Food and Agriculture Organization
of the United Nations

FAO SPECIFICATIONS AND EVALUATIONS FOR AGRICULTURAL PESTICIDES

SPINOSAD

A mixture of spinosyn A,
(2R,3aS,5aR,5bS,9S,13S,14R,16aS,16bR)-2-(6-deoxy-2,3,4-tri-O-methyl-α-L-mannopyranosyloxy)-13-(4-dimethylamino-2,3,4,6-tetradeoxy-β-D-erythropyranosyloxy)-9-ethyl-2,3,3a,5a,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-14-methyl-1H-8-oxacyclododeca[b]as-indacene-7,15-dione,
and spinosyn D,
(2R,3aS,5aR,5bS,9S,13S,14R,16aS,16bR)-2-(6-deoxy-2,3,4-tri-O-methyl-α-L-mannopyranosyloxy)-13-(4-dimethylamino-2,3,4,6-tetradeoxy-β-D-erythropyranosyloxy)-9-ethyl-2,3,3a,5a,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-4,14-dimethyl-1H-8-oxacyclododeca[b]as-indacene-7,15-dione,
with spinosyns A:D proportions in the range 50:50 to 95:5

2022

TABLE OF CONTENTS

DISCLAIMER[1]

FAO specifications are developed with the basic objective of promoting, as far as practicable, the manufacture, distribution and use of pesticides that meet basic quality requirements.

Compliance with the specifications does not constitute an endorsement or warranty of the fitness of a particular pesticide for a particular purpose, including its suitability for the control of any given pest, or its suitability for use in a particular area. Owing to the complexity of the problems involved, the suitability of pesticides for a particular purpose and the content of the labelling instructions must be decided at the national or provincial level.

Furthermore, pesticides that are manufactured to comply with these specifications are not exempted from any safety regulation or other legal or administrative provision applicable to their manufacture, sale, transportation, storage, handling, preparation and/or use.

FAO disclaims any and all liability for any injury, death, loss, damage or other prejudice of any kind that may be arise as a result of, or in connection with, the manufacture, sale, transportation, storage, handling, preparation and/or use of pesticides which are found, or are claimed, to have been manufactured to comply with these specifications.

Additionally, FAO wishes to alert users to the fact that improper storage, handling, preparation and/or use of pesticides can result in either a lowering or complete loss of safety and/or efficacy.

FAO is not responsible, and does not accept any liability, for the testing of pesticides for compliance with the specifications, nor for any methods recommended and/or used for testing compliance. As a result, FAO does not in any way warrant or represent that any pesticide claimed to comply with a FAO specification actually does so.

[1] This disclaimer applies to all specifications published by FAO.

INTRODUCTION

FAO establishes and publishes specifications* for technical material and related formulations of agricultural pesticides, with the objective that these specifications may be used to provide an international point of reference against which products can be judged either for regulatory purposes or in commercial dealings.

From 1999 onward, the development of FAO specifications follows the **New Procedure**, described first in the 5th edition of the "Manual on the development and use of FAO specifications for plant protection products" and later in the 1st edition of "Manual for Development and Use of FAO and WHO Specifications for Pesticides" (2002) - currently available as 3rd revision of the 1st edition (2016) - , which is available only on the internet through the FAO and WHO web sites.

This **New Procedure** follows a formal and transparent evaluation process. It describes the minimum data package, the procedure and evaluation applied by FAO and the Experts of the FAO/WHO Joint Meeting on Pesticide Specifications (JMPS). [Note: prior to 2002, the Experts were of the FAO Panel of Experts on Pesticide Specifications, Registration Requirements, Application Standards and Prior Informed Consent, which now forms part of the JMPM, rather than the JMPS.]

FAO Specifications now only apply to products for which the technical materials have been evaluated. Consequently from the year 1999 onwards the publication of FAO specifications under the **New Procedure** has changed. Every specification consists now of two parts namely the specifications and the evaluation report(s):

Part One: **The Specification** of the technical material and the related formulations of the pesticide in accordance with chapters 4 to 9 of the "Manual on development and use of FAO and WHO specifications for pesticides".

Part Two: The Evaluation Report(s) of the pesticide, reflecting the evaluation of the data package carried out by FAO and the JMPS. The data are provided by the manufacturer(s) according to the requirements of chapter 3 of the "FAO/WHO Manual on Pesticide Specifications" and supported by other information sources. The Evaluation Report includes the name(s) of the manufacturer(s) whose technical material has been evaluated. Evaluation reports on specifications developed subsequently to the original set of specifications are added in a chronological order to this report.

FAO specifications developed under the **New Procedure** do not necessarily apply to nominally similar products of other manufacturer(s), nor to those where the active ingredient is produced by other routes of manufacture. FAO has the possibility to extend the scope of the specifications to similar products but only when the JMPS has been satisfied that the additional products are equivalent to that which formed the basis of the reference specification.

Specifications bear the date (month and year) of publication of the current version. Evaluations bear the date (year) of the Meeting at which the recommendations were made by the JMPS.

* NOTE: PUBLICATIONS ARE AVAILABLE ON THE INTERNET AT (https://www.fao.org/pest-and-pesticide-management/guidelines-standards/faowho-joint-meeting-on-pesticide-specifications-jmps/pesticide-specifications/en/)

PART ONE

SPECIFICATIONS

SPINOSAD

SPINOSAD

INFORMATION

ISO common name

Spinosad (BSI, E-ISO, ANSI), being a mixture of spinosyns A and D, with A:D proportions in the range 50:50 to 95:5

Synonyms

None

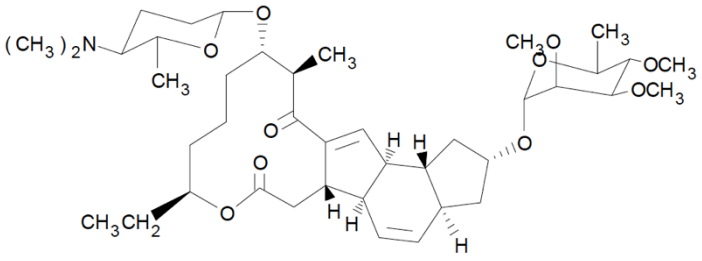
Chemical names

IUPAC A mixture of spinosyn A, (2*R*,3a*S*,5a*R*,5b*S*,9*S*,13*S*,14*R*,16a*S*,16b*R*)-2-(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyloxy)-13-(4-dimethylamino-2,3,4,6-tetradeoxy-β-D-erythropyranosyloxy)-9-ethyl-2,3,3a,5a,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-14-methyl-1*H*-8-oxacyclododeca[*b*]*as*-indacene-7,15-dione, and spinosyn D, (2*R*,3a*S*,5a*R*,5b*S*,9*S*,13*S*,14*R*,16a*S*,16b*R*)-2-(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyloxy)-13-(4-dimethylamino-2,3,4,6-tetradeoxy-β-D-erythropyranosyloxy)-9-ethyl-2,3,3a,5a,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-4,14-dimethyl-1*H*-8-oxacyclododeca[*b*]*as*-indacene-7,15-dione, with A:D proportions in the range 50:50 to 95:5

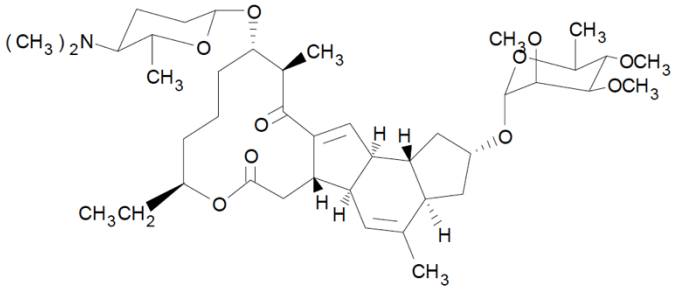
CA [2*R*-[2*R**,3a*S**,5a*R**,5b*S**,9*S**,13*S**(2*R**,5*S**,6*R**),14*R**,16a*S**,16b*R**]]-2-[(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyl)oxy]-13-[[5-(dimethylamino)tetrahydro-6-methyl-2*H*-pyran-2-yl]oxy]-9-ethyl-2,3,3a,5a,5b,6,9,10,11,12,13,14,16a,16b-tetradecahydro-14-methyl-1*H*-*as*-indaceno(3,2-*d*)oxacyclododecin-7,15-dione (spinosyn A), mixture with [2S-[2*R**,3a*S**,5a*R**,5bR*,9R*,13R*(2S*,5R*,6S*),14S*,16aR*,16bR*]]-2-[(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyl)oxy]-13-[[5-(dimethylamino)tetrahydro-6-methyl-2*H*-pyran-2-yl]oxy]-9-ethyl-2,3,3a,5a,5b,6,9,10,11,12,13,14,16a,16b-tetradecahydro-4,14-dimethyl-1*H*-*as*-indaceno(3,2-*d*)oxacyclododecin-7,15-dione (spinosyn D)

Structural formulae

spinosyn A



spinosyn D



Empirical formulae

spinosyn A: $C_{41}H_{65}NO_{10}$
spinosyn D: $C_{42}H_{67}NO_{10}$

Relative molecular mass

spinosyn A: 732.0
spinosyn D: 746.0

CAS Registry number

spinosyn A: 131929-60-7
spinosyn D: 131929-63-0

CIPAC number

636

EEC number

434-300-1

Identity tests

HPLC retention time, positive-ion ESI LC-MS.

