Molecular Design of Dinotefuran with Unique Insecticidal Properties

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ABSTRACT: Dinotefuran, (*RS*)-1-methyl-2-nitro-3-(tetrahydro-3-furylmethyl)guanidine, is a neonicotinoid insecticide developed by Mitsui Chemicals Agro. Dinotefuran provides a tetrahydrofuran (THF) moiety distinct from other neonicotinoids with a chloropyridine or chlorothiazole ring, which is considered to be an essential structural element for the neonicotinoid action. The molecular design strategy based on acetylcholine ester moiety as a lead structure has successfully led to the discovery of dinotefuran with the cyclic ether THF functional group. The unique chemical and excellent biological properties and favorable toxicological profile make dinotefuran available for pest management in wide range of crops with a variety of application methods.

KEYWORDS: acetylcholine, dinotefuran, neonicotinoid, nicotine, tetrahydro-3-furylmethyl moiety

■ INTRODUCTION

Dinotefuran, (*RS*)-1-methyl-2-nitro-3-(tetrahydro-3-furyl-methyl)guanidine (**1**), is a neonicotinoid insecticide commercialized by Mitsui Chemicals Agro in 2002 and is now increasingly utilized in more than 20 countries (Figure 1).[1] The unique structural feature of dinotefuran is a tetrahydrofuran (THF) nonaromatic cyclic ether substituent, which is distinct from those of other neonicotinoids (**2**−**7**) with chlorinated heteroaromatic counterparts.[2,3] Our research in exploring a novel functional group, replacing the heteroaromatic ring of other neonicotinoids and nicotinoids nicotine (**8a**) and epibatidine (**8b**), started with the ester moiety of endogenous cholinergic agonist acetylcholine (ACh) (**9**), because the ACh ester moiety is functionally identical to the pyridine nitrogen atom of nicotine (Figure 2).[4] The goals of the present study are to succinctly introduce the strategic molecular design and structure−activity relationships (SARs) of dinotefuran and afterward to discuss the unique biological properties.

■ MOLECULAR DESIGN STRATEGY

ACh (**9**) is inactive as an insecticide because the ester moiety undergoes hydrolysis by the ACh esterase and the cationic ammonium head is restricted to penetrate into the target area (Figure 2).[5] Our molecular design strategy primarily focused on restructuring the ACh molecule to be active as an insecticide. Accordingly, the ACh ester moiety is substituted by alkyl ether to prevent hydrolytic degradation. Second, the quaternary ammonium head is also replaced by a 2-nitromethylene-imidazolidine ring, which is intrinsically active as a receptor agonist when combined with a chloropyridinylmethyl moiety, to improve the permeability into the nervous system as with imidacloprid.[6,7] We designed the prototype structure (**10**) to consider the SAR (Figure 2). Similar to the pyridine nitrogen atom of neonicotinoid and nicotinoid, the ether oxygen atom may also serve as a hydrogen acceptor. Therefore, the position of the oxygen atom is presumably important (Figure 3). The pyridine nitrogen atom of imidacloprid nitromethylene analogue could possibly take various positions due to the flexible methylene unit. Then, we designed two types of compounds as prototypes (**10a** and **10b**) with two and three methylene units.[1]

■ STRUCTURE−ACTIVITY RELATIONSHIPS

Eight prototypes **11**−**18** were prepared and evaluated for their insecticidal activity against the small brown planthopper (*Laodelphax striatellus*) and the green rice leafhopper (*Nephotettix cincticeps*) (Table 1). All compounds showed modest insecticidal activity of a similar score against *L. striatellus*, whereas compounds with three methylene units (**15**−**18**) generally had higher potency to the *N. cincticeps* than the corresponding compounds with two methylene units (**11**−**14**). Interestingly, ether type compounds **12** and **16** showed higher activity relative to that of alcohol and ester compounds. Therefore, the methyl propyl ether moiety of compound **16** served as a primer for further structural modifications.[1]

Subsequently, we designed cyclic ether compounds (**19**−**22**) with various hydrogen-accepting oxygen points in location and direction (Figure 4). The cyclic ether counterparts were combined at this time with the chemically stable 2-nitroimino-imidazolidine moiety.[8] The tetrahydro-3-furylmethyl compound (**22**) showed highest insecticidal activity among the four cyclic ether compounds (Table 2).[1]

