

Article

Synthesis, Larvicidal Activities and Antifungal Activities of Novel Chlorantraniliprole Derivatives and Their Target in the Ryanodine Receptor

Qichao Chen ¹, Lixia Xiong ², Min Luo ¹, Jin Wang ¹, Changyan Hu ¹, Xiao Zhang ², Shujing Yu ², Yonghong Li ^{2,*} and Dequn Sun ^{1,*}

¹ Marine College, Shandong University at Weihai, No. 180, Wenhua West Road, Weihai 264209, China; E-Mails: qichao_chen@126.com (Q.C.); Min.Luo@rega.kuleuven.be (M.L.); wangjinsd@yeah.net (J.W.); sdu285@163.com (C.H.)

² State Key Laboratory of Elemento-Organic Chemistry, Institute of Elemento-Organic Chemistry, Nankai University, Tianjin 300071, China; E-Mails: xionglxia@nankai.edu.cn (L.X.); xiaojun060603@nankai.edu.cn (X.Z.); yushujing@nankai.edu.cn (S.Y.)

* Authors to whom correspondence should be addressed; E-Mails: bioassay@nankai.edu.cn (Y.L.); dequn.sun@sdu.edu.cn (D.S.); Tel./Fax: +86-22-2350-4281 (Y.L.); Tel.: +86-631-568-8537 (D.S.); Fax: +86-631-568-8303 (D.S.).

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Abstract: In order to identify novel chlorantraniliprole derivatives as potential insecticides or fungicides, 25 analogues of chlorantraniliprole were synthesized. The insecticidal activities against oriental armyworm and the antifungal activities against five typical fungi of these derivatives were tested. Compounds **2u**, **2x** and **2y** exhibited good activities against oriental armyworm, especially compounds **2u** and **2x** which showed higher larvicidal activities than indoxacarb. Moreover, all of the tested compounds exhibited activities against five typical fungi. The K_i values of all synthesized compounds were calculated using AutoDock4. The relationship between the K_i values and the results of insecticidal activities against oriental armyworm further indicated that the membrane-spanning domain protein of the ryanodine receptor might contain chlorantraniliprole binding sites.

Keywords: anthranilic diamides; chlorantraniliprole; ryanodine receptor; insecticidal activities; antifungal activities; molecular docking

1. Introduction

Lepidopteran pests such as the oriental armyworm (*Mythimna separata*) have become difficult to control because of their emerging resistance to various types of traditional insecticides [1,2]. In order to control the oriental armyworm effectively, there were currently two new types of diamide insecticides with exceptional insecticidal activities on a range of lepidopteran pests, the phthalic diamides, such as flubendiamide [3,4] and the anthranilic diamides, such as chlorantraniliprole [5] and cyantraniliprole (Figure 1) [6].

Chlorantraniliprole, discovered by DuPont, has excellent control of lepidopteran pests, low mammalian toxicity and a favorable environmental profile. The widespread use of chlorantraniliprole on lepidopteran pests in the future may result in pest resistance and residues in the field [7]. Thus, designing novel chlorantraniliprole derivatives as new insecticides has attracted considerable research attention. However, a reasonable design strategy could not be proposed because of the lack of a clear target for chlorantraniliprole. In our previous work [8], we proposed the ryanodine receptor [9] as a possible binding target for chlorantraniliprole and its derivatives using an AutoDock4 analysis, and we initially confirmed that the membrane-spanning domain protein of the ryanodine receptor might have specific binding site(s) for chlorantraniliprole derivatives. Figure 2a shows the relative position relationship between the proposed ryanodine receptor (in white color) and chlorantraniliprole (in blue color). Diamide insecticides activate insect ryanodine (Ry)-sensitive intracellular Ca^{2+} channels by affecting calcium release [10], as seen in Figure 2b. We initially identified the target by comparing the relationship between the insecticidal activities against diamondback moth and the molecular docking results.

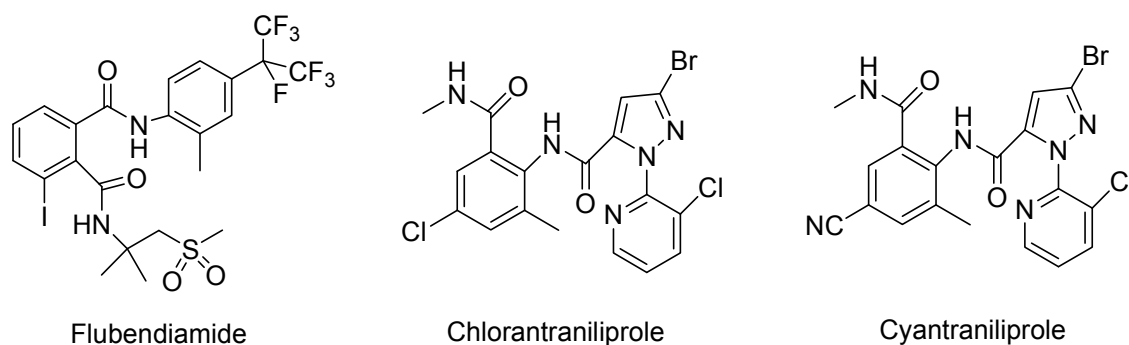


Figure 1. Insecticides acting on the insect ryanodine receptor [8]. Copyright © 2014 Elsevier Ltd.

