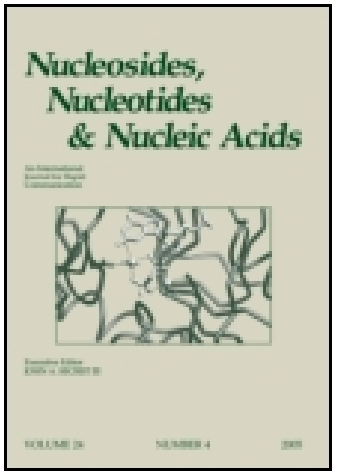


Nucleosides, Nucleotides and Nucleic Acids

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/lncn20

Synthesis and Insecticidal Activities of Novel 1,3,4-Thiadiazole 5-Fluorouracil Acetamides Derivatives: An RNA Interference Insecticide

Rong Wan [a], Jian-Qiang Zhang [a], Fen Han [a], Peng Wang [a], Peng Yu [a] & Qiu He [a]

[a] College of Science, Nanjing University of Technology, Nanjing, P. R. China

Published online: 27 May 2011.

To cite this article: Rong Wan, Jian-Qiang Zhang, Fen Han, Peng Wang, Peng Yu & Qiu He (2011) Synthesis and Insecticidal Activities of Novel 1,3,4-Thiadiazole 5-Fluorouracil Acetamides Derivatives: An RNA Interference Insecticide, Nucleosides, Nucleotides and Nucleic Acids, 30:4, 280-292, DOI: 10.1080/15257770.2011.580811

To link to this article: http://dx.doi.org/10.1080/15257770.2011.580811

Nucleosides, Nucleotides and Nucleic Acids, 30:280–292, 2011

ISSN: 1525-7770 print / 1532-2335 online
DOI: 10.1080/15257770.2011.580811

Taylor & Francis
Taylor & Francis Group

SYNTHESIS AND INSECTICIDAL ACTIVITIES OF NOVEL 1,3,4-THIADIAZOLE 5-FLUOROURACIL ACETAMIDES DERIVATIVES: AN RNA INTERFERENCE INSECTICIDE

Rong Wan, Jian-Qiang Zhang, Fen Han, Peng Wang, Peng Yu, and Qiu He
College of Science, Nanjing University of Technology, Nanjing, P. R. China

□ *A series of novel 1,3,4-thiadiazole 5-fluorouracil acetamides derivatives were designed and synthesized. Their structures were confirmed by infrared, ^1H NMR spectroscopy, and elemental analysis. The insecticidal activities against* Tetranychus cinnabarinus *and* Aphis craccivora *of these new compounds were evaluated. The bioassay tests showed that most of these title compounds possessed a good combination of stomach toxicity as well as contact toxicity against* Tetranychus cinnabarinus *and* Aphis craccivora. *In particular, the insecticidal activity of the title compound* ***IVe*** *against* Aphis craccivora *was better than the commercialized thiacloprid and was also comparable to another commercialized product, imidacloprid. The introduction of fluorines to meta and para-position of the benzene ring was essential for high bioactivity.*

Keywords 1,3,4-Thiadiazole; 5-fluorouracil; synthesis; insecticidal activities; RNA interference

INTRODUCTION

Due to the rapidly developing resistance of insects to pesticides and the desire to have compounds with less mammalian and environmental toxicity, the discovery of new leading structures with ideal properties has been one of the most important topics of research in pesticide chemistry for decades and continues to be an active area of research today.

In a variety of biological heterocyclic compounds, 1,3,4-thiadiazole derivatives have attracted considerable attention in chemical and medicinal research because of their diverse biological activities. It is reported that 1,3,4-thiadiazole derivatives possess wide spectrum insecticidal[1] and herbicidal[2] activities. Some of these compounds with broad spectrum have

Received 4 October 2010; accepted 11 April 2011.
We are thankful to Jue-ping Ni, Director of Bioassay Deptartment, Jiangsu Branch of National Pesticide Research and Development South Center, for support with bioassays. This work was supported by the Jiangsu High-Tech Research Project (Agriculture) (BG2007329) from the Science and Technology Department of Jiangsu Province.
Address correspondence to Rong Wan, College of Science, Nanjing University of Technology, Nanjing 210009, P. R. China. E-mail: rwan@njut.edu.cn

