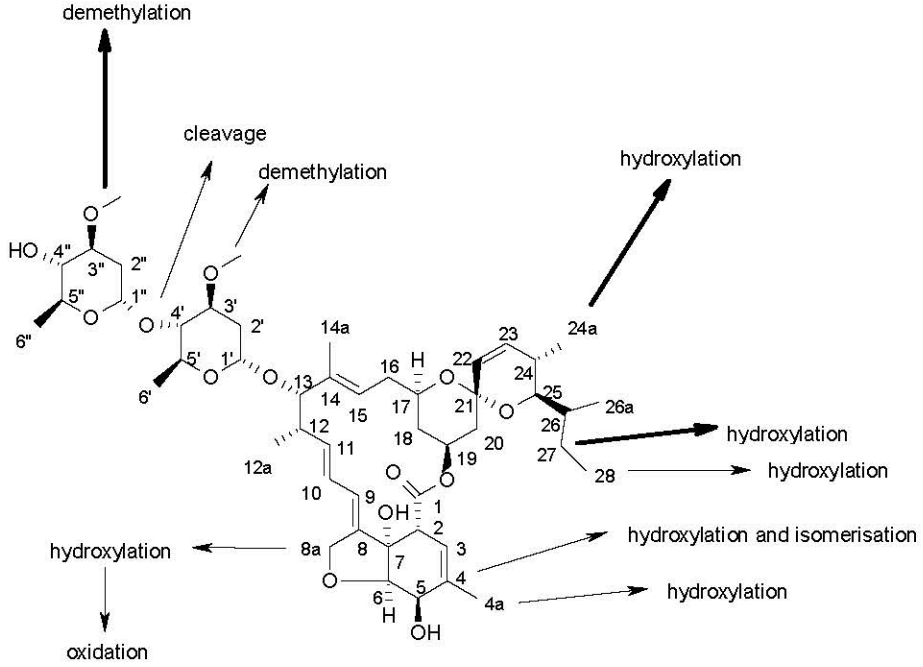
In the fat and muscle, avermectin B1a was the major component detected accounting for 92% and 72% of the radioactivity, respectively. Metabolite [3''DM] accounted for 1.7 and 19% in the fat and muscle, respectively.

Based on the structures identified, the metabolism of avermectin B_{1a} in the rat proceeds predominantly via demethylation, hydroxylation, cleavage of the oleandrosyl ring and oxidation reactions. The reaction scheme for the metabolism of avermectin B_{1a} in the rat is proposed in the figure below. The metabolites identified are the results of the following reaction steps:

- demethylation at the oleandrosyl ring as in [3"DM]
- hydroxylation at different positions as in [24aOH], [27OH], [28OH] and 8aOH (NOA 448112) combined with demethylation as in [3"DM,24aOH], [3"DM,27OH] and [3"DM,4aOH]
- cleavage of the oleandrosyl ring as in [DO,3"DM,4aOH]
- oxidation of the 8a position as in 8aOxo (NOA 448111) and 4OH,8aOxo (NOA 457465)

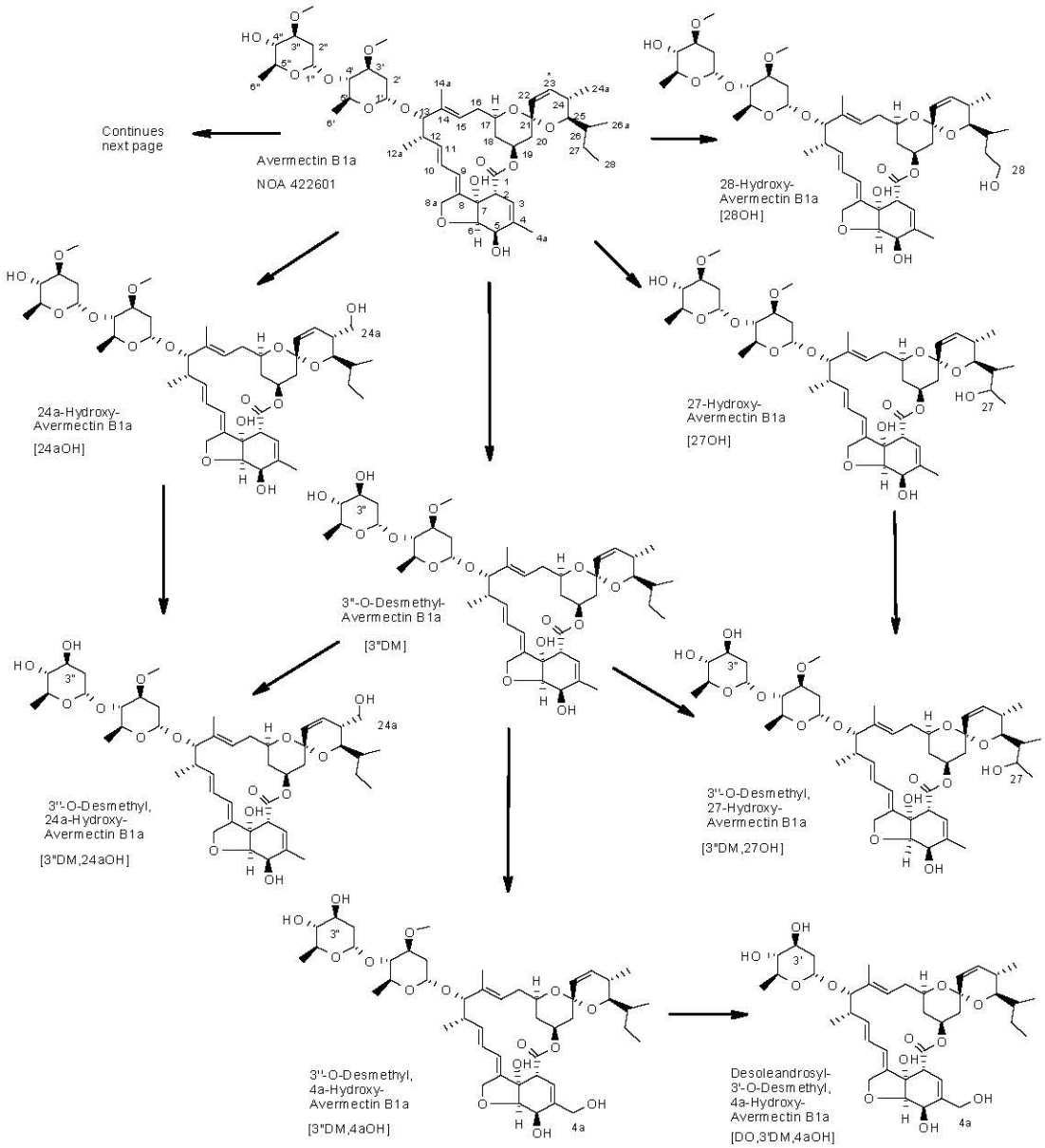
The metabolic pathways of avermectin B_{1a} in the rat are proposed in Figure 1.

