Committee for Risk Assessment
RAC

Annex 1
Background document
to the Opinion proposing harmonised classification and labelling at Community level of
Indoxacarb and
Indoxacarb
(enantiomeric reaction mass S:R 75:25)

ECHA/RAC/ CLH-O-0000001735-72-01/A1

Indoxacarb and
Indoxacarb (enantiomeric reaction mass S:R 75:25)
EC number: There are no EC-numbers assigned to Indoxacarb (S-enantiomer) or Indoxacarb (enantiomeric reaction mass S:R 75:25).
CAS number: 173584-44-6 (Indoxacarb - S-enantiomer)
There is no CAS-number assigned to Indoxacarb (enantiomeric reaction mass S:R 75:25).

Adopted
1 June 2011

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BACKGROUND TO THE PROPOSAL

Indoxacarb (enantiomeric reaction mass 75:25 S:R) has been reviewed as a new active substance under both the Biocidal Products Directive (98/8/EC) and the Plant Protection Products Directive (91/414/EEC). It was included into Annex I of Directive 91/414/EEC in 2006 and in Annex I of 98/8/EC in July 2009. The hazards of Indoxacarb have been assessed by the UK's Health and Safety Executive and the Dutch Board for the Authorisation of Plant Protection Products and Biocides as part of these regulatory programmes. These assessments were discussed and agreed by the appropriate European technical committees under each review programme.

In accordance with Article 36(2) of EC Regulation 1272/2008 on classification, labelling and packaging of substances and mixtures, Indoxacarb (enantiomeric reaction mass 75:25 S:R) should now be considered for harmonised classification and labelling. This Annex VI dossier presents a classification and labelling proposal based on the information presented following the assessment of Indoxacarb under Directives 98/8/EC and 91/414/EEC. Document IIA of the assessment under 98/8/EC and the DAR (DAR September 2000 + addendum January 2005 + post inclusion addendum 1 October 2007) under 91/414/EEC. These assessments are provided in section 13 of the IUCLID files.

It should be noted that the common name (ISO) Indoxacarb refers only to the S enantiomer (CAS: 173584-44-6, Producers code DPX-KN128) which is the insecticidally active form. During the early development of the product it was not possible to produce the pure S enantiomer on a commercial scale and, consequently, a technical material was produced consisting of approximately a 3:1 ratio of the S:R enantiomers (Indoxacarb (enantiomeric reaction mass S:R 75:25) , Producers code DPX-MP062). This technical material has been used to formulate products in the EU. However, recent developments have meant that it is now viable to manufacture a material that contains only trace amounts of the insecticidally inactive R enantiomer and this 'pure' form is now used for product formulations. Considering the available data on both Indoxacarb (S-enantiomer) and Indoxacarb (enantiomeric reaction mass S:R 75:25), different classification and labelling under Directive 67/548/EEC is appropriate. Consequently, this report contains a classification and labelling proposal for each substance.

The physical properties of Indoxacarb (enantiomeric reaction mass S:R 75:25) cause it to be glue-like and therefore difficult to handle. Consequently, it is sprayed as a solvent solution onto amorphous silica, the solvent is evaporated and the manufacturing use product (DPX-MP062 MUP) is generated. The MUP typically contains 25.6% silica (range 20-30%). A number of studies have been conducted using this material and these will be indicated in the report.

In addition, data are also available on a third technical grade, a racemic mixture [Indoxacarb (racemic mixture S:R 50:50)] CAS 144171-61-9, Producers code DPX-JW062. Data on this substance have also been used to support the classification and labelling of Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25). This report will indicate which studies have been conducted using this material.

PROPOSAL FOR HARMONISED CLASSIFICATION AND LABELLING

Substance names:	Indoxacarb (S-enantiomer) and Indoxacarb (enantiomeric reaction mass S:R 75:25)
EC number:	605-683-4 (Indoxacarb – S-enantiomer) There is no EC-number assigned to the S:R 75:25 enantiomeric reaction mass
CAS numbers:	173584-44-6 (S-enantiomer) There is no CAS-number assigned to Indoxacarb (enantiomeric reaction mass S:R 75:25)
Registration number(s):	Not applicable – both substances are in the scope of Article 15 of REACH
Purity:	Indoxacarb (S-enantiomer) > 90% Indoxacarb (enantiomeric reaction mass S:R 75:25) ≥ 88.8% 66.7-78.0% S-enantiomer 22.2-28.3% R-enantiomer
Impurities:	A number of process impurities are present in both substances (14 of the impurities can individually be present in a concentration ≥ 1% but are nominally present at < 1%. There are 19 additional impurities but they are all individually present at concentrations < 1%). These impurities have been taken into account in the proposed classification and labelling of Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R) and they are not considered to be of additional toxicological or ecotoxicological concern. The impurities are considered to be confidential so are not listed in this report. Additional information is provided in the technical dossier.

Proposed classification based on Directive 67/548/EEC:

Indoxacarb (pure S enantiomer) CAS : 173584-44-6
T; R25- 48/25,
Xn; R20
R43,
N; R50-53

Indoxacarb (enantiomeric reaction mass S:R 75:25) There is no CAS-number assigned to this substance
T; R48/25,

Xn; R20/22
R43,
N; R50-53

Proposed classification based on CLP criteria:

Indoxacarb (pure S enantiomer) CAS : 173584-44-6 and Indoxacarb (enantiomeric reaction mass S:R 75:25) There is no CAS-number assigned to this substance

Acute tox 4 - H332: Harmful if inhaled
Acute tox 3 - H301: Toxic if swallowed
Skin Sens 1B - H317: May cause an allergic skin reaction
STOT RE 1 - H372: Causes damage to organs (blood, nervous system, heart) through prolonged or repeated exposure
Aquatic Acute 1 - H400: very toxic to aquatic life
Aquatic Chronic 1 - H410: very toxic to aquatic life with long lasting effects

Proposed labelling:

According to Directive 67/548/EEC:

Indoxacarb CAS: 173584-44-6
T, N
R: 20-25-43-48/25-50/53
S: 1/2-24-37-45-60-61

Indoxacarb (enantiomeric reaction mass 75:25) there is no CAS-number assigned to this substance
T, N
R: 20/22-43-48/25-50/53
S: (2-) 24-37-45-60-61

According to Regulation EC 1272/2008:

Indoxacarb CAS : 173584-44-6 and Indoxacarb (enantiomeric reaction mass 75:25) there is no CAS-number assigned to this substance

Pictogram	: GHS06, GHS08, GHS09
Signal word	: Danger
Hazard statement codes	: H301, H317, H332, H372, , H410
Precautionary statements	: Not required as PS are not included in Annex VI.

Proposed specific concentration limits (if any):

According to Directive 67/548/EEC:

Indoxacarb CAS: 173584-44-6 and Indoxacarb (enantiomeric reaction mass 75:25) there is no CAS-number assigned to this substance

Classification of the preparation		
N, R50-53	N, R51-53	R52-53
Cn ≥ 25%	2.5% ≤ Cn <25%	0.25% ≤ Cn <2.5%

Where Cn is the concentration in the preparation.

According to Regulation EC 1272/2008:

Under CLP M factor 1 based on 0.1 <L(E)C_{50} ≤1 mg/l.

Based on the 2nd ATP of the CLP Regulation
Acute M-factor: 1 based on 0.1<L(E)C50≤1 mg/l and
Chronic M-factor: 1 based on 0.01<NOEC≤0.1 mg/l

Proposed notes (if any):

None

JUSTIFICATION

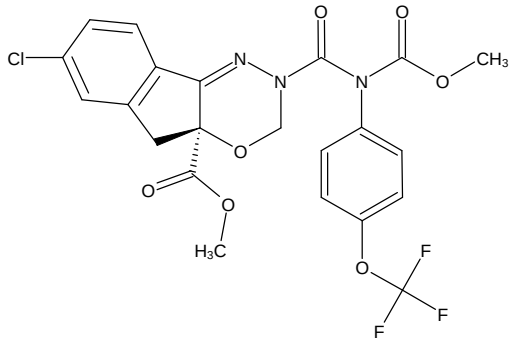
1 IDENTITY OF THE SUBSTANCE AND PHYSICAL AND CHEMICAL PROPERTIES

1.1 Name and other identifiers of the substances

Chemical Name:	Indoxacarb
EC Number:	605-683-4
CAS Number:	173584-44-6
IUPAC Name:	methyl (4aS)-7-chloro-2-({(methoxycarbonyl)[4-(trifluoromethoxy)phenyl]amino}carbonyl)-2,5-dihydroindeno[1,2-e][1,3,4]oxadiazine-4a(3H)-carboxylate
Producer code:	DPX-KN128

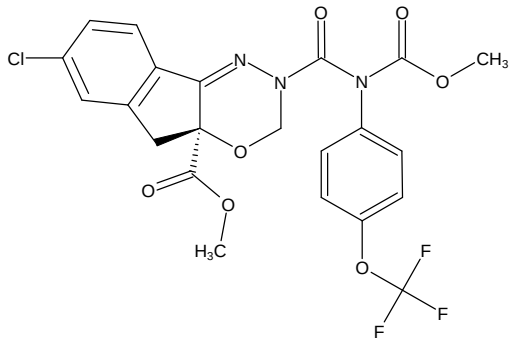
Chemical Name:	Indoxacarb (enantiomeric reaction mass S:R 75:25)
EC Number:	No EC number assigned
CAS Number:	No CAS number assigned
IUPAC Name:	reaction mass of methyl (4a*S*) and methyl (4aR)-7-chloro-2-({(methoxycarbonyl)[4-(trifluoromethoxy)phenyl]amino}carbonyl)-2,5-dihydroindeno[1,2-*e*][1,3,4]oxadiazine-4a(3*H*)-carboxylate
Producer code:	DPX-MP062

1.2 Composition of the substances

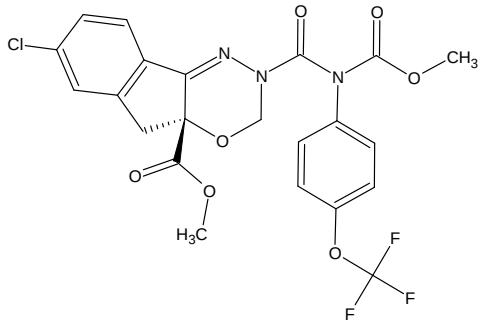
Chemical Name:	Indoxacarb
EC Number:	605-683-4
CAS Number:	173584-44-6
IUPAC Name:	methyl (4aS)-7-chloro-2-({(methoxycarbonyl)[4-(trifluoromethoxy)phenyl]amino}carbonyl)-2,5-dihydroindeno[1,2-e][1,3,4]oxadiazine-4a(3H)-carboxylate
Molecular Formula:	$C_{22}H_{17}ClF_3N_3O_7$
Structural Formula:	
Molecular Weight:	527.84
Typical concentration (% w/w)	94%

Chemical Name: Indoxacarb (enantiomeric reaction mass S:R 75:25)
EC Number: Not yet assigned
CAS Number: No CAS number assigned
IUPAC Name: reaction mass of methyl (4a*S*) and methyl (4aR)-7-chloro-2-({(methoxycarbonyl)[4-(trifluoromethoxy)phenyl]amino}carbonyl)-2,5-dihydroindeno[1,2-*e*][1,3,4]oxadiazine-4a(3*H*)-carboxylate

Molecular Formula: $C_{22}H_{17}ClF_3N_3O_7$
Structural Formula: S-enantiomer 173584-44-6 (S)



R-enantiomer 185608-75-7 (R)



Molecular Weight: 527.84
Typical concentration (% w/w) 70.8%
methyl (4aS)-7-chloro-2-({(methoxycarbonyl)[4-(trifluoromethoxy)phenyl]amino}carbonyl)-2,5-dihydroindeno[1,2-e][1,3,4]oxadiazine-4a(3H)-carboxylate
23.7%
methyl (4aR)-7-chloro-2-({(methoxycarbonyl)[4-(trifluoromethoxy)phenyl]amino}carbonyl)-2,5-dihydroindeno[1,2-e][1,3,4]oxadiazine-4a(3H)-carboxylate

1.3 Physico-Chemical properties

Table 1.1: Summary of physico-chemical properties for Indoxacarb (CAS: 173584-44-6)

REACH ref Annex, §	Property	IUCLID section	Value	Reference/Comment
VII, 7.1	Physical state at 20°C and 101.3 KPa	4.1	White powder with mild innocuous odour.	(1) Purity 99.7%
VII, 7.2	Melting / freezing point	4.2	88.1°C	(1) Purity 99.7% 92/69/EEC A1
VII, 7.4	Relative density	4.4 density	1.44	(1) Purity 99.7% 92/69/EEC A3
VII, 7.5	Vapour pressure	4.6	9.8×10^{-9} Pa at 20°C 2.5×10^{-8} Pa at 25°C	(2) Purity 99.7% 92/69/EEC A4
VII, 7.6	Surface tension	4.10	Test not conducted as water solubility is < 1 mg/l	
VII, 7.7	Water solubility	4.8	0.2 mg/l at 25°C	(1) Purity 99.7% 92/69/EEC A6
VII, 7.8	Partition coefficient n-octanol/water (log value)	4.7 partition coefficient	4.65 at 25°C	(1) Purity 99.7% 92/69/EEC A8
IX, 7.16	Dissociation constant	4.21	Not determined as the substance does not ionise in water.	(1)

Table 1.2: Summary of physico-chemical properties for Indoxacarb (enantiomeric reaction mass S:R 75:25) There is no CAS-number assigned to this substance

REACH ref Annex, §	**Property**	**IUCLID section**	**Value**	**Reference/Comment**
VII, 7.1	Physical state at 20°C and 101.3 KPa	4.1	White powder with faint ethyl acetate odour.	(4) Purity 99%
VII, 7.2	Melting / freezing point	4.2	87.1°C – 141.5°C	(5) Purity 99% 92/69/EEC A1
VII, 7.3	Boiling point	4.3	Substance decomposes at 208°C before boiling.	(5) Purity 99% 92/69/EEC A1
VII, 7.7	Water solubility	4.8	0.23 mg/l	(6) Purity 99% OECD 105

Table 1.3: Summary of physico-chemical properties for Indoxacarb (racemic mixture S:R 50:50 (CAS: 144171-61-9)

REACH ref Annex, §	**Property**	**IUCLID section**	**Value**	**Reference/Comment**
VII, 7.1	Physical state at 20°C and 101.3 KPa	4.1	Off-white powder with no discernable odour.	(7) Purity 99.6%
VII, 7.2	Melting / freezing point	4.2	140°C – 141°C	(7) Purity 98.6% 92/69/EEC A1
VII, 7.4	Relative density	4.4 density	1.34	(7) Purity 98.6% 92/69/EEC A3
VII, 7.5	Vapour pressure	4.6	1.3×10^{-10} Pa at 20°C 4.0×10^{-10} at 25°C	(8) Purity 95.6% 92/69/EEC A4
VII, 7.7	Water solubility	4.8	0.015 mg/l	(9) Purity 98.6% 92/69/EEC A6

VII, 7.8	Partition coefficient n-octanol/water (log value)	4.7 partition coefficient	4.6 at 25°C	(11) Purity 96.6% 92/69/EEC A8
VII, 7.10	Flammability	4.13	Not flammable.	(12) Purity 95% 92/69/EEC A10
VII, 7.11	Explosive properties	4.14	Not explosive.	(12) Purity 95% 92/69/EEC A14
VII, 7.12	Self-ignition temperature		No spontaneous ignition occurred.	(12) Purity 95% 92/69/EEC A16
VII, 7.13	Oxidising properties	4.15;	Examination of the chemical structure indicates that it does not contain any chemical groups typical of oxidising agents. Thus the substance can be regarded as incapable of reacting exothermically with a combustible material.	(3)
IX, 7.16	Dissociation constant	4.21	Not determined as the substance does not ionise in water.	(10)

2 MANUFACTURE AND USES

2.1 Manufacture

2.2 Identified Uses

2.3 Uses advised against.

Not necessary for this kind of dossier.

3 CLASSIFICATION AND LABELLING

3.1 Classification in Annex VI of Regulation 1272/2008

Indoxacarb and Indoxacarb (enantiomeric reaction mass S:R 75:25) are not currently listed in Annex VI of Regulation 1272/2008

3.2 Self classification(s)

The applicant has proposed the following classification for both Indoxacarb and Indoxacarb (enantiomeric reaction mass S:R 75:25)

Xn; R20/22
R43
N; R50/53

4 ENVIRONMENTAL FATE PROPERTIES

A detailed summary of the available studies has been reviewed under Directives 98/8/EC and 91/414/EEC (refer to section 13 of the IUCLID). The key information pertinent to determining a classification position is presented below.

No environmental fate studies are available using Indoxacarb (pure S enantiomer). Environmental studies presented in this section have been performed on the following substances:
Indoxacarb (enantiomeric reaction mass S:R 75:25)
Indoxacarb (racemic mixture 50:50 S:R)

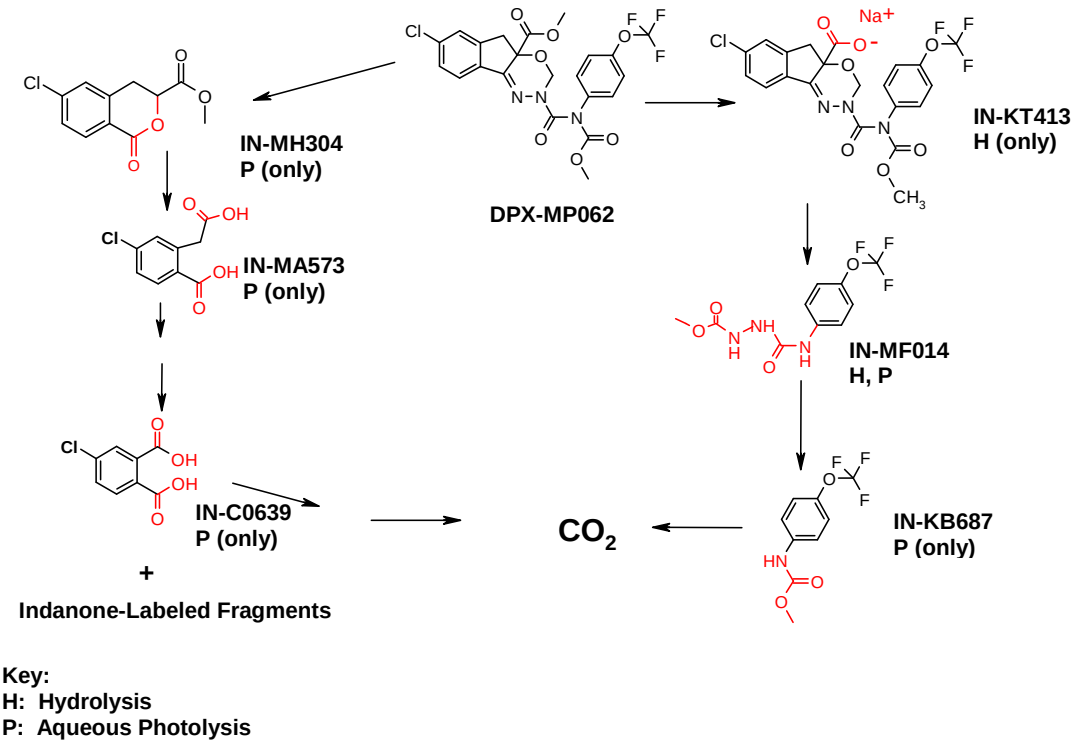
Information on a number of degradation products is included in the section below. For further information on these degradation products please refer to document IIA of the biocides assessment, Annex I to this report.

4.1 Degradation

4.1.1 Stability

A proposed aquatic photolysis and hydrolysis degradation pathway for Indoxacarb is presented below in Figure 4.1.

Figure 4.1: Proposed degradation pathway by aqueous photolysis and hydrolysis for Indoxacarb (enantiomeric reaction mass S:R 75:25)



Hydrolysis
A hydrolysis study [13] following US EPA guidelines (and largely OECD guideline 111) using ^{14}C labelled Indoxacarb (enantiomeric reaction mass 75:25 S:R), showed the substance is hydrolytically stable under acidic (pH 5) conditions with a calculated DT_{50} >365 days at 25°C and 12°C. Moderate hydrolysis was observed at neutral (pH 7) conditions with a calculated DT_{50} of 22 days at 25°C and 62.25 days at 12°C. Under alkaline (pH 9) conditions, rapid hydrolysis was observed with a calculated DT_{50} of 0.3 days at 25°C and 0.85 days at 12°C. Hydrolysis at pH 7 and pH 9 resulted in two major degradants (IN-KT413 and IN-MF014). During the study, at each pH the ratios of S:R enantiomers remained constant.

Under environmental conditions, Indoxacarb (enantiomeric reaction mass 75:25 S:R) is not considered to undergo significant hydrolysis to achieve ultimate degradation and meet the criteria of readily biodegradable. As Indoxacarb (enantiomeric reaction mass 75:25 S:R) comprises of approximately 75% S enantiomer, and S and R enantiomers were not observed to hydrolyse at differing rates, this conclusion is also applied to Indoxacarb.