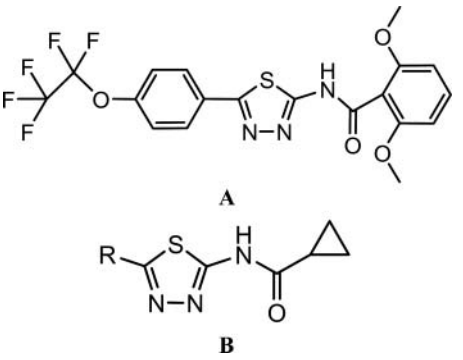


FIGURE 1 Chemical structures of compounds **A** and **B**.

already been commercialized, for instance, L 1215 (Figure 1A),[3] which is highly effective against mosquito larvae,[4] spodoptera eridania larvae,[5] and Western spruce budworm.[6] In addition, Liu et al. synthesized and tested a series of 1,3,4-thiadiazole amide derivatives (Figure 1B) and found that these compounds showed good fungicidal activities.[7] Furthermore, other 1,3,4-thiadiazole derivatives also have anti-inflammatory,[8] antiproliferative[9] and antibacterial[10–12] activities.

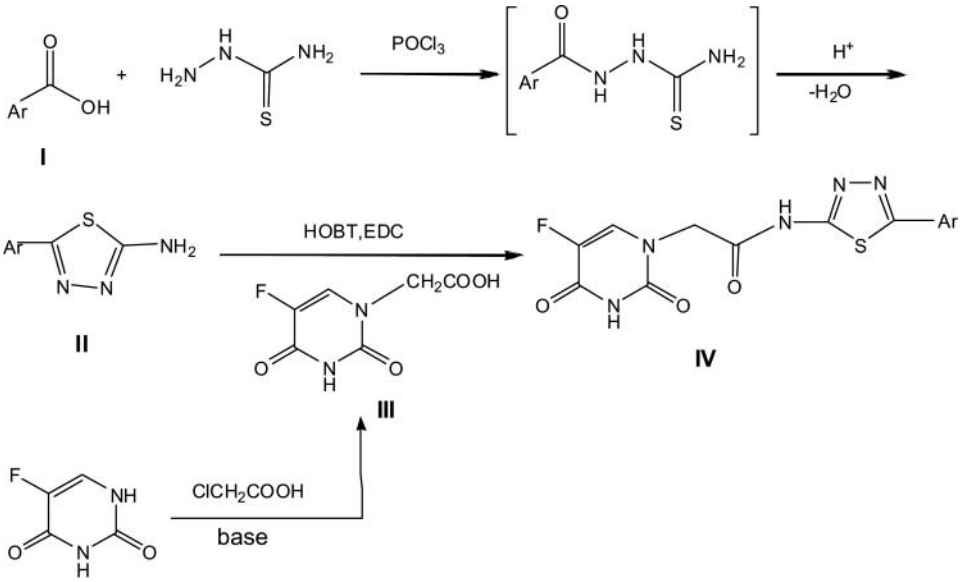
As a fluoropyrimidine, the mechanism of cytotoxicity of 5-fluorouracil has been ascribed to the misincorporation of fluoronucleotides into RNA and DNA and to the inhibition of the nucleotide synthetic enzyme thymidylate synthase. 5-Fluorouracil can be converted intracellularly to several active metabolites: fluorodeoxyuridine monophosphate (FdUMP), fluorodeoxyuridine triphosphate (FdUTP), and fluorouridine triphosphate (FUTP)—these active metabolites restrain the action of thymidylate synthase and disrupt RNA and DNA synthesis and repair.[13] Due to these satisfying advantages, 5-fluorouracil and its derivatives have been widely used as antitumor agents in the treatment of colon cancer,[14,15] gastric cancer,[16] esophageal cancer,[17] and so on.

Enlightened by the aforementioned findings and our desire to develop new insecticides with higher potency and better biological activity, we designed and synthesized a series of novel 1,3,4-thiadiazole acetamides derivatives containing 5-fluorouracil as shown in Scheme 1, and their biological activities were also evaluated.

RESULTS AND DISCUSSION

Synthesis

2-(5-fluoro-2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-N-(5-aryl- 1,3,4-thiadiazol-2-yl)acetamides **IVa–r** were synthesized as shown in Scheme 1. It is reported that 2-amino-5-alkyl-1,3,4-thiadiazole can be prepared by



SCHEME 1 General synthetic route for compounds **IVa-p**.