The tetrahydro-3-furylmethyl moiety was then fixed for the following modifications on the imidazolidine counterpart. Acyclic *N*-nitroguanidine compound dinotefuran (**1**) was outstandingly insecticidal, comparable to imidacloprid (**2**), among the other analogues (Table 3).[1] Moreover, the THF moiety of **1** was substituted with methyl and ethyl groups (**27**−**31**) and was replaced by bicyclic (**32**) and tetrahydropyran (**33**) rings. The 4-methyl (**27**) and 5-methyl (**28**) THF analogues retained some potency of the unsubstituted **1**, whereas other analogues
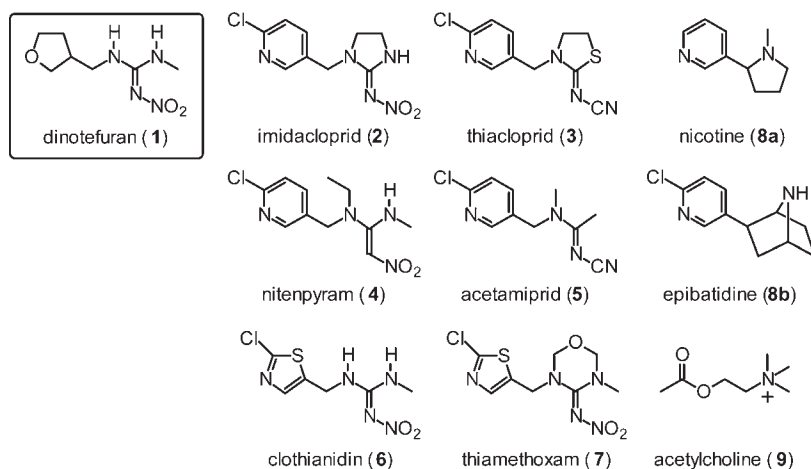
Figure 1. Structures of dinotefuran (**1**), other neonicotinoid insecticides (**2**–**7**), and nicotinic agonists nicotine (**8a**), epibatidine (**8b**), and ACh (**9**).
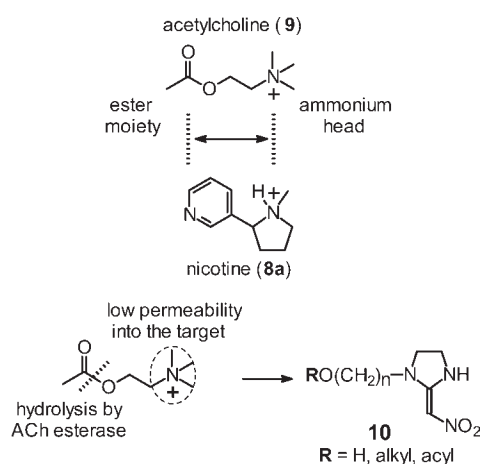

Figure 2. Molecular design strategy of dinotefuran originated from the ACh ester moiety leading to the prototype structure (**10**).
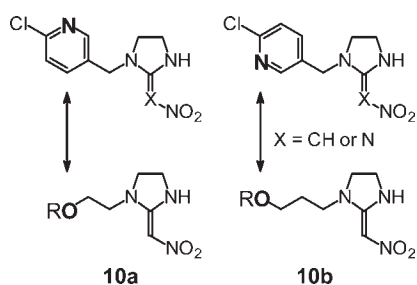

Figure 3. Design of the alkyl ether moiety of the prototype compounds based on possible conformations of chloropyridinyl neonicotinoid.

completely lost potency, except for **33** with some activity (Table 4).[9] The SAR of the acyclic nitroguanidine moiety of **1** was also examined with nine compounds (**34**–**42**) (Table 5). Markedly, *N*-acyl analogues **34** and **35** were highly potent (conceivably acting as proinsecticides). On the other hand, *N*-alkyl analogues **36**–**38** had moderate potency. In contrast, unsubstituted compound **39** resulted in greatly diminished potency. Analogue **40**, with a photolabile nitromethylene substituent, exhibited a potency identical to that of **1**. However, both types of denitrogen compounds (**41** and **42**) totally lost