Photolysis
An aquatic photolysis study [14] following US EPA guidelines using ^{14}C labelled Indoxacarb (enantiomeric reaction mass 75:25 S:R) is available. The study showed that the substance underwent photodegradation under the following conditions: pH 5 (pH at which hydrolysis is considered insignificant), 25°C, and conditions representing 15 days constant midday natural sunlight at 40°N latitude (European relevant locations: Madrid, Naples, Saloniki). The half-life derived from this study was 3 days at 25°C (or 4.5 days when adjusted for sunlight equivalent days). Adjusting the 3 days half life to assume only 12 hours of sunlight and 12°C provides a half-life of approximately 17 days or 25.5 days when adjusted for sunlight equivalent days. Five transformation products were identified (IN-KB687, IN-MF014, IN-MH304, IN-CO639, IN-MA573). During the study, the ratios of S:R enantiomers remained constant.

Although the study shows evidence of photolysis, this will only occur under conditions of strong sunlight and clear waters. With increased turbidity from increased suspended solids and lower sunlight intensity across much of Europe, the half-life is considered to be longer than 28 days under natural conditions. On this basis, Indoxacarb (enantiomeric reaction mass 75:25 S:R) is not expected to photodegrade rapidly in the environment. As Indoxacarb (enantiomeric reaction mass 75:25 S:R) comprises 75% S enantiomer, and S and R enantiomers were not observed to photolyse at differing rates, this conclusion is also applied to Indoxacarb.

Summary
Overall, the studies show that Indoxacarb (enantiomeric reaction mass 75:25 S:R) may undergo degradation through aqueous photolysis or hydrolysis in the environment but it will not rapidly degrade (>70% within a 28-day period to non-toxic substances). As the ratio of S:R enantiomers remained constant during both studies when degradation was observed, the two enantiomers are assumed to degrade at the same rate and similar degradation rates are anticipated for Indoxacarb.

The stability in air is not considered relevant for this type of dossier given that air is not considered an environmental compartment of concern for Indoxacarb *(see Section 4.2.2).*

4.1.2 Biodegradation

4.1.2.1 Biodegradation estimation

As measured data are available estimation is not relevant for this dossier.

4.1.2.2 Screening tests

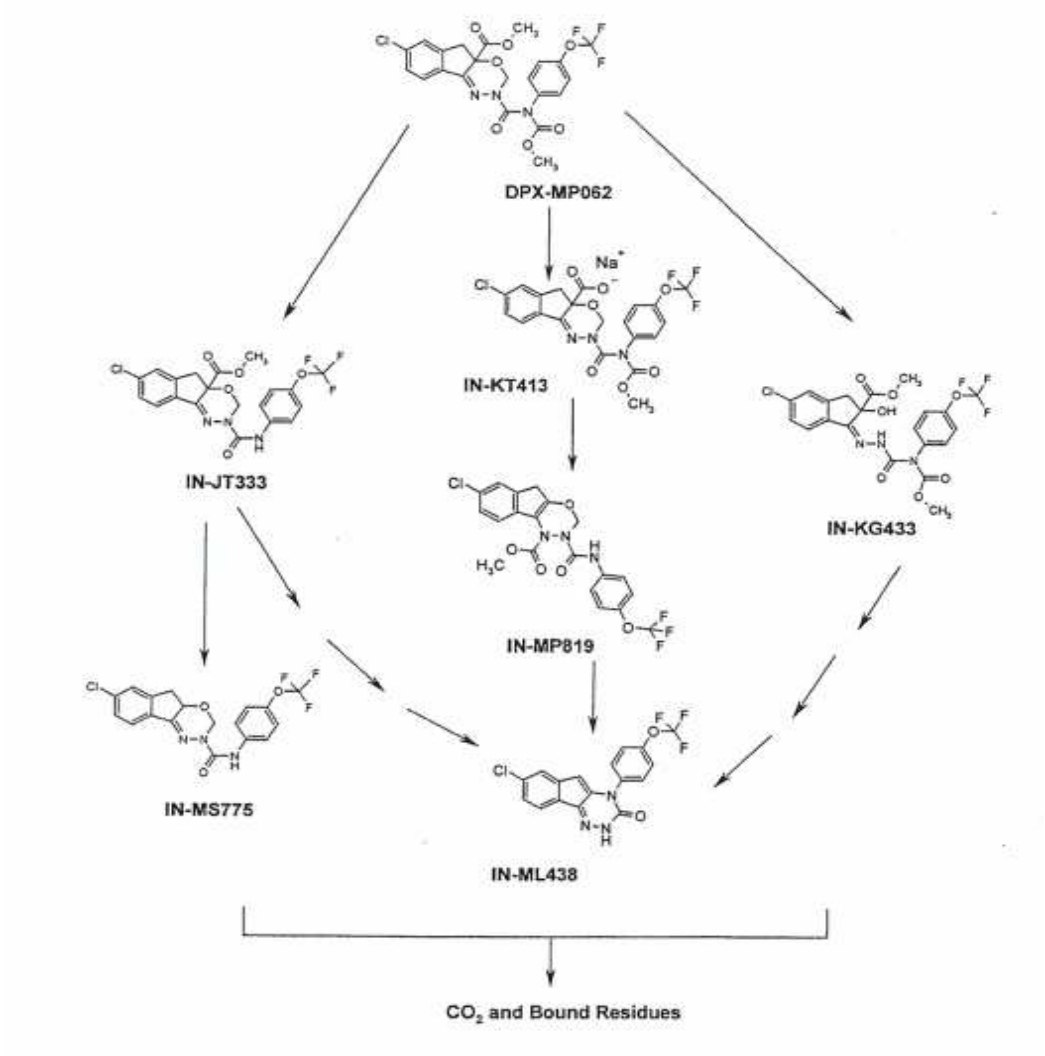
In a ready biodegradation study [15] following OECD Guideline 301c (MITI-I test) using Indoxacarb (racemic mixture 50:50 S:R) only 2-4% degradation was achieved by day 28. During the study, it was noted that the test substances was not fully dissolved in test vessels due to the low solubility of the substance in water. It is therefore possible that the low observed degradation may reflect limited availability for micro-organisms.

Assuming that all the observed degradation represented degradation of one enantiomer the maximum anticipated degradation would be 8% (i.e. twice observed degradation from study with 50:50 ratio). This means, Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R) are also considered not readily biodegradable.

4.1.2.3 Simulation tests

A proposed hydrolysis degradation pathway for Indoxacarb (enantiomeric reaction mass S:R 75:25) is presented below is Figure 4.2 [16].

Figure 4.2: Proposed degradation pathway for Indoxacarb (enantiomeric reaction mass S:R 75:25)



Study 1

Following US EPA and SETAC guidelines using two ^{14}C radiolabels the fate of Indoxacarb (enantiomeric reaction mass 75:25 S:R) was determined in water and sediment for two systems [16]; the first involved a small static pond called 'Bury' with anaerobic sediment (pH 6.9); the second involved a large lake system fed by drainage from moorland called 'Chatsworth' with aerobic sediment (pH 7.9). The laboratory simulation was run for 102 days in the dark at 20°C under flow through conditions.

'Bury' study:

In the Bury study, the water DT_{50} at 20°C was 2 days which is considered representative of primary degradation and adsorption to sediment. One major degradant (IN-KT413) was identified in water which had a DT_{50} of 26 days at 20°C. The sediment DT_{50} at 20°C was 34 days and four major degradants (IN-KT413, IN-JT333, IN-MP819, IN-MS775) were identified with DT_{50} values between 10 and >365 days at 20°C.

At 20°C, the overall total system DT_{50} for Indoxacarb (enantiomeric reaction mass 75:25 S:R) was 12 days with DT_{50} values between 10 and >365 days for identified degradants.

Using the mean of the two radiolabels the following changes in applied radioactivity (AR) were observed over the study period;

- total ^{14}C residues in water decreased from 57.1% AR at day 0 to 5.4% AR after 102 days;
- total extractable ^{14}C residues in sediment decreased from 58.9% AR at day 0 to 50% AR after 102 days; and,
- ^{14}C non-extractable residues (NER) in sediment increased from 0.1% AR at day 0 to 22.7% AR after 102 days.

Using the mean of the two radiolabels, the following applied radioactivity was observed at day 28;

- total ^{14}C residues in water were 16.4% AR; total extractable ^{14}C residues in sediment were 53.1% AR;
- ^{14}C non-extractable residues (NER) in sediment were 10.1% AR; and
- CO_2 residues were 1.8% AR.

The recovery of the two radiolabels was 89.8% (study range of 89.8 to 120.5% recovery) and 92.6% (study range of 91 to 111.5% recovery).

'Chatsworth' study:

In the Chatsworth study, the water DT_{50} at 20°C was 1 day which is considered representative of primary degradation and adsorption to sediment. One major degradant (IN-KT413) was identified in water which had a DT_{50} of 16 days at 20°C. The sediment DT_{50} at 20°C was 5 days and three major degradants (IN-MS7775, IN-KT413, IN-JT333) were identified with DT_{50} values between 19 and 29 days at 20°C.

At 20°C, the overall total system DT_{50} for Indoxacarb (enantiomeric reaction mass 75:25 S:R) was 3 days with DT_{50} values between 14 and 35 for identified degradants.

Using the mean of the two radiolabels the following changes in applied radioactivity (AR) were observed over the study period;

- total ^{14}C residues in water decreased from 55.9% AR at day 0 to 2.8% AR after 102 days;
- total extractable ^{14}C residues in sediment decreased from 62.1% AR at day 0 to 22.8% AR after 102 days;
- and, ^{14}C non-extractable residues (NER) in sediment increased from 0.8% AR at day 0 to 53.0% AR after 102 days.

Using the mean of the two radiolabels, the following applied radioactivity was observed at day 28;

- total ^{14}C residues in water were 5.4% AR; total extractable ^{14}C residues in sediment were 50.3% AR;
- ^{14}C non-extractable residues (NER) in sediment were 32% AR; and
- CO_2 residues were 7.1% AR.

The recovery of the two radiolabels were 91.9% (study range of 60.9 to 128.9% recovery) and 100.8% (study range of 90.3 to 108.5% recovery).

'Bury' and 'Chatsworth' studies

Additional calculations for sediment degradation indicate the following degradation rates for the S and R enantiomer in each system; Bury, S enantiomer DT_{50} 36 days; R enantiomer DT_{50} 29 days; and, Chatsworth, S enantiomer DT_{50} 7 days; R enantiomer DT_{50} 1 day. The difference between the S and R enantiomers is considered to be due to experimental variation as there was only one replicate per sampling event. In addition, due to the degradation of the parent mixture in sediment,

the low levels of radioactivity could have increased the variability. Comparing the 95% confidence intervals for S and R enantiomers showed an overlap indicating that there was no significant difference between the DT_{50} values for S and R enantiomers.

Indoxacarb is metabolised by target organisms to form N-decarbomethoxylated Indoxacarb called IN-JT333. IN-JT333 is insecticidally active and induces a lethal reaction. Whilst IN-JT333 was not identified in water samples, it was present as a major degradant in sediment in both systems. The calculated DT_{50} values at 20°C in aerobic and anaerobic sediments were 29 days and 23 days. The simulation study also indicates that IN-JT333 accounted for ~20% applied radioactivity in the aerobic system on day 28 and 3.35% in the anaerobic system on day 28. IN-JT333 is anticipated to undergo degradation to IN-MS775 (detected in both aerobic [DT_{50} 19 days at 20°C] and anaerobic systems [DT_{50} >1 year at 20°C]) and IN-ML438 (detected in aerobic systems, unknown DT_{50}).

Overall, the study shows that Indoxacarb (enantiomeric reaction mass 75:25 S:R) is rapidly removed from the water phase to the sediment phase where degradation occurs. Given the adsorption capacities discussed in section 4.2.1 this partitioning is considered due to adsorption.

Adjusting water DT_{50} values to an environmentally relevant temperature (12°C) results in DT_{50} values of 4 and 2 days for the Bury and Chatsworth systems respectively. The difference between systems is not considered significant and is assumed to reflect the slightly higher pH in the aerobic system with a potential increase in hydrolysis. In sediment, the degradation rate depends on the level of oxidation. Sediment DT_{50} values were 64.5 days and 9.5 days at 12°C for the anaerobic and aerobic systems respectively. Aerobic conditions appear to produce faster degradation with a total system DT_{50} for Indoxacarb (enantiomeric reaction mass 75:25 S:R) of 5.7 days at $12^{\circ C}$ compared to a DT_{50} of 22.8 days at 12°C in the anaerobic system. It should be noted that the temperature correction may not reflect actual environmental conditions where various micro-organism processes may be optimised at a range of temperatures.

Although Indoxacarb (enantiomeric reaction mass 75:25 S:R) was shown to degrade quickly, especially in aerobic systems, in water and sediment, its degradants are more persistent. Based on CO_2 evolution, by day 28, 1.8% mineralisation was observed in the anaerobic system, and 7.1% mineralisation was observed in the aerobic system. Despite primary dissipation, toxicity testing using degradants indicates the parent S enantiomer does not significantly degrade to non toxic degradants within 28 days. This means that Indoxacarb (enantiomeric reaction mass 75:25 S:R) cannot be considered to undergo significant mineralisation within 28 days and is not considered readily biodegradable for the purpose of classification and labelling.

As DT_{50} values for S and R enantiomers within each system, are not considered to be significantly different, then similar DT_{50} values to this study using Indoxacarb (enantiomeric reaction mass 75:25 S:R) are anticipated for Indoxacarb.

Study 2

A second anaerobic aquatic degradation study [17] is available using flooded soil and ^{14}C Indoxacarb (racemic mixture 50:50 S:R). This method is considered less relevant for the purpose of classification and labelling than the above study. However, it is noted that the calculated DT_{50} in water was 8 days at 20°C, which is broadly similar to results above. In addition, the S and R enantiomers were observed to degrade at equal rates under test conditions which support read-across of Indoxacarb (enantiomeric reaction mass 75:25 S:R) DT_{50} results to Indoxacarb.

4.1.3 Summary and discussion of persistence

Aquatic degradation (aqueous photolysis, hydrolysis and simulation) studies have been undertaken with Indoxacarb (enantiomeric reaction mass 75:25 S:R) and a ready biodegradation study has been undertaken with Indoxacarb (racemic mixture 50:50 S:R). As degradation rates for mixtures with S and R enantiomers broadly showed similar rates for each S and R enantiomer, the results are considered appropriate for read-across to mixtures of S:R enantiomers and the pure S enantiomer form Indoxacarb.

On the basis of a MITI (I) ready biodegradation study, the substances are not considered readily biodegradable as only 2-4% degradation was achieved.

There is evidence the substances may undergo degradation through aqueous photolysis or hydrolysis in the environment. However, on the basis of these processes, the parent is not considered to rapidly degrade, in terms of 70% degradation to non-toxic substances within 28 days.

Similarly, while the substance may quickly be adsorbed to sediment and undergo aerobic and anaerobic degradation, degradation products are more persistent. This means complete mineralisation for 70% of the parent within 28 days was not achieved. Toxicity data are available which show that the degradant IN-JT333 is more acutely toxic than either the parent S enantiomer or the S:R enantiomer mixture. IN-JT333 is anticipated to account for up to a maximum of ~25% of degradation products and has a longer DT_{50} in aerobic (19 days at 20°C) and anaerobic (>1 year at 20°C) sediments. Therefore, although primary degradation occurs in water and sediment fairly rapidly, dissipation of more toxic degradants does not occur rapidly. For the purpose of classification, the S and R enantiomers are not considered readily biodegradable.

Overall, based on the available data, Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R) are not considered to undergo significant rapid degradation to non-toxic substances. This means they are considered not readily biodegradable for the purpose of classification and labelling.

4.2 Environmental distribution

4.2.1 Adsorption / desorption

Following US EPA guidelines, an adsorption / desorption screening test [18] was performed using ^{14}C labelled Indoxacarb (racemic mixture 50:50 S:R) and four soils including sand and various matrix loams. The K_{oc} adsorption constant range was 2,500 to 9,600 l/kg. The average K_{oc} adsorption constant was 5,125 l/kg. Due to the instability of the test substance, desorption screening was not undertaken.

Adsorption coefficients were also determined for a major degradant (IN-JT333) using the same soils. The K_{oc} adsorption constant range was 8,200 to 25,000 l/kg. The average K_{oc} adsorption constant was 17,300 l/kg.

Given the S and R enantiomers structural formulas, both enantiomers are anticipated to exhibit similar adsorption properties. This means the values derived from the above study are considered representative for Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R). This is supported by similar log K_{ow} values for Indoxacarb and Indoxacarb (enantiomeric reaction mass 50:05 S:R) and low water solubility values for Indoxacarb, Indoxacarb (racemic mixture 50:50 S:R) and Indoxacarb (enantiomeric reaction mass 75:25 S:R).

The results of the study indicate that both the S and R enantiomers are likely to adsorb to various solid matrices and result in low potential mobility.

4.2.2 Volatilisation

Following OECD Guideline 104, the vapour pressure of Indoxacarb [(2)] is 9.8×10^{-9} Pa at 20°C and 2.5×10^{-8} at 25°C. The calculated Henry's Law Constant is 6.0×10^{-5} $Pa.m^3.mol^{-1}$ at 25 °C) based on measured data. Following OECD Guideline 104, the vapour pressure [(8)] of Indoxacarb (racemic mixture 50:50 S:R) is 1.3×10^{-10} Pa at 20°C and 4.0×10^{-10} at 25°C.

The presence of the R enantiomer may result in a slight decrease in vapour pressure although the difference could be due to experimental variability given the low values. A measured vapour pressure value is not available for Indoxacarb (enantiomeric reaction mass 75:25 S:R). Given the mixture of S:R enantiomers, the vapour pressure for Indoxacarb (enantiomeric reaction mass 75:25 S:R) is assumed to lie within the range for Indoxacarb and Indoxacarb (racemic mixture 50:50 S:R). On this basis both Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R) are considered unlikely to partition the air .

4.2.3 Distribution modelling

Not relevant to this type of dossier.

4.3 Bioaccumulation

4.3.1 Aquatic bioaccumulation

4.3.1.1 Bioaccumulation estimation

Using the Technical Guidance Document (TGD, 2003) [(19)] QSAR equation 74, a BCF_{fish} of 1,788 can be estimated based on a measured log K_{ow} 4.65 for the S enantiomer [(1)]. This log K_{ow} value is within the domain of the QSAR (log K_{ow} 2-6) and considered appropriate for the S and S:R enantiomer mixtures as the measured [(11)] log K_{ow} for Indoxacarb (racemic mixture 50:50 S:R) was 4.6 . However, measured BCF_{fish} data are available and are preferred.

4.3.1.2 Measured bioaccumulation data

Following OECD Guideline 305, steady state fish bioconcentration factors were determined for Indoxacarb (DPX-KN128, S enantiomer), the R enantiomer(referred to as IN-KN 127) and Indoxacarb (racemic mixture 50:50 S:R) [(20)]. The study used flow-through conditions, ^{14}C labelled Indoxacarb (racemic mixture 50:50 S:R) at 0.1 and 0.01 mgl^{-1}, and a 28 day uptake phase followed by a 21 day depuration phase. The following average fish bioconcentration factors (BCF_{fish}) were determined:
Indoxacarb (racemic mixture 50:50 S:R) = 950.3
Indoxacarb (S enantiomer) = 77.3
IN-KN127 (R enantiomer) = 1,848

It is possible that the difference in BCF_{fish} rates between S and R enantiomers is not a result of different uptake mechanisms and instead reflects metabolism of the insecticidally active S enantiomer (Indoxacarb) to IN-JT333 (N-decarbomethoxylated Indoxacarb).

During the depuration phase, Indoxacarb (racemic mixture 50:50 S:R) residues were observed to depurate rapidly with 90% removal by day 21.

The ratio of S:R enantiomers in water remained broadly 1:1 over the study period. However, the ratio of S:R enantiomers in fish tissue altered over the study period to approximately 1:19 based on measurement on days 21 and 28.

One major metabolite (IN-JT333) was observed representing ~22-28% total radioactive residues (TRR) in fish fillets and ~23-30% TRR in fish viscera. Four additional metabolites at <0.05 ppm were also observed.

The study shows that the S and R enantiomers bioaccumulate at differing rates. Whilst the S enantiomer has a relatively low bioaccumulation potential (BCF_{fish} 77.3), the R enantiomer has a considerably higher bioaccumulation potential (BCF_{fish} 1,848).

For the purpose of classification and labelling, based on a BCF_{fish} of < 100 for the S enantiomer, Indoxacarb is not considered bioaccumulative.