Both diamondback moth and oriental armyworm are lepidopteran pests [11]. Therefore, in this study, oriental armyworm was selected for the first time for a bioactivity study, which examines the relationship with the K_i values of molecular docking. On the other hand, it was reported [12] that chlorantraniliprole derivatives might also show antifungal effects. Therefore, in this work the *in vitro* antifungal activity of some chlorantraniliprole derivatives was tested too. The chlorantraniliprole derivatives obtained herein were also used as new probes to bind with the proposed acceptor to further confirm that the membrane-spanning domain protein of ryanodine receptor was the special target binding with chlorantraniliprole and its derivatives.

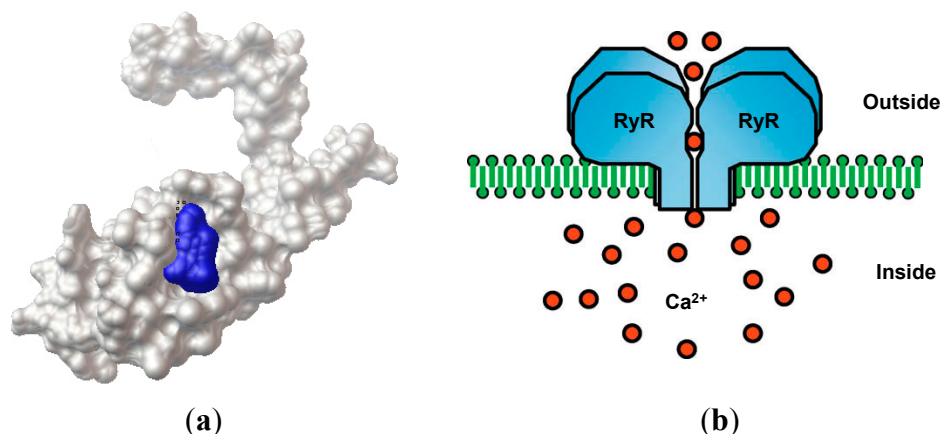


Figure 2. (a) Docking orientation of proposed ryanodine receptor(white color) and the chlorantraniliprole (blue color); (b) ryanodine receptor (RyR) and its Ca^{2+} release channels [8] Copyright © 2014 Elsevier Ltd.

In the previous work [8,13–16], most modifications of chlorantraniliprole were related to three parts: The pyrazole moiety (part A, Figure 3), amide moiety (part B, Figure 3), and anthraniloyl moiety (part C, Figure 3). Herein, new chlorantraniliprole derivatives were synthesized by modifying the amide-moiety.

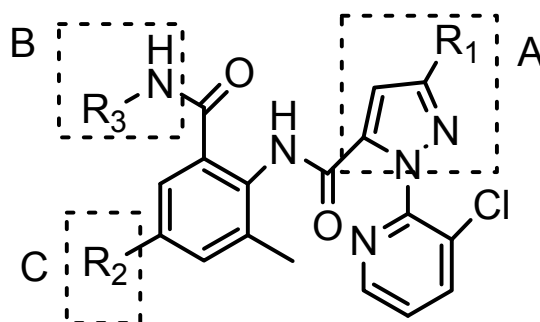
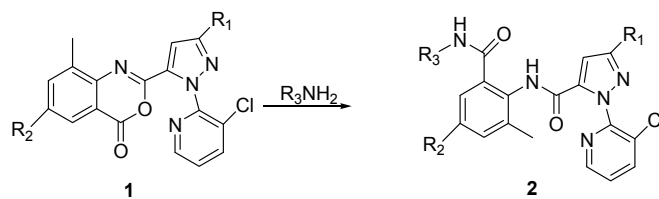


Figure 3. Modifications of chlorantraniliprole derivatives [8]. Copyright © 2014 Elsevier Ltd.

2. Results and Discussion

2.1. Chemistry

Compound **1** was obtained by literature methods (Scheme 1) [7,17–19]. The nineteen compounds **2b–t** were synthesized according to our previous work [8]. Among the new compounds **2a**, **2u–y**, compound **2u** was conveniently obtained by treatment of compound **1** with methyl 2-aminoacetate without any other catalyst or base at room temperature. Due to the insolubility of glycine in the reaction solvents (tetrahydrofuran or dichloromethane), a mixture of pyridine and water was selected as solvent in the preparation of compound **2v**. In the case of compounds **2w** and **2x**, a strong base (NaH) was needed, while triethylamine was employed for compound **2w**.