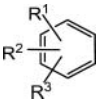
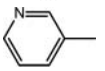
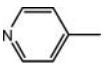
the reaction of aliphatic acid and thiosemicarbazide under the mineral acid such as sulfuric acid, hydrochloric acid or polyphosphoric acid. However, attempts to synthesize 2-amino-5-aryl-1,3,4-thiadiazoles using these acids were unsuccessful, yielding only acyl thiosemicarbazides. In this article, we obtained 2-amino-5-aryl-1,3,4-thiadiazole **II** in good yields under phosphorus oxychloride by amidation, nucleophilic addition and dehydration in one step. 5-Fluorouracil was reacted with chloroacetic acid using potassium hydroxide as alkali to yield compound **III**. The title compounds, **IVa–r**, were synthesized with the intermediate **II** and **III** in N,N-dimethylformamide (DMF) under 1-hydroxybenzotriazole (HOBT) and ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride (EDC) catalysis. Different akyl groups of the title compounds **IVa–r** are listed in Table 1.

Bioassay

Contact Toxicity Against Tetranychus cinnabarinus (Spider Mite)

Table 2 and Table 3 show the contact toxicities of the title compounds **IVa–r** and contrast compound avermectins against *Tetranychus cinnabarinus* (spider mite). The results indicate that some of the title compounds exhibit good acaricidal activities against *Tetranychus cinnabarinus*. The title compounds **IVa**, **IVc**, **IVe**, **IVk**, **IVm**, **IVn**, and **IVp** exhibit >80% mortalities at 400 mg/L. In particular, the acaricidal activities of compounds **IVe** and **IVm** against *Tetranychus cinnabarinus* are 100% at 400 mg/L, which are equal to the commercialized avermectins. Encouragingly, when the concentration

TABLE 1 Different akyl groups of the title compounds **IVa–r**

	Ar		
			
Compd.	R^1	R^2	R^3
IVa	3-OCH_3	H	H
IVb	2-Cl	4-Cl	H
IVc	3-CH_3	5-CH_3	H
IVd	4-NO_2	H	H
IVe	3-F	4-F	H
IVf	4-(n-C_5H_{11})	H	H
IVg	3-F	5-F	H
IVh	2-NO_2	4-Br	H
IVi	3-OCH_3	4-OCH_3	H
IVj	2-F	6-F	H
IVk	4-CH_3	H	H
IVl	3-OCH_3	4-OCH_3	5-OCH_3
IVm	4-OPh	H	H
IVn	4-(n-$C_{12}H_{25}$)	H	H
IVo	4-(n-C_8H_{17})	H	H
IVp	H	H	H
IVq			
IVr			

drops to 100 mg/L, **IVe** and **IVm** still had 95.65% and 97.67% acaricidal activities, respectively.

Contact Toxicity Against Aphis craccivora (Pea Aphids)

Table 4 and Table 5 show the contact toxicities of the title compounds **IVa–r** and contrast compound imidacloprid against *Aphis craccivora* (pea aphids). The results indicated that most of the title compounds exhibit good insecticidal activities against *Aphis craccivora*. The title compounds **IVc**, **IVe**, **IVg**, **IVj**, **IVk**, **IVm**, **IVn**, **IVo**, **IVp**, and **IVq** had >80% mortalities at 400 mg/L. Moreover, the insecticidal activities of compounds **IVe**, **IVg** and **IVm** are 100% at 40 mg/L against *Aphis craccivora*, which were equal to that of the commercialized imidacloprid. Particularly, the insecticidal activity of compound **IVe** is 91.03% at 2 mg/L against *Aphis craccivora*, which is comparable to that of the commercialized imidacloprid. Therefore, **IVe** stands out as the best and could be served as a promising candidate compound for further optimization.

The results of the contact toxicities of the title compounds **IVa–r** against *Tetranychus cinnabarinus* and *Aphis craccivora* imply that the bioactivity is

TABLE 2 Contact toxicities of compounds **IVa–r** and avermectins against *Tetranychus cinnabarinus* (spider mite) at 400 mg/L

	Mortality (%) at concn (mg/L)
Compd.	400
IVa	86.67
IVb	62.79
IVc	87.23
IVd	56.52
IVe	100.00
IVf	23.40
IVg	70.45
IVh	76.09
IVi	68.89
IVj	60.98
IVk	91.11
IVl	53.33
IVm	100.00
IVn	84.78
IVo	67.74
IVp	86.05
IVq	75.56
IVr	72.73
Avermectins	100.00

higher when the substituent on the benzene ring is a lower alkyl group such as CH_3, whereas it decreases as the groups are extended. Compared to the polyalkoxy substituted compounds **IVi** and **IVl**, the monoalkoxy substituted compound **IVa** exhibits a better bioactivity. But the bioactivity of the phenoxy substituted compound **IVm** is higher than these alkoxy substituted compounds **IVa**, **IVi** and **IVl**. In those electron-withdrawing groups, the fluorines substituted on meta and para position of the benzene ring exhibit better bioactivities than the ortho-substituted compounds. But the introduction of Cl, Br and nitro groups into the benzene ring results in decreased biological activity.