The BCF_{fish} value of 950.3 for the 50:50 S:R racemic mixture is influenced by the presence of the R enantiomer. On this basis, the BCF_{fish} for Indoxacarb (enantiomeric reaction mass 75:25 S:R) is anticipated to reflect the presence of the R enantiomer falling between 77.3 and 1,848. As the R enantiomer is a significant component of the Indoxacarb enantiomeric 75:25 S:R mixture and the R enantiomer BCF_{fish} is considered bioaccumulative (i.e. > 100 and > 500 under CLP), the 75:25 S:R mixture is considered bioaccumulative for the purpose of classification and labelling.

4.3.2 Terrestrial bioaccumulation

Not relevant for this type of dossier.

4.3.3 Summary and discussion of bioaccumulation

Measured log K_{ow} values of 4.65 for the S enantiomer and log K_{ow} 4.6 for a 50:50 mixture of S:R enantiomers indicate potential for bioaccumulation.

On the basis of measured BCF_{fish} data, the S and R enantiomers are considered to bioaccumulate at different rates. The S enantiomer is considered to have a relatively low bioaccumulation potential (BCF_{fish} 77.3). The R enantiomer is considered to have a higher bioaccumulation potential (BCF_{fish} 1,848). Given the presence of the R enantiomer in Indoxacarb (enantiomeric reaction mass 75:25 S:R), the mixture is considered to have a BCF_{fish} >100.

Rapid depuration of Indoxacarb (racemic mixture 50:50 S:R) residues was observed with 90% removal by day 21.

4.4 Secondary poisoning

Not relevant for this type of dossier.

5 HUMAN HEALTH HAZARD ASSESSMENT

The summaries included in this proposal are based on the Competent Authority Report (CAR) prepared by the UK in the context of the possible inclusion of Indoxacarb in Annex I of Council Directive 98/8/EC (CAR May 2008). Some details of the summaries were not included when considered not relevant for a decision on the classification and labelling of these substances. In addition, the Draft Assessment Report and Proposed Decision of the Netherlands prepared in the context of the possible inclusion of Indoxacarb in Annex I of Council Directive 91/414/EEC (DAR September 2000 + addendum January 2005 + post inclusion addendum 1 October 2007, RMS The Netherlands) was consulted. For more details the reader is referred to the CAR and DAR and its addenda (Section 13 of the IUCLID). All references to individual studies should be seen as references to the CAR and DAR and its addenda.

5.1 Toxicokinetics (absorption, metabolism, distribution and elimination)

The toxicokinetics of Indoxacarb (pure S enantiomer) have not been studied. However, radiolabelled Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) have been investigated.

Overall, it is predicted that Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R) will be well absorbed via the oral route (> 80% in rats at low doses – 5mg/kg), but dermal absorption is thought to be limited (0.88% in rats). However, in the dermal 28-day study of MacKenzie (1999) hematotoxicity was observed at doses of ≥ 50 mg/kg. Assuming that dermal absorption is below 1% would indicate that systemic toxicity after repeated dermal exposure would appear at very low internal doses of 0.5 mg/kg. LOAEL for haematotoxicity was seen in rat 90 day oral studies in the range of 1-5 mg/kg/day. For equivalency to a 28-day study design (as it was tested in the MacKenzie dermal study) equivalent LOAEL for haematotoxic effects in a 28-day study should expected to be somewhat higher. (Although dose-response relationship is unlikely to be linear taking the findings from the chronic studies into account.) If theoretically assuming the oral LOAEL for haematotoxic effects in 28-day studies will be about 5 mg/kg/day and taking 100% absorption for the oral route, then the haematotoxic effects considered as LOAEL effects were roughly comparable among 5 mg/kg/day (internal dose oral) to 0.5 mg/kg/day (internal dose dermal). Expected that haematotoxic effects would start to occur at the same internal dose, this comparison would question whether dermal absorption was underestimated and absorptions rates up to 10% may be more likely.

There are no data on absorption following inhalation, but there is evidence of systemic availability from acute inhalation studies on Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) showing that these substances are absorbed via this route. Once absorbed Indoxacarb (enantiomeric reaction mass 75:25 S:R) is extensively distributed with the potential for key metabolites to persist in fat and to bind to the red blood cells. Females were found to retain more in tissue than males. The greater retention of radioactivity in the erythrocytes might be due in part to the association of specific metabolites with red blood cells. Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) were extensively metabolised with gender and stereo specific biotransformation. The systemic biotransformation favours the insecticidally active S enantiomer and the metabolic pathway involves hydroxylation, ring opening and glucuronide and sulphate conjugation. The most important metabolite in females was IN-JT333 (N- decarbomethoxylated Indoxacarb) formed from the hydrolysis of the carboxymethyl group. Characterisation of this metabolite showed that the enantiomer derived from the insecticidally active S isomer was at a higher concentration than that of the enantiomer derived from the insecticidally inactive R isomer. The ratio of enantiomers in fat

was approximately 6:1 in samples from rats exposed to Indoxacarb (enantiomeric reaction mass 75:25 S:R), and 2:1 for exposure to the racemic compound Indoxacarb (racemic mixture 50:50 S:R). This suggests that there is greater variation in the metabolism and/or distribution of the two isomers with the insecticidally active S enantiomer being the preferred substrate in the removal of the N-methoxycarbonyl group to form IN-JT333. The production of IN-JT333 is greater in females compared to males. Indoxacarb (enantiomeric reaction mass 75:25 S:R) and/or its metabolites are slowly eliminated primarily in urine (metabolites only) and faeces (parent compound and metabolites) with no sex differences. Given the partitioning of the radiolabelled IN-JT333 in fat (as compared to other tissues) following administration and the low Log P_{ow} of Indoxacarb which indicates that it is lipophilic, it is predicted that the metabolites would likely be eliminated in breast milk.

5.2 Acute toxicity

5.2.1 Acute toxicity: oral

The summary of the results for acute oral toxicity is presented in Table 5.1.

Table 5.1: Summary of acute oral toxicity data

Test material	Method/ Guideline	Species/strain/ sex/ No per group	Dose levels/Duration of exposure	Results LD_{50}/LC_{50}	Reference
Indoxacarb Purity: 99.7%	OECD 401	Rat, CD (SD)BR/ m+f/ 5/sex/grp	M: 400, 640, 1000, 1953 mg/kg bw F: 123, 192, 300, 400 mg/kg bw 14d post-exposure period	M: LD_{50} of 843 mg/kg bw F: LD_{50} of 179 mg/kg bw (Males 1/5, 4/5, 2/5, 3/5; Females 1/5,3/5,4/5, 5/5)	Kern T.G (1997a)
Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 94.5%	OECD 401	Rat, CD (SD)BR/ m+f/ 5/sex/grp except the two lowest groups which contained 10 females	M: 1000, 3000, 5000 mg/kg bw F: 100, 250, 1000, 3000, 5000 mg/kg bw 24d post-exposure period	M: LD_{50} = 1730 mg/kg bw F: LD_{50} = 268 mg/kg bw (Males 0/5, 5/5, 5/5; Females 0/10, 7/10, 4/5, 5/5)	Sarver J.W (1996a)
Indoxacarb (enantiomeric reaction mass 75:25 S:R) – MUP Purity: 67.8% DPX-MP062	OECD 401	Rat, CD (SD)BR/ m+f/ 5/sex/grp	M: 750, 1268, 2142 mg/kg bw F: 263, 444, 750 mg/kg bw 14d post-exposure period	M: LD_{50} = 1070 mg/kg bw F: LD_{50} = 407 mg/kg bw	Kern T.G (1997c)

The study conducted on Indoxacarb (pure S enantiomer) showed it is of moderate toxicity to rats following single oral gavage, with females appearing to be more sensitive than males (LD_{50} of 179 and 843 mg/kg in females and males respectively). At a dose level of 400 mg/kg bw, 20% mortality was observed in males compared with 100% in females. Clinical signs observed in both sexes include discharge, staining and matting of fur, abnormal excretion, hypoactivity, laboured

respiration, impaired righting reflex, tremors and hair loss. Gross findings including gastrointestinal abnormalities were noted in nine rats found dead and pulmonary and external findings were reported in the three surviving rats.
The difference in sensitivity between the sexes (based on lower LD_{50} value) was also observed in the oral studies with Indoxacarb (enantiomeric reaction mass 75:25 S:R) technical (LD_{50} values of 1730 and 268 mg/kg in males and females, respectively) and Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP (LD_{50} values of 1070 and 407 mg/kg in males and females, respectively). There is no specific information on why the female rat is more susceptible than males in these studies. However, it might be due in part to the toxicokinetic differences; greater tissue distribution and retention were observed in females, and the major metabolite IN-JT333 (N-decarbomethoxylated Indoxacarb (pure S enantiomer)) has been shown to be highly toxic via the oral route following single exposure (LD50 values of 52 mg/kg and 39 mg/kg in male and female CD (SD)BR rats respectively, Sarver JW 1996b).

In addition, the neurotoxic potential of Indoxacarb (enantiomeric reaction mass 75:25 S:R) was evaluated in a single dose oral gavage study in rats. A summary of the results for the oral neurotoxicity study is presented in Table 5.2.

Table 5.2: Summary of acute oral neurotoxic toxicity data

Test material	Method/ guideline	Species/ Strain/sex / no. per group	Exposure period	Dose levels	Results	NOAEL/ LOAEL	Reference
Indoxacarb (enantiomeric reaction mass 75:25 S:R)Purity: 94.5%	US-EPA 81-8 A neurobehavioral test battery, consisting of motor activity and functional observational battery assessments, was conducted on all study rats 1 week before and 2-4 hours, 8 days, and 15 days after dosing.	Rat, Crl:CDBR/ m+f/12/sex /group	Single dose	M: 0, 25, 100 or 200 mg/kg bw F: 0, 12.5, 50 or 100 mg/kg bw	One female died at 100 mg/kg bw. Reduced forelimb grip strength and foot splay in males at 200 mg/kg bw on Day 15. ↓ motor activity in females at 100 mg/kg bw on Day 1 and 8. ↓ body weights at ≥ 50 mg/kg bw in females (males at 200 mg/kg bw) Pallor and alopecia were observed in most/all females at 100 mg/kg bw. Abnormal gait, hunched over position, hyperactivity was noted in one/few females at 100 mg/kg.	NOAEL (general toxicity): 12.5 mg/kg bw NOAEL (neurotoxicity): 50 mg/kg bw LOAEL: 50 and 100 mg/kg bw for general and neurotoxicity, respectively	Christoph, 1997 (IUCLID 5.8.3/01)

The assessments included functional observational battery, motor activity measurements and neuropathological examinations of nerve tissues and muscle. Significant reductions in body weights and food consumption of the animals were commonly observed.

In the acute gavage study, 1 female died at 100 mg/kg bw. In addition, significant reduction in forelimb grip strength and decreased foot splay were observed in males dosed with 200 mg/kg/d. The females had significantly decreased motor activity at 100 mg/kg/d. No detailed information on incidences and severity grades are reported in the available study summaries. No neurotoxicity effects were observed at 50 mg/kg/d in both sexes.

5.2.2 Acute toxicity: inhalation

The summary of the results for acute inhalation toxicity is presented in Table 5.3.

Table 5.3: Summary of acute inhalation toxicity data

Test material	Method/ Guideline	Species/strain/ sex/ No per group	Dose levels/Duration of exposure	Results LD_{50}/LC_{50}	Reference
Indoxacarb (enantiomeric reaction mass 75:25 S:R) MUP Purity: 70.7% DPX-MP062 Aerosol particlesize: <10µm (MMAD mean: 2.8µm)	OECD 403	Rat, CD (SD)BR/ m+f/ 5/sex/grp	5.5 mg/L (m+f) 4-hour, nose only exposure 14d post-exposure period	LC_{50} > 5.5 mg/L(m+f)	Scott R.S (1997)
Indoxacarb (racemic mixture 50:50 S:R) Purity 94.76% Aerosol particle size: <10µm (MMAD: 1.7 - 3.2 µm)	OECD 403	Rat, CD (SD)BR/ m+f/ 2 grps of 5 males; 3 groups with 10 females and 2 grps containing 5 females each	M: 3.3, 5.4 mg/L F: 0.45, 2.3, 3.3, 4.0, 5.4 mg/L 4-hour, nose only exposure 14d post-exposure period	M: LC_{50} > 5.4 mg/L F: LC_{50} = 4.2 mg/L	O'Neill A.J (1995)

Low to moderate acute toxicity was observed following single inhalation (nose-only) exposure of rats to aerosols of Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP (LC_{50} value > 5.5 mg/L, males and females) and Indoxacarb (racemic mixture 50:50 S:R) (LC_{50} values > 5.4 mg/L and 4.2 mg/L for males and females, respectively). Exposure to 5.5 mg/L Indoxacarb (enantiomeric reaction mass 75:25 S:R) (the only concentration tested) resulted in 20% mortality in females but no mortality in males. With Indoxacarb (racemic mixture 50:50 S:R) at concentrations of 2.3, 3.3 4.0, and 5.4 mg/L, 40% of exposed female rats died at 4.0 mg/L, while 60% mortality in females was observed at the highest dose level (5.4 mg/L). No males died during either study. There were no gross or pathological findings with either substance. Clinical signs such as nasal/ocular discharge and stained/wet perineum were commonly seen in both sexes. In females, additional clinical symptoms observed include abnormal gait/mobility, lethargy and hunched posture.

5.2.3 Acute toxicity: dermal

The summary of the results for acute dermal toxicity is presented in Table 5.4.

Table 5.4: Summary of acute dermal toxicity data

Test material	Method/ Guideline	Species/strain/ sex/ No per group	Dose levels/Duration of exposure	Results LD_{50}/LC_{50}	Reference
Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity:94.5%	OECD 402	Rat, CD (SD)BR/ m+f/ 5/sex/grp	5000 mg/kg bw 14d post-exposure period	LD_{50} > 5000 mg/kg bw (m+f)	Sarver J.W (1996c)

Indoxacarb (enantiomeric reaction mass 75:25 S:R) is of low dermal toxicity, with no evidence of systemic toxicity and no mortalities in rats at the limit dose of 5000 mg/kg. No differences between the sexes were observed.

5.2.4 Acute toxicity: other routes

No data available.

5.2.5 Summary and discussion of acute toxicity

Indoxacarb (pure S enantiomer)
Acute oral studies with Indoxacarb (pure S enantiomer) result in an LD_{50} of 179 mg/kg bw for female rats. Males were less susceptible (LD_{50} of 843 mg/kg bw), probably due to differences in toxicokinetics. Based on the low LD_{50} in female rats, according to Directive 67/548/EEC, classification with T; R25 (range 25 – 200 mg/kg bw) is appropriate and Acute Tox Cat 3; H301 according to Regulation EC 1272/2008 (range 50 – 300 mg/kg bw).

No data were available for acute inhalation exposure to Indoxacarb (pure S enantiomer). In an acute inhalation study with Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP in rats, the LD_{50} was above 5.5 mg/L, the highest dose applied. However, the concentration of Indoxacarb (enantiomeric reaction mass 75:25 S:R) in Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP is only 70% and the concentration of the S enantiomer only 52.5% (70% * 0.75). Single inhalation exposure of Indoxacarb (racemic mixture 50:50 S:R) to rats resulted in an LC_{50} above 5.4 mg/L in males and in an LC_{50} of 4.2 mg/L in females. The results with Indoxacarb (racemic mixture 50:50 S:R), are considered also relevant for Indoxacarb (pure S enantiomer) because the acute oral studies indicate that the acute toxicity of the S-enantiomer might be slightly higher compared to the R-enantiomer. Therefore, classification with Xn; R20 (harmful by inhalation) is appropriate according to Directive 67/548/EEC (range 1 – 5 mg/L) and Acute Tox Cat 4; H332 according to Regulation EC 1272/2008 (range 1 – 5 mg/L).

No data were available for acute dermal exposure to Indoxacarb (pure S enantiomer). In an acute dermal toxicity study in rats, application of Indoxacarb (enantiomeric reaction mass 75:25 S:R) at a dose of 5000 mg/kg bw did not induce mortality or evidence of systemic toxicity. These values are above the classification limits according to Directive 67/548/EEC (2000 mg/kg bw) and Regulation EC 1272/2008 (2000 mg/kg bw). The results with Indoxacarb (enantiomeric reaction mass 75:25 S:R) are considered also relevant for Indoxacarb (pure S enantiomer) even though the acute oral studies indicate that the acute toxicity of the S-enantiomer might be slightly higher compared to the R-enantiomer.a comparable acute toxicity between the two enantiomers or a somewhat higher toxicity of the S enantiomer.

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
The acute oral toxicity study with Indoxacarb (enantiomeric reaction mass 75:25 S:R) resulted in an LD_{50} of 268 mg/kg bw for female rats. The slightly higher LD_{50} than observed for Indoxacarb (pure S enantiomer) may be explained by the lower concentration of the S enantiomer in Indoxacarb (enantiomeric reaction mass 75:25 S:R). Based on these results classification with Xn; R22 (range 200 – 2000 mg/kg bw) is appropriate for Indoxacarb (enantiomeric reaction mass 75:25 S:R) according to Directive 67/548/EEC and Acute Tox Cat3; H301 according to Regulation EC 1272/2008 (range 50 – 300 mg/kg bw).

In an acute inhalation study with Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP in rats, the LD_{50} was above 5.5 mg/L, the highest dose applied. However, the concentration of Indoxacarb (enantiomeric reaction mass 75:25 S:R) in Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP is only 70% (52.5% S and 17.5% R). Single inhalation exposure of Indoxacarb (racemic mixture 50:50 S:R) to rats resulted in an LC_{50} above 5.4 mg/L in males and in an LC_{50} of 4.2 mg/L in females. The results with Indoxacarb (racemic mixture 50:50 S:R), are considered also relevant

for Indoxacarb (enantiomeric reaction mass 75:25 S:R), given the reduced concentration of Indoxacarb (enantiomeric reaction mass 75:25 S:R)-in the MUP and considering that the acute oral studies indicate that the acute toxicity of the S-enantiomer might be slightly higher compared to the R-enantiomer. Therefore, classification with Xn; R20 (harmful by inhalation) is appropriate according to Directive 67/548/EEC (range 1 – 5 mg/L) and Acute Tox Cat 4; H332 according to Regulation EC 1272/2008 (range 1 – 5 mg/L).

In an acute dermal toxicity study in rats, application of Indoxacarb (enantiomeric reaction mass 75:25 S:R) at a dose of 5000 mg/kg bw did not induce mortality. These values are above the classification limits according to Directive 67/548/EEC (2000 mg/kg bw) and Regulation EC 1272/2008 (2000 mg/kg bw).

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: T; R25 Xn; R20
CLP: Acute Tox. 3: H301
Acute Tox. 4: H332

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: Xn; R20/22
CLP: Acute Tox 3: H301
Acute Tox 4: H332

Summary and Discussion of Specific Target Organ Toxicity – Single Exposure (STOT-SE)

Indoxacarb (pure S enantiomer)
In the acute oral neurotoxicity study with Indoxacarb (enantiomeric reaction mass 75:25 S:R), which is considered as predictive for Indoxacarb (pure S enantiomer) 1/12 female rat died at 100 mg/kg/day (Christoph, 1997). In addition, measured values for motor activity were significantly reduced in females at 100 mg/kg bw and significantly reduced forelimb grip strength and foot splay were estimated in males at 200 mg/kg/day. Unfortunately, incidences and severity grades of clinical observations are not reported in the available summaries.
No effects were observed in the acute dermal or inhalation toxicity studies in rats with Indoxacarb (enantiomeric reaction mass 75:25 S:R) that would warrant classification with STOT-SE.

Acute neurofunctional studies in mice were not conducted. In repeated dose studies with Indoxacarb (racemic mixture 50:50 S:R) neurotoxicity was observed in mice at doses of ≥100 ppm (13.8 mg/kg, 18 months), 150 ppm (23 mg/kg bw/d; 90 day study) and 235 ppm (17.9 mg/kg bw/d; 28 day-study). In rats, no neurotoxicity was observed with after 90 day-administration of Indoxacarb (pure S enantiomer) up to test doses of 100 ppm (8.5 mg/kg bw/d), Indoxacarb (racemic mixture 50:50 S:R) up to test doses of 15.2 mg/kg and with Indoxacarb (enantiomeric reaction mass 75:25 S:R) up to test doses of 100 ppm (15 mg/kg bw/d).

Neurodysfunctions observed in the repeated dose studies on mice may indicate that this species could be more sensitive for these effects than rats. Clinical signs indicative for neurodysfunction were seen in repeated dose studies on Indoxacarb (racemic mixture 50:50 S:R) in mice at significant lower doses than rats received in the acute neurotoxicity study. No mouse studies were available on other Indoxacarb test compounds and no acute toxicity studies were available for the mouse. No indication of neuronal dysfunction was observed in repeated dose studies on rats receiving Indoxacarb (racemic mixture 50:50 S:R), Indoxacarb enantiomeric reaction mass 75:25 S:R) or Indoxacarb (pure S enantiomer). However the highest test doses for repeated administration were clearly below the doses that acutely affected the nervous system. The absence of findings in rats could be related to the facts that comparative dosages for female rats in repeated dose studies were about 1/10 of the acute neurotoxicity study and that not all studies included specific neurofunctional testing.