Compd. No.	R ₁	R ₂	R ₃	Compd. No.	R ₁	R ₂	R ₃
2a	Br	Cl		2n	Br	Cl	
2b	Br	Cl		2o	Br	CN	
2c	Br	Cl		2p	Br	CN	
2d	Br	Cl		2q	Br	CN	
2e	Br	Cl		2r	Br	CN	
2f	Br	Cl		2s	Br	CN	
2g	Br	Cl		2t	Br	CN	
2h	Br	Cl		2u	Br	Cl	
2i	Br	Cl		2v	Br	Cl	
2j	Br	Cl		2w	Br	Cl	
2k	Br	Cl		2x	Br	Cl	
2l	Br	Cl		2y	Br	Cl	
2m	Br	Cl					

Scheme 1. General synthetic route to the title compounds **2a–y**.

2.2. Larvicidal Activities and Structure-Activity Relationships (SARs)

The results in Table 1 indicate that all of the tested compounds **2a–y**, except for four compounds (**2a**, **2q**, **2r** and **2w**) showed the same larvicidal activities (100%) as chlorantraniliprole, indoxacarb and avermectins at the concentrations of 100 and 200 mg/L. These results reveal that most of the chlorantraniliprole derivatives in our study exhibited considerable insecticidal activities against oriental armyworm.

The larvicidal activities of all compounds except for three compounds (**2u**, **2x** and **2y**) were lower than those of chlorantraniliprole and avermectins when the concentration was under 50 mg/L, however, compounds **2u** and **2x** showed obviously better activities (both 100%) than indoxacarb (40%), even when the concentration was lowered to 5 mg/L, while, compound **2y** displayed the same

larvicidal activity (40%) as indoxacarb at this concentration. Notably, these two compounds **2u** and **2x** displayed the same activity (100%) as chlorantraniliprole and avermectins at the concentration of 5 mg/L.

Table 1. Insecticidal activities of title compounds **2a–y**, chlorantraniliprole, indoxacarb and avermectins against oriental armyworm.

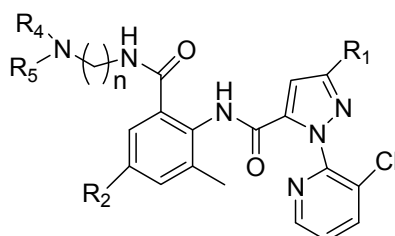
Compd. No.	Insecticidal Activities (%) at Different Concentrations					
	Concentrations (mg/L)					
2a	60					
2b	100	100	40			
2c	100	100	100	100	40	
2d	100	100	100	100	60	
2e	100	100	100	100	60	
2f	100	100	100	100	40	
2g	100	100	60			
2h	100	100	60			
2i	100	100	60			
2j	100	100	100	100	20	
2k	100	100	40			
2l	100	100	100	60		
2m	100	100	100	100	20	
2n	100	100	100	60		
2o	100	100	60			
2p	100	100	70			
2q	100	60				
2r	20					
2s	100	100	40			
2t	100	100	60			
2u	100	100	100	100	100	100
2v	100	100	40			
2w	30					
2x	100	100	100	100	100	100
2y	100	100	100	100	100	40
Chlorantraniliprole	100	100	100	100	100	100
Indoxacarb	100	100	100	100	100	40
Avermectins	100	100	100	100	100	100

In Table 2, the preliminary structure-activity relationship (SAR) data is summarized. All compounds with cyano groups ($R_2 = \text{CN}$) instead of a chloride group commonly had low insecticidal activities against oriental armyworm. For example, compounds **2o**, **2p**, **2q**, **2r**, **2s** and **2t** in Table 1 had no insecticidal activities when the concentration was less than 25 mg/L.

Most of the researchers in previous work [20–22] preserved the anthranilic amide moiety, which suggested that this structure was an important pharmacophore in those compounds [23]. The number of methylene groups in the amide moiety was also an important element for the insecticidal activity and two methylenes was the most favorable number for high activity. For example, compounds **2c**, **2d**, **2j**, **2l** and **2n** ($n = 2$) had high insecticidal activities (100%), while compounds **2g** and **2h** ($n = 3$ or 4)

showed lower activities (60%). Obviously, when R₄ and R₅ had the same substituent groups (Table 2) as in compounds **2d**, **2g** and **2h** (R₄ = R₅ = ethyl group), the insecticidal activities were dramatically reduced.