TABLE 3 Contact toxicities of compounds **IVc, IVe, IVk, IVm**, and avermectins against *Tetranychus cinnabarinus* (spider mite) at 100 mg/L

	Mortality (%) at concn (mg/L)
Compd.	100
IVc	28.89
IVe	95.65
IVk	23.40
IVm	97.67
Avermectins	100.00

TABLE 4 Contact toxicities of compounds **IVa–r** and imidacloprid against *Aphis craccivora* (pea aphids) at 400 mg/L

	Mortality (%) at concn (mg/L)
Compd.	400
IVa	77.89
IVb	76.09
IVc	88.51
IVd	45.45
IVe	100.00
IVf	56.00
IVg	100.00
IVh	41.18
IVi	53.85
IVj	81.82
IVk	93.62
IVl	75.82
IVm	100.00
IVn	86.51
IVo	81.82
IVp	89.88
IVq	80.95
IVr	70.00
Imidacloprid	100.00

Table 6 shows the contact toxicities of **IVe**, **IVg**, **IVm**, and the contrast compounds imidacloprid and thiacloprid against *Aphis craccivora*. The results of the median lethal concentrations (LC_{50}) and the toxicity regression equations of compounds **IVe**, **IVg**, **IVm**, imidacloprid, and thiacloprid are listed in Table 6. The results show that the insecticidal activities of compounds **IVe** and **IVg** against *Aphis craccivora* are 3.14- and 1.60-fold as high as that of thiacloprid on the basis of the LC_{50} values. The LC_{50} value of compound **IVm** is 2.40 mg/L, which is comparable to that of thiacloprid. Moreover, the LC_{50} value of the title compound **IVe** is 0.63 mg/L compared to 0.41 mg/L of the imidacloprid, which indicates that **IVe** possesses the same activity level as the commercialized imidacloprid.

TABLE 5 Contact toxicities of compounds **IVe, IVg, IVm**, and imidacloprid against *Aphis craccivora* (pea aphids) at 100–1 mg/L

	Mortality (%) at concn (mg/L)					
Compd.	100	40	10	4	2	1
IVe	100.00	100.00	100.00	100.00	91.03	53.06
IVg	100.00	100.00	92.70	74.14	58.66	46.24
IVm	100.00	100.00	87.21	58.18	31.96	27.88
Imidacloprid	100.00	100.00	100.00	100.00	100.00	100.00

TABLE 6 Contact toxicities of compounds **IVe, IVg, IVm,** imidacloprid, and thiacloprid against *Aphis craccivora* (pea aphids)

Compd.	$y = a + bx$	LC_{50} (mg/L)	Toxic ratio
IVe	$y = 5.4151 + 2.1162x$	0.63	3.14
IVg	$y = 4.8559 + 1.5227x$	1.24	1.60
IVm	$y = 4.4537 + 1.4332x$	2.40	0.83
Imidacloprid	$y = 5.5238 + 1.3595x$	0.41	4.83
Thiacloprid	$y = 4.5241 + 1.6090x$	1.98	1.00

Stomach Toxicity Against Aphis craccivora (Pea Aphids)

Table 7 shows the stomach toxicities of **IVe** and the contrast compounds imidacloprid and thiacloprid against *Aphis craccivora*. The results of the median lethal concentrations (LC_{50}) and the toxicity regression equations of compounds **IVe**, imidacloprid and thiacloprid are shown in Table 7. The LC_{50} value of the title compound **IVe** is 1.88 mg/L, compared to 2.04 mg/L of thiacloprid and 0.50 mg/L of imidacloprid. The results indicate that the insecticidal activity of the title compound **IVe** against *Aphis craccivora* is higher than the commercialized thiacloprid.