The observation that clinical signs indicating neurodysfunction were reported for females only is in line with the higher sensitivity of female rats in the acute oral toxicity studies. A likely interpretation of similar clinical signs such as hypoactivity, impaired righting reflex and tremors, which were reported in the acute toxicity study for rats receiving Indoxacarb (pure S enantiomer) could be that these effects indicate severe moribundity and are mortality-related. However, observations in repeated dose studies in mice demonstrate that neurodysfunctional disorders could appear as specific toxic effects, which are not exclusively related to premortal conditions.

According to the criteria for STOT-SE the clinical observations in the functional tests of the acute neurotoxicity study in rats indicate a significant toxic effect. Significantly reduced motor activity and reduced forelimp grip strength are clearly indicative of neurofunctional disorders of the nervous system. While the quality of the observed effects were in prinicipal in accordance with the criteria for STOT-SE, the lack of information on incidences and severity grades are serious limits of the study documentation. Substances should be classified if observations in experimental animals were significant and/or severe toxic effects of relevance to human health. “Significant” toxic effects are defined as “changes which clearly indicate functional disturbance or morphological changes which are toxicologically relevant”.

In conclusion, there is concern on a significant toxic effect after single exposure to Indoxacarb (pure S enantiomer) based on an acute neurotoxicity study on Indoxacarb (enantiomeric reaction mass 75-25). Effect doses (100 and 200 mg/kg for Indoxacarb enantiomeric reaction mass 75:25 S:R and 75-150 mg/kg when corrected for Indoxacarb (pure S enantiomer)) were below the guidance value for Category 1 (≤300 mg/kg).
While this necessary criteria is fulfilled, the CLP guidance recommends not to assign for STOT-SE if lethality would occur at relevant doses and classification for acute toxicity should preferentially be proposed.
Although no precise guidance is given, RAC proposes to consider effect doses for acute neurotoxicity being in the range of relevant dose for acute toxicity. LD_{50} 268 mg/kg bw for Indoxacarb (enantiomeric reaction mass 75:25 S:R) is less than 3-fold above the $LOAEL_{neurotoxicity}$ 100 mg/kg bw from the study of Christoph (1997). In this study mortality of one out of 12 females was observed. Although uncertainty remains on the treatment-relationsship of this death due to this low incidence that could occasionally happen by chance (No deaths were seen in the acute toxicity study at this dose (Sarver, 1996a) this could also be understood as if 100 mg/kg is a relevant dose for lethality.
100 mg/kg bw Indoxacarb (enantiomeric reaction mass 75:25 S:R) corresponds to 75 mg/kg bw Indoxacarb S, which also is considered to reflect the relevant acute toxic dose (LD_{50} 179 mg/kg bw, LOAEL mortality (1/5) at 123 mg/kg bw (Kern, 1997a)
Although no detailed information on incidences (and severity grades) on clinical observations is given, the fact that acute neurotoxicity was observed at relevant doses which could also induce lethality, classification for acute toxic effects is proposed and neurotoxicity observed in repeated dose studies is separately covered by STOT-RE, data are not considered to justify a classification for STOT-SE.

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
The discussion mentioned above argumentation is also valid for Indoxacarb (enantiomeric reaction mass 75:25 S:R).

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: No classification proposed
CLP: No classification proposed

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: No classification proposed
CLP: No classification proposed

5.3 Irritation

5.3.1 Skin

The summary of the results for skin irritation is presented in Table 5.5.

Table 5.5: Summary of skin irritation data

Method/Guideline/	Species/No per	Average score	**Reversibility**	**Result**	**Reference**

Test Material	group	Erythema	Oedema	(Y/N)		
OECD 404 Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity:94.5%	Rabbit, 6m	0	0	-	Non-irritating	Sarver J.W (1997a)

The skin irritation potential of Indoxacarb (enantiomeric reaction mass 75:25 S:R) (moistened with water) was investigated in a GLP and guideline compliant study in rabbits, no irritant effects were observed.

Local effects on the skin (erythema, desquamation, focal and non-focal eschar) were seen in the 28-day dermal study in female rats following semi-occlusive application of Indoxacarb (enantiomeric reaction mass 75:25 S:R) at dose levels of 1000 mg/kg/d and above, and in males at the top dose of 2000 mg/kg/d. Time of onset of these effects is not indicated However, no such effects were seen in a comparable 28-day study with the same strain of rat at dose levels up to 2000 mg/kg/d.

5.3.2 Eye

The summary of the results for eye irritation is presented in Table 5.6.

Table 5.6: Summary of eye irritation data

Method/Guideline/ Test material	**Species/ No per group**	**Average score (score at 24, 48 and 72h)**				**Reversibility (Y/N)**	**Result**	**Reference**
				conjunctiva				
		cornea	**iris**	**chemosis**	**redness**			
OECD 405 Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 94.5%	Rabbit, 6f	0.11 (0.33, 0 and 0)	0.06 (0.17, 0 and 0)	0.44 (1.0, 0.33 and 0)	0.72 (1.5, 0.67 and 0)	All effects were reversible within 72 hours post-application	Not classified	Sarver J.W (1997b)
OECD 405 Indoxacarb (enantiomeric reaction mass 75:25 S:R) MUP Purity: 67.8% MP062	Rabbit, 4m + 2f	0	0	0.61 (1.0, 0.5 and 0.33)	1.33 (1.67, 1.33 and 0)	All effects were reversible within 14 days post-application	Not Classified	Kern T.G (1997d)

The eye irritation potential of Indoxacarb (enantiomeric reaction mass 75:25 S:R) (applied as a solid) and Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP (applied in corn oil) has been investigated in standard studies in the rabbit. From the available information it can be concluded that Indoxacarb (enantiomeric reaction mass 75:25 S:R) is slightly irritating to the eyes but does not need to be classified for this effect.

5.3.3 Respiratory tract

There is no specific information regarding the respiratory tract irritation potential of Indoxacarb (pure S enantiomer), but in an acute inhalation study with Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP no evidence of irritating effects in the respiratory tract was observed; however, nasal discharge was seen with Indoxacarb (racemic mixture 50:50 S:R). Nevertheless, it is predicted that Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R) are unlikely to cause respiratory tract irritation.

5.3.4 Summary and discussion of irritation

Indoxacarb (pure S enantiomer)
For Indoxacarb (pure S enantiomer), no data on irritation potential are available. However, because irritation depends on reactivity and since the reactivity of both enantiomers is probably similar, read-across from Indoxacarb (enantiomeric reaction mass 75:25 S:R) to Indoxacarb (pure S enantiomer) is justified. Although in a repeated dermal study (28 days) with Indoxacarb (enantiomeric reaction mass 75:25 S:R) local effects on the skin were observed at doses ≥ 1000 mg/kg bw, such effects were not seen in a comparable 28-day study with the same strain of rat at dose levels up to 2000 mg/kg/d. These effects are considered to indicate sensitization rather than irritation. As Indoxacarb (enantiomeric reaction mass 75:25 S:R) (see below) is not irritating it can be expected that Indoxacarb (pure S enantiomer) is also not irritating.

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Indoxacarb (enantiomeric reaction mass 75:25 S:R) is not irritating to skin and only slightly irritating to the eyes with all effects reversible within 72 hours. In addition, in an acute inhalation study with Indoxacarb (enantiomeric reaction mass 75:25 S:R)-MUP no evidence of irritating effect in the respiratory tract was observed. It can thus be concluded that Indoxacarb (enantiomeric reaction mass 75:25 S:R) does not need to be classified for irritation.

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

5.4 Corrosivity

No corrosive effects were observed in the skin irritation study. Indoxacarb and Indoxacarb (enantiomeric reaction mass 75:25 S:R) therefore do not meet the criteria for classification as corrosive.

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

5.5 Sensitisation

5.5.1 Skin

A summary of the skin sensitisation data is presented in Table 5.7.

Table 5.7: Summary of skin sensitisation data

Method/Guideline	Species	Number of animals sensitised/Total number of animals	Result	Reference
Indoxacarb (enantiomeric reaction mass 75:25 S:R) (94.5%) OECD 406, MAGNUSSON-KLIGMAN Maximisation Test	Guinea pig	100% (w/v moistened with water: 13/20 vs 0/20 in controls (24 and 48 h after challenge) 33.3% (w/v) in propylene glycol: 7/20 vs 0/20 in controls (24 and 48 h after challenge)	Positive; sensitising Positive; sensitising	Moore G.E (1996)

A positive response in more than 30% of animals was obtained in a well-reported guinea pig maximisation test with Indoxacarb (enantiomeric reaction mass 75:25 S:R). Test animals were induced with intradermal injections of 5% Indoxacarb (enantiomeric reaction mass 75:25 S:R) in propylene glycol followed by topical application of 100% Indoxacarb (enantiomeric reaction mass 75:25 S:R) moistened with propylene glycol. Positive and negative controls produced appropriate responses.

Local effects on the skin (erythema, desquamation, focal and non-focal eschar) were seen in the 28-day dermal study in female rats following semi-occlusive application of Indoxacarb (enantiomeric reaction mass 75:25 S:R) at dose levels of 1000 mg/kg/d and above, and in males at the top dose of 2000 mg/kg/d. Time of onset of these effects is not indicated However, no such effects were seen in a comparable 28-day study with the same strain of rat at dose levels up to 2000 mg/kg/d.

5.5.2 Respiratory system

No information is available with regard to the respiratory sensitisation potential of Indoxacarb (pure S enantiomer) and Indoxacarb (enantiomeric reaction mass 75:25 S:R). Both forms of Indoxacarb have no structural relationships with known respiratory sensitisers. Therefore, there is no data available showing or indicating a potential for respiratory sensitisation.

5.5.3 Summary and discussion of sensitisation

Indoxacarb (pure S enantiomer)

The skin sensitising potential of Indoxacarb (pure S enantiomer) was not determined. Read-across of the skin sensitising potential of Indoxacarb (enantiomeric reaction mass 75:25 S:R) to Indoxacarb (pure S enantiomer) could be considered. However, it is unknown whether the R-enantiomer, the S-enantiomer or both have caused the skin sensitising potential. Sensitisation is caused by a reaction of the molecule which is normally independent of the chiral form. However, some sensitising compounds have to be activated into a reactive compound by metabolism and metabolism is often varying between enantiomers[(20a)]. Quantitative difference in metabolism between the enantiomers is shown for Indoxacarb (enantiomeric reaction mass 75:25 S:R). SAR analyses with DEREK resulted in two alerts for skin sensitisation namely “formaldehyde donor” and “hydrazine precursor”. The formation of formaldehyde may result from the initial N-decarbomethoxylation of the substance into the metabolite JT333. This metabolite is formed from both enantiomers and is an important metabolite after oral exposure but it is unknown whether this reaction also occurs after dermal exposure. However, there is no evidence from toxicokinetic studies that formaldehyde is formed during metabolism (see chapter 5.1). Secondly, there is no evidence from in vitro genotoxicity studies that Indoxacarb (pure S enantiomer or enantiomeric reaction mass 75:25 S:R) or its

metabolites are genotoxic (see chapter 5.7.1). In the presence of formaldehyde, however, this would be expected.
Read across from a positive response with Indoxacarb (enantiomeric reaction mass 75:25 S:R) to Indoxacarb is justified as it is considered that the skin sensitisation potential is independent of the chiral form. Classification of Indoxacarb (pure S enantiomer) as a skin sensitiser (R43) is appropriate according to Directive 67/548/EEC and Skin Sens Cat 1B: H317 according to Regulation EC 1272/2008.

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
A positive response in more than 30% of animals was obtained in a guinea pig maximisation test with Indoxacarb (enantiomeric reaction mass 75:25 S:R). According to the classification into potency categories as described in the Guidance on the Application of the CLP-criteria, Indoxacarb (enantiomeric reaction mass 75:25 S:R) is a moderate skin sensitizer (≥30% responding at ≥1% induction dose, Draft 2.ATP, Regulation EC 1272/2008). Therefore, classification of Indoxacarb (enantiomeric reaction mass 75:25 S:R) as a skin sensitiser (R43) is appropriate according to Directive 67/548/EEC and Skin Sens Cat 1: H317 according to Regulation EC 1272/2008 (skin sens. Category 1B:H317 under the new criteria of the CLP regulation (2nd ATP)).

The generic concentration limit of 1% is allocated.

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: Xi; R43
CLP: Skin Sens. 1 (Skin Sens 1B*) H317

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: Xi; R43
CLP: Skin Sens.1 (Skin Sens. 1B*) H317

*** according to the new criteria of CLP Regulation (2nd ATP)**

5.6 Repeated dose toxicity

5.6.1 Repeated dose toxicity: oral

A summary of repeated dose oral toxicity data is presented in Table 5.8.

Table 5.8: Summary of repeated dose oral toxicity data

Test material	Duration Method/ Guideline	Species/ strain/ sex/ no. per group	Dose levels/ frequency of dosing	Results	NOAEL/ LOAEL	Reference
Indoxacarb Purity: 99.7%	90-day OECD guideline 408	Rat/Crl: CDBR/ m+f/ 10/sex/grp	M: 0, 8, 20, 50, 100, 200 ppm equivalent to 0, 0.56, 1.4, 3.2, 6.6 and 14 mg/kg bw/day F: 0, 3, 8, 20, 50 and 100 ppm equivalent to 0, 0.25, 0.68, 1.7, 4.1 and 8.5 mg/kg bw/day	Mortality in one male at 200 ppm not considered test related. Decrease body weight and body weight gain (32-41% in females and 16-45% in males) at the two top doses. Statistically significant decrease in circulating erythrocyte mass (RBC, Hb and Ht reduced by 5-17% at ≥ 3 ppm in females and 6-10% at ≥ 50 ppm in males). Increased incidence of methaemoglobin (≤ 4%) in females at ≥ 50 ppm.	NOAEL: 20 ppm equivalent to 1.7 mg/kg bw/day LOAEL: 50 ppm equivalent to 4.1 mg/kg bw/day, based on the ≥10% reduction in RBC, Hb, and/or Ht. See text for discussion	Malek, 1997a (IUCLID 5.4/07)

Test material	Duration Method/ Guideline	Species/ strain/ sex/ no. per group	Dose levels/ frequency of dosing	Results	NOAEL/ LOAEL	Reference
				Increase incidence of pigment deposit (haemosiderin) in liver (incid.: 0,0,0,0,1,6, sign. at 100 ppm) and spleen (incid.:0,1,2,10,10,10, sign≥ 20 ppm) in females. (No data on severity grades in the DAR). Pigment deposition was increased in livers (0,0,0,0,1,8, sign. At 100 ppm) and spleen (0,1,2,6,10,10, sign. ≥50 ppm) of males. Significantly higher incidences in splenic erythrocytic and bone marrow hyperplasia at ≥ 50 ppm in both sexes. (No data on severity grades in the DAR).		
Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 94.5%	90-day OECD guideline 408	Rat/ Crl: CD (SD) BR/ m+f/ 10 /sex/grp	M: 0, 10, 50, 100 and 200 ppm equivalent to 0, 0.62, 3.09, 6.01 and 15.0 mg/kg bw/day F: 0, 10, 25, 50 and 100 ppm, equivalent to 0, 0.76, 2.13, 3.78 and 8.94 mg/kg bw/day	Treatment related mortality in females at 100 ppm (5/10) between days 8-19. Decrease body weight gain in females at ≥ 50 ppm (38-42%), and in males at 200 ppm (46%). Anaemia seen in the high dose group in both sexes. Statistically significant decrease in circulating erythrocyte mass (RBC, Hb and Ht reduced by 6-15% at ≥ 10 ppm in females and 6-16% at ≥ 50 ppm in males). Increase MCV at ≥ 100 ppm. Higher incidences of increased pigment (haemosiderin) deposit in liver (in females ≥50 ppm), in kidneys (females at 100 ppm) and spleen (in females at ≥10 ppm, males ≥100 ppm). (No data on severity grades in the DAR).	NOAEL: 25 ppm equivalent to 2.13 mg/kg bw/day LOAEL: 50 ppm equivalent to 3.78 mg/kg bw/day based on a ≥10% reduction in RBC, Hb, and/or Ht. See text for discussion	MacKenzie, 1997 (IUCLID 5.4/05)
Indoxacarb (racemic mixture 50:50 S:R) Purity: 94.7%	90-day OECD guideline 408	Rat/ Crl: CD BR/ m+f/ 10 /sex/grp	M: 0, 30, 60, 125 and 250 ppm equivalent to 0, 1.9, 3.9, 8.0 and 16.0 mg/kg bw/day F: 0, 15, 30, 60 and 125 ppm equivalent to 0, 0.99, 2.3, 4.6 and 9.5 mg/kg bw/day	No clinical signs of toxicity observed. Decrease body weight gain in 125 ppm females (35%), and in males at 250 ppm (16%). Decrease food consumption in females at 125 ppm (19%), and in males at 250 ppm (8.7%). Statistically significant decrease in circulating erythrocyte mass (RBC, Hb and Ht reduced by 7-15% at ≥ 30 ppm in females and 7-16% at ≥ 60 ppm in males). Decrease total protein and globulin concentrations in females at 125 ppm. Increase MCV and reticulocytes count at the high dose in both sexes. Increase incidence of splenic erythrocytic hyperplasia (mostly stat. significant) at ≥ 60 ppm (m: 0/10, 0/10, 4/10, 10/10, 10/10 at 0, 30, 60, 125 and 250 ppm; f: 0/10, 0/10, 1/10, 3/10 and 4/10 at 0, 15, 30, 60 and 125 ppm). Increase pigment	NOAEL: 30 ppm equivalent to 2.3 mg/kg bw/day LOAEL: 60 ppm equivalent to 3.9 mg/kg bw/day See text for discussion	Malek, 1997b (IUCLID 5.4/10)

Test material	Duration Method/ Guideline	Species/ strain/ sex/ no. per group	Dose levels/ frequency of dosing	Results	NOAEL/ LOAEL	Reference
				(haemosiderin) deposit in liver and spleen at ≥ 60 ppm		
Indoxacarb (racemic mixture 50:50 S:R) Purity: 93.05%	90-day with 21 day recovery period partly similar to OECD guideline 408	Rat/ Crl: CD BR/ m+f/ 10 /sex/grp	males: 0, 30, 60, 125, 250 mg/kg food, equivalent to 0, 1.83, 3.72, 7.49, and 15.2 mg/kg bw/d females: 0, 15, 30, 60, 125 mg/kg food, equivalent to 0, 1.23, 2.45, 4.91, and 11.6 mg/kg bw/d	Significant decrease body weight gain in 60 ppm females, and in males at 250 ppm. Significant decrease food consumption in males from 125 ppm. Statistically significant decrease in circulating erythrocyte mass (RBC, Hb and Ht reduced by 7-15% at ≥ 25 ppm in females and 6-17% at ≥ 30 ppm in males)*. All effects on circulating red blood cell parameters were reversible within the 21-day recovery period No macroscopic treatment-related findings were observed.	NOAEL: 30 ppm equivalent to 2.45 mg/kg bw/day LOAEL: 60 ppm equivalent to 4.91 mg/kg bw/day	Sarver, 1998
Indoxacarb (racemic mixture 50:50 S:R) Purity: 94.7%	90-day Not in conformity to OECD guideline 408 (Clin chemistry insufficiently addressed)	Mouse/ Crl: CD-1 (ICR)BR/m +f/10/ sex/grp	0, 35, 75, 150 and 10/300 ppm equivalent to: m: 0, 5.5, 12, 23 and 1.7/44 mg/kg bw/day f: 0, 7.0, 16, 30, 2.1/51 mg/kg bw/day The dietary concentration in the 10 ppm group was increased to 300 ppm on test day 42.	Mortality in males at 300 ppm on day 85 (1/10, probably treatment-related). Clinical signs indicative of neurotoxicity (abnormal gait, leaning to one side) in females at ≥ 150 ppm and in males at 300 ppm, all animals affected at 300 mg/kg bw/day. Decrease body weight gain in 150 ppm females (45%), and at 300 ppm in both sexes. Decrease food consumption in females at ≥ 150 ppm (12-30%) and in males at 300 ppm (18%). Increase incidence of Heinz bodies (m: 0/10, 0/10, 8/8, 9/9 and f: 0/9, 0/10, 3/9, 7/9 at 0, 75, 150 and 300 ppm, respectively). Non-statistically significant decrease RBC at ≥ 150 ppm. Small spleen (lymphoid depletion) seen in females at 300 ppm. Increase pigment (haemosiderin) deposit in spleen at ≥ 75 ppm, and the liver at 300 ppm.Increased extramedullary haemopoiesis at at ≥ 150 ppm in both sexes.	NOAEL: 35 ppm equivalent to 5.5 mg/kg bw/day. LOAEL: 75 ppm equivalent to 12 mg/kg bw/day.	Malek, 1997c (IUCLID 5.4/11)
Indoxacarb (racemic mixture 50:50 S:R) Purity:95.03%	90-day OECD guideline 409	Dog/ Beagles/ m+f/ 5/sex/grp	0, 40, 80, 160, 640 ppm equivalent to: m: 0, 1, 2, 5 and 18 mg/kg bw/day f: 0, 1, 3, 5 and 17 mg/kg bw/day	Treatment-induced anaemia at ≥ 160 ppm in males. Decrease circulating erythrocyte mass (RBC, Hb & Ht) significant at ≥ 160 ppm in males and 640 ppm in females. Increase Heinz bodies at ≥160 ppm. Increase mean platelet counts, reticulocytes and MCV at ≥ 160 ppm. Increase incidence of pigment (haemosiderin) deposit in liver, spleen and kidney. Increase haematopoiesis in spleen and hyperplasia in bone marrow.	NOAEL: 80 ppm equivalent to 2 mg/kg bw/day LOAEL: 160 ppm equivalent to 5 mg/kg bw/day	Mertens, 1996 (IUCLID 5.4/ 06)
Indoxacarb (racemic mixture 50:50 S:R)	1-year OECD guideline 452	Dog/ Beagles/ m+f/ 5/sex/grp	0, 40, 80, 640, 1280 ppm equivalent to: m: 0, 1.1, 2.3,	Decrease mean body weight at 1280 ppm. Erythrocytic changes (decrease RBC, Hb	NOAEL: 40 ppm equivalent to 1.1 mg/kg bw/day	Mertens, 1997 (IUCLID 5.4/08