SPINOSAD TECHNICAL MATERIAL

FAO Specification 636 / TC (Month 2021*)

*This specification, which is PART ONE of this publication, is based on an evaluation of data submitted by the manufacturers whose names are listed in the evaluation reports (*636/2005, 636/2021*). It should be applicable to relevant products of these manufacturers but it is not an endorsement of those products, nor a guarantee that they comply with the specifications. The specification may not be appropriate for the products of other manufacturers. The evaluation reports (*636/2005, 636/2021*) as PART TWO form an integral part of this publication.*

1 Description

The material shall consist of spinosad together with related manufacturing impurities and shall be a grey/white to tan coloured powdery material, free from visible extraneous matter and added modifying agents.

2 Active ingredient

2.1 **Identity tests** (636/TC/(M)/2, CIPAC Handbook L, p. 123, 2006)

The active ingredient shall comply with an identity test and, where the identity remains in doubt, shall comply with at least one additional test.

2.2 **Spinosad content** (636/TC/(M)/3, CIPAC Handbook L, p. 123, 2006)

The spinosad (spinosyn A + spinosyn D) content shall be declared (not less than 850 g/kg) and, when determined, the average measured content shall not be lower than the declared minimum content.

* Specifications may be revised and/or additional evaluations may be undertaken. Ensure the use of current versions by checking at: https://www.fao.org/pest-and-pesticide-management/guidelines-standards/faowho-joint-meeting-on-pesticide-specifications-jmps/pesticide-specifications/en/

SPINOSAD GRANULES

FAO Specification 636 / GR (Month 2021*)

*This specification, which is PART ONE of this publication, is based on an evaluation of data submitted by the manufacturers whose names are listed in the evaluation reports (*636/2005, 636/2021*). It should be applicable to relevant products of these manufacturers but it is not an endorsement of those products, nor a guarantee that they comply with the specifications. The specification may not be appropriate for the products of other manufacturers. The evaluation reports (*636/2005, 636/2021*) as PART TWO form an integral part of this publication.*

1 Description

The material shall consist of granules containing technical spinosad, complying with the requirements of the FAO specification 636/TC (Month 2021), together with suitable carriers and any other necessary formulants. The granules shall be free from visible extraneous matter and hard lumps, free-flowing, essentially non-dusty and intended for application by machine.

2 Active ingredient

2.1 **Identity tests** (636/GR/M/2, CIPAC Handbook L, p. 126, 2006)

The active ingredient shall comply with an identity test and, where the identity remains in doubt, shall comply with at least one additional test.

2.2 **Spinosad content** (636/GR/(M)/3, CIPAC Handbook L, p. 127, 2006)

The spinosad (spinosyn A + spinosyn D) content shall be declared (g/kg) and, when determined, the average measured content shall not differ from that declared by more than the following tolerances:

Declared content, g/kg	Tolerance
up to 25 Note: the upper limit is included in the range	± 10% of the declared content

3 Physical properties

3.1 **Pour and tap density** (MT 186, CIPAC Handbook K, p. 151, 2003)

Pour density: 0.47 to 0.61 g/ml.

Tap density: 0.52 to 0.66 g/ml.

* Specifications may be revised and/or additional evaluations may be undertaken. Ensure the use of current versions by checking at: https://www.fao.org/pest-and-pesticide-management/guidelines-standards/faowho-joint-meeting-on-pesticide-specifications-jmps/pesticide-specifications/en/

3.2 **Nominal size range** (MT 170, CIPAC Handbook F, p. 420, 1995) (Note 1)

Not less than 850 g/kg of the formulation shall be within the size range 1100 to 1600 µm.

3.3 **Dustiness** (MT 171.1, CIPAC Handbook P, p. 235, 2021)

Essentially non-dusty (Note 2).

3.4 **Attrition resistance** (MT 178, CIPAC Handbook H, p. 304, 1998)

Minimum: 98% attrition resistance.

4 Storage stability

4.1 **Stability at elevated temperature** (MT 46.4, CIPAC Handbook P, p. 232, 2021)

After storage at 54 ± 2°C for 14 days, the determined average active ingredient content must not be lower than 95% relative to the determined average content found before storage (Note 3) and the formulation shall continue to comply with the clauses for:

- nominal size range (3.2),
- dustiness (3.3),
- attrition resistance (3.4).

Note 1 Higher ratios increase the risk of segregation and adverse effects on the flow rate. This should be checked with the machine to be used. The purchaser should check that the nominal size range is suitable for his requirements, since different size ranges may affect biological activity.

Note 2 The optical method of MT 171.1 usually shows good correlation with the gravimetric method and can, therefore, be used as an alternative where the equipment is available. Where the correlation is in doubt, it must be checked with the formulation to be tested. In case of dispute the gravimetric method shall be used.

Note 3 Samples of the formulation taken before and after the storage stability test may be analyzed concurrently after the test in order to reduce the analytical error.

SPINOSAD SUSPENSION CONCENTRATE

FAO Specification 636/SC (Month 2021*)

*This specification, which is PART ONE of this publication, is based on an evaluation of data submitted by the manufacturers whose names are listed in the evaluation reports (*636/2005, 636/2021*). It should be applicable to relevant products of these manufacturers but it is not an endorsement of those products, nor a guarantee that they comply with the specifications. The specification may not be appropriate for the products of other manufacturers. The evaluation reports (*636/2005, 636/2021*) as PART TWO forms an integral part of this publication.*

1 Description

The material shall consist of a suspension of fine particles of technical spinosad complying with the requirements of FAO specification 636/TC (Month 2021), in an aqueous phase together with suitable formulants. After gentle agitation the material shall be homogeneous (Note 1) and suitable for further dilution in water.

2 Active ingredient

2.1 Identity tests (636/SC/(M)/2, CIPAC Handbook L, p. 125, 2006)

The active ingredient shall comply with an identity test and, where the identity remains in doubt, shall comply with at least one additional test.

2.2 Spinosad content (636/SC/M/3, CIPAC Handbook L, p. 125, 2006)

The spinosad (spinosyn A + spinosyn D) content shall be declared (g/kg or g/l at 20 ± 2ºC, Note 2) and, when determined, the average content measured shall not differ from that declared by more than the following tolerance:

Declared content, g/kg or g/l at 20 ± 2ºC	Tolerance
above 100 up to 250	± 6% of the declared content
above 250 up to 500	± 5% of the declared content
Note: the upper limit is included in each range	

3 Physical properties

3.1 pH range (MT 75.3, CIPAC Handbook J, p. 131, 2000)

pH range: 6.5 to 8.5.

3.2 Pourability (MT 148.1, CIPAC Handbook J, p. 133, 2000)

Maximum "residue": 5%.

* Specifications may be revised and/or additional evaluations may be undertaken. Ensure the use of current versions by checking at: ttps://www.fao.org/pest-and-pesticide-management/guidelines-standards/faowho-joint-meeting-on-pesticide-specifications-jmps/pesticide-specifications/en/.