Table 1. Insecticidal Activities of Compounds 11–18

11-18

	compd		activity score[a]	
no.	R	n	LS[b]	NC[c]
11	H	2	1	0
12	CH₃	2	1	2
13	CH₃CO	2	1	0
14	C₆H₅CO	2	1	0
15	H	3	1	1
16	CH₃	3	1	2
17	CH₃CO	3	1	1
18	C₆H₅CO	3	1	1

[a] Insecticidal activity (concentration producing 70% lethality) is graded as follows: 0, >1000; 1, 100–1000; 2, 10–100; 3, 1–10; 4, 0.1–1 ppm, respectively. [b] LS, small brown planthopper *Laodelphax striatellus*. [c] NC, green rice leafhopper *Nephotettix cincticeps*.
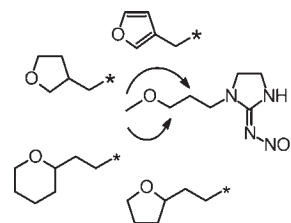

Figure 4. Design of cyclic ether neonicotinoid compounds.

potency.[10] A similar SAR pattern is reported in denitrogen heterocyclic nitromethylene compounds.[11]

Ultimately, we selected compound **1** (dinotefuran) for further development in 1995. (*S*)-(+)-Dinotefuran has higher potency relative to the (*R*)-(−)-isomer, but (*S*)-(+)- and (*RS*)-(±)-

Table 2. Insecticidal Activities of Compounds 19−22
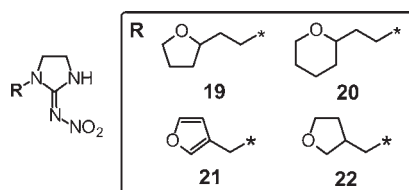
	activity score[a]	
compd no.	LS[b]	NC[c]
19	1	1
20	1	1
21	2	2
22	2	3

[a] Insecticidal activity (concentration producing 70% lethality) is graded as follows: 0, >1000; 1, 100−1000; 2, 10−100; 3, 1−10; 4, 0.1−1 ppm, respectively. [b] LS, small brown planthopper *Laodelphax striatellus*. [c] NC, green rice leafhopper *Nephotettix cincticeps*.

Table 3. Insecticidal Activities of Tetrahydro-3-furylmethyl Compounds 1, 23−26, and Imidacloprid (2)
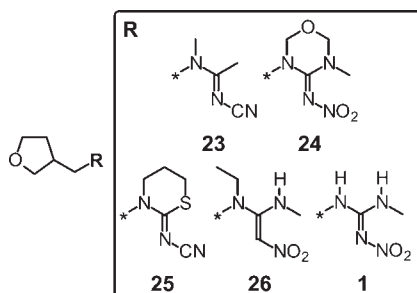
	activity score[a]	
compd no.	LS[b]	NC[c]
23	1	1
24	1	1
25	2	2
26	2	3
1 (dinotefuran)	4	4
2 (imidacloprid)	4	4

[a] Insecticidal activity (concentration producing 70% lethality) is graded as follows: 0, >1000; 1, 100−1000; 2, 10−100; 3, 1−10; 4, 0.1−1 ppm, respectively. [b] LS, small brown planthopper *Laodelphax striatellus*. [c] NC, green rice leafhopper *Nephotettix cincticeps*.

enantiomers show almost equal effectiveness in target site and insecticidal potency evaluations.[12,13]

■ BIOLOGICAL PROPERTIES

The high insecticidal activities of dinotefuran and its THF analogues are somewhat unexpected from earlier biochemical

