CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Glynn, T.P.	2006	Susceptibility of pest *Nezara viridula* (Heteroptera: Pentatomidae) and parasitoid *Trichopoda pennipes* (Diptera: Tachinidae) to selected insecticides.	Journal of economic entomology, (2006 Jun) Vol. 99, No. 3, pp. 648-57.	Does not fulfil criteria 5 (Several dose levels tested, to establish a dose response curve). Does not fulfil criteria 6 (Exposure route is environmentally relevant). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Susceptibility of the southern green stink bug, *Nezara viridula* (L.), and its endoparasitoid *Trichopoda pennipes* (F.) to thiamethoxam was compared in residual and oral toxicity tests. For the residual tests, the treatment was sprayed on the top and bottom of a plastic petri dish. Both methods are not standard Tier I and II toxicity test exposure methods. Percentages of mortality after residual exposure to 715 µg a.s./mL were reported for *T. pennipes* as 41.67% and 91.67%, respectively. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As the study methods differ from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	226

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
Ca 8.3.2	Villanueva, R.T.; Walgenbach, J.F.	2005	Development, oviposition, and mortality of *Neoseiulus fallacis* (Acari: Phytoseiidae) in response to reduced-risk insecticides.	Journal of economic entomology, (2005 Dec) Vol. 98, No. 6, pp. 2114-20.	Does not fulfil criteria 5 (several dose level tested, at least 3). Does not fulfil criteria 6 (exposure route is environmentally relevant). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). To assess the effect of insecticides on development and mortality of *N. fallacis* immatures and adults, 12h old eggs and 7 to 8 day old adults were individually placed on bean leaf disks previously dipped in insecticide solutions. Only one concentration of thiamethoxam was tested. Excised leaves were dipped in the test solutions, which is not a relevant exposure route. Additionally, only one concentration was tested so ECx and NOER values could not be calculated. As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	239

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Bostanian, N. J.; Hardman, J.M.; Ventard, E.; Racette, G.	2005	The intrinsic toxicity of several neonicotinoids to *Lygus lineolaris* and *Hyaliodes vitripennis*, a phytophagous and a predacious mirid.	Pest management science, (2005 Oct) Vol. 61, No. 10, pp. 991-6. Journal code:	Does not fulfil criteria 4 (test organisms are not previously exposed to the test material or other contaminants). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). In this study the residual toxicity of four neonicotinoids (including thiamethoxam) to *L. lineolaris* in an orchard and the toxicity to *H. vitripennis* in the laboratory was investigated. In the laboratory tests, leafs and test organisms were oversprayed until run-off; LC_{50} is given as 0.005 g a.s./L for adults. *Hyaliodes vitripennis* were collected from a commercial orchard in Dunham, Quebec, Canada where pesticide applications were minimal (no neonicotinoids used). However contamination by other pesticides cannot be excluded. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this the study methods differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	242

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Kilpatrick, A. L.; Hagerty, A. M.; Turnipseed, S. G.; Sullivan, M. J.; Bridges, W. C. Jr	2005	Activity of selected neonicotinoids and dicrotophos on nontarget arthropods in cotton: implications in insect management.	Journal of economic entomology, (2005 Jun) Vol. 98, No. 3, pp. 814-20.	Does not fulfil criteria 12 (Appropriate observation data (biological relevance) to derive endpoints). Field experiments with thiamethoxam were conducted in South Carolina, USA to examine the impact of naturally occurring insect predators in cotton at a recommended field rate. The abundance of predators were reported after 1 and 2 applications only (counts made 3-4 days after treatment). Recovery of non-target arthropods can not be demonstrated due to the limitations of the field study. Therefore this study is not considered relevent.	245
CA 8.3.2	Oliver, J.B.; Mannion, C.M.; Klein, M.G.; Moyseenko, J.J.; Bishop, B.	2005	Effect of insecticides on *Tiphia vernalis* (Hymenoptera: Tiphiidae) oviposition and survival of progeny to cocoon stage when parasitizing *Popillia japonica* (Coleoptera: Scarabaeidae) larvae.	Journal of economic entomology, (2005 Jun) Vol. 98, No. 3, pp. 694-703.	Does not fulfil criteria 4 (test organisms are not previously exposed to the test material or other contaminants). Does not fulfil criteria 5 (several dose level tested, at least 3). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). The percentage of parasitism of third instars of Japanese beetle by adult female *T. vernalis* and survival of parasitoid progeny to the cocoon stage exposed to insecticide-treated soil (0.00027 g a.s./kg soil = 0.29 kg a.s./ha), or insecticide-treated grubs was investigated. Since female wasps were collected along the perimeter of a commercial nursery near Tarlton, it cannot be excluded that they were previously exposed to any contaminants. Only one concentration was tested. As these study methods differs from the standard methods used for Tier I and II toxicity testing and no direct effects of non-target arthropods were investigated, this study is considered not considered relevant for this review.	247

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Samih, M. A.; Khajehhoseini , M.; Mahdian, K.; Alizadeh, A.	2013	Effect of Some Medicinal Plant Extracts on Stable population Parameters of *Chrysoperla Carnea* (Stephens).	Iranian Journal of Pharmaceutical Research, (Oct 2013) Vol. 12, Supp. Supplement 2, pp. 1290.	This article is an abstract published from a conference (2nd National Congress on Medicinal Plants, 2nd. Tehran, Iran, Islamic Republic of. 15 May 2013-16 May 2013). This article was not available from our copyright-cleared document delivery sources. While we placed an order to purchase this document from an advanced reference source, we were unable to obtain this article. Side effects of thiamethoxam were evaluated on stable population growth parameters of *Chrysoperla carnea* (Stephens) in controlled conditions. 1st instars larvae were treated by spray tower method. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. As this abstract was insufficiently detailed for review, this article is considered not relevant.	271

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Khajehhoseini , M.; Samih, M. A.n; Mahdian, K.; Alizadeh, A.	2013	Toxicity of Plant Extract *Rubia Tinctorum* in Comparision with Thiamethoxam on Green Lacewing *Chrysoperla Carnea* (Stephens).	Iranian Journal of Pharmaceutical Research, (Oct 2013) Vol. 12, Supp. Supplement 2, pp. 950.	This article is an abstract published from a conference (2nd National Congress on Medicinal Plants, 2nd. Tehran, Iran, Islamic Republic of. 15 May 2013-16 May 2013). This article was not available from our copyright-cleared document delivery sources. While we placed an order to purchase this document from an advanced reference source, we were unable to obtain this article. In this study, thiamethoxam was tested for their toxicity on the green lacewing. Different concentrations were prepared and first instar larva were sprayed by Potter Spray Tower. The calculated LC_{50} value was 9.09 ppm. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Furthermore, according to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. ppm). As the abstract was insufficiently detailed for review, this article is considered not relevant.	272

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Khajehhoseini , M.; Samih, M.d A.; Mahdian, K.; Alizadeh, A.	2013	The Effect of Thiamethoxam and Plant Extracts on Biological Parameters of *Chrysoperla Carnea* (Stephens) (Neu. Chrysopidae) in Iaboratory Condition.	Iranian Journal of Pharmaceutical Research, (Oct 2013) Vol. 12, Supp. Supplement 2, pp. 746.	This article is an abstract published from a conference (2nd National Congress on Medicinal Plants, 2nd. Tehran, Iran, Islamic Republic of. 15 May 2013-16 May 2013). This article was not available from our copyright-cleared document delivery sources. While we placed an order to purchase this document from an advanced reference source, we were unable to obtain this article. In this study the side effects of thiamethoxam biological parameters of *Chrysoperla carnea* (Stephens) were evaluated under controlled conditions. 1st instar larvae were treated with 750 µl/ml concentration of extracts by spray tower method. Results showed a min developmental time (egg to adult) observed for thiamethoxam of 35.03 ± 1.531 days and a max mean of larval duration of 19.68 ± 1.302. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Furthermore, according to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. µl/L). As the abstract was insufficiently detailed for review, this article is considered not relevant.	273

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Bredeson, M.M.; Reese, R. N.; Lundgren, J. G.	2015	The effects of insecticide dose and herbivore density on tri-trophic effects of thiamethoxam in a system involving wheat, aphids, and ladybeetles	Crop ProtectionCrop Protection (1 Mar 2015) Volume 69, pp. 70-76	Does not fulfil criteria 4 (Test organisms are not previously exposed to the test material or other contaminants). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Investigations on how herbivore density and insecticide dose affects the tri-trophic interactions among thiamethoxam-treated wheat, *Rhophalosiphum padi* and the predatory *Coleomegilla maculata* DeGeer. Second and fourth instar of *C. maculate* were fed aphids reared for 24 h on wheat plants treated with sub-lethal thiamethoxam soil drenches. This is not a recommended test system. Since individuals collected from a corn field near Brookings, SD, USA, it cannot be excluded that they were previously exposed to any contaminants. Test conditions (eg. temperature) were not similar to standard protocols. As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	296
CA 8.3.2	Duso, C.; Malagnini, V.; Pozzebon, A.; Buzzetti, Fi. M.; Tirello, P.	2008	A method to assess the effects of pesticides on the predatory mite Phytoseiulus persimilis (Acari Phytoseiidae) in the laboratory.	Biocontrol science and technology (2008), Vol. 18, Number 9-10, pp. 1027-1040	Does not fulfil criteria 6 (Exposure route is environmentally relevant). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) Evaluation of topical and residual effects of thiamethoxam on *P. persimilis*. Mites were treated by microimmersion and then reared in holding cells, on bean leaves previously dipped in a pesticide solution, which is not a relevant exposure route. As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	305

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Byrne, F. J.	2007	Evaluation of the Impact of Neonicotinoid Insecticides on the Glassy-winged Sharpshooter and Its Egg Parasitoids	Proceedings of the Pierce's Disease Research Symposium (2007), pp. 70-72.	Only results for imidachloprid are presented in this study. Thus, this reference is considered not relevant.	307
CA 8.3.2	Martinez, L.C.; Plata-Rueda, A.; Zanuncio, J. C.; Serrao, J. E.	2014	Comparative toxicity of six insecticides on the rhinoceros beetle (Coleoptera: Scarabaeidae)	Florida Entomologist, (SEP 2014) Vol. 97, No. 3, pp. 1056-1062.	In this lab study, the efficacy of thiamethoxam on a pest species was tested. Therefore this reference is considered not relevant.	311
CA 8.3.2	Shakoorzadeh , M.; Rafiee-Dastjerdi, H.; Golmohammadi, Gh.; Hassanpour, M.; Golizadeh, A.	2013	Lethal and sublethal effects of dinotefuran and thiamethoxam on the population growth parameters of the green lacewing, Chrysoperla carnea: (Neu.: Chrysopidae), under laboratory conditions.	Journal of Entomological Society of Iran, (2013) Vol. 33, No. 3, pp. 1-9.	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) The toxicity of thiamethoxam on first instar larvae of *Chrysoperla carnea* was investigated in the laboratory using a residual contact method. .Test concentrations were poured in both levels of glass petri dishes and left to dry *before C. carnea* larvae were introduced. The LC_{50} value estimated for thiamethoxam was 9.88 mg a.s./L. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	334

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Tinsley, N. A.; Steffey, K. L.; Estes, R. E.; Heeren, J. R.; Gray, M. E.; Diers, B. W.	2012	Field-level effects of preventative management tactics on soybean aphids (*Aphis glycines* Matsumura) and their predators.	Journal of Applied Entomology, (JUN 2012) Vol. 135, No. 5, pp. 361-371.	Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols). A 2-year field experiment was conducted in northern Illinois to evaluate the effects of host plant resistance and an insecticidal seed treatment (thiamethoxam) on soybean aphids, *Aphis glycines* Matsumura and their predators. The main aim of this study was to determine effects on aphid populations and yield. In this study only sticky traps were used to collect predator species at weekly intervals, which is not a sufficient and robust sampling method for non-target arthropod field studies. Therefore this reference is considered not relevant.	346

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Rocha, L. C. D; Carvalho, G. A; Moura, A. P; Moscardini, V. F; Rezende, D. T; Santos, O. M	2010	Physiologic selectivity of pesticides used on coffee plantations on eggs and adults of *Cryptolaemus Montrouzieri* Mulsant.	Arquivos do Instituto Biologico Sao Paulo, (2010) Vol. 77, No. 1, pp. 119-127.	English version of publication unavailable (Portugues); English abstract available. Does not fulfil criteria 5 (several dose levels tested, at least 3). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). The objective of this study was to evaluate some pesticides used on coffee plantations in regard to their lethal and sublethal effects on eggs and adults of *Cryptolaemus montrouzieri* Mulsant under laboratory conditions. Bioassays followed the methodology proposed by members of the IOBC; thiamethoxam (0.5 g/L) was applied via direct overspray. On the fifth day after spraying, thiamethoxam showed average of 6.7% survival. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	374

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Arno, J.; Roig, J.; Gabarra, R.	2008	Activity of some biorational and conventional insecticides against *Bemisia tabaci* and their compatibility with Whitefly parasitoids.	Journal of Insect Science (Tucson), (JAN 10 2008) Vol. 8, pp. 4-5.	This article is an abstract published from a conference (4th International Bemisia Workshop/International Genomic Workshop. Duck Key, FL, USA. December 03-08, 2006.). The study evaluated the efficacy of several insecticides (including thiamethoxam) against *Bemisia tabaci* and their compatibility with whitefly parasitiods. Trials were conducted under greenhouse conditions with poinsettia plants. Actara 25WG was applied by irrigation at two doses (maximum label rate and 2.5x label rate). The application type used in this study is not relevant to the uses considered in this assessment (foliar spray and seed treatment) therefore this article is not considered relevant.	394
CA 8.3.2	Lim, U. T.; Mahmoud, A. M. A.	2007	Pesticide susceptibility of Trissolcus nigripedius (Hymenoptera: Scelionidae) an egg parasitoid of Dolycoris baccarum (Heteroptera: Pentatomidae).	Entomological Research, (AUG 2007) Vol. 37, No. Suppl. 1, pp. A140.	This article is an abstract published from a conference (International Congress of Insect Biotechnology and Industry. Daegu, SOUTH KOREA. August 19-24, 2007.). Pesticides (including thiamethoxam) were evaluated for oral, residual and direct contact toxicity on *Trissolcus nigripedius* at a recommended field rate. The test methods were not significantly detailed for assessment. Therefore this article is not considered relevant.	402

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Li, W. D.; Zhang, P. J.; Zhang, J. M.; Lin, W. C.; Lu, Y. B.; Gao, Y. L.	2015	Acute and sublethal effects of neonicotinoids and pymetrozine on an important egg parasitoid, *Trichogramma ostriniae* (Hymenoptera: Trichogrammatidae).	Biocontrol Science and Technology (2015), Volume 25, Number 2, pp. 121-131	This study examined the acute and sublethal effects of thiamethoxam on adults of *Trichogramma ostriniae*. Laboratory bioassays were carried out by exposing adults to residues of the insecticides applied on glass tubes (no method references were listed). After 1 h of exposure, the wasps were transferred into a clean insecticide-free tube. After 24 h, the number and percentage mortality of dead wasps in the tubes were counted. 24h LC_{50} = 0.14 mg a.s./L To investigate the sublethal effects on reproduction and preimaginal development of offspring, adult wasps were exposed to the LC_{30} as described in the methods for the acute mortality bioassay. The adults that survived were presented rice moth egg discs and mortality of the wasps was recorded. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	419

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Tabozada, E. O.; Sayed, S. M.; El-Arnaouty, S. A.	2015	Side effects of sublethal concentration of two neonicotinoids; thiamethoxam and thiacloprid on the larval parasitoid, *Bracon brevicornis* (Hymenoptera: Braconidae).	American Journal of Experimental Agriculture (2015), Volume 5, Number 1, pp. 29-35	Does not fulfil criteria 5 (Several dose levels tested, to establish a dose response curve). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Effects of thiamethoxam at a sublethal dose (1.5 ppm) under laboratory conditions was investigated on the parasitoid, *B. brevicornis*. The parasitoid adults were exposed via direct contact (residues on a glass tube, left to dry for one hour) and indirect contact (2nd larval instars of *S. littoralis* were treated). Adult mortality after direct exposure was 1.3%, effects in the larval and pupal period did not differ significantly with the control. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. ppm). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	420
CA 8.3.2	Halikatti, G.; Pokharkar, D. S.; Shrikant V.; Vivek U.; Halikatti, G.; Vibhute, S.; Uppar, V.	2014	Toxicity of newer insecticides on adults of *Cryptolaemus montrouzieri* M. under laboratory condition.	Environment and Ecology (2014), Volume 32, Number 3, pp. 928-932	This item was not available from our copyright-cleared document delivery sources. We placed an order to purchase this document from an advanced reference source, however we were informed that this article is unavailable. Toxicity studies of insecticides (including thiamethoxam) to adults of *C. montrouzieri* by dry film method and food contamination technique was investigated under laboratory conditions. The available abstract was insufficiently detailed to determine the relevance of this article.	439

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Gontijo, P. C.; Moscardini, V. F.; Michaud, J. P.; Carvalho, G. A.	2014	Non-target effects of chlorantraniliprole and thiamethoxam on *Chrysoperla carnea* when employed as sunflower seed treatments.	Journal of Pest Science (2014), Volume 87, Number 4, pp. 711-719, 59	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). This study assessed the effects of sunflower seed treatments with thiamethoxam on *Chrysoperla carnea* by exposing larvae and adults to sunflower stem segments grown from treated seeds and the nectar secreted by their extrafloral nectaries. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route for beetle larvae would be accounted for in regulatory higher tier field studies. There are 4 for thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	445
CA 8.3.2	Ewunkem, J. A.; Jackai, L. E. N.; Osofuhene-Sintim, H.; Dingha, B. N	2014	Comparing the impact of a neonicotinoid and biorational Agroneem® on herbivorous and beneficial arthropods on cowpea and tomato.	Journal of Agricultural Science and Technology A (2014), Volume 4, Number 7, pp. 585-596	Does not fulfil criteria 13 (well defiend test material (incl. purity/content)). Does not fulfil criteria 15 (exposure route is clearly defined and suitably quantified). Does not fulfil criteria 16 (sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details)). Field experiments to evaluate and compare the effectiveness of a biorational (Agroneem®) and conventional pesticide (imidacloprid or thiamethoxam) on insect pests of cowpea and tomato were conducted. As no information was given on the formulation used, including specific application rate and number of applications, as well as information on the agricultural background of the experimental field, this reference is considered not relevant.	447

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Moscardini, V. F.; Gontijo, P. C.; Michaud, J. P.; Carvalho, G. A.	2014	Sublethal effects of chlorantraniliprole and thiamethoxam seed treatments when *Lysiphlebus testaceipes* feed on sunflower extrafloral nectar.	BioControl (2014), Volume 59, Number 5, pp. 503-511.	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). This study evaluated the performance of adult wasps fed extrafloral nectar of sunflower plants grown from seed treated with thiamethoxam. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 4 for thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	452
CA 8.3.2	Souza, J. R.; Carvalho, G. A.; Moura, A. P.; Couto, M. H. G.; Maia, J. B.	2014	Toxicity of some insecticides used in maize crop on *Trichogramma pretiosum* (Hymenoptera, Trichogrammatidae) immature stages.	Chilean Journal of Agricultural Research (2014), Volume 74, Number 2, pp. 234-239, 28	Does not fulfil criteria 8 (effects are related to a single test item). The effects of pesticides (including lambda-cyhalothrin/thiamethoxam) on *Trichogramma pretiosum* Riley immature stages were evaluated. As thiamethoxam was tested in a mixture formulation only, this reference is considered not relevant.	471

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Rahmani, S.; Bandani, A. R.; Sabahi, Q.	2013	Population statistics and biological traits of *Hippodamia variegata* (Goeze) (Coleoptera: Coccinellidae) affected by LC_{30} of thiamethoxam and pirimicarb.	Archives of Phytopathology and Plant Protection (2013), Volume 46, Number 15, pp. 1839-1847	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). This study measured adverse effects of Pirimicarb and Thiamethoxam on some biological and demographic parameters of *H. variegate.* Toxicity was assessed on the third instar larvae of *H. variegata,* using a contact method: the test solution was applied on the dorsal abdomen of the larvae using micropipette. The concentration that caused 30% mortality in the third instar larvae was 251.3 mg a.s./L. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	529
CA 8.3.2	Thiruchchelvan, N.; Mikunthan, G.; Thirukkumaran, G.; Pakeerathan, K.	2013	Effect of insecticides on bio-agent *Trichoderma harzianum rifai* under in vitro condition.	American-Eurasian Journal of Agricultural & Environmental Sciences (2013), Volume 13, Number 10, pp. 1357-1360	Does not fulfil criteria 5 (Several dose levels tested). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) An in-vitro study with Actara 25 V.G (Thiamethoxam (25%) SP) with *Trichoderma harzianum* was conducted utilizing a poison food technique. Only one concentration was tested (recommended field rate). As this study method differs from the standard methods and only one concentration was tested, this study was considered not relevant for this review.	531

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Antigo, M. de R.; Oliveira, H. N. de; Carvalho, G. A.; Pereira, F. F.; de R. Antigo, M.; de Oliveira, H. N.	2013	Repellence of pesticides used in sugarcane and their effects on the emergence of *Trichogramma galloi*. Repelencia de produtos fitossanitarios usados na cana-de-acucar e seus efeitos na emergencia de *Trichogramma galloi*.	Revista Ciencia Agronomica (2013), Volume 44, Number 4, pp. 910-916	Does not fulfil criteria 5 (Several dose levels tested). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The repellence of thiamethoxam alone and in combination with lambda-cyhalothrin was evaluated on adults of the parasitoid *T. galloi*, and its effect on the emergence of their offspring. The study method differs from the standard methods used for Tier I and II toxicity testing: e.g. eggs dipped in test solution, which is not a recommended exposure scenario and only one concentration of thiamethoxam was tested. Therefore this study was considered not relevant in this review.	541
CA 8.3.2	Shankarganesh, K.; Bishwajeet Paul; Gautam, R. D.; Paul, B.	2013	Studies on ecological safety of insecticides to egg parasitoids, *Trichogramma chilonis* Ishii and *Trichogramma brasiliensis* (Ashmead).	National Academy Science Letters (2013), Volume 36, Number 6, pp. 581-585	Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and enpoints are robust). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The adults of the egg parasitoids, *Trichogramma chilonis* and *T. brasiliensis*, were exposed to nine different insecticides (including thiamethoxam) by glass vial residue method. *T. chilonis:* LC_{50} = 0.012 mg a.s./L *T. brasiliensis:* LC_{50} = 0.011 mg a.s./L According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). Furthermore, no information is given on the concentration range tested. As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	548

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Rahmani, S.; Bandani, A. R.; Sabahi, Q.	2013	Effects of thiamethoxam in sublethal concentrations, on life expectancy (ex) and some other biological characteristics of *Hippodamia variegata* (Goeze) (Coleoptera: Coccinellidae).	International Research Journal of Applied and Basic Sciences (2013), Volume 4, Number 3, pp. 556-560,	In this study, influence of sublethal concentrations (LC_{10} and LC_{30}) of thiamethoxam was evaluated on life parameters of the beneficial insect *Hippodamia variegata*. For assessing the toxicity, one microliter of solution was applied on the dorsal abdomen of the third instar larvae. No method references were provided. Mortality and development were checked every 24 hours. After the emergence of adults, males and females were paired and checked daily in order to record their survival and their laid eggs. The experiments continued until the death of all the individuals. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 4 for thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	552

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Szczepanik, M.; Templin, J.; Napiorkowska , T.	2013	Susceptibility of developmental stages of the spider, *Tegenaria atrica* C.L. Koch to selected insecticides from different chemical groups. Wrazliwosc stadiow rozwojowych pajaka katnika wiekszego, *Tegenaria atrica* C.L. Koch na wybrane insektycydy z roznych grup chemicznych.	Progress in Plant Protection (2013), Volume 53, Number 3, pp. 519-523	English version of publication unavailable (Polish); English abstract available. The susceptibility of larvae and nymphs of the spider *Tegenaria atrica* C.L. Koch was studied under laboratory conditions. Actara WG 25 was applied by contact (insecticide-treated filter paper) or spray method at 0.05, 0.1 and 0.2% concentration. Larval mortality: at 0.05% and 2-fold higher doses, mortality does not exceed 25%. Nymph mortality: survival at 87.14-92.1%. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is thus considered not relevant for this review.	553
CA 8.3.2	Nage, S. M.; Devi, A. R.; Kumar, G. S.; Akare, U. S.	2013	Effect of different seed treatments on occurence of natural enemies in soybean ecosystem.	International Journal of Plant Protection (2013), Volume 6, Number 2, pp. 432-435,	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (appropriate sampling regime)). The effect of Actara 25 WG and 35 FS as seed treatment on germination percentage of soybean and on the population of natural enemies (coccinellids, chrysopa and spiders) was investigated. The study was undertaken under field condition in Nagpur, India. Thus, the geoclimatic conditions were not appropriate and relevant for Europe. Furthermore, not enough information on methods regarding observations of natural enemies is given. Therefore, this reference is not considered relevant for this review.	559

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Ayubi, A.; Moravvej, G.; Karimi, J.; Jooyandeh, A.	2013	Lethal effects of four insecticides on immature stages of *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) in laboratory conditions.	Tuerkiye Entomoloji Dergisi (2013), Volume 37, Number 4, pp. 399-407	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The lethal effects of thiamethoxam on the eggs and 1st instar larvae of *C. carnea* were studied in laboratory conditions. Dipping bioassay tests (not a standard method) were used for eggs and the residual contact method (test solution was applied on petris dishes) for larvae. Egg toxicity: LC_{50} = 1.9 µg a.s./L Larvae toxicity: LC_{50} = 0.55 µg a.s./L According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is thus considered not relevant for this review.	564
CA 8.3.2	Jadhav, D. S.; Shukla, A.	2013	Relative toxicity of some insecticides to *Coccinella transversalis* (F.).	Indian Journal of Entomology (2013), Volume 75, Number 4, pp. 301-303	Does not fulfil criteria 5 (Several dose levels tested). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) A study on the relative toxicity of insecticides on eggs, larvae and adults of *Coccinella transversalis* (F.) of 0.005% thiamethoxam tested as Actara 25% WG was conducted. A dry film method using petri dishes was used. Larval mortality after 48 hours: 35% at 0.005% thiamethoxam. Only one concentration was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	572

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Alexander, A.; Krishnamoorthy, S. V.; Kuttalam, S.	2013	Risk assessment of insecticides against non-target beneficials including natural enemies of papaya mealybug, *Paracoccus marginatus* Williams and Granara de Willink.	Journal of Insect Science (Ludhiana) (2013), Volume 26, Number 2, pp. 241-245	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Toxicity studies were carried out under laboratory conditions to assess the relative toxicity of thiamethoxam and its risk to non-target beneficials including natural enemies of papaya mealy bug, viz. *Cryptolaemus montrouzieri*, *Scymnus coccivora* and *Apis cerana* indica. A dry film contact toxicity method using filter paper was used which is not a recommened test procedure in standard testing. LC_{50} = 155.6 ppm According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. ppm). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	585
CA 8.3.2	Rahmani, S.; Bandani, A. R.	2013	Sublethal concentrations of thiamethoxam adversely affect life table parameters of the aphid predator, *Hippodamia variegata* (Goeze) (Coleoptera: Coccinellidae).	Crop Protection (2013), Volume 54, pp. 168-175.	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). In this study the effect of sublethal (LC_{10} and LC_{30}) concentrations of thiamethoxam on the demographic parameters of *Hippodamia variegate* were assessed and the data analyzed using the age-stage, two-sex life table procedure. A topical application method was used which is not a recommened test procedure in standard testing. Toxicity to third instars: LC_{10} = 48.2 mg a.s./L LC_{30} = 251.3 mg a.s./L LC_{50} = 788.5 mg a.s./L According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	604

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mukesh, N.; Kumawat, K. C.; Meenu Choudhary; Jat, R. G.; Nitharwal, M.; Choudhary, M.	2013	Influence of biorational and conventional insecticides on the population of *Chrysoperla carnea* (Steph.) in green gram, *Vigna radiata* (Linn.) in semi-arid conditions.	Biopesticides International (2013), Volume 9, Number 1, pp. 83-87	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Field studies were conducted in India to assess the effect of thiamethoxam formualted as Actara on the predator *Chrysoperla carnea* in the semiarid agro ecosystem of Rajasthan. As this field study was conducted in India, geoclimatic conditions were not appropriate and relevant for Europe and therefore, this reference is not considered relevant for this review.	621
CA 8.3.2	Halappa, B.; Awaknavar, J. S.; Archana, D.	2013	Safety evaluation of few insecticides against green lace wing, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) under laboratory condition.	Journal of Entomological Research (2013), Vol. 37, Number 1, pp. 73-77	Does not fulfil criteria 5 (Several dose levels tested, at leat 3, to establish a dose response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) The effects of thiamethoxam on different age classes of *C. carnea* were examined using a diet contamination method (0.2 g/L) and a dried residue method (coated glass vials, 0.2 g/L). While exposure via contaminated food is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. This exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. With regard to the tests using dry film method, according to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). Therefore this study is considered as supplemental information only.	622

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Anjum, S.; Iqbal, J.; Arshad, M.; Gogi, M. D.; Arif, M. J.; Tajammal, S.	2013	Comparative efficacy of insecticides as seed treatment against wheat aphid and its coccinellid predator.	Pakistan Entomologist (2013), Volume 35, Number 1, pp. 17-22	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). The efficacy of Actara 25 WG as seed treatment against wheat aphids and its coccinellid predator (*Coccinella septempunctata* L.) was evaluated at their recommended field doses under field conditions in Pakistan. The geoclimatic conditions were not appropriate and relevant for Europe and therefore, this reference is not considered relevant for this review.	646
CA 8.3.2	Pandi, G. G. P.; Bishwajeet Paul; Shah Vivek; Shankarganesh, K.; Paul, B.; Vivek, S.	2013	Relative toxicity of insecticides against coccinellid beetle, *Cheilomenes sexmaculata* (Fabricius).	Annals of Plant Protection Sciences (2013), Volume 21, Number 1, pp. 17-20	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) Relative toxicity of Actara 25 WG against of *C. sexmaculata* was determined in the laboratory by feeding *C. sexmaculata* with treated aphids. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	647
CA 8.3.2	Souza, C. R.; Sarmento, R. A.; Venzon, M.; Barros, E. C.; Santos, G. R. dos; Chaves, C. C.; dos Santos, G. R.	2012	Impact of insecticides on non-target arthropods in watermelon crop.	Semina: Ciencias Agrarias (Londrina) (2012), Volume 33, Number 5, pp. 1789-1801	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). In this study the effect of thiamethoxam on the soil surface and watermelon canopy arthropod community in Brazil was investigated. The geoclimatic conditions were not appropriate and relevant for Europe and therefore, this reference is not considered relevant for this review.	665

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Zhao JianWei; Zheng Yu; Li LiNa; He YuXian; Weng QiYong; Zhao, J. W.; Zheng, Y.; Li, L. N.; He, Y. X.; Weng, Q. Y.	2012	Toxicity of various classes of insecticides to *Serangium japonicum*, a predator of *Bemisia tabaci*.	Chinese Journal of Applied Entomology (2012), Volume 49, Number 6, pp.1577-1583	English version of publication unavailable (Chinese); English abstract available. Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). The toxicity of thiamethoxam to *Serangium japonicum* Chapin was tested by contact with dry residues on glass tubes. The recommended field rate was tested. Thiamethoxam caused 100% mortality at the recommended dose and at 50.06% of the recommended dose. As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	666
CA 8.3.2	Rishi Kumar; Kranthi, S.; Nitharwal, M.; Jat, S. L.; Monga, D.; Kumar, R.	2012	Influence of pesticides and application methods on pest and predatory arthropods associated with cotton.	Phytoparasitica (2012), Vol. 40, Number 5, pp. 417-424	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details, appropriate sampling scheme)). The primary aim of this study was to evaluate the difference in incidence of sucking pests in Bt and non-Bt cotton and the efficacy of two different methods of application of insecticides against the sucking pests complex of cotton and their adverse impact on the generalist predators active in cotton. Information regarding the number of sampling occasions for natural predators is not sufficient for evaluation of the study. The field study was conducted in India. Thus, the geoclimatic conditions are not appropriate and relevant for Europe. Therefore, this reference is not considered relevant for this review.	686

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Vivek S.; Bishwajeet P.; Pandi, G. G. P; Shankarganesh, K.	2012	Relative toxicity of insecticides on larval stages of green lacewing, *Chrysoperla sp*. (carnea-group) (Chrysopidae: Neuroptera).	Indian Journal of Entomology (2012), Volume 74, Number 4, pp. 394-397	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) Laboratory studies were conducted to examine the toxicity of thiamethoxam on larvae of *Chrysoperla* sp. (*carnea*-group) by a diet contamination method. While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	688

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mainali, B. P.; Kim, S. W.; Lim, U. T.	2012	Effects of combining releases of non-viable host eggs with insecticide application on *Riptortus pedestris* population and its egg parasitoids.	Journal of Asia-Pacific Entomology (2012), Volume 15, Number 2, pp. 299-305	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details, appropriate sampling regime)). Does not fulfil criteria 17 (Study conditions should not differ significantöly from recommended protocols) In this study a cage exclusion design was used to test the compatibility of combining releases of refrigerated host eggs with thiamethoxam application in soybean. The field study was conducted in Korea. Thus, the geoclimatic conditions arer not appropriate and relevant for Europe . Furthermore no information regarding the test field history are available. Information on the sampling regime are restricted and the method (whole plant counts were taken every 6 days on three randomly selected plants in each plot) does not fit with sampling methods from standard field studies. Therefore, this reference is not considered relevant for this review.	702

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Khan, M. A.; Ahmad-ur-Rahman, S.; Khan, I. A.; Saeed Khan; Qamar Zeb; Muhammad, S.; Manzoor, M.; Khan, S. Z.; Shah, S. F.; Muhammad, Sm; Zell-e-Huma; Baharullah K.	2012	Toxicity of foliar insecticides to syrphidfly predator of green peach aphid, *Myzus persicae* (Sulzer) on potato varieties.	Sarhad Journal of Agriculture (2012), Volume 28, Number 2, pp. 291-296	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details)). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) Adverse effects on syrphidfly 'syrphidae' populations were investigated in a field study conducted in Pakistan. Populations of syrphidfly were recorded on 1, 2, 3, 10 and 18 days of the post spray on ten plants per experimental unit while walking in a predetermined pattern through the field. The geoclimatic conditions are not appropriate and relevant for Europe. Furthermore no information regarding the test field history are available. The method employed to sample non-targets is not comparable to sampling methods of standard field studies. Therefore, this reference is not considered relevant for this review.	704

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Muhammadm A. K.; Ahmad-ur-Rahmanm S,; Khan, I. A.; Qamar, Z.; Muhammad, S.; Manzoor, M.; Saeed, K.; Sana, Z.; Shah, S. F.; Muhammad Saleem; Zell-e-Huma; Awan, H. U.	2012	Toxicity of foliar insecticides to ladybird beetle predator of green peach aphid, *Myzus persicae* (Sulzer) on potato varieties.	Sarhad Journal of Agriculture (2012), Volume 28, Number 2, pp. 283-290	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details)). Adverse effects on ladybird beetle populations were investigated in a field study conducted in Pakistan. Populations of Ladybird beetle were recorded on 1, 2, 3, 10 and 18 days of the post spray on ten plants per experimental unit while walking in a predetermined pattern through the field. The geoclimatic conditions are not appropriate and relevant for Europe. Furthermore no information regarding the test field history are available. The method employed to sample non-targets is not comparable to sampling methods of standard field studies. Therefore, this reference is not considered relevant for this review.	705
CA 8.3.2	Lanzoni, A; Sangiorgi, L; Luigi, V de; Consolini, L; Pasqualini, E; Burgio, G; de Luigi, V	2012	Evaluation of chronic toxicity of four neonicotinoids to *Adalia bipunctata* L. Coleoptera: Coccinellidae) using a demographic approach.	IOBC/WPRS Bulletin (2012), Volume 74, pp. 211-217	This article is an abstract published from a conference (Proceedings of the IOBC/WPRS Working Group "Integrated Protection of Fruit Crops", Subgroups "Pome-Fruit Arthropods" and "Stone Fruits" on "Sustainable protection of fruit crops in the Mediterranean area", Vico del Gargano, Italy, 12-17 September, 2010). This article is not sufficiently detailed for assessment and is therefore not considered relevant.	715

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	El-Zahi, E. S.	2012	Selectivity of some pesticides for various stages of *Chrysopeda carne*a (Steph.) using different methods of exposure.	Egyptian Journal of Biological Pest Control (2012), Volume 22, Number 2, pp. 211-216	Does not fulfil criteria 5 (Several dose levels tested, at least 3, to establish a dose-response.) Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). Thiamethoxam was evaluated against green lacewing, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) through a laboratory bioassay. *C. carnea* eggs were immersed in pesticide solutions, while second instar larvae were exposed using leaf dipping and thin film techniques using petri dishes, which are not standard methods. Only the recommended field rate was tested and the temperature in the bioassay was higher than recommended in standard protocols. As these study methods differs from the standard methods used for Tier I and II toxicity testing and no direct effects of non-target arthropods were investigated, this study is considered not relevant for this review.	716

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Seagraves, M. P.; Lundgren, J. G.	2012	Effects of neonicitinoid seed treatments on soybean aphid and its natural enemies.	Journal of Pest Science (2012), Volume 85, Number 1, pp. 125-132	Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) This study examines the effects of a thiamethoxam seed treatment on insect populations (pest and natural enemies) in soybeans over 2 years. Laboratory experiments were conducted to determine the duration that seed treatments remained effective against the soybean aphid (*Aphis glycines*, Hemiptera: Aphididae) and how thiamethoxam affected survival of the predator *Orius insidiosus*. The exposure route used in the laboratory study is environmentally relevant, but is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 4 thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations. In the field studies, effects on non targets was only monitored for approximately 2 months. Therefore, populations were not monitored long enough to possibly demonstrate recovery. Therefore this study is considered as supplemental information only.	742

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Avramova, K.; Grekov, D.; Ivanova, R.; Hristev, H.	2012	Some typical symptoms of mulberry silk worm poisoning with the neonicotinoid insecticides Confidor and Actara.	Scientific Papers, Series D. Animal Science (2012), Volume 55, pp. 107-108	Does not fulfil criteria 13 (Well defined test material (including purity/content)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate wheteher the study conclusions and endpoints are robust (e.g. pretreatmnet details, characterisation of physic-chemical parameters, replication, statistical methods and appropriate sampling scheme)). The effects of thiamethoxam on 4th and 5th instar larvae of silkworms were examined using a diet contamination method and dried residue method. As there is not given any information on the origin of the test substance or the test organisms and no information on the test field history are available, this reference is considered not relevant for this review.	748

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Wang, Y.; Yu, R.; Zhao, X.; An, X.; Chen, L.; Wu, C.; Wang, Q.	2012	Acute toxicity and safety evaluation of neonicotinoids and macrocyclic lactiones to adult wasps of four *Trichogramma* species (Hymenoptera: Trichogrammidae).	Acta Entomologica Sinica (2012), Vol. 55, Number 1, pp. 36-45	Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). To investigate the side effects of insecticides (including thiamethoxam) on trichogrammatid wasps, acute toxicities to adult *Trichogramma japonicum* Ashmead, T*richogramma ostriniae* Pang et Chen, *Trichogramma confusum* Viggiani and *Trichogramma evanescens* Westwood were determined by dry film residue method (in finger shaped tubes) under laboratory conditions. *Trichogramma japonicum*: LC_{50} = 0.40 mg a.s./L; *Trichogramma ostriniae*: LC_{50} = 2.47 mg a.s./L; *Trichogramma confusum:* LC_{50} values of 0.24 mg a.s./L; *Trichogramma evanescens*: LC_{50} = 1.12 mg a.s./L According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	768

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Redoan, A. C. M.; Carvalho, G. A.; Cruz, I.; Figueiredo, M. de L. C.; Silva, R. B. da; de L. C. Figueiredo, M.; da Silva, R. B.	2012	Selectivity of insecticides used in the control of *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) for eggs and nymphs of *Doru luteipes* (Scudder) (Dermaptera: Forficulidae). Seletividade de inseticidas utilizados no controle de *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) para ovos e ninfas de *Doru luteipes* (Scudder) (Dermaptera: Forficulidae).	Revista Brasileira de Milho e Sorgo (2012), Volume 11, Number 1, pp. 25-34	Does not fulfil criteria 8 (Effects are related to single test item). The aim of this study was to evaluate the effects of several insecticides registered for the control of *S. frugiperda* in crop corn on eggs and first instars nymphs of the predator *D. luteipes*. The viability of eggs treated with insecticides and insect survival rate in each instar were determined after 96 hours. Thiamethoxam was tested in combination with lambda-cyhalothrin, only. Thus, this reference is considered not relevant for this review.	774

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Al-Kherb, W. A	2011	Field efficacy of some neonicotinoid insecticides on whitefly *Bemicia tabaci* (Homoptera: Aleyrodidae) and its natural enemies in cucumber and tomato plants in Al-Qassim region, KSA.	Journal of Entomology (2011), Volume 8, Number 5, pp. 429-439	Does not fulfil criteria 12 (Apprpriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, replication, appropriate sampling scheme)). The effect of thiamethoxam on the whitefly, *Bemicia tabaci* and its side effects on *Orius* sp., *Paederus alfierii*, *Coccinella undicempunctata* and *Chrysoperla carnea* (Steph.) was investigated under field conditions in Onyyzah governorate, Qassim region, Saudi Arabia. The geoclimatic conditions are not appropriate and relevant for Europe. Furthermore, no information regarding the test field history are available and no information is given on the sampling method of the arthropods. Therefore, this reference is not considered relevant for this review.	808

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Scarpellini, J. R.; Andrade, D. J. de; de Andrade, D. J.	2011	The effect of insecticides on the lady beetle *Cycloneda sanguinea* L. (Coleoptera, Coccinellidae) and on the aphid *Aphis gossypii* Glover (Hemiptera, Aphididae) on cotton plants.	Arquivos do Instituto Biologico (Sao Paulo) (2011), Volume 78, Number 3, pp. 393-399	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). The toxicity of thiamethoxam on the predatory coccinellid *Cycloneda sanguinea* and on its prey, the aphid *Aphis gossypii*, in the cotton crop was investigated in field and laboratory studies. Laboratory studies were conducted with one concentration of thiamethoxam only (200 g product/ha) by using a direct overspray or a dry film residue method in glass tubes. No statistical significant effects were found. While the overspray exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Field studies were conducted in Brazil and are therefore not relevant for European climatic conditions. This study is considered as supplemental information only.	813

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mohanasundaram, A.; Sharma, R. K.	2011	Effect of newer pesticide schedules on the population of *Earias vittella* (Fabricius) and its predators on okra.	Journal of Insect Science (Ludhiana) (2011), Volume 24, Number 3, pp. 280-290	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, replication, appropriate sampling scheme)). A field study was carried out to determine the efficacy of different treatment schedules against the fruit borer, *Earias vittella* (Fabricius) and its predators on okra, *Abelmoschus esculentus* in India. The geoclimatic conditions are not appropriate and relevant for Europe. The main focus of this study was the efficacy of thiamethoxam against the fruit borer. Limited information is given on the sampling method of the arthropods, and seems to be not comparable with sampling method from standard field studies. Therefore this reference is considered not relevant for this review.	827
CA 8.3.2	Mohanasundaram, A.; Sharma, R. K.	2011	Effect of newer pesticide schedules on the population of sucking pests and predators on okra.	Pesticide Research Journal (2011), Volume 23, Number 1, pp. 55-63	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting), appropriate application method and rates (exposure) to derive endpoints). A field study was carried out to determine the efficacy of different treatment schedules against major sucking pests, leafhoppers, whitefly and red spider mite and their predators on okra in India. The geoclimatic conditions are not appropriate and relevant for Europe. Thiamethoxam was applied as a seed treatment followed by spray applications of two products with other active substances. Effects can not be related to thiamethoxam unambiguously. Therefore this reference is considered not relevant for this review.	875

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Nakahira, K.; Kashitani, R.; Tomoda, M.; Kodama, R.; Ito, K.; Yamanaka, S.; Momoshita, M.; Arakawa, R.; Takagi, M.	2011	Systemic nicotinoid toxicity against the predatory mirid *Pilophorus typicus*: residual side effect and evidence for plant sucking.	Journal of the Faculty of Agriculture, Kyushu University (2011), Volume 56, Number 1, pp. 53-55	Does not fulfil criteria 5 (several dose level tested, at least 3). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Peppers were treated with 0.5% thiamethoxam tested as Actara 5 (1 g granules) during planting in a greenhouse to investigate the effect of the residual toxicity on *P. typicus* adults. Three growing points on the stem of each pepper plant were cut 14, 21, 28, and 35 d after planting and transferred to platic containers and *P. typicus* adults were added. Death of the insects was observed 1, 3, and 5 d after their transfer. While systemic exposure via contaminated plants is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. This exposure route would be accounted for in regulatory higher tier field studies. There are 4 thiamethoxam seed treatment application field studies that examine effects on and recovery of full fauna populations. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). Therefore this study is considered as supplemental information only.	876

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	More, S. A.; Patil, P. D.; Shinde, B. D.	2011	Safety of different insecticides to the green lace wing, *Mallada boninensis* Okomoto.	Journal of Plant Protection and Environment (2011), Volume 8, Number 1, pp. 21-25	Does not fulfil criteria 5 (Several dose levels tested, at leat 3, to establish a dose response). Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and enpoints are robust). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) Effects of thiamethoxam tested as Thiamethoxam 25 WG at 0.001% on adult emergence of *M. boninensis* was investigated via dry film method at the inner surface of plastic vials. Adult emergence: 78.99% Egg Mortality: 21.93% No information on laboratory conditions are given, only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	893

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Godoy, M. S.; Carvalho, G. A.; Carvalho, B. F.; Lasmar, O.	2010	Physiological selectivity of insecticides to two lacewing species. Seletividade fisiologica de inseticidas em duas especies de crisopideos.	Pesquisa Agropecuaria Brasileira (2010), Volume 45, Number 11, pp. 1253-1258	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (study conditions should not differ significantly from recommended protocols) The effect of thiamethoxam, tested as 200 WG (0.05%), on *C. externa* and *C. cubana (* at an age up to 24 hours) was tested via direct overspray. Adult mortality and oviposition capacity were evaluated in a period of eight weeks, as well as the viability of laid eggs. *C. cubana*: 100% mortality within 24 hours of application *C. externa:*22.7 % mortality within 24 hours of application and 100% effects on egg production. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route for predatory mites would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	903

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Al-Antary, T. M.; Ateyyat, M. A.; Abussamin, B. M.	2010	Toxicity of certain insecticides to the parasitoid *Diaeretiella rapae* (Mcintosh) (Hymenoptera: Aphidiidae) and its host, the cabbage aphid *Brevicoryne brassicae* L. (Homoptera: Aphididae).	Australian Journal of Basic and Applied Sciences (2010), Volume 4, Number 6, pp. 994-1000	Does not fulfil criteria 1 (Well defined test material (including purity/content)). Does not fulfil criteria 10 (study conditions should not differ significantly from recommended protocols) Residue bioassays were conducted to determine the toxicity of thiamethoxam against cabbage aphid and its parasitoid, *D. rapae* exposed using leaf dipping. LC_{50} = 14.08 ppm According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	907
CA 8.3.2	Naveed, M.; Salam, A.; Saleem, A.; Rafiq, M.; Hamza, A.	2010	Toxicity of thiamethoxam and imidacloprid as seed treatments to parasitoids associated to control Bemisia tabaci.	Pakistan Journal of Zoology (2010), Volume 42, Number 5, pp. 559-565	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, replication, appropriate sampling scheme)). Cotton seed were treated with thiamethoxam and sown in the field. Effects on the population of whitefly and parasitoids were recorded during 2003-2005 cotton seasons. As the field studies were conducted in Pakistan, the geoclimatic conditions are not appropriate and relevant for Europe. No further information on the pretreatment of the test fields are available. The main focus of the study was on efficacy against the whitefly and sampling of non target arthropods was not conducted according to methods used in standard field studies. Therefore this reference is considered not relevant for this review.	915