3.3 **Spontaneity of dispersion** (MT 160, CIPAC Handbook F, p. 391, 1995) (Note 3)

Spontaneity of dispersion: minimum 75% after 5 min in CIPAC Standard Water D at 30 ± 2°C.

3.4 **Suspensibility** (MT 184.1, CIPAC Handbook P, p. 245, 2021) (Note 4)

Suspensibility: minimum 70% after 30 min in CIPAC Standard Water D at 25 ± 5°C.

3.5 **Wet sieve test** (MT 185, CIPAC Handbook K, p. 148, 2003) (Note 4)

Maximum: 0.5% of the formulation shall be retained on a 75 µm test sieve.

3.6 **Persistent foam** (MT 47.3, CIPAC Handbook O, p. 177, 2017) (Note 5)

Maximum: 20 ml after 1 min.

4 Storage stability

4.1 **Stability at 0°C** (MT 39.3, CIPAC Handbook J, p.126, 2000)

After storage at 0 ± 2°C for 7 days, the formulation shall continue to comply with the clauses for:

- suspensibility (3.4);
- wet sieve test (3.5).

4.2 **Stability at elevated temperature** (MT 46.4, CIPAC Handbook P, p. 232, 2021)

After storage at 54 ± 2°C for 14 days, the determined average active ingredient content must not be lower than 95% relative to the determined average content found before storage (Note 6) and the formulation shall continue to comply with the clauses for:

- pH range (3.1),
- pourability (3.2),
- spontaneity of dispersion (3.3),
- suspensibility (3.4),
- wet sieve test (3.5).

Note 1 Before sampling to verify the formulation quality, inspect the commercial container carefully. On standing, suspension concentrates usually develop a concentration gradient from the top to the bottom of the container. This may even result in the appearance of a clear liquid on the top and/or of sediment on the bottom. Therefore, before sampling, homogenize the formulation according to the instructions given by the manufacturer or, in the absence of such instructions, by gentle shaking of the commercial container (for example by inverting the closed container several times). Large containers must be opened and stirred adequately. After this procedure, the container should not contain a sticky layer of non-dispersed matter at the bottom. A suitable and simple method of checking for a non-dispersed sticky layer "cake" is by probing with a glass rod or similar device adapted to the size and shape of the container. All the physical and chemical tests must be carried out on a laboratory sample taken after the recommended homogenization procedure.

Note 2 Unless homogenization is carried out carefully, it is possible for the sample to become aerated. This can lead to errors in the determination of the mass per millilitre and in calculation of the active ingredient content (in g/l) if methods other than OECD 109 or MT 3.3 are used. If the buyer requires both g/kg and g/l at 20°C, then in case of dispute the analytical results shall be calculated as g/kg.

Note 3 Chemical assay is the only fully reliable method to measure the mass of active ingredient still in suspension. However, the simpler gravimetric method may be used on a routine basis provided

that it has been shown to give equal results to those of the chemical assay method. In case of dispute, the chemical method shall be the referee method.

Note 4 This test detects coarse particles (e.g. caused by crystal growth) or agglomerates (crust formation) or extraneous materials which could cause blockage of spray nozzles or filters in the spray tank.

Note 5 The mass of sample to be used in the test should be at the highest rate of use recommended by the supplier. The test is to be conducted in CIPAC standard water D at 25 ± 5°C.

Note 6 Samples of the formulation taken before and after the storage stability test may be analyzed concurrently after the test in order to reduce the analytical error.

PART TWO

EVALUATION REPORTS

SPINOSAD

SPINOSAD

FAO/WHO EVALUATION REPORT 636/2021

Recommendations

The Meeting recommended that

(i) The change of name of the manufacturer holding the FAO and WHO reference specifications for spinosad TC, GR and SC (FAO) and TC, and SC (WHO) from Dow AgroSciences to Corteva Agriscience should be noted by FAO and WHO.

(ii) The updated FAO specifications for spinosad TC, GR and SC should be adopted by FAO.

(iii) The updated WHO specifications for spinosad TC, GR, extended release GR, SC, mono- and bilayer DT and EC should be adopted by WHO.

Appraisal

The Meeting noted, that Corteva Agriscience (Corteva) has been formed from the merger of Dow and DuPont in 2017 and became a standalone company in June 2019[1]. The intellectual property rights for spinosad and its agricultural formulations previously owned by E.I. DuPont (DuPont) then was integrated into the portfolio of Corteva Agriscience. Its predecessor company, Dow, had been the proposer and holder of the FAO reference specifications for spinosad TC, SC and GR (FAO/WHO Evaluation Report 636/2005). Certain public heath formulations containing spinosad have been developed and are owned by Clarke International LLC (Clarke). These spinosad formulations include the EC used in public health (WHO specification 636/EC), the GR and extended release GR (WHO specifications 636/GR/1 and 636/GR/2) and the spinosad mono- and bilayer tablets (WHO specifications 636/DT/1 and 636/DT/2).

As such a transition may raise certain concerns on the continued validity of the FAO specification for spinosad technical material and formulations (see also FAO/WHO Manual, Section 2.7 on revision of specifications), Corteva was contacted by FAO and a statement on the support of the reference specifications and possible changes therein was requested.

Corteva later on provided a confirmation in writing (Corteva, 2021[2]) to FAO confirming the continued support for the FAO reference specifications for spinosad TC, SC and GR. Corteva explained, that both manufacturing site and -process for spinosad were not affected by the transition from DuPont to their company and confirmed the continued validity of the published specifications and stewardship for them.

[1] https://www.corteva.ca/en/about-corteva/our-history.html
[2] Letter Corteva to FAO and WHO, dated Feb. 22, 2021

In particular, Corteva provided the detailed information as follows:

- The intellectual property rights for spinosad TC and certain formulations used in agriculture and public health have been transferred from previously Dow AgroSciences to now Corteva.
- The manufacturing of spinosad TC and certain formulations used in agriculture and public health which are now under control of Corteva continue to comply with all specifications clauses and limits as per the data packages in support of spinosad TC and formulations evaluated by JMPS in 2005 and subsequent years.
- Corteva assure the continued support and stewardship for spinosad TC and certain formulations acquired from Dow AgroSciences.
- The public health formulations containing spinosad have been developed and owned by Clarke International LLC (Clarke). These formulations include the spinosad EC used in public health (WHO specification 636/EC), the spinosad GR and extended release GR (WHO specifications 636/GR/1 and 636/GR/2 ,and the spinosad mono- and bilayer tablets (WHO specifications 636/DT/1 and 636/DT/2). Clarke is and will remain the sole owner of these formulations captured in the related reference specifications.

For these reasons, the Meeting recommended that Corteva should be noted by FAO and WHO as new holder of the reference specifications for spinosad TC, GR and SC previously owned by Dow AgroSciences. The ownership of Clarke for the formulations used in public health (EC, mono- and bilayer DT, GR and extended release GR) should be reconfirmed.

The Meeting also noted that the specifications for spinosad TC and formulated products needed some updates. On one hand, the analytical methods for determination of the content of spinosad in TC, fast release GR and SC, the DT and the EC and extended release GR are now published in CIPAC Handbooks L, M and O respectively. On the other hand, several MT methods have been revised by CIPAC to achieve better harmonization and progress in technology (such as Suspensibility: MT 184.1 instead of MT 184, Stability at elevated temperature MT 46.4 instead of MT 46.3, Dustiness: MT 171.1 instead of MT 171, Persistent foam: MT 47.3 instead of MT 47.2).

The Meeting also considered the attrition resistance clause in the WHO specifications for a mono- and bilayer DT, respectively: currently MT 193 is used to determine that property. CIPAC have declared that method obsolete for new specifications, and recommend to use MT 178.2 instead. However, MT 178.2 comes with a size limitation: the method is applicable to tablets < 1 cm in diameter. Both the mono- and bilayer DT exceed that limit and do not qualify for that method. Therefore, the Meeting recommended to keep the clauses for attrition resistance referring to MT 193.

The Meeting also recommended to update some footnotes of the formulation specifications aligning them with the most recent versions of the specification templates in the Manual.

SPINOSAD

FAO/WHO EVALUATION REPORT 636/2005

Recommendations

The Meeting recommended that:

(i) the specifications for spinosad TC, SC and GR, proposed by Dow AgroSciences, should be adopted by FAO;

(ii) the specifications for spinosad TC, SC and GR, proposed by Dow AgroSciences, should be adopted by WHO, subject to satisfactory evaluation of these products in public health applications by WHOPES.