Table 4. Insecticidal Activities of Dinotefuran (1) and Analogues 27−33

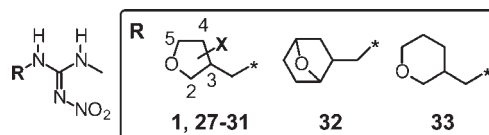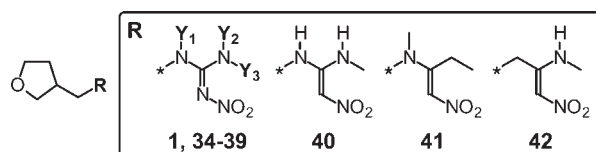| compd		activity score[a]	
no.	X	LS[b]	NC[c]
1 (dinotefuran)	H	4	4
27	4-CH₃[d−h]	2	3
28	5-CH₃[d−h]	2	3
29	4-C₂H₅[d−h]	0	0
30	2-CH₃[d−h]	0	0
31	3-CH₃	0	0
32[d−h]		0	0
33		1	2

[a] Insecticidal activity (concentration producing 70% lethality) is graded as follows: 0, >1000; 1, 100−1000; 2, 10−100; 3, 1−10; 4, 0.1−1 ppm, respectively. [b] LS, small brown planthopper *Laodelphax striatellus*. [c] NC, green rice leafhopper *Nephotettix cincticeps*. [d−h] Mixtures of diastereomers determined by ^{1}H NMR analysis: [d], 9:1; [e], 1:1; [f], trans; [g], trans; [h], exo:endo (1:1).

evaluations based on [³H]imidacloprid, [³H]epibatidine, or [³H]α-bungarotoxin binding to insect nicotinic ACh receptors (nAChRs) and from electrophysiological response in an insect/vertebrate hybrid receptor.[12−14] Finally, direct radioligand binding studies with [³H]dinotefuran were performed to determine if dinotefuran and imidacloprid and other neonicotinoids all interact with the identical site in the same way. Fascinatingly, [³H]dinotefuran displayed high affinity (~20 nM) for the insect nAChRs. Binding site specificity for THF neonicotinoid is usually identical to that for chlorinated-heteroaromatic neonicotinoid analogues, whereas a dissimilar binding mode of dinotefuran is suggested in some insect species.[15] Furthermore, the [³H]dinotefuran binding site in cockroach nerve cord nAChR has lower sensitivity to imidacloprid than to dinotefuran itself.[16] Dinotefuran has a broad insecticidal spectrum (Table 6) consistent with the neonicotinoid specificity being conserved across a wide range of insects.[3] Dinotefuran has high water solubility (54.3 g/L) and low log P value (−0.64), allowing its use in various formulations (e.g., granule, soluble granule, wettable powder, and dust) and applications (e.g., foliar, nursery box, submerged, and seed furrow) methods.

Neonicotinoid binding site interactions at chemical or atomic scale have been resolved by chemical and structural biology approaches using mollusk ACh binding protein (AChBP) as a suitable homologue of the extracellular ligand-binding domain of the nAChRs.[17] Dinotefuran binding site interactions are also simulated with the AChBP structure (Figure 5). The THF oxygen atom forms a water-bridge to the loop E amino acids at the binding domain, functionally identical to that of chloropyridine or the chlorothiazole nitrogen atom.[18] The nitroguanidine plane of dinotefuran π-stacks with the loop C Tyr aromatic ring, and the nitro tip oxygen undergoes hydrogen bonding with the Cys backbone NH.[18]

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Table 5. Insecticidal Activities of Dinotefuran (1) and Analogues 34–42

compd				activity score[a]	
no.	Y_1	Y_2	Y_3	LS^b	NC^c
1[d]	H	H	CH_3	4	4
34	CH_3CO	CH_3CO	CH_3	4	4
35	C_6H_5CO	C_6H_5CO	CH_3	3	4
36	CH_3	H	CH_3	3	3
37	CH_3	CH_3	CH_3	2	2
38	H	H	C_2H_5	1	2
39	H	H	H	0	1
40				4	4
41				0	0
42				0	0

[a] Insecticidal activity (concentration producing 70% lethality) is graded as follows: 0, >1000; 1, 100–1000; 2, 10–100; 3, 1–10; 4, 0.1–1 ppm, respectively. [b] LS, small brown planthopper *Laodelphax striatellus*. [c] NC, green rice leafhopper *Nephotettix cincticeps*. [d] Dinotefuran.