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Preetha, G.; Manoharan, T.; Kuttalam, S.	2010	Impact of chloronicotinyl insecticide, imidacloprid on egg, egg-larval and larval parasitoids under laboratory conditions.	Journal of Plant Protection Research (2010), Volume 50, Number 4, pp. 535-540	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). Thiamethoxam was tested as Thiamethoxam 25 WG at 25 g a.s./ha against three beneficial insects (*Trichogramma chilonis* Ishii, *Chelonus blackburni* Cameron and *Bracon hebetor* Say). The toxicity of thiamethoxam was evaluated by treating the parasitized eggs using an atomizer in the case of *T. chilonis* (parasitized egg cards were sprayed at a rate of 2.5 ml per mL) and glass scintillation vial residue bioassay method for the adults of C. *blackburni* and *B. hebetor*. *T. chilonis:* 93.81% adult emergence, 88.59% parasitization C. *blackburni:* 51.72 % mortality at 24 hr *B. hebetor:* 93.33 % mortality at 48 hr According to ESCORT II, the primary toxicity endpoint of the studies should be mortality, with determination of the LR_{50}. In this study only one concentration of thiamethoxam was tested, so no LC_{50} could be determined. As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	916

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Bhakray, R. B.; Thakre, S. M.; Aherkar, S. K.; Satpute, N. S.; Raut, B. T.	2010	Biosafety of systemic insecticides through seed treatment and stem smearing to some predators.	Journal of Maharashtra Agricultural Universities (2010), Volume 35, Number 3, pp. 488-489	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting). Does not fulfil criteria 16 (sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pre-treatment details, characterisation of physico-chemical parameters, statistical methods and appropriate sampling regime). Field studies were conducted in Maharashtra, India, during the 2006/07 kharif season, to evaluate the safety of thiamethoxam on the predators *Coccinella septempunctata*, *Chrysopa* sp. and spiders on cotton. As the field studies were conducted in India, the geoclimatic conditions are not appropriate and relevant for Europe. No detailed information on the origin of the test species, pre-treatment of the field, on application, sampling etc. is given. Therefore this reference is considered not relevant for this review.	919
CA 8.3.2	Schenke, D.; Baier, B.	2010	Diffusion of thiamethoxam and imidacloprid from coated sugarbeet seeds into the soil and its effects on carabid beetle larvae. Ausbreitung von Thiamethoxam und Imidacloprid aus pilliertem Zuckerruebensaat gut in den Boden und deren Auswirkungen auf Laufkaeferlarven.	Julius-Kuehn-Archiv (2010), Number 428, 462 p. ISSN: 1868-9892	This article is an abstract published from a conference (57. Deutsche Pflanzenschutztagung, Berlin, Germany, 6-9 Sep, 2010). The article is not sufficiently detailed for assessment and no data is presented. Therefore this is not considered relevant.	921

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Lima Junior, I. dos S. dos; Nogueira, R. F.; Bertoncello, T. F.; Melo, E. P. de; Suekane, R.; Degrande, P. E.; dos S. dos Lima Junior, I.; dos Lima Junior, I. dos S.; de Melo, E. P.	2010	Selectivity of pesticides over predators of cotton plant pests. Seletividade de inseticidas sobre o complexo de predadores das pragas do algodoeiro.	Pesquisa Agropecuaria Tropical (2010), Volume 40, Number 3, pp. 347-353	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). The effects of Thiamethoxam 250 WG (200 g/ha) on predators and their natural enemies were investigated in field studies in cotton in Brazil. The evaluations were carried out prior to application up to 33 days after application. Samples were collected using the cloth beat method. As the field study was conducted in Brazil, the geoclimatic conditions are not appropriate and relevant for Europe. Therefore this reference is considered not relevant for this review.	929

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Scarpellini, J. R.; Andrade, D. J. de; de Andrade, D. J.	2010	Evaluation of the effect of insecticides on lady beetles *Hippodamia convergens* uerin-Meneville (Coleoptera: Coccinellidae) in cotton plant.	Arquivos do Instituto Biologico (Sao Paulo) (2010), Volume 77, Number 2, pp. 323-330	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 12 (Apprpriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) In this study, the effects of thiamethoxam as Actara 250 WG at 50 g a.s./ha on the coccinellid predator *Hyppodamia convergens*, were evaluated under laboratory and field conditions. Laboratory test were conducted either via direct overspray or introduction of the cocinellids after treatment in cages containing a cotton plants. For field studies, *H. convergens* were placed in the field in bags of tulle (voile) before and after treatment. Field studies were not conducted according to recommended test protocols and were conducted in Brazil and are therefore not relevant for European climatic conditions. Laboratory studies were conducted with one concentration of thiamethoxam only..While direct overspray as exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route for predatory mites would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	932

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Zotti, M.J.; Gruetzmacher, A.D.; Gruetzmacher, D.D.; Dalmazzo, G.O.; Martins, J.F.S.	2010	Selectivity of insecticides used in the corn crop to adults of *Doru lineare* (Eschscholtz, 1822) (Dermaptera: Forficulidae). Seletividade de inseticidas usados na cultura do milho para adultos de Doru lineare (Eschscholtz, 1822) (Dermaptera: Forficulidae).	Arquivos do Instituto Biologico (Sao Paulo) (2010), Volume 77, Number 2, pp. 291-299	Does not fulfil criteria 8 (Effects are related to single test item). Eggs, nymphs and adults of *D. lineare* were placed on glass plates or fed on *Spodoptera frugiperda* eggs, both containing previously sprayed insecticide residues. Thiamethoxam was tested as Enego Pleno at 0.25 kg product/ha, a formulation containing lambda-cyhalothrin and thiamethoxam. As thiamethoxam was tested only in a formulation also containing lambda-cyhalothrineffects cannot unequivocally be related to thiamethoxam and therefore this refernece is considered not relevant for this review.	936
CA 8.3.2	Khajuria, D. R.; Gupta, D.; Sharma, J. P.; Editor(s): Verma, A. K.; Bhardwaj, S. P.; Gupta, P. R.	2010	Bio-efficacy of insecticides against aerial form of the woolly apple aphid, *Eriosoma lanigerum* (Hausmann) and their safety to the parasitoid, *Aphelinus mali* (Haldemann).	Pest Management and Economic Zoology (2010), Vol. 18, Number 1/2, pp. 225-228	This article was not available from our copyright-cleared document delivery sources. While we placed an order to purchase this document from an advanced reference source, we were unable to obtain this article. The efficacy of thiamethoxam against the aerial form of the woolly apple aphid, *Eriosoma lanigerum* (Hausmann), and its safety to the parasitoid, *Aphelinus mali* (Haldemann) was evaluated during September-October 2000 to 2002 in apple orchards in Kullu valley of Himachal Pradesh. As the field study was conducted in India, the geoclimatic conditions are not appropriate and relevant for Europe. Therefore this reference is considered not relevant for this review.	940

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Muthuswami, M.; Indumathi, P.; Krishnan, R.; Thangamalar, A.; Subramanian, S.	2010	Impact of chemicals used for thrips control on silkworm, *Bombyx mori* L.	Karnataka Journal of Agricultural Sciences (2010), Volume 23, Number 1, pp. 144-145	This article was not available from our copyright-cleared document delivery sources. While we placed an order to purchase this document from an advanced reference source, we were unable to obtain this article. Field trials were conducted during 2007 in Coimbatore, Tamil Nadu, India to evaluate the effects of thiamethoxam 0.025%, on the silkworm, *Bombyx mori*. As the field study was conducted in India, the geoclimatic conditions are not appropriate and relevant for Europe. Therefore this reference is considered not relevant for this review.	952
CA 8.3.2	Gorzka, D.; Olszak, R. W.	2010	Insecticide selectivity tests on spider mite destroyer (*Stethorus punctillum*) (Weise) (Coleoptera: Coccinellidae) in laboratory conditions.	IOBC/WPRS Bulletin (2010), Volume 55, pp. 109-112	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) Toxicity of thiamethoxam to adult ladybirds (*S. punctillum)* were tested via direct overspray with thiamethoxam (Actara 25 WG at 50 g a.s./ha). Lethal effects of thiamethoxam (Actara 25WG) reached 10% after 72h. Only one concentration of thiamethoxam was tested. While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route for predatory mites would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	961

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Noubar, J.; Bostanian, N. J.; Hardman, J. M.; Thistlewood, H. A.; Racette, G.	2010	The response of Neoseiulus fallacis (Garman) and *Galendromus occidentalis* (Nesbitt) (Acari: Phytoseiidae) to six reduced risk insecticides in Canada.	IOBC/WPRS Bulletin (2010), Volume 55, pp. 73-77	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) Effects of thiamethoxam on survival of adults mites *N. fallacis* and *G. occidentalis*were tested. Mites were sprayed with one concentration of thiametoxam (Actara 25 WG). Only one concentration of thiamethoxam was tested. While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route for predatory mites would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only and does not alter the existing risk assessment.	982

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Baldessari, M.; Malagnini, V.; Tolotti, G.; Angeli, G.	2010	Impact of neonicotinoid insecticides on beneficial phytoseiid mites. Insetticidi neonicotinoidi, quale l'impatto sugli acari fitoseidi utili.	Informatore Agrario (2010), Volume 66, Number 45, pp. 67-70	Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whethter the studyconclusions and endpoints are robust). Does not fulfil criteria 10 and 17 (Study conditions should not differ significantly from recommended protocols). The effects of thiamethoxam (formualetd as Actara 25 WG) on *Amblyseius andersoni* was tested in the laboratory at 37.5 g/hL and in apple and grapevine field studies at 27 and 13.3 g/hL, respectively. Thiamethoxam was slightly harmful in all tests at the given test rates. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. g/hL). In the laboratory, only one concentration was tested, therefore NOEC and LR_{50} values could not be determined. The methods used in the studies were not stated in this article, and therefore could not be evaluated. Therefore this study is considered as supplemental information only.	985

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Patel, J. K.; Patel, I. S.; Patel, G. M.	2010	Effect of Spirotetramat and Imidacloprid on survival of natural enemies of sucking pests in cotton crop.	Trends in Biosciences (2010), Volume 3, Number 1, pp. 37-38	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate wheteher the study conclusions and endpoints are robust (e.g. pretreatmnet details, characterisation of physic-chemical parameters, replication, statistical methods and appropriate sampling scheme)). Field trials were conducted during 2007 – 2009 in India to evaluate the effects of Thiamethoxam 25 WG on survival of natural enemies of sucking pests in cotton. As the field study was conducted in India, the geoclimatic conditions are not appropriate and relevant for Europe. No detailed information on field history, application or sampling regime are available. Therefore this reference is considered not relevant for this review.	1003

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Kumar, P. K. V.; Vasudev, V.; Seetharama, H. G.; Irulandi, S.; Sreedharan, K.	2010	Effect of insecticides on *Spalgis epius.*	Journal of Coffee Research (2010), Volume 38, Number 1/2, pp. 11-28	Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) Actara 25 WG (thiamethoxam) was tested for its toxicity on the coffee mealy bug *P. citri* and the indigenous Lycaenid predator *S. epius* at rates of 0.005, 0.01125 and 0.0225 % a.s. Eggs, 3rd instar caterpillars and adults of *S. epius* were exposed via direct overspray. Additionaly, a field study was conducted in coffee plants. The observations were recorded until 31 days after spray. For laboratory studies, the exposure route used is environmentally relevant, but is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar spray field studies that examine effects on and recovery of full fauna populations. In the field studies, effects on non targets was only monitored for approximately 1 month. Therefore, populations were not monitored long enough to possibly demonstrate recovery. Therefore this study is considered as supplemental information only.	1014

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	; Hegde, M.; Nidagundi, J.	2009	Effect of newer chemicals on planthoppers and their mirid predator in rice.	Karnataka Journal of Agricultural Sciences (2009), Volume 22, Number 3, pp. 511-513, 8 refs	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, appropriate sampling scheme)). Field trials were conducted for two consecutive seasons at the Agricultural Research Station in Siruguppa, India. Thiamethoxam was applied twice (15 day interval). The predatory mirid bug (*C. lividipennis*) population was recorded at ten days after application. No further information on the test substance, test conditions in the field or pretreatments of the test field are given; the field study was conducted in India, the geoclimatic conditions are not appropriate and relevant for Europe. Data on non-target arthropods were recorded in a manner not comparable to standard methods. Therefore this reference is considered not relevant for this review.	1055

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Sun DingWei; Su JianYa; Shen JingLiang; Xu JianTao; Sun, D. W.; Su, J. Y.; Shen, J. L.; Xu, J. T.	2008	Safety evaluation of insecticides to *Cyrtohinus lividipennis* (Reuter) (Hemiptera: Miridae), a predator of *Nilaparvata lugens* (Stal) (Homoptera: Delphacidae).	Scientia Agricultura Sinica (2008), Volume 41, Number 7, pp. 1995-2002	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). A dry film residue method (coated glass vials), a rice spraying method (inects were introduced after spray) and a rice stem dipping method were used to test the safety of several insecticides to *Cyrtorhinus lividipennis* under laboratory conditions. Dry film method LC_{50}: 0.507 mg a.s./L. For the rice spraying method and rice stem dipping method and application rate of 20 mg a.s./L was used. Mortality rates of 95%, 95.0%, 81.7% and 71.7% were observed for 1, 4, 7 and 10 days after spray application, respectively. Mortality after stem dipping was 100%. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). Furthermore, as the study methods differ from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	1174

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Moser, S. E.; Obrycki, J. J.	2009	Non-target effects of neonicotinoid seed treatments; mortality of coccinellid larvae related to zoophytophagy.	Biological Control (2009), Volume 51, Number 3, pp. 487-492	Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). In this study, *H. axyridis* larvae were exposed for 360 min to corn seedlings that had been grown from seeds treated with thiamethoxam and toxicity was recorded. While this exposure route is environmentally relevant, it is not considered in testing conducted for the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 4 thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations.Therefore this study is considered as supplemental information only.	1057

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Silva, M. Z. da; Oliveira, C. A. L. de; Sato, M. E.; da Silva, M. Z.; de Oliveira, C. A. L.	2009	Selectivity of the pesticides to the predaceous mite *Agistemus brasiliensis* Matioli, Ueckermann & Oliveira (Acari: Stigmaeidae). Seletividade de produtos fitossanitarios sobre o acaro predador *Agistemus brasiliensis* Matioli, Ueckermann & Oliveira (Acari: Stigmaeidae).	Revista Brasileira de Fruticultura (2009), Volume 31, Number 2, pp. 388-396	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Studies were conducted to evaluate the effect of thiamethoxam at a rate of 20 g/100 L on the predaceous mite *Agistemus brasiliensis* (Acari: Stigmaeidae) under laboratory conditions via direct overspray. Mortality = 65.1%. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1067

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Pratissoli, D; Vianna, U. R; Furtado, G. O; Zanuncio, J. C; Polanczyk, R. A.; Barbosa, W. F.; Carvalho, J. R. de; de Carvalho, J. R.	2009	Insecticide selectivity to *Trichogramma pretiosum* Riley (Hymenoptera: Trichogrammatidae) in different hosts. Seletividade de inseticidas a *Trichogramma pretiosum* Riley (Hymenoptera: Trichogrammatidae) em diferentes hospedeiros.	Boletin de Sanidad Vegetal, Plagas (2009), Volume 35, Number 3, pp. 347-353	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 8 (a quantitative relationship exists between reported endpoints and risk assessment endpoints of growth, mortality behaviour or reproduction rate). The effects of thiamethoxam on adults and immatures of *Trichogramma pretiosum* were studied using the following hosts: *Anagasta kuehniella, Sitotroga cerealella and Anticarsia gemmatalis*. Eggs of the three hosts were glued in cardboard, treated with thiamethoxam, after which they were exposed to parasitism. While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is not considered relevant for this review.	1072
CA 8.3.2	Preetha, G.; Stanley, J.; Suresh, S.; Kuttalam, S.; Samiyappan, R.	2009	Toxicity of selected insecticides to *Trichogramma chilonis*: assessing their safety in the rice ecosystem.	Phytoparasitica (2009), Volume 37, Number 3, pp. 209-215	Does not fulfil criteria 10 (study conditions should not differ significantly from recommended protocols). Thiamethoxam was tested to determine its toxicity to the parasitoid *Trichogramma chilonis* using an insecticide-coated vial (scintillation) residue bioassay. LC_{50} = 0.001 mg a.s./L According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	1091

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Kumaran, N.; Kumar, B. V.; Boomathi, N.; Kuttalam, S.; Gunasekaran, K.	2009	Non-target effect of ethiprole 10 SC to predators of rice planthoppers.	Madras Agricultural Journal (2009), Vol. 96, Number 1/6, pp. 208-212	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, replication, appropriate sampling scheme). Thiamethoxam 25WG was tested as positive control at 25 g a.s./ha under field conditions for its effect on non-target organisms, insect predators, *Cyrtorrhinus lividipennis* and *Paederus fuscipes* found in rice ecosystem. Observation on the populations of *C. lividipennis* and *P. fuscipes* was recorded in ten hills per plot before application and 7 and 14 days after application. As the field study was conducted in India, the geoclimatic conditions are not appropriate and relevant for Europe. Furthermore no information on pretreatment of the test field nor on the sampling scheme are available. Data on non-target arthropods were recorded in a manner not comparable to standard methods. Therefore this reference is considered not relevant for this review.	1092

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Thakare, S. M.; Bharti Dhoble; Thakare, A. S.; Dhoble, B.	2009	Effect of different chemicals applied by seed or stem smearing technique on natural enemies of Bt cotton.	Crop Research (Hisar) (2009), Volume 38, Number 1/3, pp. 205-207	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, replication, appropriate sampling scheme)). In this study effects of stem smearing and seed treatments of thiamethoxam against natural enemies on Bt cotton were investigated in field studies. The field study was conducted in India, therefore the geoclimatic conditions are not appropriate and relevant for Europe. Furthermore no information on pretreatment of the test field nor on the sampling scheme are available. Therefore this reference is considered not relevant for this review.	1093
CA 8.3.2	Ahire, R. V.; Bhosle, B. B.; Kadam, D. R.	2009	Impact of thiamethoxam as seed dresser on natural enemies of major pests of soybean (*Glycine max* (L.) Merill) and impact on yield.	Journal of Plant Protection and Environment (2009), Volume 6, Number 2, pp. 117-119	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, replication, appropriate sampling scheme)). In this study the effect of seed dressers containing thiamethoxam on yield and natural enemies of major pests in soybean were investigated under field conditions. The field study was conducted in India, therefore the geoclimatic conditions are not appropriate and relevant for Europe. Furthermore no information on pretreatment of the test field nor on the sampling scheme are available. Therefore this reference is considered not relevant for this review.	1114

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Agale, D. A.; Bhosle, B. B.; Kadam, D. R.	2009	Impact of different insecticides on population of natural enemies in Bt cotton and yield.	Journal of Plant Protection and Environment (2009), Volume 6, Number 2, pp. 105-107	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (e.g. pretreatment details, replication, appropriate sampling scheme)). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) This study was conducetd to examine the efficacy of different insecticides (including thiamethoxam) on thrips in Bt cotton and its effects on yield. The field study was conducted in India, therefore the geoclimatic conditions are not appropriate and relevant for Europe. Furthermore no information on pretreatment of the test field and limited information on the sampling scheme are available. The latter seems not to be consistent with methods of standard field studies (sampling only up to 7 days after treatment). Therefore this reference is considered not relevant for this review.	1117

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Shinde, C. U.; Patel, M. B.; Mehendale, S. K.	2009	Studies on relative toxicity of different insecticides on larvae of *Chrysoperla carnea* (Stephens).	Insect Environment (2009), Volume 15, Number 2, pp. 67-69	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust). Does not fulfil criteria 10 (study conditions should not differ significantly from recommended protocols) Thiamethoxam was tested on *C. carnea* under laboratory conditions at 0.005% via a dry film method: glass vials were dipped in the test solution and left drying. Larvae were introduced for 45 minutes and transferred afterwards. After 72 hours, a mortality of 29.64% was observed for the given test concentrations. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). Only one concentration was tested. As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	1133

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Wang, H. Y.; Yang, Y.; Su, J. Y.; Shen, J. L.; Gao, C. F.	2008	Assessment of the impact of insecticides on *Anagrus nilaparvatae* (Pang et Wang) (Hymenoptera: Mymanidae), an egg parasitoid of the rice planthopper, *Nilaparvata lugens* (Hemiptera: Delphacidae).	Crop Protection (2008), Volume 27, Number 3/5, pp. 514-522	Does not fulfil criteria 10 (study conditions should not differ significantly from recommended protocols) In this study, sublethal and lethal effects of thiamethoxam on adult *A. nilaparvatae* following contact (solutions were applied to scintillation vials), oral (via treated honey solution, 10 mg/L) or residual exposure (via rice plants sprayed until saturation) were investigated. LC_{50} contact = 0.52 mg a.s./L Oral toxicity: 100% mortaliy after 4 hours at 10 mg/L Residual exposure: 7 d mortality of 66.8%. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). While the oral exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1185

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	da Fonseca, P.R.B.; Bertoncello,T. F.; Ribeiro, J.F.; Fernandes, M.G.; Degrande, P.E.	2008	Selectivity of insecticides to natural enemies on soil cultivated with cotton.	Pesquisa Agropecuária Tropical, v. 38, n. 4, p. 304-309, out./dez. 2008	English version of publication unavailable (Portugues); English abstract available. Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). A field study with Tiametoxam 250 WG (0.2 kg/ha) was conducted in a soybean field in Brazil, investigating the side effects on Araneae, Formicidae and Tachinidae. Insecticides were introduced in the field at 7 days after emergence and samples were collected with traps type "modified pitfall" weekly up to 126 days after emergence. At day 133 after emergence applied the test solution was applied and observations were made at 1, 4, 7, 10, 13 and 16 days after application (DAA). Thiamethoxam 250 WG had the greatest effect on Formicidae and Tachinidae on the first day with mortality rates of 100% and 56% and was selective for Aranea. The field study was conducted in Brazil, the geoclimatic conditions are not appropriate and relevant for Europe. Additionally, the study was not carried out long enough to demonstrate potential recovery of the non-targets. Therefore this reference is considered not relevant for this review.	1195

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Hirekurubar, R.B.; Ambekar, J.S.	2008	Bio-efficacy of newer insecticides against shoot and fruit borer of okra and their impact on natural enemies	Crop. Res. 36 (1, 2 & 3): 302 – 307 (2008)	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (appropriate sampling scheme)). In this study effects of Thiamethoxam 25WG on shoot and stem borer of okra and their natural enemies (*Cocinella* sp. and *C. Carnea*) was investigated. The field study was conducted in India, therefore the geoclimatic conditions are not appropriate and relevant for Europe. Only limited information on the sampling scheme is given (made on 5 selected plants per plot before, 2, 7 and 15 days after each spray), which is not comparable to sampling in standard field studies. Therefore this reference is considered not relevant for this review.	1196
CA 8.3.2	Baldessari, M.; Maines, R.; Angeli, G. Editor(s): Lozzia, G. C.; Lucchi, A.; di Chiara, S. R.; Tsolakis, H.	2008	Comparison of two methods for the agrochemicals side effect evaluation on Phytoseiid mites in vineyards.	IOBC/WPRS Bulletin (2008), Volume 36, pp. 237-243	Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (appropriate sampling scheme)). Two assessment techniques, washing method and direct counting, were compared for estimating population size of the phytoseiids *Amblyseius andersoni* and *Kampimodromus aberrans* in vineyard. Applying the two techniques, trials on side effects of insecticides and estimation of phytoseiids on different grape variety were performed. Focus of the study was on comparison of the two sampling techniques. Information on application and field history are limited and therefore the reference is considered not relevant for this review.	1200

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mahmoud, A. M. A.; Lim, U. T.	2008	Ecotoxicological effect of fenitrothion on *Trissolcus nigripedius* (Hymenoptera: Scelionidae) an egg parasitoid of *Dolycoris baccarum* (Hemiptera: Pentatomidae).	Journal of Asia-Pacific Entomology (2008), Volume 11, Number 4, pp. 207-210	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (study conditions should not differ significantly from recommended protocols) The main focus of this study was assessment of ecotoxicological effects of fenitrothion on *T. nigripedius.* The acute toxicity of fenitrothion was compared with thiamethoxam toxicity by topical application, exposure to residues and oral ingestion at the field recommended rate. The endpoint derived from the study was the LT_{50}, the time (hours) it took for 50% of the test organisms to die. The endpoint derived from this study is not used in regulatory Tier I or II risk assessment for non-target arthropods. Therefore this study is considered as not relevant for this review.	1207

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Scarpellini, J. R.	2008	Selectivity of pesticides on lady beetles *Cycloneda sanguinea* (Linnaeus, 1763) (Coleoptera, Coccinellidae) in cotton plant. Seletividade fisiologica de aficidas sobre joaninha *Cycloneda sanguinea* (Linnaeus, 1763) (Coleoptera, Coccinellidae) em algodoeiro.	Arquivos do Instituto Biologico (Sao Paulo) (2008), Volume 75, Number 2, pp. 195-202	Does not fulfil criteria 3 (Applicable test species). Does not fulfil criteria 10 and 17 (Study conditions should not differ significantly from recommended protocols) Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). The effect of thiamethoxam 250 WG on *Aphis gossypii* and *Cycloneda sanguinea* ladybeetles in cotton plants was investigated in field, semi-field, greenhouse and laboratory studies conducted in Ribeirao Preto, Sao Paulo, Brazil. In field studies, tulle sachets containing the ladybugs were placed in the field before or after application, which is not a test procedure according to standard protocols. As the field studies were conducted in Brazil, the geoclimatic conditions are not appropriate and relevant for Europe, these are considered not relevant for this review. For semi-field studies, tulle sachets were left in the treated field for one hour and then transferred to cages with plants without treatment. In the laboratory studies and greenhouse studies, aphids were exposed via direct overspray and then fed to ladybeetles. While direct overspray of food sources is an environmentally relevant exposure route, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this reference is considered not relevant for this review.	1210

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Olszak, R. W.; Sekrecka, M.	2008	Influence of some insecticides and acaricides on beneficial mites and on *Coccinella septempunctata* (Coleoptera; Coccinellidae) larvae	IOBC/WPRS Bulletin (2008), Volume 35, pp. 101-108	This article is an abstract published from a conference (Proceedings of the IOBC/WPRS Working Group "Pesticides and Beneficial Organisms", Berlin, Germany, 10-12 October 2007.). Studies were conducted under laboratory and field conditions to assess the influence of insecticides (including thiamethoxam) on different beneficial mites and on *Coccinella septempunctata* larvae. The article is not sufficiently detailed for assessment and is therefore not considered relevant.	1227
CA 8.3.2	Bostanian, N. J; Laurin, M. C	2008	Effects of ten pesticides to *Anystis baccarum* (Acari: Anystidae).	IOBC/WPRS Bulletin (2008), Volume 35, pp. 96-100	This article is an abstract published from a conference (Proceedings of the IOBC/WPRS Working Group "Pesticides and Beneficial Organisms", Berlin, Germany, 10-12 October 2007). Laboratory toxicity studies were carried on with insecticides (including thiamthoxam) on *Anystis baccarum.* This article was not sufficiently detailed for assessment and is therefore not considered relevant.	1228

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Sun Chao; Su JianYa; Shen JinLiang; Zhang Xi; Sun, C.; Su, J. Y.; Shen, J. L.; Zhang, X.	2008	Laboratory safety evaluation of insecticides to *Trichogramma japonicum*.	Chinese Journal of Rice Science (2008), Volume 22, Number 1, pp. 93-98	English version of publication unavailable (Chinese); English abstract available. Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). The direct and indirect effects of thiamethoxam against *Trichogramma japonicum* Ashmead, an egg parasitoid of the rice stem borer, *Chilo suppressalis* Walker, were evaluated under laboratory conditions. Adults of *T. japonicum* were exposed to leaves with insecticide residues at 2 and 7 days after application. Additionally, parasitized host eggs were dipped into insecticidal solutions. As only one concentration of thiamethoxam was tested, this reference is considered as supplemental information only.	1229

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Farag, N. A.; Gesraha, M. A.	2007	Impact of four insecticides on the parasitoid wasp, *Diaertiella rapae* and its host aphid, *Brevicoryne brassicae* under laboratory conditions	Research Journal of Agriculture and Biological Sciences (2007), Volume 3, Number 5, pp. 529-533	Does not fulfil criteria 4 (Test organisms are not previously exposed to the test material or other contaminants). Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and enpoints are robust). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) Thiamethoxam (Actara 25%WG), was tested against the parasitoid wasp, *Diaertiella rapae* (McIntosh) and its host aphid, *Brevicoryne brassicae* (L). via direct overspray. *Diaertiella rapae:* LC_{50} = 0.098% Test organisms were collected from cabbage fields at Giza Governorate. No information is available on the history of the collection site regarding previous pesticide usage. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1264

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mhaske, B. M.; Pardeshi, S. R.; Bhoite, K. D.; Rasal, P. N.	2007	Biosafety of Coccinellid predators and chemical control of wheat aphids.	Agricultural Science Digest (2007), Volume 27, Number 4, pp. 264-266	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (appropriate sampling scheme). A field experiment was conducted in India to investigate the efficacy of Actara 25 WG against foliar aphids of wheat and its biosafety to coccinellid predators. As the field study was conducted in India, the geoclimatic conditions are not appropriate and relevant for Europe. No information on the preatrement history of the test field and only limited information on the sampling scheme is given. Sampling was not conducted according to standard methods for NTA field study (e.g. counts only up to 15 days after 2nd spray). Therefore this reference is considered not relevant for this review.	1280
CA 8.3.2	Choi, B. R;Lee, S. W; Park, H. M	2007	Selection of low toxic pesticides and residual toxicity to *Cotesia glomerata*.	Korean Journal of Applied Entomology (2007), Volume 46, Number 2, pp.	English version of publication unavailable (Korean); English abstract available. Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Leaf dipping method, body dipping method, and diet treatment method were set up for the toxicity evaluation of thiamethoxam against *Cotesia glomerata* adults. This is not a recommended test system according to standard methods and therefore this reference was considered not relevant for this review.	1281

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Muthukumar, M.; Sharma, R. K.; Sinha, S. R.	2007	Field efficacy of biopesticides and new insecticides against major insect pests and their effect on natural enemies in cauliflower.	Pesticide Research Journal (2007), Volume 19, Number 2, pp. 190-196	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (appropriate sampling scheme)). Field studies were conducted in New Delhi, India, to determine the efficacy of thiamethoxam against major insect pests and their effect on natural enemies in cauliflower. The field study was conducted in India, therefore the geoclimatic conditions are not appropriate and relevant for Europe. No information on the preatrement history of the test field and limited information on the sampling scheme is given. Sampling was not conducted comparable to standard methods of NTA field studies. Therefore this reference is considered not relevant for this review.	1285
CA 8.3.2	Ravi, S. K.; Manjunatha, M.; Naik, M. I.	2007	Toxic effect of insecticides on mortality of sugarcane woolly aphid, *Ceratovacuna lanigera* and its predator *Dipha aphidivora*.	Karnataka Journal of Agricultural Sciences (2007), Volume 20, Number 1, pp. 144-145	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (appropriate sampling scheme). Field experiments were conducted in India to determine the effect of insecticides (including thismethoxam) on sugarcane woolly aphid, *Ceratovacuna lanigera*, and its predator *Dipha aphidivora*. The field study was conducted in India, therefore the geoclimatic conditions are not appropriate and relevant for Europe. No information on the preatrement history of the test field or the sampling scheme is given. Therefore this reference is considered not relevant for this review.	1295

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Tamutis, V.; Ziogas, A.; Saluchaite, A.; Kazlauskaite, S.; Amsiejus, A.	2007	Epigeic beetle (Coleoptera) communities in summer barley agrocenoses.	Baltic Journal of Coleopterology (2007), Volume 7, Number 1, pp. 83-98	Does not fulfil criteria 15 (exposure route is clearly defined and suitably quantified) Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study coclusions and endpoints are robust (eg pretreatment details, characterisation of physico-chemical parameters. Replication, appropriate sampling scheme). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) A field study was conducted in Lithuania to examine the species composition, structure of dominance, abundance and activity density dynamics of epigeic beetles in different managed (Conventional: without insecticides, Conventional: with insecticide treatment, Ecological: without biopreparation treatment and Ecological: with preparation made on the basis of plant extraction treatment) summer barley fields. As this field study was only conducted for 2 months, no recovery could be demonstrated due to limited time. Information on the execution of the study is limited and therefore an evaluation of the study is not possible. Thus, this reference is considered not relevant for this review.	1311

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Gesraha, M. A.	2007	Impact of some insecticides on the coccinellid predator, *Coccinella undecimpunctata* L. and its aphid prey, *Brevicoryne brassicae* L.	Egyptian Journal of Biological Pest Control (2007), Volume 17, Number 1/2, pp. 65-69	Does not fulfil criteria 4 (Test organisms are not previously exposed to the test material or other contaminants). Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and enpoints are robust). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) Sensitivity of the coccinelled predator, *Coccinella undecimpunctata* L. and its prey *Brevicoryne brassicae* L. towards thiamethoxam (Actara 25% WG) was investigated via direct overspray. *C. undecimpunctata:* LC_{50} = 0.25% The test organisms were collected from cabbage and clover fields at Giza Governorate; no information was available on the history of the collection site regarding previous pesticide usage. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1316

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Basappa, H.	2007	Toxicity of biopesticides and synthetic insecticides to egg parasitoid, *Trichogramma chilonis I*shii, and coccinellid predator, *Cheilomenes sexmaculata* (Fabricius)	Journal of Biological Control (2007), Volume 21, Number 1, pp. 31-36	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The toxicity of thiamethoxam to *T. chilonis* and *C. sexmaculata* was evaluated under laboratory conditions via direct overspray of the eggs. Only one concentration of thiamethoxam (0.02%) was tested. Emergence of *T. chilonis* adults after exposure of 1, 3 and 7 day old eggs: 25.33% , 17.33% and 39.33% Mortality of *C. sexmaculata* after exposure of 1, 3 and 7 day old eggs: 55.55%, 93.33% and 100% According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1326

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Poletti, M.; Maia, A. H. N.; Omoto, C	2007	Toxicity of neonicotinoid insecticides to Neoseiulus californicus and *Phytoseiulus macropilis* (Acari: Phytoseiidae) and their impact on functional response to *Tetranychus urticae* (Acari: Tetranychidae).	Biological Control (2007), Volume 40, Number 1, pp. 30-36	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The objectives of this study were to evaluate the toxicity of insecticides on the phytoseiid mites *Neoseiulus californicus* (McGregor) and *Phytoseiulus macropilis* (Banks) (Acari: Phytoseiidae) and their impact on the functional response to *Tetranychus urticae* Koch eggs. A residual-contact bioassay was used to evaluate mortality of adult females of both predator mites exposed to thiamethoxam (135 mg a.s./L). Aphids were exposed by direct overspray of the leaves they were placed on. Mortality was 6% at the tested rate. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1327

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Lauziere, I.; Elzen, G.	2007	Effect of formulated insecticides on *Homalodisca vitripennis* (Germar) (Hemiptera: Cicadellidae) and its parasitoid *Gonatocerus ashmeadi* Girault (Hymenoptera: Mymaridae).	Journal of Entomological Science (2007), Volume 42, Number 1, pp. 11-19	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Thiamethoxam was tested using a bioassay of foliar insecticide residue (sprayed cowpea plants) to evaluate its impact on *Homalodisca vitripennis* nymphs and adults and on *Gonatocerus ashmeadi* parasitoid pupae and newly-emerged adults. The test rate was 0.051 kg a.s./ha, mortality for nymphs and adults was 100% after 24 -72 hours. As only one concentration of thiamethoxam was tested, this reference is considered as supplemental information only.	1333
CA 8.3.2	Carvalho, G. A.; Moura, A. P.; Bueno, V. H. P.; Editor(s): Castane, C.; Sanchez, J. A.	2006	Side effects of pesticides on *Trichogramma pretiosum* (Hymenoptera: Trichogrammatidae).	Bulletin OILB/SROP (2006), Volume 29, Number 4, pp. 355-359, 12 refs.	This article is an abstract published from a conference (Integrated control in protected crops under Mediterranean climate. Proceedings of the meeting of the International Organization for Biological and Integrated Control of Noxious Animals and Plants, West Palearctic Regional Section (IOBC/WPRS) Working Group, Murcia, Spain, 14-18 May 2006.). This study examined the effect of pesticides (including thiamethoxam) on the egg parasitiod *Trichogramma pretiosum* under laboratory conditions. Only one concentration was tested (0.05 g a.s./L; Actara 25WG). According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). Additionally, the methods used were not sufficiently detailed for review. Therefore, this reference is not considered relevant.	1357

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Hamamura, T.; Kohno, K.; Takeda, M.	2006	Insecticide susceptibility of *Pardosa astrigera* L. Koch spiderlings.	Jpn. J. Appl. Entomol. Zool Vol. 50 No.30: 253-255 (2006)	Does not fulfil criteria 5 (Several dose levels tested, at least 3, to establish a dose-response. Does not fulfil criteria 6 (Exposure route is suitably quantified). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The effects of thimathoxam on the survival of spiderlings of *Pardosa astrigera* L. Koch, were examined in the laboratory. Spiderlings were placed in plastic cups and test solution (thiamethoxam: 10%, dilution multiple: 3000) was sprayed from above. The mortality rates of the after 24 hours were 13.8 and 30%, respectively. Only one concentration of each substance was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha), not concentration (e.g. %). While this exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore, this reference is considered as supplemental information only.	1362

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Cole, P. G.; Horne, P. A.	2006	The impact of aphicide drenches on *Micromus tasmaniae* (Walker) (Neuroptera: Hemerobiidae) and the implications for pest control in lettuce crops.	Australian Journal of Entomology (2006), Volume 45, Number 3, pp. 244-248	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) Thiamethoxam (Actara) was applied to lettuce seedlings by drenching. A model aphid (*Macrosiphum euphorbiae* (Thomas), was periodically released onto the seedlings over 10 weeks. The effect of thiamethoxam on larvae of predatory brown lacewings (*Micromus tasmaniae*) was measured over 10 weeks by bioassay. This bioassay procedure was performed eight times over 75 days of the trial to evaluate the persistence of the effects of the insecticides. As this field study was not conducetd according to standard protocols (exposure, sampling), this reference is not considered relevant for this review.	1366
CA 8.3.2	Tillman, P. G.	2006	Feeding responses of *Trichopoda pennipes* (F.) (Diptera: Tachinidae) to selected insecticides.	Journal of Entomological Science (2006), Volume 41, Number 3, pp. 242-247	Does not fulfil criteria 4 (Test organisms are not previously exposed to the test material or other contaminants). Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommendfed protocols). Feeding responses of the parasitoid *Trichopoda pennipes* (F.) to thiamethoxam were determined in laboratory tests. Feeding tests with NTA organisms are neither a recommended test procedure nor can the endpoint be used in standard risk assessment. Test organisms were collected in a sorghum field without any information on pretreatment procedures. Only one concentration of thiamethoxam was tested. This reference is therefore considered not relevant for this review.	1367

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Kim, D. H.; Kim, S. S.; Kim, K. S.; Hyun, J. W	2006	Comparative toxicity of some pesticides to the predatory mites, *Neoseiulus fallacis* Garman (Acari: Phytoseiidae).	Korean Journal of Applied Entomology (2006), Volume 45, Number 2, pp. 179-188	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) In this study, contact toxicity of thiamethoxam (33.3 ppm) against *Neoseiulus fallacis* female imagoes and eggs was tested via overspray of inoculated female imagoes fed *N. fallacis*, and on inoculated Phytoseiidae on kidney bean leafs with *Tetranychus urticae* present. Egg survival after 72 hours: 82.1%; Hatchability > 90%; Adult survival after 72 hours: 24.4% According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha), not concentration (e.g. %). While direct overspray is an exposure route is environmentally relevant, it is not considered in testing conducted for the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1378

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Ersin, F.; Madanlar, N.	2006	Investigations on the effects of some pesticides used in greenhouse vegetables on predatory mite *Phytoseiulus persimilis* A.-H. (Acarina: Phytoseiidae) in laboratory conditions.	Tuerkiye Entomoloji Dergisi (2006), Volume 30, Number 1, pp. 67-80	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 6 (Exposure route is environmentally revlevant) Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). In this study, the effects of thiamethoxam (30 ml/100L) on the predatory mite *Phytoseiulus persimilis* were investigated by using residual film (on bean plant leaves), direct injection and slide dip methods. The latter two test methods are not standard methods in recommended protocols. As two study methods (direct injection and slide dip) differ from the standard methods used for Tier I and II toxicity testing, and injection is furthermore not an evironmentally relevant exposure scenario, these are considered not relevant for this review. Only one concentration of thiamethoxam (30mL/100L) was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha), not concentration (e.g. ml/L). Therefore, this study is considered as supplementary information only.	1400
CA 8.3.2	Shanmugam, P. S.; Balagurunathan, R.; Sathiah, N.	2006	Safety of some newer insecticides against *Trichogramma chilonis* Ishii.	Journal of Plant Protection and Environment (2006), Volume 3, Number 1, pp. 58-63	Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and enpoints are robust.) Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). The effect of thiamethoxam to *T. chilonis* was assessed by recording the adult emergence rate of parasitism and adult mortality after dipping exposure. As this exposure method differs from the standard exposure method used for toxicity testing, this study is not considered relevant.	1405

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Vijayaraghavan, C.; Regupathy, A.	2006	Impact of thiamethoxam on spiders in sugarcane ecosystem.	Journal of Plant Protection and Environment (2006), Volume 3, Number 1, pp. 36-39	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details)). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) The impact of thiamethoxam on the spider population in a sugarcane ecosystem was investigated in field studies in various locations in Tamil Nadu, India. No information on the pretreatment history of the test field and limited information on the sampling scheme is given, which seems to be not comparable to standard methods of NTA field studies.Therefore this reference is considered not relevant for this review.	1413
CA 8.3.2	Mafi, S. A.; Ohbayashi, N.		Toxicity of insecticides to the citrus leafminer, *Phyllocnistis citrella*, and its parasitoids, *Chrysocharis pentheus* and *Sympiesis striatipes* (Hymenoptera: Eulophidae).	Applied Entomology and Zoology (2006), Volume 41, Number 1, pp. 33-39	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The relative toxicity of 12 common insecticides (incl. thiamethoxam) against the eggs and first instar larvae of the pest, and its parasitoids of *Chrysocharis pentheus* and *Sympiesis striatipes*, was compared. Test organism were exposed via dipping of leaves. Only one concentration of thiamethoxam was tested (0.003%). According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefore considered not relevant for this review.	1414

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mullin, C. A.; Saunders, M. C., II; Leslie, T. W.; Biddinger, D. J.; Fleischer, S. J.	2005	Toxic and behavioral effects to carabidae of seed treatments used on Cry3Bb1- and Cry1Ab/c-protected corn.	Environmental Entomology (2005), Volume 34, Number 6, pp. 1626-163	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). To address potential direct nontarget effects of combined technologies, 16 Carabidae species from 10 genera (*Agonum, Amara, Bembidion, Anisodactylus, Chlaenius, Harpalus, Patrobus, Poecilus, Pterostichus, and Scarites*) field-collected from corn, were directly exposed to Bacillus thuringiensis (Bt) Cry toxin-laden pollens and seed treatments (incl. thiametoxam) in feeding bioassays. While this exposure route is environmentally relevant, it is not considered in testing conducted for the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 4 thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1426

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Moura, A. P.; Carvalho, G. A.; Rigitano, R. L. de O.; de O. Rigitano, R.L.	2005	Toxicity of insecticides used in tomato crop to *Trichogramma pretiosum*. Toxicidade de inseticidas utilizados na cultura do tomateiro a *Trichogramma pretiosum*.	Pesquisa Agropecuaria Brasileira (2005), Volume 40, Number 3, pp. 203-210	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommendfed protocols). The aim of this study was to evaluate the toxicity of thiamethoxam (at 0.05 mg/L) to *T. pretiosum*, in different immature stages in the laboratory. Eggs of *Anagasta kuehniella* were offered to females of *T. pretiosum* for 48 hours. After the parasitization, the eggs were treated by dipping them into the test solutions after they reached the egglarval, prepupal and pupal stages. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefor considered as supplemental information only.	1449

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Czepak, C.; Fernandes, P. M.; Albernaz, K. C.; Rodrigues, O. D.; Silva, L. M.; Silva, E. A. da; Takatsuka, F. S.; Borges, J. D.; da Silva, E. A.	2005	Selectivity of insecticides on the complex of natural enemies in cotton crop (*Gossypium hirsutum* L.). Seletividade de inseticidas ao complexo de inimigos naturais na cultura do algodao (*Gossypium hirsutum* L.).	Pesquisa Agropecuaria Tropical (2005), Volume 35, Number 2, pp. 123-127	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) The selectivity of thiamethoxam (300 g/ha) was evaluated in the complex of natural enemies of cotton crop in a field study. The field study was conducted in Brazil, the geoclimatic conditions are not appropriate and relevant for Europe. Samples were taken only up to 7 days after treatment, which is shorter than in standard NTA field studies, and not long enough to demonatrate recovery. This reference is thus considered not relevant for this review.	1456
CA 8.3.2	Kim SangSoo; Seo SangGi; Park JongDae; Kim SeonGon; Kim DoIk; Kim, S. S.; Seo, S. G.; Park, J. D.; Kim, S. G.; Kim, D. I.	2005	Effects of selected pesticides on the predatory mite, *Amblyseius cucumeris* (Acari: Phytoseiidae).	Journal of Entomological Science (2005), Volume 40, Number 2, pp. 107-114	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) Effects of thiamethoxam 10% WG to the predatory mite, *Amblyseius cucumeris* (Oudemans) (Acari: Phytoseiidae), were evaluated in laboratory bioassays. Only one concentration of thiamethoxam was tested (0.5 g/L). Survival was 96%, 90%, 84% and 80% after 24h, 72h, 120h and 168h, respectively. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. g/L). As this study method differs from the standard methods used for Tier I and II toxicity testing (application of the treatment was made via spray until runoff), this reference is considered not relevant for this review.	1460