Appraisal

The Meeting considered data and draft specifications for spinosad, submitted by Dow AgroSciences in 2004. Spinosad is a macrocyclic lactone insecticide that had not previously been the subject of a WHO or FAO specification. The data submitted were in accordance with the requirements of the manual (FAO/WHO 2002) and supported the proposed FAO and WHO specifications for TC, SC and GR.

The spinosad toxicology was evaluated by the FAO/WHO JMPR in 2001 (JMPR 2001). Spinosad residues data were evaluated by the FAO/WHO JMPR in 2001 (JMPR, 2001) and there are currently several Codex maximum residue limits (MRLs) for spinosad. Spinosad was reviewed and approved by the U.S. EPA in 1997 and subsequent regulatory reviews and approvals have occurred in more than 60 countries including Australia, Brazil, Canada, India, Japan, New Zealand and South Africa. Spinosad has been under evaluation by the European Commission as a new active substance since 2000 and EU member state evaluations and provisional approvals have occurred in the Netherlands, Spain and the UK. Spinosad SC and GR formulations are under development as mosquito larvicides and are currently being evaluated by WHOPES, with a report expected in 2006.

Spinosad is under patent in some countries (Australia, Japan, UK), until December 2009, and in the country of technical product manufacture (USA), until March 2015.

The ISO common name, spinosad, denotes an insecticide consisting of two components, called spinosyns A and D (which may be referred to simply as A and D, below). The spinosyns are produced by a soil bacterium, *Saccharopolyspora spinosa,* belonging to the group Actinomycetes, a large group of gram-positive filamentous or branching bacilli.

Spinosad is produced in a fermentation process, where it is obtained by extraction and purification of the whole broth. Spinosyns A and D are present in the isolated spinosad, in proportions of 65-95% and 5-35%, respectively, together with traces of spinosyn-related compounds and other materials derived from the fermentation and purification process. The specified proportions of spinosyns A and D in spinosad are in agreement with the definition of the ISO common name.

The two main spinosyns, A and D, are closely related structurally and represent more than 85% of technical spinosad and are responsible for most of its insecticidal activity. They differ only in the presence of an additional methyl group attached to the bridging carbon of the indacene moiety in spinosyn D. Spinosyns A and D are relatively high molecular weight compounds (732 and 746, respectively). The additional methyl group has a significant effect on certain properties and many of the physico-chemical data were generated using separated and purified A and D.

Spinosyns A and D have very low vapour pressures, making them essentially non-volatile. Spinosyns A and D are weak bases, with pK_as of 8.1 and 7.9, respectively. Spinosyn A has a rather low, and pH-dependent, water solubility (290 mg/l at pH 5), with that of D even lower (29 mg/l at pH 5). As may be expected for weak bases, the water solubility decreases with increasing pH in both cases. The octanol/water partition coefficient is also pH-dependent, 2.8 and 3.2 at pH 7, expressed as log P K_{ow} for A and D, respectively, with increasing log P K_{ow} with increasing pH. Both spinosyns are resistant to hydrolysis in sterile, buffered water, with no detectable hydrolysis at pH 5 and increasing but very slow hydrolysis at pH 7 and 9. Aqueous photolysis of A and D at pH 7 was rapid with a half-life of less than one day.

The Meeting was provided with commercially confidential information on the manufacturing process and 7-batch analysis data on purity and all impurities ≥1 g/kg. The Meeting noted that, although technical spinosad is of biological origin, the unaccountable fraction was 20 g/kg or less in all batches and that the data supported the proposed minimum active ingredient content of 850 g/kg. These data were confirmed as identical to those submitted for registration in Switzerland. One of the 7 batches, with a slightly higher content of D and an average content of the minor spinosyns, was utilized for the toxicity testing.

The Meeting agreed with the manufacturer that none of the impurities should be considered as relevant.

Analytical methods to determine spinosyns A and D in TC, SC and GR were adopted by CIPAC in 2005. Spinosyns A and D are determined by reversed-phase HPLC with a methanol/acetonitrile/water/acetic acid mobile phase and UV detection. The identity test is based on HPLC-separation of spinosyns A and D and detection by positive ion ESI-MS. The test is highly specific, involving comparison of the retention times of A and D in the HPLC-chromatogram, together with the mass spectra of A and D, showing proton- and sodium adducts and fragmentation.

Draft specifications were submitted for spinosad TC, SC and GR.

At the time of the meeting, the general distinction between TC and TK was still under discussion with industry, although a cut-off value for purity of 900 g/kg had been used as one criterion by the JMPS. The distinction is important because TK specifications have an upper limit for active ingredient content and TC specifications do not. The rationale has been to encourage production of TCs with the highest possible purity, because the maximum possible increase in hazard due to the active ingredient cannot exceed 10% (taken to represent a negligible increase), whereas the consequent proportional reduction in impurity levels may be very significant. This approach cannot be adopted for TK, because the maximum increase in hazard due active ingredient could exceed the 10% threshold.

On this basis, therefore, the proposed minimum content of spinosad in the technical grade active ingredient (850 g/kg) might be considered to represent a TK. The

Meeting noted that the hazards presented by spinosyns A and D are similar and therefore potential changes in their proportions do not affect the decision as to whether technical spinosad is a TC or a TK. Taking into account the manufacturing process, the nature of the impurities and the minimum content of the active ingredient, the Meeting considered that it was not necessary to introduce an upper limit for spinosad content and agreed that, exceptionally, technical spinosad should be considered to be a TC, rather than a TK.

The proportions of spinosyns A and D in technical spinosad TC were confirmed to be in agreement with the ISO definition of the spinosad common name and therefore it was not necessary to introduce a clause specifying the range of ratios.

The proposed specification for SC conformed to the guideline presented in the manual (FAO/WHO 2002) and was supported by the data held by the registration authorities in Switzerland.

The proposed specification for GR differed from the guideline given in the manual, in that the ±10% tolerance for a.i. content was narrower than the ±15% maximum. The manufacturer confirmed the proposed tolerance of ±10% for the 10 g/kg GR formulation was always met in practice and the Meeting agreed to accept it.

SUPPORTING INFORMATION
FOR
EVALUATION REPORT 636/2005

Uses

Spinosad is an insecticide, used for the control of caterpillars, thrips, beetle and fly pests in a range of fruit and vegetable crops, ornamentals, turf, and stored grains. Spinosad has contact activity on all life stages of insects, including eggs, larvae and adults. Eggs must be sprayed directly but larvae and adults can be effectively dosed through contact with treated surfaces. Spinosad is most effective when ingested. Foliar applications are not highly systemic, although trans-laminar activity is evident in certain vegetable crops and ornamental plants. Spinosad acts by altering the function of nicotinic- and GABA-gated ion channels of insect nervous systems but it does not interact with known binding sites for other nicotinic- or GABA-agonistic insecticides. It is used in agriculture, horticulture, forestry, and public health against a wide range of insects including thrips, Mediterranean fruit fly, olive fruit fly, codling moth, caterpillars, leaf miners, Colorado beetle and potato worm (Sparks *et al*. 1998).