In summary, a series of novel propesticides 2-(5-fluoro-2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-N-(5-aryl-1,3,4-thiadiazol-2-yl)acetamides were designed and synthesized by the reaction of 5-aryl-1,3,4-thiadiazol-2-amines with 5-fluorouracil-1-yl acetic acid. The contact and stomach toxicities of the title compounds **IVa–r** were evaluated. The results of bioassays show that most of these title compounds possess a combination of good stomach toxicities as well as contact toxicities. The title compound **IVe** stand out as the best, which exhibits not only a higher insecticidal activity against *Aphis craccivora* than the thiacloprid but also a comparable insecticidal activity to imidacloprid.

EXPERIMENTAL

Melting points were recorded on an X-4 binocular microscope melting point apparatus (Beijing Tech Instruments Co., Beijing, China) and were corrected. Elemental analyses were performed with a Elementer Vario EL III elemental analyzer. ^{1}H NMR spectra were recorded on a Bruker DRX500

TABLE 7 Stomach toxicities of compounds **IVe,** imidacloprid, and thiacloprid against *Aphis craccivora* (pea aphids)

Compd.	$y = a + bx$	LC_{50} (mg/L)	Toxic ratio
IVe	$y = 4.3914 + 2.2080x$	1.88	1.09
Imidacloprid	$y = 5.3248 + 1.1097x$	0.50	4.08
Thiacloprid	$y = 4.4693 + 1.7103x$	2.04	1.00

spectrometer with DMSO-d6 as solvent and TMS as internal standard. Chemical shifts were reported in ppm (δ) values. Infrared (IR) spectra in KBr were recorded on a Perkin-Elmer PE-683 infrared spectrometer. Reactions were monitored by thin layer chromatography (TLC) with visualization by ultraviolet light. Unless otherwise noted, all reagents and solvents were used as received. Yields were not optimized.

General Synthetic Procedure for 5-Aryl-1,3,4-thiadiazol-2-amine II

To a mixture of compound **I** (0.1 mol) and thiosemicarbazide (0.1 mol) was added $POCl_3$ (0.3 mol) dropwise at 0–5°C and maintained for 30 minutes. The reaction mixture was allowed to raise temperature until it was refluxed and stirred for 4 hours. After cooling to room temperature, 50 mL water was added to the reaction mixture. The pH of the reaction solution was adjusted to the range of 8 to 9 with a solution of 50% NaOH. The crude product was precipitated, filtered, washed with water, dried, and recrystallized from ethanol to afford compound **II**.

Synthetic Procedure for 5-Fluorouracil-1-yl Acetic Acid III

To a stirred solution of potassiumhydroxide (0.7 g, 0.012 mol) in water (5 mL) was added 5-fluorouracil (1.3 g, 0.01 mol) and then heated at 60°C for 30 minutes. After that, the solution of chloroacetic acid (0.85 g, 0.009 mol) in water (2 mL) was added dropwise. The reaction mixture was then allowed to raise temperature to 90°C and stirred for 5 hours. The pH of the reaction solution was maintained at the range of 9 to 10 with a solution of 50% KOH. After it was cooled to room temperature, the pH of the reaction solution was adjusted to 2 with concentrated hydrochloric acid. The crude product was then precipitated, filtered, washed with water, recrystallized from ethanol, and dried to afford compound **III** as a white solid (1.50 g, 81%); m.p. 275–277 °C. ^{1}H NMR: δ 4.37 (s, 2H, CH_2CO); 8.08 (d, $J =$ 11.0 Hz, 1H, FC=CH), 11.88 (s, 1H, $(CO)_2NH$), 13.18 (s, 1H, COOH). Anal. calcd. (%) for $C_6H_5FN_2O_4$: C, 38.31; H, 2.68; F, 10.10; N, 14.89; O, 34.02. Found (%): C, 38.27; H, 2.63; F, 10.06; N, 14.82; O, 33.95.

General Synthetic Procedure for the Title Compounds IVa-r

To a stirred solution of compound **II** (0.01 mol) in DMF (15 ml) was added compound **III** (0.0105 mol), HOBT (0.012 mol), and EDC (0.012 mol). The resulting mixture was further stirred at room temperature for 24 hours. After completion of the reaction, the mixed solution was poured into water to precipitate. The precipitate was filtered, washed successively with alkali, acid, water, dried, and recrystallized from ethanol to afford the title compounds **IVa-r**.