Proposed reaction scheme for the metabolism of avermectin B_{1a} in rats

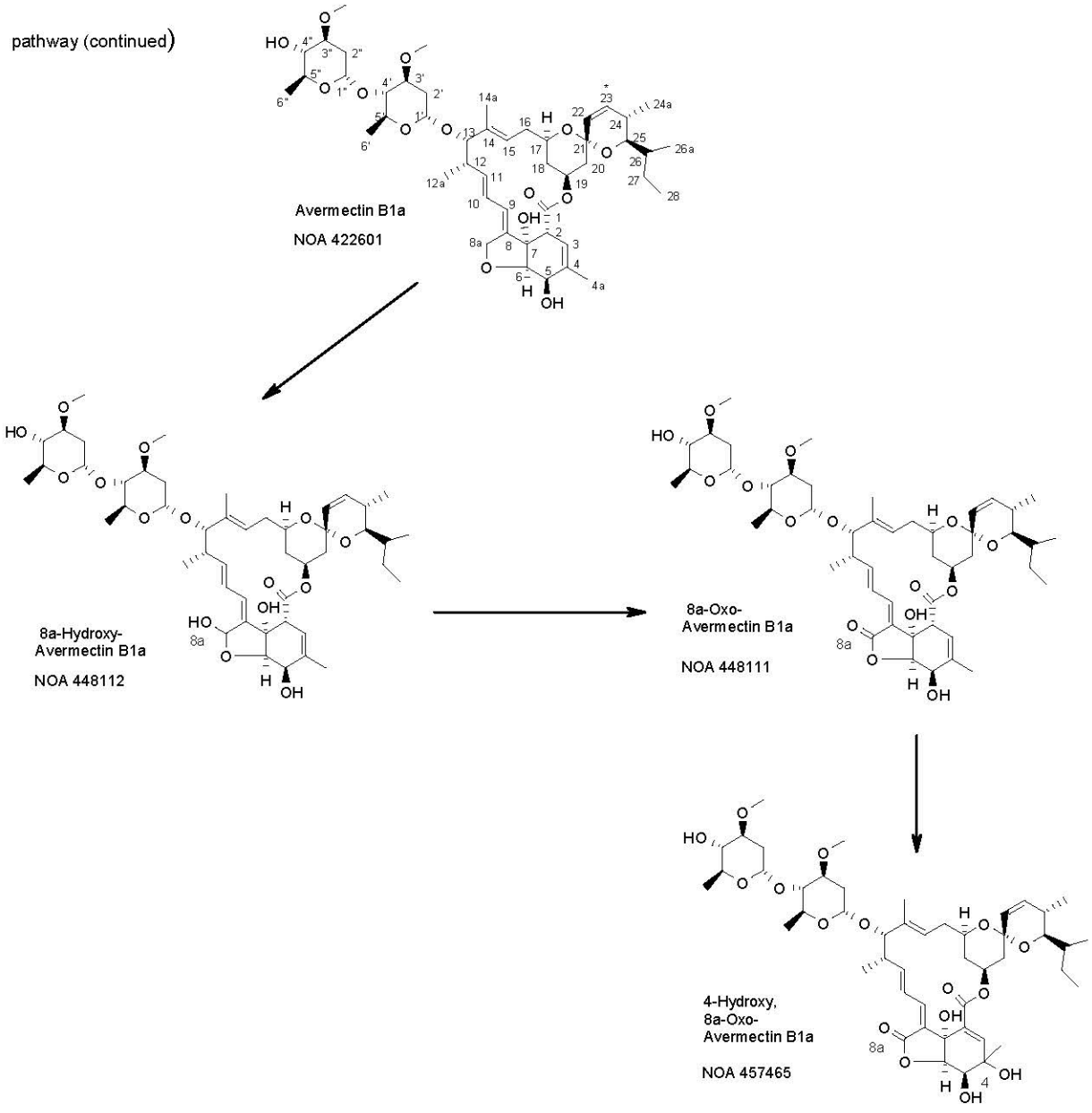


avermectin B_{1a}

Figure 1. Proposed pathways for the metabolism of avermectin B1a in rats



pathway (continued)



Acceptability

The study is considered acceptable for the overall toxicological evaluation.