Test material	Duration Method/ Guideline	Species/ strain/ sex/ no. per group	Dose levels/ frequency of dosing	Results	NOAEL/ LOAEL	Reference
Purity: 95.03%			17.5, 33.6 mg/kg bw/day f: 0, 1.3, 2.4, 18.9, 36.1 mg/kg bw/day	concentration and Ht) at ≥80 ppm. Increase Heinz bodies, reticulocytes, MCV at ≥ 640 ppm. Erythrocyte morphologic changes (increase incidence of Howell-Jolly bodies, polychromasia and hypochromasia) at ≥640 ppm. Decrease mean platelet counts at ≥640 ppm. Treatment-induced clinical anaemia at ≥640 ppm. Increase incidence of pigment deposit (haemosiderin) in liver, kidney, spleen and bone marrow at ≥ 80 ppm. Bone marrow hyperplasia and extramedullary haemopoiesis in the spleen at ≥ 80 ppm.	LOAEL: 80 ppm equivalent to 2.3 mg/kg bw/day	
Indoxacarb (racemic mixture 50:50 S:R) Purity: 95.03%	2-year/ combined chronic toxicity/carcino-genicity study OECD guideline 453	Rat/ Crl: CDBR/ m+f/ 72/sex/grp 10 male and 10 female sacrificed after 52 weeks	M: 0, 20, 40, 60, 125, 250 ppm equivalent to: 0, 0.798, 1.59, 2.40, 5.03, 10 mg/kg bw/day F: 0, 10, 20, 40, 60, 125 ppm equivalent to: 0, 0.554, 1.04, 2.13, 3.60, 7.83 mg/kg bw/day	Increased mortality in females during the first year (0/72, 1/72, 0/71, 0/72, 1/72, 7/73), seen during the 1st year of study, probably related to treatment. Bone marrow atrophy, thymic necrosis and splenic lymphoid depletion seen in those animals that died. Decrease body weight gain at ≥ 60 ppm in females (23-28%) and males at ≥ 125 ppm (17-22%). Anaemia seen in females at ≥ 60 ppm. Decrease circulating erythrocyte mass (RBC, Hb & Ht) at ≥ 20 ppm in females (6-21%) and ≥ 60 ppm in males (6-13%). Increase reticulocytes count and MCV at ≥ 60 ppm Increase incidence of pigment (haemosiderin) deposit in liver, spleen and kidney at ≥ 20 ppm.	NOAEL: 10 ppm equivalent to 0.55 mg/kg bw/day LOAEL: 20 ppm equivalent to 1.04 mg/kg bw/day	Frame, 1997a (IUCLID 5.4/09 & 5.7/02)
Indoxacarb (racemic mixture 50:50 S:R) Purity: 95.03%	18-month/ Carcinogenicity study OECD 451 (Complete microscopy was conducted on animals of the 0, 100, 200/150/125 mg groups.)	Mouse/ Crl: CD-1(ICR)BR/ m+f/ 70/sex/grp	0, 20, 100, 200/150/125 ppm equivalent to: m: 0, 2.63, 13.8, 32.2/22.7/17 mg/kg bw/day f: 0, 3.99, 20.3, 44.1/31.4/23.7 mg/kg bw/day The 200 ppm grp was reduced to 150 ppm on test day 126 and to 125 ppm on test day 287.	Increased mortality (males: 30, 20, 32 and 69%; females: 28, 27, 21 and 41%). Increase incidence of abnormal gait, head tilt and hyper-reactivity affecting most/all animals seen at ≥ 100 ppm. (No data on onset of clinical signs or time of deaths). Decrease body weight and body weight gain at ≥ 100 ppm. Decrease mean platelet counts in male mice at ≥ 100 ppm. Neuronal necrosis seen at ≥ 100 ppm (125 ppm males-2/70; in females 1/70 & 2/70 at 100 and 125 ppm respectively). Residual vacuolation of brain in 2 high-dose female.	NOAEL: 20 ppm equivalent to 2.63 mg/kg bw/day LOAEL: 100 ppm equivalent to 13.8 mg/kg bw/day	Frame, 1997b (IUCLID 5.7/01)

Test material	Duration Method/ Guideline	Species/ strain/ sex/ no. per group	Dose levels/ frequency of dosing	Results	NOAEL/ LOAEL	Reference
				Minimal to severe myocardial necrosis of the heart, with associated haemorrhage seen in males at the highest dose (12/70, assumed to occur in 12/21 surviving males). (No more data available from robust study summary. DAR reported heart effects present in most/all animals. Test substance-related causes of death included central nervous system disorder in males and females, and heart inflammation/necrosis in males only.)		
Indoxacarb (racemic mixture 50:50 S:R) Purity: 94.7%	28-day Test facility method, similar to OECD guideline 407. No haematology and clinical chemistry.	Rat/ CDBR/ m+f/5/sex/grp	0, 12, 29, 59, 118, 235 and 8/400 ppm equivalent to: m: 0, 1.02, 2.47, 5.89, 8.85, 20.6 and 0.71/23.4 mg/kg bw/day f: 0, 1.08, 2.61, 4.72, 9.29, 23.5 and 0.74/14.0 mg/kg bw/day	Treatment-related mortality in females at 235 and 8/400 ppm (2/5 on day 8 and day 21 and 3/5 days 27-28; no mortality in other groups). Decrease body weight gain at ≥ 118 ppm in females and ≥ 235 ppm in males.	NOAEL: 59 ppm equivalent to 4.72 mg/kg bw/day LOAEL: 118 ppm equivalent to 9.29 mg/kg bw/day	Reynolds, 1993a (IUCLID 5.4/01)
Indoxacarb (racemic mixture 50:50 S:R) Purity: 94.7%	28-day Test facility method, similar to OECD guideline 407. No haematology and clinical chemistry.	Mouse/ Crl: CD-1(ICR)BR/ m+f/10/sex/ grp	0, 12, 59, 118, 235, 29/400, 1225 and 2450 ppm equivalent to: m: 0, 2.06, 10.8, 17.9, 34.0 and 5.23/60.3 mg/kg bw/day f: 0, 2.52, 11.8, 21.5, 35.3 and 6.83/56.0 mg/kg bw/day	Mice in the two highest dose grps (1225 & 2450 ppm) were sacrificed in extremis after 7 days. One male and one female died at 235 ppm. Clinical signs suggestive of neurotoxicity (abnormal gait, head tilt and hyper-reactive) at ≥ 235 ppm in both sexes. Decrease Body weight gain at ≥ 118 ppm in males and ≥ 235 ppm in females.	NOAEL: 59 ppm equivalent to 10.8 mg/kg bw/day LOAEL: 118 ppm equivalent to 17.9 mg/kg bw/day	Reynolds, 1993b (IUCLID 5.4/02)
Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 94.5%	90 days US-EPA 82-7 (subacute neurotoxicity study)	Rat, Crl:CDBR/ m+f/12/sex/ group	M: 0, 10, 100 or 200 ppm equivalent to 0, 0.57, 5.62 or 11.9 mg/kg bw/day F: 0, 10, 50 or 100 ppm equivalent to 0, 0.69, 3.3 or 6.09 mg/kg bw/day	Treatment related mortality in females at 100 ppm (3/12) (no data on time of deaths) ↓ body weights in males at ≥ 100 ppm (12-16%), and females at ≥ 50 ppm (10-14%). ↓ body weight gain in males at ≥ 100 ppm (18-28%), and in females at ≥ 50 ppm (23-35%). Decrements in food consumption at ≥ 50 ppm (f) and at ≥100 ppm (m). No neurotoxic effects on assessed parameters. (Haematology was not included in the study)	NOAEL (general toxicity): 10 ppm equivalent to 0.57 mg/kg bw/day. NOAEL (neurotoxicity): Females: > 100 ppm equivalent to 6.09 mg/kg bw/day Males: > 200 ppm equivalent to 11.9 mg/kg bw/day. LOAEL (general toxicity): 50 ppm equivalent to 3.3 mg/kg bw/day	Malley, 1997 (IUCLID 5.8.3/02)

* Effects considered not severe enough to be basis of a LOAEL.

Mortality was observed in several studies. In two 90-day toxicity studies with Indoxacarb (enantiomeric reaction mass 75:25 S:R) in rats, 5/10 females died in the 100 ppm dose group (8.94

mg/kg bw/day) between days 8 and19 (MacKenzie, 1997) and 3/10 females died at 100 ppm (6.09 mg/kg bw/d) (Malley, 1997). Delayed mortality was also observed in the 28 day studies with Indoxacarb (racemic mixture 50:50 S:R) in rats and mice (2/5 female rats died at a dose of 23.5 mg/kg bw/day and 1/10 male and 1/10 female mice died at doses of 34 and 35.3 mg/kg bw/day respectively).

Test substance-induced anaemia of minimal to mild severity (more than 10% reduction in the circulating red cell mass and outside the laboratory's historical reference data range) was consistently evident across the species (rats, mice and dogs) in sub-chronic (90-day) and chronic oral dietary studies. The anaemic effects were associated with evidence of regeneration, such as reticulocytosis and macrocytosis (increased MCV). It was shown that these effects are reversible within 21 days after cessation of the exposure. The presence of Heinz bodies (mouse and dog) and methaemoglobinaemia (rat) in some studies suggest that the haemolytic effect was due to oxidative denaturation of haemoglobin. Methaemoglobuline (MetHb) formation (4%) was observed in some studies but was not determined in all studies. Effects secondary to haemolysis were also seen, including increased pigment (haemosiderin) deposit in the liver, spleen, bone marrow and sometimes the kidney. Incidence and severity grades of the haemosiderosis are not indicated in the available study summaries. Haemosiderosis is not reversible and only considered relevant for classification when also other indications of significant haemolytic anaemia are observed [(20b)]. The regenerative nature of the induced anaemia suggested that the effects might be reversible following cessation of exposure. In the study by Sarver (1998) all effects on circulating red blood cell parameters were reversible within the 21-day recovery period.

In mice, neurotoxicity was seen following repeated exposure to Indoxacarb (racemic mixture 50:50 S:R) but only at higher doses that are not relevant for classification. In a 90 day study abnormal gait and head tilt were noted from 30 mg/kg/day in females and 44 mg/kg bw/day in males. In a life time (18 month) study a low incidence of neuronal degeneration/necrosis, primarily affecting the piriform cortex and the hippocampus, occurred in the brain of both sexes at dose levels $\geq$ 20.3 mg/kg bw/day. Residual vacuolation was present in the piriform cortex of two females administered the top dose of 44 mg/kg bw/d. The neurotoxic potential of Indoxacarb (enantiomeric reaction mass 75:25 S:R) was also evaluated in a 90-day (sub-chronic) feeding study in rats (Malley (1997)). The assessments included functional observational battery, motor activity measurements and neuropathological examinations of nerve tissues and muscle. Significant reductions in body weights and food consumption of the animals were commonly observed. Treatment-related deaths (3/12 females) were seen at the high dose (100 ppm; 6.09 mg/kg/d but no neurotoxic effects were seen at the highest dose of 200 ppm (11.9 mg/kg/d) in males or 100 ppm (6.09 mg/kg/d) in females. Other effects observed included treatment-related minimal to severe myocardial necrosis of the heart in male mice in the lifetime study with Indoxacarb (racemic mixture 50:50 S:R). The heart lesion was seen only at the highest dose tested (32 mg/kg bw/day) and affected 12 (assumed to occur in 21 survivors from a total number of 70) animals.

In addition, a severe decrease in body weight gain (> 25%) was observed in most studies, mostly associated with a reduction in food intake > 10%.

5.6.2 Repeated dose toxicity: inhalation

No data available.

5.6.3 Repeated dose toxicity: dermal

A summary of repeated dose dermal toxicity data is presented in Table 5.9.

Table 5.9: Summary of repeated dose dermal toxicity data

Test material	Duration Method/ Guideline	Species/strain/ sex/ no. per group	Dose levels/ frequency of dosing	Results	NOAEL/ LOAEL	Reference
Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 95.83%	28-day OECD guideline 410	Rat/ Crl: CD(SD) IGS BR/ m+f/ 10/sex/group	0, 50, 500, 1000 and 2000 mg/kg bw/day 6h exposure/day	Increase incidences of stains in various parts of the body at ≥ 500 mg/kg bw/day. Significant decreased body wt gain in females at ≥ 500 mg/kg bw/day (45-47%; 26% in 50 mg/kg bw group). Decrease food consumption in females at ≥ 500 mg/kg bw/day. Haematological effects (↓ RBC, ↑ methaemoglobin (0.8-1.6%) and ↑ reticulocytes (44-60% females, 14-60% males) at ≥ 50 mg/kg bw/day. At 500 mg/kg ↓ in RBC, Hb, Ht were observed in both sexes (4-10%). Significant ↑ in abs + rel spleen wt, ↑ incidences of spleen discolouration. In males and females at ≥ 500 mg/kg bw/day, ↑ splenic haemopoiesis in males and females at ≥ 50 mg/kg bw/day.	NOAEL: <50 mg/kg bw/day LOAEL: 50 mg/kg bw/day	MacKenzie, 1999 (IUCLID 5.4/03)
indoxacarb (enantiomer reaction mass 75:25 S:R) Purity: 94.5%	28-day OECD guideline 410	Rat/ CDBR/ m+f/ 5/sex/grp	0, 50, 500, 1000 and 2000 mg/kg bw/day 6h exposure/day	Local: dermal irritation (erythema, desquamation, focal & non-focal eschar) in females at ≥ 1000 mg/kg bw/day and in 2000 mg/kg bw/day male group. Systemic: anaemia in females at 2000 mg/kg bw/day, seen as a decreased circulating erythrocyte mass at 2000 mg/kg bw/day and an increase MCV at 2000 mg/kg bw/day	NOAEL (systemic): 1000 mg/kg bw/day NOAEL (local): 500 mg/kg bw/day LOAEL (systemic): 2000 mg/kg bw/day LOAEL (local): 1000 mg/kg bw/day	Mertens, 1997 (IUCLID 5.4/04) (identical to Naas, 1997 in the DAR)

Following repeated dermal semi-occlusive application of Indoxacarb (enantiomeric reaction mass 75:25 S:R) in the rat, haemolysis and reduction in body weight were seen. In one of two short-term (28-day) studies, haemolytic effects (≥ 10% decrease) were only seen at the top dose (2000 mg/kg bw/d) in females only. However in a subsequent study, reduction in red cell mass (4 – 10% at and above 500 mg/kg/d) associated with increased methaemoglobin (1 – 2%) and reticulocytosis (14 – 60% increase in reticulocyte counts) were seen following application of 50 mg/kg bw/d (the lowest dose investigated). In this study, body weight was also severely reduced (26% in the 50 mg/kg dose group, although not statistically significant, and 45-47% in higher dose groups, significant). These effects were more prominent in females than males;

Local effects on the skin (erythema, desquamation, focal and non-focal eschar) were seen in the 28-day dermal study in female rats following semi-occlusive application of Indoxacarb (enantiomeric reaction mass 75:25 S:R) at dose levels of 1000 mg/kg/d and above, and in males at the top dose of 2000 mg/kg/d. However, no such effects were seen in a comparable 28-day study with the same strain of rat at dose levels up to 2000 mg/kg/d.

5.6.4 Other relevant information

The mechanism underlying the regenerative haemolytic effects of Indoxacarb (pure S enantiomer), Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) is yet to be clearly established. However, as demonstrated in toxicokinetics studies with Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R), a minor metabolic pathway involving the opening of the oxadiazine ring and subsequent cleavage results in the formation of "aniline analog" intermediate metabolites. Aniline and its related compounds are well-established haematotoxic agents known to induce haemolytic anaemia with corresponding changes in the spleen, the bone marrow and the liver in rats. Therefore, it is thought that the production of aniline-like metabolites may play an important role in the haematotoxicity effects of Indoxacarb containing compounds. *In vitro* studies with a putative "aniline analog" intermediate metabolite of the chiral compound, IN-MT713, investigating oxidation of glutathione in red blood cells and comparing relative sensitivities of rats, dogs and humans to this effect is available. The results suggest red blood cell damage through oxidation of glutathione, and that human is the least sensitive of the three species to this effect (Kemper, 2004, IUCLID, 5.9/02) This is in agreement with published reports which suggest that human haemoglobin may be less sensitive than the rat and dog to haemolysis induced by oxidative stress.

5.6.5 Summary and discussion of repeated dose toxicity

The results of the 90-day oral rat studies demonstrate that Indoxacarb (pure S enantiomer), Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) show similar dose-response characteristics. Consequently, the results of the additional studies in which animals have been exposed to Indoxacarb (racemic mixture 50:50 S:R) are considered to be relevant to the prediction of the toxicity and the classification of Indoxacarb (pure S enantiomer) and Indoxacarb (enantiomeric reaction mass 75:25 S:R).
The main findings included mortality (observed at 8.94 mg/kg bw) and haemolytic anaemia (observed at 3.78 mg/kg bw). In addition there was evidence of neurotoxicity from ≥ 30 mg/kg bw and of myocardial necrosis from ≥17 mg/kg in mice.
In most studies in the rat, females appeared to be more sensitive to the effects compared to males. Species differences were also observed with the rat more sensitive than mice and dogs to the toxicological effects.

The most sensitive effects of oral exposure to Indoxacarb (pure S enantiomer), Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) in both rats and dogs by the oral route are regenerative haemolytic anaemia and severe decreases in body weight parameters (> 25%) and/or nutritional status. Concurrent with Muller *et al.*, 2006, anaemic effects of more than 20% reduction in the circulating red cell mass (as stand-alone effect) were considered sufficient for classification[(20b)]. Methaemoglobuline (MetHb) formation (4%) was observed in some studies but was not determined in all studies. As a reduction in Haemoglobin and an increase in MetHb both result in a decrease in oxygen transport, these effects should be combined. However, since MetHb has not been determined in all studies this is difficult. Therefore, reductions in Hb somewhat below 20% are also considered sufficient as a stand-alone criteria for classification. Also premature deaths or a combination of less severe reduction in Hb (>10%) and haemolysis-related adverse effects (such as haemoglobinuria, haemosiderinuria or haemosiderosis) require classification.
Although there are in-vitro data that suggest that humans are less sensitive than rats and dogs to glutathione oxidation in red blood cells that were interpreted by the rapporteur MS in a way that humans were thought to be less sensitive to haemolysis induced by oxidative stress. In contrast, the guidance document of Muller et al., 2006 mentions that rats and mice are less sensitive to methaemoglobin production than man and dogs. The proposed mechanism underlying the

regenerative haemolytic effects of Indoxacarb (pure S enantiomer), Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) is yet to be clearly established. Therefore, for classification, these differences in sensitivity are not taken into account.

Depending on severity anaemia could be reversible by regenerative proliferation of blood cell precursors as long as no blood stem cells/early precursor cells are damaged. Indications on irreversible damage of erythrocyte precursor were seen at doses of ≥640 ppm (17.5 mg/kg bw/d) in dogs (Mertens, 1997). However, also at doses below 17.5 mg/kg bw/d haemolytic anaemia and related hemosiderin deposition are considered as serious health effects since there is no clearance mechanism for accelerated deposition of haemosiderosis in organs, and it could not be ruled out that premature deaths are related to haemotoxicity.

