APPENDIX B.

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A Critique of Dr. Richard Heintzelman, Dr. Iain D. Kelly, Dr. David L. Fischer, Dr. Christian Maus, 23 May 2012 Overview of Recent Publications on Neonicotinoids and Pollinators

by

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June 14, 2012

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The Food Quality Protection Act engendered a concerted effort on the part of the agricultural chemical industry to develop reduced-risk insecticides. Certainly one of the most effective companies in this effort was Bayer Crop Sciences with their development of the new neonicotinoid class of insecticides that have achieved unparalleled world domination in agricultural, forestry, and urban pest control. No single class of pesticides has achieved such large sales, and over the last 15 years the portended advantages of systemic action in reducing non target exposure along with greater human safety have been the hallmark of this technology. As with all new uses, however, experience has brought forth new concerns as new pest species have arisen and major beneficial species have disappeared in agricultural ecosystems and concerns have been growing about the impacts of constant exposure through pollen and nectar for pollinating insects. Of even greater concern has been the recent rapid expansion of treating GMO seeds with systemic neonicotinoids, which violates the last 25 years of integrated pest management philosophy along with compromising pesticide resistance management rationales through eliminating alternating use of mixed mode of action materials. Having been registered by the US EPA and the EU through registration systems that everyone currently admits were inadequate for determining the true risks of systemic pesticides in the environment, their use continues to escalate environmental concerns for managed honey bees and for native pollinator species safety. In addition, contentious policy decisions continue to be promulgated on an unprecedented mixture of conflicting and limited science-based understanding amid emotionally charged political landscapes that differ among countries.

Heintzelman, et al. (2012) provide an overview of 10 recent articles from the peer reviewed open scientific literature dealing with research on neonicotinoid pesticides and pollinators. These authors cite each article and then focus their discussion on one point of contention for which they provide both limited and highly selected evidence, and then use this to discredit the entire paper. Unlike a normal literature review where the pros and cons of a given paper are presented along with all pertinent literature that bear on alternate interpretations or provide conflicting results, the authors here provide only material used to refute the chosen study. This is done by carefully selecting the studies that are quoted and in further selecting specific statements to support their position without regard to the entire context or overall conclusions of the paper being cited. In addition, relevant published literature is not included or cited, and a reader not skilled in science or familiar with the pertinent literature is left with a carefully written justification for a specific viewpoint, and not an objective critique of the science pertaining to the issues raised. The authors discredit 7 of the 10 papers chosen and use the other 3 as partial justification for their positions. If the peer reviewed scientific literature were as bad as these authors would have us believe, containing 70% of papers in error, I doubt that the US would have ever been the first country to place a man on the moon! Given the limitations of space, I cannot provide a detailed critique of all 10 papers and comments by Heintzelman et al., but will address the first and last papers as indicative of the highly questionable approach used by these authors.

Sub-lethal exposure of honey bees

Heintzelman, et al. chose 3 papers on sublethal effects of neonicotinoids on honey bees for their overview. The first two papers use the newest method of RFID-tagged bees which provides an automated digital tracking system to monitor the movements of individual bees in an entirely quantitative manner, and can be used to thus track the movements of large numbers of foragers from individual hives without the requirement of individual scientists there to visually track and identify single bees. This method is relatively new to bee science, and was deemed highly desirable for development as a new colony level screening tool for risk assessment and registration of new pesticides by the lab-hazard group at the Pellston conference of 2011 (Fischer and Moriarity, 2011).

Henry et al. found that bees fed a concentration of 1.34 ng of thiamethoxam were twice as likely to fail to return to the hive after foraging as were non treated bees. Heintzelman, et al. objected to these results with the following quote: "bees were tested at a single dose that was over twenty times greater than a worst-case estimate of the acute oral dose that is field relevant, while Schneider et al. tested a range of different exposure concentration which included concentrations corresponding to field-relevant exposure scenarios." Henry tested a dose of 1.34 ng in 20 microliters of sucrose, a non-lethal dose fed to individual bees. The objection of Heintzelman, et al. is based on an unnamed estimated level derived from an unnamed acute dose for reduction in foraging flights. This violates the standard practice of giving the authors or any other reader the option to refute any calculated dose raised as an objection by checking the calculation for themselves. The authors then conclude that:

> "As might be expected, Henry, et al., concluded that exposure to thiamethoxam residues in pollen and nectar could lead to adverse effects in pollinating bees while Schneider, et al., concluded that 'at field-relevant doses for nectar and pollen no adverse effect was observed for either substance.' It is highly likely that Henry et al. would have come to the same conclusion for their test substance thiamethoxam, if they would have followed Schneider et al.'s scientifically sound approach to test a range of concentrations that also included field-relevant dose rates."

