Imidacloprid: Human Health and Ecological Risk Assessment APPENDICES

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Appendix 1: Toxicity to mammals.

A1 Table 1: Acute Oral Toxicity Studies, Experimental Mammals

Species	Exposure	Response	Reference
GAVAGE			
Technical Grade			
Rats, Wistar (Bor: WSIWSPF-Cpb), 5/sex/dose, males (7-8 weeks old, 167-187 g), females (10-12 weeks old, 168 - 194 g)	94.2% a.i., 50, 100, 250, 315, 400, 450, 500 and 1800 mg/kg bw. 14-day observation period	LD_{50}: Males: 424 mg/kg bw Females: 450-475 mg/kg bw NOAEL (mortality): 400 mg/kg bw NOAEL (toxicity): 50 mg/kg bw LOAEL (toxicity): 100 mg/kg bw Signs of toxicity: Apathy, labored breathing, accelerated breathing, decreased mobility, staggering gait, trembling, and spasms. Reversible after 2-6 days.	Bomann 1989a MRID 42055331 Used by U.S. EPA/OPP/ HED 2007a (Table A.1) to classify technical grade imidacloprid at Category II.
Mice, Bor: NMRI-SPF (Han), 5 male (4 weeks old, 21 - 25 g), and 5 female (4 - 5 weeks old, 20 - 24 g) per dose group	Technical grade imidacloprid (94.2% a.i.) Doses: 10, 71, 100, 120, 140, 160, 250 mg/kg bw. 14-day observation period	LD_{50}: Males: 131 mg/kg bw Females: 168 mg/kg bw NOAEL (mortality): 71 mg/kg bw NOAEL (toxicity): 10 mg/kg bw Signs of toxicity: Lethargy, labored or transient labored breathing, decreased “motility”, transient staggering gait, transient trembling and transient spasms. No gross pathology in survivors. No effects on body weight gain in any dose group. Pale or dark spleens and livers; patchy distended lungs in animals which died.	Bomann 1989b MRID 42256324

Appendix 1: Toxicity to mammals *(continued)*

Species	Exposure	Response	Reference
Mice, Swiss albino,	Technical grade imidacloprid (94.2% a.i.) Gavage in corn oil. 24 hour observation period.	LD_{50}: 149.76 mg/kg bw Sublethal effects include increases in the activities of several antioxidant enzymes and depletion of glutathione.	El-Gendy et al. 2010
Formulations		*Doses as formulation unless otherwise specified.*	
Rat, Sprague-Dawley (Sas:CD (SD)BR), 5 male (8 weeks old) and 5 female (10 weeks old)	Single dose: 2.5 a.i.% granular formulation in deionized water (10 ml/kg)	No mortality, signs of toxicity, or pathology. NOAEL >4820 mg formulation/kg body weight (>120.5 mg a.i./kg bw)	Sheets 1990a MRID 42055324
Rat, Sprague-Dawley, (Sas: CD (SD) BR) 5/sex/dose, approximately 11 weeks old	Single dose of 75 WP-WS formulation (76.1% a.i.) Doses: Males: 1063, 2180 and 3170 mg/kg bw Females: 1063, 2180, 2750, and 3170 mg/kg bw	LD_{50}s Males: 2591 mg/kg bw (≈1970 mg a.i./kg bw) Females: 1858 mg/kg bw (≈1410 mg a.i./kg bw) LOAEL: 1063 mg/kg bw based on Dose-related decrease in body weight gain by day 14; treatment related toxicity (tremors, labored breathing, diarrhea, increased reactivity, decreased reactivity, eyes partially shut, stained fur, salivation, lacrimation etc.) resolved (recovery) in a dose-related manner by day 14.	Sheets and Phillips 1991a MRID 42256312
Rat, Sprague-Dawley, (Sas: CD (SD) BR) 5/sex/dose	Single dose of F.S. 23.1% a.i. formulation. Doses: Males: 1030, 2100, 3595 and 4870 mg/kg bw Females: 2100, 3595 and 4870 mg/kg bw	LD_{50}s Males: > 4870 mg/kg bw (>1120 mg a.i./kg bw) Females: 4143 mg/kg bw (≈957 mg a.i./kg bw) LOAEL (females): 2100 mg/kg bw based on mortality in some females as well as lacrimation, decreased motor activity, tremors, convulsions seen on day of dosing but resolved in survivors by day 2. Dose-related decrease in body weight gain days 0 to 7, but resolved days 7 - 14 for both males and females. No gross treatment-related lesions other than lacrimation in one female.	Sheets 1990f MRID 42256313
Rat, Sprague-Dawley (Sas: CD/SD/BR), 5 or 6 male (179 - 260 g), and 6 female (171-209 g) per dose, 8-10 weeks old	Single dose of BAY T-7391 10% Pour On formulation (9.88-10.01% a.i. w/v) Doses: doses of 0, 495, 1020, 1430 (5 males treated only), 1910 or 2620 mg/kg bw	LD_{50}s Males: 1943 mg/kg bw (≈194 mg a.i./kg bw) Females: 1732 mg/kg bw (≈173 mg a.i./kg bw) LOAEL (clinical signs) = 495 mg/kg: number of rats affected and types of signs are dose-related; signs included hypoactivity, increased reactivity, labored breathing, locomotor incoordination, tremors and oral and nasal staining. Convulsions were seen in one rat at the highest dose. Signs resolved by day 3.	Warren 1995a MRID 43679601

Appendix 1: Toxicity to mammals *(continued)*

Species	Exposure	Response	Reference
Rats, 6-week old male albino, 150-170g	Confidor 20% a.i. EC formulation from Egypt. Gavage administration.	LD_{50}: 451 mg/kg bw. Working Note: Authors do not clearly indicate whether the doses are expressed as a.i. or formulation. The reported LD_{50}, however, is consistent with the reported LD50 of 424 mg/kg bw for technical grade imidacloprid from Bomann (1989a, MRID 42055331).	Mohany et al. 2012
Metabolites			
Rat, SD (Crj:CD), 5 male, 5 female per dose, 7 weeks old, fasted, non-fasted	WAK 3839 nitrosoimine metabolite. Doses: Fasted (M/F): 150, 300, and 600 and 2500 mg/kg bw Non-fasted (M/F): 150, 300, and 600, 900 and 2500 mg/kg bw. No controls used.	No mortality was observed at any dose in any sex. Non-specified toxic effects were observed as follows: non-fasted males: > 300 mg/kg; fasted males: > 150 mg/kg, non-fasted females: >350 mg/kg; fasted females: > 150 mg/kg NOAEL: Not identified. LOAEL: 150 mg/kg bw	Nakazato 1988b MRID 42256360
Rat, SD (Crj:CD), 3 or 4 males/dose, 2 or 3 females per dose, 7 weeks old	WAK 3839 nitrosoimine metabolite. Doses: Males: 300, 1000, 1400, 1800 and 2500 mg/kg bw Females: 1400 and 2500 mg/kg bw. No controls used.	LD_{50} > 2500 mg/kg. No mortality. Non-specified poisoning symptoms reported at all doses tested. Authors report "the poisoning symptoms were rather different from those seen in the study on NTN 33893 (imidacloprid: parent compound).	Nakazato 1990 MRID 42256361

Appendix 1: Toxicity to mammals *(continued)*

Species	Exposure	Response	Reference
Rat, Sprague Dawley (Crj,CD, SPF) 5/sex/dose, 7 weeks old	WAK 3839 nitrosoimine metabolite. Doses: 980, 1560, 2500 and 4000 mg/kg bw	LD_{50}s Males: 1980 mg/kg bw Females: 3560 mg/kg bw No mortality at 980 mg/kg, males, or up to 1560 mg/kg, females. All treated rats had toxic symptoms 25 minutes to 1 hour after exposure. Resolution of symptoms was dose-related: 3,3,7,7 days for males and 1,2,6 and 9 days for females at doses of 980, 1560, 2500 and 4000, respectively. Symptoms include: mydriasis, tremor, sedation, exophthalmos, and abnormal respirations. Some had convulsions prior to death. Nasal and ocular bleeding was seen only in males. Emaciation was seen at doses of 2500 and 4000 mg/kg. Necropsy revealed abnormal findings in the lung, stomach, small intestine, spleen and trachea for both sexes, and in the bladder and thymus for males. The gastrointestinal tract was essentially non-functional, as food was retained in the stomach and fecal excretion was suppressed. No pathological abnormalities were observed among surviving rats.	Ohta 1991 MRID 42286103
Mouse, ICR(Crj;CD-1), 5 week old, 5/sex/dose, for fasted and non-fasted studies	WAK 3839 (a.k.a. NTN 37571) nitrosoimine metabolite. Doses: 100, 200, 300 and 450 mg/kg bw	LD_{50}: Fasted Males: 200 (110-340) mg/kg bw Non-fasted Males: 240 (150-340) mg/kg bw Fasted Females: 200 (120-310) mg/kg bw Non-Fasted Females: ≈300 mg/kg bw Abnormal gait and respiration, exophthalmos, tremor, convulsion and click-like vocalization noted at all dose levels.	Nakazato 1988a MRID 42256325

See Section 3.1.4 for discussion.

A1 Table 2: Poisoning in Humans

Subject, age, weight / Location	Exposure/ Dose Estimate	Response	Reference
Oral			
Male, 22 years India	Intentional ingestion of 30 mL (17.8%) imidacloprid formulation. Dose Estimate: ≈5240 mg or 76 mg/kg bw assuming 70 kg.	Elevated temperature (100.4 °F), rapid heartbeat. Normal blood profile except for low potassium (2.9 mEq/L). Recovery and discharge after 5 days in hospital. No aggressive supportive care reported.	David et al. 2004
Male, in 70s, 56 kg Japan	Intentional ingestion of 20% a.i. formulation. Appears that no more than 50 mL was consumed. Dose Estimate: ≈10,000 mg or 179 mg/kg bw). Co-exposure with ethanol.	Fatal. Concentrations of 105 µg/mL in femoral blood and 58.5 µg/mL in spinal fluid. Cause of death attributed to imidacloprid consumption.	Fuke et al. 2014
Female, 69 years Taiwan	Intentional ingestion of 200 mL Confidor (imidacloprid 9.6%) in N-methyl pyrrolide). Dose Estimate: ≈19,200 mg or 320 mg/kg bw assuming 60 kg bw.	*Disoriented consciousness* with vomiting and sweating. Temperature of 35°C/95°F. Blood pressure decreased from 170/73 to <90/50 mm Hg. Increased heart rate. *Intermittent ventricular fibrillation and ventricular tachycardia.* Died 12 hours after admission. Death attributed to cardiac toxicity.	Huang et al. 2006
Male, 34 years India	Intentional ingestion. No other details provided.	Low blood pressure (NOS) slow heart rate (50 bpm) at 2 hours after ingestion. *Liver dysfunction* and *septic shock.* Despite supportive case, died on day 12.	Iyyadurai et al. 2010
Male, 67 years Turkey	Intentional ingestion. Amount unknown.	*Disorientation, drowsiness, and increased salivation* on admission to hospital. Full recovery by day 4 with supportive care.	Karatas 2009
Male, 56 years Taiwan	Intentional ingestion of 40 mL of a 9.6% a.i. formulation with N-methyl-2-pyrrolidone. Dose Estimate: ≈3840 mg a.i. or ≈55 mg a.i./kg bw assuming 70 kg.	Fever, persistent hypotension, profound dyspnea and coma. Drowsy with low blood pressure (87/56 mm Hg). Somewhat elevated white blood cell count (13,900/µL). Full recovery by day 4 with supportive care.	Lin et al. 2013

Appendix 1: Toxicity to mammals *(continued)*

Subject, age, weight / Location	Exposure/ Dose Estimate	Response	Reference
Female, 35 years Location not clear	Intentional ingestion. Amount unknown. Plasma concentration of 44.6 ng/L. Working Note: The plasma concentration is much lower than the fatal exposure reported in Fuke et al. 2014.	Respiratory failure and low blood pressure. Full recovery within 9 days with supportive care.	Mohamed et al. 2009a
Male, 37 years India	Intentional ingestion of 50 mL of 17.8% formulation of imidacloprid. Dose Estimate: 8900 mg or ≈127 mg/kg bw assuming 70 kg.	Irritable and violent. No elevation in temperature but temperature increased after admission to hospital. Peak temperature of 104 °F. Increased liver enzymes in serum. Full recovery with supportive care by day 9. Working Note: Authors suggest that many of the reported symptoms, including neuropsychiatric, may have been due to treatment with atropine.	Panigrahi et al. 2009
Male, 33 years Portugal	Intentional ingestion. Amount unknown. Concentration in blood of 12.6 µg/mL	Fatal Working Note: Concentration in blood somewhat lower than fatal case reported by Fuke et al. 2014 (58.5 µg/mL).	Proenca et al. 2005
Male, 66 years Portugal	Intentional ingestion. Amount unknown. Concentration in blood of 2.5 µg/mL	Fatal Working Note: Concentration in blood substantially lower than fatal case reported by Fuke et al. 2014 (58.5 µg/mL).	Proenca et al. 2005
Male, 35 years, 85 kg Iran	Intentional ingestion of 350 mL imidacloprid. Not clear if this specifies a formulation but the paper discusses effects of solvent. Dose Estimate: 350,000 mg (?) or 4,117 mg/kg bw. Dose could have been below 1,000 mg/kg for a <20% a.i. formulation.	Broad spectrum of neurologic effects, decreased heart rate, increased blood pressure and fever. Substantial increase in white blood cell count (up to 173,000/µL). Death by 5 days after admission.	Shadnia and Moghaddam 2008

Appendix 1: Toxicity to mammals *(continued)*

Subject, age, weight / Location	Exposure/ Dose Estimate	Response	Reference
Male, 40 years India	Intentional ingestion of 75 mL of 70% a.i. imidacloprid formulation. Dose Estimate: 52,500 mg or 750 mg a.i./kg bw assuming 70 kg.	Nausea and vomiting with abdominal cramps and difficulty breathing. Slight increase in white blood cell count (NOS). … *neuropsychiatric manifestations like agitation and delirium*. Psychiatric effects reversed after 4 days. No elevation in temperature. Recovery by day 6.	Viradiya and Mishra 2011
Male, 64 years Taiwan	Intentional ingestion of about 100 mL of 9.6% a.i. imidacloprid formulation containing N-methyl pyrrolide. Dose Estimate: 9,600 mg or 137 mg a.i./kg bw assuming 70 kg.	Drowsy, dizzy, irregular heart beat and vomiting with abdominal pain. Improved *mental status* but details not provided. No initial elevation in temperature but temperature of over 100 °F developed. Obvious damage to mucus membranes and gastrointestinal corrosion. Full recovery by day 4. Working Note: While speculative, fevers and increases in WBC counts could be due to membrane damage followed by infection.	Wu et al. 2001
Male, 67 years Taiwan	Intentional ingestion (amount not known) of 18.2% formulation of imidacloprid. History of alcohol consumption.	Disorientation, decreased heart rate, irregular heart rate followed by cardiac arrest. Development of multiple organ failure. Normal liver enzymes in blood. Family refused continued supportive care and individual died.	Yeh et al. 2010

Appendix 1: Toxicity to mammals *(continued)*

Subject, age, weight / Location	Exposure/ Dose Estimate	Response	Reference
Other			
Male, 24 years India	Spraying at 17.8% formulation of imidacloprid.	Fever (NOS) with high pulse (132 beats/min) and elevated blood pressure (166/98). Aggressive supportive care for 6 days. Toxicity attributed to … *central nicotinic stimulation* causing *severe neuropsychiatric signs.* Recovery and release.	Agarwal and Srinivas 2007
Male, 62 years Saudi Arabia	Spraying trees with no PPE for about 30 minutes with a 30% a.i. formulation of imidacloprid (Surekill). No other details of exposure reported.	High fever (up to 38.5°C/101.3 °F). Blood in urine. Increased alanine aminotransferase. Detectable (NOS) imidacloprid in serum on day 7 (after exposure). No detectable level by day 11 after exposure. Survival with aggressive supportive care.	Agha et al. 2012
Female, 48 years Poland	Purported inhalation exposure. No other details available.	Individual admitted to hospital. Mild increase in white blood cells. Asymptomatic after 2 days.	Chwaluk 2010 [Abstract only]
Male, 60 years India	Spraying pesticide in field for 1 hour with "HOTSHOT" formulation containing imidacloprid (17.80 % a.i.).	Initial difficulty in breathing followed by nausea, vomiting, and cramps. Mild increase in white blood cells. Blood chemistry indicative of liver damage. *Neuropsychiatric manifestations*. Full recovery with supportive care after 2 days. `Working note: Consistent with Agarwal and Srinivas (2007).`	Kumar et al. 2014

A1 Table 3: Subchronic and Chronic Toxicity Studies

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Subchronic			
Cow, dairy, 3/dose	Cumulative doses: 0, 5 (1 dose), 15 (3 doses) and 50 (10 doses) mg given as single 5 mg/kg bw doses of imidacloprid (97.6% a.i.)/kg via bolus capsules 28 day observation period.	No effects on body weight, food consumption or milk production. No effects relative to controls on weights of muscle, fat, liver or kidney at day 28 sacrifice.	Heukamp 1992a MRID 42556139 Murphy 1994a MRID 43143206 (additional information)
Dog, Beagle, 4 male, 4 female per group, 18 - 20 weeks old, 4.9 - 8.2 kg	Technical grade, 95.3% a.i. At 0, 200, 600 and 1800 ppm (1200 ppm from week 4 due to low food consumption) in the diet. These concentrations correspond to measured doses of 0, 65.2, 191.2 and 342.1 mg/dog/day. Duration: 13 weeks	No reduction in body weight gain in treated groups, except at the 1800 ppm concentration. There was no statistically significant difference between controls and treated dogs when the highest concentration was reduced to 1200 ppm. No mortality. No effects on hematology, liver and kidney function, histopathology. Trembling, independent of feeding time was observed in all 600 and 1800 ppm dogs up to the fifth week of the study. NOAEL: 65.2 mg/kg bw/day	Ruf 1990 MRID 42256328
Dog, Beagle, 2 male (8.6 kg, 4-6 months old) and 2 female (7.9 kg 4-6 months old) per dose	Technical grade, 92.85% a.i. 0, 200, 1000, and 5000 ppm diet. Concentrations correspond to 0, 7.3, 31.0 and 49.0 mg/kg bw/day. Duration: 28 days.	200 ppm: no clinical signs or reduction in food consumption; no effect on body weight gain. 1000 ppm: no clinical signs; transient reduction in food consumption; no effect on body weight gain; no treatment related pathology. 5000 ppm: all dogs died or were sacrificed. Tremor and ataxia. Marked weight loss. Histopathological confirmation of adverse effects on liver (atrophy, pigmentation of Kupffer cells, hypertrophy), pancreas (decreased zymogen content), testes (tubular degeneration), thyroid (follicular atrophy), bone marrow (atrophy), thymus (involution), and salivary glands (acinar atrophy).	Bloch 1987 MRID 42256330

Appendix 1: Toxicity to mammals *(continued)*

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Mice (NOS), adult, 12 per group.	Confidor 20% EC formulation (Bayer) Gavage: 0, 5, 10, and 15 mg a.i./kg bw Duration: 15 days	Slight but significant decreases in body weight at 2 higher doses (Figure 1 of paper). Significant increase in liver and kidney weights at highest dose (Figure 2) and increase in SGOT and SGPT at highest dose (Figure 3). Liver pathology at highest dose (Figure 5).	Arfat et al. 2014 China/Pakistan
Mice, BALB/c, female, 4-6 weeks old, 6-8 mice per group.	28-day gavage doses of 0, 2.5, 5, and 10 mg/kg bw/day. Technical grade imidacloprid (>98% pure) from Indofil Chemicals Company (Mumbai, India).	Significant decrease in platelet count at high dose (≈33%) and dose-related decreases (N.S.) at lower doses. Significant decreases in delayed-type hypersensitivity (increase in paw thickness) at mid and high doses and increase in T-cell proliferation response at high dose. A "*seeming dose-related depletion of lymphocytes in splenic white pulp*". Decrease in spleen weights at all dose but not statistically significant. No mortality or overt signs of toxicity at any doses. NOAEL: 2.5 mg/kg bw/day based on lack of statically significant changes in immune parameters.	Badgujar et al. 2013
Rats, albino, 150-170 g, 20 per group	28-day gavage dose of 0 or 0.21 mg/kg bw/day Confidor (20% EC), Egyptian formulation. Working Note: Not clear if dose refers to formulation or a.i.	Significant increase in leukocyte counts and total immunoglobins. Decreases in phagocytic activity. Histopathologic changes in liver, spleen, and thymus, characterized by authors as *severe*. Also increase in plasma enzymes indicative of liver damage.	Mohany et al. 2012
Mice, B6C3F, 10 male (19g) and 10 female (17 g) per dose, 5-6 weeks old	107-Day range-finding carcinogenicity study (see below). Concentrations: 0, 120, 600 or 3000 ppm TGAI (92.8% a.i.) in the diet.	NOAEL: 120 ppm, male; 600 ppm female 600 ppm: decreased body weight gain in males; 3000 ppm: decreased body weight gain in males and females; increased food consumption per kg body weight (11% males; 41% females); functional and morphological liver changes; significantly lower absolute and relative heart weights; increased frequency of death during blood withdrawal (7/10 M; 7/10 F, compared with 0/10/sex controls.).	Eiben 1988b MRID 42256337 Working Note: The CalEPA (2013) summary of this study reports the NOAEL as 600 ppm (85.7 mg/kg bw/day)

Appendix 1: Toxicity to mammals *(continued)*

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Rats, Wistar, male, 100-120 g.	28-day gavage dose of 0, and 45, and 90 mg/kg bw/day. Group designations in paper: Group I: Untreated Group II: Vehicle Control Group IV: Low Dose Group V: High Dose Technical grade imidacloprid (purity not specified) from Indofil Chemical Company, Mumbai, India	No overt signs of toxicity. Change in consistency of feces and rough body coat at high dose. Significant decrease in spontaneous locomotion (Fig. 1) and decrease in pain response (paw withdrawal threshold) (Fig. 2) at low and high dose. Decrease in AChE activity in RBCs and brain at both doses and plasma at high dose (Table 1 of paper). Dose-dependent decrease in ATP-ase activity in blood and brain as well as decrease in brain glutathione.	Lonare et al. 2014
Rat, Wistar (WISW SPF-Cpb), 10 male (69 g), 10 female (69 g) per dose, 5-6 weeks old.	98- Day range-finding study: 0, 120, 600, 3000 ppm imidacloprid (92.8% a.i.) in the diet.	120 ppm: No effects ≥600 ppm: reduced body weight gain. 3000 ppm: increased food consumption; decreased blood glucose and cholesterol levels; liver effects (multifocal group cell necrosis, elevated alkaline phosphatase); low-grade degenerative changes in testicular tubules.	Eiben 1988a MRID 42256334
Rat, Wistar (WISW, SPF Cpb), 10 male (84 g), 10 female (77 g) per concentration, 5-6 weeks old	96 day exposure to technical grade imidacloprid (95.3% a.i.) in feed at concentrations of 0, 150, 600, 2400 ppm. Recovery groups at 0 and 2400 ppm diet for 14 weeks, then 4 weeks with no exposure. Measured doses Males: 0, 14.0, 60.9 or 300.2 mg/kg bw/day Females: 0, 20.3, 83.3 or 422.2 mg/kg bw/day	NOAEL: 150 ppm, males (14 mg/kg bw/day); 600 ppm (83.3 mg/kg bw/day), females. Reduction in body weight gain (retarded growth) at concentrations > 600 ppm in males and in females at 2400 ppm. Increased food intake relative to body weight in 2400 ppm rats, both sexes, even after the recovery period. No effects on clinical signs, drinking water consumption, mortality, hematopoietic organs, blood, eyes, organs, organ weights, histopathology, cholinesterase activity in plasma, erythrocytes or brain, at any concentration, except for the following: liver toxicity (increased incidence of cell necrosis, round cell infiltrates, swollen cell nuclei and cytoplasmic changes in liver and slightly raised AST and ALT) in 400 ppm males. Reduced platelet count and blood clotting (thromboplastin times) in both sexes at 2400 ppm.	Eiben 1989 MRID 42256327

Appendix 1: Toxicity to mammals *(continued)*

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Rats, Wistar, 150-155 g., 10/dose	Imidacloprid (96%) Gavage doses: 0 (vehicle control), 5, 10, and 20 mg/kg bw/day for 90 days.	10 mg/kg bw: NOAEL 20 mg/kg bw: Decreased food consumption, decreased locomotor activity, increase in liver enzymes in blood, decrease in AChE activity in serum (16-49%) and brain (16-40%). Author's note: …*cause of this inhibition [AChE] is unknown because imidacloprid is not ChE inhibitor, since plasma AChE is synthesized in the liver, the decrease in plasma AChE activity may be related to observed changes in liver function*.	Bhardwaj et al. 2010
Rats, Wistar, 150-155 g., 10/dose	Imidacloprid (96%) Gavage doses: 0 (vehicle control), 5, 10, and 20 mg/kg bw/day for 90 days. Working Note: This may be the same study as Bhardwaj et al. (2010) with focus on different endpoints.	10 mg/kg bw: NOAEL 20 mg/kg bw/day: Significant changes in a variety of biochemical parameters (e.g., superoxide dismutase and catalase) indicative of oxidative stress.	Kapoor et al. 2010
Rats, Wistar, 150-155 g., 10/dose	Imidacloprid (96%) Gavage doses: 0 (vehicle control), 5, 10, and 20 mg/kg bw/day for 90 days. Working Note: This may be the same study as Bhardwaj et al. (2010) with focus on different endpoints.	10 mg/kg bw: NOAEL 20 mg/kg bw: Reports of endpoints associated with oxidative stress (as in Kapoor et al. 2010). In addition, decreased ovarian weights, changes in ovarian morphology, a significant increase in follicle stimulating hormone, and significant decreases in luteinizing hormone and progesterone.	Kapoor et al. 2011

Appendix 1: Toxicity to mammals *(continued)*

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Rats, female, albino, 3 months old, 100-150 g. 6 rats per group.	Confidor 40 SL (Mumbai). Imidacloprid doses of 0 (both untreated and vehicle control), 9 and 45 mg/kg bw/day for 28 days. Working Note: The % a.i. in the Confidor formulation is not specified and a label for Confidor 40 SL has not been identified.	Low dose: Decrease in body weight gain (15%, $p<0.05$) and relative liver weight (3%, N.S.) relative to untreated control. Significant increase in plasma liver aspartate aminotransferase but no significant liver pathology. High dose: Decrease in food consumption (≈6%, N.S.), body weight gain (20%, $p<0.05$), and relative liver weight (≈7%) relative to vehicle control. Increases in plasma aspartate aminotransferase and alkaline phosphatase, dilation of central vein and sinusoids. Working Note: Examined only effects on liver. No other histopathology.	Toor et al. 2013
Rats, albino, female, 3 months old, 100-150 g. 6/group	Imidacloprid in Confidor (17.8% a.i.) formulation. Gavage doses of 0, 10, and 20 mg a.i./kg bw/ day for 60 days.	10 mg/kg bw: Significant ($p<0.05$) decrease in spleen weight (12.5%). 20 mg/kg bw: Significant ($p<0.05$) decreases in spleen weight (≈21%) and heart weight (8%). Significant decrease in food consumption. Both doses: Significant and dose-related decrease in brain and plasma AChE.	Vohra et al. 2014

Appendix 1: Toxicity to mammals *(continued)*

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Metabolite			
Rat, Wistar (Bor: WISW (SpF -Cpb), 15/sex/dose, approximately 5 weeks old, 82 gram males, 78 gram females.	WAK 3839 (a.k.a. NTN 37571) nitrosoimine metabolite. Drinking water at concentrations of 0 (tap water), 100, 300 and 1000 ppm, measured concentrations were 0, 112, 339 and 1105 ppm. 12 weeks	NOAEL: 110 ppm (13 mg/kg bw/day) >300 ppm: higher lymphocyte counts and lower numbers of polymorphonuclear cells in both sexes regarded as treatment-related. >1000 ppm: reduced sodium levels in both sexes viewed as treatment-related effect on sodium balance. Lower water consumption (approximately 16% less) than controls. No thyroid effects were noted.	Krotlinger 1992 MRID 42256362 Additional details in CalEPA 2013.
Chronic			
Dog, Beagle, 4 male (6.6 - 9.2 kg) and 4 female (5.3-7.4 kg) per dose, 4-6 months old	52-Week feeding study with TGAI (94.9% a.i.). Concentrations: 0, 200, 500 and 1250/2500 ppm. The concentration in the last dose group was increased from week 17 onward. Average doses of 0, 6.1, 15 and 41/72 mg a.i./kg body weight/day	NOAEC: 500 ppm diet NOAEL: 15 mg/kg bw/day 1250/2500 ppm: slight but statistically significant elevated plasma cholesterol (females) and elevated liver cytochrome P450 (both sexes) with respect to controls. Slight but not statistically significant elevation in liver weight (both sexes) was considered treatment related.	Allen et al. 1989 MRID 42273002
Mouse, B6C3F1, 50 male (20 g) and 50 female (15 g) per dose, approximately 5 weeks old	24-Month carcinogenicity study: Concentrations: 0, 100, 330 and 1000 ppm TGAI (95.0% a.i.) in the diet. Doses: Males: 0, 20.2, 65.6, and 208.2 mg/kg bw/day Females: 0, 30.3, 103.6, and 274.4 mg/kg bw/day	NOAEL: 330 ppm 1000 ppm: reduced body weight gain (up to 10% and 5% lower for males and females, respectively. Slightly lower food and water consumption in females. No effects on incidence or timing of tumors. No effects on mortality, clinical chemistry, urinalysis, hematology, organ weights. No adverse treatment-related histopathological findings.	Watta-Gebert 1991a MRID 42256335

Appendix 1: Toxicity to mammals *(continued)*

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Mouse, B6C3F1, 50 male (25 g) and 50 female (21 g) per dose, approximately 7-8 weeks old; 10 additional mice per sex and dose were included for interim sacrifice.	Supplementary 24-month carcinogenicity study: 0 and 2000 ppm NTN 33893 (95.0% a.i.) in the diet. Equivalent to doses of 413.5 (males) and 423.9 (females) mg imidacloprid/kg body weight/day	No treatment-related effects on the incidence or timing of tumors. 2000 ppm: Adverse effects on the brain (increased incidence of mineralization of the thalamus); reduced blood cholesterol levels; statistically significant reduced mean body weight (up to 29% in males and 26% in females, with respect to controls). A "squeaking and twittering type of vocalization" was heard among the treated but not control mice from the inception of the study and throughout. No statistically significant difference in mortality between treated and control mice, but treated male mice died more frequently during manipulation (ether anesthesia for blood withdrawal, during tattooing or getting caught in automatic feeders) than did controls.	Watta-Gebert 1991b MRID 42256336
Rat, Wistar (Bor: WESW (SPF Cpb)), 50 male (81 g) and 50 female (76 g) per dose; 4 - 6 weeks old	24-months, TGAI (95.3% a.i.). Concentrations: 0, 100, 300 and 900 ppm diet. Doses: Males: 0, 5.7, 16.9 and 51.3 mg/kg bw/day Females: 0, 7.6, 24.9 and 73.0 mg/kg bw/day.	NOAEL (males): 100 ppm (thyroid) NOAEL (females): 300 ppm (thyroid) Treatment-related increased incidence of mineralization of the colloid of the thyroid follicles in males (300 and 900 ppm) and females (900 ppm). Treatment-related reductions in body weight gain were observed in both sexes at 900 ppm. No other treatment-related effects on mortality, clinical signs, clinical chemistry, ophthalmology, organ weights, tumor incidence or pathology. No effects on plasma, red cell or brain cholinesterase.	Eiben and Kaliner 1991 MRID 42256331 **This study is the basis for EPA's RfD of 0.057 mg/kg/day based on 5.7 mg/kg bw/day in low dose males.**

Appendix 1: Toxicity to mammals *(continued)*

Organism	Agent/Exposure	Response	MRID, Study Date, Classification
Rat, Wistar (Bor: WESW (SPF Cpb)), 50 male (90 g) and 50 female (84 g) per dose; 5 - 6 weeks old: an additional 10 rats/sex/dose were treated and sacrificed after 12 weeks for interim examination.	24-month supplementary chronic toxicity and carcinogenicity study. TGAI (95.3% a.i.) Concentrations: 0 or 1800 ppm. Doses: 102.6 mg/kg bw/day (males); and 143.7 mg/kg bw/day (females).	Confirms adverse effect on thyroid. Statistically significant (compared with controls) treatment-related increased incidence of mineralization in the colloid of the thyroid follicles; fewer colloid aggregation sites; parafollicular hyperplasia sites with minimal intensity. Also, retardation of growth (up to 12% reduction in body weight gain). No other treatment-related effects.	Eiben 1991 MRID 42256332

See Section 3.1.5 for discussion.

A1 Table 4: Reproductive and Developmental Studies

Species	Exposure	Response	MRID(s), (Year), Classification
Developmental			
Rabbit, Chinchilla (CHbb: CH hybrid: SPF quality), 16 females per dose, 4-6 months old, 2650 - 4064 g.	TGAI (94.2%) Doses: 0 (vehicle control), 8, 24 and 72 mg/kg bw/day, Days 6 through 18 of gestation. Sacrifice on day 28.	Maternal: NOAEL = 8 mg/kg/day. Statistically significant dose-related reduction in food consumption during treatment at 24 and 72 mg/kg/day. Reduction in body weight gain at 24 mg/kg/day (slight, during dosing period) and 72 mg/kg/day (significant on days 11-23 and 25-26 after mating); Reproductive: NOAEL = 24 mg/kg/day. At 72 mg/kg/day: 1 female aborted on Day 26 and 2 females had total litter resorptions at day 28 necropsy. This post-implantation loss results in a statistically significant reduction in the number of live fetuses per dam (32.5% in comparison with control value of 4.2%). There was also a slight but statistically significant reduction in live fetuses per dam, when only dams with live fetuses at termination were considered (10.8% versus control value of 4.2%).	Becker and Biedermann 1992 MRID 42256339
Rat, Wistar/HAN, 25 mated females per dose, 11 weeks old, 184-240 g.	TGAI (94.2%) Doses: 0 (vehicle control), 10, 30 and 100 mg/kg bw/day. Days 6 through 15 of gestation. Sacrifice on day 21.	Maternal: NOAEL= 10 mg/kg/day. Statistically significant reduction in food consumption at all doses; reductions in body weight gain at 30 (marginal) and 100 (significantly) mg/kg/day. Reproductive: NOAEL = 100 mg/kg/day. No statistically significant treatment-related effects at any dose for any variables assessed: mean number of implants, fetuses, resorptions. Fetal: NOAEL = 30 mg/kg/day. Slightly increased incidence of wavy ribs at 100 mg/kg/day (7/149 fetuses; 5/25 litters) in comparison with vehicle controls (2/159 fetuses; 1/25 litters). No other treatment-related effects.	Becker et al. 1992 MRID 42256338

Appendix 1: Toxicity to mammals *(continued)*

Species	Exposure	Response	MRID(s), (Year), Classification
Rat, Wistar (Crl:W(HAN)BR, 21 mated females, approximately 25/dose; 20 litters per dose formed from litters with at least 8 pups and 3 male and 3 females, were culled to 8 pups (as closely as possible to 4 male and 4 female)	Developmental Neurotoxicity Screening Study. Technical-grade imidacloprid (98.2 - 98.4% a.i.) administered from gestation day 0 through lactation day 21 at dietary concentrations of 0, 100, 250 and 750 ppm (measured concentrations: 0, 95.5, 227 and 691 ppm) Doses: 0, 8.0 - 8.3, 19.4 - 19.7, and 54.7 - 58.4 mg/kg bw/day; during lactation: 0, 12.8 - 19.5, 30.0 - 45.4, and 80.4 - 155.0 mg/kg bw/day.	No effects on reproduction variables including the fertility index or gestation length. Maternal: 14% reduction in food consumption at highest dose. No effect on body weight, no clinical signs. NOAEL: 250 ppm (19.4 - 19.7 mg/kg bw/day during gestation). Offspring: Decreased body weight gain and reduced activity in the figure-eight maze relative to controls at 750 ppm (54.7 - 58.4 mg/kg bw/day during gestation) on post-natal-day (PND) 17(both sexes) and PND 21(females only). No other compound-related effects (acoustic startle habituation, passive avoidance, water maze, ophthalmology, gross lesions, brain weight, brain morphology or microscopic pathology of the brain, neural tissues or skeletal muscle). The only adverse effect persisting to termination of study was a 4% deficit in body weight, relative to controls, among high-dose males. NOAEL: 250 ppm (19.4 - 19.7 mg/kg bw/day)	Sheets 2001 MRID 45537501
Rats, Sprague-Dawley (300–350 g), 5 per group	Intraperitoneal doses of imidacloprid (≈99.5% purity): 0 and 337 mg/kg bw. Single injection on Day 9 of gestation,	No mortality in dams or offspring. No overt signs of toxicity in dams. Offspring: significant sensorimotor impairment on post-natal Day 30. Included an increased AChE activity in the midbrain, cortex and brainstem (125–145% increase) and in plasma (125% increase). Working Note: No standard observations for malformations	Abou-Donia et al. 2008 [full study] Abdel-Rahman et al. 2005 [Abstract]

Species	Exposure	Response	MRID(s), (Year), Classification
Rats, Wistar, 6-8 weeks old, 160-168 g. 6 rats per group.	**Developmental immunotoxicity study** Imidacloprid (source and purity not specified) by gavage at doses of 0, 10, 30, 90 mg/kg bw on days 6-21 of gestation.	No signs of toxicity in dams at any dose. Fetal Effects Increase in malformations at 30 and 90 mg/kg bw as well as dose-related increase in post-implantation losses at these doses. No post-implantation losses in control or low dose groups. No significant effects at 10 mg/kg bw. Immune effects Decreased immune response to SRBC (sheep red blood cells) at 90 mg/kg bw/day. As lower doses, a dose-dependent decrease in hemagglutination titers. Also a decrease in immunoglobulin levels. Working Note: Malformations are not expressed in litters.	Gawade et al. 2013
As above	As above but dams doses from Day 6 of gestation to post-natal day 21.	No signs of toxicity in dams at any dose. Effects on offspring Dose-related and significant ($p<0.01$) decreases in phagocytosis and hemagglutination titer at all dose levels (Table 3 of paper). Dose-dependent increase in relative weights of thymus, decrease in relative weights of spleen (Data not given in tables but summarized on p. 64, column 1). Discussion seems unclear. Increases in plasma enzymes indicative of liver damage (AST and ALP)	Gawade et al. 2013
Reproduction			
Rat, Wistar/HAN, 30 male (123 - 169 g) and 30 female(81 - 137 g) per dose, 5-6 weeks old at start of exposure for parental generation; breeding at approximately 17 weeks old	Technical-grade imidacloprid (94.4 - 95.4% purity) 0, 100, 250 and 700 ppm in the diet.	NOAEL = 250 ppm (20 mg/kg bw/day) for reproductive effects 700 ppm: reduced food consumption in P and F1 generations, both sexes. Reduced body weight gain in first part of the treatment of P generation.; lower mean body weight in F1 throughout the study; reduced mean body weight and body weight gain in pups of all generations (F1A, F1B, F2A, F2B) throughout the study. No abnormalities in offspring were observed.	Suter et al. 1990 MRID 42256340

Species	Exposure	Response	MRID(s), (Year), Classification
Target Organ Effects			
Rats, male, Wistar, obtained a 7 days old, 6 per group	Imidacloprid (NOS), gavage doses of 0, 0.5, 2, or 8 mg/kg bw/day for 90 days.	Significant and dose-related decrease in body weight testosterone at all doses (Table 1 of paper). Decrease in testes and prostate weights not significant but significant decreases in weights of epididymis, right cauda epididymis, seminal vesicles (Figure 1 of paper). Significant increase in abnormal sperm at high dose (Table 2 of paper).	Bal et al. 2012a
Rats, male, Wistar, 8-9 weeks old, 180-210 g, 6 per group	Imidacloprid (NOS), gavage doses of 0, 0.5, 2, or 8 mg/kg bw/day for 90 days. Working Note: This paper and the above paper are similar in design but appear to be two different studies. See Section 3.3.3 for discussion.	Significant and dose-related decrease in body weights and body weight gain (Table 1). Decrease in testes and prostate weights not significant but significant decreases in weights of epididymis, right cauda epididymis, seminal vesicles (Figure 1 of paper). Significant decrease in sperm motility at highest dose and decrease in epididymal sperm concentrations at mid and high doses. Increase in abnormal sperm (Table 2).	Bal et al. 2012b

A1 Table 5: Acute and Repeated Dose Dermal Toxicity

Species	Exposure	Response	Reference
Acute			
Technical Grade			
Rat, Wistar (Bor: WSIW SPF-Cpb), 5 male (207 - 234 g), and 5 female (204 - 214 g)	Imidacloprid (94.2%) Dose: 5000 mg/kg bw for 24-hours, occluded.	No mortality. No clinical signs. No treatment-related body weight reductions. No gross pathology.	Krotlinger 1989 MRID 42055332
Formulations			
Rat, Sprague-Dawley, 5 male and 5 female, 167 - 245 g, 8-9 weeks old	Permatek IM 30 (31 g a.i./L), occluded. Dose: 2000 mg formulation/kg bw	No treatment-related mortality, clinical signs or findings at gross necropsy.	Pritchard and Donald 2004b MRID 46290904
Rabbit, New Zealand White, 5 male, 5 female	2.5% Granular formulation (2.6% a.i.) Dose: 2000 mg formulation/kg bw	No deaths. No clinical signs. All animals gained body weight. No gross lesions observed at necropsy.	Sheets 1990b MRID 42055325
Rabbit, New Zealand White, 5 male, 5 female	240 F.S. formulation. Dose: 2000 mg formulation/kg bw	No mortality. Clinical signs = erythema at the dose site of 2 females; muscle fasciculation in 1male and 1 female. Clinical signs resolved by day 2. No gross lesions	Sheets 1990g MRID 42056315
Rat, Sprague-Dawley, (Sas: CD (SD) BR), 5 male (approx. 8 weeks old), 5 female (approx. 10 weeks old)	75 WP-WS (76.1% a.i.) formulation, occluded. Dose: 2000 mg formulation/kg bw	No mortality. Urine stain in one male and 1 female was the only clinical sign. This female also developed alopecia on day 5. The alopecia persisted to the end of the study. No effects on body weight gain	Sheets and Gilmore 1991 MRID 42256314
Rat, Sprague-Dawley (Sas(CD(SD)BR), 6 male (234-271 g) and 6 female (206 - 244 g) per dose, 8-10 weeks old	BAY T-7391 10% Pour On (9.88 - 10.01% a.i.), occluded. Doses: 0 or 2000 mg formulation/kg bw	No treatment-related mortality, changes in body weigh/food consumption, clinical signs or gross lesions.	Warren 1995b MRID 43679602
Repeated Dose			
Rabbit, HC-NZW, 5 male (3.00 kg), 5 female(2.92 kg) per group, 13 weeks old	6-hr/day, 5 days/week, 3 week occluded application exposure to technical grade imidacloprid (95.0% a.i.) to shaved skin at 0 or 1000 mg/kg bw.	No treatment-related mortality. No effects on food consumption, body weight gain. No significant differences between controls and treated animals in clinical chemistry values, blood formation or cell counts, clinical chemistry, organ weights, histopathological findings, or gross pathology. No treatment related skin changes.	Flucke 1990 MRID 42256329