Indoxacarb (pure S enantiomer)
In a 90-day study with Indoxacarb (enantiomeric reaction mass 75:25 S:R) in rats, at 8.94 mg/kg bw/day mortality in females was reported (≤10 mg/kg bw/d, guidance value for CLP Cat1, 90 day). In addition, in 28-day toxicity studies with Indoxacarb (racemic mixture 50:50 S:R) in rats and mice, delayed mortality was observed at 23.5 and 34 mg/kg bw/day, respectively) ((≤30 mg/kg bw/d, guidance value for CLP Cat1, 28 day). Since the results of the repeated dose studies with Indoxacarb (pure S enantiomer), Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) show similar dose-response characteristics, the results of the additional studies in which animals have been exposed to Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb (racemic mixture 50:50 S:R) are considered to be also relevant to the prediction of the toxicity and the classification of Indoxacarb (pure S enantiomer).

In a 90-day dietary study in rats with Indoxacarb (pure S enantiomer) (only repeated dose toxicity study available for Indoxacarb (pure S enantiomer)) severely reduced body weight gain (> 32%), food intake (> 11%) and adverse effects on haematologic parameters (RBC, Hb and Ht change > 10%, max. 17%) were observed in females at doses ≥ 50 ppm (4.1 mg/kg bw/day). At the same doses, also significantly higher incidences of splenic erythrocytic and bone marrow hyperplasia was observed in males as well as females. In females increased haemosiderin deposition in liver (significantly higher incidence at 100 ppm, 8.5 mg/kg bw/d) and in spleen (significantly higher incidences ≥20 ppm, 1.7 mg/kg bw/d (10/10 females ≥20 ppm compared to 0/10 in controls, no data on severity grades) was seen. Secondary effects to the haemosiderosis were not observed in this 90 day study. The combination of effects such as Hb reduction ≥10% plus increased haemosiderosis occurred at 50 ppm (4.1 mg/kg bw/d), which is below the guidance value for Cat 1 (10 mg/kg bw/d, 90 days, CLP) and also below the guidance value of 5 mg/kg bw/d for T; R48/25 (DSD). Although no data are reported on the average severity scores, markedly increased incidences of haemolytic anemia-related effects below guidance doses are relevant for classification.

Therefore, based on the mortality observed at 8.94 mg/kg/day in the 90-day study with Indoxacarb (enantiomeric reaction mass 75:25 S:R) and supported by the mortality observed from 23.5 and 34 mg/kg/day in the 28-day studies with Indoxacarb (racemic mixture 50:50 S:R) and supported by the effects related to haemolytic anaemia in the 90-day study on Indoxacarb (pure S enantiomer), classification with T; R48/25 seems justified for Indoxacarb (pure S enantiomer) according to Directive 67/548/EEC (guidance value below 5 mg/kg bw), and STOT-RE Cat 1: H372 according to Regulation EC 1272/2008 (< 10 mg/kg bw). Proposal for classifications according to Dir 67/548/EEC and CLP are corresponding taking mortalities and haemolytic anaemia (at 50 ppm = 4.1 mg/kg bw/d), in combination with excellerated haemosiderin deposit in the spleen (≥20 ppm, 1.7 mg/kg bw/d) and in the liver (at 100 ppm, 8.5 mg/kg bw/d) into account in comparion to the guidance values (< 10 mg/kg bw for STOT-RE Cat 1 and < 5 mg/kg bw for T; R48/25).

Although the original CLH dossier of the dossier submitter proposed Xn; R48/22 for DSD, RAC considers the original proposal as a borderline case where simultaneous classification should be proposed in accordance to DSD and CLP. It is proposed to apply guidance values less strictly to differentiate Xn; R48/22 from T; R48/25. Taking into account haemotoxic/spleen/liver effects from 4.1 mg/kg bw/d in rats and as for other substances a harmonised classification on both regulations is therefore proposed.

According to the CLP criteria it is proposed to add the target organs to the hazard statement:
Causes damage to organs (blood, nervous system, heart) through prolonged or repeated exposure

No studies on repeated dermal exposure to Indoxacarb (pure S enantiomer) are reported. However, short-term (28-day) dermal exposure to Indoxacarb (enantiomeric reaction mass 75:25 S:R) in rats revealed similar effects as were observed after oral exposure, but, with a LOAEL of 50 mg/kg bw/day. Dose-related effects on body weight gain (reduction > 25%) and anaemia (decreased circulating erythrocytes and increased reticulocytes) were observed at the LOAEL. Whilst these effects occurred at doses below the classification cut-off, they are not considered severe enough (in comparison to the criteria of Muller *et al.*, 2006) or do not comply for combined effects required to justify classification for this route.
No data were available for long term inhalation exposure to Indoxacarb (pure S enantiomer), Indoxacarb (enantiomeric reaction mass 75:25 S:R) or Indoxacarb (racemic mixture 50:50 S:R).

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Based on results from 90 day studies, mortality in females was reported at 8.94 mg/kg b/day between day 8 and 19, deaths of females were also reported at 6.09 mg/kg bw/day (without any data on the time of death). Based on this mortality, and supported by the mortality data of 28 day toxicity studies with Indoxacarb (racemic mixture 50:50 S:R) in rats and mice (mortality at 23.5 and 34 mg/kg bw/day), classification with Xn; R48/22 is proposed for Indoxacarb (enantiomeric reaction mass 75:25 S:R) according to Directive 67/548/EEC (range 5-50 mg/kg bw for 90-day studies), and STOT-RE Cat 1: H372 according to Regulation EC 1272/2008 (< 10 mg/kg bw for 90-day studies).

Surviving rats of the same 90-day dietary study in rats with Indoxacarb (enantiomeric reaction mass 75:25 S:R) (only repeated dose toxicity study available for Indoxacarb (enantiomeric reaction mass 75:25 S:R)) demonstrated reduced body weight gain (≥ 38%) in females at doses ≥ 3.78 mg/kg bw/day and in males at 15 mg/kg bw/d . In addition, reduced RBC (> 10%-15%) was reported in females at ≥3.78 mg/kg bw/d, reduced RBC and Hb (-16% and -11%, resp.), as well as haemosiderin deposit in liver (in females ≥3.78 mg/kg bw/d), in kidneys (females at 100 mg/kg bw/d) and spleen (females at ≥0.67 mg/kg bw/d, males ≥6.01 mg/kg bw/d). The dose of 15 mg/kg bw/d which caused Hb reduction > 10% in males and increased incidences of haemosiderosis is below the guidance value for STOT-RE Cat 2.
Beyond findings in male rats, Hb reduction of -11% in combination with haemosiderin deposit in liver of female rats at ≥3.78 mg/kg bw/d argues for STOT-RE Cat 1. Although delayed mortilities of 5/10 females in the same study are lead for the original classification proposal from dossier submitter, haemolytic anaemia in combination with liver haemosiderosis observed in female rats at ≥3.78 mg/kg bw/d are in support of T; R48/25.

RAC considers the original proposal to classify with Xn; R48/22 as a borderline case where simultaneous classification should be proposed in accordance to DSD and CLP. It is proposed to apply guidance values less strictly to differentiate Xn; R48/22 from T; R48/25. Taking into account haemotoxic/liver effects from 3.78 mg/kg bw/d in rats and as for other substances a harmonised classification on both regulations is therefore proposed.

Although anaemic effects on blood cells could be reversible depending on its severity by regenerative proliferation of blood cell precursors, hemosiderin deposition is a non-reversible process. There is no clearance mechanism for accelerated deposition of haemosiderosis in organs, deposition in liver and kidney is a pathological finding irrespective of the severity. It could not be ruled out that premature deaths are related to haemotoxicity.
According to the CLP criteria it is proposed to add the target organs to the hazard statement:
Causes damage to organs (blood, nervous system, heart) through prolonged or repeated exposure

Similar effects were observed in rats after short-term (28d) dermal exposure to Indoxacarb (enantiomeric reaction mass 75:25 S:R) in rats, but, with a LOAEL of 50 mg/kg bw/day. Dose-related effects on body weight gain (reduction > 25%) and anaemia (decreased circulating erythrocytes and increased reticulocytes) were observed at the LOAEL. Whilst these effects occurred at doses below the classification cut-off, they are not considered severe enough (in comparison to the criteria of Muller *et al.*, 2006) or do not comply for combined effects required to justify for classification of this route.
No data were available for long term inhalation exposure to Indoxacarb (pure S enantiomer), Indoxacarb (enantiomeric reaction mass 75:25 S:R) or Indoxacarb (racemic mixture 50:50 S:R).

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: T; R48/25
CLP: STOT RE. 1; H372
Causes damage to organs (blood, nervous system, heart) through prolonged or repeated exposure

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: T; R48/25
CLP: STOT RE.1; H372
Causes damage to organs (blood, nervous system, heart) through prolonged or repeated exposure

5.7 Mutagenicity

5.7.1 In vitro data

A summary of the *in vitro* genotoxicity data is presented in Table 5.10.

Table 5.10: Summary of *in vitro* genotoxicity data

Test Material	Test system / methods / guideline	Organism / strain	Concentrations tested	Result		Remarks	Reference
				+S9	-S9		
Indoxacarb Purity: 95.47%	Bacterial reverse mutation (Ames test)/ OECD 471	*S. typhimurium*/ TA 98, TA 1535, TA 1537, TA100 *E coli*: WP2 uvr A	+S9/-S9: 75, 200, 600, 1800, 5000 µg/plate	Negative	Negative	No evidence of toxicity up to limit concentration. The number of revertants at all concentrations was similar to controls with or without metabolic activation.	Wagner and Klug (2004)
Indoxacarb	Bacterial	*S.*	+ S9/-S9: 10, 50,	Negative	Negative	No evidence of	Mathison

Test Material	Test system / methods / guideline	Organism / strain	Concentrations tested	Result		Remarks	Reference
				+S9	-S9		
(enantiomeric reaction mass 75:25 S:R) Purity: 94.5%	reverse mutation (Ames test) / OECD 471 and 472	*typhimurium*/ TA98, TA100, TA1535, TA97a E *coli*: WP2 uvr A (pKM 101)	100, 250, 500, 1000, 2500, 5000 µg/plate			toxicity up to limit concentration. The number of revertants at all concentrations was similar to controls with or without metabolic activation.	(1997)
Indoxacarb (enantiomeric reaction mass 75:25 S:R)Purity: 94.5%	Mammalian chromosome aberration test/ OECD 473	Human peripheral blood lymphocytes	+S9/-S9: 15.7, 31.3, 62.5, 125, 250, 500, 750, 1000 µg/mL	Negative	Negative	No statistically significant increase in structural chromosome aberrations at the initial or subsequent independent assay. Cytotoxicity as evidenced by mitotic inhibition (toxicity) of 76-90% observed at the highest concentration (1000 µg/mL). No significant increases in polyploidy.	Gudi and Schadly (1996)
Indoxacarb (enantiomeric reaction mass 75:25 S:R)Purity: 94.5%	Mammalian cell gene mutation test (HPRT-test)/ OECD 476	Chinese hamster ovary cell lines (CHO)	+S9/-S9: 3.1, 6.3, 12.5, 25, 100, 250 µg/mL	Negative	Negative	Cytotoxicity (i.e. low cloning efficiency) observed at the highest dose.	San and Clarke (1997c)
Indoxacarb (enantiomeric reaction mass 75:25 S:R)Purity: 94.5%	*In vitro* unscheduled DNA synthesis (UDS) assay/ OECD 482	Primary cultures: hepatocytes	1.56, 3.13, 6.3, 12.5, 25, 50, 100, 200 µg/mL	-	Negative	Evaluations for UDS were conducted at dose levels of 12.5 µg/mL and above. No significant increase in mean net nuclear counts. Cytoxicity at ≥ 100µg/mL.	San and Sly (1997a)

Indoxacarb (pure S enantiomer) gave negative responses in a well-reported bacterial gene mutation test at concentrations of up to 5000 µg/plate.
Indoxacarb (enantiomeric reaction mass 75:25 S:R) was investigated in a battery of *in vitro* tests. Negative results were observed in an Ames test, a chromosome aberration test, a mammalian cell gene mutation test and a liver UDS assay when tested up to limit concentration or concentration causing toxicity. The positive controls in all the studies gave expected results.

Additional *in vitro* investigations were conducted with the major metabolites IN-JT333 (N-decarbomethoxylated Indoxacarb) and IN-KG433 (the oxadiazine ring opened metabolite of Indoxacarb). IN-JT333 was negative in an Ames test, a mammalian cell gene mutation test and a chromosome aberration assay. IN-KG433 was negative in an Ames test, a liver UDS assay and a mammalian cell gene mutation test. Studies were conducted either to limit concentration or levels producing cytotoxicity.

5.7.2 In vivo data

A summary of the *in vivo* genotoxicity data is presented in Table 5.11.

Table 5.11: Summary of *in vivo* genotoxicity data

Test material/ method/ guideline	Species/strain / sex/ no. per group	Frequency of application	Sampling times	Dose levels	Result	Remark	Reference
Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 94.5% Bone marrow micronucleus test/ OECD 474	Mouse/ Crl:CD-1(ICR)BR/6m+ 6f per group	Single oral dose	24, 48 and 72h	M: 0, 3000, 4000 mg/kg bw F: 0, 1000, 2000 mg/kg bw	Negative	No increase in micronuclei in bone marrow cells was observed. Slight reduction in PCE/NCE ratio (10 – 20%). One 3000 mg/kg bw treated male was found dead 24h post dosing. Clinical signs of toxicity including convulsions, ataxia, abnormal gait, tremors, salivation and lethargy were observed at all dose levels in both sexes 2h after dosing.	Cox (1997)

In vivo, Indoxacarb (enantiomeric reaction mass 75:25 S:R) produced a negative result in a well-reported mouse bone marrow micronucleus test. There were no significant increases in micronucleated polychromatic erythrocytes in the bone marrow cells of treated mice. Only slight reduction in PCE/NCE ratio was observed, which may indicate that the test material did not reach the bone marrow. However, data from toxicokinetic studies indicate that following oral dosing, Indoxacarb (enantiomeric reaction mass 75:25 S:R) is distributed to the bone marrow. Therefore, it is reasonable to assume that the bone marrow has been exposed. Also, general systemic toxicity including mortality was observed in this study, hence the dose level tested is considered appropriate to detect any changes.

5.7.3 Human data

No data available.

5.7.4 Other relevant information

5.7.5 Summary and discussion of mutagenicity

Indoxacarb (pure S enantiomer)
Limited information on Indoxacarb (pure S enantiomer) (Ames test) indicates that Indoxacarb (pure S enantiomer) has no genotoxicity potential *in vitro.* Several *in vitro* studies (Ames test, chromosome aberration, gene mutation and UDS assays) and an *in vivo* bone marrow micronucleus study with Indoxacarb (enantiomeric reaction mass 75:25 S:R) were also negative. Since Indoxacarb (enantiomeric reaction mass 75:25 S:R) contains 75% of the S enantiomer, and dose ranges including relatively high concentrations of Indoxacarb (enantiomeric reaction mass 75:25 S:R) were used, these data provide further indications that Indoxacarb (pure S enantiomer) is not genotoxic *in vitro* and *in vivo.* Therefore, no classification is proposed for Indoxacarb (pure S enantiomer).

Indoxacarb (enantiomeric reaction mass 75:25 S:R)

From several *in vitro* studies on Indoxacarb (enantiomeric reaction mass 75:25 S:R) (Ames test, chromosome aberration, gene mutation and UDS assays), it can be concluded that Indoxacarb (enantiomeric reaction mass 75:25 S:R) has no genotoxicity potential *in vitro*. In addition, a negative result was obtained in an *in vivo* bone marrow micronucleus study with Indoxacarb (enantiomeric reaction mass 75:25 S:R). Therefore, no classification for Indoxacarb (enantiomeric reaction mass 75:25 S:R) is proposed.

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

5.8 Carcinogenicity

5.8.1 Carcinogenicity: oral

A summary of the results is presented in Table 5.12

Table 5.12: Summary of carcinogenicity data

Test material	Method/ Guideline	Species/ Strain/ Sex/ No per group	Dose level/ Dosing frequency	Results	Reference
Indoxacarb (racemic mixture 50:50 S:R)) Purity: 95.03%	2 year combined chronic toxicity/carcinogenicity study OECD 453	Rat/ Crl:CD(SD)BR/ m+f/ 72/sex/ concentration	M: 0, 20, 40, 60, 125, 250 ppm equivalent to: 0, 0.798, 1.59, 2.40, 5.03, 10 mg/kg bw/day F: 0, 10, 20, 40, 60, 125 ppm equivalent to: 0, 0.554, 1.04, 2.13, 3.60, 7.83 mg/kg bw/day	No treatment-related increased incidences of tumours or neoplastic changes were found at any concentration. Increased mortality was seen in females at 125 ppm during the 1st yr of study. Decreased body weight changes and haematological effects (reported in the repeated dose section) were observed.	Frame (1997a)
Indoxacarb (racemic mixture 50:50 S:R)Purity: 95.03%	OECD 451 18 month Carcinogenicity study	Mouse/ Crl:CD-1(ICR)BR/m+f/ 70/sex/ concentration	0, 20, 100, 200/150/125 ppm equivalent to: m: 0, 2.63, 13.8, 32.2/22.7/17.0 mg/kg bw/day f:0, 3.99, 20.3, 44.1/31.4/23.7 mg/kg bw/day Exposure to 200 ppm was reduced to 150 ppm on test day 126 and to 125 ppm on test day 287.	No treatment-related tumours or neoplastic changes were found at any concentration evaluated. Mortality, decreased body weight changes and neurotoxicity effects (reported in the repeated dose section) were observed. Effects on the heart (myocardial necrosis) were also seen in the male.	Frame (1997b)

Although neither Indoxacarb nor Indoxacarb (enantiomeric reaction mass 75:25 S:R) have been tested in carcinogenicity studies, the carcinogenic potential of Indoxacarb (racemic mixture 50:50 S:R) has been investigated in lifetime dietary studies in rats and mice. No increased incidences of tumours or any other neoplastic changes were observed in both species at the dietary levels causing mortality and producing general toxicity (see also Section 5.6).

5.8.2 Carcinogenicity: inhalation

No data available.

5.8.3 Carcinogenicity: dermal

No data available.

5.8.4 Carcinogenicity: human data

No data available.

5.8.5 Other relevant information

5.8.6 Summary and discussion of carcinogenicity

Indoxacarb (pure S enantiomer)
Indoxacarb (pure S enantiomer) has not been tested for carcinogenicity. However, Indoxacarb (racemic mixture 50:50 S:R) was tested for carcinogenicity in rats and mice and showed no increase in tumour incidence. It is not unknown whether a slightly higher exposure (factor 2) to the S-enantiomer in Indoxacarb (pure S enantiomer) would have an carcinogenic effect that is not observed with Indoxacarb (racemic mixture 50:50 S:R). At present, the lack of genotoxic potential (see section 5.7) would suggest that Indoxacarb (pure S enantiomer) is unlikely to be a carcinogen. Overall, the data suggest that no classification for carcinogenicity is appropriate for Indoxacarb (racemic mixture 50:50 S:R) and by extrapolation, also no classification is considered appropriate for Indoxacarb (pure S enantiomer).

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Indoxacarb (enantiomeric reaction mass 75:25 S:R) has not been tested for carcinogenicity. However, Indoxacarb (racemic mixture 50:50 S:R) was tested for carcinogenicity in rats and mice and showed no increase in tumour incidence. It is not unknown whether a slightly higher exposure (factor 1.5) to the S-enantiomer in Indoxacarb (enantiomeric reaction mass 75:25 S:R) would have an carcinogenic effect that is not observed with Indoxacarb (racemic mixture 50:50 S:R). Overall, the data suggest that no classification for carcinogenicity is appropriate for Indoxacarb (racemic mixture 50:50 S:R) and by extrapolation, also no classification is considered appropriate Indoxacarb (enantiomeric reaction mass 75:25 S:R).

Indoxacarb (pure S-enantiomer)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

5.9 Toxicity for reproduction

5.9.1 Effects on fertility

No data are available for Indoxacarb (pure S enantiomer) or Indoxacarb (enantiomeric reaction mass 75:25 S:R). The results of a two-generation reproduction study in the rat, conducted with Indoxacarb (racemic mixture 50:50 S:R), are summarized in table 5.13.

Table 5.13: Summary of fertility toxicity data

Route /Test material	Method/ guideline	Species/ Strain/sex/ no per grp	Dose levels	Exposure period	Critical effects	NOAEL Parental/ F1/ F2	Reference
Oral/diet Indoxacarb (racemix mixture 50:50 S:R) Purity: 95.3%	OECD 416	Rat, Crl:CD VAF/Plus/ m+f/ 26/sex/ group	0, 20, 60 or 100 ppm, equivalent to 0, 1.2, 3.7 or 6.5 mg/kg bw/day	70 days for F0 77 days for F1	No adverse effects on fertility parameters including mating, gestation indicies and length, number of pups born, survival or development.	Parental (F_0 + F_1): 20 ppm equivalent to 1.2 mg/kg bw/day F1(foetotxicity): 60 ppm equivalent to 3.7 mg/kg bw/day F2: > 100 ppm equivalent to 6.5 mg/kg bw/day	Breslin, 1997 (IUCLID 5.8.1/01)

No adverse effects on fertility parameters including mating, gestation indices and length, number of pups born, survival or development were observed at doses up to 100 ppm Indoxacarb (racemic mixture 50:50 S:R) in the diet (equivalent to 6.5 mg/kg/d). This dose caused marked maternal toxicity and some general signs of toxicity in males. Two F0 dams in the 100 ppm group were sacrificed in extremis during lactation with clinical signs of dehydration and signs indicative of neurotoxicity (ataxia and convulsions). Reduction in F0 maternal body weights (>10%) was seen at 100 ppm, with consequential effect on the weight of the pups (18% decrease at 100 ppm). Enlarged spleen was seen in F1 adult females at 100 ppm (3/26 animals) and the spleen weights of F0 and F1 parental animals were increased at and above 60 ppm (by 25-37%).