Identity of the active ingredient

ISO common name

Spinosad (BSI, E-ISO, ANSI), being a mixture of spinosyns A and D, with A:D proportions in the range 50:50 to 95:5

Synonyms

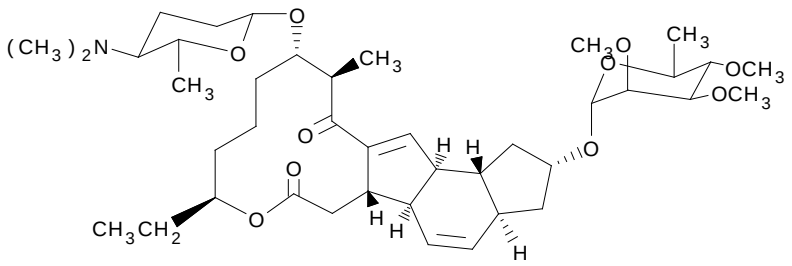
None

Chemical names

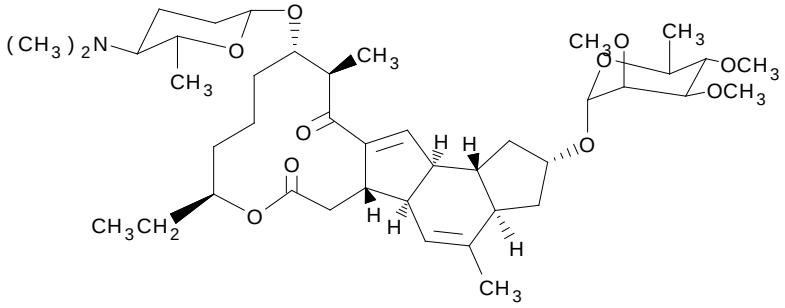
IUPAC A mixture of spinosyn A, (2*R*,3a*S*,5a*R*,5b*S*,9*S*,13*S*,14*R*,16a*S*,16b*R*)-2-(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyloxy)-13-(4-dimethylamino-2,3,4,6-tetradeoxy-β-D-erythropyranosyloxy)-9-ethyl-2,3,3a,5a,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-14-methyl-1*H*-8-oxacyclododeca[*b*]*as*-indacene-7,15-dione, and spinosyn D, (2*R*,3a*S*,5a*R*,5b*S*,9*S*,13*S*,14*R*,16a*S*,16b*R*)-2-(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyloxy)-13-(4-dimethylamino-2,3,4,6-tetradeoxy-β-D-erythropyranosyloxy)-9-ethyl-2,3,3a,5a,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-4,14-dimethyl-1*H*-8-oxacyclododeca[*b*]*as*-indacene-7,15-dione, with A:D proportions in the range 50:50 to 95:5

CA [2*R*-[2*R**,3a*S**,5a*R**,5b*S**,9*S**,13*S**(2*R**,5*S**,6*R**),14*R**,16a*S**,16b*R**]]-2-[(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyl)oxy]-13-[[5-(dimethylamino)tetrahydro-6-methyl-2*H*-pyran-2-yl]oxy]-9-ethyl-2,3,3a,5a,5b,6,9,10,11,12,13,14,16a,16b-tetradecahydro-14-methyl-1*H*-*as*-indaceno(3,2-*d*)oxacyclododecin-7,15-dione (spinosyn A), mixture with [2S-[2*R**,3a*S**,5a*R**,5bR*,9R*,13R*(2S*,5R*,6S*),14S*,16aR*,16bR*]]-2-[(6-deoxy-2,3,4-tri-*O*-methyl-α-L-mannopyranosyl)oxy]-13-[[5-(dimethylamino)tetrahydro-6-methyl-2*H*-pyran-2-yl]oxy]-9-ethyl-2,3,3a,5a,5b,6,9,10,11,12,13,14,16a,16b-tetradecahydro-4,14-dimethyl-1*H*-*as*-indaceno(3,2-*d*)oxacyclododecin-7,15-dione (spinosyn D)

Structural formulae



spinosyn A



spinosyn D

Empirical formulae

spinosyn A: $C_{41}H_{65}NO_{10}$
spinosyn D: $C_{42}H_{67}NO_{10}$

Relative molecular mass

spinosyn A: 732.0
spinosyn D: 746.0

CAS Registry number

spinosyn A: 131929-60-7
spinosyn D: 131929-63-0

CIPAC number

636

EEC number

434-300-1

Identity tests

HPLC retention time, positive-ion ESI LC-MS.

Physico-chemical properties of spinosad

Table 1. Physico-chemical properties of pure spinosad

Parameter	Value(s) and conditions	Purity %	Method	References
Vapour pressure, at 25ºC	Spinosyn A 3.0 x 10^{-8} Pa Spinosyn D 2.0 x 10^{-8} Pa	99.9 >99	OECD No. 104 EEC method A4, Knudsen-effusion/weight loss method	DAS A01, DAS A36
Melting point	Spinosyn A 84 to 99.5ºC Spinosyn D 161.5 to 170ºC Spinosyn A + D 110 to 123ºC	98.3 98.0 88.0 (A+D)	OECD No. 102 EEC method A1	DAS A03
Temperature of decomposition	Decomposition start temperature: 172ºC , 92% weight loss during heating to 400ºC	88.0 (A+D)	Thermal analysis	DAS A18
Solubility in water, at 20°C	Spinosyn A 290 mg/l at pH 5 235 mg/l at pH 7 16 mg/l at pH 9 Spinosyn D 28.7 mg/l at pH 5 0.332 mg/l at pH 7 0.053 mg/l at pH 9	98.3 99.9 99.8	OECD No. 105: flask method column elution method column elution method	DAS A20, DAS A37
Octanol/water partition coefficient, at 23°C	Spinosyn A Log P K_{ow} = 2.78 at pH 5 Log P K_{ow} = 4.01 at pH 7 Log P K_{ow} = 5.16 at pH 9 Spinosyn D Log P K_{ow} = 3.23 at pH 5 Log P K_{ow} = 4.53 at pH 7 Log P K_{ow} = 5.21 at pH 9	97.0 98.0	EPA/FIFRA subdiv. D 63.11, shake flask method	DAS A08, DAS A47
Hydrolysis characteristics, at 25°C	Spinosyn A No hydrolysis at pH 5 Half-life = 648 d. at pH 7 Half-life = 200 d. at pH 9 Spinosyn D No hydrolysis at pH 5 and 7 Half-life = 259 d. at pH 9	99.9 99.9	FIFRA guideline 161-1	DAS K05
Photolysis characteristics	Spinosyn A Half-life in dilute aqueous buffer calculated as 0.96 d. in summer sunlight (June-July, Greenfield, Indiana, 39.8°N) Spinosyn D Half-life in dilute aqueous buffer calculated as 0.84 d. in summer sunlight (June-July, Greenfield, Indiana, 39.8°N).	94.7 93.6	FIFRA Guideline 161-2	DAS K06

Table 1. Physico-chemical properties of pure spinosad

Parameter	Value(s) and conditions	Purity %	Method	References
Dissociation characteristics, at 20°C	Spinosyn A pKa = 8.1 Ka = 7.94 x 10^{-9} Spinosyn D pKa = 7.87 Ka = 1.35 x 10^{-8}	97.0 97.0	OECD guideline 112, capillary electrophoresis method	DAS A04, DAS A07

Table 2. Chemical composition and properties of technical spinosad (TC)

Manufacturing process, maximum limits for impurities ≥ 1 g/kg, 5 batch analysis data	Confidential information supplied and held on file by FAO. Mass balances were 98.0-101.6%, maximum percentage of unknowns was 0.3%.
Declared minimum spinosad content	850 g/kg (spinosyn A + spinosyn D)
Relevant impurities ≥ 1 g/kg and maximum limits for them	None
Relevant impurities < 1 g/kg and maximum limits for them	None
Stabilisers or other additives and maximum limits for them	None
Melting temperature of the TC	110 to 123ºC, (spinosyn A + spinosyn D)

Background information on toxicology/ecotoxicology

Dow AgroSciences confirmed that the toxicological and ecotoxicological data included in Annex 1, below, were derived from spinosad having impurity profiles similar to those referred to in Table 2, above.

Spinosad was evaluated for toxicology by the FAO/WHO JMPR in 2001. The JMPR concluded that spinosad has low acute toxicity. In studies with repeated doses, no acute toxicological alerts were observed that might indicate the need for establishing an acute reference dose (acute RfD). An ADI of 0–0.02 mg/kg bw was established on the basis of a NOAEL of 2.4 mg/kg bw per day in a 2-year study of toxicity and carcinogenicity in rats (Bond *et al*. 1995b, 1996d) and a 100-fold safety factor. The Swiss authorities assigned an ADI of 0-0.02 mg/kg bw/d, based on a NOEL of 2.4 mg/kg bw/d in the two year study on rats. This range is in agreement with the ADI assigned by the JMPR. The JMPR concluded that it was not necessary to assign an acute reference dose.

Maximum residue limits for spinosad have been set in Switzerland for a range of agricultural commodities. Estimated dietary intakes, based on typical food baskets, indicate that exposure of the population is expected to be well below the ADI.

The WHO hazard classification of spinosad is U, unlikely to present acute hazard in normal use (WHO 2004).