A1 Table 6: Skin Irritation and Sensitization Studies

Species	Exposure	Response	Reference
SKIN IRRITATION			
Technical Grade			
Rabbit, White (HC:NZW), 3 male	94.2% a.i., skin occluded for 4 hours, applied as paste.	No edema or irritation up to 7 days post-exposure. Not a skin irritant.	Pauluhn 1988c MRID 42055335
Formulation			
Rabbit, New Zealand White, 3 male, 3 female	2.5% Granular (2.6% a.i.), 4 hours, occluded.	No signs of erythema or edema at dose site 30 minutes, 60 minutes, or 24, 48 or 72 hours after patch removal. No signs of irritation. Primary irritation index = 0.00. Not a primary dermal irritant.	Sheets 1990d MRID 42055328
Rabbit, New Zealand White, 6 male, adult	75 WP-WS (76.1% a.i.) formulation Dose: 500 mg applied as paste.	Erythema (Grade 2) at dose site in 5/6 and edema (Grade 1) in 1/6, 1 hour after application. All irritation gone by day 7.	Sheets and Phillips 1991c MRID 42256320
Rabbit, New Zealand White, 3 male, 3 female, adult	70 WG (% a.i. not specified) Dose: 500 mg	Slight erythema in 3/6 at 4-hours, and in 2/6 at 24 hours. Slight edema in 2/6 at 4 hours. No signs of irritation at 24 hours.	Wakefield 1996b MRID 46234904
Rabbit, New Zealand White, 3 male, 3 female, adult	240 F.S. (23.1% a.i.) formulation. Dose: 500 mg	No erythema or edema in any animal.	Sheets 1990i MRID 42256321
Rabbit, New Zealand White, 6 male, young adult	0% Pour On formulation (9.88 - 10.01% a.i.) Dose: 500 mg.	Erythema in 1/6 rabbits 24 hours after removal of patch; resolved by 48 hours.	Warren 1995d MRID 43679605
Rabbit, New Zealand White, 3 male, 3 female, young adult	0.5 ml Pointer Insecticide (5% a.i.)	Slight/mild irritation.	Robbins 1996b MRID 44137602
Rabbit, New Zealand White, 2 male	0.5 ml Permatek IM30 (32 g a.i./L) formulation.	No erythema or edema in either rabbit at any observation point.	Pritchard and Donald 2004d MRID 46290906
Guinea Pig, Hartley Albino, adult male, 5/dose	75 WP-WS formulation in deionized water at doses of 1, 2.5, 5, 7.5, 10, 25, 50, 100 % (w/v).	Grade 1 erythema, red zones, or crusts at dose site in animals dosed with >10%. This was part of the skin sensitization study. See below.	Sheets and Phillips 1991d MRID 42256322
Guinea Pig, Hartley Albino, Adult male, 15 induced and 15 non-induced (control)	10% Pour On formulation, undiluted Topical induction on days 0, 7 and 14. Topical challenge on day 28.	No treatment related erythema, edema or clinical signs in any animal at any time.	Warren 1995e MRID 43679606

Appendix 1: Toxicity to mammals *(continued)*

Species	Exposure	Response	Reference
SKIN SENSITIZATION			
Technical Grade			
Guinea Pig, SPF DHPW, male (5- 8 weeks old)	Intradermal induction: 1%. Topical induction: 25%. Topical challenge: 3%,25%.	No skin reaction in either treated animals or controls.	Ohta 1988 MRID 42055336
Formulation			
Mouse, CBA/Ca strain, 5 females/dose, young adult	Local lymph node assay for sensitization. 0 (vehicle) , 25%, 50% or 100% Permatek IM 30 (32 g a.i./L) applied to dorsum of each ear for 3 consecutive days, followed by intravenous injection of 3H-methyl-thymidine 3 days later.	No mortality or clinical signs. Body weight gain considered normal. No difference between controls and any dose with regard to stimulation of T-Cell proliferation in draining auricular lymph nodes. No indication of sensitization response.	Pritchard and Donald 2004e MRID 46290907
Guinea Pig, Hartley albino, males	0.4 g imidacloprid as 2.5% granular formulation	No sensitization.	Sheets 1990e MRID 42055329
Guinea Pig, Hartley Albino, adult male, 5/dose	240 F.S. formulation in deionized water at doses of 1, 10, 25, 50, 100 % (w/v).	No evidence of irritation or sensitization at any dose.	Sheets 1990j MRID 42256323
Guinea Pig, Hartley Albino, adult male,	75 WP-WS formulation Topical induction on days 0, 7 and 14 with 7.5%BAY NTN 33893 WP-WS	No sensitization.	Sheets and Phillips 1991d MRID 42256322
Guinea Pig, Hartley Albino, Adult male, 15 induced and 15 non-induced (control)	10% Pour On formulation, undiluted	No sensitization.	Warren 1995e MRID 43679606

A1 Table 7: Eye Irritation Studies

Species	Exposure	Response	Reference
Technical Grade			
Rabbit, White (HC:NZW), 2 male, 1 female	94.2% a.i., 0.1 ml solution in conjunctival sac of one eye per rabbit. Eyes rinsed with saline 24 hr post-exposure.	Not an eye irritant, based on type, intensity and chronology of findings. No effects on the cornea, iris or conjunctiva of any rabbit at any time following exposure (up to 7 days evaluated).	Pauluhn 1988b MRID 42055334
Formulation			
Rabbit, New Zealand White, 3 male, 3 female	2.5% Granular (2.6% a.i.) formulation. 0.1 ml instilled in conjunctival sac of one eye per rabbit.	No corneal or iridal lesions. Grades 2 and 3 ocular discharge and conjunctival redness (Grade 1) in all rabbits one hour after dosing. No signs of irritation 14 days post-dosing. Classified originally as Category II Moderate eye irritant, but subsequently reduced to Category III mild irritant, due to absence of corneal or iris involvement, and resolution of irritation by day 7 post-dosing.	Sheets 1990c MRID 42055327; Astroff 1992 MRID 42674401(supplemental submission)
Rabbit, New Zealand White, 3 male, 3 female	0.5% Granular formulation (0.56% a.i.) instilled in conjunctival sac of one eye per rabbit.	No corneal or iridal lesions. Grade 2 and 3 ocular discharge, chemosis (Grades 2 and 3), and conjunctival redness (Grades 1 and 2) in all rabbits one hour after dosing. No signs of irritation 7 days post-dosing. Mild eye irritant.	Sheets and Phillips 1990 MRID 42055320
Rabbit, New Zealand White, 3 male, 3 female	0.62% Granular formulation (0.71% a.i.) instilled in conjunctival sac of one eye per rabbit.	No corneal lesions, but transient iridal lesions (grade 1) were seen in 4 rabbits at 24 hours post-instillation (resolved by 48 hours). Conjunctival redness (grade 0 - 2), chemosis (grade 1,2 or 4), and discharge (grade 2 or 3) was observed in all animals (resolved by day 7). Mild eye irritant.	Astroff and Phillips 1992 MRID 42674402
Rabbit, New Zealand White, 6 young adults	75 WP-WS, 0.1 ml (44 - 46 mg) in conjunctival sac of one eye per rabbit.	No corneal or iridal lesions. Ocular discharge (Grade 2 or 3), chemosis (Grade 1 or 2) and conjunctival redness (Grade 1) were observed in all rabbits one hour after exposure. No signs of irritation in any rabbit 14 days after test.	Sheets and Phillips 1991b MRID 42256318

Appendix 1: Toxicity to mammals *(continued)*

Species	Exposure	Response	Reference
Rabbit, New Zealand White, 3 male, 3 female, young adults	240 F.S. formulation (23.1% a.i.) in conjunctival sac of one eye per rabbit.	No corneal or iridal lesions. Transient ocular discharge (Grade 1), redness (Grade 1) and chemosis (Grade 1) of the conjunctiva in all animals, reversed in all animals by 72 hours.	Sheets 1990h MRID 42256319
Rabbit, New Zealand White, 6 young-adult males	10% Pour On formulation (9.88 - 10.01% a.i.) in conjunctival sac of one eye per rabbit.	Corneal opacity, iridal irritation, conjunctival redness, chemosis and ocular discharge in all rabbits (1-48 hours). All signs resolved by day 14.	Warren 1995c MRID 43679604
Rabbit, New Zealand White, 6 young-adult males	Pointer Insecticide (5% a.i.) in conjunctival sac of one eye per rabbit.	Corneal involvement which resolved by day 17 in all animals tested. Category II moderate eye irritant.	Robbins 1996a MRID 44137601
Rabbit, New Zealand White, 2 female adult	Permatek IM 30(32 g a.i./L) in conjunctival sac of one eye per rabbit.	No irritation in any rabbit at any time. Not an eye irritant.	Pritchard and Donald 2004c MRID 46290905

A1 Table 8: Acute and Subchronic Inhalation Toxicity

Species	Exposure	Response	Reference
Technical Grade			
Rat, Wistar (Bor: WSIW SPF-Cpb), 160 - 210 g, 8 - 10 weeks old, 5/sex/concentration; air control; vehicle control	Technical grade. Measured concentrations: 69, 1220, 2577 and 5323 mg/m^3 , with particle sizes size < 5 µm at 100, 11, 6 and 4 percent, respectively. 4-hour nose-only exposures.	No mortality. No signs or symptoms in controls or 69 or 1220 mg/m3 groups. Difficult breathing, reduced mobility, piloerection at 2577 and 5323 mg/m3. Slight tremors at 5323 mg/m3. All groups clinically normal 1 day post-exposure. Marginally reduced body weight gain in both sexes at highest concentration. No gross pathological findings at any level of exposure	Pauluhn 1988a MRID 42055333 Pauluhn 1988d MRID 42286101 (supplemental submission) Additional study details in CalEPA (2013).
Formulation			
Rat, Sprague-Dawley, 6 male, 6 female exposed; 6 male, 6 female sham-exposed	2.5% Granular (2.6% a.i.), as dust Concentration: 5092 mg formulation/m^3, measured (17,040 mg/m3 nominal); 4-hour nose-only exposure.	No deaths. No clinical signs. No statistically significant changes in body weight with respect to controls. No gross lesions at necropsy.	Warren 1990a MRID 42055326 and Warren 1990c MRID 42286102 (supplemental submission)
Rat,Sprague-Dawley (Sas: CD(SD: BR)) 6/sex/dose, 6 to 8 weeks old, 228 - 275 g males, 189 - 230 g females	240 F.S. as a liquid aerosol 4 hours Measured Concentrations: 5060 or 5330 mg/m^3	LOAEL = 5060 mg/m^3 Mortality < 50% all test groups; Hyperactivity, dyspnea, lethargy and tremors on day of exposure at both concentrations tested. Recovery by day 2. No gross lesions. No substantial reductions in body weight gain, except in low-dose males on day 3.	Warren 1990b MRID 42256317
Rat, Sprague-Dawley (Sas: CD(SD: BR)) 6/sex/dose, 6 8 weeks old, 186 - 244 g males, 177 - 230 g females	75% WP-WS formulation as a liquid aerosol. Measured Concentrations: 2110, 2810 or 2990 mg/m^3	LC_{50}: 2650 mg/m^3, males LC_{50} = 2750 mg/m^3, females LOAEL = 2110 mg/m^3, both sexes Clinical signs: ataxia, convulsions, hypoactivity, moribundity, nasal stain, tremors, unthriftiness and urine stain. Recovery by day 6. Statistically significant decreases in body weight gain on day 3 in males (all doses) and females (2990 mg/m^3). No gross lesions other than salivation and ventral wet stain in animals dying shortly after exposure.	Warren 1991 MRID 42256316
Rat, Sprague-Dawley (Sas: CD(SD)BR), 6 male (203-228 g) and 6 female (189 - 211 g) per dose, 7-8 weeks old	10% Pour On (9.88 - 10.01% a.i.) formulation 4-hours nose-only 2415 mg/m^3	Oral staining was observed in females. No changes in body weight. No mortality. No gross lesions.	Warren and Berry 1995 MRID 43679603

Appendix 1: Toxicity to mammals *(continued)*

Species	Exposure	Response	Reference
Repeated Dose			
Rat, Wistar (Bor: WSIW SPF-Cpb), 160 - 210 g, 8 - 10 wks old, 10/sex/concentration	6-hour exposures per day for 5 days to technical grade imidacloprid as powder dust. Nominal concentrations: 20, 100, 500 a.i. mg/m^3. Analytically determined concentrations: 20, 109, and 505 mg a.i./m^3 , with particle size < 5 µm at 54, 57 and 18 percent, respectively.	No mortality. No clinical signs. No effects on liver or lung to body weight ratios. No treatment-related histopathological changes in liver or lung at any concentration. No liver enzyme-related hepatotoxicity (Serum-ALAT, - ASAT, GLDH). NOAEC: 20 mg/m^3 Concentration-related induction of MFOs at 109 mg/ m3 and higher; "Transient influence on body weights" at 109 mg/m^3 and higher; Dark spleen and lower erythrocyte count at 505 mg/m^3.	Pauluhn 1988a MRID 42055333 and Pauluhn 1988d MRID 42286101 (supplemental submission)
Rat, Wistar (Bor: WISW (SPF-Cpb), 10/sex/dose, 160-200 g., 2-3 months old.	4 weeks, 6 hr/day, 5 days/week exposure to mean analytical concentrations of 5.5, 30.5 and 191.2 mg a.i./m^3 (95.5% a.i.) Particle constitution of dust was considered respirable to the rat; head-nose only exposure.	NOAEC: 5.5 mg/m^3 30.5 mg/m^3: Induction of hepatic mixed-function oxidases. 191.2 mg/m3: statistically significant reduction in body weight gain (males only); slight depression in heart and thymus weights, and increase in liver weight (females only); slight depression in hematocrit and low-grade reduction in plasma proteins attributed to slight hypervolemia (males); increased blood coagulation time and statistically significant elevation in pH of the urine with respect to controls were considered to result from functional hepatic changes (females)	Pauluhn 1989 MRID 42273001

A1 Table 9: Acute Intraperitoneal Toxicity Studies

Species	Exposure	Response	Reference
Imidacloprid			
Rat, Wistar (Bor: WISW (SPF-Cpb)), male (179 g, 8 weeks old), female (178 g, 10 weeks old), 5/sex/dose	Technical grade imidacloprid, 94.2%. Doses: Males: 10, 100, 160, 170, 180, 200, 250 and 500 mg/kg bw. Females: 10, 100, 150, 180, 200, 224, and 250 mg/kg bw.	LD_{50}s Males: 160-170 mg/kg bw Females: 186 mg/kg bw NOAEL (mortality): Males: 160 mg/kg bw Females: 100 mg/kg bw NOAEL (toxicity): 10 mg/kg bw (both sexes). Clinical signs included apathy, labored breathing, reduced motility, dyspnea, lacrimation tremors, spasms, twitching eyelids and piloerection. Transient impact on body weight gain in males at > 170 mg/kg bw and in females at > 180 mg/kg bw. No gross pathology among survivors. Gross findings on liver, lungs, spleen and GI tract among mice which died.	Krotlinger 1990 MRID 42256326
Metabolite			
Mouse, ICR(Crj;CD-1), 5 week old, 5/sex/dose	WAK 3839 (a.k.a. NTN 37571) nitrosoimine metabolite. Doses: 30 and 60 mg/kg bw	LD_{50}: 30 to 60 mg/kg bw No differences in LD_{50} values or clinical signs between sexes. Sedation, tremor and convulsion are reported for all treated mice. Authors report “no specific findings in both dead animals and survivals”.	Nakazato 1988a MRID 42256325

A1 Table 10: Neurotoxicity Studies

Species	Exposure	Response	Reference
Acute			
Rat, Sprague-Dawley (Sas: CD (SD)BR), 12/sex/dose for neurobehavioral evaluation and 6/sex/dose (satellite group) evaluated for clinical pathology, 12 weeks old	Single gavage dose of NTN 33893 Technical (97.6 - 98.8% a.i.) At confirmed doses of 0 (vehicle), 42, 151 and 307 mg/kg bw.	Mortality in 4/18 high-dose males and 10/18 high-dose females within 24 hours of exposure. Dose-related increase in clinical signs (males > 151 mg/kg and 307 mg/kg females). All clinical signs and neurobehavioral effects are attributed to acute cholinergic toxicity. Recovery from all signs and neurobehavioral effects within 7 days. NOAEL (neurofunctional battery): 42 mg/kg LOAEL (females: decreased measures of motor and locomotor activity): 42 mg/kg; NOAEL (clinical chemistry): 42 mg/kg: decreased serum triglycerides; decreased serum potassium and cholesterol for females; decreased serum ALT; NOAEL (body weight, organ weights, gross and microscopic pathology): 307 mg/kg	Sheets 1994a MRID 43170301 **Note: The LOAEL of 42 mg/kg with an uncertainty factor of 300 is the basis for EPA's acute RfD for imidacloprid.**
Rat, Sprague-Dawley (Sas: CD (SD)BR), 12 females/dose for neurobehavioral evaluation , 12 weeks old	Single gavage dose of NTN 33893 technical (97.6 - 98.8% a.i.) at confirmed doses of 0 (vehicle) and 20 mg/kg bw.	NOAEL: 20 mg/kg bw. No mortality, clinical signs, effects on body weight. No neurological effects as tested in the first study above	Sheets 1994b MRID 43285801 (Supplemental transmission)
Subchronic			
Rat, Fischer, 18/sex/group, 12/group. evaluated for neurobehaviorial characteristics, 6/group evaluated for neuropathology	13-weeks Concentrations of 0, 140, 963 and 3027 ppm technical grade imidacloprid (97.6 - 98.8% a.i.) in the diet Doses: Males: 0, 9.3, 63.3 and 196 mg/kg bw/day Females: 0, 10.5, 69.1 and 213 mg/kg bw/day	No mortality. No treatment-related clinical signs. NOAEL (body wt., food consumption): 140 ppm NOAEL (neurobehavioral functional observational battery): 963 ppm mg/kg bw/day (males); 3027 ppm mg/kg bw/day (females) NOAEL (motor/locomotor activity): 3027 ppm NOAEL (clinical chemistry): 140 ppm No treatment-related gross lesions. No microscopic lesions in skeletal muscle or neural tissues.	Sheets and Hamilton 1994 MRID 43286401

See also Developmental Neurotoxicity Screening Study, Sheets 2001, MRID 45537501, Table A1-3.

See Section 3.1.6 for discussion.

A1 Table 11: Genotoxicity/Mutagenicity/Clastogenicity Studies

System	Compound	Response	Reference
Imidacloprid			
	Technical Grade		
Hamster, Chinese, 5 male and 5 female per group (3 exposed groups; 1 negative control and 1 positive control group), 25 -35 g, 8-12 weeks old	*In vivo* evaluation of clastogenic effects on bone marrow: single gavage dose of technical grade imidacloprid (94.6% a.i.) at 2000 mg/kg bw; sacrifice at 6, 24 and 48 hours post-exposure	No clinical signs or symptoms. Eating behavior was described as "normal". Mortality in 4/34 treated animals due to NTN 33893. No increased incidence of clastogenic effects in bone marrow DNA of NTN 33893 animals relative to controls	Herbold 1989b MRID 42256344
Hamster, Chinese, 5 male and 5 female per group; 28-32 g, 8-12 weeks old	*In vivo* evaluation of sister chromatid exchange in bone marrow: single gavage dose of technical grade imidacloprid (95.0% a.i.) Doses: 0, 500, 1000 and 2000 mg/kg bw, sacrifice at 24 hours post-exposure.	No mortality. No impact on DNA relative to controls.	Herbold 1989d MRID 42256346
Mouse, NMRI, 5 male and 5 female per group,28 - 41 g, 8-12 weeks old	In vivo micronucleus test, single gavage dose of technical grade imidacloprid (95.3% a.i.) Doses: 0 and 80 mg/kg bw; sacrifice at 24, 48 and 72 hours post-exposure	Apathy, reduced motility, and difficulty breathing for up to 6 hours after exposure; no mortality. No impact on DNA relative to controls.	Herbold 1988a MRID 42256347
Mouse, NMRI, 5 males per group	In vivo germ cell cytogenetic assay, single gavage dose of NTN 33893 (95.3% a.i.) Doses: 0 and 80 mg/kg bw; sacrifice at 24, 48 and 72 hours post-exposure	No mortality reported. No chromosomal aberrations in germ cells.	Volkner 1990 MRID 42256348
Human Peripheral blood lymphocytes	Imidacloprid (99.9%) Comet assay and Micronucleus test 0.2, 2, and 20 µM (about 0.05, 0.5, and 5 mg/L)	DNA damage only at highest concentration assayed.	Costa et al. 2009
Human Peripheral blood lymphocytes	Imidacloprid (99.9%) Sister-chromatid exchange assay, 0.1 to 100 mg/L	No significant increase in induction of sister-chromatid exchange. See Table 2 of paper.	Demsia et al. 2007
Rats, Wistar, 6 weeks old, 138 ±1.54 g	Imidacloprid (99.9%) Micronucleus induction at doses of 100, 200, and 300 mg/kg bw.	Significant change in micronuclei only at 300 mg/kg bw.	Demsia et al. 2007

System	Compound	Response	Reference
Human Peripheral blood lymphocytes	Technical grade (>95% a.i.) Comet assay and Micronucleus test: 0.05 to 0.5 mg/L.	NOAEL: 0.05 mg/L Increases in micronucleus and sister chromatid exchanges at higher concentrations.	Feng et al. 2005
	Formulations		
Human Peripheral blood lymphocytes	Gaucho 70WS (Mexican formulation). Comet assay: $2.8x10^{-4}$M to $1.7x10^{-3}$ (about 71 to 435 mg/L)	Concentration related increase incidence of DNA damage.	Calderon-Sequra et al. 2012
Human Peripheral blood lymphocytes	Confidor 200 SL formulation Comet assay and Micronucleus test 0.2, 2, and 20 µM (about 0.05, 0.5, and 5 mg/L)	DNA damage only at highest concentration assayed … *slightly more severe*… than a.i. See Figures 1 and 2 of paper.	Costa et al. 2009
Human hepatoma cells (HepG2 line)	Evidences (480 g a.i./L imidacloprid, Bayer S/A) Comet assay and Micronucleus test: 0.36, 3.6, and 36 mg a.i./L for 24 hours.	Micronucleus Assess: Increase in micronuclei at all concentrations but not dose-related (Table 1 of paper). Comet Assay: Increase in score at two lower concentrations but not at highest concentration (Table 3).	Bianchi et al. 2015 Brazil
Metabolite			
Mouse, NMRI, adult male and female, 8 weeks old, 5 per sex per group	WAK 3839 (a.k.a. NTN 37571) nitrosoimine metabolite, 98.9%. Doses: 0 and 100 mg/kg bw. Sacrifice after 24, 48 and 72 hours	No mortality. Apathy, staggering gait and difficulty breathing for up to 2 hours after dosing. External appearance, behavior and physical activity returned to normal thereafter. No treatment-related clastogenic effects on bone marrow cells.	Herbold 1989f MRID 42256368
Mouse, NMRI, 5 male and 5 female per group, 31 - 41 g, 8 - 12 weeks old.	WAK 3839 (a.k.a. NTN 37571) nitrosoimine metabolite, 98.9%. Doses: 0 or 50 mg/kg bw	No mortality or symptoms of toxicity for up to 2 hours post-treatment. No clastogenic effects in bone marrow erythroblasts comparison with negative vehicle and positive controls.	Herbold 1989e MRID 42256366
Mouse, BDF1, male, 9 weeks old	WAK 3839 (a.k.a. NTN 37571) nitrosoimine metabolite, 96.4%. Doses: 0, 100, 160, 200, 300 and 400 mg/kg bw. Observations at 30 hours after dosing.	In vivo micronucleus assay. No mortality was seen among mice dosed with 100 or 160 mg/kg. No treatment-related clastogenic effects in exposed mice (second study, doses up to and including 160 mg/kg).	Usami 1988b MRID 42256369

See Section 3.1.10 for general discussion.

Appendix 2: Toxicity to birds

Notes:

Values in parentheses are 95% confidence limits unless otherwise specified.

The ecological risk assessments from EPA are cited frequently. U.S. EPA/OPP/EFED (2007a) is abbreviated to EFED (2007a). U.S. EPA/OPP/EFED (2008a) is abbreviated to EFED (2008a).

A2 Table 1: Acute Oral/Gavage Toxicity to Birds

Species	Exposure	Response	Reference[1]
Technical Grade			
Bobwhite quail (*Colinus virginianus*) 20-week old, 5 male, 5 female per dose group	Technical grade imidacloprid (97.4% a.i.) Doses: 0, 25, 50, 100, 200, 400 or 800 mg/kg bw	LD_{50} = 152 (103 - 227) mg/kg bw, NOAEL (mortality, clinical signs) = 25 mg/kg bw Clinical signs: fluffed feathers, ataxia hypo-reactivity, immobility and wing drop. Significantly reduced bw on post-exposure day 7at doses > 100 mg/kg bw, with significantly decreased food consumption at 800 mg/kg bw.	Toll 1990a MRID 42055308

Appendix 2: Toxicity to Birds *(continued)*

Species	Exposure	Response	Reference[1]
Japanese Quail, (*Coturnix coturnix japonica*) 5 male and 5 female per dose, 9-12 weeks old	Technical grade imidacloprid (95.3% a.i.) Nominal Doses: 0, 2.5, 5, 10, 20, 40, and 80 mg/kg bw	LD_{50} = 31 (22-50) mg a.i./kg body weight. NOAEL (mortality) = 5 mg/kg bw; all deaths occurred in first 24 hours NOAEL (toxic signs) = 3.1 mg a.i./kg body weight based on measured concentrations. Clinical signs, slight apathy, tumbling and ptosis at 5 mg/kg bw to unconsciousness at 80 mg/kg bw, were reversible in surviving birds. Food consumption and weight gains were comparable to controls, except for the sole surviving bird in the 80 mg/kg bw group: food consumption was almost zero during the treatment period, but returned to almost normal during post-treatment, with no effect on weight gain.	Grau 1988b MRID 43310401
Canary/finch (*Serinus canarius*), 5 per dose	Technical grade imidacloprid (94.8% a.i.) Doses: 10, 12.5, 25 and 50 mg/kg bw	LD_{50} = 25-50 mg/kg bw Mortality in 1/5 and 5/5 at 25 and 50 mg/kg bw, respectively. NOAEL (mortality) = 12.5 mg/kg bw LOAEL (clinical signs) = 10 mg/kg bw, clinical signs including apathy and "cramps" and "jerks". (translation from German to English)	Grau 1994b MRID 43310403
Pigeon (*Columba livia*) 5 males and 5 females per dose	Technical grade imidacloprid (98.4% a.i.) Doses: 12.5, 25, 50 and 100 mg/kg bw, gelatin capsules	LD_{50}: 25 mg/kg bw (female); 25-50 mg/kg bw (male) NOAEL (mortality): 12.5 mg/kg bw LOAEL (clinical signs): 12.5 mg/kg bw, clinical signs including apathy, cramps and prone position.	Grau 1994b MRID 43310404

Appendix 2: Toxicity to Birds *(continued)*

Species	Exposure	Response	Reference[1]
Formulations			
House Sparrow (*Passer domesticus)*, adult, wild-capture, 7 per dose group	2.5 Granular (2.5% a.i.) Doses: 0, 1.5, 3, 6, 12, 25 and 50 mg a.i./kg bw	LD_{50} = 41 (24-260) mg a.i./kg bw (419 granules per sparrow), NOAEL (clinical signs) = 3 mg a.i./kg bw Mortality at doses > 12 mg a.i./kg bw Clinical signs: ataxia, hypo-reactivity, loss of flight, diarrhea, immobility and decreased activity on day of administration. Surviving birds fully recovered. No statistically significant effect on bw, though weights of dead birds were not included in the analysis. Food consumption not evaluated.	Stafford 1991 MRID 42055309
Not specified			
White leghorn chicken (*Gallus gallus domesticus*) Chicks (NOS)	Imidacloprid (NOS). Source and purity not reported. Not clear if this is a formulation or technical grade imidacloprid.	Apparent LD_{50}: 50 mg/kg bw Working Note: The source of the "apparent" LD_{50} is not specified or documented.	Balani et al. 2011

A2 Table 2: Acute Dietary Toxicity to Birds

Species	Exposure	Response	Reference[1]
Bobwhite quail (Colinus virginianus), 10-day old, 10 per concentration; 2 groups of 10 unexposed controls	Technical grade imidacloprid (94.8% a.i.) 5-day dietary exposure. Nominal concentrations: 78, 156, 312, 625, 1250, 2500 and 5000 ppm Measured concentrations: 69, 145, 285, 567, 1168, 2290 and 4649 ppm a.i. Working Note: Assuming an acute food consumption factor of 0.3 (kg food/kg bw) for quail, the LD_{50} corresponds to about 426 mg a.i./kg bw using the LC_{50} from the study and 460.8 mg a.i./kg bw using the EFED 2007a. The LOAEC corresponds to 27.6 mg a.i./kg bw.	LC_{50} = 1420 (713-4503) ppm LOAEC (mortality) = 69 ppm NOAEC: not determined Mortality observed > 69 ppm; Clinical signs among dying birds include: wing drop, ataxia, hypo-reactivity, immobility and diarrhea. Significantly decreased body weight on day 5 at concentrations > 567 ppm; However, exposed birds gained weight equal to controls during the post-exposure observation period (days 5 - 13). Significantly decreased food consumption > 285 ppm during exposure period only (food aversion), with birds > 2290 ppm only continuing to have decreased consumption during the observation period.	Toll 1990b MRID 42055310 This study is cited in U.S. EPA/OPP/ EFED 2007a. The EFED 2007a risk assessment reports a slightly different LC_{50} of 1,536 ppm. See Section 4.1.2.2.1 for discussion.
Japanese Quail, (*Coturnix coturnix japonica*) 10 per concentration, 10 days old	Technical grade imidacloprid (97.7% a.i.) 5-day dietary exposure. Nominal concentrations: 0, 313, 625, 1250, 2500 and 5000 ppm diet. Working Note: Assuming an acute food consumption factor of 0.4 (kg food/kg bw) for quail, the 100% lethal dose of 625 ppm corresponds to a dose of 250 mg a.i./kg bw.	1/10 mortality at 313 ppm. 100% mortality at remaining test concentrations. No control birds died. Clinical signs included apathy, diarrhea and narcotic effects. Survivors at the lowest test concentrations were symptom free by day 6.	Grau 1994a MRID 43310402 This study is not cited in U.S. EPA/OPP/EFED 2007a, 2008a.

Appendix 2: Toxicity to Birds *(continued)*

Species	Exposure	Response	Reference[1]
Mallard Duck (*Anas platyrhynchos*) 10-day old, 10 per concentration; 2 groups of 10 unexposed controls	Technical grade imidacloprid (94.8% a.i.) 5-day dietary exposure Nominal dietary concentrations of 78, 156, 312.5, 625, 1250, 2500 and 5000 ppm Mean measured concentrations of 69, 150, 270, 622, 1228, 2474 and 4797 ppm a.i. Working Note: Assuming an acute food consumption factor of 0.3 (kg food/kg bw) for mallards, the NOAEC for weight loss is about 20.7 mg/kg bw.	LC_{50} > 4,797 ppm. No mortality. Signs of ataxia in 1/10 at 2474 ppm. No treatment-related lesions upon post-mortem examination. Significantly decreased body weight on day 5 at >150 ppm. Food consumption trends support the observed decrease in body weight and the hypothesis that imidacloprid-treated food was not palatable. NOAEC (weight loss): 69 ppm	Toll 1991a MRID 42055311 This study is cited in U.S. EPA/OPP/ EFED 2007a.

A2 Table 3: Reproductive Toxicity

Species	Exposure	Response	Reference
Bobwhite Quail (*Colinus virginianus*) 18 pens per concentration tested 1male and 1 female per pen	Technical grade imidacloprid (94.8%) One-generation study 20-week dietary exposure Nominal Concentrations: 0, 30, 60, 120 and 240 ppm Mean measured concentrations of 0, 36, 61, 126 and 243 ppm.	<u>Parental generation:</u> Significantly reduced body weight, but not feed consumption among males exposed to 243 ppm. No signs of toxicity, no treatment-related gross lesions at sacrifice. Two deaths (a male at 61 ppm and a female at 126 ppm were not considered compound-related). No other mortality. <u>Offspring:</u> Significant reduction in hatchling body weights in comparison with controls at all concentrations. However, significantly increased 14-day survivor weights at 126 and 243 ppm, in comparison with controls, and equal or greater than numbers surviving among imidacloprid-treated offspring. A small decrease in eggshell thickness at 61 (0.34 mm), 126 (0.34 mm) and 243 ppm (0.33 mm), was observed in comparison with controls (0.35 mm). The difference was statistically significant for the 61 and 243 ppm birds. However, no reduction in shell strength, increase in percentage of cracked eggs or decrease in hatchability was observed at these concentrations. NOAEC: 36 ppm (≈2.52 mg a.i./kg bw[1]) LOAEC: 61 ppm based on egg shell thinning (≈4.27 mg a.i./kg bw/day[1])	Toll 1991b MRID 42055312 Also summarized in EFED 2007a, p. 39 Classification: Core
Mallard Duck (*Anas platyrhynchos*) 15 pens with 1 male and 1 female	Technical grade imidacloprid (94.8%) One-generation study, 20-week dietary exposure Nominal concentrations: 0, 60, 120 and 240 ppm Mean measured concentrations: 0, 64, 125 and 234 ppm	No effects on parental birds other than sporadic but significant decreases in mean weekly feed consumption. 234 ppm: Significant reduction in mean number of eggs laid per hen, resulting in reductions in mean number of hatchlings per hen, percentage of normal hatchlings of viable eggs, percentage of normal hatchlings of live three-week embryos and percentage of 14-day old survivors per hen. NOAEC: 125 ppm (≈8.75 mg a.i./kg bw [1]) LOAEC: 234 ppm (≈16.38 mg a.i./kg bw [1])	Toll 1991c MRID 42055313 Also summarized in EFED 2007a, p. 39 Classification: Supplemental

Appendix 2: Toxicity to Birds *(continued)*

Species	Exposure	Response	Reference
Mallard Duck (*Anas platyrhynchos*) 15 *adult* male/female pairs per dose	Technical grade imidacloprid (96.0%) Eggshell quality one-generation study, 19 weeks. Nominal concentrations: 0, 25, 40 and 55 ppm a.i. Mean measured concentrations: 0, 22, 35 and 47 ppm a.i.	No differences in eggshell strength or thickness between controls and any treatment group. No statistically significant differences between controls and any treatment level with respect to body weight, food consumption, clinical signs (none) or mortality (none). NOAEC: 47 ppm a.i (≈3.39 mg a.i./kg bw [1]) Working Note: EFED (2007a, p. 39) cites a LOAEL for 61 ppm attributed to this study. The LOAEL of 61 ppm, however, appears to be from Stafford (1992, MRID 42480502) as summarized below. In any event, EFED (2007a) has reclassified 61 ppm as a LOAEC based on egg shell thinning.	Hancock 1994b MRID 43466501 Also summarized in EFED 2007a, p. 39 Classification: Supplemental (EFED 2008a, p. 32)..
Mallard Duck (Anas *platyrhynchos*) 15 male/female adult pairs per treatment	Technical grade imidacloprid (94.8%) One-generation study, 20-week dietary Nominal concentrations: 0, 60, 120 and 240 ppm Mean measured concentrations: 0, 61, 128 and 250 ppm	Statistically significant reduction in eggshell thickness and strength at 250 ppm. There was a statistically significant increase in number of cracked eggs at 128 ppm. No clinical signs of toxicity, no effects on mortality, no treatment-related lesions and no statistically significant differences in parental body weight, food consumption, egg production, egg viability, 21-day embryo survival, hatchability, hatchling body weight, 14-day survival or survivor body weights were observed. NOAEC: 61 ppm (≈4.27 mg a.i./kg bw [1]) LOAEC: 128 ppm (≈8.96 mg a.i./kg bw [1])	Stafford 1992 MRID 42480502 Note: No effect on eggshell thickness in Hancock (1994b), see above. Not summarized in EFED 2007a. Cited but not discussed in EFED 2008a.

[1] Dietary concentrations (ppm) converted to mg/kg bw doses using food consumption rates of 0.07 kg food/kg bw for reproduction studies in quail and mallards taken from SERA (2007b).

A2 Table 4: Subchronic Toxicity

Species	Exposure	Response	Reference
White leghorn chicken (*Gallus gallus domesticus*) Chicks (NOS), 25 per group	Imidacloprid (NOS). Source and purity not specified. Doses administered in groundnut oil (vehicle). Not clear if a formulation was used. 28 day exposure Group — Daily Dose (mg/kg bw/day) C1 — 0 C2 — 0 (Vehicle) I1 — 1.25 I2 — 1.67 I3 — 2.5 Blood samples collected at 14 and 28 days.	14 days Decrease in blood glucose at high dose only. 28 days Statistically significant and dose-related decrease in blood glucose at all doses. Statistically significant and dose-related increase in SGOT (liver toxicity) at all doses. Statistically significant decrease in total leucocyte count in high dose group (≈82% of controls). Modest (≈7%, N.S.) decrease in mid-dose group. Working Note: A complete copy of Table 2 is available on-line. See the last page of the pdf for best copy that could be downloaded.	Balani et al. 2011 India
Red-legged partridges (*Alectoris rufa*), captive born, 1 year-old. Housed as breeding pairs. 12 birds per group.	Escocet (35% a.i. w/v, Bayer CropScience, Alcacer, Spain). Imidacloprid administered in wheat seeds. Nominal Conc.: 700 and 1,400 mg a.i./kg seed Measured: 519 and 869 mg a.i./kg seed. Average consumption in birds of 25 g/day at an average body weight of 407 g. [consumption factor of 0.061 kg seed/kg bw] Average doses (from authors): 31.9 and 53.4 mg/kg bw/day Period of exposure: 10 days.	Working Note: Doses are near to or above LD_{50} for other species. See Table A2-1. Dose related increase in mortality: 0 (control), 8.3%, and 58.3%. Significant and dose-related decreases in hematocrit, albumin, alkaline phosphatase, calcium, cholesterol, and total protein. Significance decrease in aspartate aminotransferase only in high dose group. See Table 1 of paper. Significant and substantial decrease in glutathione (≈57% of controls, oxidative stress) at high dose. See Table 2 of paper. Decrease in egg shell thickness but only at low dose. Impaired cellular immune response, dose-related (Fig. 1). Dose-related decrease in number of chicks and chick survival at both doses (Table 3). Working Note: No significant decrease in glucose. Compare with Balani et al. 2011.	Lopez-Antia et al. 2013 Spain

Appendix 2: Toxicity to Birds *(continued)*

Species	Exposure	Response	Reference
Red-legged partridges (*Alectoris rufa*), captive born, 51 females and 45 males. Housed in pairs.	Escocet (35% a.i. w/v, Bayer CropScience, Alcacer, Spain). Imidacloprid administered in wheat seeds. Doses: 0, 8.8, and 44 mg/kg bw/day Test 1: November, 25 day duration Test 2: March, 10 day duration	High dose: 100% mortality in both sexes. Mean survival time of 12.7 days for males and 6.7 days for females. No effect on brain AChE. Working Note: No high dose birds survived to Test 2. Low Dose: Reduced body condition and significant (p=0.005) decrease in body weight following test 1. No effect on body condition and marginal (p=0.055) decrease in body weight following Test 2. Significant reduction in clutch size and significant increase in time to first egg (Table 2 of paper). Significant decrease in wing web swelling (assay for cellular immunity).	Lopez-Antia et al. 2015 Spain
Red Munia [a.k.a. strawberry finch] (*Amandava amandava*), 8.5±0.5 grams 8 males per treatment group	Confidor formulation (NOS), 17.8% a.i. w/w. Different groups dosed in mid-July to August (pre-breeding) and mid-September to October (breeding). Dietary exposure with measured food consumption. Dose: 0 (control) or 0.15 mg a.i./kg bw Duration: 30 days Working Note: Dose expressed as 0.5% of oral LD50 of 31 mg/kg bw. See Grau 1988b in Table A2- 1 of this appendix.	Body Weight: ≈5.5% decrease in breeding birds by DAT 30 ($p<0.01$) (Table 1 of paper). Thyroid weights: No significant change (Table 1 of paper). Decrease in T4 (thyroxine) in both groups of dosed birds. Increase in T3 (triodothyronine) in pre-breeding but decrease in breeding stage birds. Decrease in TSH (thyroid - stimulating hormone) in both groups (Figure 2 of paper). Pathologic changes in thyroid follicles and stroma that was more severe in breeding phase birds.	Pandey and Mohanty 2015 India

A2 Table 5: Food Aversion Studies

Application	Exposure	Observations	Reference
Red-Winged Blackbird (*Agelaius phoeniceus*) Wild-captured males, 8 per concentration in cup tests; 10 per flight pen in each replicate of the flight pen tests.	Concentrations: 0, 278, 833 and 2500 ppm 4-day two-cup tests: birds are presented with feed in two cups: 1) control and treated seed undyed; 2) control and treated seed both dyed; 3) Control seed undyed, treated seed dyed.	Test 1 and 2) Significantly lower consumption of treated rice compared with controls in birds given choice between untreated rice and rice treated at 833 and 2500 ppm. Test 3) Significant reduction in consumption of treated rice versus untreated rice at all levels. Dose related increase in consumption disparity between treated and untreated cups.	Avery et al. 1993a,b MRID 42856201 See supplemental information below.
Red-Winged Blackbird (*Agelaius phoeniceus*) Wild-captured males, 8 per concentration in cup tests	Concentrations: 0, 278, 833 and 2500 ppm 4-day once-cup tests: birds are presented with feed in two cups: 1) control and treated seed undyed; 2) control and treated seed both dyed; 3) Control seed undyed, treated seed dyed.	4-day one-cup test: Rice consumption measured in 4-day pre-treatment period and compared with that in 4-day treatment period. Birds given one cup at the specified treatment level, with all seed dyed. Average reduced consumption of 1.08 g/bird and 2.49 g/bird at 833 and 2500 ppm , respectively, in comparison with pre-treatment consumption levels. No difference between pre-treatment and treatment consumption rates seen at 0 or 278 ppm.	Avery et al. 1993a,b MRID 42856201 See supplemental information below.
Red-Winged Blackbird (*Agelaius phoeniceus*) Wild-captured males, 10 per flight pen in each replicate of the flight pen tests.	6 replicate Flight Pen tests: 8 plots per pen, only 2 randomly selected plots were used in a test, one treated (800 grams of 2500 ppm imidacloprid-treated rice, one untreated control (800 grams untreated rice).	Over a 4-day period, more seed was removed from control plots (mean 41.1% + 10.4% standard error) than from treated plots (mean 8.8% + 3.7% standard error).	Avery et al. 1993a,b MRID 42856201 See supplemental information below.

Appendix 2: Toxicity to Birds *(continued)*

Application	Exposure	Observations	Reference
Ringed turtle dove (*Streptopelia risoria*)	Seed avoidance 5-day pre-treatment period, followed by 2-day break, then 5-day treatment period. Nominal (measured) concentrations on wheat: 313 (228) and 1250 (1058) ppm a.i.; on sorghum: 2500 (2354) and 5000 (4612) ppm a.i. Comparison with untreated seed for controls.	Significantly reduced body weight and seed consumption in comparison with controls in both seed trials at all imidacloprid concentrations tested. Dose-related clinical signs (hypoactivity, fluffed feathers, vomiting) in all but one bird. Mortality only in trial with sorghum, with one death at 2354 ppm a.i. and 4 at 4612 ppm a.i.	Hancock 1994a MRID 43197501 See supplemental information below
House Sparrow (*Passer domesticus*)	Seed avoidance 5-day pre-treatment period, followed by 2-day break, then 5-day treatment period. Nominal (measured) concentrations on wheat: 313 (228) and 1250 (1058) ppm a.i.; on sorghum: 2500 (2354) and 5000 (4612) ppm a.i. Comparison with untreated seed for controls.	Significantly reduced body weight in comparison with controls only at 4612 ppm a.i. in the sorghum trial. Significantly reduced food consumption for all birds exposed to imidacloprid-treated seeds in comparison with controls. Clinical signs (hypoactivity, ataxia, fluffed feathers) in 2 birds at each of the imidacloprid-treated groups for the sorghum trial only. No treatment-related mortality.	Hancock 1994a MRID 43197501 See supplemental information below
Red-legged partridges (*Alectoris rufa*), housed in pairs, 6 pairs/group	Escocet (35% a.i. w/v, Bayer CropScience, Alcacer, Spain). Concentrations: 0, and 700 mg/kg wheat seed. Food avoidance: treated vs untreated.	Clear and statistically significant ($p<0.001$) preference for untreated seed. Three birds died during testing but differences in survival among groups not significant.	Lopez-Antia et al. 2014
Red-legged partridges (*Alectoris rufa*), housed in pairs, 6 pairs/group	Escocet (35% a.i. w/v, Bayer CropScience, Alcacer, Spain). Concentrations: 0, and 700 mg/kg wheat seed. Food avoidance: Variable number of feeders (2,4,8,16). Half treated and half untreated.	Substantial preference for untreated seed. See Table 1 of paper. As the number of feeders increased, the consumption of imidacloprid also increased. Avoidance may be due to taste aversion.	Lopez-Antia et al. 2014

Supplemental information for Avery et al. 1993a,b

Cage and flight pen evaluation of avian repellency and hazard associated with imidacloprid-treated rice seed.