98/8 Doc IIIA 6.2/ 07 section No.	**Percutaneous absorption (in vivo test)**	**Official use only**
91/414 Annex Point addressed	II 6.1 / 04	

Title:	Dermal penetration of avermectin B1a in the monkey (PS#1)	
Lab Report Number:	Report number not specified	
Authors:	[redacted] 1986	
Test Substance:	^{3}H-avermectin B_{1a} ([redacted]) and unlabelled avermectin B_{1a} ([redacted]). Control treated with blank EC formulation ([redacted])	
Species:	Rhesus monkey (*Macaca mulatta*)	
Guidelines:	No pertinent EC guidelines extant at time of study conduct. Deviations: None	
Date of Report:	2 June 1986	
Published:	No	
GLP:	Yes	

Characteristics

Reference	:	[redacted] (1986)	Exposure	:	1 or 10 h
Type of study	:	Dermal penetration of avermectin B1a.	Doses	:	6 or 300 µg
Year of execution	:	1985	Vehicle	:	DMSO (i.v.) or isopropanol (6.7%, dermal)
Test substance	:	5-^{3}H-avermectin B1a, avermectin B1a (purity unknown)	GLP statement	:	Yes (without date)
Route	:	dermal or i.v.	Guideline	:	-
Species	:	Rhesus monkey	Acceptability	:	acceptable
Group size	:	4 m			

Study design

A group of 4 mature male monkeys were treated according the following schedule:

Test article	Dose of Avermectin B1a (on 6 cm^2)	Route	Exposure duration (hours)
^{3}H-avermectin B_{1a} (in DMSO)	6µg	i.v.	-
^{3}H-avermectin B_{1a} (in DMSO)	300µg	i.v.	-
EC[a]	6µg	dermal	1
EC[a]	6µg	dermal	10
^{3}H-avermectin B_{1a} (in 2-propanol)	300µg	dermal	1
^{3}H-avermectin B_{1a} (in 2-propanol)	300µg	dermal	10
EC[a]	300µg	dermal	1
EC[a]	300µg	dermal	10

[a] emulsifiable concentrate formulation of ^{3}H-avermectin B_{1a}

The treatments were sequential and separated by at least a 3 week (i.v.) or a 4-week (dermal) washout period. Intravenous doses were administered via the saphenous vein and dermal doses were applied to a 6 cm^2 area of clipped skin on the left forearm. The animals were immobilised by ketamine HCl and deprived of food for 18 h prior to treatment until 8 or 10 h after treatment. Blood samples were taken from the femoral vein at 5 minutes (i.v. only), 15 and 30 minutes, 1, 2, 4, 8, 10 (10h dermal only) and 24 h after treatment, then daily for 9 days. Urine and faeces were collected seperately pre-dose, at 8 or 10 h and 24 h after treatment, then daily for up to 15 days. Samples of urine, faeces, plasma and extracts of application site skin swab were analysed by liquid scintillation counting, direct or after combustion. The extent of dermal penetration was determined by using the following equatations:

% dermal absorption (excreta)= total radioactivity excreted (topical dose) x 100% / total radioactivity excreted (i.v. dose)

% dermal absorption (plasma)= AUC (topical dose) x 100% / AUC (i.v. dose)

Results

Following i.v. administration of ^{3}H-avermectin B1a approximately 95 and 97% of the 6 and 300 µg doses, respectively, were excreted in faeces and urine within 10 days of administration. More than 96% of this radioactivity was excreted in the faeces.

After dermal application total recoveries of radioactivity ranged from 75 to 97%. Due to the low levels of

radioactivity in plasma and fluctuations in the background radioactivity it was not possible to calculate plasma AUC values for most of the animals. Only a small amount of the radioactivity was found in the urine (0.01-0.06%) and faeces (0.09-0.50%). Based on the urinary and faecal excretion of radioactivity, dermal absorption was calculated to be 0.15 to 0.50% for the tested dermal regimes. The mean % dose absorbed of the emulsifiable concentrate formulation of 3H-avermectin B1a was twice as high as the mean % dose absorbed of 3H-avermectin B1a in 2-propanol. There were no substantial differences in the extent of dermal penetration between dose levels, exposure times and formulations.

Recovery of radioactivity for the concentrate

Absorption is expressed as mean percentage of the applied dose ± SD.

Treatment	% of dose recovered from skin ± SD	% of dose excreted in faeces ± SD	% of dose excreted in urine ± SD	Recovery ± SD	Mean % absorbed
300 µg i.v.		93.73 ± 4.8	3.29 ± 1.16	97.0 ± 3.7	
300 µg abamectin B1a – 1 h	81.06 ± 6.6	0.09 ± 0.03	0.06 ± 0.09	81.2 ± 6.6	0.15
300 µg abamectin B1a – 10 h	75.17 ± 4.2	0.24 ± 0.08	0.01 ± 0.01	75.4 ± 4.1	0.25
300 µg EC – 1 h	96.70 ± 7.9	0.19 ± 0.16	0.03 ± 0.05	96.9 ± 7.9	0.22
300 µg EC – 10 h	95.74 ± 8.0	0.50 ± 0.17		96.3 ± 8.0	0.50

Recovery of radioactivity for the dilution

Absorption is expressed as mean percentage of the applied dose ± SD.