Table 2. SAR study of typical chlorantraniliprole derivatives.



Compd. No.	The Number of Methylene (n)	R ₄	R ₅	Insecticidal Activities * (%)
2c	2	CH ₃	CH ₃	100
2e	3	CH ₃	CH ₃	100
2d	2	CH ₃ CH ₂	CH ₃ CH ₂	100
2g	3	CH ₃ CH ₂	CH ₃ CH ₂	60
2h	4	CH ₃ CH ₂	CH ₃ CH ₂	60
2i	2	(CH ₃) ₂ CH	(CH ₃) ₂ CH	60
2j	2	CH ₃ CH ₂	H	100
2k	2	CH ₃ CH ₂ CH ₂	H	40
2l	2	(CH ₃) ₂ CH	H	100
2n	2	CH ₃	H	100

* The insecticidal activities against oriental armyworm at a concentration of 50 mg/L.

In addition, the substitution of R₄ or R₅ at the tail of the amide moiety was a significant factor affecting the insecticidal activities. When either one of R₄ or R₅ was substituted by hydrogen, most compounds (**2j**, **2l** and **2n**) displayed relatively high activities (100%), except compound **2k** (40%), while, when both R₄ and R₅ were substituted by alkyl groups, most of compounds (**2g**, **2h**, and **2i**) displayed relatively lower activities (60%), except compound **2d** (100%), which indicated that the secondary amine at the tail of the amide moiety was an necessary pharmacophore. In conclusion, the substitution at the terminal positions of the amide moiety and the number of methylene groups might be important factors that influence the insecticidal activities, which could be verified when more chlorantraniliprole derivatives are designed to increase the insecticidal activities against oriental armyworm.

2.3. Antifungal Activities

According to the data presented in Table 3, all of the chlorantraniliprole derivatives exhibited certain inhibitory effects against the five tested phytopathogenic fungi: *Fusarium oxysporum* (FO); *Cercospora arachidicola* (CA); *Physalospora piricola* (PP); *Alternaria solani* (AS); and *Fusarium graminearum* (FG).

The activities on FO and CA for all compounds, except **2t** and **2f** (both were 0%) showed varying inhibitory activities (ranging from 4.2% to 41.7%). Regarding the antifungal activities on PP, all tested compounds showed low to good inhibitory activities (from 5.3% to 78.9%) and two compounds (**2h**

and **2j**) showed higher activities (78.9%, 63%, respectively) than other compounds. The activities on AS for all compounds except for three (**2k**, **2l** and **2t**, no activity) showed inhibitory activities ranging from 11.1% to 50.0%.

Table 3. The antifungal activities against five fungi.

Fungi Compd. No.	Antifungal Activities (%) Against Five Fungi				
	FO	CA	PP	AS	FG
2b	11.4	20.8	31.6	38.9	51.9
2c	8.7	12.5	10.5	22.2	37.5
2d	20.0	20.8	28.1	22.2	29.6
2e	14.3	29.2	26.3	11.1	14.8
2f	8.6	0.0	22.8	38.9	37.0
2g	40.0	29.2	54.4	33.3	44.4
2h	25.7	41.7	78.9	11.1	29.6
2i	11.4	20.8	33.3	27.8	25.9
2j	20.0	29.2	63.2	22.2	40.7
2k	11.4	12.5	22.8	0	40.7
2l	11.4	29.2	33.3	0	14.8
2o	8.7	37.5	5.3	22.2	40.6
2p	13.0	31.3	7.9	16.7	31.3
2q	13.0	12.5	7.9	16.7	18.8
2r	5.7	12.5	28.1	27.8	40.7
2s	11.4	20.8	31.6	50.0	44.4
2t	0.0	4.2	24.6	0	18.5
2u	8.7	25.0	10.5	16.7	46.9
2y	8.7	6.3	7.9	11.1	25.0

FO: *Fusarium oxysporum*; CA: *Cercospora arachidicola*; PP: *Phylospora piricola*; AS: *Alternaria solani*; FG: *Fusarium graminearum*.

The activities of all tested compounds on FG were commonly higher than those on FO and AS. All compounds showed low to average inhibitory activities (from 14.8% to 51.9%) against FG and the compound **2b** showed better activity (51.9%) than the other compounds.