Formulations

The main formulation types available are suspension concentrates (SC) at 120 to 480 g spinosad/l, wettable powders (WP), water dispersible granules (WG), and

granules for direct application (GR). Spinosad may be co-formulated with other insecticide active ingredients.

The formulations are registered and sold in more than 60 countries throughout the world including the USA, Australia, Brazil, Canada, India, Japan, New Zealand, Switzerland and South Africa. Spinosad has been under EU evaluation as a new active substance since 2000, and meanwhile member state evaluations and provisional approvals have been granted in a number of EU countries including Italy, Netherlands, Spain, and the UK.

Methods of analysis and testing

Analytical methods for the identification and determination of spinosad content were adopted by CIPAC in 2005. The spinosad content (sum of spinosyns A and D) is determined by reversed-phase HPLC, using UV detection at 280 nm and external standardization. Definitive identification is by positive-ion ESI LC-MS, as no other technique is sufficiently specific.

Methods for the determination of impurities are based on reversed-phase HPLC with UV detection.

Test methods for determination of physico-chemical properties of technical spinosad were OECD/EC, while those for the formulations were CIPAC as indicated in the specifications.

Physical properties

The physical properties of the SC and GR formulations, the test methods and specification limits proposed, comply with the requirements of the manual (FAO/WHO 2002).

Containers and packaging

No special requirements for containers and packaging have been identified.

Expression of active ingredient

The active ingredient is expressed as spinosad, which is the sum of spinosyn A + spinosyn D, in g/kg or g/l at 20 ± 2ºC.

ANNEX 1

HAZARD SUMMARY PROVIDED BY THE PROPOSER

Note: The proposer provided written confirmation that the toxicological and ecotoxicological data included in the following summary were derived from spinosad having impurity profiles similar to those referred to in Table 2, above.

Table A. Toxicology profile of the spinosad technical material*, based on acute toxicity, irritation and sensitization

Species	Test	Duration and conditions	Result	References
Rat, m & f	Acute oral	OECD guideline 401 acute oral toxicity, 1987	$LD_{50} \geq 3738$ mg/kg bw (m) $LD_{50} > 5000$ mg/kg bw (f)	DAS B01, DAS B16
Mouse, m & f	Acute oral	OECD guideline 401 acute oral toxicity, 1987	LD_{50} >5000 mg/kg bw (m & f))	DAS B01, DAS B16
Rabbit, m & f	Acute dermal	OECD guideline 402 acute dermal toxicity, 1987	LD_{50} >5000 mg/kg bw (m & f)	DAS B07
Rat, m & f	Acute inhalation	EC test guideline (EC method B.2 acute toxicity (inhalation), 1984	LD_{50} >5.18 mg/l/4h	DAS B04
Rabbit, m & f	Skin irritation	OECD guideline 404 acute dermal irritation/corrosion, 1987	No irritation	DAS B05, DAS B30
Rabbit, m & f	Eye irritation	EC method B.5 acute toxicity (eye irritation), 1992	Mild transient irritation	DAS B09, DAS B32
Guinea pig, m	Skin sensitization	OECD guideline 406 skin sensitization, 1987, Buehler test	No sensitization	DAS B28
Guinea pig, f	Skin sensitization	EC test guideline (method B.6 skin sensitisation, 1996, Magnussen & Kligman test	No sensitization	DAS B33

* The spinosad TC used for the toxicity studies contained 771 g/kg A and 122 g/kg D, which was considered typical for spinosad, in terms of the ratio of spinosyns A and D and the content of other compounds.

Table B. Toxicology profile* of spinosad technical material based on repeated administration (sub-acute to chronic)**

Species	Test	Duration and conditions	Result	References
Rabbit, m & f	21-d dermal	OECD 410	NOAEL = 1000 mg/kg bw/d	DAS D05
Rat, m & f	14-d inhalation, 15-d recovery	OECD 412	NOAEL = 9.5 mg/m^3	DAS D22
Rat, m & f	13-week oral	OECD 408	NOAEL = 8.6 mg/kg bw/d LOAEL = 42.7 mg/kg bw/d	DAS D02
Rat, m & f	13-week oral	OECD 408	NOAEL = 7.7 mg/kg bw/d LOAEL = 39.1 mg/kg bw/d	DAS D20
Dog, m & f	13-week oral	OECD 409	NOAEL = 4.89 mg/kg bw/d LOAEL = 9.73 mg/kg bw/d	DAS D10
Mouse, m & f	3-month oral	OECD 408	NOAEL = 7.5 mg/kg bw/d LOAEL = 22.5 mg/kg bw/d	DAS D12
Dog, m & f	12-month oral	OECD 452	NOAEL = 2.68 mg/kg bw/d LOAEL = 8.22 mg/kg bw/d	DAS D03
Mouse, m & f	18-month oral, combined chronic toxicity and carcinogenicity	OECD 451	NOAEL = 11.4 mg/kg bw/d LOAEL = 32.7 mg/kg bw/d No carcinogenic potential	DAS I02, DAS I01, DAS I04, DAS I06
Rat, m & f	2-year oral, combined chronic toxicity and carcinogenicity	OECD 453	NOAEL = 2.4 mg/kg bw/d LOAEL = 11.4 mg/kg bw/d No carcinogenic potential	DAS I03, DAS I05
Rat	2-generation reproductive study	OECD 416	NOAEL = 10 mg/kg bw/d Reproduction NOAEL = 100 mg/kg bw/d	DAS F01
Rat	Teratogenicity	OECD 414	Maternal NOAEL = 50 mg/kg bw/d Developmental NOAEL = 200 mg/kg bw/d No teratogenic potential	DAS F03
Rabbit	Teratogenicity	OECD 414	Maternal NOAEL = 10 mg/kg bw/d Developmental NOAEL = 50 mg/kg bw/d No teratogenic potential	DAS F05
Rat, m & f	Neurotoxicity	OECD 424	No evidence of neurotoxicity in acute, sub-chronic and chronic studies	DAS B24, DAS I10, DAS D04

In addition to the data presented in Table B, the manufacturer provided data from a 28-day oral toxicity study in rats, in which the toxicity of a spinosyn A + D mixture was compared with that of spinosyn A (96.2%) and spinosyn D (93.0%). The

* In addition to the data presented, the toxicity of a spinosyn A + D mixture was compared with that of spinosyn A (96.2%) and spinosyn D (93.0%). Spinosyn A and spinosyn D were found to display similar toxicity in mammalian systems, with spinosyn A being slightly more toxic than spinosyn D at equivalent (expressed as mg/kg bw/d) dose levels (DAS D09).

** The spinosad TC used for the toxicity studies contained 771 g/kg A and 122 g/kg D, which was considered typical for spinosad, in terms of the ratio of spinosyns A and D and the content of other compounds.

mixture, spinosyn A and spinosyn D were found to display similar toxicity in mammalian systems, with spinosyn A being slightly more toxic than spinosyn D at equivalent dose levels (expressed as mg/kg bw/d.)

Table C. Mutagenicity profile of spinosad technical material* based on *in vitro* and *in vivo* tests

Species	Test	Conditions	Result	Reference
S. typhimurium TA98, TA100, TA1535,TA1537 and *E. coli* WP2uvrA	Ames test, pre-incubation *in vitro* , plate incorporation *in vitro,* OECD 471	50 to 5000 µg/plate	Negative	DAS E06
Mouse lymphoma cells, L5178Y	Mammalian cells *in vitro*, gene mutations, TK assay, OECD 476	1 to 50 µg/ml	Negative	DAS E04
Chinese hamster ovary (CHO-WB_L) cells	mammalian cells *in vitro*, cytogenic assay, OECD 473	20 to 100 µg/ml	Negative	DAS E01
Rat hepatocytes	mammalian cells *in vitro*, unscheduled DNA synthesis, OECD 482	0.1 to 5 µg/ml	Negative	DAS E02
Mouse	*In vitro* micronucleus test, OECD 474	2 daily oral doses: 500, 1000, 2000 mg/kg bw; sacrifice at 24 h after last dose	Negative	DAS E03

Based on these results, spinosad was considered to be non-genotoxic.

* The spinosad TC used for the toxicity studies contained 771 g/kg A and 122 g/kg D, which was considered typical for spinosad, in terms of the ratio of spinosyns A and D and the content of other compounds.