Treatment	% of dose recovered from skin ± SD	% of dose excreted in faeces ± SD	% of dose excreted in urine ± SD	Recovery ± SD	Mean % absorbed
6 µg i.v.		92.95 ± 5.6	2.34 ± 0.53	95.3 ± 5.8	
6 µg EC – 1 h	89.48 ± 4.4	0.17 ± 0.16	0.03 ± 0.06	89.7 ± 4.3	0.21
6 µg EC – 10 h	85.56 ± 6.9	0.42 ± 0.38	0.05 ± 0.08	86.0 ± 6.4	0.47

Acceptability

The study is considered acceptable for the overall toxicological evaluation.

Conclusion

The extent of dermal penetration of avermectin B1a is minimal in the resus monkey, amounting to less than 1% of the applied dose. The duration of exposure and applied dose do not influence the extent of dermal penetration.

98/8 Doc IIIA 6.2/ 08 section No.	**Percutaneous absorption (in vitro test)**	**Official use only**
91/414 Annex Point addressed	II 6.1 / 04	

Title:	In vitro dermal penetration of avermectin B1a.	
Lab Report Number:	CTL/JV1854/REG/REPT	
Authors:	[redacted] 2005	
Test Substance:	[^{14}C]-avermectin B_{1a}, radiochemical purity [redacted], specific activity 2.5 MBq/mg, and blank formulation Vertimec 018EC (A8612AB).	
Species:	Human skin	
Guidelines:	OECD guideline 428 Deviations: None	
Date of Report:	22 December 2005	
Published:	No	
GLP:	Yes	

Characteristics

Reference	:	[redacted] 2005)	Exposure	:	24 h; unoccluded
Type of study	:	*In vitro* dermal penetration of avermectin B1a.	Doses	:	180 µg a.i./cm^2 and 18 µg/cm^2
Year of execution	:	2005	Vehicle	:	water
Test substance	:	[^{14}C]-avermectin B1a, radiochemical purity [redacted] specific activity 2.5 MBq/mg, and blank formulation Vertimec 018EC (A8612AB)	GLP statement	:	Yes
Route	:	dermal	Guideline	:	OECD guideline 428
Species	:	Human	Acceptability	:	acceptable
Group size	:	5 replicates concentrate 5 replicates dilution			

Study design

The percutaneous absorption of [^{14}C]-avermectin B1a, formulated as Vertimec 018EC, was studied *in vitro* in glass diffusion cells. Epidermal membranes from humans were exposed to either 180 µg/cm^2 or 18 µg/cm^2. The integrity of the skin membranes was established prior to the application of the test substance, by measurement of the electrical resistance of the membranes. Cells were selected such that each applicaton was represented by five intact membranes from at least three different subjects. The formulation was applied to the skin membrane as the concentrate and as a 1:10 v/v aqueous dilution at a dose rate of 10 µl/cm^2. The test substance remained in contact with the skin for 24 hours, unoccluded. Samples of 0.5 mL of receptor fluid (50% ethanol/water) were taken at pre-treatment, 1, 2, 3, 4, 6, 8, 10, 12, 16, 20, and 24 hours after application. At the end of the exposure period, the epidermal membrane was decontaminated by gently swabbing the application site with natural sponges with 3% Teepol and further with sponges with water. The stratum corneum was removed by repeated application of adhesive tape, to a maximum of 5 strips. The exposure chambers were washed with ethanol. The receptor fluid, tissue swabs, skin washings, tape strips, the skin and the exposure chamber washes were analysed using LSC.

Results

The results are summarised in the Table below, where data are presented both in terms of absorption rate and in terms of amount and percentage of the dose applied (based on the amount in the receptor fluid).

Absorption rates and mean amount and percentage of dose absorbed.

Details of test material application	Mean absorption rates		Mean amount and percentage of dose absorbed		
	Time period (h)	Absorption rate ($\mu g/cm^2/h \pm SEM$)	Time (h)	Amount ($\mu g/cm^2$)	Percent Absorbed
Concentrate formulation					
(18 g abamectin/L)	0 - 6	0.005 ± 0.002	6	0.039	0.020
10 $\mu l/cm^2$ (180 μg ai/cm^2)	6 - 24	0.004 ± 0.002	8	0.048	0.025
Unoccluded	0 - 24	0.004 ± 0.002	10	0.057	0.029
Exposure period 24h			24	0.121	0.061
n = 5					
1:10 v/v aqueous dilution					
(1.8 g abamectin/L)	0 - 6	0.001 ± 0.000	6	0.005	0.029
10 $\mu l/cm^2$ (18 μg ai/cm^2)	6 - 24	0.001 ± 0.000	8	0.006	0.035
Unoccluded	0 - 24	0.001 ± 0.000	10	0.007	0.042
Exposure period 24h			24	0.016	0.088
n = 5					

The data for distribution in the test system are presented in the Tables below, in terms of percent of the applied dose.