2.4. Molecular Docking Results

The results in Table 4 reveal that ten compounds (**2e**, **2l**, **2m**, **2o**, **2q**, **2r**, **2s**, **2t**, **2w** and **2y**) with unavailable K_i values except for four of them (**2e**, **2l**, **2m** and **2y**) displayed low biological activities ($\leq 60\%$) against oriental armyworm, while the other 15 compounds (**2a**, **2b**, **2c**, **2d**, **2f**, **2g**, **2h**, **2i**, **2j**, **2k**, **2n**, **2p**, **2u**, **2v** and **2x**) with available K_i value, except one compound (**2a**), generally displayed medium to high biological activities (40%–100%). These results further confirm that the membrane-spanning domain protein (GenBank accession no. JF927788) of the ryanodine receptor selected in our previous work [8] might have specific binding site(s) for chlorantraniliprole and its derivatives, and identification of the possible binding site is worthy of further study from a protein molecular level point of view. However, the docking software program might affect the accuracy of the K_i values and the biological diversity of pests plays an important role in the experimental activity

tests. These reasons could explain why compounds **2e**, **2l**, **2m** and **2y** with unavailable K_i values showed high activities (100%) and compound **2a** with available K_i values showed relatively no activity.

Table 4. The K_i values and the insecticidal activities against oriental armyworm.

Compd. No.	K_i Values	Insecticidal Activities *	Compd. No.	K_i values #	Insecticidal Activities *
2h	3.64	60	2e	unavailable	100
2j	5.98	100	2l	unavailable	100
2f	8.25	100	2m	unavailable	100
2b	14.4	40	2o	unavailable	60
2a	8.41	0	2q	unavailable	0
2g	15.16	60	2r	unavailable	0
2d	17.68	100	2s	unavailable	40
2p	17.85	70	2t	unavailable	60
2x	21.07	100	2w	unavailable	0
2i	28.27	60	2y	unavailable	100
2u	63.83	100			
2n	64.92	100			
2k	99.57	40			
2c	93.86	100			
2v	916.28	40			
Chlorantraniliprole	50.12	100			

* Insecticidal activities of tested compounds against oriental armyworm at 50 mg/L; # Unavailable: The compound couldn't interact with the selected receptor and the K_i values couldn't be calculated.

3. Experimental Section

3.1. Chemistry

3.1.1. General

Melting points (mp) of the products were determined in open capillary tubes and are uncorrected. The products were purified by column chromatography by using silica gel (200–300 mesh). $^1\text{H-NMR}$ and $^{13}\text{C-NMR}$ spectra were recorded on a Varian-400 instrument at room temperature with TMS as an internal standard and CDCl_3 or $\text{DMSO-}d_6$ as solvents. Mass spectra were recorded with a JEOL MS-D 300 mass spectrometer. The reactions were monitored by analytical thin-layer chromatography TLC with ultraviolet (UV) light and the TLC was carried out on silica gel GF₂₅₄. All reagents were purchased from Acros (Geel, Belgium) or Alfa Aesar (Deisenhofen, Germany). The anhydrous solvents were dried and purified according to standard techniques before use.

3.1.2. Syntheses

The intermediate compound **1** was obtained in six steps by literature methods [7,17,18]. The nineteen compounds **2b–t** were synthesized according to our previous methods [8]. The other six new title compounds **2a**, **2u–y** were prepared as follows:

3-Bromo-N-(4-chloro-2-methyl-6-((2,4,4-trimethylpentan-2-yl)carbamoyl)phenyl)-1-(3-chloropyridin-2-yl)-1H-pyrazole-5-carboxamide (2a). 2,4,4-Trimethylpentan-2-amine (28 mg, 0.22 mmol) was added to compound **1** (50 mg, 0.11 mmol) in tetrahydrofuran (THF, 5 mL). Then the mixture was stirred at room temperature and monitored by TLC. The crude product was recrystallized from a dichloromethane/hexane (1:2) to give the product **2a** as a white solid in 55% yield. mp. 202–206 °C; ¹H-NMR (CDCl₃): δ 11.84 (s, 1H, NHCOC), 8.37 (d, *J* = 4.8 Hz, 1H, 6-H pyridine), 7.78 (d, *J* = 8 Hz, Ph-H), 7.78 (d, *J* = 8 Hz, 1H, 4-H pyridine), 7.31 (dd, *J* = 4.8, 8 Hz, 1H, 5-H pyridine), 7.23 (d, *J* = 1.6 Hz, 1H, Ph-H), 7.01 (s, 1H, pyrazole-H), 2.18 (s, 3H, Ph-CH₃), 1.56 (s, 2H, CH₂), 1.30 (s, 6H, (CH₃)₂C), 0.89 (s, 9H, (CH₃)₃C); ¹³C-NMR (CDCl₃): δ 172.42, 155.66, 148.91, 146.77, 140.10, 139.07, 136.29, 134.78, 133.14, 130.47, 129.06, 128.09, 127.95, 125.79, 110.77, 55.99, 52.97, 31.23, 31.18, 27.26, 19.44; HRMS (ESI): *m/z* [M+H]⁺ calcd for C₂₅H₂₈BrCl₂N₅O₂: 602.0701. Found: 602.1055.