IVa: m.p. 221–223°C; ^{1}H NMR (DMSO-d$_6$) δ(ppm): 3.93 (s, 3H, OCH_3), 4.03 (s, 2H, CH_2CO), 7.24–7.73 (m, 4H, Ar-H), 8.28 (d, J = 10.5 Hz, 1H, FC=CH), 12.23 (s, 1H, CONH); IR (cm^{-1}): 3305 ($\nu_{N\text{-}H}$); 3080 ($\nu_{Ar\ C\text{-}H}$); 2956, 2865 (ν_{CH3} C-H, ν_{CH2} C-H); 1721, 1697, 1665 ($\nu_{C=O}$); 1597 ($\nu_{C=N}$); 1581 ($\nu_{C=C}$); 1290 ($\nu_{C\text{-}N}$); 1253 ($\nu_{C\text{-}F}$); 1014 ($\nu_{C\text{-}O}$); 659 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{15}H_{12}FN_5O_4S$: C, 47.74; H, 3.21; N, 18.56; S, 8.50. Found (%): C, 48.80; H, 3.02; N, 18.90; S, 8.31.

IVb: m.p. 219–220°C; ^{1}H NMR (DMSO-d$_6$) δ(ppm): 4.73 (s, 2H, CH_2CO), 7.52–8.13 (m, 3H, Ar-H), 8.18 (d, J = 14.1 Hz, 1H, FC=CH), 12.00 (s, 1H, CONH), 13.19 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3213 ($\nu_{N\text{-}H}$); 3019 ($\nu_{Ar\ C\text{-}H}$); 2962 (ν_{CH2} C-H); 1720, 1693, 1662 ($\nu_{C=O}$); 1640 ($\nu_{C=N}$); 1585 ($\nu_{C=C}$); 1315 ($\nu_{C\text{-}N}$); 1273 ($\nu_{C\text{-}F}$); 1250 ($\nu_{C\text{-}Cl}$); 673 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{14}H_8Cl_2FN_5O_3S$: C, 40.40; H, 1.94; N, 16.83; S, 7.70. Found (%): C, 40.32; H, 1.99; N, 16.91; S, 7.94.

IVc: m.p. 237–239°C; ^{1}H NMR (DMSO-d$_6$) δ(ppm): 2.34 (s, 6H, $2CH_3$), 4.67 (s, 2H, CH_2CO), 7.16–7.54 (m, 4H, Ar-H), 8.09 (d, J = 11.0 Hz, 1H, FC=CH), 11.98 (s, 1H, CONH), 13.03 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3200 ($\nu_{N\text{-}H}$); 3036 ($\nu_{Ar\ C\text{-}H}$); 2946, 2839 (ν_{CH3} C-H, ν_{CH2} C-H); 1727, 1694, 1666 ($\nu_{C=O}$); 1584 ($\nu_{C=N}$); 1561 ($\nu_{C=C}$); 1315 ($\nu_{C\text{-}N}$); 1247 ($\nu_{C\text{-}F}$); 691 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{16}H_{14}FN_5O_3S$: C, 51.19; H, 3.76; N, 18.66; S, 8.54. Found (%): C, 51.02; H, 3.87; N, 18.60; S, 8.32.

IVd: m.p. 213–215°C; ^{1}H NMR (DMSO-d$_6$) δ(ppm): 4.71 (s, 2H, CH_2CO), 8.24 (m, 4H, Ar-H), 8.36 (d, J = 13.7 Hz, 1H, FC=CH), 11.97 (s, 1H, CONH), 13.24 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3253 ($\nu_{N\text{-}H}$); 3089 ($\nu_{Ar\ C\text{-}H}$); 2975 (ν_{CH2} C-H); 1728, 1697, 1673 ($\nu_{C=O}$); 1581 ($\nu_{C=N}$); 1560 ($\nu_{N=O}$); 1517 ($\nu_{C=C}$); 1340 ($\nu_{C\text{-}N}$); 1245 ($\nu_{C\text{-}F}$); 692 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{14}H_9FN_6O_5S$: C, 42.80; H, 2.31; N, 21.42; S, 8.17. Found (%): C, 42.80; H, 2.34; N, 21.53; S, 8.43.