5.9.2 Developmental toxicity

A summary of the results of developmental toxicity studies is presented in Table 5.14.

Table 5.14: Summary of developmental toxicity data

Route/test material	Method/ guideline	Species/Strain sex/no per group exposure period	Dose levels	Critical effects, dams/foetuses	NOAEL/ LOAEL maternal toxicity	NOAEL/ LOAEL developmental toxicity	Reference
Oral/gavage Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 94.5%	OECD 414	Rat; Crl:CD(SD)BR/ Female/25 per dose group Gestation days 7-21	0, 0.5, 1, 2 or 4 mg/kg bw/day (vehicle: PEG)	Dams: No mortality. ↓ body weight gain and food consumption (≈ 37%) at 4 mg/kg bw/day. No effect on reproductive outcomes. Foetuses: No mortality. Slight ↓ body weight in the 4 mg/kg bw/day grp (6%). No evidence for teratogenic potential	NOAEL: 2 mg/kg bw/day LOAEL: 4 mg/kg bw/day	Teratogenicity: NOAEL: 4 mg/kg bw/day Embryotoxicity: NOAEL: 4 mg/kg bw/day	Munley, 1997 (IUCLID 5.8.2/01)
Oral/gavage	OECD 414	Rabbit, Hra(NZW)SP	0, 250, 500 or 1000	Dams: No mortality.	NOAEL: 500 mg/kg bw/day	Teratogenicity: NOAEL > 1000	Munley, 1995

Route/test material	Method/ guideline	Species/Strain sex/no per group exposure period	Dose levels	Critical effects, dams/foetuses	NOAEL/ LOAEL maternal toxicity	NOAEL/ LOAEL developmental toxicity	Reference
Indoxacarb (racemic mixture 50:50 S:R) Purity: 94.76%		Female/23 per dose group Gestation days 7-28	mg/kg bw/day (vehicle: 0.5% methyl-cellulose)	Discoloured stools at 1000 mg/kg bw/day. ↓ body weight gain (27%) and food consumption (6%) at 1000 mg/kg bw/day. No effect on reproductive outcomes. Foetuses: No mortality. ↓ body weight at 1000 mg/kg bw/day (10%). Retarded sternebral ossification at 1000 mg/kg bw/day	LOAEL: 1000 mg/kg bw/day	mg/kg bw/day Embyrotoxicity: NOAEL= 500 mg/kg bw/day LOAEL: 1000 mg/kg bw/day	(IUCLID 5.8.2/02
Oral/gavage Indoxacarb (enantiomeric reaction mass 75:25 S:R) Purity: 94.5%	Pilot study comparable to OECD 414, no GLP	Rat, Crl:CD(SD)BR Female/8 per dose group Gestation days 7-21	0, 1, 2, 4 or 8 mg/kg bw/day (vehicle: PEG)	Dams: No mortality seen. Salivation and stained fur at ≥ 2 mg/kg bw/day. ↓ body weight gain (17-54%) and food consumption (16-29%) at ≥ 2.0 mg/kg bw/day. No effect on reproductive outcomes. Foetuses: No mortality seen. ↓ body weight at 8.0 mg/kg bw/day. No external alterations.	NOAEL: 1 mg/kg bw/day	Embyrotoxicity: 4 mg/kg bw/day	Munley, 1997
Oral/gavage Indoxacarb (racemic mixture 50:50 S:R) Purity: 94.8%	OECD 414	Rat, Crl:CD(SD)BR Female/25 per dose group Gestation days 7-21	0, 10, 100, 500, 1000 mg/kg bw/day	Dams: ≥ 100 mg/kg bw: increased mortality, decreased bw and fi, hair loss, hunched, weak animals, GIT effects (distention, haemorrhage, ulceration, unusual content) Fetuses: ≥ 100 mg/kg bw decreased live fetuses, ≥ 250 mg/kg bw decreased bw. No malformations	NOAEL: 10 mg/kg bw/day	NOAEL: 10 mg/kg bw/day	Munley, 1997

Following oral administration, Indoxacarb (enantiomeric reaction mass 75:25 S:R) was not uniquely toxic to the rat conceptus and did not produce any developmental effects. Decreases in maternal body weight gains and food consumption (37%) were observed at 4 mg/kg/d, with consequential slight reduction (6%) in mean foetal body weights.

In the rabbit, reduction in foetal bodyweight (10%) and skeletal variation (*i.e.* retarded ossification of the sternebra) were observed following oral administration of 1000 mg/kg bw/day Indoxacarb (racemic mixture 50:50 S:R) during gestation. However, maternal toxicity (*i.e.* 27% decrease in body weight gain) was evident at this dose which may be responsible for the skeletal retardation.

5.9.3 Human data

No data available.

5.9.4 Other relevant information

5.9.5 Summary and discussion of reproductive toxicity

Indoxacarb (pure S enantiomer) has not been tested for effects on reproductive toxicity. However, Indoxacarb (racemic mixture 50:50 S:R) was tested for effects on fertility in the rat. No adverse effects on fertility were reported. By extrapolation, although no reproductive toxicity is expected based on the data from testing the racemic mixture 50:50 S:R , it is unknown whether a slightly higher exposure to the S-enantiomer (which is still below the dosage that induces delayed lethalities) would have an effect that is not observed with Indoxacarb (racemic mixture 50:50 S:R). At present, no concern was derived by the lack of effects on reproductive gonads in sub-chronic repeated exposure studies. Therefore, it is concluded that no classification is appropriate on Indoxacarb (pure S enantiomer) for effects on reproduction or fertility in rats.

In addition, no severe developmental effects were observed after exposure of pregnant rats or rabbits to Indoxacarb (enantiomeric reaction mass 75:25 S:R) or Indoxacarb (racemic mixture 50:50 S:R). Some small effects on body weight gain of the foetuses were observed, as well as retarded ossification, however, only at doses that also induced maternal toxicity. These foetal effects are considered as secondary to the maternal toxicity. Consequently, no classification for effects on fertility or development is considered appropriate for Indoxacarb (pure S enantiomer).

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Indoxacarb (enantiomeric reaction mass 75:25 S:R) has not been tested for effects on fertility. However, Indoxacarb (racemic mixture 50:50 S:R) was tested for effects on fertility in the rat. No adverse effects on fertility were reported. By extrapolation, although no reproductive toxicity is expected for Indoxacarb (enantiomeric reaction mass 75:25 S:R) based on the data from testing the racemic mixture 50:50 S:R, unknown whether a slightly higher exposure to the S-enantiomer in Indoxacarb (enantiomeric reaction mass 75:25 S:R) would have an effect that is not observed with Indoxacarb (racemic mixture 50:50 S:R). At present, no concern was derived by the lack of effects on reproductive gonads in sub-chronic repeated exposure studies. Therefore, it is concluded that no classification is appropriate on Indoxacarb (enantiomeric reaction mass 75:25 S:R) for effects on reproduction or fertility in rats.

In addition, no severe developmental effects were observed after exposure of pregnant rats or rabbits to Indoxacarb (enantiomeric reaction mass 75:25 S:R), Indoxacarb (racemic mixture 50:50 S:R). Some small effects on body weight gain of the foetuses were observed, as well as retarded ossification, however, only at doses that also induced maternal toxicity. These foetal effects are considered as secondary to the maternal toxicity. Consequently, no classification for effects on fertility or development is considered appropriate for Indoxacarb (enantiomeric reaction mass 75:25 S:R).

Indoxacarb (pure S enantiomer)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

Indoxacarb (enantiomeric reaction mass 75:25 S:R)
Directive 67/548/EEC: no classification proposed
CLP: no classification proposed

5.10 Other effects

No data available.

5.11 Derivation of DNEL(s) or other quantitative or qualitative measure for dose response

Not relevant for this type of report.

6 HUMAN HEALTH HAZARD ASSESSMENT OF PHYSICOCHEMICAL PROPERTIES

6.1 Explosivity

Indoxacarb (racemic mixture S:R 50:50) was tested in a standard explosivity study (92/69/EEC. A14 [(12)]). It was found not to be explosive under the influence of a flame and was not sensitive to impact or friction.

No data are available on Indoxacarb or Indoxacarb (enantiomeric reaction mass S:R 75:25), however the chemical structures of the materials are the same and therefore the results from the studies conducted on Indoxacarb (enantiomeric reaction mass S:R 50 :50) can be used to support the classification and labelling of Indoxacarb and Indoxacarb (enantiomeric reaction mass S:R 75:25).

No classification for explosivity is proposed.

6.2 Flammability

Indoxacarb (racemic mixture S:R 50:50) was tested in a standard flammability study (92/69/EEC.A10[(12)]) study. It did not support combustion in an initial screening study and did not propagate combustion of the product train within the specified time in the main study. Consequently no classification is proposed.

Indoxacarb (enantiomeric reaction mass S:R 50:50) was tested in a standard relative self ignition temperature study (92/69/EEC.A16[(12)]). No spontaneous ignition occurred in this study and consequently no classification is proposed.

No data are available on Indoxacarb or Indoxacarb (enantiomeric reaction mass S:R 75:25), however the chemical structures of all of the materials are the same and therefore the results from Indoxacarb (enantiomeric reaction mass S:R 50 :50) can be used to support the classification and labelling of Indoxacarb and Indoxacarb (enantiomeric reaction mass S:R 75:25).

Experience in handling and use indicates that Indoxacarb and Indoxacarb (enantiomeric reaction mass S:R 75:25) are not pyrophoric and do not react with water to liberate flammable gases.

No classification for flammability is proposed.

6.3 Oxidising potential

Examination of the chemical structure of Indoxacarb and Indoxacarb (enantiomeric reaction mass S:R 75:25) indicates that they do not contain any chemical groups typical of oxidising agents. Thus, the substances can be regarded as incapable of reacting exothermically with a combustible material.

No classification for oxidising properties is proposed.

7 ENVIRONMENTAL HAZARD ASSESSMENT

A detailed summary of the available studies has been reviewed under the Directives 98/8/EC and 91/414/EEC (see Annexes I and II to this document). The key information pertinent to determining a classification position is presented below.

Indoxacarb is an insecticide intended to control insects such as cockroaches and ants.

No environmental fate studies are available using Indoxacarb (pure S enantiomer). Environmental studies presented in this section have been performed on the following substances:
Indoxacarb (enantiomeric reaction mass S:R 75:25)
IN-JT333 (metabolite and degradant)
IN-KT413 (degradant)
DPX-MP062 150SC (a formulation containing 18.8% Indoxacarb (enantiomeric reaction mass S:R 75:25).

7.1 Aquatic compartment (including sediment)

7.1.1 Toxicity test results

7.1.1.1 Fish

- Short term toxicity to fish

An acute toxicity to fish study is available following OECD Guideline 203 using Indoxacarb (enantiomeric reaction mass 75:25 S:R), *Oncorhynchus mykiss* (rainbow trout) and flow-through conditions [(21)]. Test material was dissolved in DMF (Dimethylformamide) solvent to aid dispersion - the solubility in test medium was considered to be 0.9 mg/l. Measured concentrations were 72 – 81% of nominal and results are based on mean measured concentrations. Effects were only observed at the highest exposure concentration. At this concentration, 50% mortality was observed and based on measured concentrations the 96-h LC_{50} was 0.65 mg/l. This is considered to reflect a LC_{50} value at the limit of solubility which is effectively >0.1 mg/l and <1 mg/l.

For the purpose of Indoxacarb classification, the observed toxicity is assumed to be due to the insecticidally active S enantiomer with the R enantiomer being inactive. This means the LC_{50} value is read-across to Indoxacarb.

Indoxacarb (active S enantiomer) is metabolised by target organisms to form N-decarbomethoxylated Indoxacarb called IN-JT333. IN-JT333 is insecticidally active and induces a lethal reaction. On the basis of an aquatic/sediment degradation study [16] using Indoxacarb (enantiomeric reaction mass 75:25 S:R), IN-JT333 is considered an environmental degradant. Although it was not detected in water, it was present in sediment.

An acute toxicity to fish study was performed with IN-JT333 (N-decarbomethoxylated Indoxacarb). The study used *Oncorhynchus mykiss* (rainbow trout) and flow-through conditions according to OECD Guideline 203 [22]. Solubility in test water was considered to be 0.036 mg/l. During the definitive study, exposure solutions were clear and colourless with no precipitate. The highest test concentration was nominally 0.02 mg/l corresponding to 0.029 mg/l based on mean measured data which is within 80% of the maximum water solubility in test media. At this exposure concentration 50% mortality was observed so the 96-h LC_{50} is considered to be 0.029 mg/l. Although the LC_{50} is close to the quoted limit of solubility in the test medium, given the noted absence of precipitate in solutions, the degradant appears to be more acutely toxic to fish than the parent.

A further test [23] conducted with a second degradant (IN-KT413) was performed as a static limit test using *Oncorhynchus mykiss* (rainbow trout). No mortalities were observed and the 96-h LC_{50} was > 1.06 mg/l based on measured data.

- Long-term toxicity to fish

A 90-day long-term fish toxicity study [24] using Indoxacarb (enantiomeric reaction mass 75:25 S:R), following OECD 210 and flow-through conditions is available. The NOEC of 0.15 mg/l for fingerling survival, observed abnormalities, length and weight is based on mean measured concentrations.

7.1.1.2 Aquatic invertebrates

- Short term toxicity to aquatic invertebrates

Following OECD Guideline 202, the acute toxicity to the water flea (*Daphnia magna*) was assessed using Indoxacarb (enantiomeric reaction mass 75:25 S:R) under semi-static conditions [25]. Test material was dissolved in DMF solvent to aid dispersion with the solubility in test medium considered to be ~0.8 mg/l. The 48-h EC_{50} was 0.6 mg/l based on measured concentrations.

For the purpose of Indoxacarb classification, the observed toxicity is assumed to be due to the insecticidally active S enantiomer with the R enantiomer being inactive. This means the EC_{50} value is read-across to Indoxacarb.

Two further acute toxicity studies with the water flea (*Daphnia magna*) are available for two degradants (IN-JT333 [26] and IN-KT413 [27]). Both followed OECD Guideline 202 with the first using semi-static conditions and the second using static conditions. As only 20% mortality was observed at the highest IN-JT333 exposure concentration, the 48-h EC_{50} was >0.029 mg/l based on measured concentrations.

As only 15% mortality was observed at the highest IN-KT413 exposure concentration, the 48-h EC_{50} was >0.967 mg/l based on measured concentrations. This indicates that the degradant IN-KT413 is less acutely toxic to invertebrates than the parent.

The solubility of IN-JT333 in test water was considered to be 0.036 mg/l. The highest nominal exposure concentration was 0.04 mg/l which was achieved with the aid of a solvent. This corresponded to a mean measured concentration of 0.029 mg/l which is within 80% of maximum solubility in test water. Only 20% mortality was observed at the highest IN-JT333 exposure concentration of 0.029 mg/l so the 48-h EC_{50} was >0.029 mg/l based on measured concentrations. On this basis, the degradant IN-JT333 EC_{50} appears to be greater than the water solubility and the substance appears to be less toxic to invertebrates than the parent.

Long term toxicity to aquatic invertebrates

A semi-static 21-day long-term *Daphnia magna* toxicity study following OECD 211 is available [28] using Indoxacarb (enantiomeric reaction mass 75:25 S:R). Test material was dissolved in DMF solvent to aid dispersion with the solubility in test dilution medium considered to be ~0.82 mg/l. No significant effects were observed and based on measured concentrations the NOEC is quoted as 0.09 mg/l reflecting the highest exposure concentration.

A further chronic toxicity to the water flea (*Daphnia magna*) is available for degradant IN-KT413 following OECD Guideline 202 under semi-static conditions [29]. The NOEC based on measured concentrations was 3.9 mg/l.

7.1.1.3 Algae and aquatic plants

A static algal growth inhibition study is available using *Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum*) following OECD Guideline 201 and using test material DPX-MP062 150SC (formulation containing 18.8% Indoxacarb (enantiomeric reaction mass 75:25 S:R)) [30]. The test used a single nominal concentration of 4.1 mg/l DPX-MP062 150SC (0.77 mg/l Indoxacarb (enantiomeric reaction mass 75:25 S:R and 81.2 % inert ingredients) which was considered the approximate limit of solubility determined for this formulation in test medium. Due to the slightly alkaline pH (pH 6.77-7.46 at 0h and pH 7.44-8.02 at 72h), losses were observed (60 % of the nominal in test concentrations, 42 % of the nominal in abiotic control) and are considered due to hydrolysis. In addition, due to test light conditions, losses could be due to photolysis. At 72h 7.45% inhibition was observed. Based on the geometric mean of Indoxacarb (enantiomeric reaction mass 75:25 S:R) measured concentrations at 0 and 72 hours, the study 72-h E_rC_{50} is considered to be >0.60 mg/l.
For the purpose of classification for Indoxacarb, the observed toxicity is assumed to be due to the S enantiomer and the result is read-across.

Guideline / Test method	Species / Inoculum	Endpoint / Type of test	Exposure		Results (mg l^{-1})			Remarks
			design	duration	NOE_rC	E_bC_{50} [1]	E_rC_{50} [2]	
OECD 201; EPA 123-2	*P. subcapitata*	Growth inhibition, EC_{50}	Limit Test	72 hours	0.46	> 0.60	> 0.60	Nominal concentration of 0.77 mg l^{-1}

[1] calculated from the area under the growth curve; [2] calculated from growth rate

Chronic exposure was not maintained at the nominal level and the lack of analysis at 24 and 48 h means that the rate of decline over the 72 hours cannot be determined. Therefore, a NOE_rC of 0.46 mg l^{-1} based on the arithmetic mean measured concentration at 72 hours has been used. Whilst this can be considered as a very precautionary endpoint, the data presented clearly shows that algae is not the most sensitive trophic level. Therefore, this approach will not disadvantage the assessment of DPX-MP062 but allow for a long-term algal endpoint to be considered for the classification according to the 2.ATP of the CLP Regulation.

A further algal growth inhibition study is available using *Scenedesmus subspicatus* and the degradant IN-KT413 following OECD Guideline 201 (31). Based on initial measured data the calculated 72-hour E_rC_{50} was >108 mg/l. Effects were observed at all exposure concentrations so based on initial measured data the NOE_rC is considered to be <8.10 mg/l.

7.1.1.4 Sediment organisms

Two studies following OECD Guideline 219 are available using *Chironomus riparius* (midge larvae).

The first (32) was conducted with radio-labelled Indoxacarb (enantiomeric reaction mass 75:25 S:R) under static conditions with application to the aqueous phase. Analytical measurements (based on total radioactivity) indicated that concentrations of the dose solution used to spike the water were between 99-109% of the nominal concentrations. Measured concentrations in the spiked water at the start of the test were between 26-48% of the nominal concentrations. During the study these concentrations declined and on day 28 analysis showed these were between 13-23% of the nominal concentrations. Samples of the dry sediment on day 28 contained 17-41% of the total radiolabelled test substance. Based on nominal concentrations, the NOEC for emergence and for development was 26.2 µg/l.

The decline in water concentration and partitioning to sediment, mean that the exact concentrations received by the organisms cannot be calculated. For this reason the study only provides useful supporting information.

The second study (33) was conducted with the degradant IN-JT333 over 28 days under static conditions with application of test substance to the sediment. Exposure concentrations were maintained and results are based on initial measured concentrations. The emergence rate EC_{50} was 0.4693 mg/kg sediment and the development rate EC_{50} was 0.703 mg/kg sediment.

7.1.1.5 Other aquatic organisms

Not relevant for this type of dossier.

7.1.2 Calculation of Predicted No Effect Concentration (PNEC)

Not relevant for this type of dossier.

7.2 Terrestrial compartment

Not relevant for this type of dossier.

7.2.2 Calculation of Predicted No Effect Concentration (PNEC_soil)

Not relevant for this type of dossier.

7.3 Atmospheric compartment

Not relevant for this type of dossier.

7.4 Microbiological activity in sewage treatment systems

Not relevant for this type of dossier.

7.5 Calculation of Predicted No Effect Concentration for secondary poisoning (PNEC oral)

Not relevant for this type of dossier.