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Orita, H.; Kashio, T.	2005	Toxic effect of some pesticides on adults and larvae of *Aphidoletes aphidimyza* (Rondani).	Kyushu Plant Protection Research (2005), Volume 51, pp. 83-88	English version of publication unavailable (Chinese); English abstract available. 40 insecticides (including thiamethoxam at 50 ppm) were tested on adults of *Aphidoletes aphidimyz*a by a residual contact method in test tubes at 24±1°C in the laboratory. Adults were reared in test tubes treated internally with aqueous dilutions of the test solutions. After 24 hours mortality reached 94.8%. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. ppm). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefor considered not relevant for this review.	1467
CA 8.3.2	Carvalho, G. A.; Miranda, J. C.; Moura, A. P.; Rocha, L. C. D.; Reis, P. R.; Vilela, F. Z.	2005	Control of Leucoptera coffeella (Guerin-Meneville & Perrottet, 1842) (Lepidoptera: Lyonetiidae) with soil-applied granulated insecticides and their effects on predatory wasps and parasitoids. Controle de Leucoptera coffeella (Guerin-Meneville & Perrottet, 1842) (Lepidoptera: Lyonetiidae) com inseticidas granulados e seus efeitos sobre vespas predadoras e parasitoides.	Arquivos do Instituto Biologico (Sao Paulo) (2005), Volume 72, Number 1, pp. 63-72, 14 refs. ISSN: 0020-3653	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). The efficacy of granulated insecticides applied to soil (thiamethoxam; 300 or 500 g a.s./ha), against *L. coffeella* was evaluated, and their effects on predatory wasps and parasitoids of this pest were investigated. The experiment was conducted in a coffee plantation in Brazil. Theerfore the geoclimatic conditions are not appropriate and relevant for Europe. Additionally, the application type used in this study is not relevant to the uses considered in this assessment (foliar spray and seed treatment). Therefore this reference is not consider relevant.	1475

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Wang, Y.; Wu, C.; Cang, T.; Yang, L.; Yu, W.; Zhao, X.; Wang, Q.; Cai, L.	2014	Toxicity risk of insecticides to the insect egg parasitoid *Trichogramma evanescens* Westwood (Hymenoptera: Trichogrammatidae)	Pest Management Science (2014), 70 (3), 398-404	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols) The selectivity of thiamethoxam to adults of the egg parasitoid *T. evanescens* was evaluated. Bioassays were carried out by exposing adult *T. evanescens* to residues of the insecticides applied on glass tube. 24h LC_{50} = 1.2 mg/L. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	1523
CA 8.3.2	Schenke, D.; Heimbach, U.	2014	Exposure of Coccinellidae to guttation droplets on maize seedlings with seed or granule treatment of neonicotinoids	Abstracts of Papers, 248th ACS National Meeting & Exposition, San Francisco, CA, United States, August 10-14, 2014 (2014), AGRO-359. CODEN: 69SZG4	This article is an abstract published from a conference (ACS National Meeting & Exposition, San Francisco, CA, United States, August 10-14, 2014). Field studies were carried out to estimate the frequency of guttation and measure neonictinoid residues in gutation fluid in maize. Additionally the occurance of Coccinellidae on maize seedlings were noted and analysis of neonictinoid residues in Coccinellidae was carried out. This article was not sufficiently detailed for assessment and is therefore not considered relevant.	1524

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Sun, Z.; Chen, D.; Jia, F.; Zhang, C.; Tang, S.; Ren, G.; Liu, X.	2014	Effect of six conventional insecticides on *Aphidius gifuensis* (Ashmead) in tobacco fields	Zhiwu Baohu (2014), 40 (4), 185-189. CODEN: ZBHABE; ISSN: 0529-1542	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) In order to evaluate the safety of insecticides to *Aphidius gifuensis* in tobacco fields, the impacts of six conventional insecticides (incl. thiamethoxam as 25 WG diluted by a factor of 8000) on *A. gifuensis* were investigated in laboratory using the dry film method in flasks (adults) and a dipping method (mummies). Adult contact toxicity: 97.54 % mortality after 24 hours; Dipping of mummies: Emergence rate of 16.67%, Deformity rate: 33.33% Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefore considered as supplemental information only.	1527
CA 8.3.2	Uma, S.; Jacob, S.; Lyla, K. R.	2014	Acute contact toxicity of selected conventional and novel insecticides to *Trichogramma japonicum* Ashmead (Hymenoptera: Trichogrammatidae)	Journal of Biopesticides (2014), 7 (Suppl.), 133-136.	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Eighteen insecticides (including thiamethoxam) were evaluated for their toxic effects on the hymenopteran egg parasitoid *Trichogramma japonicum* Ashmead following the residual film bioassay method with test tubes (protocol recommended by IOBC). Only the recommended field concentration was tested (25 g a.s./ha). As only one concentration was tested, this reference provides supplemental information only.	1531

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Thiruveni, T.; Kuhar, H. Ganesh; Kuttalah, S.	2014	Effect of newer indigenous thiamethoxam 25% WG formulation on spider population in mango	Pestology (2014), 38 (5), 49-51	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details, sampling regime)). Two field trials were conducted to study the effect of thiamethoxam 25% WG on field population of spiders. The field study was conducted in India, thus the geoclimatic conditions are not appropriate and relevant for Europe. No information on the pretreatment history of the test field and limited information on the sampling scheme is given, which seems to be not comparable to standard methods of NTA field studies.Therefore this reference is considered not relevant for this review.	1536
CA 8.3.2	Argolo, Poliane Sa; Jacas, Josep A.; Urbaneja, Alberto	2014	Comparative toxicity of pesticides in three phytoseiid mites with different life-style occurring in citrus: *Euseius stipulatus*, *Neoseiulus californicus* and *Phytoseiulus persimilis*	Experimental and Applied Acarology (2014)	Thiamethoxam was not a test substance in this study. Therfore this reference is not relevant for this review.	1537

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Abraham, Cheri. M.; Braman, S. K.; Oetting, R. D.; Hinkle, N. C.	2013	Pesticide compatibility with natural enemies for pest management in greenhouse gerbera daisies	Journal of Economic Entomology (2013), 106 (4), 1590-1601.	Does not fulfil criteria 1 (well defined test material (including purity/content)). Does not fulfil criteria 5 (several dose levels tested, at least 3, to establish a dose-response). Does not fulfil criteria 6 (Exposure route is clearly defined). Does not fulfil criteria 9 (Sufficient experimental information provided to substantiate and evaluate whethter the study conclusions and endpoints are robust). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommendfed protocols). *Diglyphus isaea* and *Neoseiulus californicus* were exposed to pesticides (including thiamethoxam) in laboratory assays (leaf-dip assays for the parasitoid wasps and pesticide swirl assays for predatory mite). No information can be found on the test material or exposure rate. The study methods used differ from the standard methods used for Tier I and II toxicity testing. Therefore, this reference is considered not relevant for this review.	1547
CA 8.3.2	Jenkins, S.; Hoffmann, A. A.; McColl, S.; Tsitsilas, A.; Umina, P.A.	2013	Synthetic pesticides in agro-ecosystems: are they as detrimental to nontarget invertebrate fauna as we suspect?	Journal of Economic Entomology (2013), 106 (2), 756-775	Does not fulfil criteria 8 (Effects are related to single test item). In this study, the impact of broad-spectrum pesticides and several selective pesticides on nontarget invertebrate fauna was assessed in five field trials in wheat and canola fields. Thiamethoxam was tested as a thiamethoxam-abamectin mixture, therefore effects cannot be related to thiamethoxam exclusively and therefore this reference is considered not relevant in this review.	1550

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Wang, X.; Jia, J.; Zhang, Y.; Zhou, Q.; Gao, C.	2013	Laboratory safety evaluation of eight insecticides to two predating natural enemies, *Hylyphantes graminicola* and *Coleosoma octomaculatum*	Nanjing Nongye Daxue Xuebao (2013), 36 (3), 53-58	English version of publication unavailable (Chinese); English abstract available. Does not fulfil criteria 10 (Study conditions should not differ significantly from recommendfed protocols). The toxicities to the spider *Hylyphantes graminicola* and *Coleosoma octomaculatum*, were determined in the laboratory by the spider dipping method. As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefore considered not relevant for this review.	1569
CA 8.3.2	Bajya, D. R.; Baheti, H. S.; Raza, S. K.	2013	Field efficacy of Thiamethoxam 75% SG against major sucking pests of cotton and effect on natural enemies	Pestology (2013), 37 (1), 46-50	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details, sampling regime)). A field experiment was conducted to study the efficacy of a new insecticide formulation of Thiamethoxam 75% SG against major sucking pests (jassids, thrips, whiteflies) in cotton under field conditions. The field study was conducted in India, thus the geoclimatic conditions are not appropriate and relevant for Europe. No information on the pretreatment history of the test field and limited information on the sampling scheme is given, which seems to be not comparable to standard methods of NTA field studies. Therefore this reference is considered not relevant for this review.	1571

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Anand, G. K. Sujay; Sharma, R. K.; Shankarganesh, K.	2013	Efficacy of newer insecticides against leaf hopper and whitefly infesting brinjal and its effect on coccinellids	Pesticide Research Journal (2013), 25 (1), 6-11	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whether the study conclusions and endpoints are robust (e.g. pretreatment details, sampling regime)). Efficacy of thiamethoxam (35 g a.s./ha) and toxicity against coccinellids was tested in a field study in India. The field study was conducted in India, thus the geoclimatic conditions are not appropriate and relevant for Europe. Furthermore, it doesn't follow recommended NTA field study guidelines; samples were only taken up to 13 days after spraying and the collection method was not comparable to standard methods. Therefore this reference is considered not relevant for this review.	1578
CA 8.3.2	Wang, Y.; Chen, L.; Yu, R.; Zhao, X.; Wu, C.; Cang, T.; Wang, Q.	2012	Insecticide toxic effects on *Trichogramma ostriniae* (Hymenoptera:Trichogrammatidae)	Pest Management Science (2012), 68 (12), 1564-1571.	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommendfed protocols). This study examines the toxic effects of thiamethoxam (0.05 g a.s./L) on *T. ostriniae* under laboratory conditions. Eggs of *Anagasta kuehniella* were offered to females of T. pretiosum for 48 hours. After the parasitization, the eggs were treated by dipping them into the test solutions after they reached the egglarval, prepupal and pupal stages. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. g/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefore considered not relevant for this review.	1590

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Wang, Y.; Liu, T.; Li, J.; Dong, .; Zhou, T.; Zhang, D.	2012	Selective toxicity tests and field trials of 5 kinds of insecticide to *Brevicoryne brassicae* and ladybeetles	Nongyao (2012), 51 (11), 829-831, 857	Does not fulfil criteria 8 (Effects are related to single test item). Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). The selective toxicities and field control effects of chlorantraniliprole thiamethoxam 40% WG to *B. brassicae* and ladybeetles were studied. *Harmonia axyridis* second instar larvae were tested via dipping method, *Propylaea japonica* adults were exposed for 3 hours via dry film method using finger-shaped tube and transferred afterwards to a Petri dish not to have been treated. Furthermore a field study was conducted. As this study method differs from the standard methods used for Tier I and II toxicity testing and in all cases, effects can not be related to one test substance unequivocally, this study not considered relevant for this review.	1593
CA 8.3.2	Cui, X.; Zhang, Q.; Jiang, H.; Lin, R.; Wang, K.	2012	Acute toxicity evaluation of neonicotinoid insecticides to *Bombyx mori* and observation of toxic symptoms	Canye Kexue (2012), 38 (2), 288-291	English version of publication unavailable (Chinese); English abstract available. Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) In order to identify the toxicity of neonicotinoid insecticides (including thiamethoxam) to non-target organism silkworm (*Bombyx mori*), the acute toxicity to silkworm were determined by leaf dipping method. 96 h LC_{50} = 1.31 mg/L. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). Therefore this reference is considered as supplemental information only.	1599

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Wang, Y.; Yu, R.; Zhao, X.; Chen, L.; Wu, C.; Cang, T.; Wang, Q.	2012	Susceptibility of adult *Trichogramma nubilale* (Hymenoptera: Trichogrammatidae) to selected insecticides with different modes of action	Crop Protection (2012), 34, 76-82.	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) A modifyed dry film residue method using plastic tubes was used to assess the toxicity of thiamethoxam on *T. nubilal.* 24h LC_{50} = 1.86 mg/L According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefor considered not relevant for this review.	1612

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Rocha, L. C.D.; Carvalho, G.A.; Moscardini, V.F.; Rezende, D.T.	2011	Selectivity of insecticides used in coffee crop to larvae of *Cryptolaemus montrouzieri* Mulsant	Ciencia Rural (2011), 41 (6), 939-946.	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 5 (several dose levels tested, at least 3, to establish a dose-response.) Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The selectivity of thiamethoxam (0.5 g a.s./L) for larvae of *Cryptolaemus montrouzieri* (Coleoptera: Coccinellidae) was studied under laboratory conditions via direct overspray. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). While direct overspray is an exposure route is environmentally relevant, it is not considered in testing conducted for the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1628

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Kerns, D. L.; Kesey, B. J.; Baugh, B. A.; Patman, D. R.	2011	Evaluation of insecticides against cotton aphids and convergent lady beetle larvae in cotton, 2010	Arthropod Management Tests (2011), 36, F44/1-F44/3.	Does not fulfil criteria 4 (Tets organisms are not previously expose to other contaminants). Does not fulfil criteria 8 (effects are related to a single test item). The efficacy of selected insecticides (including thismethoxam as Centric 40WG) was tested for control of cotton aphid (*Aphis gossypii*) in a cotton field and to evaluate the toxicity against convergent lady beetle (*Hippodamia convergens*). The entire site was treated with another insecticide prior to application with the thiamethoxam formulation Centric. Therefore, this reference is considered not relevant.	1642
CA 8.3.2	Kerns, D. L.; Baugh, B. A.; Patman, D. R.	2010	Evaluation of insecticides against cotton aphids and lady beetle larvae in cotton, 2009	Arthropod Management Tests (2010), 35, F17	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) Evaluation of thiamethoxam (as Centric 40WG) against cotton aphids and lady beetle larvae in cotton. The field study doesn't follow NTA field study guidelines; eg samples only taken up to 14 days after application. Therefore this reference is considered not relevant.	1666
CA 8.3.2	Kerns, D. L.; Baugh, B. A.	2008	Evaluation of insecticides against cotton aphids and predators in cotton, 2008.	Arthropod Management Tests (2009), 34, F27	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The efficacy of thiamethoxam (Centric 40WG) was tested for control of cotton aphid (*Aphis gossypii*) and for the effects on convergent lady beetle (*Hippadomia convergens*) and common green lacewing (*Chrysoperla plorabunda*). The field study doesn't follow NTA field study guidelines; eg samples only taken up to 10 days after application. Therefore this reference is considered notrelevant.	1686

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Hull, L. A.; Joshi, N.K.; Zaman, F. U.	2008	Management of internal feeding lepidopteran pests in apple, 2008	Arthropod Management Tests (2009), 34, A8	Does not fulfil criteria 8 (Effects are related to single test item). Several insecticides were evaluated against a variety of orchard pests, especially internal feeding lepidoptera, and their toxicity to a number of natural enemy species. Thiamethoxam was tested as Actara 25W in combination with Imidan 70W. Thus, effects cannot be related to thiamethoxam exclusively and therefore this reference is considered not relevant in this review.	1687
CA 8.3.2	Zhao, X.; Yu, R.; Cang, T.; Chen, L.; Wu, S.; Wu, C.; Gu, X.	2008	Effects of Nilaparvata lugens and *Cyrtorhinus lividipennis* Reuter to insecticides	Nongyao (2008), 47 (1), 74-76.	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) Lab toxicity and field control of insecticides to *Nilaparvata lugens* (Stal) were tested with several insecticides (including thiamethoxam at rates of 3.125 – 50 mg/L) and the toxicity against *Cyrtorhinus lividipennis* Reuter was determined using dry film residue method. Tubes were coated with the test solution and rice stems dipped in the test solution were added to the tubes before larvae were added. Mortality at 3.125 mg a.s./L was 93.3%. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). As this study method differs from the standard methods used for Tier I and II toxicity testing, this reference is therefor considered not relevant for this review.	1697

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mali, A.K.; Kurtadikar, J. S.; Wadnerkar, D. W.; Nemade, P. W.	2008	Studies on the safety of pesticides to grapevine mealy bug predator, *Scymnus coccivora* Aiyar	Pestology (2008), 32 (1), 37-46.	Does not fulfil criteria 5 (several dose levels tested, at least 3, to establish a dose-response.) Does not fulfil criteria 9 (Sufficient experimnental information provded to substantiate and evaluate whether the study conclusions and endpoints are robust.) Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The toxicity of thiamethoxam at 0.01% against the coccinellid predator, *Scymnus coccivora* Aiyar of grapevine mealy bug, *Maconellicoccus hirsutus* Green was studied under laboratory conditions. The grubs were exposed to sprayed grapevine leaves. No details on the method and conditions are described. Eggs and pupae were exposed via direct overspray. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). While direct overspray is environmentally relevant, it is not considered in testing conducted for the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1703

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Mali, A.K.; Kurtadikar, J. S.; Wadnerkar, D. W.; Nemade, P. W.	2008	Studies on the safety of pesticides to grapevine mealy bug predator, *Cryptolaemus montrouzieri* Aiyar	Pestology (2008), 32 (4), 17-27	Does not fulfil criteria 5 (several dose levels tested, at least 3, to establish a dose-response.) Does not fulfil criteria 9 (Sufficient experimnental information provded to substantiate and evaluate whether the study conclusions and endpoints are robust.) Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols.) The toxicity of thiamethoxam at 0.01% against Australian coccinellid predator, *Cryptolaemus montrouzieri* Aiyar of grapevine mealy bug, *Maconellicoccus hirsutus* Green was studied under laboratory conditions. The grubs were exposed to sprayed grapevine leaves. No details on the method and conditions are described. Eggs and pupae were exposed via direct overspray. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). While direct overspray is environmentally relevant, it is not considered in testing conducted for the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	1708

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Javaregowda; Naik, L. K.	2005	Bioefficacy of thiamethoxam 25 WG against paddy white backed plant hopper (WBPH) and their natural	Pestology (2005), 29 (5), 31-33	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). A field study was conducted in upland Paddy to know the bioefficacy of thiamethoxam (as 25 WG) against Paddy white backed plant hopper (WBPH) at Agricultural Research Station, Sirsi, Karnataka, India. The field study was conducted in India, thus the geoclimatic conditions are not appropriate and relevant for Europe. Addtionally, the number of non-targets were reported up to 15 days after application only. Therefore recovery of non-target arthropods can not be demonstrated due to the limitations of the field study. Therefore this study is not considered relvent.	1762
CA 8.3.2	Schenke, D; Baier, B	2009	Effect of thiamethoxam towards Poecilus cupreus larvae in comparison to imidacloprid applied as pelleted sugar beet seeds	Conference: 2009 Annual Meeting of the UK branch of the Society of Environmental Toxicology and Chemistry (SETAC 2009), Goteborg Convention Centre, Goteborg, 31 May 2009 - 4 Jun 2009	This article is an abstract published from a conference (Annual Meeting of the UK branch of the Society of Environmental Toxicology and Chemistry, SETAC 2009). This study investigated the effect of neonictinoids (incl. thiamethoxam) on *Poecilus cupreus* in laboratory tests with treated sugarbeet seeds (with Cruiser 70WS). The article is not sufficiently detailed for assessment and is therefore not considered relevant.	1789

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.4.1	Roger, P.; Alves, L.; Elke, J.B.N.; Cardoso, A.Martines, M.; Sousa, J.P.; Pasin, A.	2013	Earthworm ecotoxicological assessments of pesticides used to treat seeds under tropical conditions.	Chemosphere 90 (2013) 2674–2682	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). This study investigated the effects of seed dressing formulations including Cruiser 350FS (thiamethoxam) on survival and reproduction of *E. andrei* (earthworm), using standardized ecotoxicological tests after ISO guidelines with adaptations for tropical conditions. The acute LC_{50} was reported as >1000 mg a.s./kg soil dw. The chronic NOEC was reported as 250 mg a.s./kg soil dw and the LOEC = 500 mg a.s./kg soil dw. As tropical artificial soil and test conditions were used, the reference is therefore not considered relevant for European conditions.	44
CA 8.4.1	Zhang, P.; Chen, C. Y.; Li, H.; Liu, F.; Mu, W.	2014	Selective toxicity of seven neonicotinoid insecticides to fungus gnat *Bradysia odoriphaga* and earthworm *Eisenia foetida*.	Acta Phytophylacica Sinica (2014), Volume 41, Number 1, pp. 79-86, 23 refs.	English version of publication unavailable (Chinese); English abstract available. This study tested the effects of 13 pesticides, including thiamethoxam and clothianidin, to the fungus gnat, *Bradysia odoriphag*, and earthworms. The acute toxicity to earthworms was determined using artificial soil method. It was stated that the toxicity of thiamethoxam was low and for clothainidin the toxicity was medium. Acute toxicity to earthworms is no longer part of the risk assessment. Additionally, the fungus gnat is a pest species. Therefore this reference has been considered as not relevant for this review. There is ample regulatory and other literary references for earthworm species, none of which indicate that earthworms would drive the risk assessment.	497

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.4.1	Wang, Y.; Chen, L.; Zhao, X.; Wu,C.; Cang, T.; Yu, R.; Wu, S.; Wang, Q.	2010	Acute toxicity of neonicotinoids and avermectins to earthworm, *Eisenia foetida*	Nongye Huanjing Kexue Xuebao (2010), 29 (12), 2299-2304. ISSN: 1672-2043	English version of publication unavailable (Chinese); English abstract available Filter paper testing and artificial soil testing methods were applied to determine the acute toxicity of thiamethoxam to *E. foetida*. The 24 h and 48 h LC_{50} values of thiamethoxam to *E. foetida* were > 62.91 g/cm² in the filter paper test and > 1200 mg/kg after 7 d and 14 d in the artificial soil test. As acute toxicity to earthworms is no longer part of the risk assessment, filterpaper testing is not a recommended test protocol and the conditions of the artificial soil test cannot be confirmed from the abstract, this reference has been considered as not relevant for this review. There is ample regulatory and other literary references for earthworms species, none of which indicate that worms would drive the risk assessment.	1652
CA 8.4.2.1	Roger, P.; Alves, L.; Elke, J.B.N.; Cardoso, A.Martines, M.; Sousa, J.P.; Pasin, A.	2014	Seed dressing pesticides on springtails in two ecotoxicologicalla boratory tests	Ecotoxicology and Environmental Safety 105 (2014) 65 – 71	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). This study investigated the effects of seed dressing formulations including Cruiser 350FS (thiamethoxam) on survival and reproduction of *Folsomia candida* (Collembola), using standardized ecotoxicological tests after ISO guidelines with adaptations for tropical conditions. The acute LC_{50} was reported as >1000 mg a.s./kg soil dw. The chronic NOEC was reported as 1 mg a.s./kg soil dw and the LOEC >1 mg a.s./kg doil dw. As tropical artificial soil and tropical conditions were used, the reference is therefore not considered relevant for European conditions.	25

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.4.2.1	El-Naggar, J. B.; Nour El-Hoda; Zidan, A.	2013	Field evaluation of imidacloprid and thiamethoxam against sucking insects and their side effects on soil fauna	Journal of Plant Protection Research 53 (4), 2013	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). This field study was carried out to evaluate the effectiveness of imidacloprid and thiamethoxam, used separately as seed treatments and foliar applications at recommended rates (Cruiser 70WS at 2 gm/kg seed; Actara 25WG at 20 gm/100 L). The side effects on soil fauna was investigated as well. The study was conducted in Egypt, thus considered not relevant for European conditons. The primary aim of the study was effects on efficacy, thus sampling of non-target soil fauna was not as robust as regulatory field studies targeting effects on soil fauna. Therefore this study is not considered relvent.	566
CA 8.5	Ferreira Fernandes, M.; de Oliveira Procópio, S.; Teles, D. A.; Guedes de Sena Filho, J.; Cargnelutti Filho, A.; Clí Via Rolemberg Andrade	2013	Crescimento e fixação biológica de nitrogênio de Gluconacetobacter diazotrophicus Na presença de inseticidas utilizados na cultura da cana-de-açúcar	Rev. Cienc. Agrar., v. 56, n. 1, p. 12-18, jan./mar. 2013	English version of publication unavailable (Portugues); English abstract available Five insecticides (including thiamethoxam) were tested in their respective doses regarding the commercial impact on the growth of bacteria in liquid medium DIGs. The impact of pesticides on nitrogenase activity of *G. diazotrophicus* grown in semi-solid LGI-P-free N was also evaluated. As nitrogen fixating bacteria are not the object of investigation for regulatory purpose, this has been considered as not relevant for this review. There is ample regulatory and other literary references for nitrogen transformation, none of which indicates that nitrogen transformation would drive the risk assessment.	648

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.5	Vendan, K. T.; Sreenivas, A. G.; Nargund, V. B.; Nadaf, A. M.	2008	Impact of seed dressing chemicals on soil micro flora and sucking pests in cotton.	Annals of Plant Protection Sciences (2008), Volume 16, Number 1, pp. 212-214, 3 refs. ISSN: 0971-3573	Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). In a pot experiment the effect of thiamethoxam as a seed treatment on soil microflora was investigated. The endpoint was not nitrogen transformation but reduction of the soil bacterial population, and therefore the reference is considered not relevant.	1204
CA 8.5	Liu, G.; Yao, K.; Zheng, L.; Zhou,Q.; Zhang, F.	2005	Effects of thiamethoxam and its photo-degradation products on soil microbe activity	Nongye Huanjing Kexue Xuebao (2005), 24 (5), 870-873. ISSN: 1672-2043	English version of publication unavailable (Chinese); English abstract available. Under laboratory conditions, the effects of thiamethoxam and its photo-degrdns. on soil microbe activity were systematically investigated. As the main endpoint was carbon transformation, which is not a data requirement in ecotoxicological risk assessment anymore, this reference is considered not relevant.	1757
CA 8.6	Szczepaniec, A.; Raupp, M. J.; Parker, R.D.; Kerns, D.; Eubanks, M.D.	2013	Neonicotinoid Insecticides Alter Induced Defenses and Increase Susceptibility to Spider Mites in Distantly Related Crop Plants	PLOS One, Volume 8, Issue 5, May 2013	This study tested the impact of neonicotinoid insecticides on plant defenses to population growth of an important pest species. To test the hypothesis that neonicotinoid insecticides suppress host plant defenses against spider mites, it was shown that transcription of phenylalanine amonia lyase, coenzyme A ligase, trypsin protease inhibitor and chitinase are suppressed and concentrations of the phytohormone OPDA and salicylic acid were altered by neonicotinoid exposure. The effects of neonicotinoid applications on the population growth of *Tetranychus urticae* was also measured. The reference is not relevant, since no ecotoxicological effects of thiamethoxam or clothianidin were investigated.	270

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.6	Soare, L.C.; Dobrecu, C-M.; Burtescu, L. ; Șutan, A.N.	2013	Research on the influence of two insectides on the gametophyte of some leptosporangiate pteridophytes	Analele Ş tiinţifice ale Universităţii „Al. I. Cuza" Iaşi s. II a. Biologie vegetală, 2013, 59 , 2: 5 - 12 http://www.bio.uaic.ro/publicatii/anale_vegetala/anale_veg_index.html ISSN: 1223 - 6578, E - ISSN: 2247- 2711	Does not fulfil criteria 3 (Applicable test species). Does not fulfil criteria 6 (Exposure route is environmentally relevant). Does not fulfil crietria 10 (Study conditions should not differ significantly from recommended protocols). The reference is considered not releveant, since investigations on the influence of thiamethoxam (25%) on spore germination and gametophyte differentiation in *Asplenium scolopendrium* and *Athyrium filix*, were made. These are not recommended OECD test species. The spores were cultivated in experimental solutions, which is not an environmentally relevant exposure.	650
CA 8.6	Ford, K. A.; Gulevich, A.G.; Swenson, T. L.; Casida, J. E.	2011	Neonicotinoid Insecticides: Oxidative Stress in Planta and Metallo-oxidase Inhibition	J. Agric. Food Chem. 2011, 59, 4860 – 4867	Does not fulfil criteria 1 (well defined test material including purity/ content). Does not fulfil criteria 2 (Number of animals per group are suffient to establish a statistical significance.) Does not fulfil criteria 6 (Exposure route is environmentally relevant). Does not fulfil criteria 10 (Study condition should not differ significantly from recommended protocols). Oxidative damage of neonicotinoids in soybean seedlings and phytotoxicity of neonicotinoids in crop seedlings was investigated. No information on the test item was given. The test compounds were administered hydroponically and only three plants were tested for each test compound. Only one concentration (without any clear relevance) was tested, NOEC and ECx values could not be calculated. Therefore, this reference is considered not relevant.	1813

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.7	Yan Xun; Moens, M.; Han RiChou; Chen ShuLong; Clercq, P. de; Yan, X.; Han, R. C.; Chen, S. L.; de Clercq, P.	2012	Effects of selected insecticides on osmotically treated entomopathogenic nematodes.	Journal of Plant Diseases and Protection (2012), Volume 119, Number 4, pp. 152-158, 30 refs. ISSN: 1861-3829. URL: http://www.jpdp-online.com	Does not fulfil criteria 14 (Applicable test species). This laboratory study explored the effects of thiamethoxam on osmotically treated and untreated *S. carpocapsae* showing no negative effects on the survival and infectivity of both osmotically treated and untreated *S. carpocapsae*. This reference was considered not relevant since the objective of this study was the exploration of methods of combining the use of nematodes as a biological control agent with insecticides for Integrated Pest Management. Nematodes are not an applicable test species.	732
CA 8.7	Botelho, A. A. A.; Monteiro, A. C.	2011	Sensitivity of entomopathogenic fungi to pesticides used in management of sugarcane.	Bragantia (2011), Volume 70, Number 2, pp. 361-369	English version of publication unavailable (Portuguese); English abstract available. Does not fulfil criteria 3 (Applicable test species). Does not fulfil criteria 6 (Exposure route is environmentally relevant) Investigation of thiamethoxam effects on the fungi *Beauveria bassiana* and *Metarhizium anisopliae*. Tests on fungi are not required for European risk assessment. Moreover, fungi were grown in potato dextrose agar medium containing thiamethoxam, which is not an environmentally relevant exposure route. The test were conducted simulating tropical conditions (high temperature, high humidity). Thus, this reference is considered not relevant for this review.	817
Top-Up search Thiamethoxam						

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Goulson, D.	2015	Neonicotinoids impact bumblebee colony fitness in the field; a reanalysis of the UK's food & environment research agency 2012 experiment.	PeerJ, (2015) Vol. 2015, No. 3.	This article presents a re-analysis of the data published in another article (FERA, 2013*). The author proposes using simpler Generalized Linear Models in order to present the results as significance tests in the traditional manner. This simplified data re-analysis suggests that the data show a negative relationship between both colony growth and queen production and the levels of neonicotinoids in the food stores collected by the bees. As the reference that this article is critiquing is nto considered relevant, this reference is not considered relevant. *The FERA (2013) study was not found in the literature search as it was not indexed by the databases used. The reason for not including this additional reference in the dossier is listed below.	8
CA 8.3.1	Biocca, M.; Fanigliulo, R.; Gallo, P.; Pulcini, P.; Pochi, D.	2015	The assessment of dust drift from pneumatic drills using static tests and in-field validation.	Crop Protection, (MAY 2015) Vol. 71, pp. 109-115.	This paper reports a research study aimed at evaluating the amounts of dust emitted by a precision pneumatic drill during the sowing of maize dressed with clothianidin, thiamethoxam and others. The crop used in this study is not included in this assessment as a representative use. Additionally, maize seed is dustier than the representative crop (pelleted sugar beet) considered in this assessment. Therefore its is considered not relevant for this review.	15

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Purdy, J. R.	2014	Concentrations and distribution of Neonicotinoid residues in honeybees (*Apis melifera*) in Ontario, Canada.	Abstracts of Papers American Chemical Society, (AUG 10 2014) Vol. 248, pp. 616-AGRO. Meeting Info.: 248th National Meeting of the American-Chemical-Society (ACS). San Francisco, CA, USA. August 10 - 14, 2014	This article is an abstract published from a conference (248th National Meeting of the American-Chemical-Society (ACS). San Francisco, CA, USA. August 10 - 14, 2014). A field study was conducted to measure residues of neonicotinoids in beehives in the corn and soybean growing area of southern Ontario, and to determine if any bee loss or symptoms of stress were associated with such residues. The article is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	19
CA 8.3.2	Devi, R.; Tambe, V. J.; Srasvankumar , G.; Nage, S. M.	2014	Larvicidal effect of some newer insecticides on *Chrysoperla carnea* (Stephens).	International Journal of Plant Protection (2014), Vol. 7, Nr. 1, pp. 91-95	Does not fulfil criteria 5 (Several dose levels tested, at least 3, to establish a dose-response relationship. Does not fulfil criteria 10 (Study condition should not differ significantly from recommended protocols). Laboratory studies were conducted to investigate the toxicity of Thiamethoxam 25 WG (25 g a.s./ha) against the first and third instar larvae of lacewing, *Chrysoperla carnea* (Stephens) by leaf dip method. 1st instar larvae : 76.67% mortality. 3rd instar larvae : 26.67% mortality. According to ESCORT II, the primary toxicity endpoint of the studies should be mortality, with determination of the LR_{50}. In this study only one concentration of thiamethoxam was tested, so no LC_{50} could be determined. Furthermore, as this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	71

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Yao, F.; Zheng, Y.; Zhao, J.; He, Y.; Weng, Q.; Desneux, N.	2015	Lethal and sublethal effects of thiamethoxam on the whitefly predator *Serangium japonicum* (Coleoptera: Coccinellidae) through different exposure routes.	Chemosphere, (2015 Jun) Vol. 128, pp. 49-55.	Does not fulfil criteria 10 (Study condition should not differ significantly from recommended protocols). This study examined the lethal and sublethal effects of thiamethoxam on *S. japonicum* through three exposure routes: residue contact (glass tubes), egg-dip and systemic treatment. LC_{50} values were 6.65, 4.37 and 2.43 mg a.s./L, respectively. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). While the systemic exposure route is environmentally relevant, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route for predatory mites would be accounted for in regulatory higher tier field studies. There are 4 for thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	3

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Moscardini, V. F. G..Costa; Michaud, J P; Carvalho; G. A.	2015	Sublethal effects of insecticide seed treatments on two nearctic lady beetles (Coleoptera: Coccinellidae).	Ecotoxicology, (2015 Apr 23). Electronic Publication Date	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 6 (Exposure route is suitably quantified) This study examined the the impact on *Coleomegilla maculata* and *Hippodamia convergens* when the beetles consumed the extrafloral nectar of sunflowers grown from thiamethoxam treated seeds. While this exposure route is environmentally relevant, it is not considered in testing conducted for the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 4 thiamethoxam seed treatment field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	6

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Megha, R. R.; Basavanagoud , K.; Kulkarni, N. S.	2015	Safety evaluation of some selected insecticides against coccinellids *Cheilomenes sexmaculata* (Fab.) and *Hippodamia variegata* (Goeze).	Journal of Experimental Zoology, India (2015), Vol. 18, Number 1, pp. 315-318	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). A laboratory experiment was conducted to study the impact of some insecticides (including Thiamethoxam 25 WG at 0.2 g/L) on coccinellids, *Cheilomenes sexmaculata* (Fab.) and *Hippodamia variegate*. Exposure was made via direct overspray. Mortality after 72 hours: *Cheilomenes sexmaculata* : 76.67% *Hippodamia variegate*: 46.67% Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. mg/L). While this exposure route is environmentally relevant, it is not considered in the regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route for non-target athropods would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	34

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Dumaniya, S. G.; Patel, M. B.; Siddhapara, M. R.	2015	Toxicity of insecticides to *Cryptolaemus montrouzieri* (Mulsant).	Journal of Cotton Research and Development (2015), Vol. 29, Nr. 1, pp. 121-124	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). Study on relative toxicity of different insecticides (including thiamethoxam at 0.005%) on larvae and adults of *Cryptolaemus montrouzieri* via a dipping method. Mortality after 72 hours: 50.83% Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). As this study method differs from the standard methods used for Tier I and II toxicity testing, this study is considered as supplementary information only.	39
CA 8.3.2	Ko Ko; Liu YuDi; Hou MaoLin; Babendreier, D.; Zhang Feng; Song Kai; Ko, K.; Liu, Y. D.; Hou, M. L.; Zhang, F.; Song, K.	2015	Toxicity of insecticides targeting rice planthoppers to adult and immature stages of *Trichogramma chilonis* (Hymenoptera: Trichogrammatidae).	Journal of Economic Entomology (2015), Volume 108, Number 1, pp. 69-76	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Among others, thiamethoxam was tested under laboratory conditions for toxicity to adults and immatures of *Trichogramma chilonis* Ishii, using standard tests described by the IOBC (dry film method in glass tubes). Only the field recommended rate was tested (12.5 g a.s./ha). As only one concentration of thiamethoxam was tested, this study is considered as supplemental information only.	43

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	BuKeun, C.; Lim, E.; HeungSu, L.; ChungGyoo, P.	2014	Toxicity of several insecticides against *Halyomorpha halys* (Hemiptera: Pentatomidae) and *Gymnosoma rotundatum* (Diptera: Tachinidae).	Korean Journal of Applied Entomology (2014), Volume 53, Number 4, pp. 457-460	English version of publication unavailable (Korean); English abstract available. Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting). Toxicity of several insecticides (among them thiamethoxam) was tested against the brown marmorated stink bug, *Halyomorpha halys* (Hemiptera: Pentatomidae), and adult *Gymnosoma rotundatum* (Diptera: Tachinidae) in a sweet persimmon orchard. *G. rotundatum* were sampled using sticky traps with an aggregation pheromone and the 24h mortality was recorded. Only one concentration of thiamethoxam was tested under field conditions and a 24h mortality rate noted. The methods used here do not follow standard test methods. Additionally, the study was conducted in Korea (considered not relevant for European conditons). Thus, this reference is considered not relevant for his review.	60

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Karthick, K. S.; Kandibane, M.; Kumar, K.	2014	Safety of newer insecticides to natural enemies in the coastal rice ecosystem of Karaikal, U.T. of Puducherry.	Journal of Biopesticides (2014), Volume 7, Number 2, pp. 200-203	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). A field experiment was conducted to study the effect of thiamethoxam to natural enemies of rice pests. The population of natural enemies were recorded at ten randomly selected hills (expressed as number per hill) up to 14 days after treatment. As the study was conducted in India it is considered not relevant for European conditons. Addtionally, the number of natural enemies were reported up to 14 days after application only. Therefore recovery of non-target arthropods can not be demonstrated due to the limitations of the field study. Therefore this study is not considered relvent.	65

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Alim, Md. A.; Lim, U.T.	2015	Effects of fenitrothion on *Gryon japonicum* (Hymenoptera: Platygastridae) parasitizing non-viable, refrigerated eggs of *Riptortus pedestris* (Hemiptera: Alydidae)	Journal of Asia-Pacific Entomology (2015), 18 (2), 181-186	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). In this study the potential effects of thiamethoxam (0.05%) on the parasitization of host eggs by *G. japonicum* by direct spray, exposure to dried residues and oral ingestion was tested. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). While oral exposure and exposure via overspray are environmentally relevant exposure routes, it is not considered in the testing conducted for regulatory Tier I or II risk assessment for non-target arthropods. However, this exposure route would be accounted for in regulatory higher tier field studies. There are 5 thiamethoxam foliar application field studies that examine effects on and recovery of full fauna populations. Therefore this study is considered as supplemental information only.	77

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Salahuddin, B.; ur Rahman, H.; Khan, I.; Daud, M. K.	2015	Incidence and management of coconut scale, *Aspidiotus destructor signoret* (Hemiptera: Diaspididae), and its parasitoids on mango (*Mangifera* sp.)	Crop Protection (2015), 74, 103-109	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 17 (Study conditions should not differ significantly from recommended protocols) The efficacy of thiamethoxam 25 WG was evaluated at recommended rates against 1st and 2nd instars and mature *A. destructor* on nursery plants and mature mango trees as well as effects on its predators. As the study was conducted in India it is considered not relevant for European conditons. Furthermore, the primary endpoint was efficacy and effects on non-target arthropods were not recorded comparable to methods used in standard field tests. Therefore this study is considered as not relevant.	81
CA 8.3.2	Ohta, I.; Takeda, M.	2015	Acute toxicities of 42 pesticides used for green peppers to an aphid parasitoid, *Aphidius gifuensis* (Hymenoptera: Braconidae), in adult and mummy stages	Applied Entomology and Zoology (2015), 50 (2), 207-212	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). Acute toxicities of 42 pesticides to adult stage *Aphidius gifuensis* Ashmead were studied in the laboratory. Adult parasitoids were brought into contact with pesticide residues on glass plates, and the resultant mortalities were measured. Thiamethoxam WG (10%) diluted by a factor of 3000 was tested. Mortality was 34.7%. Only one concentration of thiamethoxam was tested. According to the Escort II guidance document, the endpoints reported should be based on the application rate (e.g. g/ha) not concentration (e.g. %). Therefore this study is considered as supplemental information only.	84

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.2	Saner, D. V.; Kabre, G. B.; Shinde, Y. A.	2014	Impact of newer insecticides on ladybird beetles (*Menochilus sexmaculatus* L.) in hybrid cotton	Journal of Industrial Pollution Control (2014), 30 (2), 269-271	Does not fulfil criteria 12 (Appropriate and relevant geoclimatic conditions (setting)). Does not fulfil criteria 16 (Sufficient experimental information provided to substantiate and evaluate whethter the study endpoints and conclusions are robust (e.g. pre-treatment details, characterisation of physico-chemical parameters, statistical methods, appropriate sampling regime)). The effect of 8 insecticides (incl. thiamethoxam) on lady bird beetles in cotton crop was investigated in a field study in India. The full text article only gives limited information on the execution of the study (no pre-treatment details, no characterisation of physico-chemical parameters, no info on the sampling regime etc. As the study was conducted in India it is considered not relevant for European conditons and thus not relevant for this review.	89
CA 8.6.2	Stamm, M. D.; Enders, L. S.; Donze-Reiner, T. J.; Baxendale, F. P.; Siegfried, B. D.; Heng-Moss, T. M.	2014	Transcriptional response of soybean to thiamethoxam seed treatment in the presence and absence of drought stress	BMC Genomics (2014), 15, 1055/1-1055/27	Does not fulfil criteria 8 (a quantitative relationship exist between reported endpoint and risk assessment endpoints of growth, mortality, behavior and/or reproduction). In this study qRT-PCR was used to compare gene expression of thiamethoxam-treated and untreated soybeans exposed to drought stress and unstressed (control) conditions of ten selected genes involved in plant defense pathways and general stress response. The main endpoint was transcriptional response, which is not a data requirement in ecotoxicological risk assessment, this reference is considered not relevant.	85

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.7	Hamlet, S. A.; Djekoun, M.; Smati, M.; Semassel, A.; Bensoltane, S. D.; Berrebbah, H.	2014	Histopathological effects of neonicotinoid insecticide in the hepatopancreas of terrestrial gastropod *Helix aspersa*.	Fresenius Environmental Bulletin (2014), Volume 23, Number 12, pp. 3041-3047	Does not fulfil criteria 3 (Applicable test species). Does not fulfil criteria 8 (a quantitative relationship exist between reported endpoint and risk assessment endpoints of growth, mortality, behavior and/or reproduction). The effect of thiamethoxam on histological changes in the hepatopancreas of the terrestrial gastropod *Helix aspersa* after a treatment of 20 days was investigated. As tests on terrestrial molluscs are not required for European risk assessment and the main endpoint was histological changes, which is not a data requirement in ecotoxicological risk assessment, this reference is considered not relevant.	50
Clothianidin search						
CA 8.1.1.3	Hoshi, N.; Hirano, T.; Omotehara, T.; Tokumoto, J.; Umemura, Y.; Mantani, Y.; Tanida, T.; Warita, K.; Tabuchi, Y.; Yokoyama, T.; Kitagawa,H.	2014	Insight into the mechanism of reproductive dysfunction caused by neonicotinoid pesticides.	Biological & pharmaceutical bulletin, (2014) Vol. 37, No. 9, pp. 1439-43	Does not fulfil criteria 8 (a quantitative relationship exists between the reported endpoint and risk assessment endpoints of growth, mortality, behaviour and/or reproduction). Investigation of effects on the reproductive function of mature male only or both young male and female quails following daily oral administration of the neonicotinoid clothianidin. The main endpoints were histological, which are not data requirements in ecotoxicological risk assessment. This reference is therefore considered not relevant.	17

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.1.1.3	Tokumoto, J.; Danjo, M.; Kobayashi, Y.; Kinoshita, K.; Omotehara, T.; Tatsumi, A.; Hashiguchi, M.; Sekijima, T.; Kamisoyama, H.; Yokoyama, T.; Kitagawa, H.; Hoshi, N.	2013	Effects of exposure to clothianidin on the reproductive system of male quails.	The Journal of veterinary medical science / the Japanese Society of Veterinary Science, (2013) Vol. 75, No. 6, pp. 755-60	Does not fulfil criteria 10 (Study should not differ significantly from recommended protocols). This study investigated the effects of a daily oral administration of clothianidin on the reproductive functions of mature male quails. Treated males were bred with untreated females to estimate the egg weights, and rates of fertilization and normal development. The testes, liver and spleen were examined histologically. There were no significant differences in apical endpoint (egg weights and fertilization rates). This study design does nto follow standard methods for determining effects on bird reproduction. Therefore this reference is considered not relevant.	49
CA 8.1.1.3	Lopez-Antia, A.; Ortiz-Santaliestra, M. E.M.; Mateo, R.; Ortiz-Santaliestra, M.E.M; Mougeot, F.	2015	Imidacloprid-treated seed ingestion has lethal effect on adult partridges and reduces both breeding investment and offspring immunity	ENVIRONMENTAL RESEARCH, (JAN 2015) Vol. 136, pp. 97-107	Neither thiamethoxam or clothianidin were tested in this study, therefore this reference is not relevant for this review.	721

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Moffat, C.; Pacheco, J. G.; Sharp, S.; Samson, A. J.; Bollan, K.A.; Huang, J.; Buckland, S.T.; Connolly, C. N.	2015	Chronic exposure to neonicotinoids increases neuronal vulnerability to mitochondrial dysfunction in the bumblebee (*Bombus terrestris*).	FASEB journal : official publication of the Federation of American Societies for Experimental Biology, (2015 May) Vol. 29, No. 5, pp. 2112-9	Does not fulfil criteria 8 (Effects are related to single test item, and a quantitative relationship exists between the reported endpoint and risk assessment endpoints of growth, mortality, behaviour and/or reproduction). This study tracks the dietary intake of clothianidin into the bumblebee brain and assesses its impact on neuronal function and colony performance, alone and in combination with raised levels of acetylcholine. The lab endpoints bear no relation to recommended endpoints used for risk assessment purpose (endpoints on growth, mortality, behavior, reproduction). Additionally, field experiments were not conducted with thiamethoxam or clothianidin. This reference is therefore considered not relevant.	10

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Rundlof M; Andersson GKS; Bommarco R; Fries I; Hederstrom V; Herbertsson L; Jonsson O; Klatt BK; Pedersen TR; Yourstone J; Smith HG	2015	Seed coating with a neonicotinoid insecticide negatively affects wild bees.	Nature, Vol. 521, No. 7550, pp. 77-80	This study investigated the effects of OSR treated seeds with clothianidin on wild and managed bees. Measurments were taken of wild bee density, nesting activity of the solitary bee *Osmia bicornis* L., colony development of the bumblebee *Bombus terrestris* L., and colony strength of the honeybee *Apis mellifera* L. To estimate exposure, the the proportion of OSR pollen collected by all three bee species was determined and the concentrations of clothianidin in bee-collected pollen and nectar was quantified. Additionally, flower samples were collected within 2 days of sowing in the permanent field borders adjacent to the OSR fields. There was a reduction in wild bee density, solitary bee nesting, and bumblebee colony growth and reproduction under field conditions. There was no significant effect on honeybee colonies. Bee colonies were exposed to a bee attractive flowering crop. This exposure is not relevant to the representative use considered in this assessment (sugar beet is harvested prior to flowering). The measured residues in pollen and nectar are from OSR, which is not a crop that is considered in this assessment. Additionally, effects from exposure to clothianidin residues in flower samples from adjacent fields is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite (not present in dust generated from sowing treated seeds).Therefore, this reference provides supplemental information only.	12