In the flight pen studies, investigators observed an inverse relationship between the number of treated seeds removed and the mean minimum temperature during the test.

Treated seed removal also appeared to be increased by the presence of predators outside the pen during trials. Residue analysis indicated that birds ingested 13-16% of the imidacloprid present on the seed. With this information, the investigators stated that birds feeding at an average rate of 6 seeds/minute (seed treated with 2500 ppm imidacloprid) would consume only a fraction of the LD50 dose (they used the house sparrow LD50 of 41 mg/kg from Mullins 1993 as the basis for comparison).

Supplemental information for Hancock 1994a:

The investigator observed that both species learned to avoid imidacloprid-treated seed through post-digestive distress. Hancock hypothesizes that doves were more sensitive than sparrows due to differences in eating habits. Doves consumed large numbers of seed during the initial visit to the feeder, while sparrows consumed fewer seeds per visit. As such, doves were exposed to higher internal doses of imidacloprid than sparrows. Due to the slower rate of ingestion, sparrows learned avoidance, which resulted in lower exposure and toxicity. Hancock estimated the dose for doves exposed to 4612 ppm-treated sorghum to be 47 mg/kg body weight (based on observed seed consumption and regurgitation, and assumes 100% absorption of non-regurgitated seed, 38% absorption of regurgitated seed and a 150 g body weight).

A2 Table 6: Field Studies

Application	Exposure	Observations	Reference
Mixed species: American Robin (*Turdus migratorius*), northern cardinal (*Cardinalis cardinalis*), gray catbird (*Dumetella carolinensis*), blue jay (*Cyanocitta cristata*), brown thrasher (*Toxostoma rufum*), northern mockingbird (*Mimus polyglottos*), rufus-sided towhee (*Pipilo erythrophthalmus*)	Merit 0.62% Granular applied to golf course turf at 0.5 lb a.i./acre. 8 golf courses, 1 treatment and 1 control plot each. Average number of birds banded = 107 (control) and 98 (treated plots). All courses were similar in species diversity. The percentage of marked birds surviving 5-7 days after treatment was determined visually and by radio telemetry. Measured maximum daily mean imidacloprid residues were: 0.38 ppm in soil, 13.36 ppm in turf verdure, 0.94 ppm in puddle water and 2.21 ppm in invertebrates.	There was no treatment-related effect on survival or percent mortality based on two null hypotheses (survival of species on treated sites is reduced by 20% or more). No difference in mortality between control and treated sites). Of the 55 intact carcasses collected after the study, only 4 had detectable residues of imidacloprid, ranging from <1% to 10% of the lowest LD_{50} for terrestrial vertebrates.	Toll and Fischer 1993 MRID 42737101 Not cited in EFED 2007a. Cited but not discussed in EFED 2008a.

Appendix 2: Toxicity to Birds *(continued)*

Application	Exposure	Observations	Reference
Insectivorous birds in Netherlands.	Population survey associating bird populations with concentrations of imidacloprid and surface water. Covered bird population data from 1994 (prior to imidacloprid use) and period from 2003 to 2010 (after imidacloprid use). **Working Note: Author's discussion notes that bird populations in Europe have been declining for the past 30 years.**	Statistically significant declines in Eurasian skylark (*Alauda arvensis*), Barn swallow (*Hirundo rustica*), Yellow wagtail (*Motacilla flava*), Common starling (*Sturnus vulgaris*), Common whitethroat (*Sylvia communis*), and Mistle thrush (*Turdus viscivorus*). **Working Note: The statistical analysis does include the Bonferroni correction for multiple comparisons.** Author's Conclusion: *At imidacloprid concentrations of more than 20 nanograms per litre, bird populations tended to decline by 3.5 per cent on average annually.* Authors discuss the possibility that the effects on birds are attributable to decreases in insect populations rather than a primary toxic effect in birds.	Hallman et al. 2014

Appendix 3: Terrestrial Invertebrates.

General Notes on Appendix 3:

Values in parentheses are 95% confidence limits unless otherwise specified.

The ecological risk assessments from EPA are cited as:

U.S. EPA/OPP/EFED (2007a) is abbreviated to EFED (2007a),
U.S. EPA/OPP/EFED (2008a) is abbreviated to EFED (2008a).

References to tables or figures typically refer to the tables or figures in the paper being addressed. Cross references to tables within this appendix are always made with reference to this appendix – e.g., "Table A3-2".

Subspecies and varieties are given in the first column of the tables when specified in the papers.

NOTE: Unlike other appendices, the doses/concentrations are identical to those given the cited publications in both the Exposure and Response columns. Take particular care when developing comparisons to the units of exposure or dosing.

A3 Table 1: Honeybee

Honey bee	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
	Oral		
2 groups of 10 each per concentration	TGAI (99.8% a.i.) Doses: 0.0015, 0.0031, 0.0063, 0.0125, and 0.025 µg/bee	48 h-LD_{50}: 0.0037 (0.0026 - 0.0053) µg/bee LOAEL: 0.0015 µg/bee (20% mortality) Working Note: This is cited in U.S. EPA/ OPP 2007a, p. 40, as a contact assay of 0.0039 µg/bee.	Cole 1990 MRID 42273003
Africanize bees Newly emerged	TGAI (92.5%) 48 hours Sucrose, oral 5 doses, 80-100 ng/µL	48-hour LD_{50}: 80.9 ng/bee Slope 2.781 Working Note: See sublethal study below in this table.	de Almeida Rossi et al. 2013 Brazil
Africanize bees >20 days old, 10 adult bees per dose	Imidacloprid (NOS) Oral doses of 0, 0.4, 0.2, 0.1, 0.05 and 0.025 µg/bee. Observation period: 120 minutes	2-hour LD50: 0.1 µg/bee. Working Note: This is an atypically short period of observation but the LD50 corresponds to 100 ng/bee, very similar to the LD50 of 80.9 ng/bee from de Almeida Rossi et al. (2013) as summarized above.	Carrillo et al. 2013 Brazil
Workers, unknown age. Late summer bees. 3 replicates per concentration, 5 conc. 180-360 bees per group.	TGAI (94-98% from different sources) Oral in sucrose Concentrations: 0.2 to 3.2 mg/L Each bee consumed 10 µL.	48-hour LD_{50}: 30.6 (26.7 - 36.3) ng/bee Working Note: See matched data below on 5-OH metabolite.	Decourtye et al. 2003 France

<table>
<tr><th>Honey bee</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Seven tests conducted with bees from seven different apiaries in Germany (5), the Netherlands (1) and the United Kingdom (1). Each test used adult workers, 14-42 days old, 10 bees per dose, 3 replicates per dose</td><td>TGAI in sucrose
Nominal doses: 0.1 - 81 ng a.i./bee for 3-4 hours.
Mortality was assessed at 4, 24 and 48 hours.

Working Note: Compare with metabolite data from this study below.</td><td>48 h-LD_{50} = 41 to >81 ng a.i./bee
See Table 2: Only one of 7 bioassays yielded a definitive LD_{50}.

NOAEL (mortality) : 1.5 ng a.i/bee

LOAEL (mortality): >3.1 ng/bee (17-50% mortality)</td><td>Nauen et al 2001</td></tr>
<tr><td>10 bees per dose,
Bees from three apiaries in the UK (a) , The Netherlands (b) and Germany (c),</td><td>TGAI
separate tests at 3 different facilities

Working Note: See paper for calculation of dietary LC_{50} values. Marginal relevance?</td><td>48-h LD_{50}s
<table><tr><th>Facility</th><th>LD_{50} (ng/bee)</th></tr><tr><td>UK</td><td>3.7</td></tr><tr><td>Netherlands</td><td>>21</td></tr><tr><td>Germany</td><td>40.9</td></tr></table>
Working Note: The U.K. data looks like Cole 1990 and the Germany data looks like Suchail et al. 2011. Exclude from analysis.</td><td>Schmuck et al. 2001</td></tr>
<tr><td>3 cages of 20 bees each per experiment, each experiment replicated 3 times</td><td>TGAI
Working Note: Compare with metabolite data from this study below.</td><td><table><tr><th>Hours</th><th>LD_{50} (ng/bee)</th></tr><tr><td>48</td><td>57</td></tr><tr><td>72</td><td>37</td></tr><tr><td>96</td><td>37</td></tr></table></td><td>Suchail et al. 2001</td></tr>
<tr><td></td><td>Contact</td><td></td><td></td></tr>
<tr><td>Appears to use 2 replicates of 10 bees per replicate per dose.
Age >20 days.</td><td>TGAI (95%)
Direct spray
24-hour exposure</td><td>24h-LC_{50}: 2.2 (1.8-3.4) % x10^{-3}
Working Note: This can be used with the study by Scott-Dupree et al. 2009 [Table A3-3] to examine sensitivities relative to other species. Same groups of investigators.
1% x 10^{-3} corresponds to 10^{-5} or 1 in 100,000. Assuming w/v units above, the LC_{50} corresponds to 22 ppm.</td><td>Bailey et al. 2005</td></tr>
</table>

Honey bee	Exposure	Response	Reference
2 groups of 10 each per concentration	TGAI (99.8% a.i.) Contact Doses: 0.025, 0.05, 0.10, 0.20 and 0.40 µg/bee	LD_{50}: 0.078 (0.0055 - 0.0119) µg/bee LOAEL: 0.025 µg/bee (20% mortality) [≈0.21 mg a.i./kg bw using a bw of 116 mg from Winston (1987)]	Cole 1990 MRID 42273003
Newly emerged, 30 bees per dose	Imidacloprid ('pure' Sigma-Aldrich) Topical Doses: 2.5, 5, 10, 20, 30, 40, and 50 ng/bee 48-hours	Contact LD_{50}: 21.21 (18.77-23.65) ng/bee (Fig. 52 in supplementary information on-line). The duration associated with the LD_{50} value is not clear.	Di Prisco et al. 2013
Adult workers n=137	TGAI (>99%) Topical, 1 µL solution	LD_{50}: 0.0179 (0.0092-0.0315) µg/bee	Iwawa et al. 2005
10 bees per dose, 3-5 replicates per dose adult workers, 14-42 days old	TGAI Topical Nominal doses: 40 to 154 ng a.i./bee. Mortality was assessed at 4, 24 and 48 hours.	48-hour LD_{50} = 62.4 (range of 42 to 104) ng a.i./bee See Table 2: 6 different assays from different facilities.	Nauen et al 2001
Formulations			
	Oral		
10 bees per dose, Bees from Germany	Bayer WG70 (700 g/kg)	48-h LD_{50}: 11.6 ng/bee	Schmuck et al. 2001
10 bees per dose, Bees from Germany	SC200 (200 g/L)	48-h LD_{50}: 21.2 ng/bee	Schmuck et al. 2001
	Contact		
Adult bees, newly emerged (24 hours for males and 24-72 hours for females). A total of 60–135 per assay, at least 5 doses	Provado 1.6F (17.4% a.i.) Topical application, 1 µL per bee 48-hours	48-h LD_{50}: 0.2 (0.1-0.3) µg/bee	Biddinger et al. 2013
10 bees per dose, Bees from Germany	Bayer WG70 (700 g/kg)	48-h LD50: 242.6 ng/bee	Schmuck et al. 2001

Honey bee	Exposure	Response	Reference
10 bees per dose, Bees from Germany	SC200 (200 g/L)	48-h LD50: 59.7 ng/bee	Schmuck et al. 2001
Longer-term			
Winter Newly emerged worker bees, 60 - 163 bees per treatment	TGAI (99.4% a.i.) 11 day exposure Concentrations: 0, 1.5, 3, 6, 12, 24, and 48 µg a.i./kg sucrose solution.	48 µg/kg solution: Significant increase in mortality (20.5%) versus controls (11.6%). NOAEC: 24 µg/kg solution (≈0.97 ng/bee) [1]	Decourtye et al. 2003
3 cages of 30 bees each per experiment, each experiment replicated 3 times	**TGAI and metabolites** 10 day exposure Concentrations: 0, 0.1, 1, and 10 µg/L food. Bee consumption: ≈1 µL/day Doses: 0.010, 0.1 and 1 ng/bee/day	Control mortality < 15%. Imidacloprid and all metabolites caused mortality within 72 hours after the onset of intoxication (trembling, tumbling, coordination problems). 50% mortality was reached by day 8 for all metabolites tested except 0.1 µg/L imidacloprid (significant lower mortality for entire duration of study in comparison with higher doses) and 0.1 µg/L 5-OH imidacloprid (reached 40% mortality by end of study). All metabolites yielded similar timing of mortality. Only imidacloprid and 5-OH-imidacloprid showed evidence of dose-response relationship.	Suchail et al. 2001
2 cages of 50 bees each for imidacloprid treatments, 3 cages, 50 bees each for controls	TGAI (99.8%) Concentrations: 0, 4 and 8 µg/L in sucrose. Measured consumption: ≈20 µL sucrose solution per bee per day. Exposures up to 60 days Average doses: 0, 0.08 and 0.16 ng a.i./bee/day.	Sharp increase in cumulative mortality at both doses by day 40 (≈80% mortality). No clear dose-response relationship. Control mortality 40%. 100% mortality between days 40 and 50. For controls, 100% mortality by Day 60. No difference in food consumption between controls and exposed bees.[2]	Dechaume Moncharmont et al 2003
Africanize bees >20 days old, 10 adult bees per dose	Imidacloprid (NOS) Oral dose at 2-hour LD_{50} dose of 0.1 µg/bee. Duration not clear but it appears to be 120 minutes.	2-hour LD50: 0.1 µg/bee. No effect on proboscis extension reflex but a decrease in response to floral odor citral (learning performance). Working Note: Based on the description on p. 433, column 1, first full paragraph, these "sublethal" effects were assayed at the LD_{50}.	Carrillo et al. 2013 Brazil

Honey bee	Exposure	Response	Reference
Brazil **Africanize bees** Newly emerged 10 bees/cage, 3 cages per dose.	TGAI (92.5%) Exposure up to 10 days. Sucrose, oral Doses: 0.809 (LD_{50}/100), 1.618 (LD_{50}/50), and 8.09 (LD_{50}/10) ng/bee	Brain cells: No effects at two lower concentrations. Cell damage (chromatin condensation, cell swelling, cell death in Kenyon cells) at highest dose. Optic lobes: Effects (cell pathology and cell death) at all doses. More rapid onset at higher doses (Table 2 of paper). Obvious potential to impact foraging.	de Almeida Rossi et al. 2013
Sublethal Effects			
30 bees per cage	Imidacloprid (NOS); might be a formulation. Contact assay. Nominal doses: 0.7, 7, and 70 µg/kg bw. Few details of experimental protocols.	Modest increase in mortality with imidacloprid. Not clearly dose-related. Possible enhanced mortality with *Nosema* pathogen (Fig. 1 of paper). Authors do not analyze based on an additive model. Separate assays for social immunity (glucose oxidase activity) significantly decreased only in combination of imidacloprid and *Nosema*.	Alaux et al. 2010
Worker bees, 10 per dose	TGAI in DMSO Topical Thorax: 0, 1.25, 2.5, 5, 10 and 20 ng/bee	1.25 µg/bee: reduced habituation of proboscis extension and increased motor activity. 2.5 - 20 ng/bee: dose-related impairment of activity	Lambin et al. 2001
Bees of various ages (4, 5, 6, 7, 8, 9 and 10 days old)	TGAI 0.1, 1 and 10 ng/bee	Irregular and age dependent alteration of proboscis extension reflex.	Guez et al. 2001
Winter bees newly emerged worker bees, 60 - 163 bees per treatment	TGAI (99.4%) 11 days feeding Concentrations: 7.5 - 240 µg/kg in sucrose Assay for olfactory learning performance	No significant change in proboscis reflex response. NOAEC: >48 µg/kg sucrose. Working note: See data on 5-OH metabolite below.	Decourtye et al. 2003
Summer bees newly emerged worker bees, 60 - 163 bees per treatment	TGAI (99.4%) 11 days feeding Concentrations: 7.5 - 240 µg/kg in sucrose Assay for olfactory learning performance	Significant decrease in proboscis reflex response at 48 mg/kg sucrose. NOAEC: 24 µg/kg sucrose. Working note: See data on 5-OH metabolite below.	Decourtye et al. 2003

Honey bee	Exposure	Response	Reference
Emerging bees.	TGAI (NOS) Concentrations: 1.3 (Exp. 1) or 2 µg/L (Exp. 2) with or without co-exposure to *Nosema ceranae* pathogen. With DMSO control. Ad libitum for 7 days Up to 12 day observations.	Treatment with imidacloprid at 2 µg/L had a marginal impact on survival (p = 0.079). Highly significant mortality with pathogen (p<0.001). Bee consumption: Uninfected bees: 45.1±17.6 ng/day/bee Infected bees: 42.2±13.5 ng/day/bee. Down-regulation of genes involved in immunity in imidacloprid-pathogen exposed groups.	Aufauvre et al. 2014
Groups of 10 bees (caged), large number of cages but distribution unclear.	TGAI (NOS) Sucrose concentrations: 0.08, 0.20, 0.51, 1.28, 3.20, 8.00, 20.0, 50, and 125 µg/L . 4 day exposure	No remarkable impact on feeding rate, locomotion, or longevity (See triangles in Figure 1 of paper). Working Note: Contrast with effects in bumblebees.	Cresswell et al. 2014
Newly emerged, 30 bees per dose	Imidacloprid ('pure' Sigma-Aldrich) Topical Doses: 2.5, 5, 10, 20, 30, 40, and 50 ng/bee	Suppression of immune function assayed by enhanced replication of deformed wing virus. NOEC/LOEC appears to be at 5/10 ng/bee (Figure 4). Contact LD50: 21.21 (18.77-23.65) ng/bee (Fig. 52 in supplementary information on-line).	Di Prisco et al. 2013
Apis mellifera ligustica 314 nectar and 209 pollen foragers from 3 colonies.	Imidacloprid (NOS) Oral (micropipette): 0.21 ng (24 ppb) or 2.16 ng (241ppb) Assayed at 1 and 24 hours post-dosing	Decrease sucrose responsiveness (proboscis extension response) after 1 hour at both doses. Most pronounced in nectar foragers. No significant effect at 24 hours post-dosing.	Eiri and Nieh 2012
Apis mellifera ligustica 65 bees from two colonies	Imidacloprid (NOS) Oral (micropipette): 0.21 ng (24 ppb) or 2.16 ng (241ppb) Assayed at 24 hours post-doing.	Fewer waggle dance circuits at 24 hours.	Eiri and Nieh 2012
Bee larvae, 24 larvae per group were	Imidacloprid (NOS) 200 ppm in larval diet.	Changes in gene expression. Significant increases in transcript levels for PROact, PGRP, Haxam, and DWV.	Gregorc et al. 2012
Emerging bees Groups of 40	Imidacloprid (NOS) Cotton (conventional) pollen at 48 ppb (ng/g) 7-day exposure	Decreased learning behavior in T-tube maze.	Han et al. 2010b

Honey bee	Exposure	Response	Reference
Emerging bees 3 replicates per treatment Number of individuals per replicate not clear.	Imidacloprid (NOS) Cotton (conventional) pollen at 48 and 96 ppb (ng/g) 7-day exposure	Significant increase in mortality at 96 ppb. Decrease in hypopharyngeal gland protein at 48 ppb (Fig. 1). No significant impact on midgut proteolytic enzyme activity.	Han et al. 2012
Full sized colonies of bees (30–40,000 adults) 30 colonies, 3 groups of 10 colonies.	Imidacloprid (NOS) Protein supplement patties: 5 and 20 ppb. 10 week exposure Challenge with *Nosema* spore suspension at week 5. Single brood frames. 30 bees per cage.	Substantial increase in Nosema growth in imidacloprid exposed bees but no dose-response relationship (see Fig. 1 of paper). No imidacloprid detected in emerged bees. Decrease in bee weight in 1 of 2 trials (Table 1).	Pettis et al. 2012
Newly emerged. 28 controls, exposed groups from 12 to 28 bees (Fig. 2) Video tracking,	TGAI (commercial source) Sucrose agar: 0.0, 0.05, 0.5, 5.0, 50, and 500 µg/L. 24-hour observations.	Altered behaviour at 2 higher doses (significant increase in time near food, decrease in distance traveled. Apparent dose-related decrease in time interacting but not statistically significant (Fig. 2).	Teeters et al. 2012
Apis mellifera mellifera Adult workers 20 to 26 bees per dose group (Table 2)	Imidacloprid (>99%) Sucrose solution. 4-days Concentrations.: 10 nmol/L, 100 nmol/L, and 1 µmol/L [≈0.256, 2.56, 25.6 µg/L]	About 80% mortality at 1 µmol/L. No increased mortality at other doses. Only two lower doses used for memory experiments. Mean sucrose consumption of 27.5 mg/day or 487 mg over experiment. Dose estimates: 1.3 ng/bee at 10 nmol/L over 6 day period. Dose-related suppression of conditioned proboscis extension response at both doses. Recovery after 3 days (Table 1).	Williamson and Wright 2013
Apis mellifera mellifera >30 bees per treatment	Imidacloprid (>99%) Sucrose solutions: Estimated dose of 1.28 ng/bee	No marked impact on learning and memory. Modest improvement of memory when combined with sublethal dose of coumaphos.	Williamson et al. 2013
Apis mellifera var. Buckfast	Imidacloprid (NOS) Sucrose Solution: 0.01, 0.1, and 1 µM. Doses: 0.401 and 3.7 ng/bee for two lower concentrations. 24 hours	Signs of neurotoxicity – i.e., loss of postural control at lowest dose. More severe response at higher doses. Not all responses show clear dose-response relationship (see figure on p. 1415).	Williamson et al. 2014

Honey bee	Exposure	Response	Reference
Metabolites			
Workers, unknown age. Late summer bees. 3 replicates per concentration, 5 concentrations 180-360 bees per group.	5-Hydroxy metabolite (99.4%) Oral in sucrose Concentrations: 1.25 to 20 mg/L Each bee consumed 10 µL.	48-hour LD_{50}: 153.5 (125.9-196.9) ng/bee Compare to matched assay on TGAI.	Decourtye et al. 2003
Winter bees newly emerged worker bees, 60 - 163 bees per treatment	5-OH metabolite (99.4%) 11 days feeding Concentrations: 7.5 - 240 µg/kg in sucrose Assay for olfactory learning performance	Significant change in proboscis reflex response. NOAEC: 24 µg/kg sucrose. LOAEC: 48 µg/kg sucrose	Decourtye et al. 2003
Summer bees newly emerged worker bees, 60 - 163 bees per treatment	5-OH metabolite (99.4%) 11 days feeding Concentrations: 7.5 - 240 µg/kg in sucrose Assay for olfactory learning performance	Significant decrease in proboscis reflex response. NOAEC: 6 µg/kg sucrose. LOAEC: 12 µg/kg sucrose.	Decourtye et al. 2003
14-42 days old 10 bees per dose, 3 replicates per dose	Olefin metabolite	LD_{50}: <36 ng a.i./bee NOAEC: 2.4 ng a.i./bee Working Note: See data above for TGAI. See data in Table A3-6 for toxicity data on metabolites in whitefly (Hemiptera).	Nauen et al 2001
14-42 days old 10 bees per dose, 3 replicates per dose	5-Hydroxy metabolite	LD_{50}: >49 ng a.i./bee NOAEC: 49 ng a.i./bee	Nauen et al 2001
14-42 days old 10 bees per dose, 3 replicates per dose	Urea metabolite	LD_{50}: >99,500 ng a.i./bee NOAEC: 1200 ng a.i./bee	Nauen et al 2001
14-42 days old 10 bees per dose, 3 replicates per dose	6-chloronicotinic acid	LD_{50}: >121,500 ng a.i./bee NOAEC: 121,500 ng a.i./bee	Nauen et al 2001

<table>
<tr><th>Honey bee</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>3 cages of 20 bees each per experiment, each experiment replicated 3 times</td><td>4,5-dihydroxy, desnitro, 6-chloronicotinic acid, and urea metabolites</td><td>All LD_{50} values > 1,000 ng/bee</td><td>Suchail et al. 2001</td></tr>
<tr><td>3 cages of 20 bees each per experiment, each experiment replicated 3 times</td><td>5-Hydroxy metabolite</td><td><table><tr><th>Hours</th><th>LD_{50} (ng/bee)</th></tr><tr><td>48</td><td>258</td></tr><tr><td>72</td><td>206</td></tr><tr><td>96</td><td>222</td></tr></table></td><td>Suchail et al. 2001</td></tr>
<tr><td>3 cages of 20 bees each per experiment, each experiment replicated 3 times</td><td>Olefin metabolite</td><td><table><tr><th>Hours</th><th>LD_{50} (ng/bee)</th></tr><tr><td>48</td><td>28</td></tr><tr><td>72</td><td>29</td></tr><tr><td>96</td><td>23</td></tr></table></td><td>Suchail et al. 2001</td></tr>
<tr><td>Mesocosm Studies</td><td>(includes hive studies)</td><td></td><td></td></tr>
<tr><td>Italy
Outdoor feeding station.
Return behavior
30 foraging bees per dose</td><td>Confidor formulation (NOS)
Sugar solutions: 100, 500, and 1000 ppb placed in feeding stations after training.</td><td>Immediately following exposure, treated bees left cages significantly more slowly than controls.
At 100 ppb, fewer bees returned to feeding station.
At 24 hours after exposure, none of the bees at the 2 higher concentrations were found at either the hive or the feeder.</td><td>Bortolotti et al 2003</td></tr>
<tr><td>France (?)
Bee colonies, ≈10,000 bees per colony
No control group comparisons.</td><td>TGAI (98%)
Oral, syrup at 48 µg/kg
Observation periods: 4 days for each of the pre-exposure, exposure, and post-exposure observations.</td><td>No impact on mortality assayed as dead bees/day before, during, and after treatment (Table 1 of paper).
Decreasing in syrup consumption and foraging activity only during exposure period (Table 2 and Fig. 1 of paper).
Possible impairment of olfactory learning during exposure but not statistically significant.
Working Note: Paper gives syrup consumption during exposure for hive but not for individuals. Cannot calculated µg/bee.</td><td>Ramirez-Romero et al. 2005</td></tr>
</table>

Honey bee	Exposure	Response	Reference
France Experimental colonies of ≈2300 bees.	Imidacloprid (NOS) Sucrose solution: 6 µg/kg sucrose. 4 day exposure.	Decrease in proportion of active bees (I/A ratio).	Colin et al. 2004
colonies in outdoor flight cages	TGAI (98%) Up to 10 days Control or 24 µg/kg sucrose	No effect on mortality. Decrease in foraging activity (measured by mean sucrose consumption) when rates were compared before treatment (186+39.3 ml, n=6), during treatment (57.9+9.7 ml, n=5) , and after treatment 38.2+5.3 ml, n=5).	Decourtye et al. 2004
2 replicates each application rate per test, approximatel y 50 bees per replicate	240 FS Alfalfa Foliar Applications: 0.045, 0.167 and 0.5 lb a.i./acre Mortalities assessed 2, 8 and 24 hours after caging bees with treated foliage after different periods of time.	<25% mortality at 0.045 lb a.i./acre after <2 hour s. <25% mortality at 0.167 lb a.i./acre after <8 hour s. <25% mortality at 0.5 lb a.i./acre after 8 hour s.	Hancock et al. 1992 MRID 42632901
Apis mellifera carnica, outdoor colonies	Imidacloprid (powder form, Bayer, NOS) Sucrose solutions: 0, 0.14, 1.5, 3, and 6 ng/bee (assuming 10µL consumption). Observation up to 3 days.	Radiofrequency tracking of individual bees. Alterations in foraging behavior at doses of 1.5 ng/bee and higher (equivalent to nectar concentrations of 115 ppb). NOAEC: 0.14 ng/bee (equivalent to 11.5 ppb in nectar).	Schneider et al. 2012
Field Studies			
Greek honey bees, sudden colony deaths. Retrospective study	Source of imidacloprid not identified. Analysis of colony samples for pathogens and pesticides.	Multiple pathogens including Some colonies contaminated with imidacloprid at 27 (14 to 39) ng/g tissue (in 60% of samples) with co-exposure to *Nosema ceranae* (Table 1).	Bacandritsos et al. 2010

Honey bee	Exposure	Response	Reference
Controlled study. Imidacloprid and uncontaminated bee hives in Belgium at different sites.	Confldor 200SL Colony dosing: Imidacloprid in sugar solution at 0.00355 µg a.i./L. Dosing in July, observations to December. Weekly or biweekly monitoring of foraging and mortality. Weekly weighing of hives.	No indication of bee mortality. Reduction in bee activity relative to control hives. See Fig. 2. The reduction appears significant at weeks 3 and 6 but not at week 10. Decrease in capped broods appears significant on week 12 (Fig. 3). No statistically significant change in colony weights (Fig. 4). No statistically or biologically significant change in foraging (Fig. 5). Working Note: Does not involve observations of overwintering.	Belien et al. 2009
Quebec Cage exposures in maize (conventionally grown) and non-cultivated fields 3 replicates per dose and controls	Admire 240F Doses: 0, 0.08, 0.16, 0.24 and 0.30 ng/bee Observations at Day 10. Based on Fig. 4a, average weight for 10 bees is about 0.9 g or about 90 mg/bee.	Increases in AChE (head) activity at lower three doses. 0.3 ng/bee: Substantial mortality (>97.5%) precluded AChE analyses. Decrease bee weights at all but the lowest dose. 10-day LD_{50}: 0.227 ±0.02 ng/bee Working Note: This is by far the lowest LD_{50}. Using a BW of about 100 mg (Fig. 3d), the LD_{50} is about 0.00227 mg/kg bw. Hyperactivity (tumbling and trembling) by day 1 at highest dose and day 3 for lower doses. Hyperactivity appears transient, peaking on Days 5-6 and close to controls by Day 10. LOAEL: 0.08 ng/bee. Significant increase in AChE activity (Fig. 4a of paper).	Boily et al. 2013
France, 5 locations Retrospective assays of pesticide exposure in colonies	Several pesticides (Table 1 of paper) including imidacloprid.	No association of hive mortality with pesticide exposure. Imidacloprid (with 6-chloronicotinic acid) was pesticide most commonly detected – i.e., 57.3% pollen loads and 29.7% in honey.	Chauzat et al. 2009
France, 5 locations Retrospective assays of pesticide exposure in colonies	Several pesticides (Table 1 of paper) including imidacloprid.	Imidacloprid was pesticide most commonly detected – i.e., 40.5% pollen loads and 21.8% in honey. No assessment of impacts on colony health.	Chauzat et al. 2011
Beehives in field	TGAI (NOS) 15 days Syrup concentrations: 2 µg/L	Increase expression of genes associated with P450 and lipid metabolism. Down regulation of genes involved in carbohydrate metabolism.	Derecka et al. 2013

<table>
<tr><th>Honey bee</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr>
<td>Maryland 2009 experiment
10 replicate colonies per treatment group.</td>
<td>Admire Pro (42.8% a.i.)
Doses: 0, 5, 20, and 100 µg/kg diet paddies (honey and Megabee powder [http://www.megabeediet.com/]).
Treatment from May to August (12 weeks or 84 days).
Observations to March of following year.
Consumption:
2009: Cumulative doses of 16.6, 63.7 and 322.6 µg for each exposure group.
Average colony populations over all dates: 17,440, 18,541, 17,813, and 18,850.

Working Note: Estimate of cumulative dose roughly proportional to concentrations.</td>
<td>Two colonies (one in the 20 µg/kg and the other in the 100 µg/kg group) did not survive to winter.
All colonies infested with Varroa mites. Mite counts significantly higher in 100 µg/kg group and a dose-related trend in infestations.
11 of 27 colonies surviving winter infested with Nosema but not dose-related.
Positive association of dose and colony size marginally significant ($_p$=0.054).
In treated colonies, foraging activity in August and September 12% lower than controls but not dose-related.
No significant impact on cells drawn, capped honey, bee bread, and capped brood cells. At the two higher doses, honey stores consistently higher.
Colony overwintering survival:
<table>
<tr><th>Dose</th><th>Mortality</th><th>Number</th></tr>
<tr><td>0</td><td>0</td><td>10</td></tr>
<tr><td>5</td><td>2</td><td>10</td></tr>
<tr><td>20</td><td>3</td><td>10</td></tr>
<tr><td>100</td><td>6</td><td>10</td></tr>
</table>
No food shortages or symptoms of colony collapse disorder associated with overwintering mortality.

Working Note: 5 ppb may be considered a NOAEC. Using the Fisher Exact Test, 0/10 vs 2/10 has a p-value of 0.236842.
Concentrations in bees of 0.3 to 2.8 µg/kg bee.

Based on the concentrations of imidacloprid in the diet, the 84 days of exposure and the average populations of bees in the colonies, the following daily doses can be estimated.
<table>
<tr><th>Cum Dose to Colonies (µg)</th><th>Number of Bees</th><th>ng/bee/ day</th><th>Dose mg/kg bw/day</th></tr>
<tr><td>16.6</td><td>18,541</td><td>0.01066</td><td>0.000092</td></tr>
<tr><td>63.7</td><td>17,813</td><td>0.04257</td><td>0.000367</td></tr>
<tr><td>322.6</td><td>18,850</td><td>0.20374</td><td>0.01756</td></tr>
</table>
Paper does not report the body weight of the bees. In the above table, mg/kg bw doses are calculated based on the body weight of 116 mg from Winston (1987). Note that ng ÷ mg = mg/kg.
Commentary on cross-contamination (p. 16): … traces of imidacloprid were detected in a few samples from control colonies in 2009, and this cross-contamination was apparently due to drifting and possibly some robbing because hives were placed close to each other in apiaries.</td>
<td>Dively et al. 2015
2009 experiment

Working Note: The cumulative doses in Dively et al. 2015 are given in the paper in units of milligrams. In a personal communication, Dively (2015) has confirmed that this is a typo-graphical error and that the correct units are micrograms.</td>
</tr>
</table>

<table>
<tr><th>Honey bee</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr>
<td>Maryland 2010 Experiment
7 replicate colonies per treatment group.</td>
<td>Admire Pro (42.8% a.i.)
Doses: 0, 5, 20, and 100 µg/kg diet paddies (honey and Megabee powder [http://www.megabeediet.com/]).
Treatment from May to August (12 weeks).
Observations to March of following year.
Consumption similar to 2009 experiment.
Average colony populations over all dates: 13,822, 14,200, 13,813, and 14,140.
Distance between hives: “isolated apiaries” (NOS). 10 meters distance between hives at the same apiary.</td>
<td>Residues of 0.3–2.2 µg/kg in bees were detected after four weeks of exposure in two colonies exposed to 100 µg/kg (p. 10 of paper).
All colonies infested with Varroa mites but no significant differences among groups. Nonetheless, a significant dose-related trend in infestations.
No significant differences in colony parameters after dosing period.
Significant decrease in brood production between controls and all treatment groups combined. No dose-effect relationship.
No impact on pollen foraging but amount of pollen collected was 41% less than 2009 experiment.
One colony in each dose group did not survive to October due to … a lack of brood and virtually no stored food, despite the fact that each hive was provisioned with sucrose syrup since mid-August.
Colony overwintering survival:
<table>
<tr><th>Dose</th><th>Mortality</th><th>Number</th></tr>
<tr><td>0</td><td>3</td><td>7</td></tr>
<tr><td>5</td><td>4</td><td>7</td></tr>
<tr><td>20</td><td>4</td><td>7</td></tr>
<tr><td>100</td><td>4</td><td>7</td></tr>
</table>
Colony mortality not significantly different among groups.
Working Note: Authors report a p-value of 0.21 for one-tailed Fishers Exact test. I get 0.412 for 3/7 vs 12/21 which checks with http://research.microsoft.com/en-us/um/redmond/projects/mscompbio/fisherexacttest/ as well as http://quantpsy.org/fisher/fisher.htm.
Residues in bees at 100 µg/kg group after 6 weeks of exposure: 0.5-1.9 µg/kg.
In 100 µg/kg group, 14–26% fewer frames of bees relative to controls and 20 µg/kg groups.
Commentary on colony survival: We contribute this higher mortality to subnormal colonies going into the winter and abnormally higher temperatures during the winter which resulted in over-consumption of the stored food.
Colonies low on food were given bee candy during winter.
Working Note: Author’s indicated that 2009/2010 pooled responses – i.e., 3/17 (control), 6/17 (5 ppb), 7/17 (20 ppb), and 10/17 (100 ppb) are not statistically significant in terms of dose-dependence. Based on the Cochran-Armitage Test using U.S. EPA (2012) Benchmark Dose Software, the dose-response relationship is significant at p=0.0136.</td>
<td>Dively et al. 2015
2010 experiment</td>
</tr>
</table>

Honey bee	Exposure	Response	Reference
France *Aphis mellifera mellifera* 4 groups of 8 to 9 hives. Extra untreated colony added to each group of environmental sentinel. Also unfed (Group Gno) colonies and fed without imidacloprid (Group G0).	Imidacloprid (purity not specified) Concentrations in sucrose: 0.5 Group G0.5) and 5 (Group G5) µg/L. Distance between hives: 30 meters. Fed during summer for about 32 days (July 12 to Aug. 14) and observed until end of following winter.	Several colonies from all groups swarmed. No significant differences among groups. No significant differences in activity. No significant difference in pollen carrying but high dose imidacloprid group had more pollen carrying days than low dose group. Significant increase in mortality in high dose group on July 20. Overall, no differences in mortality. In December, all colonies treated for *Nosema* infestation. Various decreases in capped brood area but not dose-related. No significant differences in over-wintering.	Faucon et al. 2005
Belgium Jonagold (apple) orchards with pollination hives.	Confidor 200 SL 0.046 g a.i./ha (≈0.041 lb a.i./acre) 2 Applications: during green bud stage and blossom start.	No differences in number of bee visits (i.e., no repellency).	Gobin et al. 2008
Massachusetts 4 apiaries 4 treated hives and a control hive per apiary. Note: A total of 4 hives per dose (including controls).	Imidacloprid (NOS) in sucrose. 13 week exposures to variable concentrations Concentrations: 0.1, 1, 5, and 10 µg/kg (ppb) for 4 weeks. 20, 40, 200, and 400 µg/kg (ppb) for 9 weeks. Distance between hives: 12 kilometers. Exposure initiated in July and terminated in September. Observations until March of the year following exposure.	A decrease in sealed brood cells at all concentrations and controls. All hives survived to December 22 (week 12 post-exposure). Substantial mortality starting in week 14 post-exposure. Apparent dose-response relationship. By week 21 post-exposure, 3 of 4 control hives survived but only 1 of 4 hives in the high dose group survived. By week 23, it appear that 3 of 4 controls survived all 4 colonies in the 20, 200, and 400 µg/kg dose groups were dead and only 1 in 4 of the 40 µg/kg dose group survived. See Figure 2. Working Note: Pooling all treated hives, the mortality of 15/16 in treated hives relative to 1/4 of the control hives is statistically significant (p=0.012416)using the Fisher Exact test. Without pooling, the response of 4/4 in treated hives and 1/4 in control hives is only marginally significant (p=0.071429) using the Fisher exact test. Hives separated by 30 km and all new materials used for each hive. No apparent potential for cross-contamination.	Lu et al. 2012

Honey bee	Exposure	Response	Reference
U.S. 3 apiaries 2 groups of 3 colonies per apiary: One group fed sucrose and the other high-fructose corn syrup (HFCS). Control, imidacloprid and clothianidin. Note: groups with clothianidin not considered further.	Imidacloprid (NOS) July 12: 258 µg imidacloprid in in 1.9 liter – i.e., 135 µg/L per week for 13 weeks. Colonies monitored weekly. Distance between hives: 30 km (identical to Lu et al. 2012). Experiment terminated in April of the year following dosing. Authors assume 50,000 per colony,	No differences in control and treated colonies until winter. Up to that point, no mortality or signs of toxicity. No differences in sucrose and HFSC groups. Data pooled. Treated hives continued to decline into January. Loss of 4/6 treated colonies vs 1/6 controls. Control loss apparently due to *Nosema ceranae* pathogen. Estimated average dose reported as 0.74 ng/bee/day. Working Note: This dose estimate cannot be reproduced. Given the dose of 258 µg, the 91 day period of exposure, and the estimate of 50,000 bees, the dose would be about 0.056 ng/bee/day [258,000 ng ÷ (50,000 bees x 91 days) ≈ 0.056703/bee/day].	Lu et al. 2014
Belgium 16 apiaries in the vicinity of maize fields	Imidacloprid (NOS) in agricultural applications. Average rates not specified. Analyses of honey, beeswax, and bees for imidacloprid..	No correlation of colony health with imidacloprid treatment of maize. Apparent negative but significant ($p<0.02$) correlation of mortality and area of maize treated with imidacloprid within 3000 m of field. See Fig. 3.	Nguyen et al. 2009
Uruguay Analysis of pesticide residues in active (29 apiaries, 10,000 hives.) and depopulated hives (8 apiaries, 4,800 hives).	Imidacloprid (NOS) in agricultural applications.	Imidacloprid in depopulated hive honeycombs: 377±118 µg/kg. Detected in 3 honeycombs of depopulated hives. No report of imidacloprid in active hives. Working Note: No data on populated hives.	Pareja et al. 2011

Honey bee	Exposure	Response	Reference
Sunflower plots (8 m x 30 m) with or without seeds treated with imidacloprid. Two locations, one with 3 treated fields and the other with 4 treated fields. Control fields at each locations.	TGAI 39-day exposure and observation period. Colony development using sunflower honey dosed with imidacloprid. Concentrations of 0.002, 0.005, 0.010, and 0.020 mg/kg.	No adverse effects on mortality, feeding activity, wax/comb production, breeding or colony vitality were detected at any concentration. NOAEC: 0.020 mg a.i./kg (20 ppb), highest concentration tested. Working Note: Based on calculations in this paper (p. 235, column 1), 46 ppb (0.046 mg/kg) corresponds to an acute NOAEC of 1.2 ng/bee. Based on this relationship, the NOAEC of 20 ppb would correspond to a dose of 0.52 ng/bee.	Schmuck et al. 2001
China 18-20 bees per group (Fig. 2 of paper)	Imidacloprid (>95%) Sucrose concentrations: 40, 50, 100, 200, 400, 600, 800, 1,200, 1,600, 3,000, 4,000, and 6,000 µg/L	Increase in time to return to feeding stations. Significant increase at concentrations of 50 µg/L and greater (Fig. 3). Based on consumption, altered behavior apparent at doses of 1.82 to 4.33 ng/bee (p. 1745, lower column 1). The LOAEL is 100 µg/L. Some bees failed to return to feeding stations. Dose-related. Working note: Given that 100 µg/L corresponds to doses of about 1.82 ng/bee, the NOAEC of 50 µg/L would correspond to an NOAEC of about 0.9 ng/bee.	Yang et al. 2008
Bee larvae	Imidacloprid (>95%) Sucrose solutions yielding doses of 0.0004 to 8000 ng/larva.	Significant decrease in capped brood rate, pupation rate, pupal emergence at 24 ng/larva and greater. Adult bees treated as larvae at a dose of 0.04 ng/larva evidenced changes in olfactory associative behavior.	Yang et al. 2012

A3 Table 2: Bumblebees (*Bombus* sp.)