Table D. Ecotoxicology profile* of spinosad technical material** or formulated product

Species	Test	Duration and conditions	Result	Reference
Daphnia magna (water flea)	Acute toxicity, static	48 h, FIFRA 72-2 & OECD 202 Part 1 (20 ± 2°C)	EC_{50} >1.0 mg as/l	DAS J38
Daphnia magna (water flea)	Acute toxicity, static, formulation 480SC	48 h, OECD 202 Part 1 (20 ± 2°C)	EC_{50} = 9.1 mg as/l	DAS MJ06
Daphnia magna (water flea)	Chronic toxicity	21 d, FIFRA 72-4 & OECD 202 Part 2 (20 ± 2°C)	NOEC = 0.0012 mg as/l (flow through) NOEC = 0.0080 mg as/l (semi-static)	DAS J15
Chironomus riparius (midge)	Chronic toxicity, static	25 d, OECD 219 (20 ± 0.5°C)	NOEC = 0.0016 mg as/l	DAS J51
Oncorhynchus mykiss (rainbow trout)	Acute toxicity, static	96 h, FIFRA 72-1 & OECD 203 ,12.5 ± 0.5°C	LC_{50} = 27 mg as/l	DAS J06
Lepomis macrochirus (bluegill sunfish)	Acute toxicity, static	96 h, FIFRA 72-1 & OECD 203 (21-22.1°C)	LC_{50} = 5.94 mg as/l	DAS J27
Cyprinus carpio	Acute toxicity, flow through	96 h, FIFRA 72-1 & OECD 203 (24.5-25.5°C)	LC_{50} = 4 mg as/l	DAS J05
Cyprinus carpio	Acute toxicity, static	96 h, OECD 203 (22 ± 2°C), 480 g/l SC	LC_{50} >49 mg as/l	DAS MJ16
Oncorhynchus mykiss (rainbow trout)	Early life-stage toxicity, flow through	80 day, FIFRA 72-4(a) & OECD 210 (12 ± 2°C)	NOEC = 0.5 mg as/l	DAS J12
Navicula pelliculosa (alga)	Static water	120 h, FIFRA 123-2 & OECD 201 (22 ± 1°C)	EC_{50} = 0.079 mg as/l	DAS J19
Navicula pelliculosa (alga)	Static water, formulation 480SC	120 h, OECD 201 (22 ± 1°C)	EC_{50} = 0.35 mg as/l	DAS MJ17
Anabaena flos-aquae (alga)	Static water	120 h, FIFRA 123-2 & OECD 201 (24 ± 2°C)	EC_{50} = 6.1 mg as/l	DAS J17
Selenastrum capricornutum (alga)	Static water	72 h, FIFRA 123-2 & OECD 201 (24 ± 2°C)	EC_{50} = 56 mg as/l	DAS J30
Lemna gibba (higher plant)	Static water	14 d, FIFRA 123-2 & OECD 221 (25.3 ± 0.15°C)	EC_{50} = 6.6 mg/l	DAS J16
Eisenia foetida (earthworm)	Acute toxicity	14 d, 20 ± 2°C	LC_{50} >970 mg as/kg dry soil	DAS J21

* Data were also provided on the effects of spinosad on non-target insects, including larvae of the hoverfly *Episyrphus balteatus* (DAS MJ25), the foliar-active predator *Chrysoperla carnea* (DAS MJ24), the parasitoid wasp *Aphidius colemani* (DAS MJ22) and the carabid beetle *Poecilus cupreus* (DAS MJ23).

** The spinosad TC used for the toxicity studies contained 771 g/kg A and 122 g/kg D, which was considered typical for spinosad, in terms of the ratio of spinosyns A and D and the content of other compounds.

Table D. Ecotoxicology profile* of spinosad technical material or formulated product**

Species	Test	Duration and conditions	Result	Reference
Apis mellifera (honey bee)	Oral exposure	OECD 213	LD_{50} = 0.057 µg/bee (spinosad) LD_{50} = 0.049 µg as/bee (480SC)	DAS J47
Apis mellifera (honey bee)	Contact exposure	OECD 214	LD_{50} = 0.0036 µg/bee (spinosad) LD_{50} = 0.050 µg as/bee (480SC)	DAS J20
Apis mellifera (honey bee)	Acute oral	EPPO 170	LD_{50} = 0.0057 µg/bee (spinosad) LD_{50} = 0.049 µg as/bee (480SC)	DAS MJ14
Colinus virginianus (bobwhite quail)	Acute oral toxicity	14 d, FIFRA 71-1	LD_{50} >2000 mg/kg bw	DAS J24
Colinus virginianus (bobwhite quail)	Short-term dietary toxicity	5 d, FIFRA 71-2 & OECD 205, 88% A+D	LC_{50} >5253 mg as/kg diet	DAS J26
Colinus virginianus (bobwhite quail)	Reproduction study	21 week, FIFRA 71-4(a) & OECD 206	NOEC = 550 mg/kg diet	DAS J01
Anas platyrhynchos (mallard duck)	Acute oral toxicity	14 d, FIFRA 71-1	LD_{50} >2000 mg/kg bw	DAS J23
Anas platyrhynchos (mallard duck)	Short-term dietary toxicity	5 d, FIFRA 71-2 & OECD 205	LC_{50} >5156 mg as/kg diet	DAS J25
Anas platyrhynchos (mallard duck)	Reproduction study	21 week, FIFRA 71-4(b) & OECD 206	NOEC = 550 mg/kg diet	DAS J02

The mode of action of spinosad is via activation of the nicotinic acetylcholine receptor, combined with effects on the GABA-receptor, leading to neuromuscular fatigue and paralysis in sensitive insect pests. None of the tests on mammals showed any evidence of symptoms reflecting the mode of action in target insects.

ANNEX 2. REFERENCES

Dow AgroSciences document number	Year and title of report
DAS A01	1991. Vapour Pressure of Compound 232105 measured by the Knudsen-Effusion/Weight Loss Method.
DAS A03	1994. Series 63: Physical and Chemical Characteristics of the Technical Grade of Active Ingredient XDE-105.
DAS A04	1994. Determination of the Dissociation Constant of LY-232105.
DAS A07	1994. Determination of the Dissociation Constant of XDE-105 Factor D.
DAS A08	1994. Octanol/Water Partition Coefficient Determinations of Compound 232105.
DAS A18	1997. Thermogravimetric Analysis of Spinosad and Evolved Gas Analysis by Gas Chromatography/Mass Spectrometry.
DAS A20	1993. Solubility of Compound 232105 in pH = 9 Buffer Solution for Registration.
DAS A36	1991. Vapour Pressure of Compound 275043 Measured by the Knudsen-Effusion/Weight Loss Method.
DAS A37	1994. Solubility of Compound 275043 in Water and Buffer Solutions of pH = 5, 7, and 9 for Registration.
DAS A47	1994. Octanol/Water Partition Coefficient Determinations of Compound 275043.
DAS B01	1994. XDE-105: Acute Oral Toxicity Study in Fischer 344 Rats and CD-1 Mice.
DAS B04	1992. The Acute Inhalation Toxicity in the Fischer 344 Rat of Technical XDE-105.
DAS B05	1994. XDE-105: Primary Dermal Irritation Study in New Zealand White Rabbits.
DAS B07	1994. XDE-105: Acute Dermal Toxicity Study in New Zealand White Rabbits.
DAS B09	1994. XDE-105: Primary Eye Irritation Study in New Zealand White Rabbits.
DAS B16	1996. DE-105: Acute Oral Toxicity Study in Fischer 344 Rats and CD-1 Mice.
DAS B24	1994. XDE-105: Acute Neurotoxicity Study in Fischer 344 Rats
DAS B28	1996. A Skin Sensitization Study of DE-105 in Guinea Pigs (maximisation Test).
DAS B30	1999. Spinosad (Spinosyn A&D, 50:50 Mixture): Acute Dermal Irritation Study in New Zealand White Rabbits.
DAS B32	1999. Spinosad (Spinosyn A&D, 50:50 Mixture): Acute Eye Irritation Study in New Zealand White Rabbits.
DAS B33	1999. Spinosad (Spinosyn A&D, 50:50 Mixture): Dermal Sensitisation Potential Study in Hartley Albino Guinea Pigs.
DAS D02	1994. XDE-105: 13-week Dietary Toxicity and 4-week Recovery Studies in Fischer 344 Rats.
DAS D03	1995. XDE-105: 12 Month Oral Chronic Toxicity Study in Dogs.
DAS D04	1993. XDE-105: 13-Week Dietary Toxicity 4-week Recovery and 13-week Neurotoxicity Studies in Fischer 344 Rats (Neurotoxicity Portion).
DAS D05	1994. XDE-105: Probe and 21-day Repeated Dose Dermal Toxicity Study in New Zealand White Rabbits.
DAS D09	1994. XDE-105: Factor A and Factor D:28-day Dietary Toxicity Study in Fischer 344 Rats.
DAS D10	1994. XDE-105: 13-Week Oral Subchronic Toxicity Study in Dogs.
DAS D12	1992. Subchronic Toxicity Study in CD-1 Mice Administered XDE-105 in the Diet for 3 Months.
DAS D20	1999. Spinosad (50% Spinosyn A and 50% Spinosyn D): 13-Week Dietary Toxicity Study in Fischer Rats.
DAS D22	1999. Spinosad technical (DE-105): 14-day Nose only Aerosol Inhalation Toxicity and 2-week Recovery studies in Fischer 344 Rats.
DAS E01	1992. The Effect of XDE-105 on the In Vitro Induction of Chromosome Aberrations in Chinese Hamster Ovary Cells.
DAS E02	1992. The Effect of XDE-105 on the Induction of Unscheduled DNA Synthesis in Primary Cultures of Adult Rat Hepatocytes.