To object to a specific dose on the basis of an estimated level without giving the reader the actual basis for this neglects not only good science protocol, but also obfuscates any arguments.

Objecting to the study of Henry et al. on the basis of the study of Schneider et al. by quoting the second paper as the only one dealing with environmentally relevant doses is highly misleading. All major reviews of the state of the science on neonicotinoids over the last few years, as well as two major bodies of scientists charged with reviewing the state of the science on pesticide risk assessment for pollinators, all agree that the actual field exposure levels of pesticides in pollen and nectar for pollinators are one of the major data gaps that prevent us from knowing the true risks associated with systemic neonicotinoids. (Kendembra 2009; Hopwood et al., 2012; Fischer and Moriarity, 2011; EFSA, 2012) Schneider, et al. tested imidacloprid at doses from 0.15-6 ng/bee and clothianidin at 0.05-2 ng/bee and found significant reduction in foraging activity and longer foraging flights for 3 hrs after feeding, for doses exceeding 1.5ng or 0.5 ng respectively. All of these are non-lethal doses which meet the criteria for assessing sub-lethal effects. Schneider et al. further cite work of Decourtye et al. who showed that fipronil at 0.3 ng/bee reduced the number of foraging flights to a feeder and prolonged the duration of homing flights in honey bees that lasted over a 3 day period, and the work of Yang, et al. (2008) who showed imidacloprid fed bees at doses of 40-50 ppb have a reduction in foraging flights. Further studies of El Hassani et al. (2008) show reduction in associative learning by neonicotinoids at similar doses using the PER response, a general measure of associative learning which is a major component of successful foraging behavior. All of these studies show impacts on foraging behavior within a similar range of doses for the same neonicotinoids as well as for fipronil. A new study by Eri and Nieh (2012) shows that honey bees that ingested imidacloprid (0.21 or 2.16 ng bee^{-1}) had higher sucrose response thresholds 1 h after treatment, while foragers that ingested imidacloprid also produced significantly fewer waggle dance circuits (10.5- and 4.5-fold fewer for 50% and 30% sucrose solutions, respectively) 24 h after treatment as compared with controls.

Schneider et al., do say that "no effects were seen for environmentally relevant doses," as quoted by Heintzelman et al. but then indicate they also saw significant effects at slightly higher doses. Schneider et al., further clarify their use of the term "environmentally relevant" dose as based on previous work by others that use the lowest nectar level of imidacloprid in sunflowers (Cresswell, 2011). This is clearly AN environmental dose, but not THE environmental dose representing the worst case scenario, which very likely has yet to be measured.

Recent work of Dively and Kamel (2012) show that environmental effects and method of application have major impacts on imidacloprid and thiamethoxam levels in pumpkin plants with levels reaching an average of 122 ng/gm in pollen and 17.6 ng/gm in nectar. Parent compounds represented only 15-27% of the detected residues, and since other metabolites can be as toxic as the parents, and may translocate differentially to pollen and nectar, these should also be included in the total residue levels. In another recent study on apples, the differential penetration of plant cuticle by neonicotinoids vs. insect growth regulators further emphasizes the lack of understanding of plant movements by pesticides to pollen and nectar vs. fruits (Sanchez et al., 2012). Bayer's own formulation for trees and shrubs, uses imidacloprid levels for label application rates that give 15-30 fold higher doses to ornamental plants compared to agronomic plants, so the doses in nectar and pollen of blooming ornamental plants is likely much higher than for sunflowers, and it certainly needs to be determined whether or not this represents the worst case scenario for pollinator exposure (Hopwood, et al., 2012).

When one examines the total relevant published literature for neonicotinoids impacting pollinator foraging behavior and the actual likely doses that are encountered, we see a very different picture than that depicted by Heintzelman et al. in their overview. The results of the paper of Schneider et al. confirm rather than refute the work of Henry et al. and the citation used by Heintzelman, while based on specific wording of Schneider et al., represents one narrow reference point for the term "environmentally relevant" that is not realistic for the world at large and remains yet to be determined, rather than a definitive basis for rejecting the work of Henry et al.

Heintzelman et al. attempt to further strengthen their case with the following statement:

> "The conclusions of Schneider et al. (2012) are confirmed by more than 30 field studies conducted with neonicotinoids where no effect on foraging and homing behavior of honeybees exposed to treated crops has been observed (see for instance Maus et al. 2003, Schmuck et al. 2005, Schmuck & Keppler 2003). In fact, there is no field evidence linking hive depopulations to sublethal exposures to neonicotinoids."