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Cutler G Christopher; Scott-Dupree Cynthia D; Sultan Maryam; McFarlane Andrew D; Brewer Larry	2014	A large-scale field study examining effects of exposure to clothianidin seed-treated canola on honey bee colony health, development and overwintering success.	PeerJ, Vol. 2, pp. e652	This study investigated the effects of OSR treated seeds with clothianidin on honeybee colonies. Colony weight gain, honey production, pest incidence, bee mortality, number of adults, and amount of sealed brood were assessed in each colony throughout summer and autumn. Overwintering success was also assessed. Samples of honey, beeswax, pollen, and nectar were collected and analyzed for clothianidin residues. There was no effects of exposure to clothianidin seed-treated OSR on any endpoint measured, suggesting clothianidin treated OSR seeds pose low risk to honey bees. Honeybee colonies were exposed to a bee attractive flowering crop. This exposure is not relevant to the representative use considered in this assessment (sugar beet is harvested prior to flowering). The measured residues in pollen and nectar are from OSR, which is not a crop that is considered in this assessment. Therefore, this reference provides supplemental information only.	20
CA 8.3.1	Larson, J. L.; Redmond, C. T.; Potter, D. A.	2014	Impacts of a neonicotinoid, neonicotinoid-pyrethroid premix, and anthranilic diamide insecticide on four species of turf-inhabiting beneficial insects.	Ecotoxicology (London, England), (2014 Mar) Vol. 23, No. 2, pp. 252-9	This study compares the impact of clothianidin, a premix (clothianidin + bifenthrin) and chlorantraniliprole on bumble bees. The main results of this trial were previously reported as part of a larger study (Ref ID 60, Clothianidin search) that compared both short-term and reproductive effects of exposure to clothianidin or chlorantraniliprole residues on B. impatiens. Therefore this reference is considered not relevant for this review.	25

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Larson J.L.; Redmond C.T.; Potter D.A.	2013	Assessing insecticide hazard to bumble bees foraging on flowering weeds in treated lawns.	PloS one, Vol. 8, No. 6, pp. e66375	A field study was carried out to examine the effect of clothianidin on the bumble bee (*Bombus impatiens*) when foraging on flowering weeds in a newly-treated lawn. A spray application of 450 g a.s./ha was made to turf with 30% blooming white clover. Bee colonies were introduced to the field 2 days after treatment and remained for 1 week. Existing label mitigation for thiamethoxam includes 'Mulch flowering groundcover crops immediately before treatment'. Additionally, the application rate used here far exceeds the foliar application rate for thiamethoxam supported in this assessment (20-50 g a.s./ha). Clothianidin is a plant metabolite; thus exposure residues would far exceed relevant field exposure values for clothianidin when thiamethoxam is applied. Therefore, this study provides supplemental information only.	60
CA 8.3.1	Scott-Dupree, C.D.; Conroy, L.; Harris, C.R.	2009	Impact of currently used or potentially useful insecticides for canola agroecosystems on *Bombus impatiens* (Hymenoptera: Apidae), *Megachile rotundata* (Hymentoptera: egachilidae), and *Osmia lignaria* Hymenoptera: Megachilidae).	Journal of economic entomology, (2009 Feb) Vol. 102, No. 1, pp. 177-82	Does not fulfil criteria 10 (Study condition should not differ significantly from recommended protocols). This laboratory study examined the effects of direct contact toxicity of clothianidin on 3 bee species: bumble bees [*Bombus impatiens*], alfalfa leafcutting bees [*Megachile rotundata* (F.)], and *Osmia lignaria* Cresson. 48h LD_{50} = 0.39, 0.08, 0.20 (wt:vol) (x10-3), repectively. For the contact toxicity an overspray method was used, which differs from the standard methods. Furthermore, the endpoint of the study is not given in µg/bee as used in risk assessment. Therefore, this study provides supplemental information only.	105

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Cutler G Christopher; Scott-Dupree Cynthia D	2007	Exposure to clothianidin seed-treated canola has no long-term impact on honey bees.	Journal of economic entomology, Vol. 100, No. 3, pp. 765-72	This study investigated the effects of canola treated seeds with clothianidin on honeybee colonies. Bee mortality, worker longevity, and brood development were assessed for 130 days. Overwintering success was also assessed. Samples of honey, beeswax, pollen, and nectar were collected and analyzed for clothianidin residues. Clothianidin residues were detected in honey, nectar, and pollen, while no residues were detected in any beeswax sample. There was no effects of exposure to clothianidin seed-treated canola on any endpoint measured, suggesting clothianidin treated canola seeds pose low risk to honey bees. Honeybee colonies were exposed to a bee attractive flowering crop. This exposure is not relevant to the representative use considered in this assessment (sugar beet is harvested prior to flowering). The measured residues in pollen and nectar are from canola, which is not a crop that is considered in this assessment. Therefore, this reference provides supplemental information only.	121
CA 8.3.1	Sanchez-Bayo, F.	2014	The trouble with neonicotinoids.	Science, (14 Nov 2014) Vol. 346, No. 6211, pp. 806-807	This reference is a “Perspectives” article discussing neonictinoid use and their effects on bees, as well as highlighting their effects on other non-target organisms. No new data is presented. Therefore this is not considered relevant for risk assessment purposes.	147

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Georgiadis, P.; Pistorius, J.; Heimbach, U.; Staehler, M.	2014	Manual application of insecticidal dust in semi-field experiments with honeybees	Proceedings: International conference on the German diabrotica research program. pp. 102. (2014) Tagungsband: Internationale Fachtagung zum Forschungsprogramm uber den Westlichen Maiswurzelbohrer . pp. 102. (2014)	This article is an abstract published from a conference (International Conference on the German Diabrotica Research Program. Berlin, GERMANY. November 14-16, 2012). A semi-field study was conducted to examine the effects from clothianidin treated maize seed dust (different rates and size particles) applied to flowering *Phacelia* on honeybee colonies. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Therefore this reference is not considered relevant.	170
CA 8.3.1	Dyer, D. G.; Xu, T.; Bondarenko, S.; Allen, R.	2014	Clothianidin: Potential accumulation/ bioavailability in soil and in corn and canola bee-relevant matrices.	Abstracts of Papers American Chemical Society, (AUG 10 2014) Vol. 248, pp. 606-AGRO.	This article is an abstract published from a conference (248th National Meeting of the American-Chemical-Society (ACS). San Francisco, CA, USA. August 10-14, 2014). No quantitative data is presented and it is not significantly detailed for assessment. Therefore this reference is not considered relevant.	179
CA 8.3.1	Bondarenko, S.; Rose, A.; Ansolabehere, M.; Allen, R.	2014	Clothianidin residues in pollen and nectar of cucurbits following different use patterns.	Abstracts of Papers American Chemical Society, (AUG 10 2014) Vol. 248, pp. 602-AGRO.	This article is an abstract published from a conference (248th National Meeting of the American-Chemical-Society (ACS). San Francisco, CA, USA. August 10 - 14, 2014). No quantitative data is presented and it is not significantly detailed for assessment. Therefore this reference is not considered relevant.	180

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Kimura, K.; Yoshiyama, M.; Saito, K.; Nirasawa, K.; Ishizaka, M.	2014	Examination of mass honey bee death at the entrance to hives in a paddy rice production district in Japan: the influence of insecticides sprayed on nearby rice fields.	Journal of Apicultural Research, (2014) Vol. 53, No. 5, pp. 599-606	Does not fulfil criteria 15 (Exposure route is clearly defined and suitably quantified). Field studies were conducted in a rice-producing district of northern Japan and honey bee hives were observed for mortality. Samples of pollen and honey bee corpses were analyzed for various pesticides (including clothianidin). As no information on specific application of pesticide products is recorded, the relationship of pesticide treatment and bee mortality can not be concluded from this study. Therefore this reference is not considered relevant.	181
CA 8.3.1	Pohorecka, K.; Skubida, P.; Semkiw, P.; Miszczak, A.; Teper, D.; Sikorski, P.; Zagibajlo, K.; Skubida, M.; Zdanska, D.; Bober, A.	2013	Effects of exposure on honey bee colopnies to necotinoid seed-treated maize crops.	Journal of Apicultural Science, (2013) Vol. 57, No. 2, pp. 199-208	This article was not available from our copyright-cleared document delivery sources. While we placed an order to purchase this document from an advanced reference source, we were unable to obtain this article. The effects to honeybee colonies (*Apis mellifera L.*) during and after exposure to flowering maize (*Zea mays* L), grown from seeds coated with clothianidin was assessed in field-realistic conditions. As the crop used in this study is not included in this assessment as a representative use and as the abstract is not significantly detailed for assessment, this reference is not considered relevant.	189
CA 8.3.1	Andrew, D.; Brewer, L.; Cutler, G. C.; Scott-Dupree, C. D.; Sultan, M.; McFarlane	2013	Large-scale field study examining potential impacts on honey bees of exposure to clothianidin seed-treated canola.	Abstracts of Papers American Chemical Society, (SEP 8 2013) Vol. 246, pp. 149-AGRO.	This article is an abstract published from a conference (246th National Meeting of the American-Chemical-Society (ACS). Indianapolis, IN, USA. September 08-12, 2013). No data is presented and it is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	191

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Fell, R.	2013	Honey bee colony health, bee decline, and pesticides.	Abstracts of Papers American Chemical Society, (SEP 8 2013) Vol. 246, pp. 144-AGRO.	This article is an abstract published from a conference (246th National Meeting of the American-Chemical-Society (ACS). Indianapolis, IN, USA. September 08-12, 2013). No data is presented and it is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	192
CA 8.3.1	Matsumoto, T.	2013	Short and long-term effects of necotinoid application in rice fields, on the mortality and colony collapse of honeybees (Apis mellifera)	Journal of Apicultural Science, (2013) Vol. 57, No. 2, pp. 21-35	This article was not available from our copyright-cleared document delivery sources. While we placed an order to purchase this document from an advanced reference source, we were unable to obtain this article. The effects to honeybee colonies (*Apis mellifera* L.) after exposure of clothianidin to rice fields was assessed. Honeybee hives were placed at the distance of 0, 30, 60, and 90 m. As the crop used in this study is not included in this assessment as a representative use and as the abstract is not sufficiently detailed for assessment, this reference is not considered relevant.	196
CA 8.3.1	Chauzat, M..; Martel, A.; Blanchard, P.; Clement, M.; Schurr, F.; Lair, C.; Ribiere, M.; Wallner, K.; Rosenkranz, P.; Faucon, J.	2010	A case report of a honey bee colony poisoning.	Journal of Apicultural Research, (2010) Vol. 49, No. 1, Sp. Iss. SI, pp. 113-115	This article describes an investigation into bee mortality events in Spring 2008 in Germany. Analaytical results confirmed the cause as clothianidin poisoning. The pesticide was released during sowing of poorly treated maize seeds. Additional factors contributed to a worst-case exposure scenario: poor seed dressing bonding, use of pneumatic seed drills, sowing delayed due to bad weather conditions coinciding with OSR flowering and dry windy weather. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Therefore this reference is not considered relevant.	220

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Kunz, N.; Dietzsch, A.; Frommberger, M.; Wirtz, I.; Staehler, M.; Frey, E.; Illies, I.; Dyrba, W.; Alkassab, A.; Pistorius, J.	2014	Neonicotinoids and bees: effects on honeybees, bumblebees and solitary bees in oilseed rape grown from Clothianidin-treated seed.	Berichte aus dem Julius Kuehn-Institut (2014), Nr. 177, 18 p. ISSN: 1866-590X	This article is an abstract published from a conference (Siebentes Nachwuchswissenschaftlerforum 2014, Quedlinburg,Germany, 26-28. November, 2014). Field trials and semi-field trials were conducted in five federal states in spring 2014 using the Western honeybee (*Apis mellifera* L.), the bufftailed bumblebee (*Bombus terrestris* L.) and the red mason bee (*Osmia bicornis* L.) were placed right next to flowering oilseed rape (*Brassica napus*). This abtract is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	292
CA 8.3.1	Nadaf, H. A.; Yadav, G. S.; Kaushik, H. D.; Sharma, S. K.	2013	Toxicity of new molecules of insecticides against honeybee, *Apis mellifera* L.	Trends in Biosciences (2013), Volume 6, Number 4, pp. 445-447.	Does not fulfil criteria 5 (Several dose levels tested, at least 3 to establish a dose-response). Does not fulfil criteria 10 (Study conditions should not differ significantly from recommended protocols). Insecticides (including clothianidin) were tested for their safety against the honeybee, *Apis mellifera* L., following a dry film method. Mortality after 24 hours was recorded. Clothainidin (50 WDG) at 20 g a.s./ha caused 100% mortality after 24 hours. The exposure method used does not correspond to recommended exposure routes for laboratory tests (oral or contact). Additionally, only one concentration was tested. Therefore, this reference is considered not relevant for this review.	302
CA 8.3.1	Staehler, M.; Heimbach, U.; Schwabe, K.; Pistorius, J.; Georgiadis, P. T.	2012	Ecotoxicokinetics of clothianidin on honeybees in open field - first results. Zur Oekotoxikokinetik von Clothianidin auf Bienen im Freiland – erste Ergebnisse.	Julius-Kuehn-Archiv (2012), Number 438, pp. 466-467. ISSN: 1868-9892	This article is an abstract published from a conference (Deutsche Pflanzenschutztagung, Technische Universitaet Braunschweig, 10-14 September 2012). It is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	333

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Sgolastra, F; Renzi, T; Draghetti, S; Medrzycki, P; Lodesani, M; Maini, S; Porrini, C	2012	Effects of neonicotinoid dust from maize seed-dressing on honey bees.	Bulletin of Insectology (2012), Vol. 65, Number 2, pp. 273-280	In this study, the effects on honey bees of clothianidin derived from maize seed-dressing (Poncho) in laboratory (test by indirect contact) and in semi-field conditions were investigated. Laboratory tests were also conducted with spray formulation. For the indirect contact test, exposure was done by letting the bees walk on treated apple leaves. The exposure method used does not correspond to recommended exposure routes for laboratory tests (oral or contact). Effects from exposure to clothianidin residues in flower samples from adjacent fields is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite (not present in dust generated from sowing treated seeds).Therefore, this reference provides supplemental information only.	337
CA 8.3.1	Reetz, J. E.; Zuehlke, S.; Spiteller, M.; Wallner, K.	2011	Neonicotinoid insecticides translocated in guttated droplets of seed-treated maize and wheat: a threat to honeybees?	Apidologie, Volume 42, Number 5, pp. 596-606	This study investigated residues of insecticides (including clothianidin) in guttation fluid of triticale and maize plants grown from treated seeds. Additionally, observations of water foraging bees were made during the sampling of guttation fluid. No bees were observed collecting guttated fluid from triticale or maize. But they were observed collecting the exuded fluids from *Potentilla* plants along the edge of the experimental plots. The crops used in this study are not included in this assessment as a representative use. Therefore, this study provides supplemental information only.	367

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Georgiadis, P. T.; Pistorius, J.; Heimbach, U.	2011	Dust in the wind - drift of dust containing insecticides - a risk for honey bees (*Apis mellifera* L.)? Dust in the wind - Abdrift insektizidhaltiger Staeube - ein risiko fuer Honigbienen (*Apis mellifera* L.)?	Julius-Kuehn-Archiv (2011), Number 430, pp. 15-19, 2 refs. ISSN: 1868-9892	This article is published from a conference (Third Young Scientists Forum, Quedlinburg, Germany, 23-25 November, 2010). Drift trials were conducted during the sowing of winter oilseed rape and maize seeds treated with clothianidin. The impact of dust drift on individual bees and colonies were examined. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Therefore this reference is not considered relevant.	394
CA 8.3.1	Bischoff, G.	2010	Chemical bee investigation - details of the new procedure and selected results since 2008. Chemische Bienenuntersuchung - Details des neuen Verfahrens und ausgewaehlte Ergebnisse seit 2008.	Julius-Kuehn-Archiv (2010), Number 428	This article is a short summary of a chemical bee investigation by the JKI in 2008. It is not sufficiently detailed for assessment. Additionally, details of the studies are published elsewere (Ref ID 220, 442 and 445; Clothianidin search). Therefore this reference is not considered relevant.	397
CA 8.3.1	Friessleben, R.; Schad, T.; Schmuck, R.; Schnier, H.; Schoening, R.; Nikolakis, A.	2010	An effective risk management approach to prevent bee damage due to the emission of abraded seed treatment particles during sowing of neonicotinoid treated maize seeds	Aspects of Applied Biology (2010), Nr. 99, pp. 277-282, 3 refs. ISSN: 0265-1491	This article describes a study with clothianidin treated maize seeds to investigate the impact of seed dressing quality and different maize sowing machinery on dust drift. As this study compares different maize sowing mitigation, this reference is not considered relevant to this review.	401

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Jacobs, A.; Bischoff, G.; Buettner, C.; Pestemer, W.	2010	Residue behaviour of selected pesticides in potatoes and bees. Rueckstandsverhalten von ausgewaehlten Pflanzenschutzmitteln in/auf Kartoffelpflanzen und Bienen.	Julius-Kuehn-Archiv (2010), Number 424, 34 p. ISSN: 1868-9892	This article is an abstract published from a conference (Nachwuchswissenschaftlerforum, Berlin-Dahlem, Germany, 17-19 November, 2009). It is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	418
CA 8.3.1	Georgiadis, P. T.; Pistorius, J.; Heimbach, U.	2010	Gone with the wind - drift of abrasive dust from seed treatments - a risk for honey bees (*Apis mellifera* L.)? Vom Winde verweht - Abdrift von Beizstaeuben - ein Risiko fuer Honigbienen (*Apis mellifera* L.)?	Julius-Kuehn-Archiv (2010), Number 424, 33 p., 1 refs. ISSN: 1868-9892	This article is an abstract published from a conference (Nachwuchswissenschaftlerforum, Berlin-Dahlem, Germany, 17-19 November, 2009). This article describes field and semi-field tests with clothianidin treated OSR seeds to investigate the impact of dust drift on honey bee mortality up to 48 hours. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Therefore this reference is not considered relevant.	419
CA 8.3.1	Wallner, K.;	2009	Sprayed and seed dressed pesticides in pollen, nectar and honey of oilseed rape.	Julius-Kuehn-Archiv (2009), Number 423, pp. 152-153. ISSN: 1868-9892	This article is published from a conference (Hazards of pesticides to bees. 10th International Symposium of the ICP-Bee Protection Group. Bucharest, Romania, 8-10 October, 2008). A field study was conducted using clothianidin treated OSR seed to measure the residues in pollen and nectar loads of returning honey bee foragers. Even with low quantitation limits (0.001 mg/kg), this insecticide was not detected in pollen or honey. This article is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	438

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Nikolakis, A.; Chapple, A.; Friessleben, R.; Neumann, P.; Schad, T.; Schmuck, R.; Schnier, H. F.; Schnorbach, H. J.; Schoening, R.; Maus, C.	2009	An effective risk management approach to prevent bee damage due to the emission of abraded seed treatment particles during sowing of seeds treated with bee toxic insecticides.	Julius-Kuehn-Archiv (2009), Number 423, pp. 132-148, 2 refs. ISSN: 1868-9892	This article is published from a conference (Hazards of pesticides to bees. 10th International Symposium of the ICP-Bee Protection Group. Bucharest, Romania, 8-10 October, 2008). This article describes results of a detailed analysis of the bee incidents in spring 2008 in Germany (referred to in Ref ID 220), it summarizes the outcome of a joint research initiative aiming at the development of appropriate technical solutions to ensure safety of seed dressing products and presents results of field trials conducted to evaluate the effectiveness of the developed optimizations. It also outlines an exemplary bee risk assessment under consideration of the described mitigation measures. The focus of this article is mitigation measures for treated maize seed (seed coatings and drilling), as well as clothainidin in dust particles. Clothianidin in dust is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Therefore this reference is not considered relevant.	442
CA 8.3.1	Pistorius, J; Bischoff, G; Heimbach, U; Staehler, M;	2009	Bee poisoning incidents in Germany in spring 2008 caused by abrasion of active substance from treated seeds during sowing of maize.	Julius-Kuehn-Archiv (2009), Number 423, pp. 118-126, 9 refs. ISSN: 1868-9892	This article is published from a conference (Hazards of pesticides to bees. 10th International Symposium of the ICP-Bee Protection Group. Bucharest, Romania, 8-10 October, 2008). This article reports more detailed information on the bee incidents in spring 2008 in Germany (referred to in Ref ID 220, Clothianidin search). Methods and results for the residue analysis of bees and dust are presented. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. This reference is not considered relevant.	445

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Janke, M.; Rosenkranz, P.	2009	Periodical honey bee colony losses in Germany: preliminary results from a four years monitoring project.	Julius-Kuehn-Archiv (2009), Number 423, pp. 108-117, 1 refs. ISSN: 1868-9892	This article is published from a conference (Hazards of pesticides to bees. 10th International Symposium of the ICP-Bee Protection Group. Bucharest, Romania, 8-10 October, 2008). Winter losses of bee colonies were evaluated over 4 years from the the German Bee Monitoring Project. The winter losses were significantly correlated with Varroa infestations and virus infections in autumn. It was concluded that no acute effects on honey bees have to be expected on the basis of the evaluated residue data. Thiamethoxam and clothianidin were not detected in any samples. As this article discusses interim results from a monitoring program, this reference is not considered relevant.	446
CA 8.3.1	Pistorius, J.; Bischoff, G.; Heimbach, U	2009	Bee poisoning by abrasion of active substances from seed treatment of maize during seeding in spring 2008. Bienenvergiftung durch Wirkstoffabrieb von Saatgutbehandlungsmitteln waehrend der Maisaussaat im Fruehjahr 2008.	Journal fuer Kulturpflanzen (2009), Volume 61, Number 1, pp. 9-14	This article describes an investigation into bee mortality events in Spring 2008 in Germany.. Analaytical results confirmed the cause as clothianidin poisoning. The pesticide was released during sowing of poorly treated maize seeds. Additional factors contributed to a worst-case exposure scenario: poor seed dressing bonding, use of pneumatic seed drills, bad weather and “double contamination” via oral and contact exposure. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Therefore this reference is not considered relevant.	457

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Bailey, J.; Scott-Dupree, C.; Harris, R.; Tolman, J.; Harris, B.	2005	Contact and oral toxicity to honey bees (*Apis mellifera*) of agents registered for use for sweet corn insect control in Ontario, Canada.	Apidologie (2005), Volume 36, Number 4, pp. 623-633	Bioassays were conducted to compare direct and residual contact and oral toxicities of clothianidin to honey bees. A direct contact assay using direct overspray as exposure method was conducted, as well as a residual contact assays, were forager bees were exposed to treated non-transgenic sweet corn tassels. Furthermore, oral consumption of pollen collected from treated seed was assessed. The exposure methods used do not correspond to recommended exposure routes for laboratory tests (oral or contact). Furthermore, the endpoint of the study is not given in µg/bee as used in risk assessment. Therefore, this study provides supplemental information only.	519
CA 8.3.1	Neal, K.	2013	Is planting corn killing bees?	Abstracts of Papers, 246th ACS National Meeting & Exposition, Indianapolis, IN, United States, September 8-12, 2013 (2013)	This article is an abstract published from a conference (246th ACS National Meeting & Exposition, Indianapolis, IN, United States, September 8-12, 2013). No data is presented and it is not sufficiently detailed for assessment. Therefore this reference is not considered relevant.	572
CA 8.3.1	Girolami, V.; Marzaro, M.; Vivan, L.; Mazzon, L.; Greatti, M.; Giorio, C.; Marton, D.; Tapparo, A.	2012	Fatal powdering of bees in flight with particulates of neonicotinoids seed coating and humidity implication	Journal of Applied Entomology (2012), 136(1-2), 17-26	The effect of direct aerial powdering was tested on foragers in free flight near the drilling machine. Bees were conditioned to visit a dispenser of sugar solution while a drilling machine was sowing corn along the flight path. Samples of bees were captured on the dispenser, caged and held in the laboratory. After the sowing, bee mortality in front of the hives was observed. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Therefore this reference is not considered relevant.	600

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Trenkle, A.	2009	Harm to bees in 2008 in Rheintal - analysis, causes, consequences	VDLUFA-Schriftenreihe (2009), 65 (Pt. 2, Produktivitaet und Umweltschonung in der Landwirtschaft: ein Widersprucht?), 22-41.	This article describes a bee monitoring program in Germany, that was set up following bee mortality incidents. Results indicated that mortality occurred mainly in maize growing areas, originating from dust containing clothianidin. As maize is not a represenetative crop considered here and as effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite, this reference provides supplemental information only.	641
CA 8.3.1	Knauer, K	2010	Post registration monitoring of effects of Clothianidin on bee colonies	20th Annual Meeting of the Europe branch of the Society of Environmental Toxicology and Chemistry (SETAC 2010), Palacio de Congresos y Exposiciones - FIBES, Seville, 23 May 2010-27 May 2010	This article is an abstract published from a conference (20th Annual Meeting of the Europe branch of the Society of Environmental Toxicology and Chemistry (SETAC 2010), Palacio de Congresos y Exposiciones - FIBES, Seville, 23 May 2010-27 May 2010) A field study was conducted to examine the effects of clothianidin on honeybee colonies. The focus was on effects of dust drift and guttation. Effects on bees from dust containing clothianidin is not relevant to a thiamethoxam risk assessment, as clothianidin is a plant and soil metabolite. Additionally, maize is not a representative crop considered in this assessment, thus effects on bees feeding on maize guttation is supplemental information only. Therefore this reference is not considered relevant.	713
CA 8.3.1	Anonymous	2008	Pesticide penalty	Ecologist. Vol. 38, no. 9, pp. 11-11. Nov. 2008.	This reference is a “News” article reporting the Soil Association call for the UK government to ban neonictinoid pesticides uses, as other Eurpoean countries have. No new data is presented. Therefore this is not considered relevant for risk assessment purposes.	714

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Cutler, L. G.; Scott-Dupree, C. D.; Chalmers, A.	2006	An Investigation of Potential Long-Term Impact of Clothianidin Seed-Treated Canola on Honey Bees, *Apis Mellifera*	Conference: 27th Annual Meeting of the Society of Environmental Toxicology and Chemistry (SETAC 2006), Montreal, Quebec (Canada), 3 Nov 2006 - 9 Nov 2006	This article is an abstract published from a conference (27th Annual Meeting of the Society of Environmental Toxicology and Chemistry (SETAC 2006), Montreal, Quebec (Canada), 3 Nov 2006 - 9 Nov 2006). It is not sufficiently detailed for assessment. Additionally, the data referenced is described fully in Cutler and Scott-Dupree 2007 (Ref. ID 121, Clothianidin search). Therefore this reference is not considered relevant.	715
CA 8.3.1	Feltham, H.; Park, K.; Goulson, D.	2014	Field realistic doses of pesticide imidacloprid reduce bumblebee pollen foraging efficiency	Ecotoxicology, (APR 2014) Vol. 23, No. 3, pp. 317-323	Neither thiamethoxam or clothianidin were tested in this study, therefore this reference is not relevant for this review.	724
CA 8.4	Wang Y.; Wu S.; Chen L.; Wu C.; Yu R.; Wang Q.g; Zhao X.	2012	Toxicity assessment of 45 pesticides to the epigeic earthworm Eisenia fetida.	Chemosphere, (2012 Jul) Vol. 88, No. 4, pp. 484-91	In this study, acute toxicity to earthworms was investigated, which is not a data requirement due to European risk assessment procedures anymore. This reference is therefore considered not relevant.	70
CA 8.4	Wang, Y.; Cang, T.; Zhao, X.; Yu, R.; Chen,L. .; Wu, .; Wang, Q.	2012	Comparative acute toxicity of twenty-four insecticides to earthworm, *Eisenia fetida*.	Ecotoxicology and Environmental Safety, (1 May 2012) Vol. 79, pp. 122-128	In this study, acute toxicity to earthworms was investigated, which is not a data requirement due to European risk assessment procedures anymore. This reference is therefore considered not relevant.	158
CA 8.7	Gibbons D.; Morrissey C.; Mineau P.	2015	A review of the direct and indirect effects of neonicotinoids and fipronil on vertebrate wildlife.	Environmental science and pollution research international, (2015 Jan) Vol. 22, No. 1, pp. 103-18	This article reviews 150 studies for their direct (toxic) and indirect (e.g. food chain) effects on vertebrate wildlife—mammals, birds, fish, amphibians and reptiles. The focus is on three insecticides (including clothainidin). No new data is presented. This reference is therefore considered not relevant.	7

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.7	Larson J.L.; Redmond C.T.; Potter D.A.	2012	Comparative impact of an anthranilic diamide and other insecticidal chemistries on beneficial invertebrates and ecosystem services in turfgrass.	Pest management science, (2012 May) Vol. 68, No. 5, pp. 740-8	Effects of clothianidin on non-target soil organisms were assessed under field conditions (field studies on golf courses) in Kentucky, USA. Clothianidin was applied at 150 g a.s./ha. Non-target organisms were assessed via pitfall traps (epigeal predators), Tullgren funnel extraction (soil microarthropods), hand sorting (earthworms), counting ant mounds, earthworm casts on tees and putting greens, assessing predation on sentinel pest eggs and litter bags to assess impacts on rates of decomposition. The study was conducted in the USA on a golf course. This use is not one of the representative uses considered in this assessment (vegetable crops - lettuce and potatoes). Additionally, clothianidin is a soil metabolite of thiamethoxam and the rate of clothianidin used in this study far exceeds even the representative rates of thiamethoxam considered here (20-50 g thiamethoxam/ha). Therefore this reference is considered as supplemental information only.	67
CA 8.7	Aramaki, Hironori	2014	Foreword.	Biological and Pharmaceutical Bulletin, (2014) Vol. 37, No. 9, pp. 1429	Foreword of an article on Mechanistic Understanding of Endocrine Disruption, therefore not relevant for this review.	146
CA 8.7	Cerevkova, A.; Cagan, L'.	2014	Influence of insecticides to soil nematode communities	Journal of Nematology, (JUN 2014) Vol. 46, No. 2, pp. 143.	This article is an abstract published from a conference (6th International Congress of Nematology. Cape Town, South Africa. May 04-09, 2014). Soil nematode communities were studied after application of insecticides in maize fields in Slovakia. Nematodes are not a required test species in ecotoxicological risk assessment. This reference is therefore considered not relevant.	185

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.7	So, B. H.; Kim, H. M.	2010	Two Cases of Severe Neonicotinoid Intoxication.	Clinical Toxicology, (JUL 2010) Vol. 48, No. 6, pp. 611.	This article only describes human intoxiciation from oral clothianidn exposure. Therefore it is not relevant for this review.	216
CA 8.7	Goulson, D.	2013	An overview of the environmental risks posed by neonicotinoid insecticides.	Journal of Applied Ecology (2013), Vol. 50, Nr. 4, pp. 977-987	This article provides a review of the available scientific data on neonictinoid pesticides (including thiamethoxam and clothianidin). It discusses the environmental fare of the compounds and the toxicity to a variety of non-target organisms. No new data is presented. Therefore this reference is considered not relevant.	300
CA 8.7	Cerevkova, A.; Cagan, L.	2013	The influence of Western corn rootworm seed coating and granular insecticides on the seasonal fluctuations of soil nematode communities in a maize field.	Helminthologia (2013), Vol. 50, Nr. 3, pp. 205-214	Does not fulfil criteria 14 (applicable test species). In this study the effect of thiamethoxam on nematode communities in a maize fields was investigated. Nematodes are not a required test species in ecotoxicological risk assessment. This reference is therefore considered not relevant.	306
Not found in search (but referenced)						
CA 8.1	Mineau, P., Palmer, C.,	2013	Neonicotinoid insecticides and birds: the impact of the nation's most widely used insecticides on birds.	American Bird Conservancy	This article was referenced in a review article (Ref ID 262, Initial search). It did not appear in the literature search, as it is not available in any of the databases searched. This report reviews the effects of neonictinoids (including thiamethoxam and clothianidin) on avian species. No new data is presented. Therefore this reference is considered not relevant.	n/a

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.2.2	Bose S, Nath S, Sahana SS.	2011	Toxic impact of thiamethoxam on the growth performance and liver protein concentration of a freshwater fish *Oreochromis niloticus* (Trewavas).	Ind J Fund Appl Life Sci 2011;1:274 – 80	This article was referenced in a review article (Ref ID 13, Initial search). It did not appear in the literature search, as the journal is not indexed. This study investigated the effects of thiamethoxam on growth and liver total protein of *Oreochromis niloticus* (Trewavas). It it not clear where the test organims were obtained, as they were "collected from the local market". Fish were exposed to five concentrations (12.5, 25, 50, 100, 150 mg/L), for 7 or 14 days. Growth was monitored in respect to body weight, length and breadth. Chronic toxicity tests of this length are no longer a data requirement, as a fish ELS test is now the recommended test method. The source of the test organisms is unknown, therefore they may have been previously exposed to the test material or other contaminants. Additionally, there is ample regulatory references for fish chronic toxicity, none of which indicate that fish would drive the risk assessment. Also, the effects seen were at concentrations much in exceedance of expected PECsw. Therefore this article that was not found in our literature search is considered not relevant.	n/a

CA data point number	Author(s)	Year	Title	Source	Reason(s) for not including the study in the dossier	Ref. ID
CA 8.3.1	Food & Environment Research Agency	2013	Effects of neonicotinoid seed treatments on bumble bee colonies under field conditions	Food & Environment Research Agency; Available at http://FERA.co.uk/ccss/documents/defraBumbleBeeReportPS2371V4a.pdf	This article was referenced in opinion articles (Ref ID 525, Initial search; Ref ID 8, Top-up search). It did not appear in the literature search, as it was not indexed by the databases used. This study investigated the effects of neonictinoid treated OSR seeds (including clothianidin) on bumble bee colonies. Colony mass and fitness, number of foragers and foraging activity, along with overwintering success were assessed. Samples of pollen and nectar (from colonies, foragers and flowering crop) were collected and analyzed for pesticide residues. Bumble bee colonies were exposed to a bee attractive flowering crop. This exposure is not relevant to the representative use considered in this assessment (sugar beet is harvested prior to flowering). Therefore, this reference provides supplemental information only.	n/a
	Sánchez-Bayo, F., Tennekes, H.A., Goka, K.	2013	Impact of systemic insecticides on organisms and ecosystems	Insecticides — Development of Safer and More Effective Technologies	This article was referenced in a review article (Ref ID 262, Initial search). It did not appear in the literature search, as only the chemical term imidacloprid was indexed. This article is a chapeter from the book 'Insecticides — Development of Safer and More Effective Technologies'. It reviews the effects of systemic insecticides (including thiamethoxam and clothianidin) on non-target organisms and communities. No new data is presented. Therefore this reference is considered not relevant.	n/a

All documents listed in Table 9.6-2 and not excluded (i.e. not listed in Table 9.6-4) are given below. These documents have been summarised in the relevant Document M-CA where an assessment of reliability has been conducted and the conclusions documented.

Table 9.6-5: List of references which are discussed further in MCA Section 8 (listed by data point number)

CA Data Point/ Ref Number	Authors	Year	Title	Source (Overseas evaluation report code) *	Ref ID
CA 8.1.1.1/01	Ivanova, R.; Hristev, H.; Genchev, A.	2013	Determining LD_{50} of the neonicotinoid insecticides Actara and Confidor in birds.	Agrarni Nauki (2013), Volume 5, Number 14, pp. 237-241	587 (Initial search)
CA 8.2.8/01	Barbee, G.C.; Stout, J.	2009	Comparative acute toxicity of neonicotinoid and pyrethroid insecticides to non-target crayfish (*Procambarus clarkii*) associated with rice-crayfish crop rotations.	Pest management science, (2009 Nov) Vol. 65, No. 11, pp. 1250-6. (a)	170 (Initial search)
CA 8.2.8/02	Stevens, M.M.; Helliwell, S.; Hughes, P.A.	2005	Toxicity of *Bacillus thuringiensis* var. israelensis formulations, spinosad, and selected synthetic insecticides to *Chironomus tepperi* larvae.	Journal of the American Mosquito Control Association, (December 2005) Vol. 21, No. 4, pp. 446-450.	292 (Initial search)
CA 8.2.8/03	Deng, L.; Zhang, L.; Zhang, Y.; He, W.; Feng, L.; Jiang, H.	2013	Acute immobilization of four neonicotinoid insecticides to *Daphnia magn*a Straus	Nongyao Kexue Yu Guanli (2013), 34 (6), 23-25	1576 (Initial search)
CA 8.2.8/04	Ugurlu, P.; Unlu, E.; Satar, E.I.	2015	The toxicological effects of thiamethoxam on *Gammarus kischineffensis* (Schellenberg 1937) (Crustacea: Amphipoda).	Environmental toxicology and pharmacology, (2015 Mar) Vol. 39, No. 2, pp. 720-6 (a)	1 (Top-up search)
CA 8.2.8/05	de Perre C.; Murphy T.M.; Lydy M.J.	2015	Fate and effects of clothianidin in fields using conservation practices.	Environmental toxicology and chemistry / SETAC, (2015 Feb) Vol. 34, No. 2, pp. 258-65	3 (Clothianidin search)
CA 8.2.8/06	Whiting, S.A.; Lydy, M.J.	2014	A site-specific ecological risk assessment for corn-associated insecticides.	Integrated environmental assessment and management, (2014 Dec 30).	35 (Clothianidin search)
CA 8.2.8/07	Hayasaka, D.; Suzuki, K.; Nomura, T.; Nishiyama, M.; Nagai, T.; Sanchez-Bayo, F.; Goka, K.	2013	Comparison of acute toxicity of two neonicotinoid insecticides, imidacloprid and clothianidin, to five cladoceran species.	Journal of Pesticide Science (2013), Volume 38, Number 1/2, pp. 44-47	317 (Clothianidin search)

CA Data Point/ Ref Number	Authors	Year	Title	Source (Overseas evaluation report code) *	Ref ID
CA 8.2.8/08	Riaz, M.A.; Chandor-Proust, A.; Dauphin-Villemant, C.; Poupardin, R.; Jones, C.M.; Strode, C.; Régent-Kloeckner, M.; David, J.P.; Reynaud, S.	2013	Molecular mechanisms associated with increased tolerance to the neonicotinoid insecticide imidacloprid in the dengue vector *Aedes aegypti*.	Aquatic Toxicology 126 (2013) 326– 337.	n/a
CA 8.3.1.4/01	Illarionov, A. I. ; Derkach, A. A	2008	Toxicity and Hazard of Neonicotinoids for Honeybees.	Agrokhimiya, (OCT 2008) No. 10, pp. 74-81. (b)	390 (Initial search)
CA 8.3.1.4/02	Thompson, H.M.; Fryday, S.L.; Harkin, S.; Milner, S.	2014	Potential impacts of synergism in honeybees (*Apis mellifera*) of exposure to neonicotinoids and sprayed fungicides in crops.	Apidologie (2014), Volume 45, Number 5, pp. 545-553, 30 refs (c, d)	449 (Initial search)
CA 8.3.1.4/03	Laurino, D.; Manino, A.; Patetta, A.; Porporato, M.	2013	Toxicity of neonicotinoid insecticides on different honey bee genotypes.	Bulletin of Insectology (2013), Vol. 66, Number 1, pp. 119-126.	611 (Initial search)
CA 8.3.1.4/04	Laurino, D.; Porporato, M.; Patetta, A.; Manino, A.	2011	Toxicity of neonicotinoid insecticides to honey bees: laboratory tests.	Bulletin of Insectology (2011), Volume 64, Number 1, pp. 107-113	858 (Initial search)
CA 8.3.1.4/05	Falco, J.R.P.; Hashimoto, J.H.; Fermino, F.; de Toledo, V.A.A.	2010	Toxicity of thiamethoxam, behavioral effects and alterations in chromatin of *Apis mellifera* L., 1758 (Hymenoptera; Apidae).	Research Journal of Agriculture and Biological Sciences (2010), Volume 6, Number 6, pp. 823-828	909 (Initial search)
CA 8.3.1.4/06	Laurino, D.; Manino, A.; Patetta, A.; Ansaldi, M.; Porporato, M.	2010	Acute oral toxicity of neonicotinoids on different bee strains.	Redia (2010), Volume 93, pp. 99-102, 14 (c, d)	967 (Initial search)
CA 8.3.1.4/07	Pei, H.; Ou, .X; Yu, W.; Yi, Z.; Bai, J.; Gao, D.	2013	Acute toxicity of four insecticides to honeybee *Apis mellifera*	Shijie Nongyao (2013), 35 (4), 50-51, 54.	1570 (Initial search)
CA 8.3.1.4/08	Valdovinos-Nunez, G.R.; Quezada-Euan, J.J.G.; Ancona-Xiu, P.; Moo-Valle, H.; Carmona, A.; Sanchez, E.R.	2009	Comparative toxicity of pesticides to stingless bees (Hymenoptera: Apidae: Meliponini)	Journal of Economic Entomology (2009), 102 (5), 1737-1742 (c, d)	1672 (Initial search)

CA Data Point/ Ref Number	Authors	Year	Title	Source (Overseas evaluation report code) *	Ref ID
CA 8.3.1.4/09	Thompson, H.M.; Wilkins, S.; Harkin, S.; Milner, S.; Walters, K.F.	2013	Neonicotinoids and bumblebees (*Bombus terrestris*): effects on nectar consumption in individual workers	Pest management science, (2014 Jul 30)	36 (Initial search)
CA 8.3.1.4/10	Kessler S.C; Simcock, K.L.; Softley, S.; Wright, G.A.; Tiedeken, E.J.; Stout, J.C.; Mitchell, J.	2015	Bees prefer foods containing neonicotinoid pesticides.	Nature, (2015 May 7) Vol. 521, No. 7550, pp. 74-6 (c, d)	7 (Top-up search)
CA 8.3.1.4/11	Aliouane, Y.; El Hassani, A.K.; Gary, V.; Armengaud, C.; Lambin, M.; Gauthier, M.	2008	Effects of sublethal doses of acetamiprid and thiamethoxam on the behavior of the honeybee (*Apis mellifera*).	Archives of environmental contamination and toxicology, (2008 May) Vol. 54, No. 4, pp. 653-61.	197 (Initial search)
CA 8.3.1.4/12	Aliouane, Y.; El Hassani, A.K.; Gary, V.; Armengaud, C.; Lambin, M.; Gauthier, M.	2009	Subchronic exposure of honeybees to sublethal doses of pesticides: effects on behavior.	Environmental toxicology and chemistry / SETAC, (2009 Jan) Vol. 28, No. 1, pp. 113-122	183 (Initial search)
CA 8.3.1.4/13	Williamson, S.; Willis, J.; Wright, G.A.	2014	Exposure to neonicotinoids influences the motor function of adult worker honeybees.	Ecotoxicology (London, England), (2014 Oct) Vol. 23, No. 8, pp. 1409-18.	7 (Initial search)
CA 8.3.1.4/14	Henry, M.; Beguin, M.; Requier, F.; Rollin, O.; Odoux, J.F.; Aupinel, P.; Aptel, J.; Tchamitchian, S.; Decourtye, A.	2012	A common pesticide decreases foraging success and survival in honey bees.	Science (New York, N.Y.), (2012 Apr 20) Vol. 336, No. 6079, pp. 348-50. (b, e)	98 (Initial search)
CA 8.3.1.4/15	Cresswell J.E.; Thompson H.M	2012	Comment on "a common pesticide decreases foraging success and survival in honey bees".	Science, (21 Sep 2012) Vol. 337, No. 6101, pp. 1453-c (e)	277 (Initial search)
CA 8.3.1.4/16	Fischer, .J; Muller, T.; Grunewald, B.; Spatz, A.; Greggers, U.; Menzel, R.	2014	Neonicotinoids interfere with specific components of navigation in honeybees.	PloS one, (2014) Vol. 9, No. 3, pp. e91364	34 (Clothianidin search)
CA 8.3.1.4/17	Schneider, C.W.; Tautz, J.; Grunewald, B.; Fuchs, S.	2012	RFID tracking of sublethal effects of two neonicotinoid insecticides on the foraging behavior of *Apis mellifera.*	PloS one, (2012) Vol. 7, No. 1, pp. e30023 (e)	61 (Clothianidin search)

CA Data Point/ Ref Number	Authors	Year	Title	Source (Overseas evaluation report code) *	Ref ID
CA 8.3.1.4/18	Matsumoto, T.	2013	Reduction in homing flights in the honey bee *Apis mellifera* after a sublethal dose of neonicotinoid insecticides.	Bulletin of Insectology (2013), Volume 66, Number 1, pp. 1-9 (c)	321 (Clothianidin search)
CA 8.3.1.4/19	Laycock, I.; Cotterell, K.C.; O'Shea-Wheller, T.A.; Cresswell, J.E.	2014	Effects of the neonicotinoid pesticide thiamethoxam at field-realistic levels on microcolonies of *Bombus terrestris* worker bumble bees.	Ecotoxicology and environmental safety, (2014 Feb) Vol. 100, pp. 153-8. (c)	21 (Initial search)
CA 8.3.1.4/20	Mommaerts, V.; Reynders, S.; Boulet, J.; Besard, L.; Sterk, G.; Smagghe, G.	2010	Risk assessment for side-effects of neonicotinoids against bumblebees with and without impairing foraging behavior.	Ecotoxicology (London, England), (2010 Jan) Vol. 19, No. 1, pp. 207-15.	154 (Initial search)
CA 8.3.1.4/21	Fauser-Misslin, A.; Sadd, B.M.; Neumann, P.; Sandrock, C.	2014	Influence of combined pesticide and parasite exposure on bumblebee colony traits in the laboratory.	Journal of Applied Ecology (2014), Volume 51, Number 2, pp. 450-459. (c)	455 (Initial search)
CA 8.3.1.4/22	Elston, C.; Thompson, H. M.; Walters, K.F.A.	2013	Sub-lethal effects of thiamethoxam, a neonicotinoid pesticide, and propiconazole, a DMI fungicide, on colony initiation in bumblebee (*Bombus terrestris*) micro-colonies.	Apidologie (2013), Volume 44, Number 5, pp. 563-574, 63 refs. (c, d)	551 (Initial search)
CA 8.3.1.4/23	Sandrock, C.; Tanadini, L.G.; Pettis, J.S.; Biesmeijer, J.C.; Potts, S.G.; Neumann, P.	2014	Sublethal neonicotinoid insecticide exposure reduces solitary bee reproductive success.	Agricultural and Forest Entomology (2014), Volume 16, Number 2, pp. 119-128. (c, d)	489 (Initial search)
CA 8.3.1.4/24	Sandrock, C.; Tanadini, M.; Tanadini, L.G.; Fauser-Misslin, A.; Neumann, P. ; Potts, S. G.	2014	Impact of chronic neonicotinoid exposure on honeybee colony performance and queen supersedure.	PloS one, (2014) Vol. 9, No. 8	31 (Initial search)
CA 8.3.1.4/25	Lu, C.S.; Warchol, K.M.; Callahan, R.A.	2014	Sub-lethal exposure to neonicotinoids impaired honey bees winterization before proceeding to colony collapse disorder.	Bulletin of Insectology (2014), Volume 67, Number 1, pp. 125-130	276 (Clothianidin search)
CA 8.3.1.4/26	Scholer, J.; Krischik, V.	2014	Chronic exposure of imidacloprid and clothianidin reduce queen survival,foraging, and nectar storing in colonies of *Bombus impatiens*.	PloS one, (2014) Vol. 9, No. 3, pp. e91573 (d)	33 (Clothianidin search)

CA Data Point/ Ref Number	Authors	Year	Title	Source (Overseas evaluation report code) *	Ref ID
CA 8.3.1.4/27	Abbott, V.A.; Nadeau, J.L.; Higo, H.A.; Winston, M.L.	2008	Lethal and sublethal effects of imidacloprid on *Osmia lignaria* and clothianidin on *Megachile rotundat*a (Hymenoptera: Megachilidae).	Journal of economic entomology, (2008 Jun) Vol. 101, No. 3, pp. 784-96	111 (Clothianidin search)
CA 8.3.2/01	Duso, C.; Ahmad, S.; Tirello, P.; Pozzebon, A.; Klaric, V.; Baldessari, M.; Malagnini, V.; Angeli, G.	2014	The impact of insecticides applied in apple orchards on the predatory mite *Kampimodromus aberrans* (Acari: Phytoseiidae).	Experimental & applied acarology, (2014 Mar) Vol. 62, No. 3, pp. 391-414.	14 (Initial search)
CA 8.3.2/02	Tosi, L.; Farinazzo, E.; Posenato, G.; Girolami, V.	2006	Side effects of insecticides on *Kampimodromus aberrans*. Due anni di prove in veneto sul fitoseide Effetti collaterali di insetticidi su *Kampimodromus aberrans.*	Informatore Agrario (2006), Volume 62, Number 26, pp. 54-56	1407 (Initial search)
CA 8.3.2/03	Beers, E. H.; Brunner, J. F.; Dunley, J. E.; Doerr, M.; Granger, K.	2005	Role of neonicotinyl insecticides in Washington apple integrated pest management. Part II. Nontarget effects on integrated mite control	Journal of Insect Science (Tucson, AZ, United States) (2005), 5	1769 (Initial search)
CA 8.4.1/01	Wang K.; Pang S.; Mu X.; Qi S.; Li D.; Cui F.; Wang C.	2015	Biological response of earthworm, *Eisenia fetida*, to five neonicotinoid insecticides.	Chemosphere, (2015 Aug) Vol. 132, pp. 120-6 (f)	6 (Clothianidin search)

*This code was placed if the literature was referred in the overseas evaluation report by applicant (Syngenta Japan) in 2022

(a): Thiamethoxam – Transmittal of the preliminary aquatic and non-pollinator terrestrial risk assessment to support registration review. Docket number EPA-HQ-OPP-2011-0581, dated November 2017
(b): Thiamethoxam: Tier I update review of human incidents and epidemiology for draft risk assessment. Docket number EPA-HQ-OPP-2011-0581, dated August 2017
(c): Preliminary bee risk assessment to support the registration review of clothianidin and thiamethoxam. Docket number EPA-HQ-OPP-2011-0581, dated January 2017
(d): Appendices to the Final Bee Risk Assessment for Clothianidin (PC code 044309) and Thiamethoxam (PC code 060109). Docket number EPA-HQ-OPP-2011-0581, dated January 2020
(e): Conclusion on the peer review of the pesticide risk assessment for bees for the active substance thiamethoxam. EFSA Journal 2013; 11 (1): 3067, 68 pp.
(f): Annex 1 Background document to the Opinion proposing harmonised classification and labelling at EU level of thiamethoxam (ISO); 3-(2-chloro-thiazol-5-ylmethyl)-5-methyl[1,3,5]oxadiazinan-4-ylidene-N-nitroamine, report no. CLH-O-0000006724-70-01/F, dated 5 December 2019

DA assessment part from MCA Section 8 (listed by data point number)*
*This section was prepared from MCA section 8 dossier by applicant (Syngenta Japan) in 2022

Ref ID	Reference	Summary	Categorisation & comments
587 (Initial search)	Ivanova et al. (2013) Determining LD_{50} of the neonicotinoid insecticides Actara and Confidor in birds. *Agrarni Nauki*, Volume 5, Number 14, pp. 237-241.	The acute oral toxicity of a thiamethoxam formulation (Actara 25WG) to quail survival was tested. A single oral gavage dose was administered and mortality was recorded. Actara 25WG: LD_{50} = 4 100 mg formulation/kg bw (equivalent to LD_{50} = 1 025 mg a.s./kg bw) The study did not specify if a standard method was followed. Analytical verification was not carried out. It was not clear if the study was conducted according to GLP guidelines.	(b) Study that is relevant to the data requirement, but in the opinion of the applicant provide only supplementary information that does not alter existing risk assessment parameters. The results for Actara 25WG indicate that the formulation is not more toxic than the active substance. Additionally, the endpoint is similar to a study conducted by the Notifier (Japanese quail, LD_{50} > 2 000 mg A9584C/kg bw). The endpoint should be considered with caution due to the limitations of the study. A control treatment was not included and other information is lacking for scientific reliability of results. Acute oral toxicity data with the formulation is not a data requirement and there is no evidence that the formulation is more toxic than the active substance. Therefore, this study provides supplemental information only on the acute oral toxicity of the formulation to birds.