Bumblebee	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Bombus impatiens Contact assay 4 replicates of 9-11 bees per replicate per dose.	Imidacloprid (>95%) Oral Doses: 0.1, 0.1 and 1 g/L solutions. 5 mL of solutions applied to filter paper.	About 75% mortality at low exposure and 90-100% at higher exposures (Fig. 1). Working Note: No way to meaningfully estimate doses.	Gradish et al. 2010
Bombus impatiens Direct spray.	Intercept 60 WP (600 g/kg) Topical Direct spray at 0.267 kg/ha (≈0.24 lb/acre)	100 % mortality after 72 hours.	Gradish et al. 2010
Bombus impatiens Working Note: See Figure 1. Bombus appears to be equally sensitive to honey bee. Alfalfa leafcutting bees (*Megachile rotundata,* Megachilidae) and *Osmia lignaria* are more sensitive. Again, cannot get meaningful dose in terms of ng/bee but comparison is useful.	Imidacloprid (NOS) Topical Direct spray 4-6 concentrations/ assay. NOTE: Above are expressed as % solution x 10^{-3}.or 1 in 100,000. Thus, the LC50 values multiplied by 10 give units of ppm.	48-hour LC_{50}s: Species: *Bombus impatiens* (n=299, ♀) — LC_{50} (%x10^{-3}): 3.22 (2.54-4.10) *Megachile rotundata* (n=299, ♂♀) — 0.17 (0.14-0.21) *Osmia lignaria* (n=400, ♂♀) — 0.07 (0.06-0.09) *Apis mellifera* (n=259) — 2.2 (1.8-3.4) Working Note: 1% x 10^{-3} corresponds to 10^{-5} or 1 in 100,000. Thus, the LC50 for *A. mellifera* corresponds to 22 ppm	Scott-Dupree et al. 2009 Working Note: The LC_{50} for *Apis mellifera* is from Bailey et al. (2005), from the same facility involved in this paper by Scott-Dupree et al. Note also that Bailey 2005 indicates that mortality was assessed at 24 hours.
Formulation			
Bombus terrestris, Two different colonies 5 bees per cage,	Imidacloprid (commercial product, NOS) Oral Dosing in containers with 1 bee per container, 10 µL/dose.	24-h LD_{50}: 0.04 µg/bee 72-h LD_{50}: 0.02 µg/bee	Marletto et al. 2003
Bombus terrestris, Two different colonies 5 bees per cage,	Imidacloprid (commercial product, NOS) Topical 10 µL droplet/bee	24-h LD_{50}: not calculated 72-h LD_{50}: 0.02 µg/bee Working Note: Above is not a typo. 72-h LD_{50} values are identical for oral and topical.	Marletto et al. 2003

<table>
<tr><th>Bumblebee</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Sublethal Effects</td><td></td><td></td><td></td></tr>
<tr><td>Bombus terrestris
Groups of 10 bees (caged), large number of cages but distribution unclear.</td><td>TGAI (NOS)
Sucrose concentrations: 0.08, 0.20, 0.51, 1.28, 3.20, 8.00, 20.0, 50, and 125 µg/L.
With or without acetonitrile solvent.
4 day exposure</td><td>Decreases in feeding rate and locomotion (with acetonitrile only) but no impact on longevity (See triangles in Figure 1 of paper).
Working Note: Contrast with lack of effects in honeybees.</td><td>Cresswell et al. 2014</td></tr>
<tr><td>Bombus terrestris
Both older workers and newly emerged.
In vitro culture of Kenyon cells (mushroom body neurons).</td><td>Imidacloprid (Sigma-Aldrich)
Sucrose solutions: 0, 2.5, 25, and 2500 ppb.</td><td>No effect on Kenyon cell cultures in bees exposed to 2.5 ppb. Growth impaired at 25 ppb and no growth at 2,500 ppb. More substantial impact in older workers.</td><td>Wilson et al. 2013</td></tr>
<tr><td>Mesocosm Studies</td><td></td><td></td><td></td></tr>
<tr><td>Bombus impatiens
Queenright colonies with 30-50 workers
2 weeks acclimation prior to dosing.
8 colonies per treatment.</td><td>Imidacloprid (NOS)
Sucrose solutions: 0, 10, 20, 50, and 100 ppb
11 weeks</td><td>Queens: Substantial queen mortality by week 11 at 20 ppb and higher (Figure 1 of paper).
<table><tr><th>Dose</th><th>Mortality by Week 11</th></tr><tr><td>0</td><td>1/8</td></tr><tr><td>10</td><td>2/8</td></tr><tr><td>20</td><td>5/8</td></tr><tr><td>50</td><td>7/8</td></tr><tr><td>100</td><td>8/8</td></tr></table>Workers: Slower movement relative to controls.
Colony Health: Decreases in colony weights relative to controls at all doses. Substantial and dose-related (Figure 4).</td><td>Scholer and Krischik 2014</td></tr>
<tr><td>Bombus terrestris audax
Groups of 1 queen and four workers.
6 per dose.</td><td>Imidacloprid (NOS)
Sucrose: 0.08 to 125 µg/L (9 doses)
14 day exposure followed by 14 day recovery period.</td><td>No brood production at highest dose during exposure period. Dose-related trend in fewer broods with increasing dose (Fig. 1).
Brood production inhibition:
EC_{50}: 1.44 ppb
EC_{10}: 0.15 ppb
Also a dose-dependent recovery in brood production (greater recovery at higher doses (Fig. 3).</td><td>Laycock and Cresswell 2013</td></tr>
</table>

<table>
<tr><th>Bumblebee</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Bombus terrestris
Queenless microcolonies.
128 workers
3-15 replicates per dose (p. 3). Total of 76 groups.</td><td>Imidacloprid (NOS)
Sucrose solutions: 0.08 to 125 µg/L
14 days exposure/ observation</td><td>Dose-dependent decrease in broods (Fig. 1) attributed to feeding suppression. NOAEC of about 0.1 µg/L. LOAEC at about 1 µg/L.
Imidacloprid intake can be read graphically from Fig. 2a (right axis, triangle symbols). Doses appear to be about 0.1 ng/bee/day to about 8.5 ng/bee/day.
NOAEC not given but is clearly <0.1 ng/bee and LOAEC would be about 0.7 µg/L.</td><td>Laycock et al. 2012</td></tr>
<tr><td>Bombus terrestris
Queenless microcolonies.
5 workers per next
4 nests per treatment

Without foraging (entry with foraging is given below).</td><td>Confidor 20% SC (Bayer)
Concentrations in sucrose: 0.01, 0.02, 0.2, 2, and 20 ppm
11 weeks</td><td>Time to 100% mortality:<table><tr><th>Concentration</th><th>Time</th></tr><tr><td>0.2 ppm</td><td>49 days</td></tr><tr><td>2 ppm</td><td>28 days</td></tr><tr><td>20 ppm</td><td>14 days</td></tr><tr><td>200 ppm</td><td>A few hours</td></tr></table>Chronic LC_{50}: 59 (52-68) ppb.
Chronic Reproduction EC_{50}: 37 (26-51) ppb.
Decrease drone production at 20ppb.
Reproductive NOAEC: 20 ppb.
Working Note: Be sensitive to different units used above (and in paper).</td><td>Mommaerts et al. 2010</td></tr>
<tr><td>Bombus terrestris
Queenless microcolonies.
5 workers per next
4 nests per treatment

With foraging assay (entry without foraging is given above).</td><td>Confidor 20% SC (Bayer)
Concentrations in sucrose: 0.01, 0.02, 0.2, 2, and 20 ppm
11 weeks</td><td>Time to 100% mortality:<table><tr><th>Concentration</th><th>Time</th></tr><tr><td>0.2 ppm</td><td>49 days</td></tr><tr><td>2 ppm</td><td>14 days</td></tr><tr><td>20 ppm</td><td>7 days</td></tr><tr><td>200 ppm</td><td>A few hours</td></tr></table>The above times to 100% mortality are somewhat shorter than experiment without foraging.
Chronic LC_{50}: 20 (19-21) ppb.
Less foraging in survivors.
Chronic Foraging EC_{50}: 3.7 (2.5-5.5) ppb.
NOAEC for foraging behavior not determined.</td><td>Mommaerts et al. 2010</td></tr>
<tr><td>Bombus terrestris
Queenright hives: 1 queen, 25 workers, and brood.</td><td>Confidor 20% SC (Bayer)
Concentrations in sucrose: 0, 2, 10, and 20 ppb
2 weeks</td><td>Substantial lethality at 10 and 20 ppb.
2ppb: No marked mortality or other sublethal effects including reproduction and sugar consumption.</td><td>Mommaerts et al. 2010</td></tr>
<tr><td>Bombus terrestris
Queen, 25 workers, and brood.
4 day acclimation.</td><td>Confidor (NOS)
Sucrose solution: 20 ppb
2 days</td><td>No significant change in Btfor (foraging gene) gene expression.</td><td>Tobback et al. 2011</td></tr>
</table>

Bumblebee	Exposure	Response	Reference
Bombus terrestris 25 colonies per treatment	Imidacloprid (NOS) Low dose: 6 µg/kg pollen and 0.7 µg/kg sugar solution. High dose: 12 µg/kg pollen and 1.4 µg/kg sugar solution. 2 week exposures 8 week observations	No substantial impact on colony weights during treatment. In 6 week post-treatment, substantial and dose-related decrease in colony weights (Fig 1). Significant decrease in number of new queens at both doses. As dose-response relationship it apparent (Fig. 2). Authors suggest an impact on foraging in post-exposure period.	Whitehorn et al. 2012 See field study by Feltham et al. 2014, below. Same group of investigators but different studies.
Field Studies			
Bombus impatiens One caged colony per plot, 10 paired plots (one treated, one control)	Merit 0.5 Granular Kentucky Application rate: 0.4483 kg a.i./ha (≈0.4 lb a.i./acre) with irrigation. 30 day observation period.	No effects on colony vitality measured in terms of weight, number and weight of workers, number of brood chambers and honey pots, and measures of defensive response.	Gels et al. 2002[1]
Bombus impatiens One caged colony per plot, 5 replicates of three plots. Two plots sprayed and one control plot.	Merit 75 Kentucky Application rate: 0.336 kg a.i./ha (≈0.3 lb a.i./acre) h with 1.5 cm of irrigation. 28 day observation period.	No effects on colony vitality or workers defensive response on irrigated plots. Bees on non-irrigated plots were adversely affected with respect to bees on untreated control plots: fewer honey pots and brood chambers, fewer workers, reduced biomass of workers and lower colony weight. Queen weight was not affected. Reduced defensive response to an aggressive stimulus was also observed. Foraging activity was reduced significantly on non-irrigated plots, but not on irrigated plots, with respect to controls.	Gels et al. 2002[1]
Bombus terrestris Small colonies (queen and up to 65 workers) 3 control and 3 treated nests.	Imidacloprid (NOS) Exposed colonies given sugar solution with 0.7 ppb and pollen with 6 ppb for 2 weeks prior to foraging assays.	Significant decrease in pollen gathering but no significant impact on nectar gathering (Fig. 1 of paper).	Feltham et al. 2014
Bombus terrestris 10 colonies per group	Imidacloprid (NOS) Sucrose solution: 10 µg/L 4 week exposure	27% reduction in workers starting after 2 weeks. Delay suggestive of impact on brood development. Increase (≈50%) in number of lost workers. Impaired pollen foraging efficiency.	Gill et al. 2012

A3 Table 3: Bees, Other Species

Bees, Other	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Nannotrigona perilampoides (stingless bee, Apidae) Forager, 3-5 days old, average 8.2 mg bw (p. 1739) 10 per dose, typically 2 replicates	Imidacloprid (NOS) Topical application. 24-hours	LD_{50}: 0.0011 (0.0008-0.0012) µg/bee Slope: 1.14	Valdovinos-Nunez et al. 2009
Scaptotrigona postica (stingless bee, Apidae) 3 replicates of 20 bees per replicate for each of 6 doses	Imidacloprid (NOS) Topical application. 24 and hour observations.	24-h LD_{50}: 0.0252 µg a.i./bee 48-h LD_{50}: 0.0245 µg a.i./bee Working Note: Body weights not specified in paper.	Soares et al. 2015 Brazil
Scaptotrigona postica (stingless bee, Apidae) 3 replicates of 20 bees per replicate for each of 6 doses	Imidacloprid (NOS) Dietary exposure at 1 to 120 ng a.i./µL of diet. 24 and hour observations.	24-h LC_{50}: 42.5 ng a.i./µL 48-h LD_{50}: 14.3 ng a.i./µL Working Note: Amount consumed not specified in paper. Note that ng/µL corresponds to µg/mL or mg/L. The above are very high concentrations.	Soares et al. 2015 Brazil
Formulation			
Apis ceranae 3 replicates of 10 bees per replicate	Tatamida 17.8 SL Direct spray of 50 ppm solution. Not clear if this is a.i. or formulation. See Table 1 of paper	% mortality Hours / *A. cerana* / Honey bee 24 / 60.0 % / 50% 48 / 66.67% / 66.67% Working Note: While dosing is unclear, the sensitivities are about the same.	Stanley et al. 2015

Bees, Other	Exposure	Response	Reference
Melipona quadrifasciata (stingless bee native to Brazil) Adult workers (size not specified) 3 replicates of 10 bees per dose	Brazilian formulation, 700 g a.i./L, water dispersible granules, Bayer CropScience, Brazil) Diluted in a 50% sucrose solution. Doses: 5, 10, 30, 50, 70, and 90 ng a.i./bee	LD_{50}: 23.54 ng a.i./bee LD_{55}: 5.38 ng a.i./bee In subsequent bioassays at LD_{05}, an initial decrease in activity at 3 hours followed by an increase in activity at 24 hours (Fig. 2 of paper). Also impaired flight. Significant decrease in respiration at both 3 and 24 hours (Figure 4 of paper). Working Note: Based on Contrera et al. (2006), the approximate body weight of an adult of this species is 8 mg (Figure 1 of paper). Thus, the LD_{50} is about 2.9 ng/mg or µg/g bw.	Tom et al. 2015
Osmia cornifrons (Japanese orchard bee) Adult bees, newly emerged (24 hours for males and 24-72 hours for females). A total of 60–135 per assay, at least 5 doses	Provado 1.6F (17.4% a.i.) Topical application, 1 µL per bee 48 hours	LD_{50}: 3.8 (1.7-12.6) µg/bee Working Note: In a paired assay (Table A3-1 above), *Apis mellifera* is substantially more sensitive.	Biddinger et al. 2013
Sublethal Effects			
Apis ceranae	Imidacloprid (NOS) Sucrose concentrations: 10, 20, and 40 µg/L. Upper bound estimates of doses: 0.27, 0.39, and 0.52 ng/bee. Short-term exposures: hours.	Dose-related decreases in feeding – i.e., return to feeder and volume imbibed (Fig. 1). NOAEC (volume consumed): 10 µg/L (0.27 ng/bee); LOAEC 20 µg/L (0.39 ng/bee). No aversion to imidacloprid contaminated feeders. At high dose, no significant aversion to feeders with predator species (wasp, *Velutina velutina*).	Tan et al. 2014

Bees, Other	Exposure	Response	Reference
Apis cerana ceranae Groups of 10 bees.	Imidacloprid (NOS) Topical application to thorax at a concentration of 10 mg/L.	Enhanced expression of AccGtpx-1 gene encoding phospholipid hydroperoxide glutathione peroxidase. `Working Note: This is an extraordinarily high dose.`	Wang et al. 2010
Melipona quadrifasciata anthidioides (stingless bee native to Brazil) Larval exposures	Imidacloprid formulation (700 g a.i./L, Bayer CropScience, Brazil) Oral Doses: 0.0056 to 56 µg a.i./bee (18 doses plus control).	No effect on larval survival or growth. Mushroom bodies (nerve cells) of 1 day old adults not impacted. Later development (4 and 8 days) seriously impaired – i.e., decrease in volume with increasing dose (Figure 4). NOAEC/LOAEC not clear from figure. No substantial impact of walking behavior of newly emerged adults but severe effects on older adults. Substantial and significant decreases in survival at all doses (Figure 1). Significant negative correlation between dose and survival time (Fig. 3). The dose of 0.056 ng/bee appears to be a NOAEC and the corresponding LOAEC appears to be 0.08 ng/bee.	Tome et al. 2012
Osmia lignaria Larvae/egg exposures "Field" and laboratory phases	Imidacloprid (97.5%) Concentrations: 0, 3, 30, and 300 ppb in pollen.	No lethality. In field study, longer development time at the two higher doses. Effect appears to be statistically significant (p=0.0263) only at highest dose. In laboratory phase, no significant impact on development. Significant effect among females only in time to darkening cocoons (i.e., completion of cocoon development).	Abbott et al. 2008
Mesocosm Studies			
No studies identified.			
Field Studies			
No studies identified.			

A3 Table 4: Hymenoptera, Other

Hymenoptera, Other	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
No studies identified.			
Formulation			
Aphytis melinus (parasite of the California Red Scale, Aphelinidae)	Admire 2F (240 g a.i./L) Leaf uptake bioassay 24 hour exposure At least 5 concentration/assay	LC_{50}: 0.246 (0.089-0.465) g a.i./L Working Note: Above unit designation is not a typo. This is a very peculiar bioassay. Included because the assay covered 6 species.	Prabhaker et al. 2011
Colpoclypeus florus (Eulophidae) 5 2-4-day old adult females [Ectoparasitoid of lepidopterans]	Provado 2F 48-hr contact Solution of 48 mg a.i./L	100% mortality when applied at 100% label application rate for apple trees	Brunner et al 2001
Colpoclypeus florus (Eulophidae) 5 2-3 day old females per leaf disc collected 1,3, 7, 14 and 21 days after treatment [Ectoparasitoid of lepidopterans]	Provado 2F Residues assay 3 apple trees sprayed at recommended application rate for Provado 2F 3 times in July or August. Insects evaluated 48-hours after exposure to leaf disk	No significant impact on mortality relative to controls at any of the sampling periods.	Brunner et al 2001
Diadegma insulare (wasp parasitoid; Ichneumonidae) 10 adults per treatment	Provado 2F At field solution of 0.22 mg a.i./mL) and 0.01, 0.05, 0.1 and 0.5 of field solution. Spray volume of 240 liter/ha.	24-hour LC_{50}: 0.002 (0.000 - 0.004) mg a.i./mL Working Note: Given a spray volume of 240 L/ha, the LC_{50} of 0.002 mg a.i./mL is ≈0.00048 kg a.i./ha or about 0.000428 lb/acre.	Hill and Fosler 2000
Encarsia formosa (parasitoid of whitefly, Aphelinidae)	Admire 2F (240 g a.i./L) Leaf uptake bioassay 48 hour exposure At least 5 concentration/assay	LC_{50}: 0.980 (0.267-1.53) g a.i./L Working Note: Above unit designation is not a typo. This is a very peculiar bioassay. Included because the assay covered 6 species.	Prabhaker et al. 2011
Eretmocerus eremicus (parasitic wasp of whitefly, Aphelinidae)	Admire 2F (240 g a.i./L) Leaf uptake bioassay 48 hour exposure At least 5 concentration/assay	LC_{50}: 1.93 (1.33-2.67) g a.i./L Working Note: Above unit designation is not a typo. This is a very peculiar bioassay. Included because the assay covered 6 species.	Prabhaker et al. 2011

Appendix 3: Terrestrial Invertebrates (*continued*)

Hymenoptera, Other	Exposure	Response	Reference
Gonatocerus ashmeadi (fairyfly, Mymaridae)	Admire 2F (240 g a.i./L) Leaf uptake bioassay 48 hour exposure At least 5 concentration/assay	LC_{50}: 2.63 (1.56-4.16) g a.i./L Working Note: Above unit designation is not a typo. This is a very peculiar bioassay. Included because the assay covered 6 species.	Prabhaker et al. 2011
Trichogramma nr. Brassicae (parasitoid; Trichogrammatidae) 20 -40 females per group of sprayed leaves.	Confidor 350 SC (300 g/l a.i.). Applied at field solution of 5.25 g a.i./100 L. Sprayed onto leaves.	100% mortality after 3 hours.	Hewa-Kapµge et al. 2003
Trichogramma nr. Brassicae (parasitoid; Trichogrammatidae) 15 females, tested in 3 groups of 5	Confidor 350 SC (300 g/l a.i.). Residual exposure to leaves 0, 1, 4, and 7 days after spraying. Applied at field solution of 5.25 g a.i./100 L. Sprayed onto leaves.	Significant increase in mortality (~60%) with respect to controls on day 0 only. 10-20% mortality on days 1, 4 and 7 in comparison with a 0-5% control mortality on these days.	Hewa-Kapµge et al. 2003
Trichogramma nr. Brassicae (parasitoid; Trichogrammatidae) 15 females, tested in 3 groups of 5	Confidor 350 SC (300 g/l a.i.). Residual exposure to leaves 0, 1, 4, and 7 days after spraying. Evaluation of ability to infect eggs for 24 hours. Applied at field solution of 5.25 g a.i./100 L.	The number of eggs successfully parasitized did not differ significantly from untreated controls on days 0, 1, 3 and 7 following exposure.	Hewa-Kapµge et al. 2003
Trichogramma nr. Brassicae (parasitoid; Trichogrammatidae) 5 replicates, 60 parasitized eggs each	Confidor 350 SC (300 g/l a.i.). Exposure of life stages still inside host (egg or late pupal stages): parasitized *Helicoverpa armigera* eggs dipped in solutions for 1-2 seconds.	No difference between untreated controls and imidacloprid exposed host eggs for either egg or pupal life stages of wasp.	Hewa-Kapµge et al. 2003
Trichogramma platneri (endoparasitoid; : Chalcidoidea) 5 1-2 day old females per dose	Provado 2F Contact assay 48-hours 48 ppm	100% mortality	Brunner et al 2001

Hymenoptera, Other	Exposure	Response	Reference
Trichogramma cacoeciae (egg parasitoid wasp: Trichogrammatidae) Adults	Confidor 200 SL (Bayer, Germany) Direct spray (1.28 µL/cm^2) 24-h observation	24-h LC_{50}: 1.25 (0.88-1.54) µg a.i./mL	Saber 2011
Sublethal Effects			
Psyttalia concolor (parasitoid, Braconidae)	Confidor (20% a.i., Spain)	Residual Contact Assay: 40 mg/L spray. About 28% to 75% mortality from 24 to 72 hours (Table 1). Feeding: 10 mg a.i./L. Decrease in number of emerging offspring. Other assays not useful for comparative assessments.	Adan et al. 2011
Trichogramma Cacoeciae (egg parasitoid : Trichogrammatidae) Eggs	Confidor 200 SL (Bayer, Germany) "Field rate" of 350 ppm	Reduction in adult emergence.	Saber 2011
Mesocosm Studies			
Anagyrus pseudococci (parasitic wasp, Encyrtidae)	Flowers from buckwheat following soil applications (Marathon 1%G) Label rate: 1.4 g/pot 2x rate: 2.8 g/pot Pots 10.5 cm^2 ($1.05x10^{-7}$ ha) Rates appear to be given in units of formulation and not a.i.	Substantial reduction in survival following applications intended to reflect label rate and twice label rate. For all samples combined, residues in nectar were 6,550 ppb at 1x and 12,270 ppb at 2x (Table 2 of paper). Assuming that the application rates are expressed in units of formulation, the residue rate in nectar is about 55 ppb per lb a.i./acre (1x) and 42 ppb per lb a.i./acre (2x).	Krischik et al. 2007
Tiphia vernalis (ectoparasitoid wasp, Tiphiidae) Turf exposures	Merit 75 WP 0.45 lb a.i./acre Organisms placed on foliage shortly after drying.	Significant ($p<0.0001$) but not substantial increase in mortality (≈5% in males and 4.3-5% in females) (Table 2 of paper).	Oliver et al. 2006
Field Studies			
Encarsia citrina (parasitic wasp; Aphelinidae)	Marathon 60 WP Soil drench at 0.33 g/500 ml water; Foliar application at 0.15 g/500 ml of water	No significantly impact on the number of parasitoids emerging from *Euonymus* scale (*Unaspis euonymi*) with respect to controls.	Rebek and Sadof 2003
Turf, Indiana 6 replicate 10x10 plots with irrigation.	Merit 0.34 kg a.i./ha (≈0.3 lb a.i./acre	No effect on ant populations.	Zenger and Gibb 2001

A3 Table 5: Hemiptera

Hemiptera	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Adelges tsugae (Hemlock Woolly Adelgid; Adelgidae) Branch assay 12 blocks, s 20 cm branches per block.	Imidacloprid (99.2%) Branch assay Nominal Concentrations: 0, 1, 10, and 100 ppb Results assayed at 10, 20, and 30 days	30 day LC_{50}: 242 (105-411) ppb Working Note: While this agrees with Cowles et al. (2006), the mortality data in Table 1 look like the LC_{50} should be <100 ppb. This is probably due to the use of monitored concentrations in twigs rather than nominal concentrations.	Eisenback et al. 2010
Apolygus lucorum (mirid bug; Miridae) 4 days old 5/sex/group	Imidacloprid (95%) Contact assay, pipette 0.6 µL applied to dorsum Doses: 0.1 to 90 ng. Duration not clear.	LD_{50}: 6.07 (3.25-10.04) ng/insect The following doses are used in 'sublethal studies' summarized below: LD_5: 0.38 ng/insect LD_{25}:1.96 ng/insect LD_{40}:3.97 ng/insect	Tan et al. 2012
Bemisia tabaci (whitefly; Aleyrodidae) Adults 30 insects per replicate, 5 replicates per concentration	Imidacloprid (NOS), this may be a formulation. Cotton leaves grown in different concentrations of imidacloprid.	LC_{50} values of about 1 to 13.8 mg/L. These are not comparable to other LC_{50} values but can be used to assess sensitivities in different populations of whitefly. Working Note: See Fig 6 for concentrations of imidacloprid in the leaves. It looks like 1 mg/L solution resulted in leaf residues of about 15 ng/mL or 15 µg/L. Different life-stages assayed with little difference in LC_{50} values – i.e., 0.79 mg/L (1st instars) to 4.1 mg/L (1 day egg). See Table 1 of paper for more details.	Castle et al. 2014
Bemisia tabaci (whitefly; Aleyrodidae) Adults	Imidacloprid (96.8%) Leaf disc assay Aqueous conc.: 5, 10, 20, 40, 80 and 160 mg a.i./L	LC_{50}: 39.60 (33.19-48.11) mg a.i./L LC_{10}: 5.28 (3.63-7.01) mg a.i./L Working Note: Compare to results with formulation below from He et al. 2011. Also, see sublethal studies below.	He et al. 2013
Bemisia tabaci (whitefly; Aleyrodidae) Adults	Imidacloprid (NOS) This might be a formulation. Leaf dip assay. Few bioassay details.	LC_{50} values for 9 different populations Most and least sensitive below. 0.34 mg a.i./L USA-B strain 113.56 mg a.i./L in ESP-00 (Spain) Variability related to differences in expression of P450 CYP6CM1 gene. See Section 4.1.2.4.1.1 for discussion.	Karunker et al. 2008

<table>
<tr><th>Hemiptera</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Bemisia tabaci
(whitefly; Aleyrodidae)
Adults
20 females/dose
3 replicates/ concentration</td><td>TGAI (≥97%)
Oral, sucrose solution, ad libitum
5-6 concentrations (NOS)
Observation at 48 hours</td><td>0.24 (0.064-0.59) mg/L
See data on metabolites below.</td><td>Nauen et al. 1999</td></tr>
<tr><td>Myzus persicae
(green peach aphid; Aphididae)
Resistance close (5191A) and sensitive clone (4106A).</td><td>Imidacloprid (Sigma-Aldrich, UK)
Insects dosed with 0.25 µL solutions.
Organisms treated with piperonyl butoxide (PBO) dosed with 0.1% solution 5 hours prior to imidacloprid treatment.
Duration for observations not clear.</td><td>Reported LC_{50} values (Table 1)
<table><tr><th>Strain</th><th>PBO</th><th>LC_{50} (mg a.i./L)</th></tr><tr><td>4106A</td><td>No</td><td>1.13 (0.9-1.4)</td></tr><tr><td>5191A</td><td>No</td><td>31.1 (7-69.7)</td></tr><tr><td>4106A</td><td>Yes</td><td>0.1 (0.07-0.15)</td></tr><tr><td>5191A</td><td>Yes</td><td>1.55 (0.56-2.55)</td></tr></table>
Conversion to dose ng/insect based on application of 0.25 µL.
<table><tr><th>Strain</th><th>PBO</th><th>LD_{50} (ng/insect)</th></tr><tr><td>4106A</td><td>No</td><td>0.2825</td></tr><tr><td>5191A</td><td>No</td><td>7.775</td></tr><tr><td>4106A</td><td>Yes</td><td>0.025</td></tr><tr><td>5191A</td><td>Yes</td><td>0.3875</td></tr></table></td><td>Puinean et al. 2010</td></tr>
<tr><td>Nilaparvata lugens
(brown planthopper; Hemiptera)
5 replicates of 60 insects/replicate
3rd instars
Assayed over 9 generations (for resistance)</td><td>Imidacloprid (NOS)
Direct spray
5 concentrations (NOS)</td><td>LC_{50} values:
Generation 1:
48-hour LC_{50}: ≈ 40 mg a.i./L
Generation 9:
48-hour LC_{50}: ≈ 52 mg a.i./L

Working Note: Paper gives LC_{50} values as % (w/v). Converted to mg/L above. LC_{50} values approximated from Figure 3A of paper. For example, Fig. 3A reports the 48-hour LC_{50} as 0.004%.. 1%=10,000 ppm. Thus, the LC_{50} is 40 ppm (w/v) or 40 mg a.i./L.</td><td>Bullangpoti et al. 2007</td></tr>
<tr><td>Triatoma infestans
(vector for Chagas disease; Reduviidae)
1st instar, 5-7 days old
A minimum of 3 replicates with at least 10 insects/dose.</td><td>TGAI (98%)
Topical application to abdomen.
0.2 µL at concentrations of 0.0025-0.5 mg a.i./mL.
Observations at 24, 48, and 72 hours.</td><td>Below are based on 24-h mortality.
<table><tr><th>Population</th><th>Status</th><th>LD_{50} (ng/insect)</th></tr><tr><td>Suscept.</td><td>Starved</td><td>5.2 (3.4-7.8)</td></tr><tr><td>Suscept.</td><td>Fed</td><td>4 (2.4-7)</td></tr><tr><td>Resisitant</td><td>Starved</td><td>9.2 (7.4-11.2)</td></tr><tr><td>Resisitant</td><td>Fed</td><td>10.8 (6.4-19)</td></tr></table>Note: Above assay on insects susceptible and resistant to pyrethroids. See Table on p. 764 of paper. Feeding refers to pigeon blood.
Also did residue contact assays.</td><td>Carvajal et al. 2014</td></tr>
</table>

Hemiptera	Exposure	Response	Reference
Formulation			
Adelges tsugae (Hemlock Woolly Adelgid; Adelgidae) Branch assay 5 doses 30 individuals/ replicate, 8 replicates	Bayer Advanced Garden Tree & Shrub Insect Control. Concentrations: 0.01, 0.1, 1, 10, and 100 ppm used to soak branches. 19-21 day exposure	LC_{50}: 300 (150-600) ppb	Cowles et al. 2006
Agonoscena pistaciae (pistachio psylla; Psyllidae) 1 day old 5th instar nymphs 3 replicates, 20 nymphs/replicate	Confidor (0.35%) Whole insect dip (2 seconds) Concentrations: o, 53, 70, 105, 147, and 210 mg a.i./L 24-hour observation	LC_{50}: 138.21 (100.57-233.89) mg/L Working Note: Not directly comparable to most other assays but see parallel assays on two coleopterans (*Coccinella undecimpunctata* and *Adalia bipunctata*). Somewhat less toxic to the coleopterans. Note, however, that the dosing to the coleopterans did not involve whole insect dip.	Amirzade et al. 2014
Aphis pomi (apple aphid; Aphididae) 9 populations from British Columbia Canada and Washington State, U.S.	Admire (21.4% a.i., Canada) Whole insect dip assay (2 seconds) and leaf disk assays: 5 to 7 concentrations (NOS)	72-hour insect dip LC_{50}s: Lowest: 0.38 (0.22-0.56) mg a.i./L Highest: 1.46 (1.06-2.00) mg a.i./L Variability of factor of 3.8. 72-hour Leaf disk LC_{50}s: Lowest: 0.11 (0.09-0.13) mg a.i./L Highest: 0.83 (0.63-1.05) mg a.i./L Variability of factor of 7.5. Some differences in relative sensitivity for the 9 different populations in the two types of assays.	Lowery et al. 2005
Aphis spiraecola (apple aphid; Aphididae) 2 populations from British Columbia Canada and Washington State, U.S.	Admire (21.4% a.i., Canada) Whole insect dip assay (2 seconds) and leaf disk assays: 5 to 7 concentrations (NOS)	72-hour insect dip LC_{50}s: BC: 6.90 (5.34-8.57) mg a.i./L U.S.: 3.08 (2.21-4.18) mg a.i./L 72-hour Leaf disc LC_{50}s: BC: 0.40 (0.28-0.69) mg a.i./L U.S.: 2.44 (1.86-3.11) mg a.i./L	Lowery et al. 2005
Bemisia tabaci (whitefly; Aleyrodidae) Eggs, nymphs, and adults	BIDAN 10 WP (China) Egg dip and adult leaf dip bioassays: 1.5 to 400 mg a.i./L Nymph leaf dip: 6.25-200 mg a.i./L	**Life Stage** / **LC_{50} (mg a.i./L)** Adult: 53.54 (40.18-69.24) Egg: 83.77 (63.34-111.84) Nymph: 44.98 (38.87-51.34) Also gives LC_{40} and LC_{20} values. See below for sublethal effects at these lower doses.	He et al. 2011

Hemiptera	Exposure	Response	Reference
Bemisia tabaci (whitefly; Aleyrodidae) Populations (n=17) in 4 areas of Pakistan Adults	Confidor, 200 g a.i./L (Bayer, Germany) Leaf dip assays 48-hour observations	48-hour Leaf disc LC_{50}s: (Table 1) Lab Strain: 0.60 (0.33-0.87) mg a.i./L Most Sensitive Field Strain: 2.38 (1.14 to 3.62) mg a.i./L Most Tolerant Field Strain: 4.39 (2.51 to 6.46) mg a.i./L **Life Stage** / **LC_{50} (mg a.i./L)** Adult, Lab. — 0.60 (0.33-0.87) Nymph, Lab. — 0.15 (0.03-0.20) Adult, Field — 6.08 (2.83-9.31) Nymph, Field — 0.75 (0.44-1.02) Nymphs more sensitive by factors of about 4 to 8.1	Basit et al. 2013
Dicyphus tamaninii (mirid bug; Miridae) 3rd to 4th instars nymphs, 10 nymphs per leaflet, 5 leaflets per group.	Confidor 20LS (20% a.i.) 0.5 ml/L on treated tomato leaflets. Leaves fed at 1 to 30 day post-spray	Substantial mortality ranging from 33.7% 24 hours after exposure to 1-day residues, to 91.9 % 7 days after exposure to 1-day residues. Percent mortality declined with increasing residue time, with 2 to 26.0% mortality at 24 hours and 7-days, respectively, after exposure to 30day residues	Figuls et al. 1999
Oechalia schellembergii (Pentatomidae) 3 replicates of 10 adults/replicate	Imidacloprid (350 SC, NOS) Topical, spray 0.0053% a.i. (53 mg a.i./L) Observations at 48 hours	100% mortality vs 0% mortality in controls.	James and Vogele 2001
Pristhesancus plagipennis (common assassin bug, Reduviidae) 3 replicates of 12 newly emerged 1st instar nymphs/replicate	Imidacloprid (350 SC, NOS) Topical, spray 0.0053% a.i. (53 mg a.i./L) Observations at 48 hours	0% mortality vs 6% mortality in controls.	James and Vogele 2001
Geocoris punctipes (Big-eyed bug; Geocoridae) 8 days old, 6 males and 6 females per replicate, 6 replicates	Provado 1.6 flowable 0.052 kg a.i./ha (≈0.046 lb a.i./acre) Spray chamber assay. 72 hours	11.1% and 50.0% mortality among males and females, respectively. Control mortality not specified (Table 1 of paper). Egg consumption was significantly less than that of untreated controls (Table 2 of paper).	Elzen 2001
Geocoris punctipes (big eyed bug; Geocoridae)	Admire 2F (240 g a.i./L) Leaf uptake bioassay 24 hour exposure At least 5 concentrations/ assay	24h-LC_{50}: 5.18 (2.33-10.02) g a.i./L `Working Note: Above unit designation is not a typo. This is a very peculiar bioassay. Included because the assay covered 6 species.`	Prabhaker et al. 2011

Hemiptera	Exposure	Response	Reference
Hyaliodes vitripennis (Miridae) 18 insects per concentration, 3 replicates, nymphs and adults tested separately	Admire (240 g a.i. /L) Direct spray Leaves, sidewalls of plastic cage, and insects. Concentrations: 1/256 X to X, where X = label application rate of 0.0312 g a.i./L	Nymphs : LC_{50}: 0.0023 (0.0018 - 0.0029) g a.i./L Adult LC_{50}: 0.0011 (0.0008 - 0.0017) g a.i./L	Bostanian et al. 2001
Orius insidiosus (insidious flower bug; Anthocoridae)	Admire 2F (240 g a.i./L) Leaf uptake bioassay 24 hour exposure At least 5 concentrations/ assay	24h-LC_{50}: 2.78 (1.42-4.26) g a.i./L Working Note: Above unit designation is not a typo. This is a very peculiar bioassay. Included because the assay covered 6 species.	Prabhaker et al. 2011
Myzus persicae (green peach aphid; Aphididae) 18 populations within the eastern U.S.	Admire 2F (240 g a.i./L) Leaf dip assay. Observations at 72-hours	72-hour LC_{50}s (Table 1): Lowest: 0.4 (0.1-1.0) mg a.i./L Highest: 9.3.46 (5.9-14.1) mg a.i./L Variability of factor of 25.2.	Srigiriraju et al. 2010
Myzus persicae (green peach aphid; Aphididae) 8 populations within the state of Washington	Provado (28 mg a.i./L) Leaf dip assay. Observations at 72-hours Working Note: The description of Provado formulation seems incorrect.	72-hour LC_{50}s (Table 1): Lowest: 0.172 (0.128-0.221) mg a.i./L Highest: 0.842 (0.741-0.939) mg a.i./L Variability of factor of 4.9.	Unruh and Willett 2008
Myzus persicae (green peach aphid, red clone, Aphididae) 2^{nd} to 3^{rd} instars	Intercept 60WP (Bayer, Canada) Leaf dip assay Concentrations: 0.1 to 1000 mg a.i./L	LC_{50}: 392 µg a.i./L LC_{10}: 7 µg a.i./L Working Note: These are essentially a range-finding assay for sublethal studies. See below.	Janmatt et al. 2010
Orius armatus (predator on flower trips; Anthocoridae)	Confidor 200 SC (200 g a.i./L) Residue contact with treated beans at 50 mg a.i./L solution	No significant impact on eggs. Very modest mortality in nymphs.	Broughton et al. 2014
Orius armatus (predator on flower trips; Anthocoridae)	Confidor 200 SC (200 g a.i./L) Oral exposure to treated beans at 50 mg a.i./L solution	Mortality rate less than 22%. Post-exposure, an increase (although not statistically significant) was noted in egg production (hormesis?).	Broughton et al. 2014
Orius laevigatus (Insidious flower bug; Anthocoridae) 20 5th instar nymphs and 20 adults per concentration for each test	Confidor 200 SL Oral 72 hours	Nymph LC_{50}: 1.1 (0.1 - 2.9) mg a.i./L Adult LC_{50}: 2.1(1.0 -3.8) mg a.i./L	Delbecke et al. 1997

Hemiptera	Exposure	Response	Reference
Orius laevigatus (Insidious flower bug; Anthocoridae) 20 5th instar nymphs and 20 adults per concentration for each test	Confidor 200 SL Residue contact 72 hours	Nymph LC_{50}: 0.04 (0.0002 - 1.2) mg a.i./L Adult LC_{50}: 0.3(0.2 - 0.4) mg a.i./L	Delbecke et al. 1997
Orius laevigatus (Insidious flower bug; Anthocoridae) 8 days old, 6 males and 6 females per replicate, 6 replicates	Provado 1.6 flowable 0.052 kg a.i./ha (≈0.046 lb a.i./acre) Spray chamber assay. 72 hours Consumption of *Helicoverpa zea* eggs treated with a.i.	47.8% and 62.7% mortality among males and females, respectively. Control mortality not specified (Table 1 of paper). No significant impact on egg (*Helicoverpa zea*)consumption or fecundity (Table 2 of paper).	Elzen 2001
Pseudacysta perseae (avocado lace bug; Tingidae)	Admire Pro SC (550 g/L) Leaf assay 48 and 72 hour mortality	LC_{50}: 6.1 (4.4-7.4) ng/cm^2 leaf tissue. `Working Note: See parallel study on avocado thrips.`	Byrne et al. 2010
Sitobion avenae (wheat aphid; Aphididae)	Gaucho70WS (Bayer, Germany) Wheat seeds treated at 62.5-1000 mg a.i./kg seed.	24-h LD_{50}: 188 mg/kg seed `Working Note: The LD50 is in units of mg/kg seed and not kg bw.`	Miao et al. 2014
Trialeurodes vaporariorum (greenhouse whitefly; Aleyrodidae) Adults 7 populations including reference	Confidor WG-70 Leaf assay Concentrations: 0.5 to 245 mg a.i./L.	LC_{50}s: Lowest: 0.42 (0.10-1.20) mg a.i./L Highest: 4.34 (3.35-5.54) mg a.i./L Variability of factor of 10.46.	Ovcarenko et al. 2014
Sublethal Effects			
Aphis gossypii (greenfly; Aphididae)	Imidacloprid 35SC (formulation) Leaf assay, reproduction "Recommended concentration" (NOS)	Substantial decreases in longevity and fecundity.	Gerami et al. 2005
Aphis gossypii (greenfly; Aphididae)	Imidacloprid 35SC (formulation) Leaf assay, reproduction "Recommended concentration" (NOS)	Substantial decreases in longevity and fecundity.	Gerami et al. 2005

<table>
<tr><th>Hemiptera</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Apolygus lucorum
(mirid bug;
Miridae)
4 days old
5/sex/group</td><td>Imidacloprid (95%)
Contact assay
0.6 µL applied to dorsum
Duration not clear
Doses:
LD_5: 0.38 ng/insect
LD_{25}:1.96 ng/insect
LD_{40}:3.97 ng/insect</td><td>LD_5 and LD_{25} Decrease in pre-oviposition period and increase in development time for eggs. Decreased longevity in males.
LD_{40}: Eggs developed more rapidly but decrease in hatching success. Decreased longevity in males.</td><td>Tan et al. 2012</td></tr>
<tr><td>Bemisia tabaci
(whitefly;
Aleyrodidae)
Adults</td><td>BIDAN 10 WP (China)
Leaf dip assay: LC_{20} and LC_{40} (≈8.6 and 31 mg/L per Table 1 of paper.</td><td>Substantial and dose-related decreases honeydew excretion (i.e., decreased feeding). Rapid recovery (by 24 hours) after transfer to uncontaminated leaves. Food consumption remained depressed but not significantly so. See Table 2 of paper.
Substantial and dose-related decrease egg production/female. Recovery by 48 hours with marked (≈20-30%) but not statistically significant increase in egg production (hormesis?) (Table 2).
No marked or significant impact on longevity and fecundity (Table 4).
High dose lead to modest (≈18%) decrease in male/female ratio (Table 5).</td><td>He et al. 2011</td></tr>
<tr><td>Bemisia tabaci
(whitefly;
Aleyrodidae)
Adults</td><td>Imidacloprid (96.8%)
Leaf disc assay
Aqueous conc.: 5 and 20 mg a.i./L. See above. These are about the LD_{10} and 0.5 of LC_{50}.</td><td>Significant lethality at high dose (to be expected) (Fig. 1).
Significant, substantial, and dose-related decreases in honeydew production (i.e., feeding inhibition). Fig. 2.
Decrease in fecundity/egg production (Fig. 3) do to decrease in time spent feeding. Recovery not assayed.

