ACUTE CONTACT TOXICITY TO TERRESTRIAL INVERTEBRATES (HONEY BEE)

TEST SUBSTANCE

Identity: Perfluorooctanesulfonate; may also be referred to as PFOS or FC-95. (1-Octanesulfonic acid, 1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-heptadecafluoro-, potassium salt, CAS # 2795-39-3)

Remarks field: The 3M production lot number was 217. The test substance is a white powder. Sample was stored AT 16-20^{o}C prior to testing. Purity determined to be 86.9% by LC/MS, ^{1}H-HMR, ^{19}F-NMR and elemental analyses techniques.

METHOD

Method: USEPA OPPTS 850.3020 (draft), OECD Guideline 214, EPPO Guideline 170
Test type: Acute Contact
GLP: Yes
Year Completed: 2001
Species: *Apis mellifera L.*
Analytical monitoring: None – nominal concentrations
Test honey bee source: Obtained from colony number 32 belonging to the Central Science Laboratory (CSL), Sand Hutton, York, UK, National Bee Unit.
Test honey bee age at study initiation: Young adult
Test honey bee type: Worker honey bees, free of acarine, nosema and amoeba.
Varroacide treatment: None within the 4 weeks prior to test initiation.

Test conditions

Humidity: 65% $\pm$ 5%
Temperature: 25 $\pm$ 2^{o}C
Lighting: Conducted in darkness

Stock and test solutions preparation:

Test substance: Stock solution prepared in analytical grade acetone to a final concentration of 90.4 µg PFOS/µL (nominal concentration). Final test concentrations prepared in acetone from dilutions of this solution.
Reference toxicant: Stock solution of dimethoate was prepared in deionized water containing 1 g/L Triton X-100 to a final

concentration of 3.0 µg/µL. Final test concentrations prepared from dilutions of this solution.

Stability of the test chemical solution: A dispersion test was carried out on an 86 µg PFOS/µL acetone solution before the toxicity study was performed. The homogeneity of the mixture was assessed after 2 hours. The test item formed a clear solution on mixing; after 2 hours at room temperature, slight sediment was noted. For the toxicity test, all solutions were re-mixed prior to use. The contract laboratory considered the solutions of the test doses to be homogenous for the purpose of administration.

Exposure vessels: Clean, well-ventilated, inverted petri dishes, measuring approximately 9 cm in diameter.

Feeding: 50% w/v aqueous sucrose solution, continuously available

Number of replicates: Three

Number of bees per replicate: Ten

Negative control: Undosed

Solvent control: Acetone

Reference substance: Dimethoate

Reference substance control: Triton X-100

Number of concentrations: five plus a negative and a solvent control

Dose administration: The bees were anaesthetized with carbon dioxide immediately before dosing and gently tipped out onto filter paper and counted into the petri dish cage (drones were discarded). Each bee was dosed on the thorax with a 1 µL drop of a given test item concentration or 1µL acetone before being placed into the test chamber.

Dose frequency: Once

Element basis: Mortality

RESULTS

Nominal concentrations: Negative control, acetone control (1.0 µ/bee), 1.93, 4.24, 9.30, 20.5, 45, µg PFOS/bee

Element value and 95% confidence interval :

24-hour LD_{50} = 38.9 (28.2 – 71.2) µg/bee
48-hour LD_{50} = 10.4 (8.2 – 13.0) µg/bee
72-hour LD_{50} = 6.0 (4.7 – 7.6) µg/bee
96-hour LD_{50} = 4.78 (3.8 – 5.8) µg/bee

96-hour NOEL = 1.93 µg/bee

All element values based on nominal concentrations

Statistical Evaluation: Probit mortality plotted against the logarithm of dose using the contract laboratory Probit 1 package. A least-squares

regression (Finney 1971) was fitted to these. The NOELs were estimated using Student's t-test ($p<0.05$)

Biological observations: There was significant mortality at all doses above 1.93 µg/bee with a steep dose response between 4.24 and 9.30 µg/bee

Cumulative percent mortality:

Nominal Test Conc., µg/bee	4-hours	24-hours	48-hours	72-hours	96-hours
Negative Control	0	0	3.3	3.3	3.3
Solvent Control	3.3	3.3	3.3	3.3	3.3
1.93	0	6.7	6.7	13	13
4.24	0	0	13	37	37
9.30	0	6.7	40	63	90
20.5	0	40	93	97	100
45.0	0	50	93	100	100

Sub-lethal Effects – Percent Knockdown (K) or Stumbling (S):

Nominal Test Conc., µg/bee	4-hours	24-hours	48-hours	72-hours	96-hours
Negative Control	0	0	0	0	0
Solvent Control	0	0	0	0	0
1.93	3.3 (K)	0	0	0	0
4.24	0	0	0	0	0
9.30	0	0	0	0	3.3 (K)
20.5	0	3.3 (K)	0	3.3 (K)	0
45.0	0	3.3 (K)	3.3 (K)	0	0

Control response: satisfactory

Reference toxicant response: satisfactory - dimethoate 96-hour LD_{50} = 0.19 µg/bee

CONCLUSIONS

The potassium perfluorooctanesulfonate 96-hour contact LD_{50} for the honey bee was determined to be 4.78 µg/bee with a 95% confidence interval of 3.8 – 5.8. The 96-hour no observed effect level was 1.93 µg/bee. The dose response was steep between 4.24 and 9.30 µg/bee.

Submitter: 3M Company, Environmental Laboratory, P.O. Box 33331, St. Paul, Minnesota, 55133

DATA QUALITY

Reliability: Klimisch ranking 1

REFERENCES

This study was conducted at Central Science Laboratory, Sand Hutton, York, UK, under contract by Wildlife International, Ltd, Easton, MD at the request of the 3M Company, Lab Request Number U2723, 2001.

OTHER

Last changed: 5/1/01