Ref ID	Reference	Summary	Categorisation & comments
170 (Initial search)	Barbee GC and Stout MJ (2009). Comparative acute toxicity of neonicotinoid and pyrethroid insecticides to non-target crayfish (*Procambarus clarkii*) associated with rice-crayfish crop rotations. *Pest management science* Vol. 65, No. 11, pp. 1250-6.	Juvenile *Procambarus clarkia* crayfish were exposed to thiamethoxam and clothianidin. Crayfish were examined 1, 12, 24, 48, 72 and 96 h after introduction into the exposure chambers. At each time, mortalities were recorded; sublethal effects (e.g. erraticbehavior, spasticmovement, lethargy) were noted. Data derived from each test were used to estimate the 96h LC_{50} and 95% confidence limits. The test material used was technical thiamethoxam. *Procambarus clarkia* Thiamethoxam: 96h LC_{50} = 967 µg a.s./L Clothianidin: 96h LC_{50} = 59 µg a.s./L This study was conducted according to ASTM E729-96 (static-renewal method). Analytical verification was not carried out. It was not clear if the study was conducted according to GLP guidelines.	(a) Study that provides data for refining risk assessment parameters Toxicity to crayfish which could be used if required in risk assessment. The results for thiamethoxam are similar to a study conducted by the Notifier (*Procambarus clarkii* 96h LC_{50} = 2300 µg a.s./L). The results for clothianidin suggest that the sensitivity of *Procambarus clarkii* is similar to other aquatic invertebrates tested (48h EC_{50} = 7 to 67 µg/L). Suitable for establishing a species sensitivity distribution (SSD) or calculation of geometric mean at Tier 2.

Ref ID	Reference	Summary	Categorisation & comments
292 (Initial search)	Stevens MM, Helliwell S, and PA Hughes (2005). Toxicity of *Bacillus thuringiensis* var. israelensis formulations, spinosad, and selected synthetic insecticides to *Chironomus tepperi* larvae. *Journal of the American Mosquito Control Association*, Vol. 21, No. 4, pp. 446-450.	*Chironomus tepperi* larvae were exposed to thiamethoxam and clothianidin. Larvae were assessed for mortality after 24 hours (unable to make co-ordinated response when lightly touched). Details: Glass specimen tubes (100 mm height, 25 mm diameter), 4-6 paper tissue strips (3x15 mm), 15h light 9h dark lighting, 25°C. Five concentrations tested and control (3 replicates each). 20mL of test solution and 10 individuals per replicate. No food was provided. The test material used was Actara 25WDG. *Chironomus tepperi* Thiamethoxam: 24h LC_{50} = 121 µg a.s./L Clothianidin: 24h LC_{50} = 2.83 µg a.s./L The study did not specify if a standard method was followed. The methodology described differs from the OECD 235 method. Analytical verification was not carried out. It was not clear if the study was conducted according to GLP guidelines.	(a) Study that provides data for refining risk assessment parameters Toxicity data to *Chironomus tepperi* which could be used in risk assessment if required. The results for thiamethoxam are similar to studies conducted by the Notifier on another chironomid species (*Chironomus riparius* 48h EC_{50} = 35 to 71 µg a.s./L). However, the results for clothianidin indicate that this species may be more sensitive than the existing chironmid species tested (*Chironomus riparius* 48h EC_{50} = 14 µg a.s./L). Suitable for establishing a species sensitivity distribution (SSD), calculation of geometric mean at Tier 2 or at least as data for weight-of-evidence approach.

Ref ID	Reference	Summary	Categorisation & comments
1576 (Initial search)	Deng L et al (2013). Acute immobilization of four neonicotinoid insecticides to *Daphnia magn*a Straus. *Nongyao Kexue Yu Guanli* (2013), 34 (6), 23-25.	The acute toxicity of thiamethoxam and clothianidin to *Daphnia magna* survival was tested according to OECD no. 202. The test materials used were Thiamethoxam 25% WG and Clothianidin 600 g/L SC. *Daphnia magna* Thiamethoxam: 48h EC_{50} = 0.0273 mg a.s./L Clothianidin: 48h EC_{50} = 2.1427 mg a.s./L Analytical verification was not carried out. It was not clear if the study was conducted according to GLP guidelines.	(b) Study that is relevant to the data requirement, but in the opinion of the applicant provides only supplementary information that does not alter existing risk assessment parameters. The result for Thiamethoxam 25% WG was reported in units of mg a.s./L. The current regulatory EU *Daphnia magna* acute endpoint for the 25WG formulation is >100 mg A9584C/L. The result for Clothianidin 600 g/L SC was reported in units of mg a.s./L. The current regulatory EU *Daphnia magna* acute endpoint for the metabolite CGA322704 >100 mg/L. These endpoint should be considered with caution due to the limitations of the study. In addition to not being conducted under GLP conditions and no analytical verification carried out, little information is given about the applied statistical method. Regulatory studies and other open literature indicate that *Daphnia magna* are not sensitive to thiamethoxam or its metabolite CGA322704 (Clothianidin). Therefore this report supplies supplemental information only.

Ref ID	Reference	Summary	Categorisation & comments
1 (Top-up search)	Ugurlu P, Unlu E and EI Satar (2015). The toxicological effects of thiamethoxam on *Gammarus kischineffensis* (Schellenberg 1937) (Crustacea: Amphipoda). *Environmental toxicology and pharmacology*, Vol. 39, No. 2, pp. 720-6	*Gammarus kischineffensis* were exposed to thiamethoxam for 96 h. Behavioral changes, immobility and mortality were recorded after 24, 48, 72 and 96 h of exposure. Data were used to estimate the LC_{50} and 95% confidence limits. The test material used was Actara 240SC. *Gammarus kischineffensis* Thiamethoxam: 48h LC_{50} = 23.505 mg a.s./L 96h LC_{50} = 3.751 mg a.s./L This study was conducted according to APHA (1998; static renewal method). Analytical verification was not carried out. It was not clear if the study was conducted according to GLP guidelines.	(a) Study that provides data for refining risk assessment parameters Toxicity to a freshwater amphipod which could be used if required in risk assessment. The results for thiamethoxam are similar to a study conducted by the Notifier (*Gammarus* sp. 48h EC_{50} > 2.8 mg a.s./L). The acute LD_{50} is suitable for establishing a species sensitivity distribution (SSD) or calculation of geometric mean at Tier 2. Sublethal results are suitable for weight-of-evidence approach.

Ref ID	Reference	Summary	Categorisation & comments
3 (Clothianidin search)	de Perre C, Murphy TM and MJ Lydy (2015). Fate and effects of clothianidin in fields using conservation practices. *Environmental toxicology and chemistry / SETAC*, Vol. 34, No. 2, pp. 258-65	Nontarget organisms were exposed to clothianidin to determine the acute toxicity (EC_{50}/LC_{50} values). Endpoints included lethality for all species, difficulty to swim, lack of or erratic movements for the aquatic invertebrates and fish, and growth for the earthworms. The specific test material used was not stated. Clothianidin: *Daphnia magna* 48h EC_{50} > 500 µg a.s./L *Hyalella azteca* 96h EC_{50} = 6.67 µg a.s./L *Chironomus dilutes* 96h EC_{50} = 1.85 µg a.s./L *Pimephales promelas* 96h LC_{50} > 500 µg a.s./L Based on USEPA and OECD test guidelines. It was not clear if the study was conducted according to GLP guidelines. Analytical verification was carried out. Acute toxicity of earthworms (*Eisenia fetida*), was also tested. As an acute earthworm endpoint is no longer a data requirement, this part of the study is not considered relevant. Additionally, this study exmined the environmental fate of clothianidin applied as a corn seed-coating under field conditions in the USA. Risk quotients were calculated from clothianidin concentrations measured in the field and compared with the laboratory toxicity bioassay results to assess the environmental risk of the insecticide. This is not considered relevant to the ecotoxicity of the molecule and is thus not considered relevant.	(a) Study that provides data for refining risk assessment parameters Toxicity to aquatic organisms for clothianidin; gives EC_{50}/LC_{50} for 4 species which could be used in risk assessment if required. The results for *Daphnia magna* are similar to a study conducted by the Notifier (*D. magna* 48h EC_{50} > 100 mg/L). The results for *Hyalella azteca* suggest that the sensitivity is similar to other aquatic invertebrates tested (48h EC_{50} = 7 to 67 µg/L). The results for *Chironomus dilutes* suggest that this species may be more sensitive than the previously tested species *Chironomus riparius* (*C. riparius* 48h EC_{50} = 14 µg a.s./L). The results for *Pimephales promelas* suggest that the sensitivity is similar to another fish species tested (*Oncorhynchus mykiss* 96h LC_{50} > 100 mg/L). Not suitable for use in risk assessment due to the limitations identified in the study.

Ref ID	Reference	Summary	Categorisation & comments
35 (Clothianidin search)	Whiting SA and MJ Lydy (2014). A site-specific ecological risk assessment for corn-associated insecticides. *Integrated Environmental Assessment and Management*, Published online 30 December 2014.	The amphipod *Hyalella azteca* and the Riffle beetle larvae, *Ancyronyx* spp., were exposed to clothianidin to determine the acute toxicity (EC_{50}/LC_{50} values). The specific test material used was not stated. Clothianidin: *Hyalella Azteca* 96h LC_{50} = 9.68 µg a.s./L *Ancyronyx* spp. 7d LC_{50} = 50.9 µg a.s./L Based on USEPA test guidelines and adapted literature protocols. It was not clear if the study was conducted according to GLP guidelines. Analytical verification was not carried out. Acute toxicity of earthworms (*Eisenia fetida*), was also tested. As an acute endpoint is no longer a data requirement, this part of the study is not considered relevant.	(a) Study that provides data for refining risk assessment parameters Toxicity to aquatic organisms for clothianidin; gives LC_{50} for 2 species which could be used in risk assessment if required. The results for *Hyalella azteca* and *Ancyronyx* spp. suggest that the sensitivity is similar to other aquatic invertebrates tested (48h EC_{50} = 7 to 67 µg/L). Suitable for establishing a species sensitivity distribution (SSD) or calculation of geometric mean at Tier 2.

Ref ID	Reference	Summary	Categorisation & comments
35 (Clothianidin search)	Whiting SA and MJ Lydy (2014). A site-specific ecological risk assessment for corn-associated insecticides. *Integrated Environmental Assessment and Management*, Published online 30 December 2014.	The amphipod *Hyalella azteca* and the Riffle beetle larvae, *Ancyronyx* spp., were exposed to clothianidin to determine the acute toxicity (EC_{50}/LC_{50} values). The specific test material used was not stated. Clothianidin: *Hyalella Azteca* 96h LC_{50} = 9.68 µg a.s./L *Ancyronyx* spp. 7d LC_{50} = 50.9 µg a.s./L Based on USEPA test guidelines and adapted literature protocols. It was not clear if the study was conducted according to GLP guidelines. Analytical verification was not carried out. Acute toxicity of earthworms (*Eisenia fetida*), was also tested. As an acute endpoint is no longer a data requirement, this part of the study is not considered relevant.	(a) Study that provides data for refining risk assessment parameters Toxicity to aquatic organisms for clothianidin; gives LC_{50} for 2 species which could be used in risk assessment if required. The results for *Hyalella azteca* and *Ancyronyx* spp. suggest that the sensitivity is similar to other aquatic invertebrates tested (48h EC_{50} = 7 to 67 µg/L). Suitable for establishing a species sensitivity distribution (SSD) or calculation of geometric mean at Tier 2.

Ref ID	Reference	Summary	Categorisation & comments
317 (Clothianidin search)	Hayasaka D, Suzuki K, Nomura T, Nishiyama M, Nagai T, Sanchez-Bayo F, and K Goka (2013). Comparison of acute toxicity of two neonicotinoid insecticides, imidacloprid and clothianidin, to five cladoceran species. *Journal of Pesticide Science*, Volume 38, Number 1/2, pp. 44-47	Five cladoceran species were exposed to clothianidin to determine the acute toxicity (EC_{50} values). The test material used was the commercial formulation Dantotsu®. Clothianidin: *Ceriodaphnia dubia* 48h EC_{50} = 1 691.3 µg a.s./L *Ceriodaphnia reticulate* 48h EC_{50} = 29 474 µg a.s./L *Daphnia magna* 48h EC_{50} = 67 564 µg a.s./L *Daphnia pulex* 48h EC_{50} = 31 448 µg a.s./L *Moina macrocopa* 48h EC_{50} = 61 106 µg a.s./L Based on OECD guideline no. 202. It was not clear if the study was conducted according to GLP guidelines. Analytical verification was not carried out.	(a) Study that provides data for refining risk assessment parameters Toxicity to 5 cladoceran species which could be used in risk assessment if required. The results suggest that the sensitivity of the five cladoceran species tested are similar to the cladoceran species tested by the Notifier (*D. magna* 48h EC_{50} > 100 000 µg a.s./L) and less sensitive than other aquatic invertebrates tested (48h EC_{50} = 7 to 67 µg/L). Suitable for establishing a species sensitivity distribution (SSD) or calculation of geometric mean at Tier 2.

Ref ID	Reference	Summary	Categorisation & comments
n/a	Riaz et al. (2013) Molecular mechanisms associated with increased tolerance to the neonicotinoid insecticide imidacloprid in the dengue vector *Aedes aegypti*. *Aquatic Toxicology* 126 (2013) 326– 337.	The acute toxicity of different insecticides (including thiamethoxam) to the mosquito *Aedes aegypti* was tested. Fourth stage larvae of an imidacloprid resistant and a susceptible strain were used. Mortality was recorded after 24 hours. The specific test material used was not stated. Thiamethoxam: *Aedes aegypti* Imidacloprid resistant strain: 24h LC_{50} = 806 µg a.s./L Susceptible strain: 24h LC_{50} = 183 µg a.s./L The study did not specify if a standard method was followed. Analytical verification was not carried out. It was not clear if the study was conducted according to GLP guidelines.	(a) Study that provides data for refining risk assessment parameters Toxicity to mosquito larvae which could be used if required in risk assessment. The results suggest that the sensitivity of *Aedes aegypti* is similar to other aquatic insects tested (48h EC_{50} = 14 to 7300 µg/L). The acute toxicity of "susceptible" (i.e. not conditioned to imidacloprid) aquatic larval stages of *Aedes aegypti* is suitable for establishing species-sensitivity distributions (SSD) or calculating a geometric mean at tier 2 of the risk assessment on aquatic invertebrates.

Ref ID	Reference	Summary	Categorisation & comments
390 (Initial search)	Illarionov and Derkach (2008) Toxicity and Hazard of Neonicotinoids for Honeybees. *Agrokhimiya*, No. 10, pp. 74-81.	The acute oral and contact toxicity of thiamethoxam as Actara 25WG was determined for honeybees. Thiamethoxam *Apis mellifera* L. Oral LD_{50} = 0.034 µg a.s./bee Direct contact LD_{50} = 0.0025 µg a.s./bee Residual contact LD_{50} = 0.038 µg a.s./cm^2 A field study was carried out in fields of flowering *Phacelia*. The plants were treated with thiamethoxam at the recommended field rates (0.06, 0.4 and 0.8 kg/ha). Existing label mitigation for thiamethoxam includes 'No application during flowering'. Additionally, the application rate used here exceeds the foliar application rate supported in this assessment (20-50 g a.s./ha). Therefore, this aspect of the study provides supplemental information only and is not considered further.	(b) Study that is relevant to the data requirement, but in the opinion of the applicant provide only supplementary information that does not alter existing risk assessment parameters. The oral and direct contact results derived from this study are similar to the existing EU endpoints: Thiamethoxam Oral 48h LD_{50} = 0.005 µg a.s./bee Direct contact 48h LD_{50} = 0.024 µg a.s./bee The residual contact method is not an exposure route addressed in the risk assessment and does not produce an endpoint useful for risk assessment. The toxicity test methods do not follow the standard OECD guideline and are not conducted to GLP. This study provides supplemental information only on the acute toxicity to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
449 (Initial search)	Thompson et al. (2014). Potential impacts of synergism in honeybees (*Apis mellifera*) of exposure to neonicotinoids and sprayed fungicides in crops. *Apidologie*, Volume 45, Number 5, pp. 545-553.	The acute oral and contact toxicity of thiamethoxam and clothianidin was determined for honeybees following standard OECD guidelines. The test material used in the study was technical active substance. *Apis mellifera* Thiamethoxam Oral: 48h LD_{50} = 0.0112 µg a.s./bee (95% CI: 0.00915–0.0135) Contact: 48h LD_{50} = 0.124 µg a.s./bee (95% CI: 0.0768–0.3280) Clothianidin Oral: 48h LD_{50} = 0.00739 µg a.s./bee (95% CI: 0.00607–0.00903) Contact: 48h LD_{50} = 0.0350 µg a.s./bee (95% CI: 0.0155–0.0607) Acute oral and contact studies were also carried out in combination with fungicides to examine the potential for synergism. As mixture toxicity studies are not relevant for this review, this aspect of the study is not considered further.	(b) Study that is relevant to the data requirement, but in the opinion of the applicant provide only supplementary information that does not alter existing risk assessment parameters. The experimental design complies with OECD standard guidelines. However, the study was not performed under GLP conditions. There are no limitations in the technical quality of the experiments. Additionally, the results derived from this study are similar to the existing EU endpoints: Thiamethoxam Oral 48h LD_{50} = 0.005 µg a.s./bee Direct contact 48h LD_{50} = 0.024 µg a.s./bee Clothianidin Oral 48h LD_{50} = 0.0168 µg a.s./bee Direct contact 48h LD_{50} = 0.0275 µg a.s./bee This study provides supplemental information only on the acute toxicity to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
611 (Initial search)	Laurino et al. (2013). Toxicity of neonicotinoid insecticides on different honey bee genotypes. *Bulletin of Insectology*, Vol. 66, Number 1, pp. 119-126.	The acute oral and contact toxicity of thiamethoxam and clothinidin was determined for different honey bee genotypes belonging to the following subspecies: *Apis mellifera mellifera* L., *Apis mellifera ligustica* Spinola, and *Apis mellifera carnica* Pollmann. The test materials used in the study were commercial products Actara 25WG (thiamethoxam) and Dantop 50WG (clothianidin). The oral toxicity test methods are similar to the standard test guideline; however the contact toxicity is based on a residual contact method. Therefore only the acute oral toxicity is considered relevant. <u>Thiamethoxam</u> *A. mellifera mellifera* L. Oral: 48h LD_{50} = 3.4 ng a.s./bee *A. mellifera ligustica* Spinola Oral: 48h LD_{50} = 1.65 – 5.08 ng a.s./bee *A. mellifera carnica* Pollmann Oral: 48h LD_{50} = 5.56 – 9.07 ng a.s./bee <u>Clothianidin</u> *A. mellifera mellifera* L. Oral: 48h LD_{50} = 6.72 ng a.s./bee *A. mellifera ligustica* Spinola Oral: 48h LD_{50} = 1.11 – 5.07 ng a.s./bee	(a) Study provides data for refining a risk assessment parameter. The oral and contact toxicity test methods do not follow the standard OECD guideline and were not conducted to GLP. The oral toxicity results for the 3 sub species are similar to the results for the standard test species used to derive the existing EU endpoint: <u>Thiamethoxam</u> Oral 48h LD_{50} = 0.005 µg a.s./bee <u>Clothianidin</u> Oral 48h LD_{50} = 0.0168 µg a.s./bee The endpoints are suitable to be used in the risk assessment as part of a weight-of-evidence approach only due to the exposure regime used in the toxicity tests.

Ref ID	Reference	Summary	Categorisation & comments
858 (Initial search)	Laurino et al. (2011). Toxicity of neonicotinoid insecticides to honey bees: laboratory tests. *Bulletin of Insectology*, Volume 64, Number 1, pp. 107-113.	The acute oral and contact toxicity of thiamethoxam and clothinidin was determined for honeybees (*A. mellifera)*. The test materials used in the study were commercial products Actara 25WG (thiamethoxam) and Dantop 50WG (clothianidin). *Apis mellifera* <u>Thiamethoxam</u> Oral: 48h LD_{50} = 4.411 ng a.s./bee (95% CI: 3.612-5.252) Residual contact LC_{50} = 3.313 ng a.s./µL (95% CI: 2.786-3.806) <u>Clothianidin</u> Oral: 48h LD_{50} = 2.689 ng a.s./bee (95% CI: 1.749-3.679) Residual contact LC_{50} = 2.967 ng a.s./µL (95% CI: 2.398-3.467)	(b) Study that is relevant to the data requirement, but in the opinion of the applicant provide only supplementary information that does not alter existing risk assessment parameters. The oral and contact toxicity test methods do not follow the standard OECD guideline. The contact toxicity test was performed in a fashion of test procedures for other non-target arthropods but not as for bees. Additonally they were not conducted to GLP. The oral toxicity results are similar to the existing EU endpoints: <u>Thiamethoxam</u> Oral 48h LD_{50} = 0.005 µg a.s./bee <u>Clothianidin</u> Oral 48h LD_{50} = 0.0168 µg a.s./bee This study provides supplemental information only on the acute toxicity to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
909 (Initial search)	Falco et al. (2010). Toxicity of thiamethoxam, behavioral effects and alterations in chromatin of *Apis mellifera* L., 1758 (Hymenoptera; Apidae). *Research Journal of Agriculture and Biological Sciences*, Volume 6, Number 6, pp. 823-828.	The acute oral toxicity of thiamethoxam was determined for honeybees (*Apis mellifera* L.) at different ages. The specific test material used was not identified. Four ages of bees were included in the study (0, 7, 14 and 21 day old). The percent mortality of bees following acute exposure to six test concentrations was presented. However, 24h LD_{50} values were not calculated. Effects on feeding behaviour are addressed below in a separate evaluation. The effect of acute oral exposure on critical electrolyte concentration (CEC) in the chromatin of cells from Malpighian tubules was also determined. This is not relevant to the risk assessment and therefore not considered further.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. The oral toxicity test methods do not follow the standard OECD guideline and not conducted to GLP. An endpoint relevant to the risk assessment is not included.

Ref ID	Reference	Summary	Categorisation & comments
967 (Initial search)	Laurino et al. (2010) Acute oral toxicity of neonicotinoids on different bee strains. *Redia*, Volume 93, pp. 99-102.	The acute oral toxicity of thiamethoxam and clothinidin was determined for different honey bee strains of *Apis mellifera ligustica* Spinola. The test materials used in the study were commercial products Actara 25WG (thiamethoxam) and Dantop 50WG (clothianidin). <u>Thiamethoxam</u> 1: 48h oral LD_{50} = 2.644 ng a.s./bee 2: 48h oral LD_{50} = 3.018 ng a.s./bee 3: 48h oral LD_{50} = 4.383 ng a.s./bee <u>Clothianidin</u> 1: 48h oral LD_{50} = 4.671 ng a.s./bee 2: 48h oral LD_{50} = 3.789 ng a.s./bee 3: 48h oral LD_{50} = 4.507 ng a.s./bee	(a) Study provides data for refining a risk assessment parameter. The oral toxicity test methods do not follow the standard OECD guideline and not conducted to GLP. The oral toxicity results are similar to the results for the standard test species used to derive the existing EU endpoints. <u>Thiamethoxam</u> Oral 48h LD_{50} = 0.005 µg a.s./bee <u>Clothianidin</u> Oral 48h LD_{50} = 0.0168 µg a.s./bee The endpoints are suitable to be used in the risk assessment as part of a weight-of-evidence approach only.

Ref ID	Reference	Summary	Categorisation & comments
1570 (Initial search)	Pei et al (2013) Acute toxicity of four insecticides to honeybee *Apis mellifera*. *Shijie Nongyao*, 35 (4), 50-51, 54.	The acute oral and contact toxicity of thiamethoxam was determined for honeybees (*A. mellifera)*. The test material used in the study was technical active substance. Thiamethoxam *Apis mellifera* Oral: 48h LD_{50} = 0.262 mg a.s./L (95% CI: 0.240 - 0.289) Contact: 48h LD_{50} = 0.016 µg a.s./bee (95% CI: 0.014 - 0.020)	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. The oral and contact toxicity test methods do not follow the standard OECD guideline and not conducted to GLP. The oral toxicity results are not given in units useful for risk assessment. The contact toxicity results are similar to the existing EU endpoints. Thiamethoxam Oral 48h LD_{50} = 0.005 µg a.s./bee Contact 48h LD_{50} = 0.024 µg a.s./bee This study provides supplemental information only on the acute toxicity to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
1672 (Initial search)	Valdovinos-Nunez et al. (2009). Comparative toxicity of pesticides to stingless bees (Hymenoptera: Apidae: Meliponini). *Journal of Economic Entomology*, Volume 102 (5), 1737-1742.	The acute contact toxicity of thiamethoxam was determined for the stingless bee species *Nannotrigona perilampoides*. The test material used in the study was technical active substance. Thiamethoxam *Nannotrigona perilampoides* Contact: 24h LD_{50} = 0.004 µg a.s./bee	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. This species of bee is localized to Central America and not relevant to a European risk assessment. The contact toxicity test methods do not follow the standard OECD guideline and are not conducted to GLP. Additionally, only 2 x 10 individuals per dose were tested with implications on robustness of the results. This study provides supplemental information only on the acute toxicity to the stingless bee species *Nannotrigona perilampoides*.

Ref ID	Reference	Summary	Categorisation & comments
36 (Initial search)	Thompson et al. (2014). Neonicotinoids and bumblebees (*Bombus terrestris*): effects on nectar consumption in individual workers. *Pest management science*, (2014 Jul 30)	The effect of a 4d oral exposure to thiamethoxam and clothianidin on bumble bee (*Bombus terrestris*) feeding behaviour was examined. The amount of sucrose consumed per day was recorded over 4 days. The test material used in the study was technical active substance. Due to 100% mortality at 100 µg/L, analysis was only conducted when bees were exposed to 1 and 10 µg/L. <u>Thiamethoxam</u> No consistent avoidance/antifeedant response was detected at the 1 and 10 µg/L dose rates. <u>Clothianidin</u> No reduction in food intake was recorded at 1 µg/L, but reduced consumption was noted at 10 µg/L.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. There are no comparable regulatory studies that address anti-feedant effects or food choice. Results indicate that thiamethoxam does not have an anti-feedant effect on bumblebees at concentrations up to and including 10 µg/L, while clothianidin does not have an anti-feedant effect on bumblebees at concentrations up to and including 1 µg/L. However, the endpoints measured in this study do not allow any conclusion regarding exposure to bees and has a negligible impact on the risk assessment.

Ref ID	Reference	Summary	Categorisation & comments
7 (Top-up search)	Kessler et al. (2015). Bees prefer foods containing neonicotinoid pesticides. *Nature*, Vol. 521, No. 7550, pp. 74-6.	The effect of a sublethal acute oral exposure to thiamethoxam and clothianidin on honeybee (*Apis mellifera*) and bumble bee (*Bombus terrestris*) feeding behaviour was examined. The test material used in the study was technical active substance. Observations on whether honeybees or bumble bees avoid sucrose solutions containing neonicotinoids were recorded using a two-choice test. Four concentrations were tested (1 nM, 10nM, 100nM and 1 µM). <u>Thiamethoxam</u> Results indicate that honeybees and bumble bees do not avoid nectar-relevant concentrations in food. And both bumblebees and honeybees showed a preference for thiamethoxam solutions over sucrose alone. <u>Clothianidin</u> Results indicate that honeybees and bumble bees do not avoid nectar-relevant concentrations in food.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. There are no comparable regulatory studies that address anti-feedant effects or food choice. Results indicate that thiamethoxam and clothianidin do not have an anti-feedant effect on honeybees and bumblebees at concentrations up to and including 1 µM (highest tested). However, the endpoints measured in this study do not allow any conclusion regarding exposure to bees and has a negligible impact on the risk assessment. The study provides supplemental information only on anti-feedant effects of thiamethoxam and clothianidin on *Apis mellifera* and *Bombus terrestris*.

Ref ID	Reference	Summary	Categorisation & comments
197 (Initial search)	El Hassani et al. (2008). Effects of sublethal doses of acetamiprid and thiamethoxam on the behavior of the honeybee (*Apis mellifera*). *Archives of environmental contamination and toxicology*, Vol. 54, No. 4, pp. 653-61.	The effect of sublethal acute exposure (oral and contact) to thiamethoxam on honeybee (*Apis mellifera*) behaviour was examined. Bees were exposed to three concentrations (0.1, 0.5, or 1 ng a.s./bee). Observations on locomotor activity were recorded 60 min after after a single topical application or oral dose. The test material used in the study was technical active substance. Thiamethoxam No effect on bee behavior under the conditions used. Observations on feeding behaviour (sucrose and water responsiveness (proboscis extension) and olfactory learning) were also recorded and are considered in a separate evaluation.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam does not effect honeybee motor behaviour when exposed (orally and topically) to 0.1-1 ng a.s./bee for 60 min. Behavioral effects of acute exposure are addressed in regulatory acute toxicity studies. The results from these studies indicate that 24 and 48 hours after exposure, surviving bees exhibited no behavioural abnormalities. Sublethal effects are more properly considered in chronic risk assessments than for acute toxicity. The study provides supplemental information only on behavioural effects of acute exposure to thiamethoxam.

Ref ID	Reference	Summary	Categorisation & comments
183 (Initial search)	Aliouane et al. (2009). Subchronic exposure of honeybees to sublethal doses of pesticides: effects on behavior. Environmental toxicology and chemistry / SETAC, Vol. 28, No. 1, pp. 113-122	The effect of sublethal chronic exposure (oral and contact) to thiamethoxam on honeybee (*Apis mellifera*) behavior was examined. The test material used in the study was technical active substance. Observations on water and sucrose responsiveness (proboscis extension), and olfactory learning abilities (olfactory conditioning of the proboscis extension reflex) were recorded. The exposure period was 11 days and two concentrations were tested (0.1 and 1.0 ng a.s./bee) tested. <u>Thiamethoxam</u> Thiamethoxam by contact induced either a significant decrease of olfactory memory 24 h after learning at 0.1 ng/bee or a significant impairment of learning performance with no effect on memory at 1 ng/bee. Responsiveness to antennal sucrose stimulation was significantly decreased for high sucrose concentrations in honeybees treated orally with thiamethoxam (1 ng/bee). Observations on locomotor activity were also recorded and are considered in a separate evaluation.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. There are no comparable regulatory studies that address effects on feeding behavior. Results indicate that sub-chronic exposure (oral and contact) to thiamethoxam does have an effect on honeybee feeding behaviour at concentrations of 0.1 ng a.s./bee. There is no link between proboscis extension responsiveness and bee behaviour which can be used in risk assessment. Therefore, this study provides supplemental information only on sub-lethal behavioral effects of thiamethoxam on *Apis mellifera*.
183 (Initial search)	Aliouane et al. (2009). Subchronic exposure of honeybees to sublethal doses of pesticides: effects on behavior. Environmental toxicology and chemistry / SETAC, Vol. 28, No. 1, pp. 113-122	The effect of sublethal chronic exposure (oral and contact) to thiamethoxam on honeybee (*Apis mellifera*) behavior was examined. The exposure period was 11 days and two concentrations were tested (0.1 and 1.0 ng/bee) tested. Observations on locomotor activity were recorded at the end of the exposure period. The test material used in the study was technical active substance. <u>Thiamethoxam</u> No significant effect on the three parameters of locomotor activity compared to controls, regardless of dose or exposure route. Observations on feeding behaviour (water and sucrose responsiveness, olfactory learning abilities) were also recorded and are considered in a separate evaluation.	(a) Study provides data for establishing or refining risk assessment parameters. Results indicate that thiamethoxam does not effect honeybee motor behaviour when exposed (orally and topically) to 0.1 and 1 ng a.s./bee for 11 days. Behavioral effects of a 10 d chronic oral exposure are recorded in regulatory chronic toxicity studies. This study provides information on sub-lethal behavioral effects of thiamethoxam on *Apis mellifera*. The results could be used for deriving a NOEC value for effects on locomotor activity.

Ref ID	Reference	Summary	Categorisation & comments
7 (Initial search)	Williamson et al. (2014). Exposure to neonicotinoids influences the motor function of adult worker honeybees. *Ecotoxicology*, Vol. 23, No. 8, pp. 1409-18.	The effect of acute oral exposure to thiamethoxam and clothianidin on honeybee (*Apis mellifera*) motor function was examined. Observations on walking, flying, remaining still, falling upside down and grooming were recorded. Due to high mortality at 100 nM, behavioural analysis was only conducted when bees were exposed to 10 nM for 24 hours. The test material used in the study was technical active substance. <u>Thiamethoxam</u> 10 nM = 2.92 ppb or 0.481 ng a.s./bee/24h No significant effects on walking, flying, or standing still behaviour. However bees were more likely to lose postural control and fail to right themselves. Bees also spent more time grooming. <u>Clothianidin</u> 10 nM = 2.50 ppb or 0.344 ng a.s./bee/24h No significant effects on walking, flying, or standing still behaviour. However bees were more likely to lose postural control and fail to right themselves.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam and clothianidin effect honeybee motor behaviour when exposed orally to 10 nM for 24 hours (only concentration tested). Behavioral effects of acute oral exposure are addressed in regulatory acute toxicity studies. The results from these studies indicate that 24 and 48 hours after exposure, surviving bees exhibited no behavioural abnormalities. Sublethal effects are more properly considered in chronic risk assessments than for acute toxicity; full recovery is likely if effects are only acute. The study provides supplemental information only on behavioural effects of acute exposure to thiamethoxam and clothianidin.

Ref ID	Reference	Summary	Categorisation & comments
98 (Initial search)	Henry et al. (2012). A common pesticide decreases foraging success and survival in honey bees. *Science*, Vol. 336, No. 6079, pp. 348-50.	Effects from acute exposure to thiamethoxam on honeybee (*Apis mellifera*) homing behaviour was investigated under semi-field conditions. Homing behaviour was exmined by releasing tagged honeybees (up to 1 km away from their respective colony) and using RFID readers to detect tagged honey bees enetering the hive. Foragers received a dose of 1.34 ng a.s./bee. The specific test material used was not reported. The authors also assessed the effects of post-exposure homing failure in combination with natural forager mortality, on colony dynamics using a honey bee population model. <u>Thiamethoxam</u> The proportion of treated foragers returning to the colony was significantly lower than control. Additionally post-exposure homing failure (m_{hf}) was greater in treated foragers that were unfamiliar with the foraging site, as indicated by their significantly lower homing proportions as compared with familiar foragers. When implementing post-exposure homing failure into a honey bee population model, all tested scenarios predicted a major deviation from the expected dynamic (e.g. colony collapse).	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam has an effect on honeybee homing behaviour when orally exposed to 1.34 ng a.s./bee. In this study the bees consumed the total amount of active substance within a relatively short period and not administered over a longer period i.e. a day. EFSA concluded that this method of exposure could lead to more severe effects than what may occur when bees are foraging. These observations of acute sublethal effects are rather not suitable for risk assessment due to a negligible impact of such temporally and indidually limited effects on bee colonies. The study provides supplemental information only on behavioural effects of acute exposure to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
277 (Initial search)	Cresswell and Thompson (2012). Response to comment on "a common pesticide decreases foraging success and survival in honey bees". *Science*, Vol. 337, No. 6101, pp. 1453-c.	This article is a “Technical comment” critiquing the study by Henry et al 2012 (Ref. ID 98, Initial search). The authors recalculate the bee population model presented by Henry et al. 2012 with parameter values appropriate to the season when most pesticide treated flowering crops are in bloom (spring). The author’s data suggest that dietary thiamethoxam would not precipitate collapse in healthy colonies in spring.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results from re-parameterizing the bee population model used by Henry et al. 2012 indicated that exposure to thiamethoxam would not precipitate collapse in healthy colonies in spring. As the study that this article is critiquing is considered supplemental information, due to over dosing of the test organisms, this reference also only provides supplemental information.

Ref ID	Reference	Summary	Categorisation & comments
34 (Clothianidin search)	Fischer et al. (2014). Neonicotinoids interfere with specific components of navigation in honeybees. *PloS one*, Vol. 9, No. 3, pp. e91364	Effects from an acute exposure to clothianidin on honeybee (*Apis mellifera*) navigation was investigated under semi-field conditions. A catch and release experimental design was applied. The flight paths of individual bees were tracked with harmonic radar. Foragers received a dose of 0.2 mM (corresponding to 2.5 ng/bee) clothianidin over the course of 90 minutes prior to release. The test material used in this study was technical active substance. <u>Clothianidin</u> The proportion of bees returning to the hive differed between the control and treated groups (88% of control bees vs. 78% treated bees). There was a significant difference in the flight direction compared to control (i.e. flights were less straight). Additionally, the total flight path during the homing phase was significantly longer length and duration in treated bees.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that clothianidin has an effect on honeybee navigation when orally exposed to 2.5 ng clothianidin/bee. In this study the bees consumed the total amount of active substance within a relatively short period and not administered over a longer period i.e. a day. EFSA concluded that this method of exposure could lead to more severe effects than what may occur when bees are foraging. These observations of acute sublethal effects are rather not suitable for risk assessment due to a negligible impact of such temporally and indidually limited effects on bee colonies. The study provides supplemental information only on behavioural effects of acute exposure to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
61 (Clothianidin search)	Schneider et al. (2012). RFID tracking of sublethal effects of two neonicotinoid insecticides on the foraging behavior of *Apis mellifera*. *PloS one*, Vol. 7, No. 1, pp. e30023	Effects from an acute exposure to clothianidin on honeybee (*Apis mellifera*) foraging behavior was investigated under semi-field conditions. Foraging behaviour was exmined by releasing tagged honeybees and using RFID readers at both the hive and feeder. The number of foraging trips from the hive to feeder, duration of foraging trip, and time interval a bee spent inside the hive between foraging trips was recorded. Additionally, flight time to feeder, duration of stay at feeder, and flight time back to the hive were also recorded. Foragers received a one-time oral exposure (consumption of dose within 20 min) prior to analysis of foraging behaviour. Behavior was measured for a 3 hr period immediately after and between 24 h and 48 h after exposure. The doses tested were 0.05, 0.5, 1 and 2 ng clothianidin/bee. <u>Clothianidin</u> There was a significant reduction of foraging activity and increase in foraging flight time at doses of ≥0.5 ng/bee during the first 3 hrs after treatment. Bees disappeared after receiving the 1 and 2 ng/bee doses. There were no significant effects on the number of feeder visits or the total foraging time at 0.05 ng/bee.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that clothianidin has an effect on foraging behavior when orally exposed to ≥0.5 ng clothianidin/bee. In this study the bees consumed the total amount of active substance within a relatively short period and not administered over a longer period i.e. a day. EFSA concluded that this method of exposure could lead to more severe effects than what may occur when bees are foraging. These observations of acute sublethal effects are rather not suitable for risk assessment due to a negligible impact of such temporally and indidually limited effects on bee colonies. The study provides supplemental information only on behavioural effects of acute exposure to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
321 (Clothianidin search)	Matsumoto (2013). Reduction in homing flights in the honey bee *Apis mellifera* after a sublethal dose of neonicotinoid insecticides. *Bulletin of Insectology*, Volume 66, Number 1, pp. 1-9	Effects from acute exposure to clothianidin on honeybee (*Apis mellifera*) homing flight was investigated under semi-field conditions. A catch and release experimental design was applied (up to 500 m away from their hives). The number of number of returning bees within 30 min and time of arrival was recorded. Foragers received a one-time topical exposure (1 µL drop on dorsal side of thorax) prior to analysis of homing flight. Behavior was measured between 3 and 6 hours after exposure. The doses tested were ½, ¼, 1/10th, 1/20th and 1/40th of the measured LD_{50} (= 21.8 ng/head). <u>Clothianidin</u> Flight times were not significantly different among treatments at any dose. While the proportion of successful homing flights were significantly reduced at doses of 1/10th (2.18 ng/head) and greater.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that clothianidin has an effect on homing flight when topically exposed to 2.18 ng/head (and up to 10.9 ng/head). The doses given are not reported in units typically considered in the risk assessment. Sublethal effects are more properly considered in chronic risk assessments than for acute topical exposure. The study provides supplemental information only on behavioural effects of acute exposure to *Apis mellifera*.

Ref ID	Reference	Summary	Categorisation & comments
21 (Initial search)	Laycock et al. (2014) Effects of the neonicotinoid pesticide thiamethoxam at field-realistic levels on microcolonies of *Bombus terrestris* worker bumble bees. *Ecotoxicology and environmental safety*, Vol. 100, pp. 153-8.	Sublethal effects from chronic exposure to thiamethoxam on bumble bee (*Bombus terrestris*) micro-colonies was investigated under laboratory conditions. Effects on brood (eggs and larvae) production, food consumption and days survived by workers were recorded. Bees were fed for 17 days on syrup, ad libitum, at at a range of test concentrations (0.06 to 98 µg a.s./kg). The test material used in the study was technical active substance. <u>Thiamethoxam</u> There was a significant reduction in feeding and brood production in when exposed to a dietary concentration of 39.4 µg a.s./kg or above for 17 days. At lower dosages (15.7 µg a.s./kg and lower), microcolonies consumed syrup and pollen at normal control rates and brood production was not detectably dose-dependent. The number of days survived by workers was only significantly reduced at 98.4 µg a.s./kg. 15.7 µg a.s./kg = 5.101 ng a.s./bee/day 39.4 µg a.s./kg = 7.379 ng a.s./bee/day 98.4 µg a.s./kg = 14.785 ng a.s./bee/day	(b) Study that only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam has an effect on bumble bee brood production when orally exposed to ≥39.4 µg a.s./kg (in artificial nectar) under laboratory conditions. Effects on larval and brood development for bumble bees is not a data requirement. The data from this study would not impact the honeybee risk assessment. Therefore the study provides supplemental information only on reproduction effects of chronic exposure to bumble bees *(Bombus terrestris)*.