Working Note: Compare to results with formulation (above) from He et al. 2011.</td><td>He et al. 2013</td></tr>
<tr><td>Bemisia tabaci
(whitefly;
Aleyrodidae)
Adults, <2 days old
11 replicates involving a total of 167 organisms.</td><td>Admire Pro 4.6 SC
Contact assay: filter paper at rate equivalent to 0.089 kg a.i./ha (≈0.08 lb a.i./acre)
48 hour observation period</td><td><table><tr><th>Time (hrs)</th><th>% Mortality</th><th>% Feeding Inhibition</th></tr><tr><td>0-1.5</td><td>2</td><td>54</td></tr><tr><td>1.5-2</td><td>8</td><td>80</td></tr><tr><td>3-3.5</td><td>NR</td><td>92</td></tr><tr><td>24-24.5</td><td>39</td><td>97</td></tr><tr><td>48</td><td>74</td><td>98</td></tr></table>Significant feeding inhibition and mortality. Unremarkable.</td><td>Cameron et al. 2013</td></tr>
</table>

Hemiptera	Exposure	Response	Reference
Myzus persicae (green peach aphid; Aphididae) First instars (24 h old) 3 replicates, 5/dose	Admire 240 SC (240 g a.i./L) Leaf dip (contact) assay Concentrations: 0.025, 0.1, 0.25, 1.0, 2.5, 10, and 25 µg/L Observation period: 4 generations (Table 1)	Authors suggest a hormetic response in terms of increases in reproduction parameters. Note substantial scatter.	Ayyanath et al. 2013
Myzus persicae (green peach aphid; Aphididae) First instars (24 h old) 3 replicates, 5/dose	Admire 240 SC (240 g a.i./L) Topical, direct spray Concentrations: 0.2, 0.6, 2.0, 6.0, 20, 60, and 200 µg/L Observation period: 2 generations (Table 1)	Authors suggest a hormetic response in terms of increases in reproduction parameters. Note substantial scatter.	Ayyanath et al. 2013
Myzus persicae (green peach aphid, red clone, Aphididae) Adults	Intercept 60WP (Bayer, Canada) Leaf dip assay Concentrations: 0, 1, 3, 6, and 9 µg a.i./L 3 day exposures	No significant effects on mortality or offspring per adult (Table 1) `Working Note: These are sublethal doses based on preliminary acute lethality studies. See above.`	Janmatt et al. 2010
Myzus persicae (green peach aphid, red clone, Aphididae) 1st instar nymphs	Intercept 60WP (Bayer, Canada) Nymphs taken from adults assay (see above). Concentrations: 0, 1, 3, 6, and 9 µg a.i./L 6 day exposures	Significant increase in mortality at 2 highest doses (Table 2)	Janmatt et al. 2010

Hemiptera	Exposure	Response	Reference
Metabolites			
Bemisia tabaci (whitefly; Aleyrodidae) Adults 20 females/dose 3 replicates/ concentration	olefin metabolite (≥97%) [Compound 1 in paper] Oral, sucrose solution, ad libitum 5-6 concentrations (NOS) Observation at 48 hours	0.025 (0.017-0.032) mg/L Working Note: See data above for TGAI. See data in Table A3-1 for toxicity data on metabolites in honeybee. This is the only metabolite markedly more toxic than imidacloprid.	Nauen et al. 1999
Bemisia tabaci (whitefly; Aleyrodidae) Adults 20 females/dose 3 replicates/ concentration	4-hydroxy metabolite (≥97%) [Compound 2 in paper] Oral, sucrose solution, ad libitum 5-6 concentrations (NOS) Observation at 48 hours	0.15 (0.039-0.32) mg/L Working Note: See data above for TGAI. See data in Table A3-1 for toxicity data on metabolites in honeybee. This is modestly more toxic than imidacloprid.	Nauen et al. 1999
Bemisia tabaci (whitefly; Aleyrodidae) Adults 20 females/dose 3 replicates/ concentration	5-hydroxy metabolite (≥97%) [Compound 3 in paper] Oral, sucrose solution, ad libitum 5-6 concentrations (NOS) Observation at 48 hours	2.4 (0.67-7.2) mg/L Working Note: See data above for TGAI. See data in Table A3-1 for toxicity data on metabolites in honeybee. This is less toxic than imidacloprid by a factor of 10.	Nauen et al. 1999
Bemisia tabaci (whitefly; Aleyrodidae) Adults 20 females/dose 3 replicates/ concentration	dihydroxy metabolite (≥97%) [Compound 4 in paper] Oral, sucrose solution, ad libitum 5-6 concentrations (NOS) Observation at 48 hours	>60 mg/L Working Note: See data above for TGAI. See data in Table A3-1 for toxicity data on metabolites in honeybee. This is less toxic than imidacloprid by a factor of >250.	Nauen et al. 1999
Bemisia tabaci (whitefly; Aleyrodidae) Adults 20 females/dose 3 replicates/ concentration	urea metabolite (≥97%) [Compound 5 in paper] Oral, sucrose solution, ad libitum 5-6 concentrations (NOS) Observation at 48 hours	>60 mg/L Working Note: See data above for TGAI. See data in Table A3-1 for toxicity data on metabolites in honeybee. This is less toxic than imidacloprid by a factor of >250.	Nauen et al. 1999
Mesocosm Studies			
No studies identified			
Field Studies			

Hemiptera	Exposure	Response	Reference
Eastern hemlocks for control of hemlock woolly adelgid.	Merit 75 WP 1.5 g imidacloprid per 2.5 cm of tree DBH (full rate) as well doses half and one-quarter of this dose.	Clear dose-related decrease in live HWA, significant mortality at two higher doses (Fig 2 of paper). Mean concentration in trees with <30% proportion of shoots infested was 187 ppb. Mean concentration in trees with 0% infested was 211 ppb. Abstract states no infestation in trees with >413 ppb.	Eisenback et al. 2014
Pseudacysta perseae (avocado lace bug; Tingidae) Commercial avocado groves, California	Admire Pro SC (550 g/L) Bioassays on leaves with imidacloprid residues 0.560 kg/ha (≈0.5 lb a.i./acre) or 0.280 kg/ha (≈0.25 lb a.i./acre)	Levels of imidacloprid in leaves sufficient to control lace bug. Working Note: See parallel observations on avocado thrip.	Byrne et al. 2010

A3 Table 6: Coleoptera

Coleoptera	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Anoplophora glabripennis (Asian longhorned beetle) 5th instars	Imicide (10% a.i.) Dietary conc: 0, 0, 0.16, 1.6, 16, and 160 mg/kg diet. Included formulation control. 12 weeks	LC_{50}: 4.92 mg/kg diet Increasing weight loss with increasing concentrations due to decreased food consumption (antifeedant effect). Mortality inversely correlated with concentration. Food consumption given in Fig. 2. Working note: Cannot reliably calculate LD_{50} values.	Poland et al. 2006a
Harpalus pennsylvanicus (Carabid beetle) 10 adult beetles per replicate; 4 replicate plots each treatment plus controls	Imidacloprid (NOS) Dietary: pellets, with imidacloprid at label rate (0.336 kg/ha) and 0.5 label rate – i.e., ≈ 0.15 and 0.3 lb a.i./acre. Observations at 4 hr, 12 hr, and daily for 7 days.	Intoxication of all imidacloprid treated beetles (both doses) between 4 hours and 1 day post-exposure; most beetles were recovered by day 7.	Kunkel et al. 2001
Harpalus pennsylvanicus (Carabid beetle) 10 adult beetles per replicate; 3 replicate plots each treatment plus controls	Imidacloprid (NOS) Contact: Spray with rates equivalent to ≈ 0.075, 0.15, and 0.3 lb a.i./acre. Observations at 4 hr, 12 hr, and daily for 7 days.	Most beetles incapacitated within 4 hours, appearing dead or nearly dead. All beetles incapacitated by 1 day followed by recovery within 4 days for more than 85% of the beetles.	Kunkel et al. 2001
Harpalus pennsylvanicus (Carabid beetle) 3 replicate pairs, 10 beetles per replicate	Imidacloprid (NOS) Residue: Plots sprayed at 0.3 lb a.i./acre, with and without irrigation. Observations at 48 hrs.	Significant residual toxicity with respect to controls was observed on non-irrigated plots only, Most of the intoxicated beetles (80%) recovered.	Kunkel et al. 2001
Harpalus pennsylvanicus (Carabid beetle) Control and treated beetles, 3 replicates, 15 beetles each	Imidacloprid (NOS) Predation by ants following oral exposures of beetle to pellets treated at ≈0.3 lb/acre	Intoxicated beetles, but not untreated controls, were captured by predatory ants.	Kunkel et al. 2001
Harpalus pennsylvanicus (Carabid beetle) Male/female pairs,	Imidacloprid (NOS) Fecundity Oral exposures to pellets treated at ≈0.3 lb/acre for 1 day followed by untreated food.	No effect on egg fecundity or time to first oviposition. Egg hatch similar to controls.	Kunkel et al. 2001

Coleoptera	Exposure	Response	Reference
Hippodamia convergens (lady beetle)	TGAI (95%) Topical, micropipette Concentrations: 10, 50, 100, 200, 300 and 800 mg a.i./L.	LD_{50} values **Hours** / **LD_{50} (µg/g bw)** 24 / 1.8 (1.0-2.8) 48 / 0.7 (0.4-1.1) 72 / 0.4 (0.1-1.0) Working Note: Results given as µg/g bw but insect body weights not reported in paper.	Kaakeh et al. 1996
Laricobius nigrinus (HWA predator ;Derodontidae) Average bw: 0.71 mg	Imidacloprid (99.2%) Topical application, abdomen. Doses: 0, 0.005, 0.05, 0.5, 5, and 50 ng/beetle	6-Day LD_{50}: 1.8 (1.7-133.7) ng/beetle	Eisenback et al. 2010
Sasajiscymnus tsugae (predator of HWA, Coccinellidae) Average bw: 0.39 mg	Imidacloprid (99.2%) Topical application, abdomen. Doses: 0, 0.005, 0.05, 0.5, 5, and 50 ng/beetle	6-Day LD_{50}: 0.71 (0.5-1.7) ng/beetle	Eisenback et al. 2010
Formulation			
Adalia bipunctata (predator on pistachio psylla; Coccinellidae) 4th instar larvae 3 replicates, 20 larvae/replicate	Confidor (0.35%) Topical (micro-syringe) Concentrations: o, 53, 70, 105, 147, and 210 mg a.i./L 24 hour observation period Working Note: Volume of application not specified.	LC_{50}: 218.89 (116.25-350.37) mg/L Working Note: See parallel assays on Hemipteran, *Agonoscena pistaciae*. Note that dosing to *A. pistaciae* involved whole insect dip.	Amirzade et al. 2014
Agriotes obscurus (wireworm; Elateridae) Late instars, at least 16 mm long	Imidacloprid (NOS) Topical assay/spray.	LC_{50} = 0.83 (0.69-0.97) % Working note: Used a 19:1 solution of acetone:olive oil and report LC_{50}s as percent. Not clear if the % refers to w/w or w/v.	Van Herk et al. 2008
Agrilus planipennis (emerald ash borer; Buprestidae)	Imicide (10% a.i.) with 14C-imidacloprid injected into ash. Leaf residues from ≈27 µg/g (DAT 1) to ≈1 µg/g (one-year).	Substantial mortality at imidacloprid residues as low at about 1 µg/g. Nearly complete mortality as residues approached 200 µg/g.	Mota-Sanchez et al. 2009

Coleoptera	Exposure	Response	Reference
Anoplophora glabripennis (Asian longhorned beetle) Adults, 15/dose About 46 days old.	TGAI Oral Exposure: 1 µl doses of 0, 2, 10, or 50 ppm Above corresponds to doses of 0, 2, 10, and 50 ng/insect Single dose	Mortality (Fig. 1 of paper Controls: Only about 5% mortality over 25 days. 2 ng: Over 50% mortality by 25 days. 10 ng: Complete mortality by day 20 50 ng: Complete mortality by day 10.	Ugine et al. 2011
Anoplophora glabripennis (Asian longhorned beetle) Female Adults,	TGAI Oral Exposure: 1 µl doses of 0, 2, 10, 20, or 30 ppm Above corresponds to doses of 0, 2, 10, 20, or 30 ng/insect Repeated daily doses for 10 weeks.	10-60% mortality by day 70 (See Fig. 3 of paper. Mortality at high dose was only about 20%. Mortality at 20 ng/insect was about 60%. Mortality at 2 ng/insect was only about 10%.	Ugine et al. 2011
Anoplophora glabripennis (Asian longhorned beetle) Adults, including young and mated	Imidacloprid formulation (NOS) Dietary following tree injection. Feeding on twig bark from injected trees.	14-day LC_{50}: 4.0 (3.1-4.8) ppm 21-day LC_{50}: 1.3 (1.0-1.5) ppm	Ugine et al. 2012
Coccinella transversalis (transverse ladybird; Coccinellidae) 3 replicates of 10 adults/replicate	Imidacloprid (350 SC, NOS) Topical, spray 0.0053% a.i. (53 mg a.i./L) Observations at 48 hours	50% mortality vs 5% mortality in controls	James and Vogele 2001
Coccinella undecimpunctata (predator on pistachio psylla; Coccinellidae) 4th instar larvae 3 replicates, 20 larvae/replicate	Confidor (0.35%) Topical (micro-syringe) Concentrations: o, 53, 70, 105, 147, and 210 mg a.i./L 24 hour observation period Working Note: Volume of application not specified.	LC_{50}: 447.82 (290.64-1022.79) mg/L Working Note: See parallel assays on Hemipteran, *Agonoscena pistaciae*. Note that dosing to *A. pistaciae* involved whole insect dip.	Amirzade et al. 2014
Dicranolaius bellulus (red and blue beetle [Australia]; Myridae) 3 replicates of 10 adults/replicate	Imidacloprid (350 SC, NOS) Topical, spray 0.0053% a.i. (53 mg a.i./L) Observations at 48 hours	0% mortality in both control and treated organisms.	James and Volel 2001

Coleoptera	Exposure	Response	Reference
Leptinotarsa decemlineata (Colorado potato beetle; Chrysomelidae) 47 populations, field collected plus laboratory strain. 1st instars 10/dose	Admire F (240 g/L) Dietary exposures. At least 3 replicates per assay with 5 doses each Duration unclear.	Laboratory strain: LC_{50}: 0.6 (0.32-149) mg/L Field populations: 0.12 to 11.71 mg/L Sensitivities of field populations highly correlated with sensitivity to thiamethoxam ($p<0.0001$, $r^2=0.82$). See Fig. 1 in paper. Working Note: Presumed resistance by up to a factor of about 100 based on LC_{50} values.	Alyokhin et al. 2007
Plectrodera scalator (Cottonwood beetle) 5th instars	Imicide (10% a.i.) Dietary conc: 0, 0, 0.16, 1.6, 16, and 160 mg/kg diet. Included formulation control. 12 weeks	LC_{50}: 1.78 (0.0067 - 21.88) mg/kg diet. Increasing weight loss with increasing concentrations due to decreased food consumption (antifeedant effect). Food consumption given in Fig. 2. Working note: Cannot reliably calculate LD_{50} values.	Poland et al. 2006a
Rodolia cardinalis (cardinal ladybird beetle; Coccinellidae) 10-15 adults per replicate 3 replicates plus untreated controls;	Provado 1.6 Flowable 72-hour contact assay Citrus leaves from trees treated with by soil drench (0.56 kg a.i./ha) or foliar spray application (0.14 kg a.i/ha). Leaves collected on 26, 35, 42, 51, 77 and 86 days post-treatment	48-hour post-treatment adult mortality and 7-day post-treatment assessment of emerged larvae and number of progeny per female beetle: foliar application significantly reduced adult survival and progeny per female 26 days after treatment.	Grafton-Cardwell and Gu 2003
Rodolia cardinalis (cardinal ladybird beetle; Coccinellidae) 3 replicates, cottony cushion scale larvae provided every 2-3 days;	Provado 1.6 Flowable 72-hour contact assay 20-day contact only exposure to treated or untreated leaves, as above. Larvae placed on scale-infested leaves 6 days after plants and scale were treated	Larval mortality and stage of development evaluated every 2-3 days for 20 days exposure to treated or untreated leaves. No larvae survived in either treatment. All died within 2-3 days following exposure to leaves and insects treated by soil drench, and within 8 days following exposure to insects and leaves treated by foliar application	Grafton-Cardwell and Gu 2003

Coleoptera	Exposure	Response	Reference
Sublethal Effects			
Serangium japonicum (predator on white fly; Coccinellidae)	BIDAN 10WP (China) Direct spray: 40 ppm (field solution) sprayed on to *B. tabaci* eggs on leaves.	No mortality after consuming treated eggs. Decrease rate of egg consumption (feeding inhibition). Effects were rapidly reversible on transfer to uncontaminated media. Also did glass residue studies which demonstrated clear dose-response relationship (Fig. 1 of paper).	He et al. 2012
Mesocosm Studies			
Coccinella septempunctata (a ladybird beetle; Coccinellidae) 2nd instar larvae	Imidacloprid (97.3%) Direct spray of microcosms. Single applications. Application rates: 0.85, 1.71, 3.42, 6.83, and 13.66 g a.i./ha. 18 day observation period.	72-h LD_{50}: 683.2 (596.8-790.4) g/ha Decreases in survival at two highest application rates. NOAEC (mortality): 3.42 g a.i./acre Note that NOAEC is based on lack of a statistically significant difference from control mortality. A dose-related trend in mortality was apparent. A more reasonable NOAEC would be 1.71 g a.i./acre. See Figure 1 of paper. EC_{50} for egg production: 26.63 g a.i./ha.	Yu et al. 2014
Field Studies			
Michigan, ash trees (Fraxinus spp.) for the control of *Agrilus planipennis* (emerald ash borer; Buprestidae)	Tree injections and bark applications with and without Pentra-Bark	Effective (EAB at 57-68% of controls) when applied in two consecutive years. Not effective when applied in only one year. Pentra-Bark did not significantly improve control.	McCullough et al. 2011
Turf, Indiana 6 replicate 10x10 m plots with irrigation. Japanese beetle control	Merit 0.34 kg a.i./ha (≈0.3 lb a.i./acre	Significantly fewer white grubs and eggs of Japanese beetles. Imidacloprid-treated plots had no grubs at all, in comparison with an average of 10.2 grubs per control plot.	Zenger and Gibb 2001
Tuff, Kentucky Japanese beetle control @@ Field plots, ≈1x1 m	Merit 75 WP 0.34 kg a.i./ha (≈0.3 lb a.i./acre	Substantial reduction in total eggs and viable females. Correlation of levels of imidacloprid in soil (0.1 to 2 ppm) and group mortality.	George et al. 2007
Tree injection, loblolly pine Southern pine engraver beetles (Curculionidae) and wood borers (Cerambycidae)	IMA-jet (5%) Tree injection 0.08 g active per cm DBH	Ineffective in reducing infestations. See esp. Tables 4 and 6 of paper.	Grosman and Upton 2006

A3 Table 7: Other Insects

Insects, Other	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Aedes aegypti (mosquito; Culicidae: Diptera) 4-day old females, 25 per dose	Imidacloprid (NOS) Topical, application of 0.3 µL imidacloprid solutions	Reported 24-hour LC_{50} values (Table 1) Sensitive strain: 6.830 (5.577-7.964) mg/L Tolerant strain: 8.352 (7.221-9.463) mg/L 24-hour LD_{50} values based on 0.3 µL application Sensitive strain: ≈ 2.05 ng/insect Tolerant strain: ≈ 2.5 ng/insect Working Note: Tolerance was seen in the larvae. The larval data are summarized in Appendix 8 (aquatic invertebrates).	Riaz et al. 2013
Archimantis sp. (Praying mantis [Australia] , Mantodea: Mantidae) 2 replicates of 10 newly emerged 1sr instar nymphs/ replicate	Imidacloprid (350 SC, NOS) Topical, spray 0.0053% a.i. (53 mg a.i./L) Observations at 48 hours	100% mortality vs 0% mortality in controls	James and Vogele 2001
Blattella germanica (German cockroach, Blattodea: Blattellidae) 7-14 day old male adults	TGAI (95%) Topical, 1 µL solution thorax (2 µL of 50% concentration of test solution) Observations at 24 hours.	24h-Reported LC_{50} 0.0216 (0.0145-0.0337) Working Note: Units of LC_{50} expressed at percent. Thus, the LC_{50} would correspond to 216 mg/L or 0.216 mg/mL or 0.216 µg/µL. Estimated LD_{50} based on 1 µL/insect LD_{50}: 0.216 µg/insect or 216 ng/insect.	Sims and Appel 2007
Ctenocephalides felis (cat flea; Siphonaptera: Pulicidae) Adults 10 populations	TGAI (97.8%) Micro-syringe application. 24 hour period	24h-LD_{50} values (Table 3 of paper): Most sensitive: 0.02 (0.015-0.031) ng/flea Most tolerant: 0.19 (0.15-0.22) ng/flea Range of about 9.5.	Rust et al. 2014
Ctenocephalides felis (cat flea; Siphonaptera: Pulicidae) Larva 10 populations	TGAI (97.8%) Incorporated in rearing medium. Duration of exposure not clear.	24h-LC_{50} values (Table 5 of paper): Most sensitive: 0.11 (0.07-0.154) mg/kg medium Most tolerant: 0.21 (0.133-0.272) mg/kg medium Range of about 1.9.	Rust et al. 2014

Insects, Other	Exposure	Response	Reference
Drosophila melanogaster (Diptera: Drosophilidae) 2nd instar larvae, 3 replicates of 30 insects per replicate. Three strains from Oregon.	TGAI (96.4%) Oral in food (artificial diet medium). 7 doses per assay. Results appear to be expressed as µg/g food.	**Strain** / **LD_{50} (mg/kg food)** CS: 0.4 (0.31-0.56) w^{1118}: 1.39 (1.03-1.86) Oregan: 0.81 (0.52-1.11)	Arain et al. 2014
Drosophila melanogaster (Diptera: Drosophilidae) Adults, 4 replicates of 3-4 insects per replicate. Three strains from Oregon.	TGAI (96.4%) Oral assay (wells with sucrose solution) 7 doses per assay. Results appear to be expressed as µg/g sucrose. Observations at 24 h.	**Strain** / **24 h-LD_{50} (µg/g solution)** CS: 4.58 (2.61-6.68) w^{1118}: 4.57 (1.64-6.73) Oregan: 4.12 (3.12-5.41) Working Note: For both adults and larvae, authors give RQs based on application rate (g a.i./ha) ÷ LD_{50} (µg/g food). Appears to be a categorization score. Does not make sense as standard RQ from EPA.	Arain et al. 2014
Drosophila melanogaster (Diptera: Drosophilidae) 3-4 days old adult male and female, ≈20/sex/dose	Imidacloprid (99.5%) Oral, sucrose solution 18 hour exposure 10 concentrations of 7.8 µM to 3.1 mM (≈ 2 to 792 mg a.i./L)	Acute LC_{50}s Males: 1304 µM (333 mg a.i./L) Females: >3.1 mM (>792 mg a.i./L) Working Note: As with housefly, females more tolerant than males. See Kavi et al. (2014) below.	Charpentier et al. 2014
Drosophila melanogaster (Diptera: Drosophilidae) 3rd instar larvae ≈20/dose	Imidacloprid (99.5%) Oral, sucrose solution 18 hour exposure 7 concentrations of 11.1 µM to 0.5 mM (≈2.8 to 792 mg a.i./L)	Acute LC^{50}s 157 µM (≈40 mg a.i./L) Working Note: Larvae much more sensitive than adults.	Charpentier et al. 2014
Musca domestica (housefly; Diptera) 400 to 1275 flies per assay 9 strains in Florida 3-5 days old 25 per dose Minimum of 4 replicates/bioassay	TGAI (99.5%) Sugar cube feeding. 72-hour observation	Strain / Male LC_{50} (µg/g food) / Female LC_{50} (µg/g food) Lab: 0.0029 / 0.012 KS8S3: 0.39 / 28 Resistance Factor: 134 / 2333 Only least and most resistant strains summarized above. Confidence intervals not summarized above but given in Table 2 of paper.	Kavi et al. 2014

Insects, Other	Exposure	Response	Reference
Musca domestica (housefly; Diptera) Females, 25 per dose. Two strains: Univ. of California at Riverside (UCR) and California dairy strain (BS)	TGAI (NOS, Chem Service Inc., West Chester, PA) Cotton wicks soaked in sugar with various concentrations of imidacloprid. 72-hour observation	<table><tr><th>Strain</th><th>Choice LC_{50} (mg/L)</th><th>No choice LC_{50} (mg/L)</th></tr><tr><td>UCR</td><td>15.1 (13-17.5)</td><td>69.2 (38.4-117.3)</td></tr><tr><td>BS</td><td>155.9 (110-194)</td><td>N/A[1]</td></tr></table>[1] Insufficient mortality.	Gerry and Zhang 2009
Formulation			
Scirtothrips perseae (avocado thrip; Thysanoptera: Thripidae)	Admire 240 SC (240 g/L) Application: 1.17 L/ha (≈0.25 lb a.i./acre) Oral exposure to contaminate leaf disks.	Marked mortality (80%) from leaves with residues of 6 ng/cm^2. By day 47, only 10% mortality although concentrations on leaves were about 7 µg/cm^2.	Byrne et al. 2005
Scirtothrips perseae (avocado thrip; Thysanoptera: Thripidae)	Admire Pro SC (550 g/L) Leaf assay 48 and 72 hour mortality	LC_{50}: 72.7 (61.5-86.7) ng/cm^2 leaf tissue. Working Note: See parallel study on avocado lace bug.	Byrne et al. 2010
Chronic			
Drosophila melanogaster (Diptera: Drosophilidae) 5 virgin males and 5 virgin females, <6 hours old	Imidacloprid (99.5%) Oral, sucrose solution 5 day exposure 10 concentrations of 7.8 µM to 3.1 mM (≈ 2 to 792 mg a.i./L)	Chronic LC_{50}s Males: 45 µM (11.5 mg a.i./L) Females: 18 µM (4.6 mg a.i./L) Larvae: 3 µM (0.77 mg a.i./L) Decrease in fecundity at 1.96 nM (0.0005 mg a.i./L) Mating increased at 0.391 nM (≈100 ng/L) Working Note: Compare with acute exposures. Males not the more sensitive than females in this chronic study. As with acute, larvae the most sensitive. Authors note that increased mating in *Drosophila* has also been observed with lead.	Charpentier et al. 2014

<table>
<tr><th>Insects, Other</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Sublethal Effects</td><td></td><td></td><td></td></tr>
<tr><td>Tryporyza incertulas (yellow stem borer, Lepidoptera: Pyralidae)
On spotted rice plants</td><td>10% imidacloprid WP (Yangnong Chemical Group Ltd. Co., China)
Treatment of spotted rice plants.
Application rates: 0.015 kg/ha (≈0.013 lb a.i./acre) and 0.0375 kg/ha (≈0.033 lb a.i./acre)</td><td>Increase in fecundity and larval weights.
No adverse effects</td><td>Wang et al. 2005a</td></tr>
<tr><td>Mesocosm Studies</td><td></td><td></td><td></td></tr>
<tr><td>Reticulitermes flavipes (eastern subterranean termite, Blattodea: Isoptera: Rhinotermitidae)
Workers, 24 per dose, 25 replicates</td><td>Premise 75 WP (Bayer, Missouri)
75% a.i. based on Bayer label.
Bioassays in different soils with concentrations of 0.1 to 50 ppm)
Observation period up to 21 days.</td><td>NOAEL/LOAEL (mortality) at 21 days
<table>
<tr><th>Soil</th><th>NOAEL (ppm soil)</th><th>LOAEL (ppm soil)</th></tr>
<tr><td>Sand</td><td>0.1</td><td>0.5</td></tr>
<tr><td>Sandy loam</td><td>5.0</td><td>10.0</td></tr>
<tr><td>Loam</td><td>5.0</td><td>10.0</td></tr>
<tr><td>Silty clay loam</td><td>N/D</td><td>2.5</td></tr>
</table></td><td>Ramakrishnan et al. 2000</td></tr>
<tr><td>Field Studies</td><td></td><td></td><td></td></tr>
<tr><td>Coptotermes formosanus (Formosan subterranean termite; Blattodea, Isoptera, Rhinotermitidae)
Louisiana, USA
40 ha total area (other pesticides examined)</td><td>Premise (NOS)
0.1% (1,000 mg a.i./L) foam treatment of 57 trees.</td><td>Termites collected from treated areas did not feed and did not survive beyond 14 days.</td><td>Osbrink and Lax 2003</td></tr>
<tr><td>Coptotermes formosanus (Formosan subterranean termite; Blattodea, Isoptera, Rhinotermitidae)
Louisiana, USA
40 ha total area (other pesticides examined)</td><td>Premise 75 (Bayer)
0.05% (500 mg a.i./L) soil treatments around buildings.</td><td>No substantial impact on termite populations.
Field NOAEC: 500 mg/L (applied solution).
Working Note: This is the concentration of imidacloprid in the solution applied to soil and not the concentration in the soil. This cannot be directly compared to Ramakrishnan et al. (2000) assay in termites or soil assays in other species.</td><td>Osbrink et al. 2005</td></tr>
</table>

Insects, Other	Exposure	Response	Reference
Rhyacionia frustrana (pine tip moth, Lepidoptera)	SilvaShield Forestry Tablets (Bayer), 20% a.i. Placed in planting hole during planting.	Substantial reduction in damage to trees for two seasons (Figure 1).	Asaro and Creighton 2011
Scirtothrips perseae (avocado thrip; Thysanoptera: Thripidae) Commercial avocado groves, California	Admire Pro SC (550 g/L) Bioassays on leaves with imidacloprid residues 0.560 kg/ha (≈0.5 lb a.i./acre) or 0.280 kg/ha (≈0.25 lb a.i./acre)	Levels of imidacloprid in leaves insufficient to control thrips. Working Note: See parallel observations on avocado lace bug.	Byrne et al. 2010

A3 Table 8: Mites and Spiders

Mites/Spiders	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Pardosa pseudoannulata (spider, Araneae: Lycosidae) Sub-adults Groups of 60-150 per dose	Imidacloprid (93.8%) Dip assay (20 seconds) 0, 12.5, 25, 50, 100, and 200 mg/L Observations at 24 hours	LC_{50}: 40.44 mg a.i./L Sublethal studies on survivors described below.	Chen et al. 2012
Formulation			
Amblyseius victoriensis (mite; Acari: Phytoseiidae) 5-10 females per treated leaf disc, two leaf discs per treatment, test conducted 3 times	Confidor 350 SC (5.25 g/100L or 0.0053% a.i.) Sprayed on grape leaf discs at field rate to control aphids and 10X this rate	No mortality observed in controls or at field application rate. 34.4% mortality observed at 10X field rate	James 1997
Anystis baccarum (predatory mite, Acari: Anystidae)	Admire 24% (Bayer) Petri dish contact assay. Concentrations of 0.01689 to 2.70 mg a.i./L	No mortality in excess of control (Table 2 of paper).	Laurin and Bostanian 2007
Typhlodromus dossei (Australian mite, Phytoseiidae) Total of 55 to 58 animals per dose. Replicates not clear	Imidacloprid (350 SC, NOS) Topical, spray 0.0053% a.i. (53 mg a.i./L) and 0.053% a.i. (530 mg a.i./L) Observations at 48 hours	No mortality at either dose or in controls.	James and Vogele 2001

Mites/Spiders	Exposure	Response	Reference
Typhlodromus dossei (Australian mite, Phytoseiidae) Total of 55 to 62 animals per dose. Replicates not clear.	Imidacloprid (350 SC, NOS) Topical, spray 0.0053% a.i. (53 mg a.i./L) and 0.053% a.i. (530 mg a.i./L) Observations at 48 hours	No mortality in low dose or controls. 19% mortality at high dose.	James and Vogele 2001
Sublethal Effects			
Amblyseius victoriensis (mite; Acari: Phytoseiidae) 50 females per grapefruit leaf platform, three platforms per treatment	Confidor 350 SC 0.0053% a.i. "field rate" – i.e., 53 ppm Egg toxicity assay Sprayed on leaves. Eggs recorded 12 days post-exposure.	Egg production in imidacloprid-exposed females (1.9 - 2.0eggs per female per day) was significantly increased with respect to untreated controls (1.3 - 1.6 eggs per female per day).	James 1997
Neoseiulus fallacis (mite; Acari: Phytoseiidae) 7-8 day old females 4 replicates on separate dates	Provado 1.6 F (Bayer) 60 ppm a.i. Detached leaves dipped into imidacloprid. Insects placed on leaves.	Significant decrease in oviposition rate (Table 3). No remarkable effects in other sub-experiments.	Villanueva and Walgenbach 2005
Pardosa pseudoannulata (spider, Araneae: Lycosidae) Sub-adults Groups of 60-150 per dose	Imidacloprid (93.8%) Emersion assay 0, 12.5, 25, 50, 100, and 200 mg/L Observations at 24 hours	Survivors from acute bioassay (summarized above under Acute Lethality studies). Dose-related decrease in number of eggs produced. NOAEC: 12.5 mg/L (Fig. 1). Dose-related delayed development of larvae. At lower doses, delay seen only in late (i.e., 7^{th}) instars. NOAEC not defined (Table 2 of paper). Significant increase in prey attack rate at 12.5 mg/L (hormesis?). At higher concentrations, attack rates significantly diminished in dose-related manner. Decrease in carboxylesterase, AChE and mixed function oxidase activities (Table 4).	Chen et al. 2012
Tetranychus cinnabarinus (carmine spider mite, Acari: Tetranychidae) eggs	10% WP formulation (Jiangsu Wujiang Pesticide Ltd. Co., China). Leaf dip assays Concentrations: 0, 0.5778, 1.4247, and 2.7308 mg a.i./L	Significant increase in hatch rate of eggs relative to controls.	Zeng and Wang 2010

Mites/Spiders	Exposure	Response	Reference
Tetranychus cinnabarinus (carmine spider mite, Acari: Tetranychidae) Adults	10% WP formulation (Jiangsu Wujiang Pesticide Ltd. Co., China). Leaf dip assays Concentrations: 0, 0.5778, 1.4247, and 2.7308 mg a.i./L	Small but statistically insignificant increase in egg production. No effect on longevity.	Zeng and Wang 2010
Greenhouse Studies			
Tetranychus urticae (red spider mite; Acari: Tetranychidae)	Confidor 200 SL (Bayer) Soil Drench: 100 mg/L Foliar Spray: Application rate unclear.	Reduced egg laying in some strains of mite. No overall or remarkable change in in fecundity. Egg viability not impacted. Working Note: Study motivated by concern that the use of imidacloprid may cause outbreaks in mite populations. See Szczepaniec et al. (2011) as well as Szczepaniec and Raupp (2013).	Ako et al. 2006
Eurytetranychus buxi (boxwood mite, Acari: Tetranychidae) feeding on boxwood	Marathon 60 WP 3,300 mg/L applied to soil.	Increase in fecundity in mites on treated plants but no effect on fecundity in mites directly sprayed with imidacloprid. No effect on longevity in mites exposed by either route (Fig. 1 of paper). Mechanism of enhancement not clear. See notes on separate field study below.	Szczepaniec and Raupp 2013
Field Studies			
Amblyseius victoriensis (mite; Acari: Phytoseiidae) 185 trees in imidacloprid-sprayed section of orchard; 185 trees in unsprayed section; 8 trees randomly selected from each section of analysis of leaves	Confidor 350 SC (5.25 g/100L or 0.0053% a.i.) Sprayed at label instructions at rate to control aphids (15 ml/100 L or 0.0053% a.i.).	Imidacloprid significantly reduced the population 4 weeks following application. The population recovered at 5-6 weeks following application, and was more than twice the size of the untreated control population (in another area of the orchard) by 9-12 weeks post-application	James 1997
Eurytetranychus buxi (boxwood mite, Acari: Tetranychidae) feeding on boxwood	Merit 75 WP Application rate specified as 2 g/0.3 m of shrub height in 1 liter of water.	Increase in abundance of mites by about 1 month after application.	Szczepaniec and Raupp 2013

Appendix 3: Terrestrial Invertebrates (*continued*)

Mites/Spiders	Exposure	Response	Reference
Tetranychus schoene (herbivorous mite; Acari: Tetranychidae) New York and Maryland	14,000 applications of imidacloprid between 2005 and 2007 for the control of Asian long-horned beetle (*Anoplophora glabripennis*)	Increase in abundance of mite on elm trees. Increase in mite fecundity. Increase in mite populations could be partially due to adverse effects on mite predators (coleopterans of the Coccinellidae and Chrysopidae families). This, however, is not directly demonstrated in this study. No remarkable impacts on Aphididae (aphids), Saproglyphidae (scavenger mites), Chrysopidae (green lacewings), Cecidomyiidae (predatory midges), Thripidae (thrips) and Coccinellidae (lady beetles in the genus Stethorus).	Szczepaniec et al. 2011

A3 Table 9: Other Arthropods

Arthropods, other	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
No studies encountered.			
Formulation			
Folsomia candida (springtail, Hexapoda; Collembola) Laboratory culture. 5 replicates plus control per dose.	Gaucho 600 Forest Service (600 g a.i./L, Bayer) Artificial soil assay 14-day exposure.	LC_{50}: 20.96 (9.51-32.13) mg/kg soil NOAEC:: 10 mg a.i./kg soil LOAEC (mortality): 100 mg a.i./kg soil.	Alves et al. 2014
Sublethal Effects			
Folsomia candida (springtail, Hexapoda; Collembola) Laboratory culture. 5 replicates plus control per dose.	Gaucho 600 Forest Service (600 g a.i./L, Bayer) Artificial soil assay 28-day exposure. Concentrations: 0.06, 0.12, 0.25, 0.5, and 1 mg/kg soil	Statistically significant decrease in number of juveniles at all concentrations. Not clearly dose-related at concentrations above 0.12 mg a.i./kg soil. LOAEC: 0.06 mg a.i./kg soil. NOAEC: not defined.	Alves et al. 2014
Porcellio scaber (sowbug, Isopoda: Porcellionidae) Wild caught Adults, 30-64 mg	Imidacloprid (99.8%) Contaminated leaves 14 days Nominal concentrations: 0 (n=36), 10 (n=42) and 25 (n=22) mg a.i./kg dry weight	No significant increase in mortality. NOAC (decrease in feeding rate): Not determined. LOAECC (decrease in feeding rate): 10 mg/kg food 0.24 mg/kg bw/day Note: No decrease in body weight gains even though food consumption was depressed. Increase in glutathione S-transferase activity at 25 mg/kg food.	Drobne et al. 2008
Porcellio scaber (sowbug, Isopoda: Porcellionidae) Wild caught Juveniles, 12.5 to 30 mg	Imidacloprid (99.8%) Ground leaves 14 days Nominal concentrations: 2.5, 5, 10, and 50 mg a.i./kg dry weight	No significant increase in mortality. NOAC (decrease in weight gain): 5 mg/kg food 0.3 mg/kg bw/day LOAECC (decrease in weight gain): 10 mg/kg food 0.5 mg/kg bw/day	Drobne et al. 2008
Mesocosm Studies			
No studies encountered.			
Field Studies			
No studies encountered.			