IVe: m.p. 211–212°C; ^{1}H NMR (DMSO-d$_6$) δ(ppm): 4.71 (s, 2H, CH_2CO), 7.56–8.07 (m, 3H, Ar-H), 8.11 (d, J = 11.5 Hz, 1H, FC=CH), 11.99 (s, 1H, CONH), 13.13 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3178 ($\nu_{N\text{-}H}$); 3024 ($\nu_{Ar\ C\text{-}H}$); 2842 (ν_{CH2} C-H); 1732, 1697, 1662 ($\nu_{C=O}$); 1620 ($\nu_{C=N}$); 1576 ($\nu_{C=C}$); 1318 ($\nu_{C\text{-}N}$); 1249 ($\nu_{C\text{-}F}$); 659 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{14}H_8F_3N_5O_3S$: C, 43.87; H, 2.10; N, 18.27; S, 8.37. Found (%): C, 43.94; H, 2.23; N, 18.29; S, 8.54. In order to further confirm the structure, ^{13}C NMR, HSQC, and HMBC of compound **IVe** were recorded as a representative. ^{13}C NMR (300 MHz, DMSO-d$_6$) δ(ppm): 49.91, 115.90, 118.64, 124.34, 127.50, 130.57, 140.90, 149.78, 151.42, 152.45, 153.14, 157.50, 160.13, 166.51. HSQC (δ_H to δ_C) δ(ppm): 4.71–49.91, 7.64–118.64, 7.80–124.34, 8.04–115.90, 8.11–130.57. HMBC (δ_H to δ_C) δ(ppm): 4.71–130.57, 149.78, 166.51; 7.64–127.50, 151.42; 7.80–115.90, 152.45, 160.13; 8.04–124.34, 151.42, 160.13; 8.11–49.91, 140.90, 157.50.

IVf: m.p. 246–248°C; ^{1}H NMR (DMSO-d$_6$) δ(ppm): 0.85 (t, J = 7.5 Hz, 3H, CH_3), 1.29–1.58 (m, 6H, $3CH_2$), 2.55 (t, J = 10.6 Hz, 2H, CH_2), 4.67 (s,

2H, CH_2CO), 7.33–7.84 (m, 4H, Ar-H), 8.09 (d, $J = 11.0$ Hz, 1H, FC=CH), 11.98 (s, 1H, CONH), 13.02 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3195 ($\nu_{N\text{-}H}$); 3029 ($\nu_{Ar\ C\text{-}H}$); 2927, 2842 (ν_{CH3} C-H, ν_{CH2} C-H); 1731, 1699, 1662 ($\nu_{C=O}$); 1582 ($\nu_{C=N}$); 1550($\nu_{C=C}$); 1351 ($\nu_{C\text{-}N}$); 1227 ($\nu_{C\text{-}F}$); 685 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{19}H_{20}FN_5O_3S$: C, 54.67; H, 4.83; N, 16.78; S, 7.72. Found (%): C, 54.78; H, 4.80; N, 16.66; S, 7.84.

IVg: m.p. 218–219°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 4.68 (s, 2H, CH_2CO), 7.41–7.70 (m, 3H, Ar-H), 8.09 (d, $J = 11.0$ Hz, 1H, FC=CH), 12.00 (s, 1H, CONH), 13.19 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3250($\nu_{N\text{-}H}$); 3050 ($\nu_{Ar\ C\text{-}H}$); 2980 (ν_{CH2} C-H); 1716, 1698, 1661 ($\nu_{C=O}$); 1660 ($\nu_{C=N}$); 1558 ($\nu_{C=C}$); 1321 ($\nu_{C\text{-}N}$); 1200 ($\nu_{C\text{-}F}$); 675 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{14}H_8F_3N_5O_3S$: C, 43.87; H, 2.10; N, 18.27; S, 8.37. Found (%): C, 43.98; H, 2.20; N, 18.39; S, 8.12.

IVh: m.p. 220–222°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 4.69 (s, 2H, CH_2CO), 7. 65–8.10 (m, 3H, Ar-H), 8.38 (d, $J = 3.0$ Hz, 1H, FC=CH), 11.99 (s, 1H, CONH), 13.25 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3255 ($\nu_{N\text{-}H}$); 3095 ($\nu_{Ar\ C\text{-}H}$); 2950 (ν_{CH2} C-H); 1704, 1683, 1653 ($\nu_{C=O}$); 1617 ($\nu_{C=N}$); 1564 ($\nu_{N=O}$); 1526 ($\nu_{C=C}$); 1348 ($\nu_{C\text{-}N}$); 1292 ($\nu_{C\text{-}F}$); 1090 ($\nu_{C\text{-}Br}$); 668 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{14}H_8BrFN_6O_5S$: C, 35.86; H, 1.71; N, 17.83; S, 6.08. Found (%): C, 35.80; H, 1.65; N, 17.77; S, 6.32.