DAS E03 1992. The Effect of XDE-105 on the In Vivo Induction of Micronuclei in Bone Marrow of ICR Mice.

DAS E04 1992. The Effect of XDE-105 on the Induction of Forward Mutation at the Thymidine Kinase Locus of L5178Y Mouse Lymphoma Cells.

DAS E06 1996. Mutagenicity Test on XDE-105 in the Salmonella - Escherichia coli /Mammalian Microsome Reverse Mutation Assay Preincubation Method with a Confirmatory Assay).

DAS F01 1994. XDE-105: Two Generation Dietary Reproduction Study in Sprague-Dawley Rats.

DAS F03 1993. XDE-105: Oral Gavage Teratology Study in Sprague-Dawley Rats.

DAS F05 1994. XDE-105: Oral Gavage Teratology Study in New Zealand White Rabbits.

DAS I01 1996. XDE-105: 18 Month Dietary Oncogenicity Study in CD-1 Mice.

DAS I02 1995. XDE-105: 18 Month Dietary Oncogenicity Study in CD-1 Mice.

DAS I03 1995. XDE-105: Two-year Chronic Toxicity Chronic Neurotoxicity and Oncogenicity Study in Fischer 344 Rats.

DAS I04 1996. XDE-105: 18-Month Dietary Oncogenicity Study in CD-1 Mice (Report Supplement).

DAS I05 1996. XDE-105: Two-year Chronic Toxicity Chronic Neurotoxicity and Oncogenicity Study in Fischer 344 Rats-Supplemental Statistical Analysis of Histopathology Data.

DAS I06 1996. XDE-105: 18 Month Dietary Oncogenicity Study in CD-1 Mice-Supplemental Statistical Analysis of Histopathology Data.

DAS I10 1995. XDE-105: Chronic Neurotoxicity Study in Fischer 344 Rats.

DAS J01 1994. XDE-105 Insecticide: A Reproduction Study with the Northern Bobwhite (Colinus virginianus).

DAS J02 1994. XDE-105 Insecticide: A Reproduction Study with the Mallard (Anas platyrhynchos).

DAS J05 1994. Evaluation of the Acute Toxicity of XDE-105 Insecticide to the Japanese Carp Cyprinus carpio.

DAS J06 1993. Evaluation of the Acute Toxicity of XDE-105 Insecticide to the Rainbow Trout. Oncorhynchus mykiss Walbaum.

DAS J12 1993. Evaluation of the Toxicity of XDE-105 Insecticide to the Early Life Stages of the Rainbow Trout Oncorhynchus mykiss Walbaum.

DAS J15 1995. Evaluation of the Chronic Toxicity of XDE-105 Insecticide to the Daphnid Daphnia magna Straus following flow-through exposure.

DAS J16 1994. The Toxicity of XDE-105 Insecticide (Lot # ACD13651) to the Aquatic Plant Duckweed Lemna gibba G-3.

DAS J17 1993. The Toxicity of XDE-105 Insecticide to Anabaena flos-aquae.

DAS J19 1994. The Toxicity of XDE-105 Insecticide to Navicula pelliculosa.

DAS J20 1992. XDE-105 Insecticide: An Acute Contact Toxicity Study with the Honey Bee.

DAS J21 1993. Acute Toxicity of XDE-105 Insecticide to the Earthworm Eisenia foetida.

DAS J23 1992. The Toxicity of XDE-105 to Mallards in a 14-Day Acute Oral Study.

DAS J24 1992. The Toxicity of XDE-105 to Bobwhite in a 14-Day Acute Oral Study.

DAS J25 1992. The Toxicity of XDE-105 to Juvenile Mallards in a 5 -Day Dietary Study.

DAS J26 1992. The Toxicity of XDE-105 to Juvenile Bobwhite in a 5-Day Dietary Study.

DAS J27 1992. The Acute Toxicity of XDE-105 to Bluegill (Lepomis macrochirus) in a Static Test System.

DAS J30 1992. Toxicity of XDE-105 to a Freshwater green Alga (Selenastrum capricornutum) in a 7-Day Static Test System.

DAS J38 1992. The Acute Toxicity of XDE-105 to Daphnia magna in a Static Test System.

DAS J47 1998. Spinosad Technical Acute Toxicity to Honey Bees (Apis mellifera).

DAS J51 1999. DE-105 - The Chronic Toxicity to Midge (Chironomus riparius) Under Static Conditions.

DAS K05 1994. Hydrolysis of XDE-105 Factors A and D in Aqueous Buffer.

DAS K06 1994. Photodegradation of XDE-105 Factors A & D in pH 7 Buffer.

DAS MJ06 1996. Evaluation of the Acute Toxicity of NAF-85 to the Daphnid Daphnia magna Straus.

DAS MJ14 1998. NAF-85 (480 g/L SC of spinosad) Acute toxicity to honey bees.

DAS MJ16 1999. NAF-85, Acute Toxicity to Fish.

DAS MJ17 1999. NAF-85 Algal Growth Inhibition Assay (Navicula pelliculosa).

DAS MJ22 1999. Extended Laboratory Bioassay to Evaluate the Effects of Spinosad (Formulated as NAF-85, 480 g/L SC) on the Parasitoid Aphidius colemani.

DAS MJ23 1999. An Extended Laboratory Test to Evaluate the Side-effects of Repeated Applications of Spinosad(Formulated as NAF-85, 480 g/L SC) on the Carabid Beetle Poecilus Cupreus.

DAS MJ24 1999. An Extended Laboratory Test to Evaluate the Side-effects of the Insecticide Spinosad 480 SC(NAF-85), a suspension Concentrate Formulation Containing 480 g/L DE-105, on the Foliar-Active Predator, Chrysoperla Carnea.

DAS MJ25 1991. Testing of an Experimental Insecticide, XDE-105, for Side Effects to Larvae of the Hoverfly, Episyrphus balteatus with Reference to BBA guideline VI, 23-2.1.7.

JMPR 2001 WHO/PCS/02.1 , Joint FAO/WHO Meeting on Pesticide Residues, Evaluations 2001, Part II – Toxicological, pp. 183-227.

Sparks *et al*. 1998 Sparks, T.C., Thompson, G.D., Kirst, H.A., Hertlein, M.B., Larson, L.L., Worden, T.V., Thibault, S.T. Biological activity of the spinosyns, new fermentation derived insect control agents, on tobacco budworm (Lepidoptera: Noctuidae) larvae. *J. Econ. Entomol*. **91**, 1277-1283 (1998).

WHO 2004 The WHO recommended classification of pesticides by hazard and guidelines to classification 2002-2004. WHO/PCS/01.5. WHO, Geneva.