A3 Table 10: Earthworms

Earthworms	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Eisenia fetida 350 and 500 mg,	TGAI (95.3%) Soil 14-day exposures	7-day LC_{50}: 2.75 (2.94-7.98) mg/kg dry soil 14-day LC_{50}: 1.99 (1.67-2.49) mg/kg dry soil Working Note: Also did contact filter paper assay (not summarized here)	Chen et al. 2014b
Eisenia foetida 6-10 worms per concentration	TGAI (>95%) Direct exposure to liquid for up to 48 hours. Solution concentrations: 0.24, 0.48, 0.96, 2.00 mg/L	24-hour LC_{50}: 1.23 mg/L 48-hour LC_{50}: 0.77 mg/L	Luo et al 1999; Zhang et al. 2000
Eisenia foetida 6-10 worms per concentration	TGAI (>95%) Contact Filter Paper: acetone control, 0.004, 0.020, 0.100, 0.500 µg/cm^2	24-hour LC_{50}: 0.100 µg/cm^2 48-hour LC_{50}: 0.034 µg/cm^2	Luo et al 1999; Zhang et al. 2000
Eisenia foetida 6-10 worms per concentration	TGAI (>95%) Soil Concentrations: control, 1,2,4,8, 16 mg/kg dry soil	7-day LC_{50}: 3.48 mg/kg dry soil 14-day LC_{50}: 2.30 mg/kg dry soil	Luo et al 1999
Eisenia fetida, 10 per replicate, 3 replicates per concentration	TGAI (95.3%) Contact Filter paper Observation 48 hours	48-hour LC_{50}: 0.027 µg/cm^2	Wang et al. 2012 China
Eisenia fetida, 10 per replicate, 3 replicates per concentration	TGAI (95.3%) Soil Concentrations (other than range finding) not explicitly stated. Observations at 7 and 14 days	7-Day LC_{50}: 3.15 mg/kg dry soil 14-Day LC_{50}: 2.82 mg/kg dry soil Slopes given but nature of dose transformation (log_{10} or ln) not specified.	Wang et al. 2012 China
Eisenia fetida, 10 per replicate, 3 replicates per concentration	TGAI (97%, China) Artificial soil 14-day exposure Concentrations: 1.5 to 4.6 mg/kg soil.	14-day LC_{50}: 2.75 mg/kg dry soil Working Note: This study as well as Wang et al. 2015b is focused on application of concentration addition with ternary mixtures.	Wang et al. 2015a China

Earthworms	Exposure	Response	Reference
Formulation			
Allolobophora icterica	Confidor (200 g a.i./L) Soil Concentrations: 0, DMSO control, 0.1, 0.5, and 1 ppm 2 week observations	14-day LC_{50}: 2.81 (1.94-4.05) mg/kg dry soil Swelling along body surface after 2-7 days. Reversible after transfer to clean soil. Concentration-related weight loss.	Capowiez et al. 2005
Aporrectodea nocturna	Confidor (200 g a.i./L) Soil Concentrations: 0, DMSO control, 0.1, 0.5, and 1 ppm 2 week observations	14-day LC_{50}: 3.74 (3.41-4.08) mg/kg dry soil Swelling along body surface after 2-7 days. Reversible after transfer to clean soil. Concentration-related weight loss.	Capowiez et al. 2005
Dendrobaena octaedra	Merit Solupak (750 g a.i./kg, Bayer) Soil and leaf litter Observations at 35 days. Concentrations: 0, 1.4, 14, 140 and 1400 mg/kg soil.	35-day LC_{50} of 5.7mg/ kg soil 35-day LC_{10} of 2 mg/ kg soil LOAEL (weight loss): 3 mg/kg soil LOAEL (litter loss): 7 mg/kg soil	Kreutzweiser et al. 2008b
Eisenia andrei (tiger worm, European earthworm)	Gaucho 600 Forest Service (600 g a.i./L) Artificial soil 14 day exposure Concentrations: 6.25, 12.5, 25, 50, 100 mg/kg soil	14-day Toxicity values (mg a.i./kg soil LC_{50}: 25.53 (24.44-25.53) NOAEC: 12.50 LOAEC: 25	Alves et al. 2013
Eisenia fetida	Merit Solupak (750 g a.i./kg, Bayer) Soil and leaf litter Observations at 35 days. Concentrations: 0, 1.4, 14, 25 and 45mg/kg soil.	35-day LC_{50} of 25 mg kg NOAEC (mortality): 14 mg/kg soil	Kreutzweiser et al. 2008b
Pheretima group earthworms (*Amynthas hawayanus, A. aeroginosus and A. diffringens*) Note: these are prevalent in South Africa 10 worms per bucket, 5 buckets per concentration	Formulation (350 g a.i./L, NOS) Artificial soil Soil Concentrations: 0, 3.5, 5.25, 7.0, 8.75, 10.50 mg a.i./kg soil	**Days** / **LC_{50} (mg/kg soil)** 1 / 155 2 / 5 7 / 3	Mostert et al. 2000

Earthworms	Exposure	Response	Reference
Sublethal Effects			
Allolobophora icterica	Confidor (200 g a.i./L) 2-D terraria Concentrations: 0, solvent control, 0.5 and 1 mg/kg soil 7 day observations	Significant decrease in burrowing behavior (mean burrow length, distance, and burrow reuse (Fig. 1 of paper). Response of A. icterica significantly greater than that of *A. nocturna* in terms of burrow length only at 1 mg a.i./kg soil.	Capowiez et al. 2003
Allolobophora icterica	Confidor (200 g a.i./L without additives in DMSO) Soil Concentrations: 0, DMSO control, 0.5, and 1 mg a.i./kg dry soil	Burrowing activity greatly reduced with 24 hours. Clear dose-related response for area burrowed and maximum depth burrowed. See Table 2 of paper. LOAEL: 0.5 mg a.i./kg soil	Capowiez and Bérard 2006
Aporrectodea nocturna	Confidor (200 g a.i./L) 2-D terraria Concentrations: 0, solvent control, 0.5 and 1 mg/kg soil 7 day observations	Significant decrease in burrowing behavior (mean burrow length, distance, and burrow reuse (Fig. 1 of paper). Significant increase in total protein after 1 week. Dose-related but statistically significant only at 1 ppm.	Capowiez et al. 2003
Aporrectodea nocturna	Confidor (200 g a.i./L without additives in DMSO) Soil Concentrations: 0, DMSO control, 0.5, and 1 mg a.i./kg dry soil	Burrowing activity reduced with 24 hours, albeit not as severely a with *Allolobophora icterica*. Clear dose-related response for area burrowed and maximum depth burrowed. See Table 2 of paper. See Table 2 of paper. LOAEL: 0.5 mg a.i./kg soil	Capowiez and Bérard 2006
Aporrectodea caliginosa 25 worms/group	TGAI (99.9%) Soil 7 day exposure Concentrations: 0, 0.2, 0.66, 2 mg a.i./kg dry soil	NOAEC: 0.2 mg/kg dry soil 0.66 mg/kg dry soil: Slight (9%) but statistically significant decrease in relative body weight (relative to initial weights). 2.0 mg/kg dry soil: About 30% decrease in body weights (Table 1 of paper).	Dittbrenner et al. 2010

Earthworms	Exposure	Response	Reference
Aporrectodea caliginosa	Imidacloprid (NOS) Soil Concentrations: 0.2, 0.66, and 2 mg/kg dry soil Pre-exposure periods of 1, 7, 14 days, followed by sublethal assays	Short-term (24-96 hours) post-exposure (Table 1 of paper) 0.2 mg/kg soil: Decrease in burrow length after 24 but not 96 hours following a 1 day exposure. Increases in burrow depth with longer periods of pre-exposure. Clear NOAEC not determined. Longer-term observations (6 weeks after exposure): Dose-related decrease in macropore volume. Trend is significant ($p<0.01$) but the magnitude of the differences do not appear to be statistically significant based on author discussion.	Dittbrenner et al. 2011
Eisenia andrei (tiger worm, European earthworm)	Gaucho 600 Forest Service (600 g a.i./L) Artificial soil Avoidance assay 48 hour exposure Concentrations: 0.125, 0.25, 0.5. 1, 2 mg/kg soil	Toxicity values (mg a.i./kg soil AC_{50} (50% avoidance): 0.11 (confidence interval not determined) NOAEC: Not determined LOAEC: 0.13	Alves et al. 2013
Eisenia foetida 6 worms per concentration	TGAI (>95%) Soil 10 day exposure Concentrations: 0, 0.1, 0.2, and 0.5 mg/kg dry soil for 10 day	Dose-related increase in sperm deformity. Statistically significant increase in percentage of deformed sperm with respect to controls at 0.2 and 0.5 mg/kg dry soil. NOAEC = 0.1 mg/kg dry soil	Luo et al 1999
Eisenia fetida 300-600 mg	Imidacloprid (98.5%, Germany) Artificial soil Concentrations: 0, 0.2, 0.66, 2, and 4 mg/kg soil	0.2 mg/kg soil Transient increase in peroxidase on Day 7 but not Day 14. 0.66 mg/kg soil Significant and prolonged increases in superoxide dismutase, peroxidase, and cellulose. General increase in oxidative enzymes (i.e., oxidative stress) with increasing concentration up to 2 mg/kg soil. See Figure 1 of paper.	Zhang et al. 2014

Earthworms	Exposure	Response	Reference
Lumbricus terrestris 25 worms/group	TGAI (99.9%) Soil 7 day exposure Concentrations: 0, 0.2, 0.66, 2, 4 mg a.i./kg dry soil	NOAEC: 0.2 mg/kg dry soil 0.66 mg/kg dry soil: Slight (8.4%) but statistically significant *increase* in relative body weight (relative to initial weights) (hormesis?). 2.0 mg/kg dry soil: Slight (5.5%) but statistically significant decrease in relative body weight (relative to initial weights). More sever weight loss at higher concentrations (Table 2 of paper). Cast production was increased at 0.2 mg/kg dry soil (hormesis?) but significantly decreased at higher concentrations. Working Note: Cast production may be viewed as surrogate for activity level in earthworms.	Dittbrenner et al. 2010
Lumbricus terrestris 3.35 ± 0.85 g Collected	Imidacloprid (NOS) Soil Concentrations: 0.2, 0.66, 2, and 4 mg/kg dry soil Pre-exposure periods of 1, 7, 14 days, followed by sublethal assays	Short-term (24-96 hours) post-exposure (Table 2 of paper) No effect with only 1 day pre-exposures at any concentration. 2 mg/kg soil: Increases in burrow depth and length with 7-day pre-exposure at 24 but not 48 hours after observation. Longer-term observations (6 weeks after exposure): Dose-related decrease in macropore volume. Trend is significant ($p<0.01$) but the magnitude of the differences do not appear to be statistically significant based on author discussion.	Dittbrenner et al. 2011

Earthworms	Exposure	Response	Reference
Chronic Exposures			
Eisenia andrei (tiger worm, European earthworm)	Gaucho 600 Forest Service (600 g a.i./L) Artificial soil 56 day exposure Concentrations: 0.75, 1.25, 2.50, 5, 10, 20 mg/kg soil	Toxicity values (mg a.i./kg soil EC_{50} (reproduction): 4.07 (2.42-5.72) NOAEC: Not determined LOAEC: 0.75 Decrease in body weights. Dose-response for decreases in number juveniles given in Fig. 1 of paper. Biomass loss was less than control group at 0.75 mg a.i./kg soil (hormesis?).	Alves et al. 2013
Mesocosm Studies			
Composting simulations (cattle manure) with *Eisenia fetida*	Imidacloprid (99%) Concentrations: 0, 2, 4, and 8 mg/kg (wet weight) Observation period up to 49 days.	LOAEL: 2 mg/kg media No mortality but impaired reproduction. Clear but not dose-related decrease in worm populations relative to controls by week 15 (Fig. 3 of paper) `Working Note: This appears to be a well-conducted study but the exposure media is not relevant to the current risk assessment.`	Fernandez-Gomez et al. 2011
Sugar maple litter with 2 earthworms, *Dendrobaena octaedra*	Imidacloprid (NOS) 35 day exposure Low Field Rate: Two leaves added to microcosm. Conc. in leaves of 3.2 (1.4-5.4, range) mg/kg.	No mortality and no effect on cocoon production. Weight loss by Day 35 but not statistically significant (Fig. 2 of paper). Dose-related and statistically significant decrease in leaf loss attributable to earthworms.	Kreutzweiser et al. 2008a
Sugar maple litter with 2 earthworms, *Dendrobaena octaedra*	Imidacloprid (NOS) 35 day exposure Imidacloprid (NOS) High Field Rate: 2 contaminated fallen leaves added to system. Conc. in leaves of 11 (6.4-18.5, range) mg/kg.	No mortality and no effect on cocoon production. Weight loss by Day 35 but not statistically significant (Fig. 2 of paper). Dose-related and statistically significant decrease in leaf loss attributable to earthworms.	Kreutzweiser et al. 2008a
Sugar maple litter with 2 earthworms, *Dendrobaena octaedra*	Imidacloprid (NOS) 35 day exposure Imidacloprid (NOS) Overdose Field Rate: (NOD): Two leaves added to microcosm. Conc. in leaves of 132 (86.6-188, range) mg/kg.	No mortality. Weight loss apparent by Day 14 and statistically significant by Day 28. Dose-related and statistically significant decrease in leaf loss attributable to earthworms.	Kreutzweiser et al. 2008a

Earthworms	Exposure	Response	Reference
Sugar maple litter with 2 earthworms, *Dendrobaena octaedra* 5 replicates	EcoPrid (experimental EC formulation, 50 mg a.i./mL) [based on reference to 2007 paper] Low-dose 0.125g/cm DBH; Leaf residue: 18.0 µg/g High-dose 0.25g/cm DBH; Leaf residue: 122.9 µg/g Observations up to 35 days.	Mortality 3/10 at low dose 1/10 at high dose Significant decrease in mass loss of leaf material (Fig. 2 of paper). Greater loss of uncontaminated leaf material relative to contaminated leaf material but significant only in the high-dose group.	Kreutzweiser et al. 2009
Field Studies			
Kentucky bluegrass in Kentucky with 5 replicate 2 x 2 m plots per formulation or untreated control	Merit 75 WP Application rates: 0.34 and 0.45 kg a.i./ha (≈0.3 and 0.4 lb a.i./acre). Observations 9 and 40 days after application. Fall and spring applications.	A temporary suppression in earthworm abundance in fall (40-50%) in fall but not spring. Earthworm abundance was no different than that of controls by the second sampling date (day 40 or 36 for fall and spring, respectively).	Kunkel et al. 1999
Kentucky bluegrass in Kentucky with 5 replicate 2 x 2 m plots per formulation or untreated control	Merit 0.5% granular Application rates: 0.34 kg a.i./ha (≈0.3 a.i./acre). Observations 9 and 40 days after application. Fall and spring applications.	A temporary suppression in earthworm abundance in fall (40-50%). Significant reduction in earthworm abundance in spring application. Earthworm abundance was no different than that of controls by the second sampling date (day 40 or 36 for fall and spring, respectively).	Kunkel et al. 1999

A3 Table 11: Other Invertebrates

Invertebrates, Other	Exposure	Response	Reference
Acute Lethality			
Technical Grade			
Steinernema carpocapsae (entomopathogenic nematode; Phylum Nematoda) 2 ml of suspension (150 infective juveniles per ml) per concentration for mortality test, 0.05 ml suspension (180 - 200 infective juveniles per ml) for infectivity test	TGAI (90%) Mortality assay: 48-hour exposure to 0, 10 and 100 µg/ml in solution. Infectivity, development and reproduction assay:100 µg/ml in solution for 24 hours	Mortality assay: No significant mortality in comparison with controls at any concentration tested. Nematodes treated with 100 µg/ml imidacloprid were no different than untreated controls in their ability to kill newly molted last instar cutworms (S. litura) in 3 trials conducted with 10 cutworms each treatment/control group.	Zhang et al. 1994
Helix aspersa (garden snail; Gastropoda: Helicidae) Wild caught 1.56 ±0.24 g 6 replicates, 10 per dose per replicate	TGAI (98.7%, Bayer, Germany) Doses: 25, 50, 100, 150, 200, 250, and 300 mg/snail Mortality assayed at 48 hours. Topically applied inside shell cavity.	48h-LD_{50}: 109.2 (97.85-121.90) µg/snail Working Note: Taking the average bw of 0.00156 kg the LD_{50} is approximately 70 (62.7-78.1) mg a.i./kg bw. This dose is somewhat lower than mammalian oral LD_{50} values of about 130-424 mg/kg bw. See sublethal phase of study below.	Radwan and Mohamed 2013
Formulation			
Heterorhabditis bacteriophora (entomopathogenic nematode; Phylum Nematoda)	Imidacloprid (NOS) [appears to have used formulation but this is not clear] Mortality and infectivity assays Concentrations: 0, 10, 40 or 160 mg a.i./l. 40 mg a.i. /L.	Imidacloprid did not affect nematode mortality with respect to controls. In addition, imidacloprid did not adversely impact the infective ability of nematodes (penetration of wax larvae of target moths) with respect to unexposed controls. In a separate greenhouse tests, imidacloprid was shown to act synergistically with the nematode in controlling white grubs in turfgrass.	Koppenhofer and Kaya 1998

Invertebrates, Other	Exposure	Response	Reference
Sublethal Effects			
Helix aspersa (garden snail; Gastropoda: Helicidae) Wild caught 1.56 ±0.24 g 6 replicates, 10 per dose per replicate	TGAI (98.7%, Bayer, Germany) Doses specified as 0.2 and 0.6 of acute LD_{50}. See entry above. Approximate doses: 22 and 66 mg/snail Topically applied inside shell cavity.	Dose-related decreases in AChE activity at 1, 3, and 7 days after dosing. Statistically significant at all doses and observation periods (Fig. 1 of paper). Dose-related increases in catalase activity at 1, 3, and 7 days after dosing. Significant at all observation periods for high dose. At low dose, significant only on Day 1. (oxidative stress) Significant decreases in glycogen levels as high dose (all observation periods) and low dose (Day 7 only). No pronounced dose-response relationship (Table 2 of paper). Significant and dose-related decreases in total lipids at both doses and all observation periods (Table 3 of paper). Significant increase in total protein only at high dose (Table 4 of paper)	Radwan and Mohamed 2013
Mesocosm Studies			
No studies encountered.			
Field Studies			
No studies encountered.			

A3 Table 12: Multispecies Mesocosm and Field Studies

Mesocosm/Field	Exposure	Response	Reference
30 eastern hemlocks in Tennessee. Treated for control of HWA	Soil injection, soil drench, and tree injection. Survey of 293 species, 226 genera, 75 families, and 9 orders which are associated with eastern hemlocks.	Soil Drench: Significant decrease in invertebrate abundance. Soil Injection and Tree Injection: Decrease in abundance relative to controls but not to horticultural oil. Across all groups examined, soil drench had a greater impact than other application methods. Soil injection impacted hematophaga (blood sucking) and transient phytophaga (plant eating) guilds. Tree injection: Impacted only transient phytophaga guild. Working Note: Complex paper but basic results seem intuitive. Effects on phytophagous insects will occur on hemlocks with any effective treatment with imidacloprid.	Dilling et al. 2009
Cotton fields for the control of various pests including *Helicoverpa zea* (bollworm, Lepidoptera: Noctuidae). Small plots, single or multiple (up to 3) applications.	Trimax 4F (Bayer) 0.05 kg a.i./ha (≈0.045 lb a.i./acre)	Single Applications: No significant impact on arachnids, fire ants (Hymenoptera), or big-eyed bugs (Hemiptera). Significant but not substantial decrease in insidious flower bugs (Hemiptera) (Fig. 1a). Multiple Applications: No significant impact on arachnids or big-eyed bugs. Decreases in fire ants insidious flower bugs (Fig. 1b). No significant impact on predators of bollworm larvae (Fig. 3) or viability of bollworm eggs.	Kilpatrick et al. 2005
Kentucky bluegrass in Kentucky with 5 replicate 2 x 2 m plots per formulation or untreated control	Merit 75 WP Application rates: 0.34 and 0.45 kg a.i./ha (≈0.3 and 0.4 lb a.i./acre). Observations 9 and 40 days after application. Fall and spring applications.	No effect of imidacloprid treatment on the abundance of soil micro-arthropods (Collembola, Mesostigmatid and Orbatid mites).	Kunkel et al. 1999

Mesocosm/Field	Exposure	Response	Reference
Kentucky bluegrass in Kentucky with 5 replicate 2 x 2 m plots per formulation or untreated control	Merit 0.5% granular Application rates: 0.34 kg a.i./ha (≈0.3 a.i./acre). Observations 9 and 40 days after application. Fall and spring applications.	No effect of imidacloprid treatment on the abundance of soil micro-arthropods (Collembola, Mesostigmatid and Orbatid mites).	Kunkel et al. 1999
Golf course in Kentucky	Merit 0.5G 0.336 kg a.i./ha (≈0.3 lb a.i./acre) by drop spreader, followed by 1.5 cm irrigation Applications on two different year (1996 and 1997)	There was no difference in pre-treatment counts for any group of predatory arthropods and scarabaeid grubs in either year. The abundance of beneficial predators (ants, carabids, spiders, and staphylinids) essentially was not impacted in either year. There was no difference between controls and imidacloprid-treated plots with respect to scavenging of black cutworm eggs or Japanese beetle eggs.	Kunkel et al. 1999
Orchard, Australia	Imidacloprid (350 SC, NOS) Applied to stone fruit as a 0.0053% a.i. or 53 ppm a.i. solution by air blast. Application rate in units of mass/area not specified.	Clear adverse effects on some coleopterans (particularly some ladybird beetle larvae). Populations of *Stethorus vagans* (Australian predatory ladybug) greatly reduced. Not all coleopterans impacted. No substantial adverse effect on *Dicranolaius bellulus* (Australian red and blue beetle). No remarkable impact on spiders or wasps (Hymenoptera).	James and Vogele 2001
Turf, 1.3 ha, New York 10 m x 10 m experimental plots separated by 10 m border. Several pesticides assayed.	Merit 0.5 G Drop spreader applications. Once per year for 3 years at 0.37 kg/ha (≈0.33 lb a.i./acre)	Decrease in populations of all hexapods (factor of 2.2), Collembola (factor of 2.6), Coleoptera adults (factor of 2.4), and Thysanoptera [thrips] (factor of 2.4) with respect to controls. See Figure 2 of paper. Some indication of recovery over time, particularly in Collembola and Thysanoptera [thrips] (See Figure 3). No substantial effects on mites, Hemiptera (mostly mealybugs), Hymenoptera (mostly ants), Diptera (mostly larvae), Coleoptera larvae.	Peck 2009

Mesocosm/Field	Exposure	Response	Reference
Mesocosm (small cups) with wild strawberry (*Fragaria vesca*), wood crickets (*Nemobius sylvestris*), and nursery web spider (*Pisaura mirabilis*)	Confidore WG 70 Two treatment levels: 1 mg/microcosm. Corresponds to 0.24 g/m^2 or ≈2.14 lb a.i./acre. 10 mg/microcosm. Corresponds to 2.39 g/m^2 or ≈21.32 lb a.i./acre.	Crickets: Dose related decrease in cricket feeding and growth. Statistically significant decrease in feeding and growth only at higher treatment level (Figure 1 of paper). Higher survival in response to spider predation at low treatment relative to controls but this was only marginally significant (p=0.045). Spiders: No statistically significant effects on behavior. At tendency toward increased activity but this was not statistically significant.	Uhl et al. 2015

Appendix 4: Toxicity to fish.

Notes:

Values in parentheses are 95% confidence limits unless otherwise specified.

The ecological risk assessments from EPA are cited frequently. U.S. EPA/OPP/EFED (2007a) is abbreviated to EFED (2007a). U.S. EPA/OPP/EFED (2008a) is abbreviated to EFED (2008a).

A4 Table 1: Acute Toxicity

Species	Exposure	Response	Reference
Bluegill (*Lepomis macrochirus*) Mean length 27mm, mean weight 0.46 g 10 per concentration	Technical grade imidacloprid (97.4%) Solvent: dimethylformamide Nominal Concentrations: 0, 0, (solvent control), 16, 27, 45, 75 and 125 mg a.i./L Mean measured concentrations of 0, 0 (solvent control), 14, 25, 42, 68 and 105 mg a.i./L	96-hour LC_{50} > 105 mg/L (greater than the limit of solubility) 96-hour NOAEC = 25 mg/L 42 mg/L and higher: mortality, dark discoloration, fish on the bottom of test chamber, erratic swimming, surfacing, quiescence, rapid fin movement, labored respiration. A surface film and precipitate on the bottom were noted at these concentrations.	Bowman and Bucksath 1990a MRID 42055314 Core
Rainbow Trout (*Oncorhynchus mykiss*) Mean length 44 mm, mean weight 1.07 g. 10 per concentration	Technical grade imidacloprid (97.4%) Solvent: dimethylformamide Nominal Concentrations: 0, 0, (solvent control), 16, 27, 45, 75 and 125 mg a.i./L Mean measured concentrations of 0, 0 (solvent control), 15, 27, 42, 64 and 83 mg/L	96-hour LC50 > 83 mg/L (greater than the limit of solubility) 96-hour NOAEC = 42 mg/L 64 mg/L and higher: mortality, dark discoloration, fish on the bottom of test chamber, erratic swimming, and quiescence. A surface film and precipitate on the bottom were noted at concentrations at and above 42 mg/L.	Bowman and Bucksath 1990b MRID 42055315 Core
Rainbow Trout (*Oncorhynchus mykiss*) Mean length 53 mm, mean weight 1.3 g 10 per concentration.	Technical grade imidacloprid (95.3%) Nominal Concentrations: 0, 50, 89, 158, 281, 500 mg a.i./L Mean measured concentrations of greater than 80% nominal.	96-hour LC_{50} = 211 (158 – 281) mg a.i./L 96-hour NOAEC = 50 mg a.i./L 89 mg/L and higher: apathy, irregular swimming behavior, lying on side/back, staggering 281 mg/L and higher: mortality	Grau 1988a MRID 42055316 Not cited in EFED 2007a. Cited but not discussed in EFED 2008a.

Appendix 4: Toxicity to fish *(continued)*

Species	Exposure	Response	Reference
Sheepshead Minnow (*Cyprinodon variegatus*) Young adult, mean length 29 mm, mean weight 0.77 g, 10 per concentration	Technical grade imidacloprid (96.2%) Mean measured concentrations: Control, solvent control, 22.4, 35.2, 58.2, 105 and 195 mg a.i./L	96-hour LC_{50} = 163 (58.2-∞) mg a.i./L NOAEC = 58.2 mg a.i./L on the basis of mortality and signs (lethargy, dark coloration) at higher concentrations.	Ward 1990a MRID 42055318
Zebra fish (*Danio rerio*) Embryos	Imidacloprid (NOS but clearly a technical grade) 96-hour Concentrations: 0, 200; 215; 260; 280; and 300 mg/L	96-hour LC_{50}: 241 (224-257) mg a.i./L 96-hour LC_{10}: 201 mg a.i./L	Tisler et al. 2009 Slovenia
Zebra fish (*Danio rerio*) Embryos	Confidor 200 SL (Bayer) 96-hour	96-hour LC_{50}: 214 (202-230) mg a.i./L 96-hour LC_{10}: 194 mg a.i./L	Tisler et al. 2009 Slovenia
Zebra fish (*Danio rerio*) Eggs in 40 mL petri dishes. 10 embryos per dish, 4 dishes per level.	Imidacloprid (Sigma-Aldrich, Germany) Nominal concentrations at 26°C: 1, 5, 10, 15, 20, 30, 40, and 50 mg/L. Nominal concentrations at 28°C: 5, 15, and 30mg/L. Nominal concentrations at 30°C and 33°C: 5, 10, 25 and 25mg/L. [Note: Repeating the two 25 mg/L concentrations may be a typo in the publication.] No solvent. 96-hour observation/ exposure period.	No effects at any concentrations or temperatures. Author discussion: …*the absence of detrimental effects might also be due to the fact that the embryos remained in the egg for approximately three quarters of the test duration and were protected by the chorion.* `Working Note: Above discussion does not address the factor that adverse effects were seen with nickel chloride.`	Scheil and Kohler 2009

A4 Table 2: Chronic toxicity

Species	Exposure	Response	Reference
Rainbow Trout (*Oncorhynchus mykiss*) Newly fertilized eggs <4 hours old, 4 replicates of 35 eggs each per concentration, plus an additional 50 eggs per each of the 4 control replicates (egg viability determination)	Technical grade imidacloprid (95%) 98-Day flow-through (early-life stage) Nominal concentrations: 0, 1.3, 2.5, 5.0, 10 and 20 mg a.i./L. Mean measured concentrations: 0, 1.2, 2.3, 4.9, 9.8 and 19 mg/L	**Original conclusions:** NOAEC = 9.8 mg/L LOAEC = 19 mg/L (statistically significant reduction in length at 36 and 60 days post-hatch, and body weight at 60 days post-hatch). No statistically significant biologically important effects on egg viability, hatch, survival or behavioral variables were observed. <u>1992 re-evaluation</u> Day 36 growth was most sensitive endpoint. Based on re-evaluation of this endpoint: NOAEC = 1.2 mg a.i./L LOAEC = 2.3 mg a.i./L EFED 2008a (p. 17) and EFED 2007a (p. 41) uses the re-evaluation from 1992.	Cohle and Bucksath 1991 MRID 42055320 Gagliano 1992 MRID 42466501 Supplemental

A4 Table 3: Field Studies

Species	Exposure	Response	Reference
Japanese medaka (*Oryzias latipes*) 4 rice paddy mesocosms, 2 treated and 2 control. 10 male and 10 female fish per paddy.	Admire GR (1% a.i.) Rice mesocosms (5.2 m x 1.6 m), 4 cm deep planted with rice seedlings. Application rate: 215 g a.i./ha (≈0.2 lb a.i./acre). Observation period up to 118 days. Working Note: Admire GR appears to be a Japanese formulation. Could not identify a label for a 1% granular Admire formulation.	Water concentrations diminished rapidly from 239.2 µg/L (0.1 days after application) to 1.1 µg/L by Day 118. Calculated half-life of 4 days due to dissipation and not degradation. See Table 2 of paper. 2/40 fish dead in first 2 days with concentrations of >30 µg/L. No mortality in controls. [Working Note: This mortality is not significant based on one-tailed Fisher Exact test, p=0.246835] Subsequent mortality attributed to predation by herons. No malformations in "*abundant*" (NOS) offspring. Increase in rates of microbial ciliate parasite (*Trichodina domerguei*) infestations in imidacloprid exposed group. Authors indicate difference is significant. (See Table 3. Based on these data, this effect is significant, p=0.0005). Authors suggest immune suppression. Increase in blood lactate – i.e., anaerobic metabolism, stress response – possibly due to gill damage.	Sanchez-Bayo and Goka 2005 Japanese study

Appendix 5: Toxicity to amphibians.

Values in parentheses are 95% confidence limits unless otherwise specified.
The ecological risk assessments from EPA are cited as follows.
U.S. EPA/OPP/EFED (2007a) is abbreviated to EFED (2007a).
U.S. EPA/OPP/EFED (2008a) is abbreviated to EFED (2008a).
The designation of amphibian stages corresponds to the Gosner (1960) system (e.g., http://froglet.us/Development/gosner_stages.html)

A5 Table 1: Acute Toxicity

<table>
<tr><th>Species</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Rana limnocharis
Tadpoles,
10 per concentration,
3 replicates per concentration</td><td>Technical grade (>95%)
96-hour acute exposure
Concentrations: 16.7, 30, 54, 97.2, 174.9, 314.9, and 556.8 mg a.i./L</td><td><table><tr><th>Time (hours)</th><th>LC_{50} (mg/L)</th></tr><tr><td>24</td><td>235</td></tr><tr><td>48</td><td>165</td></tr><tr><td>72</td><td>116</td></tr><tr><td>96</td><td>82</td></tr></table>See Table 1 of paper for confidence intervals.

96-hours
NOAEC (mortality) = 16.7 mg/L
LOAEC = 30 mg/L (1/10 died)</td><td>Feng et al. 2004

China</td></tr>
<tr><td>Rana nigromaculata Hallowell
Tadpoles,
10 per concentration,
3 replicates per concentration</td><td>Technical grade (>95%)
96-hour acute exposure
Concentrations: 30, 45, 67.5, 101.2, 151.8, 227.8, and 341.7 mg a.i./L</td><td><table><tr><th>Time (hours)</th><th>LC_{50} (mg/L)</th></tr><tr><td>24</td><td>268</td></tr><tr><td>48</td><td>219</td></tr><tr><td>72</td><td>177</td></tr><tr><td>96</td><td>129</td></tr></table>See Table 2 of paper for confidence intervals.

96-hours
LOAEC = 45 mg/L (mortality 1/10 vs 0/10 in controls)</td><td>Feng et al. 2004

China</td></tr>
<tr><td>Rana hallowell
Tadpoles`</td><td><u>Micronucleus assay</u>
As above but concentrations of 2, 8, and 32 mg a.i./L.
7-day in vivo exposure of tadpoles</td><td>Significant increase in incidence of micronuclei at 8 and 32 mg/L (Table 3 of paper).</td><td>Feng et al. 2004

China</td></tr>
</table>

Appendix 5: Toxicity to Amphibians *(continued)*

<table>
<tr><th>Species</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Rana hallowell
Tadpole erythrocytes</td><td><u>Comet Assay</u>
As above but concentrations of 0.05, 0.1, 0.2, and 0.5 mg a.i./L.
Comet Assay, 1-hour exposure for blood cells.</td><td>Dose-related and significant (p<0.01) increase in DNA damage (Comet assay) scores at all concentrations.

See Table 4 in publication.</td><td>Feng et al. 2004

China</td></tr>
<tr><td>Xenopus laevis (African clawed frog)
Embryos (FETAX assay)
10 eggs/concentration Stages 8-11</td><td>Imidacloprid (NOS)
96-hours at 24°C or until control eggs reached Stage 46 (pre-adult).

Working Note: The source, purity, and type (a.i. vs formulation) are not indicated.</td><td>LC_{50}: 17.4 (14.6-20.6) mg a.i./L
EC_{50}: 10 mg/L
Note: EC_{50} is for malformations.
Toxicity values given on p. 51 but doses used are not specified. Malformations specified as inhibited egg development, a failure of the mouth to develop, and lack of pigment in the eye (p. 57 of paper).</td><td>Channing 1998

South Africa</td></tr>
<tr><td>Bufo americanus (American Toad), larvae</td><td>Imidacloprid (Merit, 75% a.i. powder)</td><td>48-hour LC_{50}: 468.0 mg/L
Days to metamorphosis:
4.68 mg a.i./L: No effect
46.8 mg a.i./L: Slight but significant increase (≈4.5 %)</td><td>Howard et al. 2003.
Julian 2000</td></tr>
<tr><td>Pseudacris triseriata (Western chorus frog), larvae</td><td>Imidacloprid (Merit, 75% a.i. powder)</td><td>48-hour LC_{50}: 388.5 mg/L
Days to metamorphosis:
3.89 mg a.i./L: No effect
39.9 mg a.i./L: Slight but significant increase (≈1.6 %)
Increase in deformities at high concentration but not significant based on analysis of variance (p.33).</td><td>Howard et al. 2003
Julian 2000</td></tr>
<tr><td>Rana berlandieri (Rio Grande leopard frog), larvae</td><td>Imidacloprid (Merit, 75% a.i. powder)</td><td>48-hour LC_{50}: 184.5 mg a.i./L</td><td>Howard et al. 2003.
Julian 2000</td></tr>
<tr><td>Hypsiboas pulchellus (Montevideo tree frog)
Tadpoles, Stage 36 at start of study.</td><td>Glacoxan Imida (35% a.i.). Formulation from Punch Química S.A.,Argentina
96-hour exposures
Concentrations: 25, 37.5, 50, 75, 100, and 124.5 mg a.i./L)</td><td>LC_{50} values
<table><tr><th>Time (hours)</th><th>LC_{50} (mg/L)</th></tr><tr><td>24</td><td>69.4</td></tr><tr><td>48</td><td>58.2</td></tr><tr><td>72</td><td>56.8</td></tr><tr><td>96</td><td>52.6</td></tr></table></td><td>Perez-Iglesias et al. 2014

Argentina</td></tr>
</table>

Appendix 5: Toxicity to Amphibians *(continued)*

Species	Exposure	Response	Reference
Hypsiboas pulchellus (Montevideo tree frog) Tadpoles, Stage 36 at start of study.	Glacoxan Imida (35% a.i.). Formulation from Punch Química S.A.,Argentina Concentrations of 12.5, 25, 37.5 mg/L for assay of sublethal effects.	<u>Micronuclei assay (Table 1 of paper)</u> Increase in incidence of micronuclei at 25 mg a.i./L at 96 hours but no effect as lower (12.5 mg a.i./L) or higher (37.5 mg a.i./l) concentrations at 96 hours. <u>Comet Assay (Table 2 of paper).</u> 12.5 mg/L: No effect on rate of damaged cells but a transient (48 but not 96 hours) in index of genetic damage. 25 mg/L: Increase in incidence of cell damage at both 48 and 96 hours. Transient (48 but not 96 hours) in index of genetic damage. 37.5 mg/L: Increase in incidence of cell damage and genetic damage at both 48 and 96 hours. Working Note: Index of genetic damage based on sister-chromatid exchanges, micronuclei, and the Comet assay (adopted from Pitarque et al. 1999).	Perez-Iglesias et al. 2014 Argentina

Appendix 6: Toxicity to aquatic invertebrates

Notes:

Values in parentheses are 95% confidence limits unless otherwise specified.

The ecological risk assessments from EPA are cited as:

U.S. EPA/OPP/EFED (2007a) is abbreviated to EFED (2007a),

U.S. EPA/OPP/EFED (2008a) is abbreviated to EFED (2008a).

Unless otherwise specified, reported slopes do not specify whether a natural or common log transformation was used.

All values are reported in mg a.i./L unless otherwise specified.

The term "Mean Conc." is used as an abbreviations for "Mean Measured Concentration".

All responses are expressed as mg/L regardless of units used in study. The original units as reported are maintained in the exposure column.

A6 Table 1: Daphnids and other Cladocera, Acute Toxicity

Note: Several studies in this table cite OECD guidelines. The OECD (2004) guidelines call for temperatures of 18-22 °C. These recommendations have been consistent for many years.

Species	Exposure	Response	Reference
Standard LC_{50}s			
Technical Grade			
Daphnia magna, <24 hours old 4 replicates, 10 organism per replicate	Imidacloprid (99%, Sigma Aldrich) Concentrations: 0, 0.40, 1.20, 3.70, 11.1, 33.3, 100 mg/L 20 ±1 °C Static renewal 7-days	Day 3: Mortality only at 100 mg/L. Day 7: 100% mortality at all concentrations No EC_{50} calculated.	Agatz and Brown 2013b U.K.
Daphnia magna, <24 hours old	Imidacloprid (Bayer, purity/nature not specified) Five conc. (NOS) from 60 to 125 mg/L.	48-hour LC_{50} 97 mg/L Slope: 1.5	Loureiro et al. 2010 Portugal

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
Daphnia magna	Imidacloprid (99.5%) 22±1 °C.	48-hour EC_{50} 11.822 (0.464-301.256) mg a.i./L Normal photoperiod	Sanchez-Bayo and Goka 2006a Japan
Daphnia magna 4 replicates per concentration 10 animals each species per replicate	Imidacloprid (>95% purity) 48-hours, static 20 °C	48-hour LC_{50} 10.44 (6.97 - 17.71) mg/L Slope: 0.91	Song et al 1997
Daphnia magna, <24 hours old Numbers not clearly specified but appears to be 20 per concentration.	Imidacloprid (NOS but clearly a technical grade) 48-hours 21±1 °C Concentrations: 0, 10, 40, 70, 100, 130 mg L/1	24-Hour EC_{50}: 97.9 (81.4-127.7) mg/L 48-Hour EC_{50}: 56.6 (34.4-77.2) mg/L Working Note: Compare to companion study on Confidor below. Confidor somewhat more toxic.	Tisler et al. 2009 Slovenia
Daphnia magna 2 flasks per concentration 10/flask	Imidacloprid (95.9% purity) 48-hours, static Mean Conc: 0, 15, 25, 42, 71 and 113 mg/L	48-hour LC_{50} 85 (71 - 113) mg/L NOAEC: 42 mg/L (immobility)	Young and Hicks 1990 MRID 42055317
Formulation			
	***Ceriodaphnia* sp.**		
Ceriodaphnia dubia 4 replicates, 10 animals per replicate	Admire Pro (Bayer) Imidacloprid (99.5%) 48-hour static Temperature N.S. Nominal conc.: 0.5, 1, 2, 4, 10, and 20 µg/L.	LC_{50}: 0.00207 (0.00114-0.0034) mg a.i./L Slope: 0.78 (log_{10}) LC_{50} based on measured not nominal concentrations. Authors determined mortality based on microscopic examination for heart rate.	Chen et al. 2010 U.S.
Ceriodaphnia dubia, <24 hours old 4 replicates, 5 organisms per replicate Mean body length of 0.34±0.06 mm (see Table 3 of paper).	Admire Flowable, imidacloprid/water and surfactant (20:80, v/v)], BASF Japan Ltd. 48-hours static renewal 22 ± 1°C Concentrations: 390.63 to 6250 [5 levels]	48-hour EC_{50}: 0.57162 (0.2896 to 0.8412) mg a.i./L Working Note: This assay is *C. dubia*. The study below in the same paper is *C. reticulata*.	Hayasaka et al. 2012b
Ceriodaphnia reticulata, <24 hours old 4 replicates, 5 organisms per replicate Mean body length of 0.37±0.07 mm (see Table 3 of paper).	Admire Flowable, imidacloprid/water and surfactant (20:80, v/v)], BASF Japan Ltd. 48-hours static renewal 22 ± 1°C Concentrations: 781.25 to 50000 µg/L [7 levels]	48-hour EC_{50}: 5.5529 (4.2133 to 7.3878) mg a.i./L	Hayasaka et al. 2012b

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
	Daphnia magna		
Daphnia magna	Confidor 200 SL Few experimental details.	48-hour EC_{50}: 84 mg a.i./L	Daam et al. 2013
Daphnia magna, neonates, <24 hrs 20 per concentration Neonates from ephippia (Daphtoxkit F test kit).	Confidor (Bayer Hellas AG). Composition of formulation not specified. 20 °C Concentrations: 7.81, 15.6, 31.3, 62.5 and 125 mg/L	48-hour LC_{50} 64.6 (43.3–122.5) mg/L	Kungolos et al. 2009 Greece
Daphnia magna, neonates, <24 hrs 10 per concentration	Confidor 200 SL (Bayer) OECD Guidelines Concentrations: 0, 25, 50, 75, 100, 125, 150, 175 and 200 mg/L	48-hour EC_{50} (no predator cues) 96.5 (87.83-105.6) mg/L 48-hour EC_{50} (with predator cues) 90.68 (82.04-99.03) mg/L	Pestana et al. 2010 Portugal/Canada
Daphnia magna, <24 hours old Numbers not clearly specified but appears to be 20 per concentration.	Confidor 200 SL (Bayer) 48-hours 21±1 °C Concentrations: 0, 10, 40, 70, 100, 130 mg a.i./L.	24-Hour EC_{50}: 38 (32-48) mg/L 48-Hour EC_{50}: 30 (28-44) mg/L `Working Note: Compare to companion study on TGAI above. Confidor somewhat more toxic.`	Tisler et al. 2009 Slovenia
Daphnia magna, <24 hours old 4 replicates, 5 organisms per replicate	Admire Flowable, imidacloprid/water and surfactant (20:80, v/v)], BASF Japan Ltd. 48-hours static renewal 22 ± 1°C Concentrations: 12500 to 400000 µg/L [6 levels]	48-hour EC_{50}: 43.265 (34.302 to 53.592) mg a.i./L	Hayasaka et al. 2012b
Daphnia pulex, <24 hours old 4 replicates, 5 organisms per replicate	Admire Flowable, imidacloprid/water and surfactant (20:80, v/v)], BASF Japan Ltd. 48-hours static renewal 22 ± 1°C Concentrations: 6250 to 200000 µg/L [6 levels]	48-hour EC_{50}: 36.872 (28.399 to 48.106) mg a.i./L	Hayasaka et al. 2012b

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
	Moina macrocopa	Note: Very small cladoceran.	
Moina macrocopa, <24 hours old 4 replicates, 5 organisms per replicate	Admire Flowable, imidacloprid/water and surfactant (20:80, v/v)], BASF Japan Ltd. 48-hours static renewal 22 ± 1°C Concentrations: 6250 to 20000 µg/L [6 levels]	48-hour EC_{50}: 45.271 (34.378 to 62.218) mg a.i./L	Hayasaka et al. 2012b
Sublethal Effects			
Daphnia magna, neonates, <24 hours old	Imidacloprid (99%, Sigma Aldrich) 24-hour 20 ±1 °C Concentrations: 0, 0.078, 1.56, 7.8, 31.2, 156mg/L	Inhibition of Feeding 24-hr EC_{50}: 1.83 mg/L 24-hr EC_{05}: 0.19 mg/L 24-hr EC_{95}: 8.7 mg/L Approximately 50% inhibition of feeding at 1.56 mg/L (Fig. 1 of paper). No inhibition at 0.078 mg/L. No feeding at two higher concentrations.	Agatz and Brown 2013b
Daphnia magna, <24 hours 25 neonates per group	Confidor 200 SL (Bayer) OECD Guidelines Concentrations: 0, 2.2, 4.4, and 8.8 mg a.i./L	Substantial and concentration related decrease in feeding at all concentrations (see Fig.1 of paper). Predation cues modestly augmented inhibition of feeding and increased respiration (see Figs.1 and 2 of paper).	Pestana et al. 2010 Portugal/Canada
Daphnia magna, <24 hours old Groups of 5, 4-5 days old, 3 replicates	Imidacloprid (Bayer, purity/nature not specified) OECD Guidelines 24 hour exposures 10 concentrations	EC_{50} (feeding inhibition) 3.7 mg/L Slope: 0.77	Loureiro et al. 2010 Portugal
Saltwater			
Chydorus sphaericus (salt-water Cladocera)	Imidacloprid (99.5%) 22±1 °C	Dark 48-hour EC_{50} 0.832 (0.274-2.522) mg a.i./L Normal photoperiod 48-hour EC_{50} 2.209 (1.289-3.787) mg a.i./L	Sanchez-Bayo and Goka 2006a Japan

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

<table>
<tr><th>Species</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Other studies</td><td></td><td></td><td></td></tr>
<tr><td>Daphnia magna</td><td>Imidacloprid (NOS)</td><td><table><tr><th>Concentration (mg/L)</th><th>Estimated Time to 50% Mortality (days)</th></tr><tr><td>0.25</td><td>384.7</td></tr><tr><td>0.75</td><td>69.7</td></tr><tr><td>2.22</td><td>18.6</td></tr><tr><td>6.7</td><td>15</td></tr><tr><td>20</td><td>18.4</td></tr><tr><td>60</td><td>3</td></tr></table>From Table 2 of paper with concentrations converted from µg/L to mg/L.
Author notes that estimated time exceed the lifespan of the organism.</td><td>Sanchez-Bayo 2009

Australia</td></tr>
<tr><td>Daphnia magna</td><td>Imidacloprid (NOS)</td><td><table><tr><th>Concentration (mg/L)</th><th>Estimated Time to 50% Mortality (days)</th></tr><tr><td>0.004</td><td>5.15</td></tr><tr><td>0.016</td><td>2.67</td></tr><tr><td>0.064</td><td>3.19</td></tr><tr><td>0.25</td><td>2.26</td></tr><tr><td>1</td><td>1.98</td></tr><tr><td>4</td><td>0.89</td></tr></table>Above modified from Table 3 of paper converting time in hours to days.</td><td>Sanchez-Bayo and Goka 2007

