

**DATA EVALUATION RECORD  
HONEY BEE - ACUTE CONTACT & ORAL LC<sub>50</sub> TEST  
§141-1**

1. **CHEMICAL**: Orthosulfamuron PC Code No.: 108209

2. **TEST MATERIAL**: IR5878 Purity: 98.54 ± 0.51%

3. **CITATION**:

Author: S. Schmitzer

Title: Laboratory Testing for Toxicity (Acute Contact and Oral LD<sub>50</sub>) of IR5878 on Honey Bees (*Apis Mellifera* L.)

Study Completion Date: July 10, 2001 (Revised March 3, 2003)

Laboratory: Institute for Biological Analysis and Consulting  
IBACON GmbH  
Rossdorf, Germany

Sponsor: ISAGRO SpA,  
Centro Uffici San Siro  
Milano, Italy

Laboratory Report ID: 8225036

DP Barcode: D304186

MRID No.: 46219046

4. **REVIEWED BY**: Rebecca Bryan, Staff Scientist, Dynamac Corporation

**Signature:**

**Date:** 12/21/04

**APPROVED BY**: Gregory S. Hess, Staff Scientist, Dynamac Corporation

**Signature:**

**Date:** 12/28/04

5. **APPROVED BY**: Christopher J. Salice

**Signature:**



**Date:** 7/28/06



**6. STUDY PARAMETERS:**

**Scientific Name of Test Organism:** *Apis mellifera*  
**Age or Size of Test Organism at Test Initiation:** 4-6 weeks old  
**Type of Concentrations:** Nominal  
**Definitive Study Duration:** 48 hours

**7. CONCLUSIONS:**

The honey bee, *Apis mellifera* L., was exposed to IR5878 (Orthosulfamuron) for 48 hours in both oral and contact toxicity tests. In the oral and contact tests, the nominal test concentration was 100 µg a.i./bee. By 48 hours in the oral test, 4% mortality was observed in the 100 µg a.i./bee treatment group, compared to 8% solvent control mortality. By 48 hours in the contact test, 10% mortality was observed in the 100 µg a.i./bee treatment group, compared to 0% negative control and 2% solvent control mortality.

**The LD<sub>50</sub> value for the oral test was >109.4 µg a.i./bee. The LD<sub>50</sub> value for the contact test was >100 µg a.i./bee. As a result, IR5878 (Orthosulfamuron) is categorized as practically non-toxic to honeybees on both an acute oral and contact basis.**

**This acute contact study is classified as ACCEPTABLE.** This study is scientifically sound and it satisfies the EFED guideline requirements for a contact toxicity test with honey bees (Subdivision L, §141-1 or 850.3020). **The acute oral study is scientifically sound and is classified as SUPPLEMENTAL.**

**Reported Statistical Results - Oral Test:**

LD<sub>50</sub>: >109.4 µg a.i./bee      95% C.I.: N/A  
NOAEC: 109.4 µg a.i./bee      Probit Slope: N/A

**Reported Statistical Results - Contact Test:**

LD<sub>50</sub>: >100 µg a.i./bee      95% C.I.: N/A  
NOAEC: 100 µg a.i./bee      Probit Slope: N/A

**8. ADEQUACY OF THE STUDY:**

**A. Classification:** This acute contact study is classified as ACCEPTABLE. This study is scientifically sound and it satisfies the EFED guideline requirements for a contact toxicity test with honey bees (Subdivision L, §141-1 or 850.3020). The acute oral study is scientifically sound and is classified as SUPPLEMENTAL.

**B. Rationale:** This acute oral study is scientifically sound and is classified as Supplemental because the study is a non-guideline study and does not fulfill an OPP guideline requirement.

**C. Repairability:** N/A

**9. GUIDELINE DEVIATIONS:**

No deviations noted.

**10. SUBMISSION PURPOSE:** This study was submitted to provide data on the acute oral and contact toxicity of IR5878 (Orthosulfamuron) to honeybees for the purpose of chemical registration.

**11. MATERIALS AND METHODS:****A. Test Organisms**

<b>Species:</b> Species of concern ( <i>Apis mellifera</i> , <i>Megachile rotundata</i> , or <i>Nomia melanderi</i> )	<i>Apis mellifera</i>
<b>Age at beginning of test:</b>	4-6 weeks old
<b>Supplier:</b>	Laboratory colonies (IBACON)
<b>All bees from the same source?</b>	Yes

**B. Test System**

<b>Cage size adequate?</b>	The cages were stainless steel chambers with a removable glass sheet front and perforated bottom (1 mm holes). Cage dimensions were 10 x 8.5 x 5.5 cm.
<b>Lighting:</b>	Continuous darkness (except during observations).
<b>Temperature:</b>	25°C
<b>Relative humidity:</b>	40-46%