Ref ID	Reference	Summary	Categorisation & comments
154 (Initial search)	Mommaerts et al. (2010). Risk assessment for side-effects of neonicotinoids against bumblebees with and without impairing foraging behavior. *Ecotoxicology*, Vol. 19, No. 1, pp. 207-15.	Sublethal effects from chronic exposure to thiamethoxam on bumble bee (*Bombus terrestris*) micro-colonies was investigated under laboratory conditions. Effects on worker survival, nest development and reproduction (numbers of drones produced per nest) were recorded for a period of 11 weeks. Worker bumblebees exposed via the drinking of treated sugar water (0.01 to 100 ppm). The test material used in the study was the formulation Actara (25% WG). Additionally, effects on survival and reproduction when the assay also included foraging were recorded when bees were exposed to 0.1 ppm only. The daily consumption of sugar water per bumblebee worker was determined to be 277 ± 16 µL. <u>Thiamethoxam</u> Survivial: 11 week LC_{50} = 0.12 ppm Reproduction: total loss of reproduction at 0.5 to 100 ppm due to high worker mortality. Significant effects were observed at 0.1 ppm and there were no effects on reproduction at 0.01 ppm. When foraging involved: 85% mortality and a significant effect on reproduction at 0.1 ppm.	(b) Study that only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam has an effect on bumble bee brood production when orally exposed to ≥0.1 ppm (in artificial nectar) under laboratory conditions. Effects on larval and brood development for bumble bees is not a data requirement. The data from this study would not impact the honeybee risk assessment. Therefore the study provides supplemental information only on reproduction effects of chronic exposure to bumble bees *(Bombus terrestris)*.

Ref ID	Reference	Summary	Categorisation & comments
455 (Initial search)	Fauser-Misslin et al. (2014). Influence of combined pesticide and parasite exposure on bumblebee colony traits in the laboratory. *Journal of Applied Ecology*, Volume 51, Number 2, pp. 450-459.	Sublethal effects from chronic exposure to thiamethoxam and clothianidin on bumble bee (*Bombus terrestris*) micro-colonies was investigated under laboratory conditions. Effects on worker production and longevity, colony sexual investment, queen longevity, pollen and sugar water collection were recorded over a 9 week period. Worker bumblebees were provided with spiked sugar water and pollen patties ad libitum (4 µg thiamethoxam/kg and 1.5 µg clothianidin/kg). The test materials used in the study were technical active substance. <u>Thiamethoxam and Clothianidin</u> Worker production: Significant effect at weeks 4 through 7 and 9 (worker production initially increased before declining towards the end of the colony cycle). Worker longevity: Lower survival rate compared to controls. Colony sexual investment: Significant decrease (results indicate a population-level loss of about 43% in males and 77% in queens). Male production was significantly decreased but queen production per colony was not significant (despite fewer queens). Pollen and sugar water collection: Sugar water collection was consistently lower across all weeks in exposed colonies. Pollen collection was initially the same, but diverged over the course of the experiment, with exposed colonies collecting less pollen per week per bee.	(b) Study that only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam and clothianin in mixture has an effect on bumble bee brood production when orally exposed to 4 µg/kg and 1.5 µg/kg, respectively (in artificial nectar and pollen) under laboratory conditions. Effects on larval and brood development for bumble bees is not a data requirement. The data from this study would not impact the honeybee risk assessment. Therefore the study provides supplemental information only on reproduction effects of chronic exposure to bumble bees *(Bombus terrestris)*.

Ref ID	Reference	Summary	Categorisation & comments
551 (Initial search)	Elston et al. (2013). Sub-lethal effects of thiamethoxam, a neonicotinoid pesticide, and propiconazole, a DMI fungicide, on colony initiation in bumblebee (*Bombus terrestris*) micro-colonies. *Apidologie*, Volume 44, Number 5, pp. 563-574.	Sublethal effects from chronic exposure to thiamethoxam on bumble bee (*Bombus terrestris*) micro-colonies was investigated under laboratory conditions. Effects on worker mortality, nestbuilding activity and egg laying were recorded over a 28 day period. Worker bumblebees were provided with spiked artificial nectar solution and pollen dough ad libitum (1 or 10 µg a.s./kg). The test material used in the study was technical active substance. <u>Thiamethoxam</u> Nectar consumption: Signifcant decrease at both test doses. Survival: No significant effect during the 28-day period at both test doses. Nest building and brood production: In the 10 µg/kg dose, there was a significant difference between the time to initiation of nest building activity, significantly fewer eggs and larvae were produced over the experimental period and resulted in the production of no larvae during the experiment. Significantly fewer honey pots were produced in micro-colonies exposed to both doses.	(b) Study that only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam has an effect on bumble bee brood production when orally exposed to 10 µg a.s./kg (in artificial nectar) under laboratory conditions. Effects on larval and brood development for bumble bees is not a data requirement. The data from this study would not impact the honeybee risk assessment. Therefore the study provides supplemental information only on reproduction effects of chronic exposure to bumble bees *(Bombus terrestris)*.

Ref ID	Reference	Summary	Categorisation & comments
489 (Initial search)	Sandrock et al. (2014). Sublethal neonicotinoid insecticide exposure reduces solitary bee reproductive success. *Agricultural and Forest Entomology*, Volume 16, Number 2, pp. 119-128.	Sublethal effects from chronic exposure to thiamethoxam and clothianidin on solitary bee (*Osmia bicornis*) reproduction was investigated under laboratory conditions. Effects on mortality and reproductive output (number of fully developed cocoons, number of under developed offspring, hatching success, sex ratio, and body weight of emerged offspring) were recorded. Adult bees were provided with spiked sugar water (2.87 µg thiamethoxam/kg and 0.45 µg clothianidin/kg) and pollen pellets ad libitum. The test materials used in the study were technical active substance. <u>Thiamethoxam and Clothianidin</u> Adult mortality: no effect on adult females' longevity. Nest production: 22% reduction in nests completed in the treated population Brood: Completed nests contained 43.7% fewer total brood cells compared to control. Relative offspring mortality was almost two-fold higher in the treated population. The proportion of offspring that completed larval development and/or were able to hatch after hibernation was significantly lower compared to the control. Significantly male-biased offspring sex ratios were detected across nests within the treated population, resulting in a significantly lower proportion of daughters overall compared to control. Taken together, the treatment population produced 47.7% fewer offspring than control. However there was no effect on mean body weight of emerged offspring.	(b) Study that only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that thiamethoxam and clothianidin in mixture has an effect on solitary bee reproduction when orally exposed to 2.87 µg/kg and 0.45 µg/kg, respectively (in artificial nectar) under laboratory conditions. Effects on larval and brood development for solitary bees is not a data requirement. The data from this study would not impact the honeybee risk assessment. Therefore the study provides supplemental information only on reproduction effects of chronic exposure to solitary bees *(Osmia bicornis)*.

Ref ID	Reference	Summary	Categorisation & comments
31 (Initial search)	Sandrock et al. (2014). Impact of chronic neonicotinoid exposure on honeybee colony performance and queen supersedure. *PloS one*, Vol. 9, No. 8	Sublethal effects from chronic exposure to thiamethoxam on honeybee (*Apis mellifera*) colonies were evaluated under semi-field conditions. Effects on colony growth, honey production and pollen consumption were recorded over a 1 year period. Chronic exposure through in-hive pollen feeding was performed for 46 days (1.5 months) in order to cover two brood cycles, thereby resulting in total provisions of 8 kg of pollen patties per colony. Test concentrations were 5.0 and 2.0 ppb for thiamethoxam and clothianidin, respectively. The test materials used in the study were technical active substances. After the exposure phase (mid-May 2011 to beginning of July 2011) the colonies were maintained for a further year. Colony assessments were performed in mid-October 2011, end March 2012, late April 2012 and June 2012. <u>Thiamethoxam and Clothianidin</u> *Colony growth:* After 1.5 months of exposure there was a significant decrease in number of adult bees (-28%). Also a significant overall decrease in total brood (-13%). After 3.5 months there were no effects on adults or total brood. After 1 year there was again a significant effect on number of adult bees and total brood. *Honey production and pollen consumption:* During the exposure period, overall honey production (-29%) and pollen collection (-19%) declined. Overall, the mean honey production over the entire season remained ower in the treated colonies. There was no indication that pollen storing and pollen consumption during the experimental pollen feeding was influenced by neonicotinoid exposure. *Pollen collection:* Both treatment groups collected similar amounts of pollen during the first 3 weeks of the exposure period but treated colonies consistently collected less pollen later on. *Supersedure of queens and tendency to swarm:* There was a significant association of neonicotinoid exposure and queen supersedure, as well as a significant association with a reduced propensity to swarm. Honeybee colonies exhibited a short-term decline in colony performance, but colonies recovered in the medium term and overwintered successfully. However the following spring colony performance was once again negative impacted.	(a) Study provides data for establishing or refining risk assessment parameters. Results indicate that thiamethoxam and clothianidin in mixture has an effect on honeybee colony health when orally exposed to 25.0 and 2.0 ppb, respectively (in artificial pollen) under semi-field conditions. There are limitations to this study, involving test design. Spiked pollen was provided as the source of exposure. Pollen traps were placed in the hives to prevent other pollen being collected. The pollen levels during the treatment period (when pollen traps in place) increased by 66% in the control but only by 20% in the treated hives. This suggests an effect on pollen storing, which was continued once the pollen traps were removed with normal pollen collection reduced by 50% (which could reflect lower brood levels). Pollen availability affects brood production (in the absence of sufficient pollen larvae are cannabalised) and brood presence affects pollen collection. Thus, there are knock-on effects if brood levels are reduced and colonies build up slower. The effects seen in this study may be a result of anti-feedant effects from the pollen used, rather than from the test item. The study results may only be considered in a weight-of-evidence approach of chronic risk to honeybee colonies due to the limitations identified in the study design.

Ref ID	Reference	Summary	Categorisation & comments
276 (Clothianidin search)	Lu et al. (2014). Sub-lethal exposure to neonicotinoids impaired honey bees winterization before proceeding to colony collapse disorder. *Bulletin of Insectology*, Volume 67, Number 1, pp. 125-130.	Sublethal effects from chronic exposure to clothianidin on honeybee (*Apis mellifera*) colonies were evaluated under semi-field conditions. Effects on colony growth, strength, mortality and morbidity plus over wintering success were recorded over a 13 month period (end June 2012 to early August 2013). Chronic exposure through in-hive artificial nectar feeding was performed for the first 13 weeks (3 months) of the study. The test concentration was 258 µg a.s./1.9L artificial nectar solution (equivalent to 0.74 ng a.s./bee/day, assuming 50,000 bees per colony). The test material used in the study was technical active substance. <u>Clothianidin</u> No effects on colony performance from June through December 2012. There was a significant difference in the number of frames containing bees from January to April 2013. The decreased cluster size led to the loss of 50% of the colonies in the treatment group compared to control (17% loss). In the treated colonies, a majority of the bee colonies had been abandoned the following spring (2013) and those with bees only conatiend small coloies without quuens or had no brood.	(b) Study that is relevant to the data requirement but only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that clothianidin has an effect on honeybee colony health when orally exposed to 258 µg a.s./1.9L (in artificial nectar) under semi-field conditions. The dose used in this study (equivalent to 0.74 ng a.s./bee/day, assuming 50,000 bees per colony) is not how test doses are reported in standard studies. This dose is considered to be well above environmentally relevant concentrations. The study also has other limitations, including lack of details regarding the source of the colonies, there are no clear details on colony size, and data are presented for adult bess but no information are presented on levels of brood. Therefore the study provides supplemental information only for effects on honeybee *(Apis mellifera)* colony health from chronic exposure.

Ref ID	Reference	Summary	Categorisation & comments
33 (Clothianidin search)	Scholer and Kirschik (2014). Chronic exposure of imidacloprid and clothianidin reduce queen survival, foraging, and nectar storing in colonies of *Bombus impatiens*. *PloS one*, Vol. 9, No. 3, pp. e91573	Sublethal effects from chronic exposure to clothianidin on bumble bee (*Bombus impatiens*) colonies were evaluated under semi-field conditions. Effects on individual behavior and colony health were recorded over an 11 week period in a greenhouse. Chronic exposure through in-hive artificial nectar (sugar syrup) feeding was performed for 11 weeks. The test concentrations were 10, 20, 50 and 100 ppb. The test material used in the study was technical active substance. <u>Clothianidin</u> *Queen mortality and movement:* There was a significant effects on queen mortality at test concentrations at and above 20 ppb after 11 weeks. At 6 weeks, significant effects were seen at 50 and 100 ppb. No significant effects on movement throughout the test. *Worker behaviour:* There was a significant decrease in the speed of bee movements at test concentrations at and above 20 ppb. Significantly more sugar syrup was consumed in weeks 2, 4, 6, and 8 in the control compared to all treatments. *Colony health:* There was a significant decrease in colony weight at test concentrations at and above 20 ppb after 11 weeks. There was a significant decrease in weight of syrup in wax syrup pots and number of stored syrup pots added at all test concentrations. Clothianidin was not toxic to brood, as dead brood was not significantly different among treatments. However, at week 11 the amount of alive brood was significantly reduced at test concentrations at and above 20 ppb, reflecting premature queen mortality. Total brood (dead and alive) was significantly decreased at 50 and 100 ppb. Daughter queen production was not significantly different among treatments. Additionally, the number of workers produced was not significantly different among treatments. However, the mean number of males produced was significantly decreased at 50 and 100 ppb. The numbers of bees on nest showed a significant interaction of week and treatment. However, when weeks were individually analyzed only at week 6, were significantly less bees on the nest in the 50 and 100 ppb treatments.	(b) Study that only supplies supplementary information that does not alter the risk assessment parameters. Results indicate that clothianidin has an effect on bumble bee colony health when orally exposed to 10 µg a.s./kg (in artificial nectar) under semi-field conditions. Effects on bumble bees colonies is not a data requirement. The data from this study would not impact the honeybee risk assessment. Therefore the study provides supplemental information only for effects on bumble bees *(Bombus terrestris)* colony health from chronic exposure.

Ref ID	Reference	Summary	Categorisation & comments
14 (Initial search)	Duso et al (2014). The impact of insecticides applied in apple orchards on the predatory mite *Kampimodromus aberrans* (Acari: Phytoseiidae). *Experimental & applied acarology*, (2014 Mar) Vol. 62, No. 3, pp. 391-414.	The effects of a number of insecticides (including thiamethoxam) on predatory mites (*Kampimodromus aberrans*) were evaluated under laboratory and field conditions. In the laboratory, mites were exposed to fresh residues on apple leaves at one application rate (30 g/hL). Effects on mortality and fecundity were recorded. As only the field application rate was tested and the endpoint is not given in g/ha, this aspect of the study provides supplemental information only and is not considered further. In the field, Actara 25WG was applied at 30 g/hL (1-3 applications) to an apple orchard (Italy). Populations were monitored by collecting leaves from each replicate and counting mites under a dissecting microscope for approximately 10 weeks after the 1st application. Single or multiple applications caused no detrimental effects on predatory mites. The study did not specify if a standard method was followed. It was not clear if the study was conducted according to GLP guidelines.	a) Study that provides data for refining risk assessment parameters The results from the field study indicate initial effects on mite populations (*Kampimodromus aberrans*), followed by recovery between 28 and 56 days after the second application. These results are similar to the results of a field study on the predatory mite *Typhlodromus pyri* submitted by the Notifier where there were intial effects followed by recovery at 56 days after treatment (2 x 100 g a.s./ha, 28 d interval). However the study is not reliable. Little information is given regarding the mite population and results are not transparent or completely reported. Addtionally the results that are reported do not give endpoints (ie. NOEAER) for use in risk assessment.

Ref ID	Reference	Summary	Categorisation & comments
1407 (Initial search)	Tosi et al (2006) Side effects of insecticides on *Kampimodromus aberrans*. *Informatore Agrario* (2006), Volume 62, Number 26, pp. 54-56	A field study was conducted to assess the impact of insecticides (including thiamethoxam) applied to grapevines in a single treatment on populations of *K. aberrans*. The study with thiamethoxam (Actara 25WG; 20 g/hL) was conducted in 2005 in a vineyard located in Verona, Italy. Samples were taken before and after treatment (on days 5, 10, 20, 40 and 60) in order to evaluate both the short- and long-term effect of the product. The phytoseiid population remained consistently at levels similar to those observed for the control group throughout the study duration. The study did not specify if a specific method was followed. It was not clear if the study was conducted according to GLP guidelines.	a) Study that provides data for refining risk assessment parameters The results from the field study indicate no effects on mite populations (*Kampimodromus aberrans*). These results are similar to the results of a field study on the predatory mite *Typhlodromus pyri* submitted by the Notifier where there were intial effects followed by recovery at 56 days after treatment (2 x 100 g a.s./ha, 28 d interval). Suitable as supporting data in a weight of evidence for the risk assessment.

Ref ID	Reference	Summary	Categorisation & comments
1769 (Initial search)	Beers et al (2005) Role of neonicotinyl insecticides in Washington apple integrated pest management. Part II. Nontarget effects on integrated mite control. *Journal of Insect Science*, 5	The effect of neonicotinyl insecticides (including thiamethoxam as 25 WG at 96 g a.s./ha) on integrated mite control in Washington apple was examined from 2000-2004. In a series of field trials designed primarily to look at efficacy against the codling moth, *Cydia pomonella*. Mite densities were assessed at 1- to 2-wk intervals for approximately 2 months. The mite species evaluated included the European red mite, *Panonychus ulmi* (Koch); twospotted spider mite, *Tetranychus urticae* Koch; McDaniel spider mite, *Tetranychus mcdanieli* McGregor; the predatory mites *Galandromus occidentalis* (Nesbitt) and *Zetzellia mali* Ewing; and the eriophyid apple rust mite, *Aculus schlechtendali* (Nalepa). The predatory mite populations in the thiamethoxam treatment were not different than the control. The study did not specify if a specific method was followed. It was not clear if the study was conducted according to GLP guidelines.	a) Study that provides data for refining risk assessment parameters The results from the field study indicate no effects on mite populations. These results are similar to the results of a field study on the predatory mite *Typhlodromus pyri* submitted by the Notifier where there were intial effects followed by recovery at 56 days after treatment (2 x 100 g a.s./ha, 28d interval). However the study is not reliable. The results are not suitable for use in risk assessment due to insufficient information provided and mite densities in the control were too low for robust statistical evaluation.

Ref ID	Reference	Summary	Categorisation & comments
6 (Clothianidin search)	Wang K, Pang S, Mu X, Qi S, Li D, Cui F and C Wang (2015). Biological response of earthworm, *Eisenia fetida*, to five neonicotinoid insecticides. *Chemosphere*, Vol. 132, pp. 120-6	Earthworms (*Eisenia fetida*) were exposed to clothianidin for 56 days. The data evaluated from the test was used to determine the effects on reproduction. Technical clothianidin was used as the test material. *Eisenia fetida* Clothianidin: 56d NOEC = 0.01 mg a.s./kg (based on mean cocoon weight) Based on OECD guideline no. 222. It was not clear if the study was conducted according to GLP guidelines. Analytical verification was carried out. Acute toxicity of earthworms (*Eisenia fetida*), was also tested. As an acute endpoint is no longer a data requirement, this part of the study is not considered relevant. Measurement of cellulose activity and effects on intestinal and epidermal histology were conducted. As these endpoints are not a data requirement or considered in the risk assessment, this part of the study is not considered relevant.	(b) Study that is relevant to the data requirement, but in the opinion of the applicant provide only supplementary information that does not alter existing risk assessment parameters. The results for clothianidin are similar to a study conducted by the Notifier (*Eisenia fetida* 56d NOEC = 0.06 mg/kg). The toxicity test was not conducted to GLP. This study provides supplemental information only on the chronic toxicity to *Eisenia fetida*. Additionally, higher tier field data is used to in the risk assessment.

Study Summaries and Reliability Evaluations part*
*This section was prepared from MCA section 8 dossier by applicant (Syngenta Japan) in 2022

Ref ID: 170

	Submitted because data concern toxicity to aquatic invertebrates:
Report:	K-CA 8.2.4.2/03, Barbee GC and Stout MJ (2009). Comparative acute toxicity of neonicotinoid and pyrethroid insecticides to non-target crayfish (*Procambarus clarkii*) associated with rice-crayfish crop rotations. (Syngenta File No. CGA293343_11854) Published, Pest Management Science Vol. 65, No. 11, pp. 1250-6.

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Most insecticides used to control rice water weevil (*Lissorhoptrus oryzophilus* Kuscel) infestations are pyrethroids. However, pyrethroids are highly toxic to non-target crayfish associated with rice–crayfish crop rotations. One solution to the near-exclusive relianceonpyrethroids in a rice–cray fish pest management program is to incorporate neonicotinoid insecticides, which are insect specific and effective against weevils but not extremely toxic to crayfish. This study aimed to take the first step to assess neonicotinoids as alternatives to pyrethroids in rice–crayfish crop rotations by measuring the acute toxicities of three candidate neonicotinoid insecticides, clothianidin, dinotefuran and thiamethoxam, to juvenile *Procambarus clarkii* (Girard) crayfish and comparing them with the acute toxicities of two currently used pyrethroid insecticides, lambda-cyhalothrin and etofenprox.

RESULTS: Neonicotinoid insecticides are at least 2–3 orders of magnitude less acutely toxic (96 h LC_{50}) than pyrethroids to juvenile *Procambarid* crayfish: lambda-cyhalothrin (0.16 μg a.s./L) = etofenprox (0.29 μg a.s./L) >>clothianidin (59 μg a.s./L) > thiamethoxam (967 μg a.s./L) > dinotefuran (2032 μg a.s./L).

CONCLUSION: Neonicotinoid insecticides appear to be much less hazardous alternatives to pyrethroids in rice–crayfish crop rotations. Further field-level neonicotinoid acute and chronic toxicity testing with crayfish is needed.

Material

Test material	Thiamethoxam (CAS no. 153719-23-4) Clothianidin (CAS no. 210880-92-5)
Source:	Thiamethoxam : Chem Sevice, Inc., West Chester, PA, USA Clothianidin: Sigma-Aldrich, St. Louis, MO, USA
Purity:	Thiamethoxam : 99.5% Clothianidin: 99%

Treatments

Test concentrations:	Thiamethoxam : 0, 110, 200, 363, 660, 1200 µg a.s./L Clothianidin: 0, 500, 800, 1000, 1200, 1250 µg a.s./L
Solvent:	Acetone
Negative control:	Dilution water without test item
Method of administration:	Nominal insecticide test concentrations were prepared by spiking the appropriate volume of stock solution using a micropipette (Cole-Parmer, Chicago, IL, USA) into the known volume of chamber dilution water.
Treatment level(s):	Five plus control

Test organism

Species:	Rice-crayfish (*Procambarus clarkii*)
Source:	Canals at the Louisiana State University Aquaculture Research Station (LSU AgCenter ARS, BatonRouge)
Life stage of test organisms at study initiation:	Juvenile, Thiamethoxam: 3 months, Clothianidin: 3.5 months
Acclimatisation prior to testing:	Minimum 14 days to ensure that they were disease free and to slowly acclimatize them to test conditions
Feeding:	Fish were fed twice weekly with Wardley Shrimp Pellets Formula™ (The Hartz Mountain Corp., Secaucus, NJ, USA) during holding and acclimatisation.
Sex:	Male and female
Size:	Thiamethoxam: The average body length of the fish was 6.8 cm (measured form the tip of the rostrum to the end of the telson), the average body wet weight was 8.3 g Clothianidin: The average body length of the fish was 7.6 cm (measured form the tip of the rostrum to the end of the telson), the average body wet weight was 13.2 g
Comments remarks:	Natural crayfish aggression and cannibalism were minimized during quarantine by placing solvent-washed PVC pipe couplings in the coolers.

Test design

Test principle:	Acute toxicity test
Test units:	20 L glass jars with Teflon®-lined cork lids (Web Infusions, Inc., Irving, TX, USA) with 10 L test solution.
Water/medium type:	Not stated.
Number of replicates:	Two
Medium:	Static-renewal
Individuals per replicate:	10
Study duration:	96 hours
Method (guideline):	ASTM E729-96
Environmental conditions	
Feeding:	Crayfish were not fed 48 hours prior to toxicity test initiation or during 96 hours exposure period.
Water/medium parameters:	Total hardness 268 mg/L

Laboratory conditions:	Total alkalinity	214 mg/L
	Total chlorine (Cl_2)	< 0.05 mg/L
	pH	7.4
	Photoperiod	16 hours light: 8 hours dark
	Temperature	Thiamethoxam: average 21.2°C (min. 20.1 °C, max 22.2 °C) Clothianidin: average 21.4°C (min. 20.3 °C, max 22.5 °C)
Aeriation:	Before insecticide addition, dilution water was intensively aerated for 24 h to ensure dissolved oxygen levels were adequate (i.e. 60–100% saturation) for crayfish survival.	
Comments remarks:	Preliminary range-finding toxicity tests were performed to bracket the LC_{50} concentration for the definitive toxicity test	
Analytical verification		
Method (guideline/protocol):	Not stated.	
Reference item:	Not stated.	
Recovery:	Not stated.	
Limit of detection:	Not stated.	
Limit of quantification:	Not stated.	
Assessments		
Endpoints / biological parameters:	Lethal and sublethal effects (e.g. erratic behavior, spastic movement, lethargy)	
Measurement frequency:	1, 12, 24, 48, 72 and 96 hours after test initiation	
Statistical analyses:	Data derived from each 96 h definitive toxicity test were used to estimate the median lethal concentration to 50% of the crayfish test population (LC_{50}) and 95% confidence limits. The maximum likelihood regression method with probit transform (i.e. probit analysis) was performed using TOXCALC® (Tidepool Scientific Software, McKinleyville, CA, USA). The no-observed-effect concentration (NOEC) was determined if test data were sufficient. The NOEC was the highest tested concentration at and below which there were no toxicant-related mortalities or observed behavioral abnormalities with respect to the dilution water/solvent control organisms.	

Results and Discussion

The estimated 96 h LC_{50} values and 95% confidence limits for thiamethoxam and clothianidin, and NOEC, are presented in the table below. For definitive acute toxicity tests, no mortality or sublethal effects were observed among crayfish exposed to the dilution water/solvent control.

Sublethal concentrations of thiamethoxam caused distinct behavioral abnormalities in the crayfish. Concentrations from about 500 to 700 µg a.s./L, caused extreme aggressiveness in crayfish, which resulted in significant mortality (up to 80%), likely owing to aggression and cannibalism but not to direct chemical toxicity.

Remarkable behavioural abnormalities were not observed in crayfish exposed to sublethal concentrations of clothianidin, and definitive toxicity test data were not sufficient to estimate a NOEC value.

Estimated 96 h LC_{50} values for thiamethoxam and clothianidin in juvenile crayfish

Test substance	Estimated LC_{50} (µg a.s./L)	95% confidence interval (µg a.s./L)	NOEC (µg a.s./L)
Thiamethoxam	967	879 – 1 045	<500
Clothianidin	59	6 – 137	Not determined*

* NOEC was not determined since data was insufficient

Validity Criteria

Not applicable, no guideline followed.

Conclusions

Neonicotinoid insecticides appear to be much less hazardous alternatives to pyrethroids in rice–crayfish crop rotations. Further field-level neonicotinoid acute and chronic toxicity testing with crayfish is needed. Based on the findings, the 96 h LC_{50} of thiamethoxam and clothianidin to juvenile crayfish (*Procambarus clarkii*) exposed in a static-renewal water system was 967 and 59 µg a.s./L, respectively.

(Barbee G et al, 2009)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		Score	Evaluator's comments on criteria (optional)
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	1	
10	Is information on the source/origin of the substance given?	1	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	
	Subtotal Test Substance	**3**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	

13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	1	
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**4**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**22**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	Minimal requirements given
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		

Date/period of evaluation:	03.06.2015
Evaluator information	
Name	Sebastian Schadt
Affiliation	Dr. Knoell Consult

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable • 22 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Well-documented study meeting all criteria for a highly scientific reliability.

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Limitations		Results are based on nominal test concentration and no analytical verification of the adminisitered doses was carried out.
GLP		No
Relevance	Supplemental data for acute toxicity to aquatic invertebrates	Reported LC_{50} values are suitable as additional toxicity data for tier-2 RAC derivation by calculating geometric mean or establishing a species sensitivity distribution (SSD).

Ref ID: 292

	Submitted because data concern toxicity to aquatic invertebrates:
Report:	K-CA 8.2.4.2/04, Stevens MM, Helliwell S, and PA Hughes (2005). Toxicity of *Bacillus thuringiensis* var. israelensis formulations, spinosad, and selected synthetic insecticides to *Chironomus tepperi* larvae. (Syngenta File No. CGA293343_11857) Published, Mosquito Control Association, Vol. 21, No. 4, pp. 446-450.

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Three *Bacillus thuringiensis* var. israelensis (BTi) formulations, the bacterial metabolite spinosad, and 7 synthetic insecticides were bioassayed against 4th instars of *Chironomus tepperi*, a serious pest of rice in southern Australia. The BTi formulations returned 48-h product median lethal concentration (LC_{50}) values (25 ± 1°C) of between 0.59 mg/liter (VectoBac® water-dispersible granule [WDG], 3,000 international toxic units [ITU]/mg) and 2.15 mg/liter (Teknar® suspension concentrate [SC], 1,200 ITU/mg). When LC_{50} values were adjusted to reflect nominal ITU values of the 3 products, there was still substantial variation, with LC_{50} values ranging from 1,770 ITU/liter (VectoBac WDG) to 2,580 ITU/liter (Teknar SC). Aquabac® SC (1,200 ITU/mg) showed intermediate activity. Differential activity between formulations may reflect faster settling rates in the more active formulations, which may be a beneficial characteristic when controlling benthic species such as *C. tepperi*. Spinosad (24 h LC_{50} = 28.9 µg a.s./L) and the synthetic insecticides we evaluated were all substantially more active than BTi. The highest activity was shown by the neonicotinoid compounds thiacloprid, acetamiprid, and clothianidin, which all returned 24 h LC_{50} values between 1 and 3 µg a.s./L. Indoxacarb and thiamethoxam showed the lowest activity of the synthetic compounds evaluated.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam and clothianidin are included below.

Materials

Test material	Clothianidin and Thiamethoxam
Source:	Clothianidin: TI 435, 200 g a.s./L SC, lot 0018; Bayer CropScience Australia Pty. Ltd. (Melbourne, Australia) Thiamethoxam: Actara® 250 g a.s./kg WDG, lot KW 107076; Syngenta Crop Protection Pty. Ltd. (Sydney, Australia)
Purity:	No information
Water solubility:	No information

Treatments	
Test concentrations:	No information
Solvent:	1 x Martin's/ thiamine hydrochloride
Control:	1 x Martin's/ thiamine hydrochloride
Method of administration:	Test suspension, prepared by serial dilution

Test organism

Species:	*Chironomus tepperi*
Source:	Temporary water bodies in the vicinity of Yanco Agricultural Institute (34°37'S, 146°26'E)
Age/life stage at study initiation:	4th instars
Holding conditions prior to test:	Temporary water bodies in the vicinity of Yanco Agricultural Institute (34°37'S, 146°26'E)
Acclimatisation prior to testing:	Rearing in aquaria containing 10 liters of 1 X Martin's rearing solution supplemented with thiamine hydrochloride (1.2 mg/L) lined with ethanol washed paper tissues for dietary bulk and tunnel building material. 4-6 egg masses/ aquarium, with a small quantity of K9® fish food (Carnation Pty. Ltd., Melbourne, Australia). Aquaria were covered with plastic film to reduce evaporation and prevent contamination. Aeration through hypothermic needle attached by plastic tubing to an aquarium aerator. 2-3 day intervals during larval development, bioassays used actively feeding early 4th instars of *C. tepperi* between 10-14 days old.

Test design

Test units:	Flat bottomed glass specimen tubes 100 mm high, 25 mm in diameter (Australian Entomological supplies Pty. Ltd., Bangalow, Australia)
Test medium:	1 x Martin's/ thiamine hydrochloride solution, paper tissue strips (3 X 15 mm), soaked in ethanol for 24 h, drained, air dried (min. 3 days), as substrate
Replication:	18 per final bioassay, 3 untreated controls, 3 for each of 5 concentration levels, 4 replicates per concentration
Individuals per replicate:	10 *C. tepperi* 4^{th} instars per container, (= total of 120 larvae to each concentration)
Duration:	Larvae assessed for mortality after 24 h
Exposure regime:	Static

Environmental conditions

Medium temperature:	25±1°C
pH:	No information
Feeding:	No feeding during pesticide exposure
Laboratory conditions:	15/9 h light/dark regimen

Analytical verification

Method:	No method for analytical verification is reported.

Assessments

Endpoints:	Acute toxicity: Median lethal concentration LC_{50}, 90% lethal concentration LC_{90}
Measurement frequency:	24 h after test initiation
Statistical analyses:	Chi-square
Analytical parameters:	No information

Results and Discussion

In this study clothianidin and thiamethoxam amongst a range of other compounds were bioassayed in the laboratory to obtain baseline data on their activity and to determine their potential for chironomid control in rice crops. Clothianidin was more toxic than thiamethoxam, which was the least toxic out of all tested compounds. Clothianidin's low acute LC_{50} (<3 µg/L) makes it interesting as an alternative for chironomid

control. Lower acute toxicity of thiamethoxam was attributed to reduce uptake and/or limited conversion under the given bioassay conditions.

Acute toxicity (24 h) of clothianidin and thiamethoxam to 4th instars of *C. tepperi*.

Test substance	LC_{50} (µg/L)[1,2]	LC_{90} (µg/L)[1,2]	Slope[3]	Chi-square (3df)
Clothianidin	2.83 (2.60-3.08)	5.19 (3.95-6.83)	4.6 (0.5)	2.2
Thiamethoxam	121 (108-136)	171 (155-187)	8.2 (0.7)	0.7

[1] Numbers in parentheses next to median lethal concentration (LC50) and 90% lethal concentration (LC90) values are 95% fiducial limits,
[2] Active substance concentration,
[3] Standard error of slope in parentheses.

Conclusions

Clothianidin was more toxic to *Chironomus tepperi* than thiamethoxam. Clothianidin's low acute LC_{50} (<3 µg/L) makes it interesting as an alternative for chironomid control.

(Stevens et al, 2005)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		**Score**	**Evaluator's comments on criteria (optional)**
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	
3	Is the administration route given?	1	directly applied to water media but without details
4	Are doses administered or concentrations in application media given?	0	test concentrations are not stated explicetely; only number of test concentrations is given
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**7**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	only nominal content
10	Is information on the source/origin of the substance given?	1	including batch number
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	not relevant
	Subtotal Test Substance	**2**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	not relevant, larvae were used for toxicity testing

13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	source of collection and rearing conditions reported
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	1	
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	not relevant but anyways reported
	Subtotal Test Organisms	**4**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	0	no informatin given about water parameters
17	Are sufficient details of the administration scheme given to judge the study?	0	no details given about administration scheme
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	not relevant; however, analytical verification are recommended for aquatic systems but is not reported
	Subtotal Study design / Test method	**1**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	0	results are based on nominal concentrations, anlytical results would be needed for completeness
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**2**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**17**	
	A Numerical result leads to initial Category:	**2**	
	B Cut-off criteria restricts Category:	**Yes**	Minimal criteria not met
	C Evaluator's proposed Category:	**2**	
	D Justification in case evaluator deviates from B:	Study is considered reliable with restriction even though the cut-off criterion for test concentrations is not sufficiently stated. However, it is reported that 5 concentrations were used providing a range of mortality allowing LCx calculations.	
Date/period of evaluation:		29.07.2015	
Evaluator information			
Name		Stefan Kroder	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 2	Reliable with restrictions. • 17 out of 22 reliability criteria are met according to

Qualitative assessment		
Study assessment	**Score**	**Rationale**
		ToxRTool (Schneider et al. 2009) but 1 cut-off criterion was not sufficiently met. • Even though test concentrations are not explicitely reported study meets basic scientific principles. The information given provides confidence in properly chosen test concentrations for scientific reliability. This is further supported by the goodness of fit in results of probit analysis.
Limitations		Test concentrations are not clearly stated and no information reported about test media used for experiments and the exact administration scheme. Results are based on nominal concentrations in the lack of analytical verification.
GLP		No
Relevance	Supplemental data for acute toxicity to aquatic invertebrates	Acute LC_{50} value of an additional species of aquatic invertebrates that is suitable for use in risk assessment to establish a species sensitivity distribution (SSD) and geometric mean calculations at tier 2 or at least as data for weight-of-evidence approach.

Ref ID:1

	Submitted because data concern toxicity to aquatic invertebrates:
Report:	K-CA 8.2.4.2/05. Ugurlu P, Unlu E and EI Satar (2015). The toxicological effects of thiamethoxam on *Gammarus kischineffensis* (Schellenberg 1937) (Crustacea: Amphipoda). (Syngenta File No. CGA293343_11858), Published, Environmental toxicology and pharmacology, Vol. 39, No. 2, pp. 720-6.

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Neonicotinoids are a new group of insecticides, and little is known about their toxicity to nontarget freshwater organisms and potential effects on freshwater ecosystems. The aim of this study is to establish the acute toxicity and histopathological effects of thiamethoxam-based pesticide on the gill tissue of *Gammarus kischineffensis*. In this study *G. kischineffensis* samples were exposed to 2.5, 5, 7.5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 60, 70, 80, 90 and 100 mg/L of commercial grade thiamethoxam for 96 h. The 24, 48, 72 and 96 h LC_{50} values were determined as 75.619, 23.505, 8.048 and 3.751 mg/L, respectively. In the histopathological study the individuals were exposed to 0.004, 0.04 and 0.4 mg/L thiamethoxam concentrations for 14 days. The results showed that the most common changes at all doses of thiamethoxam were vacuolization and hemostatic infiltration in the gill tissue of *G. kischineffensis.*

Materials

Test material	Thiamethoxam (Actara® 240 SC)
Source:	Gani Pesticides Marketing Ltd. Company
Purity:	240 g/L
Water solubility:	Not stated
Storage conditions:	Not stated
Comments remarks:	The stock thiamethoxam solution was prepared from commercial total pesticide mixture by dissolving this mixture pesticide in distilled water. It was taken into account that there was 240 g thiamethoxam in 1 L of pesticide mixture while test solutions were being prepared. The test concentration was obtained without any dilution with water.
Treatments	
Test concentrations:	Acute toxicity test: 2.5, 5, 7.5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 60, 70, 80, 90 and 100 mg a.s./L. Histopathological study: 0.004, 0.04 and 0.4 mg/L
Solvent:	None
Positive control:	None
Method of administration:	2L glass jars were filled with 1L test solution. 20 (acute toxicity test) or 30 (histopathological study) individuals per jar. The test design was static renewal.
Evidence for unsolved material:	Not stated
Comments remarks:	Before acute toxicity test, range finding tests have been performed.

	In the histopathological study, the sublethal concentrations of thiamethoxam were determined according to LC_{50} value. 1/10, 1/100 and 1/1000 of the LC_{50} value were used as sublethal concentrations of thiamethoxam.

Test organism

Species:	*Gammarus kischineffensis*
Source:	Tigris River in Diyarbakır, Turkey. (with long-handled sieves and dip nets in an unpolluted slow-running stream under stones and leaves)
Age/life stage at study initiation:	Not stated
Holding conditions prior to test:	The animals were maintained in a controlled room at 18±1 °C under an artificial light regime (13 h light:12 h dark). Animals were fed with decomposed willow (Salix sp.) leaves.
Acclimatisation prior to testing:	In the laboratory undamaged and active individuals were placed in 40×35×40 cm glass aquariums containing 50:50 mixture field-sampled freshwater and dechlorinated tap water. Each aquarium was aerated with air stones. Approximately 50% of each aquarium's water was substituted by dechlorinated tap water daily. The animals were acclimated to the laboratory conditions for 2 weeks before the experiments.
Comments remarks:	The decomposed willow (Salix sp.) leaves were collected from the sampling site and conditioned in field-sampled water for at least 2 weeks prior to use.

Test design

Test units:	2L glass jars.
Test medium:	1L dechlorinated tap water.
Replication:	Both in acute toxicity test and histopathological study: 3 replicates
Individuals per replicate:	Acute toxicity test: 20 individuals Histopathological study: 30 individuals
Duration:	96 h acute toxicity test and 14 d histopathological study
Exposure regime:	Static renewal
Comments remarks:	Static renewal: About 50% of the water in the all aquaria was substituted at 24 h intervals to maintain water quality and to prevent degradation of the pesticide. After replenishment, in order to maintain the concentration, 50% of pesticide concentration was added to the test solutions.

Environmental conditions

Medium temperature:	18±1 °C
pH:	7.94 ± 0.505
Feeding:	With decomposed willow (Salix sp.) leaves.
Other medium parameters:	DO: 7.5 ± 0.38 mg/L Total chlor: 42.6 mg/L Total hardness: 287 ± 2.35 mg/L $CaCO_3$ Mg: 36 mg/L Conductivity: 7.94 Mmho/cm NO_3-N: 2.1 mg/L NO_2-N: 0.002 mg/L
Laboratory conditions:	The animals were maintained in a controlled room at 18±1 °C under an artificial light regime (13 h light:12 h dark).
Comments remarks:	The decomposed willow (Salix sp.) leaves were collected from sampling site and conditioned in field-sampled water for at least 2 weeks prior to use.

Analytical verification

Method: No analytical verifications reported

Assessments

Endpoints: Acute toxicity test: behavioural changes, immobility and mortality
Histopathological study: histopathological alterations

Measurement frequency: Acute toxicity test: 24h intervals (after 24, 48, 72 and 96 h of exposure)
Histopathological study: 7d intervals (at 7th and 14th day)

Statistical analyses: Acute toxicity test: Probit analysis in SPSS statistical software
Histopathological study: none

Analytical parameters: Acute toxicity test: mortality
Histopathological study: none

Comments remarks: Determination of histopathological alterations: at 7th and 14th day 5 samples were taken randomly from exposure and control groups, the organisms were sacrificed by dissecting them from 2nd and 7th thoracic segments. Then the sample were fixed in formalin solution for 24 h and prepared according to the n-butyl alcohol technic. Sections of 5 µm were prepared from paraffin blocks by using a rotary microtome. These sections were then stained with hematoxylin–eosin, examined under a light microscope and the histopathological alterations were photographed.

Results and Discussion

Acute toxicity test:

In the control group, there was no death for 96 h. The death rate increased with the increasing concentration. 100% mortality was observed at 1000 mg/l after 24 h. All individuals at 500 mg/L died after 48 h. At 25, 30 and 35 mg a.s./L concentrations the mortality was 78% after 72 h. The % mortality of the individuals exposed to increasing grades of thiamethoxam was given in Table 8.2.4-10.

The % mortality of individuals exposed to increasing grades of thiamethoxam

Concentration (mg a.s./L)	Mortality (%)			
	24h	48h	72h	96h
2.5	0	17	30	35
5	7	20	41	53
7.5	7	27	53	58
10	7	28	63	76
15	11	46	68	79
20	13	51	73	84
25	15	53	78	82
30	18	62	78	93
35	30	65	78	95
40	20	70	85	93
45	30	73	100	100
50	33	88	100	100
60	38	95	100	100
70	43	95	100	100
80	50	100	100	100
90	58	100	100	100
100	63	100	100	100

500	83	100	100	100
1000	100	100	100	100

After Probit analyses, 24, 48, 72 and 96 h LC_{50} values and 95% confidence limit of commercial grade thiamethoxam for *G. kischineffensis* were given in Table 8.2.4-11 .

LC_{50} value and 95% confidence limit of commercial grade thiamethoxam for *G. kischineffensis.*

Hours	LC_{50} value (mg a.s./L)	95% confidence limit (mg/L)
24	75.619	66.678 - 88.504
48	23.505	18.843 - 27.731
72	8.048	1.319 - 12.744
96	3.751	3.506 - 8.332

Histopathological study:

No histopathological changes were observed in the gill of the control group relative to experiment group. Histopathological results indicated that gill was the primary target tissue affected by thiamethoxam. The most common changes at all doses of thiamethoxam were vacuolization and hemostatic infiltration. At the 7th day of exposure to 0.004, 0.04 and 0.4 mg/L thiamethoxam, the gills of experimental samples showed vacuolization in many areas and collapse of the pillar cells. At the 14th day of exposure to 0.004 and 0.04 mg/L thiamethoxam, necrosis and hemostatic infiltration was noticed. In the group exposed to 0.4 mg/L thiamethoxam at the 14th day, since more than 10% of the *G.kischineffensis* individuals were dead, results of this experimental group were not included in the test results.

Conclusions

Results have revealed that thiamethoxam is highly toxic to *G.kischineffensis*, the 96h LC_{50} value was 3.751 mg a.s./L. This chemical has rapid and acute effects on the individuals of *G. kischineffensis*. Accidental leakage of this chemical, even at very low concentrations, can cause lethal consequences for this species.

All the histopathological observation indicated that exposure to lethal concentrations of thiamethoxam caused destructive effect in the tissues of *G. kischineffensis*. Tissue alterations, such as those observed in this study and findings from previous studies, may result in severe functional problems, ultimately leading to the death of organism.

(Ugurlu et al, 2015)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		**Score**	**Evaluator's comments on criteria (optional)**
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	*Gammarus kischineffensis*
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	

5	Are frequency and duration of exposure as well as time-points of observations explained?	1	96 h acute toxicity test and 14 d sublethal toxicity test
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	Not stated.
10	Is information on the source/origin of the substance given?	1	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	0	E.g. no purity and solubility is stated.
	Subtotal Test Substance	**1**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	0	Not stated.
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	0	Not stated.
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**2**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	0	The acute test was conducted as a static renewal test however no analytics were performed.
	Subtotal Study design / Test method	**2**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	

	Subtotal Plausibility	**1**	
	Total Score	**17**	
	A Numerical result leads to initial Category:	**2**	
	B Cut-off criteria restricts Category:	**No**	Minimal requirements given
	C Evaluator's proposed Category:	**2**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		13.07.2015	
Evaluator information			
Name		Sebastian	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 2	Reliable with restrictions. • 17 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Well-documented and scientifically sound study.
Limitations		No data given for purity or solubility of the test substance. Some information on the test species (e.g. sex, age) are missing. No analytical verification was conducted.
GLP		No
Relevance	Supplemental data for acute and chronic toxicity to aquatic invertebrates.	Acute LD_{50} values of additional species are suitable for establishing a species sensitivity distribution (SSD) or geometric mean calculation at Tier 2. Sublethal results are suitable for weight-of-evidence approach.

Ref ID: 3

	Submitted because data concern toxicity to aquatic invertebrates:
Report:	K-CA 8.2.8/08, de Perre C, Murphy TM and MJ Lydy (2015). Fate and effects of clothianidin in fields using conservation practices. (Syngenta File No. CGA322704_10074) Published, Environmental toxicology and chemistry / SETAC, Vol. 34, No. 2, pp. 258-65.

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Despite the extensive use of the neonicotinoid insecticide clothianidin, and its known toxicity to beneficial insects such as pollinators, little attention has been given to its fate under agricultural field conditions. The present study investigated the fate and toxicity of clothianidin applied every other year as a corn seed-coating at 2 different rates, 0.25 mg/seed and 0.50 mg/seed, in an agricultural field undergoing a corn–soybean annual rotation, and conservation tillage. Concentrations were measured in soil, surface runoff, infiltration, and groundwater from 2011 to 2013. Clothianidin was detected at low concentrations in soil and water throughout the 2-yr corn and soybean rotation. Low and no-tillage had little or no effect on clothianidin concentrations. Laboratory toxicity bioassays were performed on nontarget species, including *Daphnia magna, Hyalella azteca, Chironomus dilutus, Pimephales promelas,* and *Eisenia fetida*. Risk quotients were calculated from clothianidin concentrations measured in the field and compared with the laboratory toxicity bioassay results to assess the environmental risk of the insecticide. The risk quotient was found to be lower than the level of concern for *C. dilutus*, which was the most sensitive species tested; therefore, no short-term environmental risk was expected for the species investigated in the present study.