Summary of avermectin B1a distribution for the concentrate

Test Compartment	Percentage of Dose Recovered (%)						
						Mean	SEM
	Cell 3	Cell 15	Cell 17	Cell 25	Cell 32		
Donor Chamber	1.28	0.750	0.228	0.834	0.011	0.620	0.225
Skin Wash at 24h	106	107	101	96.3	112	104	2.68
Stratum Corneum	0.061	0.224	0.092	0.076	0.051	0.101	0.032
Remaining Epidermis	0.151	0.222	0.176	0.232	0.012	0.159	0.040
Absorbed [1]	0.012	0.109	0.040	0.128	0.017	0.061	0.024
Total % potentially absorbed [2]						**0.258**	
TOTAL	108	108	101	97.6	112	105	2.58

[1] receptor fluid

[2] receptor fluid + remaining epidermis + tape strips 3 – 5 (see text below)

Summary of avermectin B1a distribution for the 1/10 dilution

Test Compartment	Percentage of Dose Recovered (%)						
						Mean	SEM
	Cell 42	Cell 43	Cell 46	Cell 50	Cell 60		
Donor Chamber	0.017	0.016	0.153	1.13	0.047	0.272	0.251
Skin Wash at 24h	94.0	99.3	96.7	87.9	91.8	93.9	1.97
Stratum Corneum	0.113	0.113	0.340	0.340	0.113	0.204	0.055
Remaining Epidermis	0.055	0.067	0.545	0.495	0.367	0.306	0.104
Absorbed [1]	0.045	0.057	0.023	0.249	0.057	0.086	0.041
Total % potentially absorbed [2]						**0.479**	
TOTAL	94.3	99.6	97.7	90.1	92.4	94.8	1.72

[1] receptor fluid

[2] receptor fluid + remaining epidermis + tape strips 3 – 5 (see text below)

Total mean recovery of the concentrate and 1/10 dilution was 105 and 95%. Absorption of avermectin B1a

through human epidermis was very slow during the 24 hour exposure period. The absorption rate was similar throughout the 24 hour exposure period. For the concentrate 0.061% had penetrated through the human skin during 24 hours of exposure. For the dilution 0.086% of the applied dose penetrated within 24 hours through human skin.

The amount in the remaining epidermis may become systemically available and will therefore be considered as potentially absorbed. In this *in vitro* study, tape stripping was performed, but in the report no results are presented for the individual tape strips. In general for an *in vitro* study, only the first two tape strips are regarded as not absorbed. The notifier additionally provided the results of the individual tape strips. For the concentrate, 0.041 and 0.022% of the applied dose was present in tape strip 1 and 2, respectively. For the dilution, 0.068 and 0.049% of the applied dose was present in tape strip 1 and 2, respectively. Avermectin B1a in the epidermal membranes and in the tape strips 3 – 5 should be included into the potentially absorbed amount. The potentially absorbed dose for the concentrate and the dilution is therefore 0.3% and 0.5% (rounded values), respectively.

Acceptability

The study is considered acceptable.

Conclusion

The extent of dermal penetration of avermectin B1a is minimal through human skin *in vitro*, amounting to 0.3 and 0.5% of the applied dose for the concentrate and 1/10 dilution, respectively.

Overall conclusion on dermal absorption of abamectin

The dermal absorption studies are performed with avermectin B1a. Abamectin is a mixture of avermectin B1a (>80%) and avermectin B1b (<20%). Since avermectin B1a is the major component and structurally almost identical to avermectin B1b, the results with avermectin B1a can be used to derive a dermal absorption value for abamectin.

The correct dose level (180 µg/cm^2) was tested for the concentrate in the *in vitro* study. It should be noted that this dose level is relatively low for a concentrate, due to the low amount of abamectin in the formulation Vertimec (18 g/L). In the *in vitro* study, a 1/10 dilution was also tested. In practice, the formulation Vertimec will be further diluted for the spray solution. It is however not feasible to apply a lower concentration than a 1/10 dilution because of the technical limitations imposed by the low amount applied and the specific activity of the radiolabel. But because the dose level of the concentrate is already relatively low, it is not expected that there is still a significant dose-dependent difference in dermal absorption. This is illustrated by the study results which show no significant difference in dermal

absorption between the concentrate and the 1/10 dilution in the *in vitro* study and between the concentrate and the 1/50 dilution in the *in vivo* study.

It should furthermore be taken into account that the exposure period in the *in vitro* study was 24 hours. Based on the *in vivo* study in monkeys and *in vitro* study with human skin, it can be concluded that the dermal absorption of abamectin will be less than 1%.

The poor penetration of avermectin B1a through the skin is consistent with its physicochemical properties. Avermectin B1a is a large, hydrophobic macrocyclic compound (MW 873 and log Pow 4.4) and it is not expected that a molecule of this size would easily diffuse through the skin.

For the risk assessment, a dermal absorption value of 1% will be used.

98/8 Doc IIIA 6.2/09 section No.	Oral study in monkeys – plasma levels	Official use only
91/414 Annex Point addressed	No corresponding annex point	

Title:	MK-0933 and MK-0936 oral toxicity and plasma level study in monkeys	
Lab Report Number:	TT 85-013-0	
Authors:	[redacted] (1985d)	
Test Substance:	Abamectin (batch no. [redacted], purity [redacted]	
Species:	Rhesus monkeys (*Macaca mulatta)*	
Guidelines:	Not applicable (investigative study)	
Date of Report:	6 December 1985	
Published:	No	
GLP:	Yes	

In DAR : STUDY 8 (B.6.8 Further toxicological studies, B.6.8.2 Supplementary studies)

Characteristics

Reference/notifier	:	[redacted] (1985d)	Exposure	:	29 weeks
Type of study	:	Oral toxicity and plasma level study in monkeys.	Doses	:	0.2 to 24.0 mg/kg bw/day (see dosing schedule)
Year of execution	:	1985	Vehicle	:	Sesame oil
Test substances	:	Abamectin (purity [redacted]) or ivermectin (purity [redacted])	GLP statement	:	yes
Route	:	Oral (gavage)	Guideline	:	Not applicable (in house investigative study)
Species	:	Monkey (Macca mulatta)	Acceptability	:	acceptable
Group size	:	2/sex abamectin and 2/sex ivermectin	$NOAEL_{abamectin}$	:	1.0 mg/kg bw
			$NOAEL_{ivermectin}$	:	1.0 mg/kg bw