7.6 Conclusion on the environmental classification and labelling

For the purpose of Indoxacarb classification, the observed toxicity for tested mixtures of S:R enantiomers is assumed to be due to the S enantiomer. Therefore the results from studies with various ratios of S:R enantiomers can be read-across.

Ecotoxicity studies conducted using Indoxacarb (enantiomeric reaction mass 75:25 S:R) show that the substance is acutely toxic to fish and invertebrates with a 96-h LC_{50} of 0.65 mg/l and a 48-h EC_{50} of 0.6 mg/l. Both values are above the experimental solubility in pure water (0.225 ± 0.036 mg/l at 20°C) but within the solubility limit in the test water. Although effects were observed in algal growth inhibition studies using Indoxacarb (enantiomeric reaction mass 75:25 S:R), the E_rC_{50} is considered to be above the solubility of this substance in test media.

Chronic toxicity tests were conducted using Indoxacarb (enantiomeric reaction mass 75:25 S:R). A 90-day NOEC for fish is 0.15 mg/l, a 21-day NOEC for Daphnia magna is 0.09 mg/l and a 72-h NOE_rC for algae is 0.46 mg/l.

An acute toxicity to fish study was performed with major degradant IN-JT333 (N-decarbomethoxylated Indoxacarb) which is formed in soil and sediment. The 96-h LC_{50} was 0.029 mg/l. Other degradants were observed to exhibit less acute toxicity than the parent substance.

It is noted that the S enantiomer is insecticidally active and following metabolism to IN-JT333 induces death after a few hours. The available acute testing includes limited testing with insects and it is recognised that other insect species may be more sensitive.

There is some experimental evidence to suggest that the S and R enantiomers are likely to undergo removal processes in the aquatic environment through aqueous photolysis, hydrolysis, adsorption to sediment and anaerobic and aerobic degradation. In water this is considered to occur at the same rates for both enantiomers. Most removal reflects adsorption to sediment and primary degradation with water DT_{50} values between 2 and 4 days at 12°C. Sediment DT_{50} values are longer, ranging between 64.5 days under anaerobic conditions and 9.5 days under aerobic conditions at 12°C.

However, various degradants are more persistent. This includes IN-JT333 which although not detected in water, was detected in aerobic and anaerobic sediment samples during a simulation study. The calculated DT_{50} values of IN-JT333 at 20°C in aerobic and anaerobic sediments are 29 days and 23 days. IN-JT333 is anticipated to further degrade to substances with DT_{50} values of 19 days and >1 year at 20°C.

The simulation study showed that mineralisation of the substance was minimal over 28 days. At day 28 the mean CO_2 residues were 1.8% AR in the anaerobic system and 7.1% AR in the aerobic system.

Therefore, although primary degradation of S and R enantiomers occurs in water and sediment fairly rapidly, significant degradation to non-toxic components does not occur with 28 days.

Based on available information, neither Indoxacarb nor Indoxacarb (enantiomeric reaction mass 75:25 S:R) are considered readily degradable for the purpose of classification.

Similarly under the CLP Regulation, neither Indoxacarb nor Indoxacarb (enantiomeric reaction mass 75;25 S:R)) are considered to undergo rapid degradation to non-toxic substances in the aquatic environment.

Measured fish BCF data indicate the S enantiomer (Indoxacarb) is not highly bioaccumulative (BCF_{fish} 77.3) whilst the R enantiomer (IN-KN127) is significantly more bioaccumulative (BCF_{fish} 1,848). As the BCF_{fish} is <100 for the S enantiomer, Indoxacarb is not considered bioaccumulative for the purpose of classification and labelling. The measured BCF_{fish} for Indoxacarb (racemic mixture 50:50 S:R) is 950. The BCF_{fish} for Indoxacarb (enantiomeric reaction mass 75:25 S:R) is anticipated to reflect the presence of the R enantiomer falling between 77.3 and 1,848. As the R enantiomer is a significant component of the Indoxacarb enantiomeric 75:25 S:R mixture and the R enantiomer BCF_{fish} is considered bioaccumulative (i.e. > 100 and > 500 under CLP), the 75:25 S:R mixture is considered bioaccumulative for the purpose of classification and labelling.

Classification

The acute toxicity of both substances to aquatic organisms lies between 0.1 and 1 mg/l based on the 96-h LC50 of 0.65 mg/l to fish and on the 48-h EC50 of 0.6 mg/l to Daphnia magna. Both substances are not readily degradable neither do they fill the classification criteria of degrading biologically and/or abiotically in the aquatic environment to a level > 70 % within a 28 day period. The BCF 77.3 of Indoxacarb is below the classification limits 100 and 500 in Directive 67/548 and CLP Regulation respectively. However, the Indoxacarb (enantiomeric reaction mass 75:25 S:R) is considered bioaccumulative according to both criteria based on the BCF 1,848 of R enantiomer.

The specific concentration limits and M-factor are based on the acute toxicity values between 0.1 and 1 mg/l.

Classification of manufactured material Indoxacarb (enantiomeric reaction mass 75:25 S:R):
Following Directive 67/548/EEC, Indoxacarb (enantiomeric reaction mass 75:25 S:R) should be classified Dangerous for the Environment with the following risk and safety phrases:
N Dangerous for the Environment
R50 Very toxic to aquatic organisms
R53 May cause long term effects in the environment
S60 This material and its container must be disposed of as hazardous waste
S61 Avoid release to the environment. Refer to special instructions/Safety Data Sheet
Specific concentration limits:
N; R50-53: C ≥ 25 %; N; R51-53: 2.5 % ≤ C < 25 %; R52-53: 0.25 % ≤ C < 2.5 %

Based on the CLP Regulation , Indoxacarb (enantiomeric reaction mass 75:25 S:R) should be classified as: Aquatic Acute 1, Aquatic Chronic 1
With the following hazard phrases: H400 'Toxic to aquatic life' and H410 'Very toxic to aquatic life with long lasting effects'
Signal word: 'Warning'
Pictogram with the Environment symbol.
Additionally: M factor 1 based on $0.1<L(E)C_{50}\leq 1$ mg/l should apply.

Classification of Indoxacarb (pure S form)
Following Directive 67/548/EEC, Indoxacarb should be classified Dangerous for the Environment with the following risk and safety phrases:
N Dangerous for the Environment
R50 Very toxic to aquatic organisms
R53 May cause long term effects in the environment
S60 This material and its container must be disposed of as hazardous waste
S61 Avoid release to the environment. Refer to special instructions/Safety Data Sheet
Specific concentration limits:
N; R50-53: C ≥ 25 %; N; R51-53: 2.5 % ≤ C < 25 %; R52-53: 0.25 % ≤ C < 2.5 %

Based on the CLP Regulation , Indoxacarb should be classified as:
Aquatic Acute 1, Aquatic Chronic 1
With the following hazard phrases: H400 'Toxic to aquatic life' and H410 'Very toxic to aquatic life with long lasting effects'
Signal word: 'Warning'
Pictogram with the Environment symbol
Additionally: M factor 1 based on $0.1<L(E)C_{50}\leq 1$ mg/l should apply.

Classification according to the 2.ATP of the CLP Regulation

Acute hazard
There are no changes to the acute classification in the 2.ATP of the CLP Regulation. Consequently both substances are classified Acute Aquatic 1, M-factor 1 as described above in this Chapter.

Long-term hazard
Adequate chronic toxicity data for all three trophic levels is available for Indoxacarb (enantiomeric reaction mass 75:25 S:R) and Indoxacarb. In this case the classification is fone according to Table

4.1.0 (b)(i) or (ii) depending on rapid degradation. The substances are non rapidly degradable and consequently Table 4.1.0 (b)(i) is used. The cut-off for chronic NOEC for classifying to Category Chronic 1 is ≤ 0.1 mg/l. Chronic NOEC values available for these substances are a 90-day NOEC for fish of 0.15 mg/l, a 21-day NOEC for Daphnia magna of 0.09 mg/l and a 72-h NOE_rC for algae of 0.46 mg/l. Based on the 21-day NOEC to Daphnia of 0.09 mg/l the substances will be classified to Category Chronic 1 with a chronic M-factor of 1.

The classification criteria in Table 4.1.0 (b)(iii) are the same as in CLP Regulation. The classification is based on acute toxicity, rapid degradability and bioaccumulation. The classification of these two substances according to this Table is Aquatic Chronic 1 as described above.

Classification of manufactured material Indoxacarb (enantiomeric reaction mass 75:25 S:R):

Based on the 2. ATP of the CLP Regulation, Indoxacarb (enantiomeric reaction mass 75:25 S:R) should be classified as: Aquatic Acute 1, Aquatic Chronic 1
With the following hazard phrases: H400 'Toxic to aquatic life' and H410 'Very toxic to aquatic life with long lasting effects'
Signal word: 'Warning'
Pictogram with the Environment symbol.
Additionally: Acute M-factor:
1 based on $0.1<L(E)C_{50}\leq 1$ mg/l and
Chronic M-factor:
1 based on $0.01<NOEC\leq 0.1$ mg/l

Classification of Indoxacarb (pure S form)
Based on the 2.ATP of the CLP Regulation, Indoxacarb should be classified as:
Aquatic Acute 1, Aquatic Chronic 1
With the following hazard phrases: H400 'Toxic to aquatic life' and H410 'Very toxic to aquatic life with long lasting effects'
Signal word: 'Warning'
Pictogram with the Environment symbol
Additionally: Acute M-factor:
1 based on $0.1<L(E)C_{50}\leq 1$ mg/l and
Chronic M-factor:
1 based on $0.01<NOEC\leq 0.1$ mg/l

JUSTIFICATION THAT ACTION IS REQUIRED ON A COMMUNITY-WIDE BASIS

Indoxacarb has been reviewed as a new active substance under both the Biocidal Products Directive (98/8/EC) and the Plant Protection Products Directive (91/414/EEC). It was included into Annex I of Directive 91/414/EEC in 2006. The hazards of Indoxacarb have been assessed by the UK's Health and Safety Executive and the Dutch Board for the Authorisation of Plant Protection Products and Biocides as part of these regulatory programmes. These assessments were discussed and agreed by the appropriate European technical committees under each review programme.

In accordance with Article 36(2) of EC Regulation 1272/2008 on classification, labelling and packaging of substances and mixtures, Indoxacarb should now be considered for harmonised classification and labelling. This Annex XV dossier presents a classification and labelling proposal based on the information presented following the assessment of Indoxacarb under Directives

98/8/EC and 91/414/EEC. Document IIA of the assessment under 98/8/EC and the DAR under 91/414/EEC are attached to the IUCLID files.

OTHER INFORMATION

REFERENCES

1. Schmuckler,M.E.,Cooke, L.A.(1997a), Physical and Chemical Characteristics of DPX-KN128, E. I. du Pont de Nemours and Company, Report No.AMR 4141-96, 2 July 1997.

2. Cobranchi, D.P.,Schmuckler, M.E.(1997), Vapour Pressure of DPX-KN128 and Calculation of Henry's Law Constant, E. I. du Pont de Nemours and Company, Report No.AMR 4169-96, 8 May 1997.

3. Gravell, R.L.(1996),Autoflammability, Flammability, Explosive and Oxidising Properties of DPX-JW062 (A Racemic Mixture of KN128 and KN127), E. I. du Pont de Nemours and Company, Report No.AMR 3339-95, 22 February 1996.

4. Schmuckler,M.E.,Cooke, L.A.(1997b), Solubility in Organic Solvents, Colour, Physical State and Odour of DPX-MP062, E. I. du Pont de Nemours and Company, Report No.AMR 4426-97, 30 June 1997.

5. Dobbin, J.(2002),DPX-MP062: Determination of the Melting Point/ Melting Range and Decomposition Temperature, ABC Laboratories Europe, Coleraine, UK, Report No.DuPont-7557 Revision 1, 27 March 2002.

6. Sarff, P.(2002), DPX-MP062: Determination of the Aqueous Solubility, ABC Laboratories Inc., USA, Report No. DuPont-7497, 4 March 2002.

7. Schmuckler, M.E.,Moore, L.A.(1997), Melting Point, Relative Density, Appearance (Colour, Physical State, Odour) and pH of DPX-JW062, E. I. du Pont de Nemours and Company, Report No.AMR 3032-94, 2 July 1997.

8. Cobranchi, D.P.,Schmuckler, M.E.(1996), Vapour Pressure Determination of DPX-JW062 (A racemic mixture of KN128 and KN127), E. I. du Pont de Nemours and Company, Report No. AMR 3309-95, 21 February 1996.

9. Schmuckler,M.E.(1996), Water Solubility of DPX-JW062 (A racemic Mixture of KN128 and KN127), E. I. du Pont de Nemours and Company, Report No. AMR 2951-94, 21 February 1996.

10. Cooke, (1996a): AMR 3494-95.

11. Cooke, L.A.(1996b),Octanol Water Partition Coefficient of DPX-JW062 (A Racemic Mixture of KN128 and KN127), E. I. du Pont de Nemours and Company, Report No. AMR 3192-94, 2 April 1996

12. Gravell, R.L.(1996), Autoflammability, Flammability, Explosive and Oxidising Properties of DPX-JW062 (A Racemic Mixture of KN128 and KN127), E. I. du Pont de Nemours and Company, Report No.AMR 3339-95, 22 February 1996.

13. Lentz. N.R., (2002a), Hydrolysis of ^{14}C-DPX-MP062 in Buffer Solutions of pH 5, 7 and 9., Ricerca Biosciences, LLC, Metabolism Division, Concord, OH 44077-1000. Report No. DuPont-9800, 9 October 2002.

14. Lentz. N.R., (2002b) Photodegradation of ^{14}C-DPX-MP062 in pH Buffer by Simulated Light, Ricerca Biosciences, LLC, Metabolism Division, Concord, OH 44077-1000, Report No. DuPont-9801, 9 October 2002

15. Yoshida, T. (2005) Biodegradation Study of DPX-JW062 by Microorganisms, Kurume Laboratory, Chemicals Evaluation and Research Institute, Japan, Report No. 14300, 21 January 2005

16. Shaw, D., (2002) ^{14}C DPX-MP062 (A 3:1 Mixture of DPX-KN128 and IN-KN127):Degradability and Fate in the Water/Sediment System, Huntingdon Life Sciences Ltd., Woolley Road, Alconbury, Huntingdon, Cambridgeshire, PE28 4HS, England, Report No. DuPont-8417, 17 October 2002

17. Smyser, B.P.(2002a), Anaerobic Aquatic Metabolism of [Indanone-1-^{14}C]DPX-JW062 and [Trifluoromethoxyphenyl(U)-^{14}C]DPX-JW062, a Racemic (50:50) Mixture of DPX-KN128 and IN-KN127, Critical Path Services, LLC, 100 East Market Street, Suite #3, Newport, Delaware 19804, Report No.AMR 3236-94 RV2, 22 October 2002

18. Priester, T.M.; Djanegara, T.K.S.; Cooper, P.A. (1996), Batch Equilibration Study of DPX-JW062 (A Racemic Mixture of DPX-KN128 and DPX-KN127) and its Major Soil Degradate, DuPont Experimental Station, DuPont Agricultural Products, Experimental Station, Wilmington, DE, 19880-0402 USA, Report No. AMR 3489-95 Revision 1, 15 November 1996.

19. EU Technical Guidance Document 2003

20. Premkumar, N.D.; Cole, K.C.; Madsen, T.J. (1997), Bioconcentration and Metabolism of [Indanone-1-^{14}C]DPX-JW062 and [Trifluoromethoxyphenyl(U)-^{14}C]DPX-JW062 (Racemic Mixture of DPX-KN128 and DPX-KN127) in Fish, ABC Laboratories, Inc., 7200 E. ABC Lane, Columbia, MO 65202 USA, Report No. AMR 3663-95 Revision 1, 12 November 2002

20a. Aptula, A.O.; Patlewicz, G.; Roberts, D.W. (2005), Skin sensitization: Reaction Mechanistic applicability domains for structure-activity relationships, Chem. Res. Toxicol. 18: 1420-26.

20b. Muller, A.; Jacobsen, H.; Healy, E.; McMickan, S.; Istace, F.; Blaude, M.N.; Howden, P.; Fleig, H.; Schulte, A. (2006), Hazard classification of chemicals inducing haemolytic anaemia: An EU regulatory perspective. Regul. Toxicol. Pharmacol. 45: 229-41.

21. Hoke, R.A. (1996a), DPX-MP062 Technical (Approximately 75% DPX-KN128, 25% DPX-KN127): Flow-Through, Acute, 96-Hour LC_{50} to Rainbow Trout, Oncorhynchus mykiss, DuPont Haskell Laboratory, 1090 Elkton Road, P.O. Box 50, Newark, DE 19714 USA, Report No. HLR 911-96 Revision No. 2, 29 October 1997

22. Hoke, R.A. (1997b), IN-JT333-20: Flow-Through, Acute, 96-Hour LC_{50} to Rainbow Trout, Oncorhynchus mykiss, DuPont Haskell Laboratory, Elkton Road, P.O. Box 50, Newark, DE 19714 USA, Report No. HL-1997-00180, 19 June 1997

23. Boeri, R.L.; Magazu, J.P.; Ward, T.J.(1999a), IN-KT413 a Metabolite of DPX-MP062: Acute, Static, 96-Hour Toxicity Test to Rainbow Trout, Oncorhynchus mykiss, T.R. Wilbury Laboratories Inc., Marblehead, Massachusetts 01945, Report No. DuPont-1311, 24 February 1999

24. Hoke, R.A. (1997d), DPX-MP062 Technical (Approximately 75% DPX-KN128, 25% DPX-KN127): Early Life-Stage Toxicity to Rainbow Trout, Oncorhynchus mykiss, DuPont Haskell Laboratory, Elkton Road, P.O. Box 50, Newark, DE 19714 USA, Report No. HLR 598-96 Revision 1, 30 October 1997

25. Hoke, R.A. (1996b), DPX-MP062 Technical (Approximately 75% DPX-KN128, 25% DPXKN127) Static, Acute, 48-Hour EC_{50} to Daphnia magna, DuPont Haskell Laboratory, 1090 Elkton Road, P.O. Box 50, Newark, DE 19714 USA, Report No. HLR 603-96 Revision 2, 29 October 1997

26. Hoke, R.A. (1997c), IN-JT333-20: Static-Renewal, Acute, 48-Hour EC_{50} to Daphnia magna, DuPont Haskell Laboratory, Elkton Road, P.O. Box 50, Newark, DE 19714 USA, Report No. HL-1997-00006, 21 May 1997

27. Boeri, R.L.; Magazu, J.P.; Ward, T.J.(1999b), IN-KT413, a Metabolite of DPX-MP062: Acute, Static, 48-Hour Toxicity (EC50) Test to Daphnia magna, T.R. Wilbury Laboratories Inc., Marblehead, Massachusetts 01945, Report No. DuPont-1309, 3 March 1999

28. Hoke, R.A. (1997f), DPX-MP062 (Approximately 75% DPX-KN128, 25% IN-KN127): Chronic Toxicity to Daphnia magna, DuPont Haskell Laboratory, Elkton Road, P.O. Box 50, Newark, DE 19714 USA, Report No. HLR 597-96, 19 June 1997

29. Boeri, R.L.; Wyskiel, D.C.; Ward, T.J.(2003), IN-KT413: Chronic, Static-Renewal Toxicity to the Daphnid, Daphnia magna, T.R. Wilbury Laboratories Inc., Marblehead, Massachusetts 01945, Report No. DuPont-12041, 3 July 2003

30. Sloman T.E. (1999), DPX-MP062 150SC: Influence on Growth and Growth Rate of the Green Alga Selenastrum capricornutum, E.I. du Pont de Nemours and Company, Haskell Laboratory for Toxicology and Industrial Medicine, Elkton Road, P.O. Box 50, Newark, Delaware 19714-0050, Report No. DuPont-3154, 01 October 1999 (unpublished)

31. Boeri, R.L., Ward, T.J.(2000), IN-KT413: Influence on Growth and Growth Rate of the Algae, Selenastrum capricornutum, T.R. Wilbury Laboratories Inc., Marblehead, Massachusetts 01946, Report No. DuPont-3935, 16 October 2000.

32. Radford, K. (2001), DPX-MP062 : To Assess The Toxicity To The Sediment Dwelling Phase of the Midge Chironomus Riparius, Huntingdon Life Sciences Ltd, Woolley Road, Alconbury. Huntingdon, Cambridgeshire, UK, Report No. DuPont-4055, 05 December 2000

33. Radford, K. (2001), IN-JT333 : To Assess The Toxicity To The Sediment Dwelling Phase of the Midge Chironomus Riparius, Huntingdon Life Sciences Ltd, Woolley Road, Alconbury. Huntingdon, Cambridgeshire, UK, Report No. DuPont-4054, 11 January 2001

Competent Authority Report UK. Effects Assessment for the Active Substance INDOXACARB. May 2008.

European Commission. Draft Assessment Report Indoxacarb, prepared by The Netherlands, September 2000.

European Commission. Draft Assessment Report Indoxacarb, prepared by The Netherlands, Addendum January 2005.