Data on the non-relevant substances and non-relevant environmental media included in this study are not summarized here. Only data on the ecotoxicological effects of clothianidin in the environment are included below.

Materials

Test material	Clothianidin
Source:	e.g. Sigma-Aldrich Laborchemikalien, Seelze, Germany (if available Lot/Batch number)
Purity:	No information
Water solubility:	No information
Treatments	
Test concentrations:	7 concentration levels, more when no toxicity was observed at highest level.
Solvent:	None
Control:	Negative and solvent control, no further details
Method of administration:	Spiked water and soil
Verification of test concentration:	Concentrations were checked immediately before addition of the animals and at the end of each bioassay after the field sample protocols.

Test organism

Species:	*Daphnia magna, Hyalella azteca, Chironomus dilutus,* and *Pimephales promelas*
Source:	No information
Age/life stage at study initiation:	No information
Holding conditions prior to test:	No information
Acclimatisation prior to testing:	No information

Test design

Test units:	No information
Test medium:	*D. magna* and *C. dilutus* moderately hard water *H. azteca* and *P. promelas* hard water
Replication:	Triplicate for each concentration level
Individuals per replicate:	10
Duration:	*D. magna* 48 h *H. azteca, C. dilutus,* and *P. promelas* 96 h
Exposure regime:	static, exposed to spiked water

Environmental conditions

Medium temperature:	No information
pH:	No information
Feeding:	None during the experiment
Comments remarks:	*H. azteca* were fed at the beginning of the bioassays, and after 48 h with 0.2mL per beaker of a yeast, Cerophyl, and trout chow solution. All of the other species were not fed during the tests

Assessments

Endpoints:	Lethality (LC_{50} and EC_{50}), difficulty to swim, lack of or erratic movements
Measurement frequency:	No information
Statistical analyses:	Median to calculate LC_{50} or EC_{50} from probit regression results.
Analytical parameters:	Clothianidin concentrations in test solution immediately before addition of animals and at end of bioassays.

Analytical verification

Method:	liquid chromatography
Conduction:	1260 Agilent liquid chromatography–diode array detection equipped with a guard column and an Agilent (Agilent Technologies) Prep-C18 4.6- mm x 250-mm, 5-mm column. 20 mL of each sample was injected, and a constant flow of 0.75mL/ minute was used during the 28-min run. Solvent A consisted of a 95:5 (v/v) water – acetonitrile mixture, and solvent B was 100% acetonitrile. Mobile phase gradient: 13.7% of B for 1 min, increased to 36.8% of B in 7 min, increased to 63.2% of B for 6 min, increased to 100% of B in 1 min and held for 5 min; at 20 min, decreased back to 13.7% of B within 2 min and held until the end of the run. Visible lamp was off, only the ultraviolet lamp was used as the detector. Three wavelengths were acquired during the run, including 242nm, 252nm, and 269 nm, with a bandwidth and slit of 4 nm. The spectrum from 190 nm to 400 nm was recorded at a step of 1 nm for each peak (threshold of 0.1 mAU). The peak width was greater than 0.1 a minute, equivalent to a 2-s response time. Clothianidin was quantified using λ=269 nm (confirmed with λ=252 nm).

Reference item:	Clothianidin stock (99.9% pure, ChemService) in 0.1% trifluoroacetic acid water – acetonitrile 80:20 (v/v/v) surrogates: 2011: imidacloprid (99.5% pure, ChemService) and thiacloprid (99.9% pure, ChemService) 2012: acetamiprid (99.9% pure, ChemService)
Recovery:	80% to120%,
Limit of quantification:	Not stated (neither limit of detection)

Results and Discussion

The toxicity of clothianidin to reference nontarget species was relatively low. Clothianidin was shown to cause no sublethal or lethal effects to *D. magna* or *P. promelas*, at concentrations as high as 500 mg/L.

Clothianidin was more toxic to the other aquatic species tested, including *H. azteca* and *C. dilutus*, with EC_{50}s and LC_{50}s from 1.85 mg/L to 12.52 mg/ L.

Conclusions

Neonicotinoid insecticides are widely used due to their low toxicity to many nontarget species, despite their great efficiency toward target insects. This is especially true for clothianidin. The results of the present study confirm this point in that very high concentrations that were not environmentally relevant did not result in mortality to *D. magna* or *P. promelas*. The nontarget aquatic insect *C. dilutus* may be affected by clothianidin to a greater extent.

(de Perre et al, 2015)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		**Score**	**Evaluator's comments on criteria (optional)**
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	Clothianidin (metabolite of Thiamethoxam)
2	Is the species given?	1	*Daphnia magna, Hyalella azteca, Chironomus dilutus, Pimephales promelas, and Eisenia fetida*
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	0	No administered doses or concentration stated.
5	Are frequency and duration of exposure as well as time-points of observations explained?	0	Frequencey is not stated.
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**6**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	Not stated.
10	Is information on the source/origin of the substance given?	0	Not stated.

11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	0	E.g. purity, solubility are not stated.
	Subtotal Test Substance	**0**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	Not relevant.
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	0	Not stated.
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	0	Not stated.
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	0	It`s not sure whether the studies were conducted with repeated dose therefore the score is set to 0.
	Subtotal Test Organisms	**1**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	0	Study description is not sufficient.
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	0	It`s not sure whether the studies were conducted with repeated dose therefore the score is set to 0.
	Subtotal Study design / Test method	**1**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**12**	
	A Numerical result leads to initial Category:	**3**	
	B Cut-off criteria restricts Category:	**Yes**	Minimal criteria not met
	C Evaluator's proposed Category:	**3**	
	D Justification in case evaluator deviates from B:	Two cut-off criteria are not met. Therefore the study is not reliable.	
Date/period of evaluation:		14.07.2015	
Evaluator information			
Name		Sebastian Schadt	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 3	Not reliable • 2 reliability cut-off criteria are not met according to ToxRTool (Schneider et al. 2009) • Important information is not reported. Study does not meet basic scientific principles.
Limitations		Important information is missing, e.g. information about the test item and test species. Treatment doses and concentrations are not given and administration route is not reported.
GLP		No
Relevance	Not relevant	Ecotoxicological endpoints not suitable for risk assessment.

Ref ID: 35

	Submitted because data concern toxicity to aquatic invertebrates:
Report:	K-CA 8.2.4.2/06, Whiting SA and MJ Lydy (2014). A site-specific ecological risk assessment for corn-associated insecticides. (Syngenta File No. CGA322704_10071) Published, Integrated Environmental Assessment and Management, Published online 30 December 2014.

Guidelines

USEPA. 2000. Methods for measuring the toxicity and bioaccumulation of sediment-associated contaminants with freshwater invertebrates. Duluth (MN): USEPA. EPA 600/R-99/064.

GLP: No. Published study (peer-reviewed article).

Executive Summary

A site-specific ecological risk assessment (ERA) was conducted to examine the simultaneous use of genetically modified corn (Bt corn) with a neonicotinoid seed coating, clothianidin, and use of a granular insecticide, tefluthrin, to protect crops from pest damage. A field study was conducted on site, and exposure data from the literature were summarized to determine the matrices and exposure concentrations that nontarget species could typically experience within an agricultural ecosystem. To determine ecological effects on nontarget species, acute toxicity bioassays were conducted on earthworms (*Eisenia fetida*), amphipods (*Hyalella azteca*), and Elmid riffle beetle larvae (*Ancyronyx spp.*) in which the test species were exposed to single insecticides as well as the mixture of the 3 insecticides. In the risk characterization section of the ERA, stressor–response profiles for each species tested were compared with field distributions of the insecticides, and a margin of safety at the 10th percentile (MOS10) was calculated to estimate risk. No acute toxicity was observed in any of the 3 nontarget species after exposure to senescent Bt corn leaf tissue. Large MOS_{10} values were calculated for clothianidin to the nontarget species. When bioassays were compared with tefluthrin field distributions, very low MOS_{10} values were calculated for earthworms (0.06) and *H. azteca* (0.08) because the environmental concentrations often exceeded the stressor–response profile. No increased toxicity was observed when nontarget species were exposed to a mixture of the 3 insecticides. In summary, the genetically modified corn insecticidal proteins and clothianidin were not found at environmental concentrations exceeding benchmark values for ecological effects, but tefluthrin was consistently detected in the environment at levels that could be causing toxicity to nontarget species, especially if this pyrethroid is able to travel off site.

Data on the non-relevant substances and non-relevant environmental media included in this study are not summarized here. Only data on the ecotoxicological effects of clothianidin in the environment are included below.

Materials

Test material	Clothianidin
Source:	Not stated
Purity:	Analytical grade, but no detail
Storage conditions:	Not stated

Other specifications:	Clothianidin: water solubility is 340 mg/L at 20℃; half-life of photolysis is less than 1 day; hydrolysis stable; Koc is 84 to 129; vapour pressure is 9.8×10^{-10} mm Hg at 25℃.

Treatments

Test concentrations:	Single compound of clothianidin: not stated
Solvent:	Acetone (pesticide grade)
Method of administration:	Prepared the soil or solution with test concentrations using acetone, and put the test organisms on the surface of the soil or into the test solution.
Verification of test concentrations:	HPLC with a diode array detector according to Whiting et al. (2014).
Comments remarks:	If concentrations were within 10% of nominal values, the nominal value was used for the calculation of the LC_{50}; otherwise, the measured concentration was used in the toxicity calculations.

Test organism

Species:	Amphipod (Hyalella azteca), and riffle beetle (Ancyronyx spp.).
Source:	Amphipods were obtained from an established culture at Southern Illinois University. Riffle beetles were collected from woody material found in high flow area of the upper Cache River near Vienna, IL, USA.
Age/life stage at study initiation:	Amphipods: 7 to 10 days old. Individuals that passed through the 1 mm sieve, but remained on the 0.5 mm sieve were estimated to be the appropriate age for the bioassays. Riffle beetles: 3 to 7 mm.
Holding conditions prior to test:	Amphipods: not stated Riffle beetles: water temperature of 23 °C, and at least 7 days.
Acclimatisation prior to testing:	Amphipods: not stated Riffle beetles: larvae were taken back to the laboratory, transferred to a tank with filtered stream water, collected from the Cache River (IL, USA) at the same time as the beetle collection, and gradually acclimated (approximately 1°C/h) to a water temperature of 23 °C. Larvae were allowed to acclimate to the laboratory environment for at least 7 days before testing.
Comments remarks:	For riffle beetles, prior to testing, larvae were identified under a dissecting microscope to confirm genus, and their total body length was measured to the nearest mm using a 1-mm grid.

Test design

Test units:	Amphipods: beaker with 500 mL dosed water. Riffle beetles: glass 200-mL jelly jars.
Test medium:	Amphipods: moderately hard reconstituted water. Riffle beetles: stream water collected from the Cache River (IL, USA) at the same time as the beetle collection.
Replication:	Amphipods and riffle beetles: not stated
Individuals per replicate:	Amphipods: 10 amphipods Riffle beetles: not stated
Duration:	Amphipods: 96 hours Riffle beetles: 7 days
Exposure regime:	Amphipods: static Riffle beetles: a 100-mL water renewal of dosed water was conducted each day of the test.

Environmental conditions

Test temperature / relative humidity:	Amphipods: according to USEPA 600/R-99/064 Riffle beetles: 23 °C
Photoperiod:	Amphipods: according to USEPA 600/R-99/064 Riffle beetles: 18-h light and 6-h dark
Lighting:	Amphipods: according to USEPA 600/R-99/064 Riffle beetles: not stated
pH:	not stated
Feeding:	Amphipods: according to USEPA 600/R-99/064 Riffle beetles: larvae were not fed supplementary diets during the test.
Comments remarks:	Amphipods: a small piece of mesh was placed in each replicate for the water-only bioassays so that the *H. azteca* had a substrate to attach to within the beaker. Riffle beetles: stream water was filtered using qualitative P8 filter paper prior to use in the bioassays; approximately 5 to 6 aquarium pebbles were also added as a substrate to each replicate.

Analytical verification

Method:	No analytical verification carried out.

Assessments

Endpoints:	LC_{50}
Measurement frequency:	Amphipods: according to USEPA 600/R-99/064 Riffle beetles: not stated
Statistical analyses:	Single compound: PROC PROBIT with SAS® software
Analytical parameters:	mortality
Comments remarks:	For riffle beetles, the larvae were categorized as dead if they did not respond to prodding with forceps, did not move for at least 60 consecutive seconds, and were a transparent colour.

Results and Discussion

Effect characterization:

The amphipod toxicity test with clothianidin yielded an LC_{50} of 9.68 µg/L (95% confidence interval: 7.64-11.8 µg/L).

The riffle beetle toxicity test with clothianidin yielded an LC_{50} of 50.9 µg/L (95% confidence interval: 26.6-97.3 µg/L).

Uncertainties:

The bioassays conducted as part of the risk assessment were all based on mortality; therefore, it is unclear if the individual insecticides could potentially causing chronic effects, such as reproductive toxicity. As with all laboratory bioassays, some bias is present in testing species that in some cases are more or less sensitive than wild populations, but this is why the wild-caught riffle beetle larvae were also tested.

Validity Criteria

Not stated.

Conclusions

The amphipod (*Hyalella azteca*) 96h LC_{50} value was calculated to be 9.68 µg clothianidin/L. The riffle beetle (*Ancyronyx* spp.) 7d LC_{50} value was calculated to be 50.9 µg clothianidin/L.

(Whiting and Lydy, 2014)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		**Score**	**Evaluator's comments on criteria (optional)**
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	amphipods (*Hyalella azteca*), and Elmid riffle beetle larvae (*Ancyronyx* spp.)
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**7**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	Not stated.
10	Is information on the source/origin of the substance given?	0	Not stated.
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	0	E.g. source, purity, solubilty are not stated.
	Subtotal Test Substance	**0**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	Not relevant.
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	1	
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**4**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	

18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**19**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**Yes**	Minimal criteria not met
	C Evaluator's proposed Category:	**2**	
	D Justification in case evaluator deviates from B:	Insufficient information about test items.	
Date/period of evaluation:		15.07.2015	
Evaluator information			
Name		Sebastian Schadt	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 2	Reliable • 18 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Well-documented and scientifically sound study, however single information is missing (see below).
Limitations		Information about the administered concentrations are not clearly described and the source of test item, its purity neither any physico-chemical properties given.
GLP		No
Relevance	Supplemental data for acute toxicity to aquatic invertebrate.	Acute LC_{50} values are suitable for weight-of-evidence approach or to be used in clothianidin risk assessment to establish a species sensitivity distribution (SSD) or geometric mean calculation.

Ref ID: 317

	Submitted because data concern toxicity to aquatic invertebrates:
Report:	K-CA 8.2.4.2/07, Hayasaka D, Suzuki K, Nomura T, Nishiyama M, Nagai T, Sanchez-Bayo F, and K Goka (2013). Comparison of acute toxicity of two neonicotinoid insecticides, imidacloprid and clothianidin, to five cladoceran species. (Syngenta File No. CGA322704_10073), Published, Journal of Pesticide Science, Volume 38, Number 1/2, pp. 44-47.

Guidelines

OECD 202 (2004)

GLP: No. Published study (peer-reviewed article).

Executive Summary

The acute toxicity (48hr) of old (imidacloprid) and new (clothianidin) neonicotinoid insecticides to five cladoceran species and species sensitivity distribution (SSD) for cladocerans and other aquatic organisms to these insecticides are compared here. The sensitivities to both insecticides were in the following descending order: *Ceriodaphnia>Daphnia>Moina*. Differences in the 5% hazardous concentration (HC_5) threshold between the two species taxa to each compound indicated that clothianidin was 4 times less toxic than imidacloprid only to cladocerans.

Data on the non-relevant substances included in this study are not summarized here. Only data on clothianidin are included below.

Materials

Test material	Clothianidin
Source:	Kyoyu Agri Co., Ltd., Kanagawa, Japan.
Purity:	Dantosu® Flowable, clothianidin/water, and surfactant (20:80, v/v)
Water solubility:	Not stated
Comments remarks:	The wettable powder of clothianidin was used to make the insecticidal solutions.
Treatments	
Test concentrations:	No details, but 5 to 7 concentrations, and the ratio between them were 2.0.
Solvent:	None
Method of administration:	Five neonates, consisting of four replicates of each concentration, were introduced into each beaker containing 20 mL of the test solutions (see Hayasaka *et al.*) for details of the methodology for the bioassays).
Evidence for unsolved material:	Not stated
Test organism	
Species:	*Ceriodaphnia dubia*, *Ceriodaphnia reticulata*, *Daphnia magna*, *Daphnia pulex*, and *Moina macrocopa.*
Source:	National Institute for Environmental Studies (NIES), Tsukuba, Japan
Age/life stage at study initiation:	Female neonates (<24-hr old) from the second or later broods were used in all study.

Holding conditions prior to test:	Not stated
Acclimatisation prior to testing:	Not stated
Comments remarks:	The stock cultures of test organisms have been in NIES over three decades.

Test design

Test units:	Beaker containing 20 mL of the test solutions
Test medium:	Not stated, but for the controls, only dechlorinated tap water was used.
Replication:	4 replicates
Individuals per replicate:	5 neonates
Duration:	48 hours
Exposure regime:	Static

Environmental conditions

Medium temperature:	Not stated
pH:	At the beginning of the tests: 7.65 ± 0.24 At the end of the tests: 7.85 ± 0.04
Feeding:	Test individuals were not fed during the test period.
Other medium parameters:	Dissolved oxygen was 8.19 ± 0.40 to 7.65 ± 0.31 at the beginning and the end of the tests respectively.
Laboratory conditions:	Not stated

Analytical verification

Method:	No method for analytical verification reported.

Assessments

Endpoints:	EC_{50}, the relationship between toxicant sensitivity and body lengths, HC_5.
Measurement frequency:	Not stated
Statistical analyses:	The acute EC_{50} values were determined by the Probit method using the program EcoTox-Statics ver. 2.5. The relationship between toxicant sensitivity (48-hr EC_{50} values) and body lengths was analysed by Pearson's correlation coefficient. Differences in the susceptibility between test cladocerans and aquatic organisms other than cladocerans to clothianidin were compared using the 5% hazardous concentration (HC_5) values of both species groups.
Analytical parameters:	Immobilization and body length.
Comments remarks:	30-50 female neonates of each species were selected randomly, and their body lengths were measured using a dissecting microscope. The body length means from the crown of the head to the base of the tail spine. The HC_5 is calculated from the species sensitivity distribution (SSD) of each compound. The acute toxicity data (LC_{50} and EC_{50}) of clothianidin obtained for other aquatic organisms from other literatures comprise 3 crustaceans, *Procambarus clarkia*, *Hyalella azteca*, and *Paratya compressa improvisa*, 3 fish, *Cyprinus carpio*, *Lepomis macrochirus*, and *Oncorhynchus mykiss*, and 2 insects, *Cheumatopsyche brevilineatus* and *Chironomus riparius*.

Results and Discussion

Inter-species variation of acute toxicity:

The toxicity of clothianidin was in the following decreasing order: *C. dubia>C. reticulata>D. pulex>M. macrocopa>D. magna*. After Probit analyses, 48 hr EC_{50} values and 95% confidence limits of clothianidin for five test cladocerans were given in the table below. The inter-species variations of acute toxicity values were small, generally less than a factor of 2 to 3 (maximum 8).

EC_{50} values and 95% confidence limits of clothianidin for five test cladocerans

Species	EC_{50} value (µg/L)	95% confidence limit (µg/L)
Ceriodaphnia dubia	1 691.3	1 077.1 – 19 844
Ceriodaphnia reticulata	29 474	21 076 – 49 968
Daphnia magna	67 564	48 762 – 98 441
Daphnia pulex	31 448	20 881 – 46 463
Moina macrocopa	61 106	42 582 – 106 290

In this study, significant relationship between the EC_{50} values of each species for clothianidin and their body lengths was not found (r^2=0.593, p=0.129).

Differences in SSD between cladocerans and other aquatic organisms:

SSDs have been used to determine hazardous concentrations for the protection of ecosystems and are frequently used for the quantitative determination of the ecological risks of toxicants. The SSDs of clothianidin showed a clear difference between the five cladocerans tested here and other aquatic vertebrates and invertebrates (excluding cladocerans) reported in the existing literature. Although cladocerans are more sensitive to metal and organophosphate pesticides than other aquatic vertebrates and invertebrates, the opposite tendencies are found for clothianidin. The HC_5 values for clothianidin to the two taxa groups were 1929.72 µg/L and 0.34 µg/L, respectively, suggesting that freshwater aquatic taxa other than cladocerans were about 2,000 times more sensitive to clothianidin. The toxicity patterns of clothianidin for aquatic organisms, except cladocerans, might be the same as those of imidacloprid due to their identical mode of action.

Conclusions

The toxicity of clothianidin was in the following decreasing order (48 hr EC_{50} from 1691.3 to 67564 µg/L): *C. dubia>C. reticulata>D. pulex>M. macrocopa>D. magna*.

There are no significant relationship between the EC_{50} values of each species for clothianidin and their body lengths (r^2 = 0.593, p = 0.129).

The HC_5 values for clothianidin to the five cladocerans tested here and other aquatic vertebrates and invertebrates (excluding cladocerans) were 1929.72 µg/L and 0.34 µg/L, respectively. This is the opposite tendencies comparing with metal and organophosphate pesticides.

Clothianidin may be safe only to cladocerans, the evaluation of the ecotoxicological risks of clothianidin based on acute toxicity to cladocerans can systematically underestimate the risk of the pesticide to all other aquatic species and communities. The community effects of pesticides should not be linked to the lethal effects alone because sublethal responses of individual species and indirect effects on other species in the community also play a role.

(Hayasaka et al, 2013)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		**Score**	**Evaluator's comments on criteria (optional)**
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	*Ceriodaphnia dubia*, *C. reticulata*, *Daphnia magna*, *D. pulex*, and *Moina macrocopa*
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	Treatment concentrations are not explicitely reported but spacing factor is given and reference to OECD guideline methods.
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	
10	Is information on the source/origin of the substance given?	1	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	0	E.g. purity, solubility is not stated.
	Subtotal Test Substance	**1**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	0	Age/life stage not stated.
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**3**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	

	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**19**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	Minimal requirements given
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		14.07.2015	
Evaluator information			
Name		Stefan Kroder	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable • 18 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Acceptable publication, statistical and experimental design meets basic scientific principles.
Limitations		Age and life stage of test organisms as well as treatment levels are not explicitely reported but reference is given to standard OECD guideline methods and the used spacing factor for test concentrations.
GLP		No
Relevance	Additional laboratory acute toxicity data of aquatic invertebrates.	Acute EC_{50} values are suitable for establishing a species sensitivity distribution (SSD) or calculation of geometric mean at Tier 2 effect assessment for aquatic invertebrates.

Ref ID: n/a

	Submitted because data concern toxicity to aquatic invertebrates
Report:	K-CA 8.2.4.2/08, Riaz M.A., Chandor-Proust A., Dauphin-Villemant C., Poupardin R., Jones C.M., Strode C., Régent-Kloeckner M., David J.P., Reynaud S. (2013) Molecular mechanisms associated with increased tolerance to the neonicotinoid insecticide imidacloprid in the dengue vector *Aedes aegypti*, (Syngenta File No. CGA293343_11855) Published, Aquatic Toxicology, 126: 326-337 http://dx.doi.org/10.1016/j.aquatox.2012.09.010

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Mosquitoes are vectors of several major human diseases and their control is mainly based on the use of chemical insecticides. Resistance of mosquitoes to organochlorines, organophosphates, carbamates and pyrethroids led to a regain of interest for the use of neonicotinoid insecticides in vector control. The present study investigated the molecular basis of neonicotinoid resistance in the mosquito *Aedes aegypti*. A strain susceptible to insecticides was selected at the larval stage with imidacloprid. After eight generations of selection, larvae of the selected strain (Imida-R) showed a 5.4-fold increased tolerance to imidacloprid while adult tolerance level remained low. Imida-R larvae showed significant cross-tolerance to other neonicotinoids but not to pyrethroids, organophosphates and carbamates. Transcriptome profiling identified 344 and 108 genes differentially transcribed in larvae and adults of the Imida-R strain compared to the parental strain. Most of these genes encode detoxification enzymes, cuticle proteins, hexamerins as well as other proteins involved in cell metabolism. Among detoxification enzymes, cytochrome P450 monooxygenases (CYPs) and glucosyl/glucuronosyl transferases (UDPGTs) were over-represented. Bioassays with enzyme inhibitors and biochemical assays confirmed the contribution of P450s with an increased capacity of the Imida-R microsomes to metabolize imidacloprid in presence of NADPH. Comparison of substrate recognition sites and imidacloprid docking models of six CYP6s over-transcribed in the Imida-R strain together with *Bemisia tabaci* CYP6CM1vQ and *Drosophila melanogaster* CYP6G1, both able to metabolize imidacloprid, suggested that CYP6BB2 and CYP6N12 are good candidates for imidacloprid metabolism in *A. aegypti*. The present study revealed that imidacloprid tolerance in mosquitoes can arise after few generations of selection at the larval stage but does not lead to a significant tolerance of adults. As in other insects, P450-mediated insecticide metabolism appears to play a major role in imidacloprid tolerance in mosquitoes.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam are included below.

Materials

Test material	Thiamethoxam
Source:	Not stated
Purity:	Not stated

Treatments

Test concentrations:	Four doses, but no detail.
Method of administration:	Put the larval into 50 mL insecticide solution.

Test organism

Species:	Mosquito (*Aedes aegypti*)
Source:	Strain Bora-Bora (susceptible): French Polynesia Strain Imida-R: Bora-Bora larvae were selected with imidacloprid (Sigma-Aldrich, Germany) for several generations at the larval stage to obtain the Imida-R strain.
Age/life stage at study initiation:	9^{th} generations (G_9), fourth stage larvae.
Holding conditions prior to test:	Standard insectary conditions (26 °C, 14h/10h light/dark period, 80% relative humidity) in tap water (larvae) and net cages (adults). Larvae and adults were fed with hay pellets and papers impregnated with honey, respectively. Blood feeding of adult females was performed on mice.
Acclimatisation prior to testing:	Selection was performed by exposing third-fourth-stage larvae for 24 h to imidacloprid. The concentration of imidacloprid (500-900 µg/L) was adjusted at each generation to cause 70-80% mortality. Surviving larvae were transferred in tap water, fed with standard larval food and allowed to emerge. Adults were allowed to reproduce for 4 days and blood fed to obtain eggs for the next generation. In order to limit bottleneck effects, each generation was started with more than 7000 individuals before selection. Considering the high number of mosquitoes required for bioassays, biochemical assays and transcriptome profiling, these analyses were performed on individuals from the 9^{th}, 10^{th} and 11^{th} generations, respectively (G_9, G_{10}, G_{11}). In order to only consider constitutive tolerance mechanisms, individuals used for these analyses were not exposed to imidacloprid. The selection process was continued until G_{14}. Evolution of resistance of the Imida-R strain across three generations without insecticide selection was also monitored from G_{11} to G_{14}.

Test design

Test units:	Not stated, but containing 50 mL test solution
Test medium:	Not stated
Replication:	3 replicates
Individuals per replicate:	25 individuals
Duration:	24 hours
Exposure regime:	static

Environmental conditions	Not stated
Analytical verification	Not stated

Assessments

Endpoints:	Mortality
Measurement frequency:	After 24 h of exposure
Statistical analyses:	LC_{50} and 95% confidence intervals (CI 95%): Probit approach using XL-Stat (Addinsoft, Paris, France). TR_{50}: The LC_{50} values were compared between the two strains.

Results and Discussion

The 24 h LC_{50} values and 95% confidence intervals of thiamethoxam for each strain are given in the table below. The 24 h LC_{50} values of thiamethoxam on two strains (susceptible and Imida-R) of larvae of

mosquito (*Aedes aegypti*) were 183 µg/L and 806 µg/L, respectively. Imida-R larvae showed cross-tolerance to thiamethoxam (TR_{50} of 4.4).

Cross-tolerance of G_9 Imida-R larvae to thiamethoxam

Insecticide	Strain	LC_{50} (µg a.s./L) (CI 95%)	TR_{50} (CI 95%)
Thiamethoxam	Susceptible	183 (162-205)	-
	Imida-R	806 (701-910)	4.40 (3.42-5.62)

Conclusions

Thiamethoxam is moderately toxic to untreated larvae of mosquitos. The 24 h LC_{50} values of thiamethoxam on two strains (susceptible and Imida-R) of mosquito larvae (*Aedes aegypti*) were 183 µg a.s./L and 806 µg a.s./L, respectively. Imida-R larvae showed cross-tolerance to thiamethoxam (TR_{50} of 4.4).

(Riaz et al, 2013)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		Score	Evaluator's comments on criteria (optional)
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	no data given
10	Is information on the source/origin of the substance given?	0	no data available
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	
	Subtotal Test Substance	**1**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	

14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	1	
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**4**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**20**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	Minimal requirements given
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		04.08.2015	
Evaluator information			
Name		Lukas Jeker	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable • 20 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Well-documented and scientifically sound study
Limitations		Source and purity of the test substance is not given
GLP		No

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Relevance	Additional endpoint for acute toxicity to aquatic invertebrates	Acute toxicity of thiamethoxam to “susceptible” (=not conditioned to imidacloprid) aquatic larval stages of *Aedes aegypti* was investigated in laboratory bioassays. The endpoint expressed as LC_{50} is suitable for establishing species-sensitivity distributions (SSD) or calculating a geometric mean at tier 2 of the risk assessment on aquatic invertebrates.

Ref ID: 611

	Submitted because data concern toxicity to bees
Report:	K-CA 8.3.1.1/01. Laurino D, Manino A, Patetta A, Porporato M. (2013) Toxicity of neonicotinoid insecticides on different honey bee genotypes, (Syngenta File No. CGA293343_11856) Published, Bulletin of Insectology, 66 (1): 119-12621-8861

Guidelines

No standard guideline used.

GLP: No

Executive Summary

Toxicity effects of the neonicotinoid insecticides clothianidin, imidacloprid, and thiamethoxam were tested in the laboratory on different honey bee (*Apis mellifera* L.) genotypes belonging to the following subspecies: *Apis mellifera mellifera* L., *Apis mellifera ligustica* Spinola, and *Apis mellifera carnica* Pollmann. Oral and indirect contact trials were carried out on adult worker honey bees for each pesticide, using commercial formulations. The acute oral toxicity (AOT) LD_{50} and the acute indirect contact toxicity (ICT) LC_{50} were calculated.

Mean AOT LD_{50} values at 24 hours (clothianidin 3.53 ng/honey bee; imidacloprid 118.74 ng/honey bee; thiamethoxam 4.40 ng/honey bee), 48 hours (clothianidin 3.35 ng/honey bee; imidacloprid 90.09 ng/honey bee; thiamethoxam 4.27 ng/honey bee), and 72 hours (clothianidin 3.28 ng/honey bee; imidacloprid 69.68 ng/honey bee; thiamethoxam 4.16 ng/honey bee) from test start were of the same order of magnitude of those reported in the literature for all three neonicotinoids. Statistically significant differences emerged in a few instances between groups of honey bees coming from the different hives tested for clothianidin, between the groups of honey bees coming from the single *A. m. mellifera* hive and the four *A. m. ligustica* hives tested for imidacloprid, and more extensively between the two *A. m. carnica*, the single *A. m. mellifera*, and the six *A. m. ligustica* groups of honey bees tested for thiamethoxam. ICT LC_{50} values were obtained for a reduced number of hives: the single *A. m. mellifera* and two *A. m. ligustica* hives for clothianidin, the single *A. m. mellifera* and one *A. m. ligustica* hive for imidacloprid, the single *A. m. mellifera*, three *A. m. ligustica* hives, and one *A. m. carnica* hive for thiamethoxam. Nevertheless statistically significant differences were observed for clothianidin and thiamethoxam, but not for imidacloprid.

The results confirm that genetic differences in the response to pesticide toxic action exist in the honey bee, but they do not constitute the key factor involved in the uneven results observed in toxicity tests. In any case, the LD_{50} or other similar toxicity indexes should not be determined on a single colony.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam and clothianidin are included below.

Materials

Test material	Actara 25 WG (active substance: thiamethoxam) Dantop 50 WG (active substance: clothianidin),
Source:	Commercial product

Content:	Actara 25 WG: 25.0% thiamethoxam Dantop 50 WG: 50.0% clothianidin
Treatments	
Test concentrations:	Acute oral toxicity: Thiamethoxam: 0.02, 0.05, 0.1, 0.2, 0.5, 1 and 2 ppm Clothianidin: 0.015, 0.0375, 0.075, 0.15, 0.375, 0.75 and 1.5 ppm Indirect contact toxicity: Thiamethoxam: 2, 5, 10 and 20 ppm Clothianidin: 1.5, 3.75, 7.5 and 15 ppm
Solvent:	Not stated
Positive control:	None
Negative control:	1. Oral test: pure 25% sucrose solution 2. Contact test: leaves were sprayed with pure water.
Calibration of sprayer / Verification of application rates:	Not stated
Test organism	
Species:	*Apis mellifera mellifera, Apis mellifera ligustica, Apis mellifera carnica* (honey bee)
Source:	Not particularly stated, bees were taken from 9 hives/colonies.
Diet:	Bees were fed with sugar candy throughout the tests.
Comments remarks:	Hives/colonies were periodically checked for most common bee diseases. Morphometric analysis was used to assess the honey bee subspecies used in the trials.
Test design	
Test principle:	Acute oral and indirect contact toxicity
Test units:	Cages, not further described.
Treatment level(s):	Acute oral toxicity: seven and a control Indirect contract toxicity: four and a control
Method of administration:	1. Oral test: bees were fed to 25% sucrose solution with given amounts of a.s. 2. Contact test: chestnut (*Castanea sativa*) leaves were sprayed with suspensions of the product.
Number of replicates:	2 – 3containing 2 – 3 test cages
Individuals per replicate:	10
Study duration:	72 hours
Comments remarks:	Preliminary tests were carried out to determine the concentration range between 100% mortality and a mortality level not significantly different from that of the untreated control-
Environmental conditions	
Test units (type and size):	Cages, not further described.
Temperature:	28 - 30°C
Relative humidity	70%
Photoperiod:	Dark
Assessments	
Endpoints:	$LD_{50,}$ mortality (through oral and contact exposure)

Measurement frequency:	After 24, 48 and 72 hours
Statistical analyses:	LC_{50} values after 24, 48 and 72 hours were calculated using probit analysis. Since in oral toxicity tests each honey bee ingests on the average 35 µL of sucrose syrup during the allowed feeding period, the ingestion LD_{50} was obtained from the relative LC_{50}. Pairwise oral LD_{50} and contact LC_{50} ratios were calculated and their statistical significance determined, under the null hypothesis that they are identical, for each pairwise comparison the null hypothesis was checked at three probability levels ($p < 0.05$, $p < 0.01$, and $p < 0.001$). The Bonferroni correction was adopted as a safeguard against the risk deriving from performing multiple tests of statistical significance on the same data.
Comments remarks:	LD_{50} could not be calculated for the contact toxicity tests because the absorbed amount of the various a.s. cannot be determined. Oral toxicity test consumption was assumed rather than measured and therefore actual intake is not reported

Results and Discussion

Thiamethoxam:

The lowest LD_{50} after 72 hours for acute oral toxicity was 1.54 ng a.s./honey bee. *A. m. carnica* colonies yielded LD_{50} values significantly higher than those of *A. m. mellifera* and of most *A. m. ligustica.* All results are presented in table below.

Acute oral toxicity LD_{50} at 24, 48 and 72 hours of thiamethoxam to honey bee

Hive (species)	LD_{50} (ng a.s./honey bee)		
	24 hours	48 hours	72 hours
A. m. ligustica 1	4.32	3.90	3.59
A. m. ligustica 2	2.26	2.31	2.15
A. m. ligustica 3	5.01	5.08	4.52
A. m. ligustica 4	4.13	3.68	4.27
A. m. ligustica 5	2.48	2.44	2.44
A. m. ligustica 6	1.99	1.65	1.54
A. m. mellifera 1	3.40	3.40	3.36
A. m. carnica 1a	9.00	9.07	8.86
A. m. carnica 1b	5.73	5.56	5.46
A. m. carnica 2	5.71	5.64	5.36

Overall lowest LC_{50} after 72 hours for indirect contact toxicity was 2.75 ng a.s./honey bee. LC_{50} values were rather uneven in the tested colonies, but several statistically significant differences emerged, mainly at 72 hours. All results are presented in table below.

Indirect contact toxicity LC_{50} at 24, 48 and 72 hours of thiamethoxam to honey bee

Hive (species)	LC_{50} (ng a.s./honey bee)		
	24 hours	48 hours	72 hours
A. m. ligustica 1	5.36	3.53	2.75
A. m. ligustica 3	5.27	5.30	5.27
A. m. ligustica 4	6.03	4.61	4.17
A. m. mellifera 1	3.38	3.31	3.09
A. m. carnica 1	4.44	3.75	3.11

The overall lowest LD_{50} and LC_{50} for Thiamethoxam after 72 hours for acute oral and indirect contact toxicity was 1.54 and 2.75 ng a.s./honeybee, respectively.

Clothianidin:

The lowest LD_{50} after 72 hours for acute oral toxicity was 1.25 ng a.s./honey bee. Clothianidin LD_{50} values were rather uniform in the tested colonies, also when comparing results at 24, 48 and 72 hours; on the contrary, confidence intervals were narrower for some colonies and wider for others. Therefore the LC_{50} ratios are not statistically significant in most cases. All results are presented in table below.

Acute oral toxicity LD_{50} at 24, 48 and 72 hours of clothianidin to honey bee

Hive (species)	LD_{50} (ng a.s./honey bee)		
	24 hours	48 hours	72 hours
A. m. ligustica 1	1.24	1.11	1.25
A. m. ligustica 2	2.75	2.82	2.79
A. m. ligustica 3	5.37	5.07	4.83
A. m. ligustica 5	2.85	2.61	2.50
A. m. ligustica 6	2.20	2.91	2.16
A. m. mellifera 1	6.76	6.27	6.13

Overall lowest LC_{50} after 72 hours for indirect contact toxicity was 2.96 ng a.s./honey bee. LC_{50} values were rather uneven in the tested colonies, but statistically significant differences emerged, at 72 hours. All results are presented in table below.

Indirect contact toxicity LC_{50} at 24, 48 and 72 hours of clothianidin to honey bee

Hive (species)	LC_{50} (ng a.s./honey bee)		
	24 hours	48 hours	72 hours
A. m. ligustica 1	4.53	3.12	2.96
A. m. ligustica 3	4.71	4.64	4.29
A. m. mellifera 1	4.08	3.28	3.03

The overall lowest LD_{50} and LC_{50} after 72 hours for acute oral and indirect contact toxicity was 1.25 and 2.96 ng a.s./honeybee, respectively.

Validity Criteria

Not applicable, no guideline followed.

Conclusions

The results confirm that genetic differences in the response to pesticide toxic action exist in the honey bee, but they do not constitute the key factor involved in the uneven results observed in toxicity tests. In any case, the LD_{50} or other similar toxicity indexes should not be determined on a single colony.

(Laurino et al, 2013)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		Score	Evaluator's comments on criteria (optional)
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	1	
10	Is information on the source/origin of the substance given?	1	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	
	Subtotal Test Substance	**3**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	0	
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	0	
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	0	
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**1**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	

17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**19**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		03.06.2015	
Evaluator information			
Name:		Sebastian Schadt	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable. • 19 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Well-documented and scientifically sound study.
Limitations		No details reported about the test organisms' source of strain, sex, age or life stage. Exposure regime deviates from standard laboratory studies. In the oral test consumption was not quantified. The contact test was performed in a fashion of test procedures for other non-target arthropods but not as for bees (dorsal application).
GLP		No
Relevance	Supplemental data for acute toxicity to adult honeybee.	Acute oral and contact LD_{50} values are suitable for weight-of-evidence approach but rather not to be used in risk assessment due to the exposure regime used in toxicity tests.

Ref ID: 967

	Submitted because data concern toxicity to bees
Report:	K-CA 8.3.1.1/02. Laurino, D., Manino, A., Patetta, A., Ansaldi, M., Porporato, M. (2010) Acute oral toxicity of neonicotinoids on different honey bee strains, (Syngenta File No. CGA293343_11850) Published, REDIA, XCIII, 2010: 99-102

Guidelines

No standard guideline used.

GLP: No

Executive Summary

Neonicotinoids are a class of relatively new insecticides, designed in the `80s, characterized by their excellent feedback, since they are highly systemic and with long-term persistence. Theses insecticides, however, show very strong toxicity to pollinating insects and in particular to the honey bee, besides causing also other effects which are often not easily identifiable, such as behavioural disturbance, orientation difficulties, and impairment of social activities. During the past few years, in many countries, alarming bee mortality rates were recorded. In some cases this was clearly due to the use of neonicotinoids either for see dressing or crop spraying. It was therefore considered appropriate to test bon three strains of *Apis mellifera* of Piedmontese origin and referable to *A. m. ligustica* in the laboratory both the acute oral toxicity of imidacloprid, which had been, in use for many years, clothianidin and thiametoxam, only more recently introduced to Italy. To reach this aim, methods developed at the Di.Va. P.R.A. and applied in several previous studies, were used. Commercial products were used throughout the tests. They were tested at the recommended concentration for field treatments. When mortality was higher in the tested bees than in the untreated ones, the active substanceactive substances were tested at gradually decreasing concentrations so as to reach a mortality not significantly different from that found in the untreated controls. The LD_{50} calculated for clothianidin and thiametoxam were lower than for Imidacloprid; the differences between the honey bee strains for the same active substance were slight.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam and clothianidin are included below.

Materials

Test material:	Actara 25 WG (hydro dispersible granules) Dantop 50 WG (hydro dispersible granules)
Source:	Commercial product
Content:	Actara 25 WG: 25.0% thiamethoxam Dantop 50 WG: 50.0% clothianidin

Treatments

Test concentrations:	Thiamethoxam: 0.01, 0.02, 0.05, 0.1, 0.2, 0.5, 1 and 100 ppm Clothianidin: 0.0075, 0.015, 0.0375, 0.075, 0.15, 0.375, 0.75 and 75 ppm
Negative control:	Oral test: pure 25% sucrose solution

Test organism

Species:	*Apis mellifera ligustica* (honey bee)
Source:	Not particularly stated, bees were taken from 3 hives/colonies of the Piedmontese region, Italy.
Diet:	Throughout the tests bees were fed with sugar candy.
Comments remarks:	Hives/colonies were periodically checked for most common bee diseases.

Test design

Test principle:	Acute oral toxicity
Treatment level(s):	Eight and a control
Method of administration:	Bees were fed with 25% sucrose solution with given amounts of a.s..
Number of replicates:	Four
Individuals per replicate:	10
Study duration:	72 hours

Environmental test conditions

Test units (type and size):	Cages (20 x 20 x 30 cm)
Temperature:	28 - 30°C
Relative humidity	70%
Photoperiod:	Dark

Assessments

Endpoints:	Mortality (LD_{50}) and behaviour (e.g. shaking, tremors)
Measurement frequency:	Tests started at 12.00 h; mortality was checked at 13.00 h, 15.00 h, and 18.00 h on the first day of the trial and at 9.00 h, 12.00 h, 15.00 h, and 18.00 h during the following days.
Statistical analyses:	Acute oral LC_{50} were calculated by means of probit analysis. Considering that each honey bee ingested 35 µg of sucrose syrup during the allowed one hour feeding period, the acute LD_{50} was obtained from the relative LC_{50}.

Results and Discussion

Thiamethoxam:

Honeybees exposed to thiamethoxam showed obvious symptoms of poisoning, such as shaking and tremors, uncoordinated and uncontrolled movements, inability to take up a correct position of the body, and prolonged frenetic movement of the legs and rotation when being in the supine position. Direct observation of the behaviour of the honey bees in cages proved that it was transient at a lower concentration. Moreover, the highest concentration caused extensive vomiting by honey bees.

Thiamethoxam caused the death of all tested honey bees even at the concentration of 0.5 ppm, within 6 hours after the beginning of the test. This product caused a statistically significant mortality up to 0.05 ppm in two strains and up to 0.02 ppm in the third. All results are presented in table below.

Acute oral toxicity LD_{50} at 24, 48 and 72 hours of thiamethoxam to honey bee

Hive	LD_{50} (ng a.s./honey bee)

	24 hours	48 hours	72 hours
Beehive 1	2.761	2.644	2.556
Beehive 2	3.336	3.018	2.936
Beehive 3	4.546	4.383	3.151

The overall lowest LD_{50} after 72 hours for acute oral toxicity of thiamethoxam was 2.556 ng a.s./honeybee.

Clothianidin:

Clothianidin caused the death of all tested honey bees within 3 hours from the start of the trial at the field concentration of 75 ppm, and was still toxic within 72 hours at the concentration of 0.015 ppm, 5000 times lower. Mortality at the concentration of 0.75 ppm 1 hour after the beginning of the test was similar to that at the concentration of 75 ppm. This phenomenon was consistent for all three colonies. All results are presented in table below.

Acute oral toxicity LD_{50} at 24, 48 and 72 hours of clothianidin to honey bee

Hive	LD_{50} (ng a.s./honey bee)		
	24 hours	48 hours	72 hours
Beehive 1	4.930	4.671	4.514
Beehive 2	3.885	3.789	3.747
Beehive 3	4.627	4.507	4.369

The overall lowest LD_{50} after 72 hours for acute oral toxicity of clothianidin was 3.747 ng a.s./honeybee.

Validity Criteria

Not applicable, no guideline followed.

Conclusions

The LD_{50} calculated for clothianidin and thiametoxam revealed only slight differences between the honey bee strains for the same active substance.

(Laurino et al, 2010)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		Score	Evaluator's comments on criteria (optional)
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	3 including thiametoxam
2	Is the species given?	1	*Apis mellifera ligustica*
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	

6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	1	
10	Is information on the source/origin of the substance given?	1	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	
	Subtotal Test Substance	**3**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	0	Assumably worker bees used for tests but not clearly stated
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	0	No informations given
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	0	Age of bees necessary but not reported
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**1**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	

	Total Score	**19**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	Minimal requirements given
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		03.06.2015	
Evaluator information			
Name		Sebastian Schadt	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable • 19 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Study with acceptable experimental and statistical approach meeting basic scientific principles
Limitations		Lack of information about bees used for the toxicity tests, i.e. missing details for source of strains, age of individuals and sex of individuals (even though use of worker bees can be assumed). These informations are particularly essential as the study is aimed at the comparison among different honeybee strains. No record of consumption in oral tests.
GLP		No
Relevance	Supplemental data for acute oral toxicity to adult honeybee.	Acute oral LD_{50} values are suitable as supportive data in a weight-of-evidence approach but rather not to be used as endpoint in the risk assessment.

Ref ID: 183

	Submitted because data concern toxicity to bees
Report:	K-CA 8.3.1.4/03. Aliouane, Y.; El Hassani, A. K.; Gray, V.; Armengaud, C.; Lambin, M.; Gauthier, M.; (2009) Subchronic exposure of honeybees to sublethal doses of pesticides: effects on behaviour, (Syngenta File No. CGA293343_11851) Published, Environmental Toxicology and Chemistry, Vol. 28, Issue 1, pp. 113-122.

Guidelines

No standard guideline used.

GLP No. Published study (peer-reviewed article).