Table 6. Insecticidal Spectrum of Dinotefuran (1)

insect pest	activity grade[a]	insect pest	activity grade[a]
Hemiptera		**Isoptera**	
leafhoppers	++++	termites	++++
planthoppers	++++	**Coleoptera**	
whiteflies	++++	flea beetles	+++
aphids	++++	weevils	+++
mealybugs	++++	leaf beetles	++
rice bugs	++++	**Diptera**	
plant bugs	++++	leafminer flies	+++
Thysanoptera		houseflies	++
thrips	+++	**Dictyoptera**	
Lepidoptera		cockroach	+++
leafminers	+++	**Saitatoria**	
rice borers	++	grasshoppers	++
diamondback moth	++	**Siphonaptera**	
peach fruit moth	+++	flea	++++

[a] Activity definition: ++++, excellent; +++, very good; ++, fairly good.

Dinotefuran has no cross-resistance against imidacloprid-resistant insect pests,[19] probably attributable to an activated cytochrome P450 monooxygenase detoxification system.[20] Alternatively, a possible nAChR leafhopper mutation associated with lower imidacloprid mortality and receptor sensitivity than the wild type involves Tyr to Ser (adjacent to loop B Trp) of α subunit. This mutation influences the agonist potency of several chloropyridinyl or chlorothiazolyl neonicotinoids and
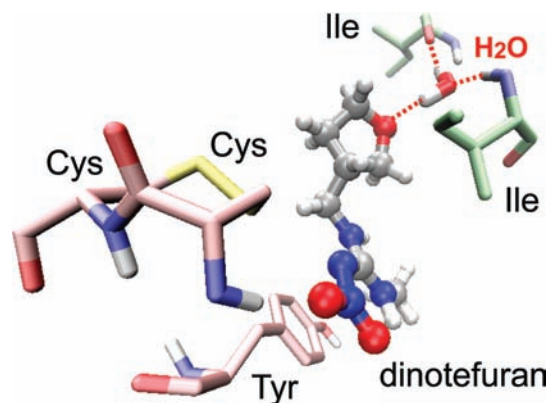

Figure 5. Binding site interactions of (S)-(+)-dinotefuran with the agonist binding pocket of the *Aplysia californica* AChBP,[18] which is an appropriate structural surrogate of the insect nAChR extracellular domain.[17] The THF oxygen atom of dinotefuran forms a water-bridge between the loop E Ile-106 backbone carbonyl oxygen and Ile-118 backbone NH. Nitroguanidine plane π-stacks with the loop C Tyr-188 aromatic ring, and nitro tip oxygen undergoes hydrogen bonding with the Cys-190 backbone NH. This image is regenerated in a different format on the basis of the data published by Ohno et al.[18]

Table 7. Mammalian Acute Toxicity of Dinotefuran

	LD_{50} (mg/kg)	
	male	female
oral		
mice	2450	2275
rat	2804	2000
dermal		
rat	>2000	>2000

Table 8. Avian and Aquatic Toxicity of Dinotefuran

Japanese quail (acute oral)	LD_{50}, mg/kg	>2000
mallard duck (dietary)	LC_{50}, mg/L (120 h)	>5000
bluegill sunfish	LC_{50}, ppm (96 h)	>100
Daphnia	EC_{50}, ppm (48 h)	>1000

epibatidine, but not of two nonchlorinated analogues (nicotine and dinotefuran) or ACh.[21] On the basis of analogy to the AChBP model, this single mutation conceivably induces conformational change in the area between loops B and E, where it is in close proximity to the binding position for the neonicotinoid chloropyridinyl/chlorothiazolyl ring.[22] Interestingly, a similar pattern is noted for another leafhopper with much higher sensitivity to THF compounds and ACh when assayed with [³H]dinotefuran than with [³H]imidacloprid.[15] Neonicotinoids are possible replacements for mosquito control if they have suitable potency and little or no cross-resistance, and dinotefuran is effective for pyrethroid-resistant mosquitoes.[23,24]

Selective toxicity of dinotefuran is clearly evident from the favorable toxicology profiles (Tables 7 and 8). Dinotefuran is rapidly detoxified by mammalian aldehyde oxidase and possibly cytochrome P450.[25] Moreover, dinotefuran and the major metabolites are almost inactive to the vertebrate neuronal α4β2 and α7 nAChR subtypes.[15,26] A selective neonicotinoid interaction mechanism for imidacloprid and thiacloprid

(presumably for dinotefuran) depends on multiple binding conformations.[27]

CONCLUDING REMARKS

Dinotefuran was discovered and commercialized by Mitsui Chemicals Agro. The molecular design originated from the ACh ester moiety as a lead structure, and the subsequent cyclization of an ether moiety of the prototype compound was the determining step, thereby ultimately conferring the distinctive THF chemotype neonicotinoid with unique biological properties and maximal safety.