The fact that EPA has no standardized protocol for conducting a field study with honey bee colonies raises the distinct possibility that failure to detect any change in colony performance is a measure of the failure of the test system to measure the correct endpoints or to have the correct statistical discriminating power to quantify the degree of change given the inherent variability of the system. Cresswell (2011) performed a meta analysis of the major studies of neonicotinoids impacts on bees and thoroughly documents this lack of statistical discrimination among past studies to detect sub-lethal impacts. He further provides the 4 reasons why the known dose of neonicotinoids that bees are being exposed to and its impacts at the colony level still represent unknown data gaps that prevents us from knowing the true risks to pollinators, and argues for the needed research in much the same way as do Hopwood et al., and the EFSA working group. Cresswell summarizes it thus:

> "Given our current inability to resolve any of the preceding four complications it is not currently possible to be precise about the impact of trace dietary neonicotinoids on the health and fitness of honey bees in the field and these are issues for future research".

Neonicotinoids in Bees: Review and Risk Assessment

Heintzelman, et al. cite two reviews, that of Blacquiere, et al. summarizing 15 years of research on neonicotinoids and that of Cresswell, et al. using Hill's epidemiological causality criteria to examine the evidence that the agricultural use of neonicotinoids is a cause of the recently observed decline in honey bees. For the Blacquiere et al. review, Heintzelman et al. posit that "The authors conclude that the reported levels of neonicotinoids in nectar and pollen are below acute and chronic toxicity levels and the levels in bee-collected pollen, bees and bee products are low. They encourage collection of additional residue data before drawing final conclusions. Blacquiere, et al., note that laboratory studies have shown many lethal and sublethal effects of neonicotinoids, but no effects have been observed in the field studies with field-realistic dosages."

Here again, it seems that the levels of neonicotinoids used and field realistic doses used are based largely on agricultural field crop levels. The newest paper on pumpkins shows just one example in which multiple routes of treatment and environmental conditions exceed these levels (Dively and Kamel, 2012). The review also seems to ignore the potential for much higher levels in ornamentals and trees where repeated treatments using formulations and label directions that permit 15-30 fold higher doses than for ag crops are common place (Hopwood, et al. 2012; page 21). Cresswell (2011) gives a meta analysis of all relevant imidacloprid studies with extracted dose response curves that include doses from 1-100+ ng/bee, far exceeding the environmentally relevant doses cited in the Blacquiere, et al. review. If one considers the other relevant reviews and working group results for risk assessment that bear directly on major data gaps for actual exposure routes and quantities that bee foragers, nurse bees, and larvae actually acquire, clearly, the case is far from being defined (Kindembra, 2009; Hopwood, et al., 2012; Fischer and Moriarity, 2011; EFSA, 2012).

The citation of Cresswell, et al. 2012 by Heintzelman et al. is "the question of whether neonicotinoids cause bee population declines would be settled beyond reasonable doubt if realistically dosed honey bee colonies showed sufficient harm under field conditions." Based on their assessment of the available data they "conclude that dietary neonicotinoids cannot be implicated in honey bee declines". This is a gross misquote of Cresswell et al. and is opposite to the intent of the paper as given by them in their conclusion section of the abstract as follows: " It is concluded that dietary neonicotinoids cannot be implicated in honey bee declines, but this position is provisional because important gaps remain in current knowledge. Avenues for further investigations to resolve this longstanding uncertainty are therefore identified." They go on to give 4 specific suggestions for how research can address these areas, but clearly their intent is not that stated by Heintzelman et al.

The same detailed critique of the other papers cited in the overview by Heintzelman et al. could be made given more time and space. Clearly this overview is not a balanced, objective review of the papers cited or interpretations of them put into the context of all of the current scientific literature and the major unresolved questions that need further research, as is typical of a balanced scientific critique. For me this raises real concerns that the neonicotinoids that are currently being used in the market place were registered by a risk assessment process that was seriously flawed in its capacity to evaluate systemic pesticides. The plethora of unanswered questions about their environmental fates, exposures to honey bees and native bees, potential sub-lethal impacts, interactions with other pesticides, and contributions of their formulation inerts and adjuvants leave little confidence that their continued use will be free from additional complications for pollinators and perhaps for humans as well. The unusually high number of beekeeper reported bee kills in association with corn planting across seven states in the Midwest so far this year, indicates that beekeepers bear the brunt of undue risks from this class of pesticides.

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