Japan</td></tr>
</table>

A6 Table 2: Amphipods, Acute Toxicity

<table>
<tr><th>Species</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Technical Grade</td><td></td><td></td><td></td></tr>
<tr><td>Hyalella azteca (scud)
2-3 mm juveniles, 2 replicates per concentration, 10 per replicate</td><td>Technical grade imidacloprid
96-hours
Mean Conc: 0.00035, 0.00097, 0.0035, 0.010, 0.034, 0.100, 0.340, 1.000 and 3.100 mg/L</td><td>96-hour LC_{50}: 0.526 (0.194 - 1.263) mg/
96-hour EC_{50}: 0.055 (0.034 - 0.063) mg/L
96-hour NOAEC (immobilization and abnormal effects, such as lethargy or surfacing) = 0.00035 mg/L</td><td>England and Bucksath 1991
MRID 42256303</td></tr>
<tr><td>Gammarus pulex
Wild caught, 3.8 mg – 15 mg bw.</td><td>Imidacloprid (99%).
96 hr exposure with 3 day post-exposure observation period.
13±1 °C
Concentrations: 0, 0.81. 2.7. 9.0, 30.0, and 100 µg/L</td><td>EC_{50}’s for feeding inhibition.<table><tr><th>Hrs.</th><th>EC_{10} (mg/L)</th><th>EC_{50} (mg/L)</th></tr><tr><td>24</td><td>0.00905</td><td>0.01896</td></tr><tr><td>48</td><td>0.00328</td><td>0.02059</td></tr><tr><td>72</td><td>0.00203</td><td>0.01050</td></tr><tr><td>96</td><td>0.00205</td><td>0.00534</td></tr></table>Except for highest concentration, recovery in feeding during 3-day recovery period. See Fig. 1 in paper.</td><td>Agatz et al. 2014</td></tr>
<tr><td>Gammarus pulex
Wild caught, 3.8 mg – 15 mg bw.</td><td>Imidacloprid (N0S)
Seven concentrations (not specified)
13±1 °C
96 hour exposure

Working Note: The higher EC50 values at 96-hours implies recovery.</td><td>EC_{50} for immobility based on measured concentrations.<table><tr><th>Hrs.</th><th>$E5_{50}$ (nmol/L)</th><th>EC_{50} (mg/L)</th></tr><tr><td>24</td><td>404</td><td>0.103</td></tr><tr><td>48</td><td>430</td><td>0.110</td></tr><tr><td>72</td><td>405</td><td>0.104</td></tr><tr><td>96</td><td>514</td><td>0.131</td></tr></table>Data from Table 1 converted from nmol/L to mg/L using MW of 255.66
Note: Data for mortality could not be used to calculate an LC_{50} …Due to the different concentration ranges of mortality and immobility.</td><td>Ashauer et al. 2011

Switzerland/Australia</td></tr>
<tr><td>Gammarus pulex
Wild caught</td><td>Imidacloprid (analytical grade)
15±2 °C
96-hours</td><td>LC_{50}:
0.27 (0.17-0.45) mg/L
Lethality based on lack of all movement not just immobility.</td><td>Beketov and Liess 2008

Germany</td></tr>
<tr><td>Gammarus roeseli
Wild caught
Different sizes (column 3)
10 per dose group</td><td>Imidacloprid (NOS)
Stream water
12 °C
Concentrations: 6, 12, 24, 48, 96, 192, 384, and 768 µg/L</td><td>96-hour EC_{50} (immobility)
6 mm size
0.0142 (0.0064-0.0312) mg/L
9 mm size:
0.0019 (0.0001-0.0335) mg/L
11 mm size:
≈0.028 mg/L (Fig.1)</td><td>Bottger et al. 2012

Germany</td></tr>
</table>

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
Gammarus roeseli Wild caught Different sizes 10 per dose group	Imidacloprid (NOS) Artificial water 17 °C Concentrations: 6, 12, 24, 48, 96, 192, 384, and 768 µg/L	6 mm size 96-hour EC_{50} (immobility) 0.125 mg a.i./L (read from Fig. 2 of paper) Authors also give 24-hour values. `Working Note: Lower toxicity at higher temperature is unusual.`	Bottger et al. 2012 Germany
Formulations			
Gammarus fossarum (stream scud)	**Confidor SL 200** 48 hours 10 °C Concentrations: 1, 3, 10, 30, 100 mg/L `Working Note: Above unit of mg/L is not a typo. See p. 221, column 1 of paper.`	48-hours LC_{50}: 0.8 mg/L 24-hours EC_{50}: 0.07 mg/L Respiration movement Above values from Table 1 of paper. `Working Note: The above values do not seem sensible given the test concentrations used.`	Lukancic et al. 2010a,b Slovenia
Gammarus pulex 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 10, 30, 100, 300, 100 µg a.i./L	96-h LC_{50}: 0.316 (0.216-0.461) mg/L 96-h LC_{10}: 0.0995 (0.0.0322-0.307) mg/L 96-h EC_{50}: 0.0183 (0.00884-0.0378) mg/L 96-h EC_{10}: 0.00363 (0.000916-0.0144) mg/L EC for immobilization	Roessink et al. 2013

A6 Table 3: Midges, Acute Toxicity

Species	Exposure	Response	Reference
Midge (*Chironomus tentans*), Diptera; second instar 2 replicates per concentration, 10chironomids per replicate	Technical grade (NOS) Static renewal 10-day exposure Imidacloprid ((95.0%) Control, solvent control, measured concentrations of 0.00067, 0.00124, 0.00339, 0.0102, 0.0345, 0.100, and 0.329 mg a.i./L	96-hour LC_{50}: 0069 mg/L **Working Note: The previous Forest Service risk assessment (SERA 2005) had recorded an EC50 0.0105 (0.0077 - 0.0144) mg/L. U.S. EPA/OPP/EFED (2007a, p. 41) reports a higher 48-hour EC_{50} of 0.069 mg/L. It is not unusual for EFED to reanalyze data. The current risk assessment defers to EFED.**	Gagliano 1991 MRID 42256304 As summarized in U.S. EPA/OPP/ EFED (2007a, p. 41). Basis for EPA risk characterization.
Midge (*Chironomus tentans*), cultured, ≈7 days old 5 replicates	Imidacloprid (99.2% purity) 96-hours static 23±1 °C Concentrations: 0, 1, 5, 29, 145, 725 µg a.i./L	LC_{50}: 0.00575 (0.0041-0.00808) mg/L LC_{25}: 0.00246 mg/L NOEC: 0.00103 mg/L LOEC: 0.00439 mg/L Comparable to Admire formulation based on LC_{50}	Stoughton et al. 2008
Chironomus dilutus (midge, Diptera) Cultured, about 10 days old 5 replicates per concentration, 7-10 organisms per replicate	Admire 240F (Bayer) 96 hours 23 °C Concentrations: 0, 0.842, 1.39, 5.76, 11.2, 22.2 mg/L	96-h LC_{50}: 0.00265 (0.0016–0.00358) mg/L	Leblanc et al. 2013
Chironomus riparius (midge; Diptera) 5 replicates, 25 organisms per replicate	Confidor 200 SL (Bayer, Germany) 96-hour bioassay 10-day feeding 14.5-14.9 °C Concentrations: 0, 0.4, 1.2 and 3.7 µg/L. With and without predator cues.	96-hour EC_{50}: Without cues: 0.01294 (0.00974-0.01822) mg/L With cues: 0.01406 (0.01074-0.02018) mg/L 10-day feeding NOAEC: 0.004 mg/L LOAEC: 0.0012 mg/L No significant interaction with predator cues.	Pestana et al. 2009a
Chironomusriparius (midge, Diptera) Late 3rd instar.	Confidor 200 SL (Bayer, Germany) 20±2 °C 48-hours exposure with 144 hour recovery period. 0, 0.3, 0.55, and 1.2 µg/L	1.2 µg/L: Decrease in movements at 96 hours which persisted in 144 hour post-exposure period (Fig. 1 of paper). Decrease in ventilation frequency at all doses during recovery period. LOAEL (ventilation freq.): 0.3 µg a.i./L NOAEL: not determined	Azevedo-Pereira et al. 2011a

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
Chironomusriparius (midge, Diptera) Early 2nd instar.	Confidor 200 SL (Bayer, Germany) 20±2 °C Constant 10 day exposure. 4 pulse exposures followed by 6 day recovery period. Measured conc.: 0, 0.39, 0.74, and 2.15 µg/L	Decrease in growth at 2.15 µg/L in pulse and constant exposures. No significant impact at lower concentrations. Full recovery in post-exposure period. NOAEL: 0.74 µg/L LOAEL (growth): 2.15 µg/L.	Azevedo-Pereira et al. 2011b
Midge (*Chironomus tentans*), cultured, ≈7 days old 5 replicates	**Admire (240 g a.i./L)** 96-hours static 23±1 °C Concentrations: 0, 1, 5, 29, 145, 725 µg a.i./L	LC_{50}: 0.0054 (0.00401-0.00728) mg/L LC_{25}: 0.002285 mg/L NOEC: 0.00511 mg/L LOEC: 0.02359 mg/L Comparable to TGAI based on LC_{50}	Stoughton et al. 2008

A6 Table 4: Other Diptera, Acute Toxicity

Species	Exposure	Response	Reference
Freshwater			
Mosquito Larvae (*Aedes aegypti*); Diptera 4 replicates per concentration 10 animals each species per replicate	Imidacloprid (NOS) Temperature not specified 24-hours, static	24-hour LC_{50} Sensitive strain 0.339 (0.261 – 0.465) mg a.i./L Tolerant strain 1.833 (1.634 – 2.057) mg a.i./L Increased toxicity with piperonyl butoxide but not substantial – i.e., factor of 1.17 for sensitive strain and 2.77 for tolerant strain.	Riaz et al. 2013
Mosquito Larvae (*Aedes aegypti*); Diptera 4 replicates per concentration 10 animals each species per replicate	Imidacloprid (>95% purity) 27 °C (80.6 °F) 48-hours, static	48-hour LC_{50} 0.044 (0.041 – 0.047) mg a.i./L Slope: 4.02	Song et al 1997 ; Song and Brown 1998 (one study published in 2 papers).
Simulium latigonium (Diptera)	Imidacloprid (analytical grade) 15±2 °C 96 hours	96-hour LC_{50}: 0.00373 (0.00154- 0.00905) mg/L	Beketov and Liess 2008 Germany
Simulium vittatum (Diptera) 5th instar	Imidacloprid (analytical grade) 19.9-22 °C 96 hours	48-hour LC_{50} (3 replicates, based on measured concentrations) LD_{50} (mg a.i./L) / Slope 0.00825 (0.00756-0.00887) / 10.27 0.00675 (0.00604-0.00741) / 6.36 0.00954 (0.00871-0.01057) / 6.48 Geometric mean: 0.0081 mg a.i./L	Overmyer et al. 2005
Chaoborus obscuripes (Diptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 0.294 (0.247-0.350) mg/L 96-h LC_{10}: 0.178 (0.0661-0.481) mg/L 96-h EC_{50}: 0.284 (NR) mg/L 96-h EC_{10}: 0.223 (NR) mg/L EC for immobilization	Roessink et al. 2013
Saltwater			
Marsh mosquito, *(Aedes taeniorhynchus*); Diptera 4 replicates per concentration 10 animals each species per replicate	Imidacloprid (>95% purity) 27 °C 48-hours, static	48-hour LC_{50} 0.013 (0.01 –0-0.016) mg a.i./L Slope: 3.63	Song et al 1997; Song and Brown 1998 (one study published in 2 papers).

A6 Table 5: Ostracods, Acute Toxicity

Species	Exposure	Response	Reference
Cypretta seurati; Ostracoda, field caught	Imidacloprid (99.5%) 22±1 °C	Dark 48-hour EC_{50} 0.001 (0.0004-0.002) mg a.i./L Normal photoperiod 48-hour EC_{50} 0.016 (0.007-0.039) mg a.i./L	Sanchez-Bayo and Goka 2006a Japan
Cypridopsis vidua, Ostracoda	Imidacloprid (99.5%) 22±1 °C	Dark 48-hour EC_{50} 0.010 (0.0013-0.073) mg a.i./L Normal photoperiod 48-hour EC_{50} 0.003 (0.0005-0.015) mg a.i./L	Sanchez-Bayo and Goka 2006a Japan
Ilyocypris dentifera, Ostracoda	Imidacloprid (99.5%) 22±1 °C	Dark 48-hour EC_{50} 0.003 (0.0002-048) mg a.i./L Normal photoperiod 48-hour EC_{50} 0.003 (0.001-0.011) mg a.i./L	Sanchez-Bayo and Goka 2006a Japan
Heterocypris incongruens (Ostracoda)	Confidor SL 200 6-days Few experimental details	6-day EC_{50} (growth): 0.01-0.0015 mg a.i./L 6-day LC_{50}: >0.0015 mg a.i./L	Daam et al. 2013

A6 Table 6: Ephemeroptera, Acute Toxicity

Species	Exposure	Response	Reference
Freshwater			
Baetis rhodani (mayfly larvae) (Ephemeroptera) Wild caught	Imidacloprid (analytical grade) 15±2 °C 48-hours	48-hour LC_{50}: 0.00849 (0.00445- 0.0162) mg/L	Beketov and Liess 2008 Germany
Cloeon dipterum (mayfly; Ephemeroptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 0.00668 (0.00419-0.0106) mg/L 96-h LC_{10}: 0.00255 (0.000952-0.00685) mg/L 96-h EC_{50}: 0.00177 (0.00105-0.00299) mg/L 96-h EC_{10}: 0.000325 (0.000105-0.001) mg/L EC for immobilization	Roessink et al. 2013 Netherlands
Caenis horaria (mayfly; Ephemeroptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 3, 10, 30, 100 µg a.i./L	96-h LC_{50}: 0.0263 (0.0177-0.0391) mg/L 96-h LC_{10}: 0.00616 (0.00269-0.0141) mg/L 96-h EC_{50}: 0.00102 (0.00046-0.00228) mg/L 96-h EC_{10}: 0.0001 (0.000018-0.000554) mg/L EC for immobilization	Roessink et al. 2013 Netherlands
Epeorus longimanus (mayfly; Ephemeroptera) Early instars collected in spring 3 replicates, 5 organisms per replicate	**Admire (240 g a.i./L)** 24-hours 20 ±1 °C Concentrations: 0, 0.1, 0.5, 1, 5, 10, 100, and 240 µg/L.	24-hour LC_{50}: 0.0021 mg a.i./L	Alexander et al. 2007 Canada
Epeorus longimanus (mayfly; Ephemeroptera) Late instars collected in summer 3 replicates, 5 organisms per replicate	**Admire (240 g a.i./L)** 96-hours 20 ±1 °C Concentrations: 0, 0.1, 0.5, 1, 5, 10, 100, and 240 µg/L.	24-hour LC_{50}: 0.0021 mg a.i./L (identical to early instar) 96-hour LC_{50}: 0.00065 mg a.i./L	Alexander et al. 2007 Canada

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
Epeorus longimanus (mayfly; Ephemeroptera) Early and late instars 5 replicates, 5 organisms per replicate	**Admire (240 g a.i./L)** 24-hour pulse exposure 20 ±1 °C Concentrations: 0, 0.1, 0.5, 1, 5, and 10 µg a.i./L.	Feeding inhibition assay in artificial stream. Concentration related inhibition significant at 1 µg a.i./L and higher. Increased feeding in late but not early instars at 0.0001 and 0.0005 mg a.i./L Only larvae at 0.0001 mg/L recovered to pre-exposure feeding levels (by 4 days).	Alexander et al. 2007 Canada

A6 Table 7: Other Freshwater Invertebrates, Acute Toxicity

Species	Exposure	Response	Reference
Isopoda			
Asellus aquaticus (water louse; Isopoda)	**Confidor SL 200** 48 hours 10 °C Concentrations: 1, 3, 10, 30, 100 mg/L	48-hours LC_{50}: 8.5 mg/L 24-hours EC_{50}: 0.8 mg/L Respiration movement	Lukancic et al. 2010a,b
Asellus aquaticus (water louse; Isopoda) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SC) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 10, 30, 100, 300, 100 µg a.i./L	96-h LC_{50}: 0.316 (0.216-0.461) mg/L 96-h LC_{10}: 0.0616 (0.0341-0.11)) mg/L 96-h EC_{50}: 0.119 (NR) mg/L 96-h EC_{10}: 0.0247mg/L EC for immobilization	Roessink et al. 2013
Hemiptera			
Micronecta spp. (Hemiptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 0.0282 (0.0176-0.0452 mg/L 96-h LC_{10}: 0.00887-0.00343-0.0229) mg/L 96-h EC_{50}: 0.0108 (0.00972-0.012) mg/L 96-h EC_{10}: 0.00941 (0.00834-0.0106) mg/L EC for immobilization	Roessink et al. 2013
Notonecta spp. (Hemiptera) 15 replicates, 1 organism per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: >10 mg/L 96-h LC_{10}: > 10 mg/L 96-h EC_{50}: 0.0182 (0.00924-0.0357) mg/L 96-h EC_{10}: 0.003 (0.000779-0.0115) mg/L EC for immobilization	Roessink et al. 2013
Plea minutissima (pygmy backswimmers; Hemiptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 0.0375 (NR) mg/L 96-h LC_{10}: 0.0323 (NR) mg/L 96-h EC_{50}: 0.0359 (0.0311-0.0415) mg/L 96-h EC_{10}: 0.0304 (0.0261-0.0354) mg/L EC for immobilization	Roessink et al. 2013

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
Megaloptera			
Sialis lutaria (alderfly; Megaloptera) 15 replicates, 1 organism per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: >10 mg/L 96-h LC_{10}: > 10 mg/L 96-h EC_{50}: 0.0506 (0.0309-0.0828) mg/L 96-h EC_{10}: 0.0157 (0.00695-0.0354) mg/L EC for immobilization	Roessink et al. 2013
Tricoptera			
Sericostoma vittatum (caddisfly; Trichoptera) 10 replicates, 1 organism per replicate	Confidor 200 SL (Bayer, Germany) 96-hour bioassay 10-day feeding 14.5-14.9 °C Concentrations: 0, 1.9, 3.9, and 7.8 µg/L. With and without predator cues.	96-hour EC_{50}: Without cues: 0.04722 (0.03417-0.07074) mg/L With cues: 0.03586 (0.02547-0.05215) mg/L 10-day feeding NOAEC: 0.0.0039 mg/L LOAEC: 0.0078 mg/L Reduced oxygen consumption. Inhibition of burrowing at highest concentration. Predator cues increased burrowing.	Pestana et al. 2009a
Limnephilidae sp. (caddisfly; Trichoptera) 2 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 96 hours 18±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 0.0257 (0.0181-0.0365) mg/L 96-h LC_{10}: 0.00986 (0.00509-0.0191) mg/L 96-h EC_{50}: 0.00179 (0.000993-0.00322) mg/L 96-h EC_{10}: 0.000532 (0.00022-0.00129) mg/L EC for immobilization	Roessink et al. 2013
Annelids			
Blackworm (*Lumbriculus variegatus*) Annelida	Imidacloprid (NOS) 10 day exposure 20 °C Sediment concentrations: 0, 0.04, 0.5, 1.0, 2.5, and 10 mg/kg sediment.	Mortalities of about 10% or more at lower concentrations and approximately 35% at 5 mg/kg. Growth inhibition at all concentrations.	Sard and Soares 2010 Portugal
Lumbriculus variegatus (blackworm , Annelida) 3 replicates, 25 organisms per replicate	**Admire (240 g a.i./L)** 24-hour pulse exposure 20 ±1 °C Concentrations: 0, 0.1, 0.5, 1, 5, 10, 100, and 240 µg/L.	Feeding inhibition assay Inhibition at >5 µg a.i./L Delayed recovery at 0.5 µg/L (2 days) and 1 µg/L (4 days).	Alexander et al. 2007 Canada

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
Lumbriculus variegatus (blackworm , Annelida) 3 replicates, 25 organisms per replicate	**Admire (240 g a.i./L)** 96-hours 20 ±1 °C Concentrations: 0, 0.1, 0.5, 1, 5, 10, 100, and 240 µg/L.	96-hour EC_{50} (immobility): 0.0062 mg a.i./L	Alexander et al. 2007 Canada
Gastropods			
Marias cornuarietis (Giant Ramshorn snail) Gastropod Embryos 20 eggs (5 from 4 different egg sacks) per concentration.	Imidacloprid (Sigma-Aldrich, Germany) 24±1 °C 14 days Concentrations: 0, 10, 25, and 50 mg/L	No significant mortality of developmental effects. Two higher concentrations caused a significant decrease in heart rate (Figure 3 of paper) NOAEC: 10 mg/L	Sawasdee and Kohler 2009

A6 Table 8: Other Saltwater Invertebrates, Acute Toxicity

Species	Exposure	Response	Reference
Shrimp			
Shrimp *(Artemia* species); Anostraca 4 replicates per concentration 10 animals each species per replicate	Imidacloprid (>95% purity) 27 °C 48-hours, static	48-hour LC_{50} 361.23 (308 – 498) mg a.i./L Slope: 3.47	Song et al 1997 ; Song and Brown 1998 (one study published in 2 papers).
Daggerblade grass shrimp, (*Palaemonetes pugio*) Malacostraca	Imidacloprid (99.5% purity) 96-hours 25 °C Static renewal Concentrations: 0, 100, 200, 400, 600, 800 µg/L.	Larvae LC_{50}: 0.3088 (0.2736-0.3486) mg/L NOEC: 0.1 mg/L LOEC: 0.2 mg/L Adult LC_{50}: 0.5635 (0.4781-0.6642) mg/L	Key et al. 2007
Mysid Shrimp, (*Mysidopsis bahia*); Mysida, < 24 hours old 10 per concentration.	Imidacloprid (96.2% purity) Mean Conc: solvent control, 0.032, 0.0584, 0.0937, 0.146 and 0.249 mg a.i./L	96-hour LC_{50} = 0.0377 (0.0267 - 0.0464) mg a.i./L NOAEC not determined.	Ward 1990b MRID 42055319 Initial assay
Mysid Shrimp, (*Mysidopsis bahia*); Mysida, < 24 hours old 10 per concentration.	Imidacloprid (96.2% purity) Mean Conc: control, solvent control, 0.00842, 0.0133, 0.0229, 0.0372 and 0.0634 mg a.i./L	96-hour LC_{50} = 0.0341(0.0229 - 0.0372) mg a.i./L, NOAEC = 0.0133 mg a.i./L on the basis of mortality and loss of equilibrium at higher doses.	Ward 1990b MRID 42055319 Second assay
Mysid shrimp (*Mysidopsis bahia*); Mysida, < 24 hours old 2 replicates per concentration, 10 per replicate	**240 FS Formulation** Nominal (measured) Conc.: control, solvent control, 18 (21), 29 (31), 49 (56), 82 (78), 136 (125) and 227 (219) µg a.i./L nominal	96-hour LC_{50} = 0.036 mg a.i./L, 95% CI = 0.031 - 0.042 mg a.i./L NOAEC (mortality) = 0.021 mg a.i./L	Lintott 1992 MRID 42528301
Bivalves			
Eastern Oyster (*Crassostrea virginica*); Ostreoida 20 per concentration	Imidacloprid (95.8% purity) 96-hour flow-through Mean Conc.: control, solvent control, 2.93, 5.14, 8.19, 14.2, and 23.3 mg a.i./L	100% survival; No effects on new shell growth. NOAEC: 23.3 mg/L	Wheat and Ward 1991 MRID 42256305
Eastern Oyster (*Crassostrea virginica*); Ostreoida 20 per concentration	Imidacloprid (95.8% purity) 96-hour flow-through Mean Conc.: control, 145.0 mg a.i./L mg a.i./L,	100% survival; new shell growth of exposed was 22% less than controls. This was statistically significant.	Wheat and Ward 1991 MRID 42256305

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Exposure	Response	Reference
Mediterranean mussel (*Mytilus galloprovincialis*)	Imidacloprid (technical grade from Bayer) 4 days 16 °C Semi-static Concentrations: 0, 0.1, 1, and 10 mg/L	EC_{50}: 1.8 mg/L EC_{25}: 0.46 mg/L Endpoints not clear.	Dondero et al. 2010

A6 Table 9: Chronic Toxicity

Species	Chronic Exposure	Response	Reference
Cladocera			
	Technical Grade		
Daphnia magna, 4 replicate jars per concentration, 6 first instar daphnids each jar	Technical grade imidacloprid Static renewal Concentrations: Control, solvent control, 0.46, 0.86, 1.8, 3.6, and 7.3 mg/L	21-day EC_{50} (immobilization) >7.3 mg/L MATC = 2.5 mg/L (1.8 - 3.6 mg/L) NOAEC = 1.8 mg/L LOAEC = 3.6 mg/L 3.6 and 7.3 mg/L: Significantly reduced adult daphnid length in comparison with pooled controls 7.3 mg/L: Significantly reduced survival; significantly reduced mean young/adult reproduction days in comparison with pooled controls. No effects on time to first brood at any concentration	Young and Blake 1990 MRID 42055321
Daphnia magna, <24 hours old, 3replicates, 5 neonates per replicate	Imidacloprid (NOS), appears to have been technical grade, no solvent used Nominal Conc.: 1.8, 25, 45, 60, 85 and 140 mg/L. 20 °C 21-day period of exposure.	High Food Quality: EC_{50}: 37.24 (31.83-43.58) mg/L EC_{10}: 47.16 (39.72- 54.60) mg/L Lowest Food Quality: EC_{50}: 28.38 mg/L EC_{10}: 29.62 mg/L Several experiments on survival indicate that higher food quality improves survival. See Table 2 of paper. Maximum difference in EC_{50} values, however, is only a factor of 1.31 [37.24 mg/L ÷ 28.38 mg/L] `Working Note: The reported EC50 values are consistently lower than the reported EC10 values. See Table 2 of paper. Correspondence with author indicates that the EC10 values indicated 10% survival (Ieromina 2015).`	Ieromina et al. 2014 Netherlands
Daphnia magna, <24 hours 10 container, 1 daphnid per container	Imidacloprid technical grade (>99% pure) Static renewal 21±1 °C 21-days Concentrations: 0, 0.625, 1.25, 2.5, 5, 10, 20, 40 mg/L	NOAEC: 1.25 mg/L LOAEC (neonate production) 2.5 mg/L LOAEC (mortality) 40 mg/L See Table 2 of paper for other endpoints. Increase in activities of activities of cholinesterases, glutathione S-transferase and catalase. **Working Note: Compare a.i. for formulation in text. Unusual and highly relevant study. Different relationships based on endpoint.**	Jemec et al. 2007 Slovenia

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Chronic Exposure	Response	Reference
Daphnia magna, <24 hours 10 container, 1 daphnid per container	Imidacloprid (NOS), appears to be technical grade. See Pavlaki et al. 2014 20 °C 21 days Nominal Conc.: 0, 2, 4, 6, 8, and 10 mg/L	Body length: Dose-related decrease NOAEC: 4 mg/L LOAEC: 6 mg/L Decrease in reproduction: EC_{50}: 5.5 mg/L NOAEC and LOAEC not specified. Delay in neonate production: NOAEC: 6 mg/L LOAEC: 8 mg/L, 2-day delay.	Pavlaki et al. 2011 Pavlaki et al. 2014 Portugal
Daphnia magna, neonates, <24 hours old 10 replicates, 1 organism/replicate	Imidacloprid (99%, Sigma Aldrich) Higher food density (0.05 TOC/d) Static renewal 7 day exposure 34 day observation Concentrations: 0, 0.15, and 12.0 mg/L	0.15 mg/L: No effect on feeding. No significant effect on time to maturation. Decrease (NS) in number of offspring 12 mg/L: pronounced feeding inhibition (≈97%) (Table 2 of paper). Substantial and significant increase in time to maturation (≈12 d control, 16.6 d exposed). (Table 3 of paper).	Agatz and Brown 2013b
Daphnia magna, neonates, <24 hours old 10 replicates, 1 organism/replicate	Imidacloprid (99%, Sigma Aldrich) Lower food density (0.035 TOC/d) Static renewal 7 day exposure 27 day observation Concentrations: 0, 0.15, 0.40, 1.3, 4.0, 12.0 mg/L	Concentration-related decrease in feeding at all concentrations – i.e., from ≈15% at lowest concentration to 40% at highest concentration (Table 2 of paper). Significant increase in time to maturation only at highest concentration. Significant decrease in offspring at two higher concentrations. Significant decrease in number of offspring/adult only at two higher concentrations (See Table 3 of paper).	Agatz and Brown 2013b
	Formulation		
Ceriodaphnia dubia 4 batches of 10 neonates	Admire Pro (Bayer) Imidacloprid (99.5%) Exposure Period: 8 days. Temperature not specified. Conc. 0. And 8,093 µg/L	Mortality of 15% Significant reduction in population growth rate relative to controls (p<0.001). See Table 2: 0.282 day^{-1} in exposed vs 0.313 day^{-1} in controls. Reduction of ≈10%. Significant synergism by R-11.	Chen et al. 2010 U.S.
Daphnia magna, <24 hours 10 container, 1 daphnid per container	Confidor SL 200 (200 g a.i./L) Static renewal 21-days 21±1 °C Concentrations: 0, 1.25, 2.5, 5, 10, 20, 40 mg a.i./L	NOAEC: 2.5 mg/L LOAEC (neonate production) 5 mg/L LOAEC (mortality) 10 mg/L See Table 2 of paper for other endpoints. Increase in activities of activities of cholinesterases, glutathione S-transferase and catalase.	Jemec et al. 2007 Slovenia

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

<table>
<tr><th>Species</th><th>Chronic Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Daphnia magna, <24 hours
OECD protocols (10 container, 1 daphnid per container)</td><td>Confidor 200 SL (Bayer)
21-days
Static renewal (every 2 days)
Concentrations: 0, 2.2, 4.4, and 8.8 mg a.i./L</td><td>Number of broods
NOAEC: 2.2 mg a.i./L
LOAEC 4 mg/L
Note: No effects or lesser responses with predator cues.
Inhibition of feeding rate
LOAEC: 2.2 mg a.i./L
Note: Somewhat greater inhibition with predator cues.
Feeding rate increased respiration which may have led to higher food consumption.</td><td>Pestana et al. 2010

Portugal/Canada</td></tr>
<tr><td>Amphipoda</td><td></td><td></td><td></td></tr>
<tr><td></td><td>Technical Grade</td><td></td><td></td></tr>
<tr><td>Gammarus pulex
Wild caught
3 replicates plus controls.</td><td>Imidacloprid (99.9% purity, Sigma-Aldrich)
21 days
13 °C
Pulse concentration of 90 µg/ with 4 days between pulses</td><td>No significant impact on feeding overall. Organisms began to eat two days after pulse exposure.

Note: TWA exposures were intended to be identical to continuous exposures.</td><td>Nyman et al. 2013

U.K./Switzerland</td></tr>
<tr><td>Gammarus pulex
Wild caught
3 replicates plus controls.</td><td>Imidacloprid (99.9% purity, Sigma-Aldrich)
21 days
13 °C
Pulse concentration of 90 µg/ with 8 days between pulses</td><td>No significant impact on feeding overall. Organisms began to eat two days after pulse exposure.

Note: TWA exposures were intended to be identical to continuous exposures.</td><td>Nyman et al. 2013

U.K./Switzerland</td></tr>
<tr><td></td><td>Formulation</td><td></td><td></td></tr>
<tr><td>Hyalella azteca, cultured
6 replicate beakers</td><td>Admire (240 g a.i./L)
28 day static renewal
23±1 °C
Measured Concentrations: 0, 0.3, 1.3-1.46, 3.53-3.54, and 11.95-11.46 µg a.i./L
Hyphens separate 10 and 28 day measurements.</td><td><table><tr><th>Days</th><th>NOAEC (mg/L)</th><th>LOAEC (mg/L)</th></tr><tr><td>Continuous</td><td></td><td></td></tr><tr><td>10</td><td>0.00353</td><td>0.01195</td></tr><tr><td>28</td><td>0.00344</td><td>0.01146</td></tr><tr><td>Pulse</td><td></td><td></td></tr><tr><td>10</td><td>0.01193</td><td>N.D.</td></tr><tr><td>28</td><td>0.00353</td><td>0.01193</td></tr></table>Above endpoint is survival which is more sensitive than dry weights (see Table 4 of paper).</td><td>Stoughton et al. 2008</td></tr>
</table>

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

<table>
<tr><th>Species</th><th>Chronic Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Gammarus pulex
3 replicates, 10 organisms per replicate.</td><td>Soluble concentrate (SL) 200 g a.i./L (source not specified)
28-days
20±1 °C
Concentrations: 10, 30, 100, 300, 100 µg a.i./L</td><td>96-h LC_{50}: 0.0338 (0.0209-0.0546) mg/L
96-h LC_{10}: 0.00577 (0.00192-0.0173) mg/L

96-h EC_{50}: 0.0154 (0.0098-0.0241) mg/L
96-h EC_{10}: 0.00295 (0.00115-0.00759) mg/L
EC for immobilization</td><td>Roessink et al. 2013</td></tr>
<tr><td>Diptera</td><td></td><td></td><td></td></tr>
<tr><td></td><td>Technical Grade</td><td></td><td></td></tr>
<tr><td>Midge (Chironomus tentans), second instar
2 replicates per concentration, 10 organisms per replicate</td><td>Imidacloprid (95.0% purity)
Static renewal
Mean Conc.: control, solvent control, 0.00067, 0.00124, 0.00339, 0.0102, 0.0345, 0.100, and 0.329 mg a.i./L</td><td>10-day LC_{50}: 0.00317 (0.00124 - 0.0102) mg/L
10-day survival NOAEC: 0.00124 mg/L
10-day growth NOAEC: 0.00067 mg/L (basis = dry weight of survivors)</td><td>Gagliano 1991
MRID 42256304</td></tr>
<tr><td>Midge (Chironomus tentans), cultured
7 replicate beakers</td><td>Admire (240 g a.i./L)
28 day static renewal
23±1 °C
Measured Concentrations: 0, 0.36-0.37, 1.17-1.14, and 357-3.46µg a.i./L
Hyphens separate 10 and 28 day measurements.</td><td><table><tr><th>Days</th><th>NOAEC (mg/L)</th><th>LOAEC (mg/L)</th></tr><tr><td>Continuous</td><td></td><td></td></tr><tr><td>10</td><td>0.00117</td><td>0.00357</td></tr><tr><td>28</td><td>0.00114</td><td>0.00346[1]</td></tr><tr><td>Pulse</td><td></td><td></td></tr><tr><td>10</td><td>0.00347</td><td></td></tr><tr><td>28</td><td>0.00347</td><td></td></tr></table>Above are for dry weight, more sensitive than survival. See Table 4 of paper.
[1] All animals died.</td><td>Stoughton et al. 2008</td></tr>
<tr><td></td><td>Formulation</td><td></td><td></td></tr>
<tr><td>Chaoborus obscuripes (Diptera)
3 replicates, 10 organisms per replicate.</td><td>Soluble concentrate (SL) 200 g a.i./L (source not specified)
28 days
20±1 °C
Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L</td><td>96-h LC_{50}: 0.0126 (0.00733-0.0216) mg/L
96-h LC_{10}: 0.00199 (0.000523-0.0076) mg/L

96-h EC_{50}: 0.0118 (0.00817-0.0171) mg/L
96-h EC_{10}: 0.00457 (0.00205-0.0102) mg/L
EC for immobilization</td><td>Roessink et al. 2013</td></tr>
</table>

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Chronic Exposure	Response	Reference
Isopods			
Asellus aquaticus (water louse; Isopoda) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 28 days 20±1 °C Concentrations: 10, 30, 100, 300, 100 µg a.i./L	96-h LC_{50}: 0.0203 (0.00861-0.0479) mg/L 96-h LC_{10}: 0.00135 (0.000164-0.011) mg/L 96-h EC_{50}: 0.0119 (0.00594-0.0237) mg/L 96-h EC_{10}: 0.00171 (0.000386-0.00755 mg/L EC for immobilization	Roessink et al. 2013
Megaloptera			
Sialis lutaria (alderfly; Megaloptera) 15 replicates, 1 organism per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 28 days 20±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 32.5 mg/L 96-h LC_{10}: 25.1 mg/L 96-h EC_{50}: 0.00346 (0.00186-0.00644) mg/L 96-h EC_{10}: 0.00128 (0.000382-0.00431) mg/L EC for immobilization	Roessink et al. 2013
Hemiptera			
Plea minutissima (pygmy backswimmers; Hemiptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 28 days 20±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 0.0098 (0.00761-0.0126) mg/L 96-h LC_{10}: 0.00435 (0.00266-0.00711) mg/L 96-h EC_{50}: 0.00645 (0.00481-0.00864) mg/L 96-h EC_{10}: 0.00203 (0.00126-0.00328) mg/L EC for immobilization	Roessink et al. 2013
Ephemeroptera			
Cloeon dipterum (mayfly; Ephemeroptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 28 days 20±1 °C Concentrations: 0, 1, 10, 30, 100, 300 µg a.i./L	96-h LC_{50}: 0.000195 (0.000113-0.000338) mg/L 96-h LC_{10}: 0.000041 (0.000013-0.000124) mg/L 96-h EC_{50}: 0.000123 (0.000075-0.000201) mg/L 96-h EC_{10}: 0.000033 (0.000012-0.000090) mg/L EC for immobilization	Roessink et al. 2013
Caenis horaria (mayfly; Ephemeroptera) 3 replicates, 10 organisms per replicate.	**Soluble concentrate (SL) 200 g a.i./L** (source not specified) 28 days 20±1 °C Concentrations: 0, 1, 3, 10, 30, 100 µg a.i./L	96-h LC_{50}: 0.000316 mg/L 96-h LC_{10}: 0.235 mg/L 96-h EC_{50}: 0.000126 (0.000070-0.000228) mg/L 96-h EC_{10}: 0.000024 (0.000006-0.000091) mg/L EC for immobilization	Roessink et al. 2013

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Species	Chronic Exposure	Response	Reference
Saltwater Shrimp			
Mysid shrimp (*Mysidopsis bahia*), < 24 hours old 4 replicates per concentration, 15 per replicate	Flow-through chronic Imidacloprid (96.2% purity) Mean Conc.: control, solvent control, 560, 1290, 2850, 5080 and 10100 ng a.i./L Initial assay Working Note: Concentrations are specified in nanograms/L ins study. Results column converted to mg/L.	0.00129 mg/L and higher: Significantly reduced number of offspring per female reproductive day 0.00508 mg/L and higher: significantly reduced growth of first generation mysids as total length and as dry weight 0.0101 mg/L: Statistically increased mortality in comparison with pooled controls for first generation. No effects on mortality in second generation. U.S. EPA/OPP/EFED 2007a NOAEC: 0.0006 mg a.i./L LOAEC: 0.0013 mg a.i./L	Ward, 1991 MRID 42055322
Mysid shrimp (*Mysidopsis bahia*), < 24 hours old 4 replicates per concentration, 15 per replicate	Flow-through chronic Imidacloprid (96.2% purity) Nominal Conc.: 36.8, 78.4, 163, 326 and 643 ng a.i./L Second assay Working Note: Concentrations are specified in nanograms/L ins study. Results column converted to mg/L.	No effects on number of offspring per female reproductive day. 0.000326 and 0.000643 mg/L: Significantly reduced growth of first generation as total length and as dry weight in comparison with pooled controls 0.000643 mg/L: Statistically increased mortality in comparison with pooled controls for first generation. No effects on mortality in second generation. No real explanation for discrepancy between first and second tests with regard to growth.	Ward, 1991 MRID 42055322

A6 Table 10: Mesocosm/Mixed Species Studies

Sorted by author(s)

Type/Species	Exposure	Response	Reference
Artificial stream *Baetis rhodani* (mayfly larvae) (Ephemeroptera) Wild caught	Imidacloprid (analytical grade) Artificial stream Pulse exposure, 0.97 µg/L Concentration below LC_{50} by factor of about 8.	LOAEC: 0.00097 Increased drift (not quantified)	Beketov and Liess 2008
Artificial stream *Gammarus pulex*	Imidacloprid (analytical grade) Artificial stream Pulse exposure, 30 µg/L Concentration below LC_{50} by factor of about 9.	LOAEC: 0.030 Increased drift (not quantified)	Beketov and Liess 2008
Benthic sediment/water microcosms, outdoor 3 week colonization phase 14 control mesocosms and 7 mesocosms per treatment.	Imidacloprid (NOS) 3-week treatment phase with three pulses at 1 week intervals. 7-week observation phase. Nominal Pulse Concentrations: 0, 0.6, 1.4, 3.2, 7.5, 17.3, and 40 µg/L. TWA Concentrations: 0.0, 0.2, 0.4, 1.0, 2.3, 5.2, 12 µg/L (see Table 2 of paper).	Chironomidae (midges): Significant decrease in species diversity at two highest concentrations. Decrease in abundance only at highest concentration (Fig 2 of paper). Orthocladiinae (subfamily of Chironomidae): Significant decreases in diversity and abundance only at highest concentration (Fig 3 of paper). *Ablabesmyia* sp. (midge): Significant decreases in diversity and abundance only at highest concentration (Fig 4 of paper). *Radix* sp. (snails): Increase in abundance at highest concentration (Fig 5 of paper). *Caenis* sp. (Ephemeroptera): Significant decrease in emergence at 3.2 µg/L and higher. Significant decrease in abundance at two higher concentrations. NOAEC: Nom. 0.0014 mg/L (all species) TWA: 0.0004 mg/L (all species) LOAEC: Nom. 0.0032 mg/L (Ephemeroptera) TWA: 0.001 mg/L (Ephemeroptera)	Colombo et al. 2013 Australia

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Type/Species	Exposure	Response	Reference
Rice paddy mesocosm, outdoor 6 paddies, 5.2 m x 1.6 m. flow-through systems without recycling of water	Admire (100g/box NOS) Two applications 1 year apart. Initial peak concentrations of about 19 µg/L in year one and 15 µg/L in year two (Fig. 1 of paper) **Working Note: See Hayasaka et al. 2012c: Admire (1% granular imidacloprid, Bayer CropScience, Tokyo, Japan)**	Decrease in benthic organisms in both years. Decreased growth of medaka fish. Number of coleopteran species decreased in first year but increased in second year relative to controls. Bivalves and gastropods no well-represented. Low abundance of ostracods and chironomids. Overall decrease in some species in Year 1 could be due to dry weather. No significant differences in overall community structure scores. LOAEC: 0.0019 mg a.i./L (peak)	Hayasaka et al. 2012a Japan
Rice paddy mesocosm, outdoor 6 paddies, 5.2 m x 1.6 m. flow-through systems without recycling of water	Admire (1% granular imidacloprid, Bayer CropScience, Tokyo, Japan) Initial peak concentrations of about 49 µg/L (at 2 hours) dropped rapidly to 1 µg/L by Day 3. 119 day observation period	Low turbidity, higher pH, and higher dissolved oxygen. From Day 28 to Day 56, zooplankton abundance in imidacloprid-treated fields significantly lower than controls (i.e., 1 µg a.i./L). Recovery of abundance by Day 112. Significant decrease in leafhoppers and aphids and scavenger species such as Chironomidae, Sarcophagidae, Ephemeroptera and Gastropoda. Also significant decrease in organisms at air-water interface (i.e., neuston species). Decrease in abundance of actively swimming (nekton) species. Significant drop in Shannon-Wiener diversity index. LOAEC (transient) Peak: 0.049 mg a.i./L TWA: 0.001 mg a.i./L	Hayasaka et al. 2012c
Glass aquaria, 20°C, natural stream water and sediment, alder twigs. Stonefly (*Pteronarcys dorsata* [Plecoptera] and crane fly (*Tipula* sp., [Diptera], larvae as representative leaf-shredding insects. 9 organisms of each species	EcoPrid (experimental EC formulation, 50 mg a.i./mL), tree injection. Leaves from green ash, 0.06 g a.i./cm diameter dbh, recommended rate. 14 days 12 ash leaves from treated trees added to 6 L of natural stream water. No imidacloprid detected in water.	Mortality: Stonefly: 6.7% *Tipula* sp.: 2.2% No morbidity. Not significantly different from controls. No effect on leaf shredding, microbial respiration, or microbial decomposition rates. NOAEC: Not expressible in units of concentration. Compare to direct water exposure study (below). Consistent with 0.012 mg a.i./L or less.	Kreutzweiser et al. 2007

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

<table>
<tr><th>Type/Species</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Glass aquaria, 20°C, natural stream water and sediment, alder twigs.
Stonefly (Pteronarcys dorsata [Plecoptera] and crane fly (Tipula sp., [Diptera], larvae as representative leaf-shredding insects.
9 organisms of each species</td><td>Merit Solupak, 750 mg a.i./g, soil injection,
Leaves from green ash, 0.56 g a.i./cm dbh (recommended rate).
14 days
12 ash leaves from treated trees added to 6 L of natural stream water.
No imidacloprid detected in water.</td><td>Mortality:
Stonefly: 8.9%
Tipula sp.: 6.7%
No morbidity.
Not significantly different from controls.
No effect on leaf shredding, microbial respiration, or microbial decomposition rates.