Study design

Two groups of 2 male and 2 female 2-3 year old rhesus monkeys were treated orally, by gavage, with

repeated single doses of abamectin or ivermectin in sesame oil, at 2 - 3 week intervals for a total of 13 doses, according to the schedule given in the table below:

Dosing Schedule

Week of study	Dose level (mg/kg bw)
1, 4, 6	0.2
8	0.5
10	1.0
13	2.0
15	4.0
17	2.0
19	6.0
21	8.0
24	8.0
27	12.0
29	24.0

After the first 2 doses of either abamectin or ivermectin, treatment of the groups was crossed-over in order to elucidate slight pupil dilatation in 2 animals treated with ivermectin. Thereafter the groups continued to receive the same test substance. Clinical signs were recorded daily. Food consumption was estimated 5 times/week. Body weights were recorded twice each week up to week 9 and weekly thereafter. Ocular examinations for mydriasis were performed using a pen-light light source pre-dose and at 2, 4 and 24 h post-dose for all doses except 24.0 mg/kg bw (8 and 48 h examinations). Analysis of plasma for abamectin or ivermectin concentration in plasma samples was determined 15 and 30 minutes, 1, 2, 4, 8, 24, 48, 72 and 96 h after treatment at 2.0 mg/kg bw in week 17. Plasma samples were also taken 8, 24 and 48 h after treatment at 8.0 mg/kg bw in week 24, and after 8, 24, 48, 72 and 96 h following treatment at 24.0 mg/kg bw in week 29.

Results

Both abamectin and ivermectin produced emesis at dose levels of ≥ 2.0 mg/kg bw with a dose-related increase in incidence and decrease in time of onset. Emesis is considered to be the most sensitive indicator of toxicity in rhesus monkeys and the minimum toxic dose for both test substances is considered to be 2.0 mg/kg bw. Mydriasis occurred only in response to abamectin at 24.0 mg/kg bw. Pupil dilatation and/or decreased pupil constriction occurred at dose levels of ≥ 6.0 mg/kg bw abamectin and ≥ 12.0 mg/kg bw ivermectin, but there was no relationship between dose level and time of onset and

duration of effect. The incidence of mydriasis was higher in the group treated with abamectin than in the ivermectin-treated group. Three animals in each group displayed transient reduced activity or sedation after treatment at 24.0 mg/kg bw. The effect had a variable time of onset and persisted for up to 25 hours. There were no other treatment-related clinical signs and no effect on food consumption and body weight at any dose level in either group.

Plasma concentrations of abamectin and ivermectin were similar for 4 hours after a dose of 2.0 mg/kg bw but thereafter the plasma concentrations of ivermectin were generally higher than abamectin. The peak plasma concentrations of abamectin and ivermectin occurred 8-24 h post-treatment. There was an approximate proportionality between dose and plasma concentration for both compounds. There was a poor correlation between plasma levels in individuals and the occurrence of emesis and the incidence and severity of mydriasis.

Acceptability

The study is considered acceptable as exploratory study.

Conclusions

Clinical signs of toxicity elicited by abamectin and ivermectin are confined to emesis, mydriasis and sedation. The most sensitive indicator of abamectin and ivermectin toxicity in rhesus monkeys is emesis, observed at and above 2.0 mg/kg bw. Clinical signs of toxicity seen in mice and rats (tremors and convulsions) did not occur. The maximum plasma concentrations of both abamectin and ivermectin occur between 8 and 24 hours after oral administration, increasing generally less than proportionally with dose.

SYNGENTA CONCLUSIONS

Conclusion:	Clinical signs of toxicity elicited by abamectin and ivermectin are confined to emesis and mydriasis at dose levels up to 12.0mg/kg and, additionally, sedation at 24.0mg/kg. The most sensitive indicator of abamectin and ivermectin toxicity in rhesus monkeys is emesis, as clinical signs of toxicity seen in mice and rats (tremors and convulsions) did not occur. The minimum toxic dose of both abamectin and ivermectin is 2.0 mg/kg and the highest NOEL is 1.0 mg/kg. The maximum plasma concentrations of both abamectin and ivermectin occur between 8 and 24 hours after oral administration. Plasma	

	concentrations increase with increasing dose but generally less than proportionally with dose. Approximate 5-fold and 7-fold increases in the plasma concentrations of abamectin and ivermectin, respectively, over those produced by the minimum toxic dose, do not elicit more severe clinical signs of toxicity, other than slight to moderate sedation	

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	*6 November 2007; updated January 2009*
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]
Acceptability	[redacted]
Remarks	[redacted]
	COMMENTS FROM ...
Date	*Give date of comments submitted*

Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion. Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

List of identified compounds

See study 6.

Proposed metabolic pathway of abamectin in the rat

See study 6.

Summary from abamectin PPP DAR and/or revised addendum (Febr. 2008). This is included for information.

The absorption, tissue distribution and elimination of avermectin B1a were investigated in the rat using ^{3}H- and ^{14}C-radiolabeled compounds. Samples derived from this study were subsequently analysed in a further study to investigate the metabolism of avermectin B1a in rat tissues. The dermal penetration of avermectin B1a in isopropanol or emulsifiable concentrate (EC) formulation was investigated in rhesus monkeys. Furthermore, an *in vitro* dermal absorption study with human skin was performed.