3-Bromo-N-(4-chloro-2-((2-(2-(methoxy)oxoethyl))carbamoyl)-6-methylphenyl)-1-(3-chloropyridin-2-yl)-1H-pyrazole-5-carboxamide (2u). To a solution of methyl glycinate (29 mg, 0.33 mmol) in dichloromethane (DCM, 2 mL), a solution of **1** (148 mg, 0.33 mmol) in DCM (3 mL) was added dropwise at 0 °C. Then the resulting mixture was stirred at room temperature and monitored by TLC. The reaction mixture was evaporated to remove most of the DCM to give the crude product, which was recrystallized from *n*-hexane to give **2u** as a white solid in 40% yield. mp. 142–146 °C; ¹H-NMR (CDCl₃): δ 9.84 (s, 1H, CONH-Ar), 8.42 (d, *J* = 7.2 Hz, 1H, pyridyl-H), 7.84 (d, *J* = 8 Hz, 1H, pyridyl-H), 7.36 (dd, *J* = 4.8 Hz, 8 Hz, 1H, pyridyl-H), 7.29 (s, 1H, Ph-H), 7.25 (d, *J* = 5.2 Hz, 1H, Ph-H), 7.05 (s, 1H, pyrazolyl-H), 6.76 (br, s, 1H, NHCO-Ar), 4.12 (d, *J* = 5.6 Hz, 2H, COCH₂NH), 3.77 (s, 3H, CH₃O), 2.17 (s, 3H, Ph-CH₃); ¹³C-NMR (CDCl₃): δ 169.55, 167.89, 156.34, 156.18, 148.90, 146.75, 138.98, 138.56, 133.37, 132.20, 131.79, 131.19, 128.94, 128.13, 125.67, 124.78, 110.91, 52.62, 41.69, 18.82; HRMS (ESI): *m/z* [M+H]⁺ calcd for C₂₀H₁₇BrCl₂N₅O₄: 553.0521. Found: 553.0526.

Methyl2-(2-(2-(3-bromo-1-(3-chloropyridin-2-yl)-1H-pyrazole-5-carboxamido-5-chloro-3-methylbenzamido)acetic acid (2v). To a solution of glycine (24 mg, 0.33 mmol) in a mixed solvent of pyridine/water (1:1.5, 5 mL), a solution of **1** (0.11 mmol) in pyridine/water (1:1.5, 2 mL) was added dropwise at 0 °C. Then the mixture was stirred at room temperature and monitored by TLC. When the reaction mixture was adjusted to pH 2–3 by addition of 1 mol/L hydrochloric acid, a white solid precipitated, which was collected and dried to give **2v** in 52% yield. mp. 212–214 °C; ¹H-NMR (DMSO-*d*₆): δ 12.67 (s, 1H, COOH), 10.54 (s, 1H, CONH-Ar), 8.83 (s, 1H, Ar-CONH), 8.49 (d, *J* = 4.4 Hz, 1H, pyridyl-H), 8.17 (d, *J* = 4 Hz, 1H, pyridyl-H), 7.61 (dd, *J* = 4.8 Hz, 8 Hz, 1H, pyridyl-H), 7.53 (s, 1H, pyrazolyl-H), 7.50 (s, 1H, Ph-H), 7.39 (s, 1H, Ph-H), 3.82 (d, *J* = 5.6 Hz, 2H, COCH₂NH), 2.16 (s, 3H, Ph-CH₃); ¹³C-NMR (DMSO-*d*₆): δ 170.55, 165.68, 155.54, 148.34, 146.88, 139.07, 135.57, 131.33, 130.94, 127.74, 126.58, 126.40, 125.63, 110.83, 40.97, 17.60; HRMS (ESI): *m/z* [M+H]⁺ calcd for C₁₉H₁₅BrCl₂N₅O₄: 527.9664. Found: 527.9668.

3-Bromo-N-(4-chloro-2-((2-(5-(dimethylamino)naphthalen-1-ylsulfonyl))carbamoyl)-6-methylphenyl)-1-(3-chloropyridin-2-yl)-1H-pyrazole-5-carboxamide (2w). To a solution of 5-(dimethylamino)naphthalene-1-sulfonamide (94 mg, 0.4 mmol) and NaH (14 mg, 0.6 mmol) in THF (5 mL), a solution of **1** (180 mg, 0.4 mmol) in THF (3 mL) was added dropwise at 0 °C. Then the resulting mixture was