NOAEC:
Not expressible in units of concentration. Compare to direct water exposure study (below). Consistent with 0.012 mg a.i./L or less.</td><td>Kreutzweiser et al. 2007</td></tr>
<tr><td>Glass aquaria, 20°C, natural stream water and sediment, alder twigs.
Stonefly (Pteronarcys dorsata [Plecoptera] and crane fly (Tipula sp., [Diptera], larvae as representative leaf-shredding insects.
9 organisms of each species</td><td>Merit Solupak, 750 mg a.i./g, soil injection,
Leaves from green ash, 10x recommended rate.
14 days
12 ash leaves from treated trees added to 6 L of natural stream water.
Monitored concentrations of 0.009 to 0.30 mg/L over 14-day period with some no detect.</td><td>Mortality:
Stonefly: 88.9%
Tipula sp.: 13.3%
Morbidity
Stonefly: 0.0%
Tipula sp.: 77.8%
Combined mortality and morbidity for Tipula sp. was 91.1%, significantly (p<0.05) greater than controls.
Mortality in stonefly was significantly (p<0.05) greater than controls.
No effect on microbial respiration or microbial decomposition rates.

LOAEC:
Not expressible in units of concentration. Compare to direct water exposure study (below). Consistent with 0.135 mg a.i./L or higher.</td><td>Kreutzweiser et al. 2007</td></tr>
<tr><td>Glass aquaria, 20°C, natural stream water and sediment, alder twigs.
Stonefly (Pteronarcys dorsata [Plecoptera] and crane fly (Tipula sp., [Diptera], larvae as representative leaf-shredding insects.
9 organisms of each species</td><td>EcoPrid (experimental EC formulation, 50 mg a.i./mL)
14 days
Imidacloprid added directly to water.
Mean Conc.: 0, 0.001, 0.012, 0.135, 1.55, and 15.4 mg a.i./L

Working Note: Mortality here consistent with exposures to contaminated leaves. See below.</td><td>Mortality at Day 14:
<table>
<tr><th>Conc. (mg a.i./L)</th><th>Stonefly % dead</th><th>Tipula % dead</th></tr>
<tr><td>0</td><td>4.4</td><td>0</td></tr>
<tr><td>0.001</td><td>8.3</td><td>2.8</td></tr>
<tr><td>0.012</td><td>7.4</td><td>0</td></tr>
<tr><td>0.135</td><td>94.4</td><td>100</td></tr>
<tr><td>1.55</td><td>100</td><td>94.4</td></tr>
<tr><td>15.4</td><td>100</td><td>100</td></tr>
</table>
Above is total response (mortality and morbidity). Responses in bold significant greater than controls.

NOAEC (mortality):
0.012 mg a.i./L</td><td>Kreutzweiser et al. 2007</td></tr>
</table>

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Type/Species	Exposure	Response	Reference
Glass aquaria, stream water and sediment Stonefly (*Pteronarcys dorsata* [Plecoptera] and crane fly (*Tipula* sp., [Diptera], larvae as representative leaf-shredding insects. 18.8 to 19.9°C 5 replicates per group. 9 organisms of each species	Imidacloprid (NOS) Low Field Rate: Maple trees treated at recommended rate of 25 g/m dbh by stem injection in fall and harvested in two weeks (to minimize translocation). Contaminated fallen leaves (n=9) added to 6 liters of stream water. Conc. in leaves of 3.2 (1.4-5.4, range) mg/kg. 14 days	Mortality: Stonefly: 11.1% *Tipula* sp.: 22.2% Not significantly different from controls. No significant effect on leaf decomposition (insects and microorganisms combined). NOAEC: Not expressible in units of concentration. Compare to direct water exposure study (below). Consistent with a concentration of <0.12 mg a.i./L.	Kreutzweiser et al. 2008a
Glass aquaria, stream water and sediment Stonefly (*Pteronarcys dorsata* [Plecoptera] and crane fly (*Tipula* sp., [Diptera], larvae as representative leaf-shredding insects. 18.8 to 19.9°C 5 replicates per group. 9 organisms of each species	Imidacloprid (NOS) High Field Rate: Maple trees treated at recommended rate of 25 g/m dbh by stem injection in June and harvested in October. Contaminated fallen leaves (n=9) added to 6 liters of stream water. Conc. in leaves of 11 (6.4-18.5, range) mg/kg. 14 days	Mortality Exposed: Stonefly: 8.8% *Tipula* sp.: 31.1% Not significantly different from controls. Significant effect on leaf decomposition (insects and microorganisms combined). NOAEC: Not expressible in units of concentration. Compare to direct water exposure study by Kreutzweiser et al. (2008c), summarized below. The NOAEC in this study is consistent with a concentration of <0.012 mg a.i./L based on Kreutzweiser et al. (2008c).	Kreutzweiser et al. 2008a

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

<table>
<tr><th>Type/Species</th><th>Exposure</th><th>Response</th><th>Reference</th></tr>
<tr><td>Glass aquaria, stream water and sediment
Stonefly (Pteronarcys dorsata [Plecoptera] and crane fly (Tipula sp., [Diptera], larvae as representative leaf-shredding insects.
18.8 to 19.9°C
5 replicates per group.
9 organisms of each species per replicate</td><td>Imidacloprid (NOS)
Overdose Field Rate: (NOD)
Contaminated fallen leaves (n=9) added to 6 liters of stream water. Conc. in leaves of 38.6 (31.2-5.4, range) mg/kg.
14 days</td><td>Mortality Exposed:
Stonefly: 83.3%
Tipula sp.: 83.3%
Note: Above rates are identical in both species and were significantly higher than in controls.
Significant effect on leaf decomposition (insects and microorganisms combined). All leaf degradation attributed to microorganisms based on lack of visual leaf damage by insects.

LOAEC:
Not expressible in units of concentration. Compare to direct water exposure study (below). Consistent with a concentration of 0.096 mg a.i./L or greater.</td><td>Kreutzweiser et al. 2008a</td></tr>
<tr><td>Glass aquaria, stream water and sediment
Stonefly (Pteronarcys dorsata [Plecoptera] and crane fly (Tipula sp., [Diptera], larvae as representative leaf-shredding insects.
20 (±3)°C
9 organisms of each species per replicate
3 replicates per concentration</td><td>Confidor 200SL (200 g/L)
Nominal concentrations: 0, 12, 24, 48, and 96 µg/L.

14 days

Working Note: Static exposure. Concentrations in water at the end of the 14-day period were somewhat less than 50% of the nominal concentration.</td><td><table><tr><th>Conc. (mg a.i./L)</th><th>Stonefly % dead</th><th>Tipula % dead</th></tr><tr><td>0</td><td>3.7</td><td>11.1</td></tr><tr><td>0.012</td><td>3.7</td><td>7.4</td></tr><tr><td>0.024</td><td>7.4</td><td>7.4</td></tr><tr><td>0.048</td><td>40.7</td><td>18.5</td></tr><tr><td>0.096</td><td>70.4</td><td>33.3</td></tr><tr><td></td><td>mg/L</td><td>mg/L</td></tr><tr><td>LC_{50}</td><td>0.071</td><td>0.139</td></tr><tr><td>LC_{10}</td><td>0.0208</td><td>0.0162</td></tr></table>Significant reduction in leaf loss at all concentrations. No visible signs of insect leaf shredding at two higher concentrations. Signs of insect shredding at lower concentrations but less so than in controls.

Working Note: See Figure 1a. The effect of imidacloprid on leaf decomposition is clear but there does not seem to be a concentration-response relationship. This could be due to a stimulation of microbial degradation which is suggested in Figure 1b.</td><td>Kreutzweiser et al. 2008c</td></tr>
</table>

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Type/Species	Exposure	Response	Reference
Identical to Kreutzweiser et al. 2007 5 replicates	EcoPrid (experimental EC formulation, 50 mg a.i./mL) [based on reference to 2007 paper] Low-dose 0.125g/cm DBH Leaf residue: 18.0 µg/g 14 days	Mortality Exposed: Stonefly: 8.9% *Tipula* sp.: 15.5% Mortality not significantly different from control. Surviving *Tipula* sp. seemed less active than controls. Significant decrease in loss of leaf mass for both species. NOAEC: Not expressible in units of concentration in water.	Kreutzweiser et al. 2009
Identical to Kreutzweiser et al. 2007 5 replicates	EcoPrid (experimental EC formulation, 50 mg a.i./mL) [based on reference to 2007 paper] High-dose 0.25g/cm DBH Leaf residue: 122.9 µg/g 14 days	Mortality Exposed: Stonefly: 17.8% *Tipula* sp.: 53.3% Mortality significantly higher than control. Stonefly survivors seemed less active than controls. Surviving *Tipula* sp. were moribund. Significant decrease in loss of leaf mass for both species. LOAEC: Not expressible in units of concentration in water.	Kreutzweiser et al. 2009
Stream mesocosm, indoor Mixed invertebrate population dominated by dipterans and crustaceans 4 replicates of treated and control.	Imidacloprid (NOS) 2 sets of 12 hour pulses, 1 week apart. First set on Day 0, second set on Day 50 All pulses of 12 µg/L 86 day observation period Started in spring (15.7°C). Second pulse in summer had higher temperature (17.4°C).	Abundance data indicated stronger effects in the second series of 3 pulses, perhaps due to higher temperature. Caddisfly (Trichoptera) most sensitive species. Ephemeroptera and Diptera responded only with repeated exposures. Emergence of Ephemeroptera adversely affected (decreased abundance and emergence). General decrease in taxa over time due to emergence of Diptera. Non-emerging invertebrates (gammarids/amphipods) increased over time. *On the basis of the population count data alone, no pulse effects on taxa numbers and gammarid abundance were evident in both pulse series.* Working Note: Based on discussion in paper, the exposures cannot be viewed as a NOAEC. LOAEC: 0.012 mg a.i./L	Mohr et al. 2012 Germany

Appendix 6: Toxicity to Aquatic Invertebrates *(continued)*

Type/Species	Exposure	Response	Reference
Multiple-species: phytoplankton, zooplankton, macroinvertebrates, including Hyalella azteca; 3 tanks each for control and 5 concentration levels	19-Week microcosm study with technical grade imidacloprid (95.8% a.i.) Four surface applications at 2-week intervals at nominal concentrations of 0, 0.002, 0.006, 0.020, 0.060 and 0.180 mg a.i./L Mean Conc.: 0, 0.0015, 0.0047, 0.019, 0.058 and 0.180 mg a.i./L	Amphipods were the most sensitive species, with statistically significant impacts at the lowest concentration tested. Impacts (statistically significant decrease in population) on cyanophytes (blue-green algae) and copepods at the 3 highest concentrations. Statistically significant decrease in populations of total macroinvertebrates as well as individual macroinvertebrate taxa (Mayfly, Midge, Caddisfly, and Beetle) at the three highest concentrations. Working Note: Study authors recommend 0.006 mg/L as NOEC for regulatory action. A mean concentration of 0.0047 mg/L seems more reasonable for groups except amphipods. NOAEC (other groups): 0.0047 mg a.i./L NOAEC (other groups): 0.019 mg a.i./L NOAEC (amphipods): Not determined LOAEC (amphipods): 0.002 mg a.i./L.	Moring et al. 1992 MRID 42256306 Not cited or discussed in EFED 2007a, 2008a
Stream mesocosm, outdoors Mixed benthic organisms. 16 replicates	Imidacloprid (NOS) 12-hour pulses to 1.63 and 17.6 µg a.i./L, every 7 days.	Concentration-related decreases in invertebrate density but significant only at higher concentration. Low Concentration: No significant effects. High Concentration: Diptera: No significant effects. Coleoptera: relatively tolerant. Oligochaetes: relatively sensitive. NOAEC: 0.00163 mg a.i./L LOAEC: 0.0176 mg a.i./L	Pestana et al. 2009a
Artificial rice paddies, outdoor 2 replicates for exposed and control	Admire GS, 1% a.i., 15 kg/ha (≈0.134 lb a.i./acre) Peak initial concentration of 109 µg/L dropping to 0.51 µg/L by 4 months. 4 month observation period.	Peak effects by weeks 2-4. Highly erratic patterns with respect to controls (Fig. 1). By end of observation period, benthic communities had recovered (p. 671). NOAEC/LOAEC: unclear.	Sanchez-Bayo and Goka 2012

A6 Table 11: Metabolites, Acute Toxicity

Species	Exposure	Response	Reference
Hyalella azteca (amphipod), 14 - 21 days old, Two replicates per concentration, 10 organisms per replicate	96-hour static acute toxicity of NTN 33519 urea metabolite Nominal (measured) concentrations of 0, 6.25 (5.81), 12.5 (11.80), 25 (23.46), 50 (46.80), and 100 (94.83) mg a.i./L	96-hour LC*50*: > 94.83 mg a.i/L, 96-hour EC*50* (immobilization): > 94.83 mg a.i/L, 96-hour NOAEC: 94.83 mg a.i./L	Dobbs and Frank 1996a MRID 43946603
Hyalella azteca (amphipod), 14 - 21 days old, two replicates per concentration, 10 organisms per replicate	96-hour static acute toxicity of NTN 33823 hydroxyl metabolite at mean measured concentrations of 0, 5.6, 11.0, 22.1, 43.8 and 86.8 mg/L	96-hour LC_{50}: 51.8 mg a.i/L, 95% CI = 44.0 - 60.9 mg a.i./L 96-hour EC_{50} (immobilization): 29.0 mg a.i./L, 95% CI = 24.7 - 34.0 mg a.i./L 96-hour NOAEC (mortality): 22.1 mg a.i./L	Rooney and Bowers 1996 MRID 43946601
Midge (*Chironomus tentans*) 2 replicates per concentration, 10chironomids per replicate	96-hour static NTN 33823 hydroxyl metabolite at mean nominal (measured) concentrations of 0, 0.1 (0.12), 1.0 (0.87), 10.0 (8.19) and 100 (82.8) mg a.i./L	96-hour LC_{50}: >82.8 mg a.i/L,96-hour EC50 (sub-lethal effects)): 17.0 mg a.i./L, 95% CI = 10.3 - 28.1mg a.i./L 96-hour NOAEC (mortality and sub-lethal effects): 8.19 mg a.i./L, sub-lethal effects included mottled coloration and erratic behavior.	Bowers 1996a MRID 43946602
Midge (*Chironomus tentans*), ≈ 16 days old 2 replicates per concentration, 10chironomids per replicate	96-hour static acute toxicity of NTN 33519 urea metabolite Nominal (measured) concentrations of 0, 0.1 (0.10), 1 (1.0), 10 (10.04) and 100 (99.80) mg a.i./L	96-hour LC_{50}: > 99.80 mg a.i/L, 96-hour EC_{50} (sub-lethal effects): >99.80 mg a.i/L, 96-hour NOAEC: 99.80 mg a.i./L	Dobbs and Frank 1996b MRID 43946604
Midge (*Chironomus tentans*)	6-chloronicotinic acid (97% a.i.) 96-hour static acute	96-hour LC_{50}: > 1 mg a.i./L NOAEC = 1 mg a.i./L	Bowers and Lam 1988 MRID 44558901

Appendix 7: Toxicity to Aquatic Plants

A7 Table 1: Algae

Species	Exposure	Response	Reference
Blue-Green Algae (*Anabaena flos-aquae*)	Merit 2F (21.6% a.i.) Mean measured concentrations: 0, 24.9, 40.5, 68.2, 121.3, and 193.3 mg a.i./L	96-hr EC_{25} 26.7(18.9-29.2) mg a.i./L 96-hr EC: 32.8(30.4-34.6) mg a.i./L 96-hr NOEC = 24.9 mg a.i./L	Bowers 1996b MRID 44187101
Diatom (*Navicula pelliculosa*)	Merit 2F (21.6% a.i.) Mean measured concentrations: 0, 0.16, 0.42, 1.05, 2.64, 6.69, and 17.0 mg a.i./L	96-hr NOAEC: 6.69 mg a.i./L 96-hr LOAEC: 9.88 mg a.i./L	Hall 1996 MRID 44187102
Green algae (*Scenedesmus subspicatus*)	Technical grade imidacloprid (92.8% a.i) Nominal concentrations: 0, 0.1, 1, and 10 mg a.i./L	96-hr NOAEC: 10 mg a.i../L LOAEC: not determined Working Note: Used by U.S. EPA/OPP/EFED (2008a, p. 17) and U.S. EPA/OPP/EFED (2007a, p. 24) for risk characterization.	Heimbach 1989 MRID 42256374
Green algae (*Selenastrum capricornutum*, a.k.a. *Pseudokirchneriella subcapita*)	Technical grade imidacloprid (92.8% a.i) Mean measured concentrations: 0, 14.1, 24.1, 41.1, 69.5, and 119 mg a.i./L	5-day NOAEC: >119 mg/L 5-day LOAEC: Not determined	Gagliano and Bowers 1991 MRID 42256374
Green algae (*Selenastrum capricornutum*, a.k.a. *Pseudokirchneriella subcapita*)	Confidor 200 SL (200 g a.i./L)	72-hour EC_{50} (growth): >600 mg a.i./L	Daam et al. 2013
Green algae (*Selenastrum capricornutum*, a.k.a. *Pseudokirchneriella subcapita*)	Confidor (Bayer Hellas AG). Composition of formulation not specified.	IC_{50}: >1000 mg/L IC_{50} for inhibition of growth. Not clear if data are expressed as a.i. or formulation.	Kungolos et al. 2009 Greece

Species	Exposure	Response	Reference
Green algae (*Desmodesmus subspicatus*)	Imidacloprid (NOS but clearly a technical grade)	72-hour IC_{50}: 389 mg/L 72-hour IC_{10}: 106 mg/L Working Note: The term IC_{xx} used in this paper designates inhibition of growth and is identical to the more common EC_{xx} designation.	Tisler et al. 2009 Slovenia
Green algae (*Desmodesmus subspicatus*)	Confidor 200 SL (Bayer)	72-hour EC_{50}: 116 mg a.i./L 72-hour EC_{10}: 5.6 mg a.i./L	Tisler et al. 2009 Slovenia
Vibrio fischeri (bioluminescent marine bacteria)	Confidor (Bayer Hellas AG). Composition of formulation not specified. Concentrations of 4.40, 8.79,17.6, 35.2, 70.3,140, 281 and 562 mg/L.	226 (159–322) mg/L Not clear if data are expressed as a.i. or formulation.	Kungolos et al. 2009 Greece

A7 Table 2: Field/Mesocosm Studies

Species	Exposure	Response	Reference
Algae (mixed populations)	19-Week microcosm study with technical grade imidacloprid (95.8% a.i.) Four surface applications at 2-week intervals at nominal concentrations of 0, 0.002, 0.006, 0.020, 0.060 and 0.180 mg a.i./L Mean Conc.: 0, 0.0015, 0.0047, 0.019, 0.058 and 0.180 mg a.i./L	Transient decreases in population density. Accompanied by population decreases in invertebrates (See Appendix 6). Impacts (statistically significant decrease in population) on blue-green algae at the 3 highest doses. NOAEC: 0.0047 mg/L Working Note: This study is not cited in U.S. EPA/OPP/EFED (2007a, 2008a).	Moring et al. 1992 MRID 42256306

A7 Table 3: Macrophytes

Species	Exposure	Response	Reference
Lemna minor (duckweed)	Confidor 200 SL (200 g a.i./L)	7-day EC_{50} (frond number and area): 740 mg a.i./L	Daam et al. 2013 Portugal

Appendix 8: Gleams-Driver Modeling, Soil Injection

Soil Injection

Table 1: Effective Offsite Application Rate (lb/acre)

Site	Clay	Loam	Sand
Dry and Warm Location	1.87E-07 (0 - 1.89E-05)	0 (0 - 8.00E-07)	0 (0 - 0)
Dry and Temperate Location	9.50E-08 (0 - 2.83E-05)	0 (0 - 2.97E-06)	0 (0 - 0)
Dry and Cold Location	1.16E-06 (0 - 1.37E-05)	0 (0 - 7.00E-07)	0 (0 - 0)
Average Rainfall and Warm Location	0.00044 (0.000134 - 0.0009)	0.000054 (7.70E-06 - 0.000198)	0 (0 - 4.30E-09)
Average Rainfall and Temperate Location	0.00027 (0.0001 - 0.00056)	0.000032 (1.73E-06 - 0.000116)	0 (0 - 0)
Average Rainfall and Cool Location	0.000071 (1.02E-05 - 0.000207)	0.000004 (1.78E-09 - 0.000046)	0 (0 - 0)
Wet and Warm Location	1.93E-06 (9.90E-08 - 0.000068)	1.39E-08 (0 - 6.50E-06)	0 (0 - 0)
Wet and Temperate Location	0.000111 (0.000039 - 0.000237)	1.15E-05 (2.01E-06 - 0.000041)	0 (0 - 0)
Wet and Cool Location	0 (0 - 2.80E-07)	0 (0 - 3.50E-09)	0 (0 - 0)
Average of Central Values:	9.90E-05	1.13E-05	0
25th Percentile:	1.87E-07	0	0
Maximum:	0.0009	1.98E-04	4.30E-09
Summary:	9.90E-05 (1.87E-07 - 0.0009)	1.13E-05 (0 - 1.98E-04)	0 (0 - 4.30E-09)

Soil Injection

Table 2: Concentration in Top 12 Inches of Soil (ppm)

Site	Clay	Loam	Sand
Dry and Warm Location	0.41 (0.36 - 0.45)	0.37 (0.32 - 0.4)	0.37 (0.33 - 0.39)
Dry and Temperate Location	0.46 (0.43 - 0.49)	0.41 (0.38 - 0.43)	0.4 (0.36 - 0.42)
Dry and Cold Location	0.51 (0.5 - 0.51)	0.45 (0.44 - 0.45)	0.45 (0.44 - 0.45)
Average Rainfall and Warm Location	0.4 (0.36 - 0.43)	0.33 (0.289 - 0.37)	0.27 (0.24 - 0.306)
Average Rainfall and Temperate Location	0.43 (0.39 - 0.46)	0.36 (0.32 - 0.39)	0.279 (0.247 - 0.34)
Average Rainfall and Cool Location	0.43 (0.4 - 0.46)	0.36 (0.33 - 0.39)	0.28 (0.251 - 0.32)
Wet and Warm Location	0.315 (0.287 - 0.35)	0.25 (0.238 - 0.271)	0.231 (0.231 - 0.233)
Wet and Temperate Location	0.33 (0.294 - 0.36)	0.256 (0.239 - 0.285)	0.232 (0.231 - 0.235)
Wet and Cool Location	0.35 (0.313 - 0.39)	0.267 (0.243 - 0.294)	0.232 (0.226 - 0.237)
Average of Central Values:	0.4	0.34	0.305
25th Percentile:	0.35	0.267	0.232
Maximum:	0.51	0.45	0.45
Summary:	0.4 (0.35 - 0.51)	0.34 (0.267 - 0.45)	0.305 (0.232 - 0.45)

Soil Injection

Table 3: Concentration in Top 36 Inches of Soil (ppm)

Site	Clay	Loam	Sand
Dry and Warm Location	0.138 (0.12 - 0.151)	0.122 (0.108 - 0.132)	0.123 (0.11 - 0.132)
Dry and Temperate Location	0.155 (0.143 - 0.162)	0.136 (0.128 - 0.142)	0.136 (0.122 - 0.143)
Dry and Cold Location	0.17 (0.167 - 0.171)	0.15 (0.148 - 0.151)	0.15 (0.147 - 0.152)
Average Rainfall and Warm Location	0.138 (0.123 - 0.147)	0.119 (0.105 - 0.127)	0.118 (0.102 - 0.128)
Average Rainfall and Temperate Location	0.153 (0.14 - 0.159)	0.133 (0.124 - 0.14)	0.133 (0.121 - 0.138)
Average Rainfall and Cool Location	0.159 (0.149 - 0.164)	0.14 (0.135 - 0.145)	0.139 (0.13 - 0.143)
Wet and Warm Location	0.142 (0.125 - 0.151)	0.122 (0.11 - 0.13)	0.091 (0.081 - 0.11)
Wet and Temperate Location	0.156 (0.147 - 0.162)	0.135 (0.126 - 0.141)	0.096 (0.082 - 0.118)
Wet and Cool Location	0.165 (0.157 - 0.168)	0.143 (0.134 - 0.147)	0.109 (0.088 - 0.13)
Average of Central Values:	0.153	0.133	0.122
25th Percentile:	0.142	0.122	0.109
Maximum:	0.171	0.151	0.152
Summary:	0.153 (0.142 - 0.171)	0.133 (0.122 - 0.151)	0.122 (0.109 - 0.152)

Soil Injection

Table 4: Maximum Penetration into Soil Column (inches)

Site	Clay	Loam	Sand
Dry and Warm Location	18 (8 - 30)	18 (8 - 30)	18 (8 - 36)
Dry and Temperate Location	24 (12 - 30)	24 (8 - 36)	30 (12 - 36)
Dry and Cold Location	30 (24 - 30)	30 (24 - 36)	36 (30 - 36)
Average Rainfall and Warm Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Average Rainfall and Temperate Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Average Rainfall and Cool Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Wet and Warm Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Wet and Temperate Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Wet and Cool Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Average of Central Values:	32	32	33.3
25th Percentile:	30	30	36
Maximum:	36	36	36
Summary:	32 (30 - 36)	32 (30 - 36)	33.3 (36 - 36)

Soil Injection

Table 5: Stream, Maximum Peak Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	0.0007 (0 - 0.04)	0 (0 - 0.0026)	0 (0 - 0.02)
Dry and Temperate Location	0.0004 (0 - 0.03)	0 (0 - 0.006)	0 (0 - 0.18)
Dry and Cold Location	0.0023 (0 - 0.019)	0 (0 - 0.0016)	0.0005 (0 - 0.01)
Average Rainfall and Warm Location	0.11 (0.05 - 0.28)	0.2 (0.016 - 1.12)	12.9 (2.08 - 34)
Average Rainfall and Temperate Location	0.1 (0.04 - 0.29)	0.2 (0.009 - 1.66)	12 (3.3 - 28)
Average Rainfall and Cool Location	0.05 (0.008 - 0.4)	0.4 (0.009 - 2.41)	12.2 (5.8 - 28.7)
Wet and Warm Location	2.32 (0.8 - 5.4)	6.9 (3.3 - 12)	30.5 (19.3 - 56)
Wet and Temperate Location	3.1 (1.14 - 6.9)	9 (3.8 - 15.9)	33 (23.4 - 45)
Wet and Cool Location	5.3 (2.48 - 10.8)	15.7 (8.3 - 23.5)	41 (31.1 - 51)
Average of Central Values:	1.22	3.6	15.7
25th Percentile:	0.0023	0	0.0005
Maximum:	10.8	23.5	56
Summary:	1.22 (0.0023 - 10.8)	3.6 (0 - 23.5)	15.7 (0.0005 - 56)

Soil Injection

Table 6: Stream, Annual Average Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	2.3E-06 (0 - 0.00016)	0 (0 - 0.000008)	0 (0 - 0.00011)
Dry and Temperate Location	1.3E-06 (0 - 0.00018)	0 (0 - 0.00002)	0 (0 - 0.0012)
Dry and Cold Location	0.000014 (0 - 0.0001)	0 (0 - 0.000005)	2.5E-06 (0 - 0.00007)
Average Rainfall and Warm Location	0.0028 (0.001 - 0.011)	0.004 (0.00015 - 0.04)	0.5 (0.06 - 1.6)
Average Rainfall and Temperate Location	0.0018 (0.0009 - 0.011)	0.006 (0.00016 - 0.06)	0.5 (0.12 - 1.63)
Average Rainfall and Cool Location	0.0009 (0.00017 - 0.013)	0.015 (0.00024 - 0.11)	0.7 (0.26 - 1.97)
Wet and Warm Location	0.28 (0.09 - 0.8)	1 (0.4 - 1.94)	4 (2.73 - 5.6)
Wet and Temperate Location	0.3 (0.09 - 1)	1.11 (0.4 - 2.43)	4.9 (3.6 - 6.7)
Wet and Cool Location	0.6 (0.23 - 1.44)	2.09 (1.02 - 3.7)	6.6 (5.1 - 7.7)
Average of Central Values:	0.132	0.47	1.91
25th Percentile:	1.40E-05	0	2.50E-06
Maximum:	1.44	3.7	7.7
Summary:	0.132 (1.40E-05 - 1.44)	0.47 (0 - 3.7)	1.91 (2.50E-06 - 7.7)

Soil Injection

Table 7: Pond, Maximum Peak Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	0.0002 (0 - 0.02)	0 (0 - 0.0011)	0 (0 - 0.016)
Dry and Temperate Location	0.0001 (0 - 0.023)	0 (0 - 0.0028)	0 (0 - 0.2)
Dry and Cold Location	0.0012 (0 - 0.011)	0 (0 - 0.0008)	0.00025 (0 - 0.006)
Average Rainfall and Warm Location	0.5 (0.13 - 1.37)	0.6 (0.03 - 5.2)	61 (6.6 - 216)
Average Rainfall and Temperate Location	0.26 (0.09 - 1.15)	0.6 (0.019 - 6.4)	53 (10.9 - 175)
Average Rainfall and Cool Location	0.13 (0.022 - 1.22)	1.46 (0.025 - 10.5)	69 (25.3 - 200)
Wet and Warm Location	11.7 (3.2 - 29.8)	46 (12.8 - 92)	223 (92 - 320)
Wet and Temperate Location	8.1 (2.14 - 28)	35 (12.4 - 97)	196 (93 - 282)
Wet and Cool Location	33 (12.9 - 74)	99 (56 - 169)	245 (151 - 350)
Average of Central Values:	5.97	20.3	94.1
25th Percentile:	0.0012	0	0.00025
Maximum:	74	169	350
Summary:	5.97 (0.0012 - 74)	20.3 (0 - 169)	94.1 (0.00025 - 350)

Soil Injection

Table 8: Pond, Annual Average Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	0.00006 (0 - 0.007)	0 (0 - 0.0006)	0 (0 - 0.01)
Dry and Temperate Location	0.000018 (0 - 0.009)	0 (0 - 0.0011)	0 (0 - 0.09)
Dry and Cold Location	0.0005 (0 - 0.006)	0 (0 - 0.0004)	0.0001 (0 - 0.0021)
Average Rainfall and Warm Location	0.21 (0.06 - 0.4)	0.15 (0.012 - 1.81)	22 (2.21 - 99)
Average Rainfall and Temperate Location	0.12 (0.06 - 0.4)	0.15 (0.008 - 2.3)	21.7 (2.47 - 78)
Average Rainfall and Cool Location	0.05 (0.007 - 0.4)	0.3 (0.009 - 2.97)	23.8 (7.8 - 77)
Wet and Warm Location	3.9 (1.06 - 10.7)	17.5 (5.4 - 37)	105 (56 - 175)
Wet and Temperate Location	3.8 (1 - 14.3)	16.2 (5.7 - 41)	100 (57 - 137)
Wet and Cool Location	7.4 (2.72 - 17.8)	25.7 (13.2 - 48)	111 (82 - 151)
Average of Central Values:	1.72	6.67	42.6
25th Percentile:	0.0005	0	0.0001
Maximum:	17.8	48	175
Summary:	1.72 (0.0005 - 17.8)	6.67 (0 - 48)	42.6 (0.0001 - 175)

Appendix 9: Gleams-Driver Modeling, Directed Foliar Application

Table 1: Effective Offsite Application Rate (lb/acre)

Site	Clay	Loam	Sand
Dry and Warm Location	0.0034 (0 - 0.037)	0 (0 - 0.0057)	0 (0 - 0)
Dry and Temperate Location	0.0045 (0 - 0.028)	1.87E-06 (0 - 0.0047)	0 (0 - 0)
Dry and Cold Location	0.0063 (0.000169 - 0.0254)	0 (0 - 0.00125)	0 (0 - 0)
Average Rainfall and Warm Location	0.043 (0.0162 - 0.118)	0.005 (0.00126 - 0.0302)	0 (0 - 1.91E-07)
Average Rainfall and Temperate Location	0.046 (0.015 - 0.1)	0.0062 (0.00079 - 0.0262)	0 (0 - 0)
Average Rainfall and Cool Location	0.0279 (0.0106 - 0.07)	0.0022 (0.000083 - 0.014)	0 (0 - 0)
Wet and Warm Location	0.029 (0.0144 - 0.073)	0.0028 (0.0009 - 0.0126)	1.65E-09 (0 - 2.60E-07)
Wet and Temperate Location	0.029 (0.0138 - 0.072)	0.00209 (0.00039 - 0.0116)	0 (0 - 8.50E-10)
Wet and Cool Location	0.147 (0.108 - 0.268)	0.0243 (0.0114 - 0.064)	0 (0 - 5.10E-08)
Average of Central Values:	0.037	0.0047	1.83E-10
25th Percentile:	0.0063	1.87E-06	0
Maximum:	0.268	0.064	2.60E-07
Summary:	0.037 (0.0063 - 0.268)	0.0047 (1.87E-06 - 0.064)	1.83E-10 (0 - 2.60E-07)

Directed Foliar Application

Table 2: Concentration in Top 12 Inches of Soil (ppm)

Site	Clay	Loam	Sand
Dry and Warm Location	0.284 (0.247 - 0.311)	0.26 (0.226 - 0.288)	0.256 (0.217 - 0.283)
Dry and Temperate Location	0.316 (0.289 - 0.34)	0.295 (0.263 - 0.312)	0.293 (0.268 - 0.31)
Dry and Cold Location	0.36 (0.34 - 0.37)	0.33 (0.312 - 0.34)	0.33 (0.313 - 0.34)
Average Rainfall and Warm Location	0.268 (0.24 - 0.298)	0.248 (0.22 - 0.269)	0.214 (0.188 - 0.256)
Average Rainfall and Temperate Location	0.304 (0.28 - 0.32)	0.275 (0.249 - 0.3)	0.233 (0.199 - 0.275)
Average Rainfall and Cool Location	0.32 (0.299 - 0.34)	0.291 (0.269 - 0.307)	0.234 (0.197 - 0.275)
Wet and Warm Location	0.236 (0.215 - 0.264)	0.199 (0.181 - 0.221)	0.171 (0.16 - 0.176)
Wet and Temperate Location	0.259 (0.23 - 0.302)	0.207 (0.182 - 0.241)	0.171 (0.161 - 0.177)
Wet and Cool Location	0.268 (0.226 - 0.299)	0.223 (0.193 - 0.258)	0.172 (0.161 - 0.181)
Average of Central Values:	0.291	0.259	0.23
25th Percentile:	0.268	0.223	0.172
Maximum:	0.37	0.34	0.34
Summary:	0.291 (0.268 - 0.37)	0.259 (0.223 - 0.34)	0.23 (0.172 - 0.34)

Directed Foliar Application

Table 3: Concentration in Top 36 Inches of Soil (ppm)

Site	Clay	Loam	Sand
Dry and Warm Location	0.095 (0.082 - 0.104)	0.087 (0.075 - 0.096)	0.085 (0.072 - 0.094)
Dry and Temperate Location	0.105 (0.096 - 0.112)	0.098 (0.088 - 0.104)	0.098 (0.09 - 0.103)
Dry and Cold Location	0.12 (0.114 - 0.123)	0.11 (0.104 - 0.113)	0.111 (0.104 - 0.113)
Average Rainfall and Warm Location	0.09 (0.081 - 0.1)	0.085 (0.075 - 0.093)	0.086 (0.075 - 0.093)
Average Rainfall and Temperate Location	0.104 (0.095 - 0.11)	0.096 (0.087 - 0.103)	0.097 (0.088 - 0.103)
Average Rainfall and Cool Location	0.111 (0.101 - 0.116)	0.102 (0.094 - 0.106)	0.102 (0.095 - 0.106)
Wet and Warm Location	0.093 (0.083 - 0.102)	0.087 (0.079 - 0.095)	0.069 (0.061 - 0.083)
Wet and Temperate Location	0.109 (0.101 - 0.114)	0.1 (0.092 - 0.105)	0.074 (0.061 - 0.091)
Wet and Cool Location	0.106 (0.096 - 0.113)	0.103 (0.096 - 0.109)	0.084 (0.068 - 0.097)
Average of Central Values:	0.104	0.096	0.09
25th Percentile:	0.095	0.087	0.084
Maximum:	0.123	0.113	0.113
Summary:	0.104 (0.095 - 0.123)	0.096 (0.087 - 0.113)	0.09 (0.084 - 0.113)

Directed Foliar Application

Table 4: Maximum Penetration into Soil Column (inches)

Site	Clay	Loam	Sand
Dry and Warm Location	18 (8 - 24)	18 (8 - 30)	18 (8 - 36)
Dry and Temperate Location	18 (12 - 30)	18 (8 - 30)	24 (12 - 36)
Dry and Cold Location	18 (18 - 24)	24 (18 - 24)	30 (24 - 36)
Average Rainfall and Warm Location	36 (30 - 36)	36 (36 - 36)	36 (36 - 36)
Average Rainfall and Temperate Location	36 (30 - 36)	36 (36 - 36)	36 (36 - 36)
Average Rainfall and Cool Location	36 (30 - 36)	36 (36 - 36)	36 (36 - 36)
Wet and Warm Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Wet and Temperate Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Wet and Cool Location	36 (36 - 36)	36 (36 - 36)	36 (36 - 36)
Average of Central Values:	30	30.7	32
25th Percentile:	18	24	30
Maximum:	36	36	36
Summary:	30 (18 - 36)	30.7 (24 - 36)	32 (30 - 36)

Directed Foliar Application

Table 5: Stream, Maximum Peak Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	9.5 (0 - 45)	0 (0 - 9)	0 (0 - 0.004)
Dry and Temperate Location	9.5 (0 - 34)	0.007 (0 - 8.4)	0 (0 - 0.03)
Dry and Cold Location	15.4 (0.8 - 45)	0 (0 - 4.3)	0.000014 (0 - 0.0007)
Average Rainfall and Warm Location	33 (8.7 - 75)	5.2 (0.5 - 20.4)	6.4 (0.4 - 21.6)
Average Rainfall and Temperate Location	30.2 (12 - 62)	4.6 (0.6 - 16.7)	4.8 (0.7 - 15.5)
Average Rainfall and Cool Location	20.6 (8.1 - 54)	2.14 (0.08 - 12.3)	5.8 (1.34 - 15.7)
Wet and Warm Location	18.3 (6.5 - 48)	4.1 (1.6 - 11.1)	18.7 (12.1 - 34)
Wet and Temperate Location	16.5 (7 - 45)	5.4 (2.11 - 12.4)	22.4 (16.4 - 36)
Wet and Cool Location	51 (33 - 78)	12.9 (6 - 25.8)	28.6 (19.8 - 37)
Average of Central Values:	22.7	3.82	9.63
25th Percentile:	15.4	0.007	1.40E-05
Maximum:	78	25.8	37
Summary:	22.7 (15.4 - 78)	3.82 (0.007 - 25.8)	9.63 (1.40E-05 - 37)

Directed Foliar Application

Table 6: Stream, Annual Average Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	0.04 (0 - 0.22)	0 (0 - 0.04)	0 (0 - 0.000019)
Dry and Temperate Location	0.05 (0 - 0.18)	0.000021 (0 - 0.03)	0 (0 - 0.00014)
Dry and Cold Location	0.07 (0.0025 - 0.19)	0 (0 - 0.013)	7.0E-08 (0 - 0.000004)
Average Rainfall and Warm Location	0.26 (0.13 - 0.5)	0.029 (0.006 - 0.1)	0.17 (0.009 - 1.01)
Average Rainfall and Temperate Location	0.25 (0.11 - 0.5)	0.028 (0.004 - 0.1)	0.17 (0.023 - 0.8)
Average Rainfall and Cool Location	0.17 (0.07 - 0.4)	0.012 (0.0008 - 0.06)	0.28 (0.05 - 0.9)
Wet and Warm Location	0.22 (0.14 - 0.4)	0.4 (0.11 - 1)	2.65 (1.67 - 3.8)
Wet and Temperate Location	0.24 (0.14 - 0.6)	0.5 (0.15 - 1.33)	3.5 (2.54 - 4.9)
Wet and Cool Location	0.5 (0.4 - 0.9)	0.9 (0.4 - 1.93)	4.6 (3.4 - 5.8)
Average of Central Values:	0.2	0.208	1.26
25th Percentile:	0.07	2.10E-05	7.00E-08
Maximum:	0.9	1.93	5.8
Summary:	0.2 (0.07 - 0.9)	0.208 (2.10E-05 - 1.93)	1.26 (7.00E-08 - 5.8)

Directed Foliar Application

Table 7: Pond, Maximum Peak Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	3.7 (0 - 42)	0 (0 - 6.1)	0 (0 - 0.0031)
Dry and Temperate Location	4.6 (0 - 28.6)	0.002 (0 - 5)	0 (0 - 0.04)
Dry and Cold Location	6.5 (0.18 - 25.2)	0 (0 - 1.37)	0.000007 (0 - 0.0004)
Average Rainfall and Warm Location	42 (17.5 - 124)	5.3 (1.24 - 31.5)	25.1 (1.06 - 129)
Average Rainfall and Temperate Location	43 (12.4 - 104)	5.9 (0.7 - 26.3)	17.9 (2.49 - 94)
Average Rainfall and Cool Location	23.7 (8.1 - 66)	2.27 (0.13 - 13.2)	26 (5.4 - 94)
Wet and Warm Location	21.1 (9.9 - 55)	17.9 (4.3 - 47)	146 (61 - 238)
Wet and Temperate Location	18.9 (8 - 55)	14.6 (3.02 - 44)	148 (86 - 214)
Wet and Cool Location	22.1 (14.3 - 38)	50 (21.9 - 95)	171 (98 - 264)
Average of Central Values:	20.6	10.7	59.3
25th Percentile:	6.5	0.002	7.00E-06
Maximum:	124	95	264
Summary:	20.6 (6.5 - 124)	10.7 (0.002 - 95)	59.3 (7.00E-06 - 264)

Directed Foliar Application

Table 8: Pond, Annual Average Concentration in Surface Water (ug/L or ppb)

Site	Clay	Loam	Sand
Dry and Warm Location	1.74 (0 - 18.8)	0 (0 - 2.44)	0 (0 - 0.0012)
Dry and Temperate Location	1.92 (0 - 15.3)	0.00016 (0 - 2.95)	0 (0 - 0.023)
Dry and Cold Location	2.91 (0.08 - 12.3)	0 (0 - 0.6)	2.3E-06 (0 - 0.00018)
Average Rainfall and Warm Location	26.7 (8.7 - 81)	3.08 (0.4 - 19.4)	7.8 (0.4 - 49)
Average Rainfall and Temperate Location	25.3 (8.5 - 63)	3.13 (0.32 - 14.3)	5.5 (0.6 - 39)
Average Rainfall and Cool Location	13.6 (5.4 - 41)	1.2 (0.06 - 7.4)	8.4 (1.39 - 44)
Wet and Warm Location	8.1 (3.7 - 20.7)	6.4 (1.68 - 15.7)	66 (34 - 122)
Wet and Temperate Location	6.5 (2.47 - 17.2)	7.1 (1.49 - 21.4)	75 (41 - 105)
Wet and Cool Location	5.2 (3.4 - 11.2)	11.5 (5.7 - 24)	73 (51 - 102)
Average of Central Values:	10.2	3.6	26.2
25th Percentile:	2.91	0.00016	2.30E-06
Maximum:	81	24	122
Summary:	10.2 (2.91 - 81)	3.6 (0.00016 - 24)	26.2 (2.30E-06 - 122)