Orally-administered avermectin B1a is absorbed relatively quickly from the gastrointestinal tract and is distributed throughout all major tissues and organs sampled, with the highest residues found in fat. Maximum concentrations in blood are achieved within 4-8 hours after administration. The half lives for elimination of radioactivity from blood were 9 and 18 h in males and 13 and 27 h in females in the low and high dose, respectively. Avermectin B1a is rapidly eliminated from the body, almost exclusively in the faeces (more than 92% of the dose within 7 days, urinary excretion accounting for 0.9-1.6% of dose in males and 0.5-1.0% in females of low and high dose groups) and there is no evidence for tissue accumulation on repeated administration. After multiple dosing, the tissue residues reached a plateau after 7 days of dosing for all tissues, except for fat, ovaries and spleen, and residues declined rapidly after the dosing period. The rate of excretion was slower in females as compared to males, with males excreting 81% and 73% of the dose within 48h after administration and females excreting within the

same period 69% and 48% of the low and high dose, respectively. The excretion via expired air accounted for only 0.01% of the dose within 48h after administration. Residue levels were greatest in all tissues 24h after treatment and were dose-dependent, whereas the depletion rates were independent of dose level. The tissue residue levels of female rats were generally higher than the corresponding residue levels in male rats. Tissue half-lives were mostly within the range 1.2 ± 0.3 days. So, with the exception of dose-dependence for tissue residue levels and excretion by urine, the toxicokinetic profile is not influenced by sex, dose level or treatment regime.

The comparison of urinary excretion after oral or intravenous administration indicated almost complete oral absorption, with a **calculated oral bioavailability of 0.86 (86% oral absorption)**. Intravenous administration confirmed the non biliary excretion of the majority of absorbed avermectin B1a directly into the intestinal tract with ultimate elimination in the faeces. The tissue half-lives of avermectin B1a are lower in males (12 to 17 h) than in females (13 to 33h). Avermectin B1a and/or metabolites do not accumulate in liver, kidneys, muscle or fat on repeated administration of a low dose. Seven days after the last of 14 daily consecutive doses less than 1% of the total administered dose was present in tissues and organs, except for fat (more than 10 times higher levels compared to other tissue residue levels).

A comparative distribution and clearance study with avermectin B1b following single oral doses showed that the toxicokinetic profile was essentially the same as that of avermectin B1a.

The metabolite pattern in urine, faeces and bile was complex, and 11 metabolites were isolated. In faeces, avermectin B1a accounted for 25 to 45% of the dose and the metabolites [3''DM], [24aOH], [27aOH], [3''DM,24aOH] and [3''DM,27aOH] accounted each for 3-27% of the dose. The major faecal components avermectin B1a and metabolite [3''DM] were not present in urine, and the metabolites [24aOH], [27aOH], [3''DM,24aOH] and [3''DM,27aOH] each accounted for 0.02-0.4% of the dose. In bile, [3''DM,24aOH], [3''DM,27aOH], [3''DM] and avermectin B1a each accounted for 0.1-1.2% of the dose. In the fat and muscle, avermectin B1a was the major component (92% and 72%, respectively), and metabolite [3''DM] accounted for 1.7 and 19% in the fat and muscle, respectively.

Based on the structures identified, the major reactions involved in the biotransformation of avermectin B1a in the rat are demethylation, hydroxylation, cleavage of the oleandrosyl ring and oxidation reactions. The proposed metabolic pathway is given.

In rat the 8,9-Z isomer of abamectin B1a is not formed.

The extent of dermal penetration of avermectin B1a is minimal in the rhesus monkey, amounting to less than 1% of the applied dose. The duration of exposure, applied dose and formulation do not influence the extent of dermal penetration. The *in vitro* study with abamectin formulated as Vertimec 018EC confirms the results of the *in vivo* study.

The poor penetration of avermectin B1a through the skin is consistent with its physicochemical properties. Avermectin B1a is a large, hydrophobic macrocyclic compound (MW 873 and log Pow 4.4)

and it is not expected that a molecule of this size would easily diffuse through the skin.
For the risk assessment, a dermal absorption value of 1% will be used.

A study in the juvenile rhesus monkey showed that the most sensitive indicator of abamectin toxicity in rhesus monkeys is emesis, which occurs at dose levels at and above 2.0 mg/kg bw. The incidence of emesis is dose-related and the time of onset generally decreases with increasing dose level. Typical clinical signs of abamectin toxicity, tremors and convulsions, are absent in the rhesus monkey and pupil dilation or decreased pupil constriction occur only at dose levels at and above 6.0 mg/kg bw. Rhesus monkeys seem to be relative insensitive for the acute effects of abamectin since clinical signs of neurotoxicity are confined to transient sedation and marked mydriasis at 24 mg/kg bw. Based on the occurence of emesis at and above 2.0 mg/kg bw, the NOAEL in this study is 1.0 mg/kg bw. The maximum plasma concentrations of abamectin occurred 8 - 24 h after oral administration and plasma concentrations increase with increasing dose, but less than proportionally. Despite increases in plasma concentration of abamectin with increasing dose level, the severity of clinical signs did not markedly increase. Therefore, the dose-response relationship for abamectin in monkeys appears to be much flatter than in dogs and rodent species.