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ABBREVIATIONS USED

ACh, acetylcholine; AChBP, ACh binding protein; LS, *Laodelphax striatellus* (small brown planthopper); NC, *Nephotettix cincticeps* (green rice leafhopper); nAChR, nicotinic ACh receptor; SAR, structure−activity relationship; THF, tetrahydrofuran.

REFERENCES

(1) Wakita, T.; Kinoshita, K.; Yamada, E.; Yasui, N.; Kawahara, N.; Naoi, A; Nakaya, M.; Ebihara, K.; Matsuno, H.; Kodaka, K. The discovery of dinotefuran: a novel neonicotinoid. *Pest Manag. Sci.* **2003**, *59*, 1016–1022.

(2) Kagabu, S. Molecular design of neonicotinoids: past, present, and future. In *Chemistry of Crop Protection*; Voss, A., Ramos, G., Eds.; Wiley-VCH: Weinheim, Germany, 2003; pp 193−212.

(3) Tomizawa, M.; Casida, J. E. Selective toxicity of neonicotinoids attributable to specificity of insect and mammalian nicotinic receptors. *Annu. Rev. Entomol.* **2003**, *48*, 339–364.

(4) Beers, W. H.; Reich, E. Structure and activity of acetylcholine. *Nature* **1970**, *228*, 917–922.

(5) Tomizawa, M.; Yamamoto, I. Binding of nicotinoids and the related compounds to the insect nicotinic acetylcholine receptor. *J. Pestic. Sci.* **1992**, *17*, 231–236.

(6) Tomizawa, M.; Yamamoto, I. Structure−activity relationships of nicotinoids and imidacloprid analogs. *J. Pestic. Sci.* **1993**, *18*, 91–98.

(7) Liu, M.-Y.; Lanford, J.; Casida, J. E. Relevance of [3H]imidacloprid binding site in house fly head acetylcholine receptor to insecticidal activity of 2-nitromethylene- and 2-nitroimino-imidazolidines. *Pestic. Biochem. Physiol.* **1993**, *46*, 200–206.

(8) Kagabu, S.; Medej, S. Stability comparison of imidacloprid and related compounds under simulated sunlight, hydrolysis conditions, and to oxygen. *Biosci., Biotechnol., Biochem.* **1995**, *59*, 980–985.

(9) Wakita, T.; Kinoshita, K.; Kodaka, K.; Yasui, N.; Naoi, A.; Banba, S. Synthesis and structure−activity relationships of dinotefuran derivatives: modification in the tetrahydro-3-furylmethyl part. *J. Pestic. Sci.* **2004**, *29*, 356–363.

(10) Wakita, T.; Kinoshita, K.; Yasui, N.; Yamada, E.; Kawahara, N.; Kodaka, K. Synthesis and structure−activity relationships of dinotefuran derivatives: modification in the nitroguanidine part. *J. Pestic. Sci.* **2004**, *29*, 348–355.

(11) Zhang, N.; Tomizawa, M.; Casida, J. E. α-Nitro ketone as an electrophile and nucleophile: synthesis of 3-substituted-2-nitromethylenetetrahydrothiophene and -tetrahydrofuran as *Drosophila* nicotinic receptor probes. *J. Org. Chem.* **2004**, *69*, 876–881.

(12) Mori, K.; Okumoto, T.; Kawahara, N.; Ozoe, Y. Interaction of dinotefuran and its analogues with nicotinic acetylcholine receptors of cockroach nerve cords. *Pest Manag. Sci.* **2002**, *58*, 190–196.

(13) Kiriyama, K.; Nishiwaki, H.; Nakagawa, Y.; Nishimura, K. Insecticidal activity and nicotinic acetylcholine receptor binding of dinotefuran and its analogues in the housefly, *Musca domestica*. *Pest Manag. Sci.* **2003**, *59*, 1093–1100.