stirred for 10 min at room temperature. The reaction mixture was washed with 1 mol/L hydrochloric acid to pH 5. The aqueous phase was extracted with DCM (20 mL) and then dried to give the crude residue that was recrystallized from a DCM/hexane (1:1.5) mixture to give the product **2w** as a yellow solid in 77% yield. mp. 142–146 °C; ¹H-NMR (DMSO-*d*₆): δ 10.23 (s, 1H, CONH-Ar), 8.53 (dd, *J* = 1.6, 4.8 Hz, 1H, pyridyl-H), 8.17 (dd, *J* = 1.6, 8Hz, 1H, pyridyl-H), 8.32 (s, 1H, Ph-H), 8.30 (s, 1H, Ph-H), 8.10 (dd, *J* = 4.8Hz, 8 Hz, 1H, pyridyl-H), 7.252–7.690 (m, 6H, pyridine-H), 6.95 (s, 1H, pyrazolyl-H), 3.84 (s, 6H, 2NCH₃), 2.11 (s, 3H, Ph-CH₃); ¹³C-NMR (DMSO-*d*₆): δ 163.90, 155.66, 151.34, 148.00, 146.93, 139.20, 139.15, 132.86, 132.67, 131.93, 130.77, 128.89, 128.18, 127.44, 126.58, 126.32, 126.01, 123.24, 118.06, 115.06, 110.52, 109.19, 44.93, 17.37; HRMS (ESI): *m/z* [M+H]⁺ calcd for C₂₉H₂₄BrCl₂N₆O₄S: 701.0140. Found: 701.0092.

3-Bromo-N-(4-chloro-2-((2-cyanoacetyl)carbamoyl)-6-methylphenyl)-1-(3-chloropyridin-2-yl)-1H-pyrazole-5-carboxamide (2x). To a solution of 2-cyanoacetamide (11 mg, 0.13 mmol) and NaH (1 mg, 0.17 mmol) in THF (3 mL), a solution of **1** (0.11 mmol) in THF (1 mL) was added dropwise at room temperature. Then the mixture was stirred for 30 min and monitored by TLC. The reaction mixture was evaporated to give the product **2x** as a white solid in 50% yield. mp. 163–166 °C; ¹H-NMR (DMSO-*d*₆): δ 10.31 (s, 1H, CONH-Ar), 8.49 (d, *J* = 4.4 Hz, 1H, pyridyl-H), 8.16 (d, *J* = 4 Hz, 1H, pyridyl-H), 7.60 (dd, *J* = 4.8 Hz, 8 Hz, 1H, pyridyl-H), 7.57 (s, 1H, pyrazolyl-H), 7.48 (s, 1H, Ph-H), 7.33 (s, 1H, Ph-H), 3.58(d, *J* = 5.6 Hz, 2H, COCH₂NH), 2.23 (s, 3H, Ph-CH₃); ¹³C-NMR (DMSO-*d*₆): δ 182.92, 169.50, 152.51, 145.12, 143.97, 136.17, 135.92, 129.07, 127.88, 124.62, 123.57, 123.40, 122.90, 107.58, 36.50, 14.46; HRMS (ESI): *m/z* [M+H]⁺ calcd for C₂₀H₁₄BrCl₂N₆O₃: 553.9610. Found: 554.9688.

3-Bromo-N-(4-chloro-2-((2-heptyl)carbamoyl)-6-methylphenyl)-1-(3-chloropyridin-2-yl)-1H-pyrazole-5-carboxamide (2y). n-Heptylamine (25 mg, 0.22 mmol) was added to a solution of compound **1** (50 mg, 0.11 mmol) in THF (5 mL) at 50 °C. After 5 h, TLC showed the complete consumption of compound **1**. The mixed solution was evaporated to remove the THF. The residue was dissolved in DCM (20 mL). The organic layer was washed with H₂O, and dried to give a crude product that was then recrystallized from DCM/hexane (1:1.5) to give the product **2y** as a white solid in 58% yield; mp. 162–168 °C; ¹H-NMR (CDCl₃): δ 10.12 (s, 1H, NHCOCH₂), 8.44 (dd, *J* = 4.8 Hz, 1.6 Hz, 1H, 6-H pyridine), 7.83 (dd, *J* = 4.8 Hz, 1.6 Hz, 1H, 4-H pyridine), 7.36 (dd, *J* = 4.8 Hz, 8 Hz, 1H, 5-H pyridine), 7.21 (s, 1H, Ph-H), 7.17 (d, *J* = 6.4 Hz, 1H, NHCO), 7.15 (s, 1H, Ph-H), 6.14 (s, 1H, pyrazole-H), 3.35 (m, *J* = 7.2 Hz, 2H, CH₂NH), 2.16 (s, 3H, Ph-CH₃), 1.52 (m, *J* = 7.2 Hz, 2H, CH₂CH₃), 1.28 (m, *J* = 14 Hz, 8H, (CH₂)₄), 0.89 (t, *J* = 7.2 Hz, 3H, CH₃); ¹³C-NMR (CDCl₃): δ 167.84, 156.56, 149.09, 146.79, 138.81, 138.72, 133.22, 132.65, 132.26, 131.27, 129.00, 128.21, 125.63, 124.35, 111.03, 40.34, 31.68, 29.37, 28.91, 26.92, 22.59, 18.70, 14.06; HRMS (ESI): *m/z* [M+H]⁺ calcd for C₂₄H₂₆BrCl₂N₅O₂: 566.0725. Found: 566.0740.