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	*6 November 2007; updated January 2009*
Materials and Methods	[redacted]
Results and discussion	[redacted]
Conclusion	[redacted]
Reliability	[redacted]

Acceptability	[redacted]
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion. Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

3. SHORT TERM TOXICITY

98/8 Doc IIIA 6.3.1/ 01 section No.	Short term repeated dose toxicity	Official use only
91/414 Annex II Point addressed 5.3.1 / 01	repeated dose toxicity - oral	

Title:	MK-0936 - 8-week dietary range-finding study in rats	
Lab Report Number:	No. TT 82-075-0-1	
Authors:	[redacted] (1984b)	
Test Substance:	Abamectin (MK-0936, batch no. [redacted], purity [redacted] by UV fluorescence)	
Species:	Rat	
Guidelines:	No applicable guideline, but deviates from 88/302/EEC, B.26, sub-chronic oral toxicity test in rodents (May 1988) in the following respects: Ophthalmoscopy, haematology, clinical chemistry, urinalysis, necropsy, post mortem examination, organ weight analysis and histopathology not performed.	
Date of Report:	3 July 1984	
Published:	No	
GLP:	No	

28-day oral studies

STUDY 1
Characteristics

Reference/notifier	:	[redacted] (1984b)	Exposure	:	8 or 4 weeks
Type of study	:	8-week/4 week oral range-finding study	Dose[1]	:	0, 5, 10, 15, 20/25, 40 (and 60) ppm (mean weekly achieved

					dose 0, 0.3-0.7, 1.0-1.4, 1.6-2.2, 1.7-2.7 and 4.1-5.8 mg/kg bw/day)
Year of execution	:	1982	Vehicle	:	acetone
Test substance	:	Abamectin technical (purity [redacted])	GLP statement	:	no
Route	:	Oral (diet)	Guideline	:	-
Species	:	Rat, CRCD	Acceptability	:	As range-finding study only
Group size	:	10/sex/dose	NOAEL	:	-

1: a dose level range could not be calculated for the 60 ppm group. The 20 ppm concentration was increased to 25 ppm at the start of week 7.

Study design

In this study, only body weight and food consumption were determined, and clinical signs were described but not presented individually.

Groups of 10/sex were orally dosed with abamectin at concentrations of 0, 5, 20/25, 40 or 60 ppm. The 20 ppm concentration was increased to 25 ppm at the start of week 7. In the 40 and 60 ppm groups severe toxicity and mortality occured, and surving animals in these groups were sacrificed on day 15 and 5 respectively. Two additional groups of animals were placed on test in week 5 and were treated for 4 weeks at 10 and 15 ppm.

Results

In week 1, all animals of the 60 ppm group and 6m/4f of the 40 ppm group died. The remaining 4m/6f of the 40 ppm group were sacrificed in week 3. One female in the 15 ppm group was killed in moribund condition on day 16 (hydrocephalus). Treatment-related clinical signs were observed in animals dosed 40 and 60 ppm on day 2, and included tremors, decreased activity, red-coloured discharge in the oro-nasal region and staining of the urogenital area. Slight tremors and/or urogenital staining also occurred in some animals treated at 15 and 20/25 ppm, but were no longer apparent after week 3. One female of the 5 ppm dose group showed slight tremors on day 2 only.

In week 1 and 2 body weight gain was reduced in both sexes by approximately 50% in the 40 ppm group and in the 15 and 20/25 ppm groups by 9-18% and 15-19% respectively. In week 1 and 2 food consumption was reduced by 25-47% in both sexes of the 40 ppm group.

Acceptability

The study is acceptable as range-finding study only.

Conclusions

No clinical signs or reduced body weight gain was observed in rats dosed up to and including 10 ppm (1.0-1.4 mg/kg bw/day).

Reliability Indicator	1	
Data Protection Claim	Yes	

28-day dermal studies

No study submitted.

Justification below not/partially reported in DAR

98/8 Doc IIIA 6.3.2/ 01 Repeat dose dermal section No.	**Official use only**

Section 6.3 Annex Point IIA 6.3.2/01	**Repeat dose dermal**	
	JUSTIFICATION FOR NON-SUBMISSION OF DATA	Official use only
Other existing data [X]	**Technically not feasible [ ] Scientifically unjustified [ ]**	
Limited exposure [ ]	**Other justification [ ]**	
Detailed justification:	[redacted]	

98/8 Doc IIIA 6.3.3/ 01 section No.	Repeat dose inhalation	Official use only

Section 6.3 Annex Point IIA 6.3.3	Repeat dose inhalation Section 6.3.3/01 Repeat Dose Inhalation, 5 day inhalation study in the rat	
Title:	5 Day Preliminary Inhalation Toxicity Study In The Rat	
Lab Report Number:	No. MR0236	
Authors:	[redacted] (2006)	
Test Substance:	Abamectin technical	
Species:	Rat	
Guidelines:	No (Preliminary study)	
Date of Report:	August 2006	
Published:	No	
GLP:	Yes	

STUDY 1

Characteristics

Reference/notifier	:	[redacted] 2006a	Exposure	:	inhalation (nose-only), 6h/day, 5 days
Type of study	:	sub-acute inhalation (range finding)	Doses	:	actual concentrations: 0, 1.03, 3.71, 9.59 and 24.7 µg/L
Year of execution	:	2006	Vehicle	:	none
Test substance	:	Abamectin technical, purity [redacted]	GLP statement	:	Yes
Route	:	Inhalation	Guideline	:	none
Species	:	Rat (HsdBrlHan:WIST)	Acceptability	:	acceptable as preliminary study
Group size	:	2/sex/dose	$NOAEL_{neurotoxicity}$	:	-

Study design