(14) Kagabu, S.; Matsuda, K.; Komai, K. Preparation of dinotefuran related compounds and agonistic action on SADβ2 hybrid nicotinic acetylcholine receptors expressed in *Xenopus laevis* oocytes. *J. Pestic. Sci.* **2002**, *27*, 374–377.

(15) Honda, H.; Tomizawa, M.; Casida, J. E. Insect nicotinic acetylcholine receptors: neonicotinoid binding site specificity is usually but not always conserved with varied substituents and species. *J. Agric. Food Chem.* **2006**, *54*, 3365–3371.

(16) Miyagi, S.; Komaki, I.; Ozoe, Y. Identification of a high-affinity binding site for dinotefuran in the nerve cord of the American cockroach. *Pest Manag. Sci.* **2006**, *62*, 293–298.

(17) Tomizawa, M.; Casida, J. E. Molecular recognition of neonicotinoid insecticides: the determinants of life or death. *Acc. Chem. Res.* **2009**, *42*, 260–269.

(18) Ohno, I.; Tomizawa, M.; Durkin, K. A.; Casida, J. E.; Kagabu, S. Neonicotinoid substituents forming a water bridge at the nicotinic acetylcholine receptor. *J. Agric. Food Chem.* **2009**, *57*, 2436–2440.

(19) Foster, S. P.; Cox, D.; Oliphant, L.; Mitchinson, S.; Denholm, I. Correlated responses to neonicotinoid insecticides in clones of the peach-potato aphid, *Myzus persicae* (Hemiptera: Aphididae). *Pest Manag. Sci.* **2008**, *64*, 1111–1114.

(20) Puinean, A. M.; Foster, S. P.; Oliphant, L.; Denholm, I.; Field, L. M.; Millar, N. S.; Williamson, M. S.; Bass, C. Amplification of a cytochrome P450 gene is associated with resistance to neonicotinoid insecticides in the aphid *Myzus persicae*. *PLoS Genet.* **2010**, *6* (6), e1000999.

(21) Liu, Z.; Williamson, M. S.; Lansdell, S. J.; Han, Z.; Denholm, I.; Millar, S. N. A nicotinic acetylcholine receptor mutation (Y151S) causes reduced agonist potency to a range of neonicotinoid insecticides. *J. Neurochem.* **2006**, *99*, 1273–1281.

(22) Casida, J. E.; Tomizawa, M. Insecticide interactions with g-aminobutyric acid and nicotinic receptors: predictive aspects of structural models. *J. Pestic. Sci.* **2008**, *33*, 4–8.

(23) Corbel, V.; Duchon, S.; Zaim, M.; Hougard, J.-M. Dinotefuran: a potential neonicotinoid insecticide against resistant mosquitoes. *J. Med. Entomol.* **2004**, *41*, 712–717.

(24) Liu, H.; Cupp, E. W.; Guo, A.; Liu, N. Insecticide resistance in Alabama and Florida mosquito strains of *Aedes albopictus*. *J. Med. Entomol.* **2004**, *41*, 946–952.

(25) Honda, H.; Tomizawa, M.; Casida, J. E. Neonicotinoid metabolic activation and inactivation established with coupled nicotinic receptor-CYP3A4 and -aldehyde oxidase systems. *Toxicol. Lett.* **2006**, *161*, 108–114.

(26) Kanne, D. B.; Dick, R. A.; Tomizawa, M.; Casida, J. E. Neonicotinoid nitroguanidine insecticide metabolites: synthesis and nicotinic receptor potency of guanidines, aminoguanidines, and their derivatives. *Chem. Res. Toxicol.* **2005**, *18*, 1479–1484.

(27) Tomizawa, M.; Maltby, D.; Talley, T. T.; Durkin, K. A.; Medzihradszky, K. F.; Burlingame, A. L.; Taylor, P.; Casida, J. E. Atypical nicotinic agonist bound conformations conferring subtype selectivity. *Proc. Natl. Acad. Sci. U.S.A.* **2008**, *105*, 1728–1732.