Executive Summary

Laboratory bioassays were conducted to evaluate the effects on honeybee behavior of sublethal doses of insecticides chronically administered orally or by contact. Emergent honeybees received a daily dose of insecticide ranging from one-fifth to one-five-hundredth of the median lethal dose (LD_{50}) during 11 d. After exposure to fipronil (0.1 and 0.01 ng/bee), acetamiprid (1 and 0.1 µg/bee), or thiamethoxam (1 and 0.1 ng/bee), behavioral functions of honeybees were tested on day 12.

Fipronil, used at the dose of 0.1 ng/bee, induced mortality of all honeybees after one week of treatment. As a result of contact treatment at 0.01 ng/bee, honeybees spent significantly more time immobile in an open-field apparatus and ingested significantly more water. In the olfactory conditioning paradigm, fipronil-treated honeybees failed to discriminate between a known and an unknown odorant. Thiamethoxam by contact induced either a significant decrease of olfactory memory 24 h after learning at 0.1 ng/bee or a significant impairment of learning performance with no effect on memory at 1 ng/bee. Responsiveness to antennal sucrose stimulation was significantly decreased for high sucrose concentrations in honeybees treated orally with thiamethoxam (1 ng/bee). The only significant effect of acetamiprid (administered orally, 0.1 µg /bee) was an increase in responsiveness to water.

The neonicotinoids acetamiprid and thiamethoxam tested at the highest dose (one-tenth and one-fifth of their oral LD_{50}, respectively) and fipronil at one-fivehundredth of LD_{50} have limited effects on the motor, sensory, and cognitive functions of the honeybee. Our data on the intrinsic toxicity of the compounds after chronic exposure have to be taken into account for evaluation of risk to honeybees in field conditions.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam is included below.

Materials

Test material: Thiamethoxam

Source: Cluzeau Info Lab, Sainte-Foy-La-Grande, France.

Purity: 97%

Treatments

Test concentrations: 0.1 and 1 ng/bee

Solvent (source):	Acetonitrile (Sigma Aldrich, France)
Negative control:	1. Topical application: Control animals received 1 µl of water containing the solvent (10% v/v). 2. Oral application: Control animals were fed with 33 µl/bee/day of sucrose solution containing the solvent (0.3% v/v).
Calibration of sprayer / Verification of application rates:	Not stated.

Test organism

Species:	*Apis mellifera* (honey bee)
Source:	Not stated.
Life stage of test organisms at study initiation:	Newly emerged bees collected from cells of brood frame.
Holding conditions:	Bees were collected from hives and placed in controlled room temperature (23°C). Bees were caged in groups of 40 individuals and maintained in darkness under controlled conditions (40% relative humidity, temperature 33°C). Pollen and sucrose solution (50% w/v) were provided ad libitum.
Diet during exposure period:	Bees were fed with sucrose solution (50% w/v) and water. Feeders were changed daily with fresh solutions.

Test design

Test principle:	Lethal und sublethal effects
Test medium:	1. Topical application: water containing the solvent (10% v/v) 2. Oral application: sucrose solution containing the solvent (0.3% v/v).
Treatment level(s):	Two and a control
Method of administration:	1. Topical application: Stock solutions were dissolved in water and 1 µl of the final solution was deposited onto the thorax of the honeybee. Control animals received 1 µl of water containing the solvent (10% v/v). 2. Oral application: Stock solutions were dissolved in sucrose solution (50%, w/v) that was used to feed honeybees with 33 µl/bee/day. Control animals were fed with pure sucrose solution containing the solvent (0.3% v/v).
Number of replicates:	None.
Individuals per replicate:	22 – 65 for each experimental design.
Study duration:	11 day of exposure followed by the experiments
Comments remarks:	Locomotor activity: Honeybees were tested in an open-field-like apparatus (30 x 30 x 4 cm) standing vertically and illuminated from above.

Environmental test conditions

Test units (type and size):	1. Locomotor activity: Individuals were introduced into a 5 mL syringe 2. Sucrose and water sensitivity: Individuals were fixed in a small tube by depositing a drop of wax-colophony mixture onto the dorsal part of the thorax. 3. Olfactory learning: see sucrose sensitivity
Temperature :	Not stated.
Relative humidity:	Not stated.
Photoperiod:	Not stated.

Assessments

Endpoints:	Mortality, locomotor activity, sucrose and water sensitivity, olfactory learning
Measurement frequency:	1. Mortality: Throughout exposure period mortality was observed daily. 2. Locomotor activity: 3 min observation period. Position was of the honey bee was recorded every 3 s on the screen of a computer using specially adapted softwatre. 2. Sucrose sensitivity: 60 min after treatment honeybee` antennae were stimulated every 3 min with sucrose solution. 3. Water sensitivity: 60 min after treatment and 3 hours after the first test. 4. Olfactory learning: 3 h prior to olfactory conditioning. Classical olfactory conditioning was carried out. A five-trial paradigm with an intertrial interval of 1 min, which leads to long-term memory, was used. Memory tests were performed 1, 24 and 48 hours after the training phase.
Statistical analyses:	Comparison of the mortality curves between the control and treated groups was performed with the Kaplan–Meier test. Locomotor activity was evaluated through analysis of three relevant parameters: the path length during the 3-min observation period, the duration of immobility, and the time spent in each level of the box. Student's t tests were performed for mean comparison between treated and control group values after variance comparison with Levene's test. The daily values of consumed water were compared between the treated and the control groups with a Student's t test. Responsiveness to water was compared between control and treated groups at 1 and 3 h with a chi-square test. The comparisons between the groups for sucrose responsiveness were conducted using Fisher's exact test, which directly yields a p value. For olfactory learning, the values were compared between control and treated groups for acquisition (from the second to the fifth trial) and for each retention test (at 1, 24, and 48 h) using Fisher's exact test. Within-group comparison for level response to conditioned odorant versus new odorant was performed using McNemar's test [29]. For each of these tests, a p value of less than 0.050 was considered significant. All the statistical tests were performed with SPSS®12 software (SPSS Science, Chicago, IL, USA).

Results and Discussion

Mortality:

At the end of the exposure period with thiamethoxam (11th day), the percentage of mortality in bees treated orally and topically with 1 ng was 10% (data not shown). The mortality in these groups was not different from that of controls (4 and 10% respectively). A maximum of 20% of dead bees was observed in the groups exposed orally and topically to 0.1 ng thiamethoxam. The same mortality level was observed in control animals (15% for oral and 20% for topic exposure).

Locomotor activity:

Thiamethoxam had no significant effect on the three parameters of locomotor activity compared to control, regardless of dose (0.1 and 1 ng/bee for thiamethoxam) or exposure route (oral delivery or topical application). In all cases, honeybees seemed less active than controls in the box and spent less time in the sixth upper level of the box and more time in levels 1 and 2. The time spent in immobility was increased, and the distance covered was reduced in treated honeybees. None of these differences were significant. We never observed honeybees trembling on the floor, fallen backward, or displaying abnormal movements of wings, legs, or body.

Sucrose sensitivity:

Oral thiamethoxam (1 ng/bee) induced a decrease of honeybees' sucrose responsiveness to 3 and 10% sucrose concentrations (Fisher's exact test, p = 001 and 0.008, respectively). Oral exposure to a dose of 0.1 ng/bee and contact exposure to thiamethoxam (0.1 and 1 ng/bee) had no effect on sucrose responsiveness (for contact 1 ng/bee).

Water sensitivity:

Thiamethoxam induced no effect on water consumption and responsiveness.

Olfactory learning:

Comparison of performance in the control group between the 1-, 24-, and 48-h memory tests revealed a decay of memory with time. Oral treatment of thiamethoxam (0.1 and 1 ng) induced a slight and nonsignificant decrease of performance during learning. Only with topical application did we observe a significant decrease of learning performance. For a dose of 0.1 ng/bee, the learning of topically treated animals was not different from the control. The memory test performed 24 h after learning showed a significant decrease in performance in the treated group compared to controls (Fisher's exact test, p = 0.020), but at 48 h, there were no more differences between the two groups. At the dose of 1 ng/bee, topical application of thiamethoxam induced a significant decrease in learning performance for the third and fourth trials (Fisher's exact tests, p = 0.025 and 0.033, respectively). At the end of the learning session, control bees reached 70% response rate, whereas thiamethoxam-treated bees reached only 50% response rate. Consequently, memory performance of the latter was lower than that of controls at 1, 24, and 48 h, but the difference was not significant.

Validity Criteria

Not applicable, no guideline followed.

Conclusions

Thiamethoxam by contact induced either a significant decrease of olfactory memory 24 h after learning at 0.1 ng/bee or a significant impairment of learning performance with no effect on memory at 1 ng/bee. Responsiveness to antennal sucrose stimulation was significantly decreased for high sucrose concentrations in honeybees treated orally with thiamethoxam (1 ng/bee). The neonicotinoid thiamethoxam tested at the highest dose (one-fifth of theoral LD_{50}) has limited effects on the motor, sensory, and cognitive functions of the honeybee.

(Aliouane Y et al, 2009)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		**Score**	**Evaluator's comments on criteria (optional)**
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	*Apis mellifera*
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	1	
10	Is information on the source/origin of the substance given?	1	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	
	Subtotal Test Substance	**3**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	0	Only honey bee was mentioned. However, study was performed with female workers
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	1	
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	

	Subtotal Test Organisms	**3**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**21**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		04.06.2015	
Evaluator information			
Name		Lukas Jeker	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable • 21 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Well-documented and scientifically sound study.
Limitations		The sex of the bees was not clearly defined, only honey bee was mentioned. However, study was apparently performed with female workers.
GLP		No
Relevance	Supplemental data for sublethal effects on behaviour of	Behavioral effects after sub chronic contact and oral exposure to sub lethal doses tested on locomotive activity in an open field-like apparatus might be used as NOEC values for sublethal

Qualitative assessment		
Study assessment	**Score**	**Rationale**
	honeybees	effects. There is no link between proboscis extension responsiveness and bee behaviour which can be used in risk assessment.

Ref ID: 31

	Submitted because data concern toxicity to bees
Report:	K-CA 8.3.1.3/07. Sandrock, Ch.; Tanadini, M.; Tanadini, L.G.; Fauser-Misslin, A.; Potts, S.G.; Neumann, P. (2014), Impact of Chronic Neonicotinoid Exposure on Honeybee Colony Performance and Queen Supersedure, (Syngenta File No. CGA293343_11853) Published, PLOS ONE, Vol. 9, Issue 8, DOI No. 10.1371/journal.pone.0103592

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Background: Honeybees provide economically and ecologically vital pollination services to crops and wild plants. During the last decade elevated colony losses have been documented in Europe and North America. Despite growing consensus on the involvement of multiple causal factors, the underlying interactions impacting on honeybee health and colony failure are not fully resolved. Parasites and pathogens are among the main candidates, but sublethal exposure to widespread agricultural pesticides may also affect bees.

Methodology/Principal Findings: To investigate effects of sublethal dietary neonicotinoid exposure on honeybee colony performance, a fully crossed experimental design was implemented using 24 colonies, including sister-queens from two different strains, and experimental in-hive pollen feeding with or without environmentally relevant concentrations of thiamethoxam and clothianidin. Honeybee colonies chronically exposed to both neonicotinoids over two brood cycles exhibited decreased performance in the short-term resulting in declining numbers of adult bees (-28%) and brood (-13%), as well as a reduction in honey production (-29%) and pollen collections (-19%), but colonies recovered in the medium-term and overwintered successfully. However, significantly decelerated growth of neonicotinoid-exposed colonies during the following spring was associated with queen failure, revealing previously undocumented long-term impacts of neonicotinoids: queen supersedure was observed for 60% of the neonicotinoid-exposed colonies within a one year period, but not for control colonies. Linked to this, neonicotinoid exposure was significantly associated with a reduced propensity to swarm during the next spring. Both short-term and long-term effects of neonicotinoids on colony performance were significantly influenced by the honeybees' genetic background.

Conclusions/Significance: Sublethal neonicotinoid exposure did not provoke increased winter losses. Yet, significant detrimental short and long-term impacts on colony performance and queen fate suggest that neonicotinoids may contribute to colony weakening in a complex manner.

Materials

Test material	Thiamethoxam and clothianidin
Source:	PESTANAL, Fluka
Purity:	99.7% and 99.9% for thiamethoxam and clothianidin, respectively

Storage conditions:	Dissolved in distilled water (1 mg/L) and then stored at room temperature and protected from light.
Treatments	
Test concentrations:	Target concentrations were 5.0 and 2.0 ppb for thiamethoxam and clothianidin, respectively.
Solvent:	For stock solution: distilled water For test solution: the stock solution was mixed with pollen patties (consisted of 55% honeybee pollen, 5% brewer's yeast and approximately 40% sucrose).
Positive control:	Plain pollen patties
Method of administration:	Two test materials were applied in combination. In total, 20 mixtures of plain and neonicotinoid-spiked pollen were prepared and fed batch-wise over time. Three times per week (each Monday, Wednesday and Friday) all colonies were provided with two 200 g pollen patties loosely packed in cellophane paper and placed between the two lower hive bodies. One group received plain pollen, while the other received patties containing thiamethoxam and clothianidin. Chronic neonicotinoid exposure through in-hive pollen feeding was performed for 46 days (1.5 months) in order to cover two brood cycles.
Verification of test concentrations:	LC/MS-MS
Test organism	
Species:	*A. m. carnica, A. m. mellifera*
Source:	One group of 14 queens from a region in eastern Germany, and one group of 10 queens from an alpine region in central Switzerland.
Age/life stage at study initiation:	Honeybee colonies established using artificial swarms
Test area	
Location:	The apiary was established on the land of the research station Agroscope Reckenholz-Tänniken in a rural area near the city of Zurich, Switzerland (47u25938N 8u31911E).
Field history:	Newly built for this study
Test design	
Plot size:	Two groups of each 12 hives were placed in a single row. Hives within groups were separated by 1 m, and both groups of hives were separated by approximately 20 m distance.
Test medium:	A apiary
Replication:	Two groups of each 12 hives.
Duration:	46 days to cover two brood cycles
Exposure regime:	Semi-field
Environmental conditions	No information about environmental conditions reported.
Assessments	
Endpoints:	Numbers of adult honeybees, pupae and eggs and larvae
Sampling technique:	Colony strength: visually estimation The contents of all pollen traps: collection
Sampling frequency:	The contents of all pollen traps: each time when new pollen patties were provided, i.e. three times per week.

	The first colony assessment: 1st : spring 2011, three days before the experiment treatment initiated 2nd for short-term effects: summer 2011, two days after the last pollen patties were fed 3rd for medium-term effects: autumn 2011, 3.5 months after the exposure 4th for long-term effects: spring 2012, one year after the treatment
Statistical analyses:	Linear regression performed using R.
Analytical parameters:	Explanatory variables includes treatment (control and neonicotinoids), honeybee strain (A and B), and assessment date (spring 2011, summer 2011, autumn 2011 and spring 2012) as fixed effects, and colony as a random effect. Residual analysis of all response variables indicated the need for variance stabilization and variables were transformed accordingly.

Results and Discussion

Residue analyses

No unexpected additional exposure of thiamethoxam or clothianidin from outside.

No residues in the forager bee and pupae samples collected directly after the treatment and 3 weeks after the treatment.

Colony growth

The average amount of total brood had declined by 13% after the exposure. Average worker populations were 28% smaller in the neonicotinoid treatment compared to the control. The number of eggs and larvae also has a significant overall decrease. There was no significant effect on the amount of pupae.

After 3.5 months of the experimental pollen feeding, no effects of the exposure were detected.

After 1 year, the effect was stronger than directly after the exposure.

Model-based estimates of contrasts and corresponding significance levels are summarized in the table below.

Model-based estimates of contrasts and corresponding significance levels of the treatment effect (neonicotinoid versus control) and honeybee genetics (strain A vs. strain B).

	Adult bees			Eggs and larvae			Pupae		
	Summer 2011	**Autumn 2011**	**Spring 2012**	**Summer 2011**	**Autumn 2011**	**Spring 2012**	**Summer 2011**	**Autumn 2011**	**Spring 2012**
Neonicotinoids *vs* Control	-60.56***	0.73	-82.96 ***	-0.31*	-0.01	-0.49 ***	-4.36	1.84	-15.31**
Strain A *vs* strain B							5.31	2.65	7.91
Treatment within strain A	-14.07	-1.33	-28.59 *	-0.10	-0.03	-0.06			
Treatment within strain B	-46.49***	0.59	-54.37 ***	-0.21	-0.05	-0.42 ***			

Results are shown in the transformed scale for the three response variables adult bees, eggs and larvae and pupae assessed directly after the 1.5 months of treatment (Summer 2011), 3.5 months later (Autumn 2011) and 1 year later (Spring 2012). For adult bees and eggs and larvae (the models that included a significant threefold interaction between treatment, honeybee strain

and assessment date) contrasts for treatment effects were also computed within individual honeybee strains at each assessment date. P values are adjusted for multiple testing. ***P<0.001; **P<0.01; *P<0.05; · 0.05<P<0.1.

Honey production

There is an overall decrease of -1.0±3.8 kg per colony (increase of 1.8±2.1 kg on average for strain A, and decrease of -4.9±1.0 kg on average for strain B) on average during the exposure. Overall, the mean honey production over the entire season remained 29% lower in the neonicotinoid-exposed colonies (23.7±2.5 kg) compared to the control (33.4±5.1 kg).

Pollen consumption

No indication of influences.

Pollen collections

Colonies collected less pollen with mean pollen collections barely reaching more than 50% of the control group.

Supersedure of queens and tendency to swarm

6 out of 10 queens of the colonies were replaced within one year after treatment. The result remained significant when overall queen loss was assessed, i.e. also including the two colonies (one per treatment group) that lost their queen during winter (P= 0.02).

A negative association of neonicotinoid exposure and swarming events during spring following experimental treatment was found (P= 0.005): in the control group 9 out of 10 colonies swarmed until end of May 2012 (5 out of 6 colonies of strain A and all 4 colonies of strain B), while only 2 colonies (one of strain A and B each) of the group that was exposed to thiamethoxam and clothianidin in the previous season swarmed.

Conclusions

The results indicate that neonicotinoids negatively impact on honeybee colony performance after chronic sublethal exposure throughout two brood cycles.

(Sandrock C et al, 2014)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		Score	Evaluator's comments on criteria (optional)
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	thiamethoxam, clothianidin
2	Is the species given?	1	
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	

6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	1	
10	Is information on the source/origin of the substance given?	1	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	
	Subtotal Test Substance	**3**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	*A. m.carnica* / *A. m. mellifera*
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	1	honeybee colonies established using artificial swarms
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	
	Subtotal Test Organisms	**4**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	
	Subtotal Study design / Test method	**3**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**22**	

	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		28.05.2015	
Evaluator information			
Name		Lukas Jeker	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable • 22 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Well-documented and scientifically sound study.
Limitations		There are limitations to this study involving test design. Spiked pollen was provided as the source of exposure. Pollen traps were placed in the hives to prevent other pollen being collected. The pollen levels during the treatment period (when pollen traps in place) increased by 66% in the control but only by 20% in the treated hives. This suggests an effect on pollen storing, which was continued once the pollen traps were removed with normal pollen collection reduced by 50% (which could reflect lower brood levels). Pollen availability affects brood production (in the absence of sufficient pollen larvae are cannabalised) and brood presence affects pollen collection. Thus, there are knock-on effects if brood levels are reduced and colonies build up slower. The effects seen in this study may be a result of anti-feedant effects from the pollen used, rather than from the test item.
GLP		No
Relevance	Sub-lethal effect after chronic exposure on honeybee colony	Impact on honey bee colony performance and queen supersedure after chronic sub lethal exposure throughout two brood cycles. It is supposed, exposure through pollen has a stronger impact at honey bee colony level compared to nectar substitute feeding. The study results may only be considered in a weight-of-evidence approach of sublethal chronic risk to honeybee colonies due to the limitations identified in the study design.

Ref ID: 14

	Submitted because data concern toxicity to non-target arthropods other than bees
Report:	K-CA 8.3.2.2/02. Duso C, Ahmad S, Tirello P, Pozzebon A, Klaric V, Baldessari M, Malagnini V and Angeli G. (2014) The impact of insecticides applied in apple orchards on the predatory mite *Kampimodromus aberrans* (Acari: Phytoseiidae), (Syngenta File No. CGA293343_11874) Published, Experimental and Applied Acarology, 62:391–414. DOI 10.1007/s10493-013-9741-3

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Kampimodromus aberrans is an effective predatory mite in fruit orchards. The side-effects of insecticides on this species have been little studied. Field and laboratory experiments were conducted to evaluate the effects of insecticides on *K. aberrans*. Field experiments showed the detrimental effects of etofenprox, tau-fluvalinate and spinosad on predatory mites. Spider mite (*Panonychus ulmi*) populations reached higher densities on plots treated with etofenprox and tau-fluvalinate than in the other treatments. Single or multiple applications of neonicotinoids caused no detrimental effects on predatory mites. In the laboratory, spinosad and tau-fluvalinate caused 100 % mortality. Etofenprox caused a significant mortality and reduced fecundity. The remaining insecticides did not affect female survival except for imidacloprid. Thiamethoxam, clothianidin, thiacloprid, chlorpyrifos, lufenuron and methoxyfenozide were associated with a significant reduction in fecundity. No effect on fecundity was found for indoxacarb or acetamiprid. Escape rate of *K. aberrans* in laboratory was relatively high for etofenprox and spinosad, and to a lesser extent thiacloprid. The use of etofenprox, tau-fluvalinate and spinosad was detrimental for *K. aberrans* and the first two insecticides induced spider mite population increases. The remaining insecticides caused no negative effects on predatory mites in field trials. Some of them (reduced fecundity and repellence) should be considered with caution in integrated pest management programs.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam are included below.

Materials

Test material	Thiamethoxam: Actara®
Source:	Not stated
Purity:	Thiamethoxam: 25 WG
Storage conditions:	Not stated
Other specifications:	Not stated
Treatments	
Test concentrations:	Thiamethoxam: 30 g/hL
Solvent:	None
Method of administration:	Field study: applied according to codling moth control timing

	Laboratory study: apple leaves were treated with insecticides and then mated *K. aberrans* females were transferred onto the leaves to expose them to fresh insecticide residues. To prevent mite escape and leaf desiccation, leaves were placed onto wet cotton prior to mite transfer.
Verification of test concentrations:	Not stated
Test organism	
Species:	Predatory mite (*Kampimodromus aberrans*)
Source:	Not stated
Age/life stage at study initiation:	Not stated
Holding conditions prior to test:	Not stated
Acclimatisation prior to testing:	Not stated
Test area	
Location:	Trentino
Field history:	Golden Delicious
Pesticides used on fields:	Not stated
Test design	
Test units (laboratory study):	climatic chamber at 25 ± 2 °C, 70 ± 10 % relative humidity and 16L:8D photoperiod.
Plot size (field study):	Not stated
Replication:	Field study: 3 Laboratory study: 1
Individuals per replicate:	Field study: not stated Laboratory study: 45-50 females
Duration:	Field study: growing season Laboratory study: 7 days
Exposure regime:	Laboratory study: apple leaves were treated with insecticides and then mated *K. aberrans* females were transferred onto the leaves to expose them to fresh insecticide residues. To prevent mite escape and leaf desiccation, leaves were placed onto wet cotton prior to mite transfer.
Comments remarks:	In Field study, the process for leaves and mites: a total of 60 leaves per treatment (15 leaves per replicate) were removed and transferred to the laboratory where predatory and phytophagous mites, eventually present, were counted under a dissecting microscope. The phytoseiids were mounted on slides, in Hoyer's medium, and identified under a phase contrast microscope.
Environmental conditions	
Test temperature / relative humidity:	Field study: not recorded Laboratory study: 25 ± 2 °C
Photoperiod:	Field study: not stated Laboratory study: 16L:8D
Lighting:	Not stated
pH:	Not stated
Feeding:	Field study: not stated

	Laboratory study: Fresh *Typha sp*. pollen was provided every 2 days as food for predatory mites.
Analytical verification	Not stated
Assessments	
Endpoints:	Field study: the difference of population between the treatments Laboratory study: survival, fecundity, escape rate (number of escaped or drowned females/initial number of females) and egg hatching.
Measurement frequency:	Not stated
Statistical analyses:	Field study: the experiment was analysed using a REML repeated measures model where treatments, times and their interactions were considered as sources of variation and F tests were used to evaluate their effects (α=0.05). Degrees of freedom were estimated using the Kenward–Roger method. Mite densities were considered as dependent variables with repeated measurements at different times. Data were checked for normality assumption and thus the number of phytoseiids per leaf was log (x+1) transformed prior to the analyses. The SLICE option of the LSMEANS statement was used to test treatment effect variation during observation periods. The differences among treatments were evaluated with a t test with Bonferroni adjustment (α=0.05) to least square means, while for the second experiment pairwise comparison between treatments in the period after treatment were performed using contrasts (α=0.05). Laboratory study: one-way ANOVA with F test (P = 0.05) to evaluate the effect of insecticides on mite survival, fecundity, escape rate (number of escaped or drowned females/initial number of females) and egg hatching using GLM procedure of SAS. Treatments were compared using Tukey–Kramer test (P=0.05). In order to meet the ANOVA assumptions, data on survival were arcsin-transformed while square-root transformation was applied to data on fecundity. The formula was used for fecundity calculation. The overall toxicity of each insecticide was expressed as: E=100%-(100%-M)▪R, where E is the coefficient of toxicity; M is the corrected mortality according to Abbott; R is the ratio between the average number of hatched eggs produced by treated females and the average number of hatched eggs produced by females in the control group.
Analytical parameters:	Field study: population Laboratory study: survival, fecundity, escape rate (number of escaped or drowned females/initial number of females) and egg hatching.

Results and Discussion

Field studies:

A significant variation in *K. aberrans* numbers was found among treatments ($F_{18, 321} = 42.34$; $P < 0.0001$) and over time ($F_{9, 321} = 36.03$; $P < 0.0001$). There was also a significant interaction between treatments and time ($F_{162, 286} = 2.10$; $P < 0.0001$). No differences among treatments were found prior to the first insecticide application ($F_{18, 373} = 1.13$; $P = 0.32$) while significant differences emerged later (see table below). In particular, insecticide applications induced low numbers of predatory mites compared with the control, independently of application frequency (see table below).

Result of pairwise *t*-test on the least square means of *K. aberrans* population observed in different treatments during 2011

Active substance	Number of applications	Thiamethoxam		
		1	2	3

Control	***t***		5.22	4.91	5.27
	P		<0.0001	<0.0001	<0.0001
Thiamethoxam	**1**	***t***		-0.31	0.05
		P		0.7569	0.9567
	2	***t***			0.36
		P			0.716

df = 121.

There was no effect of treatments before the first insecticide application ($F_{18, 340}$=0.05; P=1) but later higher spider mite densities were associated with applications of tau-fluvalinate (see table below).

Result of pairwise *t*-test on the least square means of *Panonychus ulmi* population observed in different treatments during 2011

Active substance	**Number of applications**		**Thiamethoxam**		
			1	**2**	**3**
Control	***t***		2.03	0.58	0.40
	P		0.0468	0.5615	0.6935
Thiamethoxam	**1**	***t***		1.44	1.63
		P		0.1537	0.1077
	2	***t***			0.19
		P			0.8517

df = 63.9.

Laboratory studies:

Insecticides affected *K. aberrans* survival ($F_{11, 503}$ = 33.80; P <0.0001) and fecundity ($F_{9, 258}$ = 21.76; P <0.0001). Thiamethoxam did not affect female survival, where a survival of 81.7% was observed along with a low level of fecundity (see table below). Thiamethoxam was associated with a significant reduction in fecundity (see table below).

Escape rates of thiamethoxam were not significantly different compared with the control. Egg hatching rate ranged from 77 to 91% among treatments and was independent from applied insecticides ($F_{11, 345}$ = 0.49; P = 0.90).

Effects of insecticides on survival and fecundity of *K. aberrans*; the coefficient of toxicity is also reported

Treatments	**Corrected morality (%) M**	**Corrected fecundity R**	***E (%)**
Thiamethoxam	3.33	0.28	73.03

*E = 100 % - (100 % - M) R

Conclusions

Thiamethoxam caused no negative effects on predatory mites in field trials.

In laboratory studies, thiamethoxam did not affect female survival, but it was associated with a significant reduction in fecundity.

The present study shed light on lethal and sub-lethal effects of pesticides frequently used in orchards. For most pesticides results were consistent with those of field trials available in the literature. This suggests

that laboratory studies with *K. aberrans* can predict most effects induced by field applications of pesticides on this beneficial.

(Duso et al, 2014)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		Score	Evaluator's comments on criteria (optional)
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	0	Time-points of observations for field study 2011 not sufficiently stated
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	0	Replicates for the laboratory study is not given (but for field study)
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	0	It is not appropriate to use maximum field rates for laboratory experiments
	Subtotal Test Substance	**5**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	
10	Is information on the source/origin of the substance given?	0	
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	not relevant
	Subtotal Test Substance	**1**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	reported for laboratory study, not relevant in field study
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	0	not stated for laboratory study, too few information given in field study
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	0	not reported
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	not relevant
	Subtotal Test Organisms	**2**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	apple plants and leaves
17	Are sufficient details of the administration scheme given to judge the study?	0	no details about equipment and way of application

18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	1	not relevant
	Subtotal Study design / Test method	**2**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	
20	Is the description of the study results for all endpoints investigated transparent and complete?	0	Results for field experiments are insufficient, only statistical output of pairwise comparisons given
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**2**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**13**	
	A Numerical result leads to initial Category:	**3**	
	B Cut-off criteria restricts Category:	**Yes**	Minimal criteria not met
	C Evaluator's proposed Category:	**3**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		28.07.2015	
Evaluator information			
Name		Stefan Kroder	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 3	Not reliable. • 13 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) as well as cut-off criteria were not met for both field and laboratory experiments. • Study design is not appropriate and reported information is insufficient for reliable conclusions.
Limitations		For laboratory experiments, replicates and number of individuals not given. The study design is not appropriate as maximum field rates of insecticides were used and no endpoint for risk assessment can be derived. For field experiments time-points of observations are not sufficiently described, too few information is given regarding the predatory mite population and results are not transparent neither completely reported.
GLP		No
Relevance	Not relevant	Results do not give endpoints (ie. NOEC or ERx) for use in risk assessment.

Ref ID: 1407

	Submitted because data concern toxicity to non-target arthropods other than bees
Report:	K-CA 8.3.2.2/03. Tosi L, Farinazzo E, Posenato G, Girolami V. (2006) Side effects of insecticides on *Kampimodromus aberrans*, (Syngenta File No. CGA293343_11846) Published, L'Informatore Agrario, 26: 54-56.

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

Experiments in 2004 and 2005 investigated the effects of thiamethoxam, chlorpyrifos, spinosad, etofenprox, methoxyfenozide and abamectin on *Kampimodromus aberrans*, a species useful in maintaining a biological balance in north eastern Italian vineyards. Methoxyfenozide and thiametoxam had no significant effects on *K. aberrans*; nor did abamectin once the period immediately following application had passed. Chlorpyrifos and spinosad had some adverse effect but biological control was maintained. Etofenprox suppressed *K. aberrans* for a considerable period and has been dropped from further experiments.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam is included below.

Materials

Test material	Thiamethoxam
Source:	Actara 25 WG, Syngenta
Purity:	25% as water dispersible granules
Treatments	
Test concentrations:	20 g/hL, 25% a.s.
Method of administration:	A FOX 320 motorised pump, 1200 L/ha water, applied on 6 July 2005
Verification of test concentrations:	None
Test organism	
Species:	*K. aberrans*
Source:	There has been a population in the test area for many years.
Age/life stage at study initiation:	No information.
Test area	
Location:	In a vineyard (with spacing of 4.5 x 1.1m) located in the municipality of Montecchia di Crosara, in the eastern part of the province of Verona, in the Soave DOC region.
Field history:	The vineyard contains Garganega vines trained on Veronese pergolas, planted in 1970.

Pesticides used on fields: No information.

Test design

Plot size: Each plot was made up of seven vines.

Replication: Four repetitions per treatment group

Duration: 60 days

Environmental conditions No information

Assessments

Endpoints: Number of juveniles, adults and mobile forms of *K. aberrans* on the leaves

Sampling technique: At each sampling, ten leaves were collected per repetition (40 per group), from the five central vines, at the midway point between two central branches.

Sampling frequency: Before treatment and then 5, 10, 20, 40 and 60 days after treatment

Statistical analyses: The data collected and stored using CAMPO-lab software were subjected to analysis of variance (ANOVA) and compared using Tukey's test, after square-root transformation as necessary.

Results and Discussion

The test results are shown in the table below. Five days after treatment – sampling performed on 6 July – a smaller number of phytoseiids was observed in the treatment group than in the control. At T+10, the *K. aberrans* population did not differ from the control. The same trend was also observed at +20 and +40 days from treatment. At T+60, the groups showed substantially equal values.

The situation for juvenile phytoseiids appeared to be more complex, partly due to their low population.

Mean number of juveniles, adults and mobile forms of *K. aberrans* per leaf

Group	7 Jul (PreT)	11 Jul (T+5)	16 Jul (T+10)	28 Jul (T+20)	18 Aug (T+40)	5 Sep (T+60)
A - Juveniles						
Thiamethoxam	1.8	0.2 ab	0.2	0.5 a-A	0.0 bc	0.9
Control	2.2	1.0 a	0.2	0.2 ab-AB	0.6 abc	1.1
B - Adults						
Thiamethoxam	3.4	2.8 a-A	1.1 a	1.3 a-A	0.8	1.6
Control	4.9	4.3 a-A	1.1 a	1.0 a-AB	1.6	1.3
C - Mobile forms (juveniles + adults)						
Thiamethoxam	5.2	3.0 a-A	1.3 a-AB	1.8 a-A	0.9 ab	2.5
Control	7.1	5.3 a-A	1.3 a-AB	1.2 a-A	1.1 A	2.4

Different letters indicate significant differences, Tukey's test. Lower case letters $p < 0.05$, upper case letters $P < 0.01$.

Conclusions

In the thiamethoxam group, limited to 2005, the phytoseiid population remained consistently at levels similar to those observed for the control group.

(Tosi et al, 2006)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)

Criteria		Score	Evaluator's comments on criteria (optional)
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	
2	Is the species given?	1	
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	1	
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	
	Subtotal Test Substance	**8**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	Only nominal content reported
10	Is information on the source/origin of the substance given?	0	Only manufacturer stated
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	Not relevant
	Subtotal Test Substance	**1**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	Natural population at experimental field site
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	1	Population status presented in pre-treatment sample
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	Not relevant in field experiment
	Subtotal Test Organisms	**4**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	
17	Are sufficient details of the administration scheme given to judge the study?	1	
18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	0	No methods for verifying exposure levels or dispersion of spray solutions are presented.
	Subtotal Study design / Test method	**2**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	1	

20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**3**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**19**	
	A Numerical result leads to initial Category:	**1**	
	B Cut-off criteria restricts Category:	**No**	Minimal requirements given
	C Evaluator's proposed Category:	**1**	
	D Justification in case evaluator deviates from B:		
Date/period of evaluation:		10.08.2015	
Evaluator information			
Name		Stefan Kroder	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 1	Reliable. • 19 out of 22 reliability criteria are met according to ToxRTool (Schneider et al. 2009) • Acceptable, well documented study.
Limitations		There is a lack of information about source and purity of test items, only stating the nominal content of active substance and the commercial availability of plant protection products. Exposure levels were not verified neither dispersion of spray solution on investigated leaf areas.
GLP		No
Relevance	Additional field data on non-target arthropods (NTA)	The results from this field study about side effects of thiamethoxam on predatory mite populations in vineyards are suitable as supporting data in a weight of evidence for the risk assessment.

Ref ID: 1769

	Submitted because data concern toxicity to non-target arthropods other than bees
Report:	K-CA 8.3.2.2/04. Beers EH, Brunner JF, Dunley JE, Doerr M, Granger K (2005) Role of neonicotinyl insecticides in Washington apple integrated pest management. Part II. Nontarget effects on integrated mite control. The publisher has been contacted to obtain copyright clearance, however Syngenta is unable to provide this reference at this time. Published, Journal of Insect Science, 5: 16-26.

Guidelines

No standard guideline used.

GLP: No. Published study (peer-reviewed article).

Executive Summary

The effect of neonicotinyl insecticides on integrated mite control in Washington apple was examined from 2000-2004. In a series of 20 field trials (54 treatments) designed primarily to look at efficacy against the codling moth, *Cydia pomonella*, nearly half of the treatments using four or more applications of acetamiprid had peak mite densities exceeding the economic threshold of 5 mites per leaf. Overall, acetamiprid treatments had 4.6-fold higher mite densities than the standard organophosphate insecticide treatment. Of the treatments with high mite populations, *Panonychus ulmi*, the European red mite, and *Tetranychus urticae*, the two-spotted spider mite, were the dominant species in roughly equal numbers of cases. Only 11.1% of the thiacloprid treatments exceeded 5 mites per leaf; these experimental treatments included eight applications, whereas the current label restricts the number of applications at the rate for *C. pomonella* to two applications. One out of six clothianidin treatments caused a significantly higher mite density than the standard treatment; however, this material appeared to suppress predatory mites. Neonicotinyl insecticides did not eliminate predatory mites, but they inhibited their ability to respond normally to increasing prey populations. In field trials designed specifically to examine mite population densities where neonicotinyl insecticides were used, significantly higher levels of tetranychid mites occurred in one or more acetamiprid treatments (one, two or four applications) in five out of six trials. In the sixth trial (in a commercial orchard), only two acetamiprid applications were made, and mite populations were low in all treatments. While elevated mite densities were more likely to occur with four applications, in one case it occurred following a single application. The predominant tetranychid mite species (either *P. ulmi* or *T. urticae*) varied from trial to trial; however, there was no apparent bias regarding stimulation of the two species. Horticultural mineral oil was used with acetamiprid in some trials in an attempt to mitigate mite outbreaks. However, the addition of oil did not counteract the tendency of acetamiprid to increase tetranychid mite populations, and in one trial, had a negative effect on predatory mite densities. Seasonal tetranychid mite density was positively related to the total grams a.s. (or number of applications) of acetamiprid, thus reducing the number of applications per season should lower the probability of mite outbreaks.

Data on the non-relevant substances included in this study are not summarized here. Only data on thiamethoxam are included below.

Materials

Test material	Thiamethoxam.
Source:	Thiamethoxam: Actara, Syngenta.
Purity:	Thiamethoxam: 25 WDG
Treatments	
Test concentrations:	Thiamethoxam: 96 g a.s./ha
Solvent:	Horticultural mineral oil
Standard:	Azinphosmethyl (Guthion, Bayer CropScience) Phosmet (Imidan, Gowan Co.)
Positive control:	Esfenvalerate (Asana, DuPont)
Method of administration:	Spray
Verification of test concentrations:	Not stated
Test organism	
Species:	Phytophagous tetranychids European red mite, *Panonychus ulmi* (Koch); twospotted spider mite, *Tetranychus urticae* Koch; McDaniel spider mite, *Tetranychus mcdanieli* McGregor; the predatory mites *Galandromus occidentalis* (Nesbitt) and *Zetzellia mali* Ewing; and the eriophyid apple rust mite, *Aculus schlechtendali* (Nalepa).
Source:	Natural field population
Age/life stage at study initiation:	Not stated
Test area	
Location:	TF, Tree Fruit Research and Extension Center (home farm); CV, WSU Columbia View Farm.
Field history:	Not stated
Pesticides used on fields:	Not stated
Test design:	Mite-specific field experiments
Plot size:	Not stated
Test medium:	Not stated
Replication:	4 replications
Individuals per replicate:	5 trees (single row)
Duration:	Years 2000 to 2004
Exposure regime:	Sprayed using an airblast sprayer (Rears Pak-Blast, Rears Mfg., Eugene, OR). Plots were either multiple trees in a single row, or multiple trees per row in three rows. Plots had buffer trees and rows in the experimental layout to ensure treatments did not contaminate neighboring plots. Spray applications were directed at the first and second generation of *C. pomonella*. The first application for each generation was based on a degree-day model. Subsequent applications for the generation were made at an interval of days based on the length of residual control being evaluated. Two applications per generation (four per season) were made. A neonicotinyl insecticide was substituted for one, two, or all four of the cover sprays, with phosmet used for the remaining cover sprays.
Environmental conditions	Not stated
Analytical verification	Not stated

Assessments

Endpoints: Mite populations and densities: mites that fell on a revolving glass plate were counted with a binocular microscope. In the mite-specific trials, the variable cumulative mite-days (CMD) was used to summarize the seasonal densities for these trials, using the following equation:

$$CMD=\Sigma 0.5(P_a+P_b)D_{a-b}$$

Where P_a is the population density (mean mites per leaf at time *a*), P_b is the population density at time *b*, and Da-b is the number of days between evaluations.

Sampling and measurement: Mite densities were assessed by randomly selecting 20 to 60 leaves per plot, with the exception of one of the commercial orchard trials, where 100- to 200-leaf samples were taken from the 2-ha plots, avoiding the plot borders. The major differences between the two groups of experiments was that in the mite-specific trials, phosmet was the standard organophosphate insecticide used (vs. azinphosmethyl), and mite samples were taken at 1- to 2-wk intervals (vs. 2 to 3 times per season).

Statistical analyses: Cumulative mite-days (CMD) Data were analyzed using analysis of variance (SAS Institute 1982) and the Waller-Duncan *k*-ratio *t*-test mean separation. Data were tested for homogeneity of variance using Levene's (1960) test. Data with unequal variances were transformed (log (*y*+0.5)) prior to analysis.

Results and Discussion

Mite densities were low in the control throughout the season, never exceeding 0.3 mites per leaf, which is typical of minimally sprayed apple orchards in Washington. The predatory mite populations in the thiamethoxam treatments were not different than the control. Apple rust mite densities were high, but with no consistent trend in differences among treatments. Shown on the table below.

Effect of thiamethoxam on phytophagous and predatory mites, Aug 2001

Test substance	g a.s./ha	CM cover sprays [X]	Cumulative mite days						
			Two-spotted spider mite [Y]	European red mite [Y]	Total tetranychid mites [Y]	*G. occidentalis* [Y]	*Z. mali*	Total predatory mites [Y]	Apple rust mite
Thiamethoxam 25 WDG	96	C1, C2, C3, C4	9 cd	18 bc	30 bc	55 ab	43 ab	99 a	18 008 c
Check	-	-	3 de	4 d	8 d	85 a	21 bcd	105 a	22 035 bc

Means within columns not followed by the same letter are significantly different (Waller-Duncan *K*-ratio *t*-test).
Experimental design: RCB, 4 replicates, 5 tree (single row); sampled 20 leaves/plot; airblast 1.871 liters/ha.
Tetranychid mite species composition (seasonal average): 59% ERM, 38% TSM, 3% MCD.
[a] Locations: TF, Tree Fruit Research and Extension Center (home farm); CV, WSU Columbia View Farm.
[b] Method: H, Handgun, A, Airblast (liters/ha).
[c] Species composition; figure in column is percentage European red mite, remainder was twospotted spider mite.
[d] Mean comparison statistically different at P=0.10 but not at P=0.05.
[e] Rates for the 1st and 2nd generation of codling moth, respectively.
[Y] Data transformed log (y+0.5) due to unequal variances.

Conclusions

The predatory mite populations in the thiamethoxam treatments were not different than the control.

(Beers et al, 2005)

Reliability Assessment of Ecotoxicity Studies Based on ToxRTool (Schneider et al., 2009)			
Criteria		**Score**	**Evaluator's comments on criteria (optional)**
No.	**Cut-off criteria**		
1	Was the test substance identified?	1	Description in materials and methods is confusing and doesn't match with result tables.
2	Is the species given?	1	
3	Is the administration route given?	1	
4	Are doses administered or concentrations in application media given?	1	
5	Are frequency and duration of exposure as well as time-points of observations explained?	0	Information in parts is insufficient, giving only vague information (e.g. 4 applications during season against 1st and 2nd codling moth generation).
6	Were negative (where required) and positive controls (where required) included (give point also, when absent but not required)?	1	
7	Is the number of replicates and/or organisms per group given?	1	
8	Is the study design chosen appropriate for obtaining the substance-specific data aimed at?	1	Study aims at the compatibility of insecticide use for codling moth control including predatory mite populations, and thus only indirectly aimed at risk to non-target organisms.
	Subtotal Test Substance	**7**	**If not 8, study is not reliable.**
	Criteria Group I: Test substance identification		
9	Is the purity of the substance given?	0	Only nominal content given.
10	Is information on the source/origin of the substance given?	0	Only producer given.
11	Is all information on the nature and/or physico-chemical properties of the test item given, which you deem indispensable for judging the data?	1	Not relevant
	Subtotal Test Substance	**1**	
	Criteria Group II: Test organism characterisation		
12	Is the sex or the sex ratio of the test organisms given?	1	Not relevant in field experiments
13	Is information given on the source or strain of test organisms plus, if considered necessary to judge the study, other specifications?	1	Natural populations at experimental field sites
14	Is age, life-stage, growth stage, body weight of the test organisms at the start of the study given?	0	No status reported and no pre-treatment sampling given.
15	For repeated dose toxicity studies only (give point for other study types): Is information given on the housing or feeding conditions?	1	Not relevant in field experiments
	Subtotal Test Organisms	**3**	
	Criteria Group III: Study design description		
16	Is the test media clearly described (water, soil, plant)?	1	Not relevant
17	Are sufficient details of the administration scheme given to judge the study?	0	Materials and methods are confusing referring to result tables but the tables do not seem to match with descriptions in material and methods.

18	For repeated dose toxicity studies only (give point for other study types): Were exposure concentrations analytically verified or was stability of the test substance otherwise ensured or made plausible?	0	Not reported
	Subtotal Study design / Test method	**1**	
	Criteria Group IV: Study results documentation		
19	Are the study endpoint(s) and their method(s) of determination clearly described?	0	Sampling methods only vaguely described
20	Is the description of the study results for all endpoints investigated transparent and complete?	1	
21	Are the statistical methods applied for data analysis given and applied in a transparent manner (give also point, if not necessary/applicable)?	1	
	Subtotal Study Result Documentation	**2**	
	Criteria Group V: Plausibility of study design and results		
22	Are the quantitative study results reliable?	1	
	Subtotal Plausibility	**1**	
	Total Score	**15**	
	A Numerical result leads to initial Category:	**2**	
	B Cut-off criteria restricts Category:	**Yes**	Minimal criteria not met
	C Evaluator's proposed Category:	**3**	
	D Justification in case evaluator deviates from B:	Exposure regime insufficiently described	
Date/period of evaluation:		10.08.2015	
Evaluator information			
Name		Stefan Kroder	
Affiliation		Dr. Knoell Consult	

Qualitative assessment		
Study assessment	**Score**	**Rationale**
Reliability/Repeatability	Klimisch 3	Not reliable. • One reliability cut-off criterion is not met according to ToxRTool (Schneider et al. 2009) • Experimental design and statistics meet basic scientific principles but important information about the exposure regime is unclear.
Limitations		Material and methods lists a number of insecticides which were not used in most of the experiments. This is not clearly reported. Exposure regimes for a number of experiments are only vaguely described and therefore not suitable for use in risk assessment. Thiamethoxam was only used in “mite trial #2, experimental orchards” in which mite densities in the control were too low for robust statistical evaluation.
GLP		No
Relevance	Supplemental data for toxicity on non-target arthropods (NTA)	Impact of insecticide use on predatory mite populations was investigated for the purpose of IPM strategies in apple orchards. The results are not suitable for use in risk assessment due to insufficient information about exposure regimes.