### C. Test Design

<b>Range finding test?</b>	No range finding study was conducted.
<b>Reference toxicant test?</b>	The reference toxicant was dimethoate, Perfekthion EC (nominal 400 g/L). In the <u>oral test</u> and <u>contact test</u> , the concentration of dimethoate was 0.02 µg a.i./bee.
<b>Method of administration:</b>	<u>Oral test</u> : the test substance was mixed with 1 part solvent (acetone) and 19 parts commercial syrup (30% Saccharose, 31% Glucose, and 39% Fructose). The food and test substance were offered in syringes.  <u>Contact test</u> : 5 µL of test substance in solvent (acetone) was applied to the ventral thorax of each bee using a Burkard-Applicator.
<b>Nominal doses:</b>	<u>Oral test</u> : 100 µg a.i./bee  <u>Contact test</u> : 100 µg a.i./bee
<b>Controls:</b> Negative control and/or diluent/solvent	<u>Oral test</u> : solvent (acetone) control

control	<u>Contact test</u> : negative control and solvent (acetone) control
<b>Number of colonies per group:</b>	5 replicates; 10 bees/replicate
<b>Solvent:</b> The following solvents: acetone, dimethylformamide, triethylene glycol, methanol, ethanol.	Acetone
<b>Feeding:</b>	<u>Oral test</u> : After treated solutions were consumed (<5 hours), bees were supplied with syrup (30% Saccharose, 31% Glucose, and 39% Fructose), <i>ad libitum</i> .  <u>Contact test</u> : A syrup (30% Saccharose, 31% Glucose, and 39% Fructose) was provided <i>ad libitum</i> .
<b>Observations period:</b>	48 hours

**12. REPORTED RESULTS:**

<b>Quality assurance and GLP compliance statements were included in the report?</b>	Yes
<b>Control performance:</b>	<u>Oral test</u> : 8% solvent control mortality by 48 hours. <u>Contact test</u> : 0% negative control and 2% solvent control mortality by 48 hours.
<b>Raw data included:</b>	Data were provided.
<b>Signs of toxicity (if any) were described?</b>	No sublethal effects were recorded.

**Mortality - Oral Test**

Test Substance (IR5878, Orthosulfamuron)				
Solvent Control	50	4	8	8
100 (109.4)	50	0	0	4
Toxic Standard (Dimethoate):				
0.2 (0.23)	50	36	88	94

Observations: By 48 hours, 4% mortality was observed in the nominal 100 µg a.i./bee treatment group, compared to 8% solvent control mortality.

**Mortality - Contact Test**

Test Substance (IR5878, Orthosulfamuron)				
Negative Control	50	0	0	0
Solvent Control	50	0	0	2
100	50	0	6	10
Toxic Standard (Dimethoate):				
0.2	50	20	76	78

Observations: By 48 hours, 10% mortality was observed in the nominal 100 µg a.i./bee

treatment group, compared to 0% negative control and 2% solvent control mortality.

Statistical method: The LD<sub>50</sub> values were visually estimated based on mortality and sublethal effects data in the oral and contact toxicity tests.

**Reported Statistical Results - Oral Test:**

LD<sub>50</sub>: >109.4 µg a.i./bee      95% C.I.: N/A  
NOAEC: 109.4 µg a.i./bee      Probit Slope: N/A

**Reported Statistical Results - Contact Test:**

LD<sub>50</sub>: >100 µg a.i./bee      95% C.I.: N/A  
NOAEC: 100 µg a.i./bee      Probit Slope: N/A

**13. VERIFICATION OF STATISTICAL RESULTS:**

Statistical Method: The LD<sub>50</sub> and NOAEC values for the oral and contact test were visually determined because mortality was ≤10% in all treatment and control groups at 48-hours for both the contact and oral toxicity tests.

**Results - Oral Test:**

LD<sub>50</sub>: >109.4 µg a.i./bee      95% C.I.: N/A  
NOAEC: 109.4 µg a.i./bee      Probit Slope: N/A

**Results - Contact Test:**

LD<sub>50</sub>: >100 µg a.i./bee      95% C.I.: N/A  
NOAEC: 100 µg a.i./bee      Probit Slope: N/A

**14. REVIEWER'S COMMENTS:**

The reviewer's conclusions were identical to the study authors. **The LD<sub>50</sub> value for the oral test was >109.4 µg a.i./bee. The LD<sub>50</sub> value for the contact test was >100 µg a.i./bee. As a result, IR5878 (Orthosulfamuron) is categorized as practically nontoxic to honeybees on both an acute oral and contact basis.**

**15. REFERENCES:**

- Barrett, K.L., Grandy, N., Harrison, E.C., Hassan, S., and Oomen, P. 1994: SETAC-Guidance document on regulatory testing procedures for pesticides with non-target arthropods. 28-30 March 1994, IAC Wageningen, The Netherlands
- Chemikaliengesetz der Bundesrepublik Deutschland (ChemG), Anhang 1, in der Fassung der Bekanntmachung vom 25. Juli 1994 (BGBl. IS. 1703) mit Änderungen vom 27. September 1994 (BGBl. IS. 2705) und 14 Mai 1997 (BGBl. IS. 1060)
- Commission Directive 96/12/EC. 1996. amending Council Directive 91/414/EEC. Official Journal of The European Communities No. L 65: 20-37
- ICPBR (2000) Hazards of pesticides to bees, 7<sup>th</sup> International Symposium of the ICPBR Bee Protection Group, Avignon (France), 07-09 September 1999; Les Colloques d'INRA (in press)
- EPA 1996: OPPTS 850.3020, Honey Bee Acute Contact Toxicity, United States Environmental Protection Agency, EPA 712-C-96-147, April 1997.
- EPPO 1992: Guideline on test methods for evaluating the side-effects of plant protection products on honeybees Bulletin OEPP/EPPO Bulletin 22, 203-215 1992, No. 170.
- OECD Principles of Good Laboratory Practice, adopted by Council on 26<sup>th</sup> November 1997 [C(97)186/Final], Environment Directorate, Organisation for Economic Co-operation and Development, Paris 1998