3.2. Biological Tests

3.2.1. Biological Assay Methods

All of the tested compounds had the purity of more than 95%, and all bioassays were performed on representative test organisms reared in the laboratory. Evaluations were based on a percentage scale of

0–100, in which 0 = no activity and 100 = total kill. The standard deviations of the tested biological values were $\pm 5\%$.

3.2.2. Larvicidal Activities against Oriental Armyworm (*Mythimna separata* Walker)

The larvicidal activities against oriental armyworm were tested by foliar application [7]. The test was repeated at $25\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$ according to statistical requirements. The tested compounds were dissolved in acetone and diluted with water to the required concentrations from 200 to 5 mg/L for bioassay. Individual corn leaves were placed on moistened pieces of filter paper in Petri dishes. The leaves were then dipped in the test solution and allowed to dry. The dishes were infested with 10 fourth-instar oriental armyworm larvae. Percentage mortalities were evaluated 24 h after treatment. Each treatment was replicated for three times. For comparative purposes, chlorantraniliprole, indoxacarb and avermectins were selected as the controls under the same conditions.

3.2.3. Antifungal Bioassay: Inhibitory Effects on Phytopathogenic Fungi

The five phytopathogenic fungi chosen included *Fusarium oxysporum* (FO), *Cercospora arachidicola* (CA), *Physalospora piricola* (PP), *Alternaria solani* (AS), and *Fusarium graminearum* (FG). All the fungi are typical and often occur in the Chinese agro-ecosystem. The antifungal activities of nineteen chlorantraniliprole derivatives (**2b–l**, **2o–u** and **2x**) were tested in vitro by the poisoned food technique at the concentration of 50 $\mu\text{g}/\text{mL}$ by dissolving the compounds in DMSO and sterilized water (containing 1% Tween) and diluting with PDA in a Petri dish [24–26].

3.3. Molecular Docking Methods

In our study, the membrane-spanning domain protein of the ryanodine receptor was selected from GenBank (accession No. JF927788) [8] as the possible specific receptor, which was proposed to bind with small molecules to give the K_i values using AutoDock4. For the AutoDock4 docking methods readers can refer to our previous work [8]. The K_i values were used to evaluate the binding energy between the small molecules and their possible receptor. The analysis of the relationship between the K_i values and the insecticidal activities against oriental armyworm could thus afford further information about the specific receptor.

4. Conclusions

A series of novel chlorantraniliprole derivatives containing different amide groups and anthraniloyl moieties were designed and synthesized. Their insecticidal activities against oriental armyworm and the antifungal activities against five typical fungi were evaluated. The results indicated that all tested compounds except three (**2a**, **2r** and **2w**) exhibited favorable insecticidal activities against oriental armyworm. In particular, compounds **2u** and **2x** showed obviously better activities than indoxacarb even when the concentration was lowered to 5 mg/L. The preliminary structure-activity relationship of the title compounds indicated that compounds with a cyano group ($R_2 = \text{CN}$) had low insecticidal activities. Two methylene groups and a secondary amine in the amide moiety were necessary factors for increasing the insecticidal activities. Moreover, all of the tested compounds exhibited activities

against five typical fungi. The molecular docking results revealed that most of the compounds with available K_i values exhibited moderate to high activities against oriental armyworm. The relationship between the K_i values and the insecticidal activities suggested that the proposed membrane-spanning domain protein (GenBank accession no. JF927788) of the ryanodine receptor has special binding site(s) and might be the receptor of chlorantraniliprole and its analogs. Confirmation of this is a worthy topic for further study.

Supplementary Materials

Supplementary materials can be accessed at: <http://www.mdpi.com/1420-3049/20/03/3854/s1>.

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Author Contributions

Conceived, designed the synthesis: Dequn Sun. Performed the chemical experiments and spectroscopic analysis: Qichao Chen, Min Luo, Jin Wang and Changyan Hu. Designed, performed and analysed the biological experiments: Lixia Xiong, Xiao Zhang, Shujing Yu and Yonghong Li. Performed the molecular docking part: Qichao Chen. Wrote the paper: Qichao Chen and Dequn Sun. All authors contributed to the paper and approved the manuscript.

Conflicts of Interest

The authors declare no conflict of interest.

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Sample Availability: Samples of the compounds **2a–y** are available from the authors.

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