IVi: m.p. 229–230°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 3.84 (s, 6H, $2OCH_3$), 4.66 (s, 2H, CH_2CO), 7.06–7.50 (m, 3H, Ar-H), 8.09 (d, $J = 11.5$ Hz, 1H, FC=CH), 11.98 (s, 1H, CONH), 13.00 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3198 ($\nu_{N\text{-}H}$); 3055 ($\nu_{Ar\ C\text{-}H}$); 2950, 2841 (ν_{CH3} C-H, ν_{CH2} C-H); 1717, 1690, 1653 ($\nu_{C=O}$); 1584 ($\nu_{C=N}$); 1526 ($\nu_{C=C}$); 1273 ($\nu_{C\text{-}N}$); 1247 ($\nu_{C\text{-}F}$); 1022 ($\nu_{C\text{-}O}$); 662 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{16}H_{14}FN_5O_5S$: C, 47.17; H, 3.46; N, 17.19; S, 7.87. Found (%): C, 47.09; H, 3.38; N, 17.07; S, 7.95.

IVj: m.p. 215–216°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 4.62 (s, 2H, CH_2CO), 7.14–7.58 (m, 3H, Ar-H), 8.01 (d, $J = 11.5$ Hz, 1H, FC=CH), 11.92 (s, 1H, CONH), 13.15 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3182 ($\nu_{N\text{-}H}$); 3031 ($\nu_{Ar\ C\text{-}H}$); 2850 (ν_{CH2} C-H); 1720, 1708, 1653 ($\nu_{C=O}$); 1569 ($\nu_{C=N}$); 1554 ($\nu_{C=C}$); 1319 ($\nu_{C\text{-}N}$); 1242 ($\nu_{C\text{-}F}$); 694 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{14}H_8F_3N_5O_3S$: C, 43.87; H, 2.10; N, 18.27; S, 8.37. Found (%): C, 43.84; H, 2.03; N, 18.11; S, 8.45.

IVk: m.p. 230–232°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 2.37 (s, 3H, CH_3), 4.68 (s, 2H, CH_2CO), 7.32–7.82 (m, 4H, Ar-H), 8.05 (d, $J = 10.5$ Hz, 1H, FC=CH), 11.82 (s, 1H, CONH), 12.89 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3190 ($\nu_{N\text{-}H}$); 3068 ($\nu_{Ar\ C\text{-}H}$); 2910, 2805 (ν_{CH3} C-H, ν_{CH2} C-H); 1716, 1696, 1635 ($\nu_{C=O}$); 1589 ($\nu_{C=N}$); 1550 ($\nu_{C=C}$); 1311 ($\nu_{C\text{-}N}$); 1244 ($\nu_{C\text{-}F}$); 657 ($\nu_{C\text{-}S}$); Anal. calcd. (%) for $C_{15}H_{12}FN_5O_3S$: C, 49.86; H, 3.35; N, 19.38; S, 8.87. Found (%): C, 49.98; H, 3.50; N, 19.00; S, 8.35.

IVl: m.p. 235–237°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 2.37 (s, 3H, CH_3), 4.68 (s, 2H, CH_2CO), 7.32–7.82 (m, 4H, Ar-H), 8.05 (d, $J = 10.5$ Hz, 1H, FC=CH), 11.82 (s, 1H, CONH), 12.89 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3300 ($\nu_{N\text{-}H}$); 3060 ($\nu_{Ar\ C\text{-}H}$); 2946, 2837 (ν_{CH3} C-H, ν_{CH2} C-H); 1716, 1687, 1652

($\nu_{C=O}$); 1578 ($\nu_{C=N}$); 1510 ($\nu_{C=C}$); 1307 (ν_{C-N}); 1249 (ν_{C-F}); 1126 (ν_{C-O}); 693 (ν_{C-S}); Anal. calcd. (%) for $C_{17}H_{16}FN_5O_6S$: C, 46.68; H, 3.69; N, 16.01; S, 7.33. Found (%): C, 46.70; H, 3.73; N, 16.00; S, 7.56.

IVm: m.p. 246–248°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 4.67 (s, 2H, CH_2CO), 7.13–7.95 (m, 9H, Ar-H), 8.08 (d, J = 11.5 Hz, 1H, FC=CH), 11.98 (s, 1H, CONH), 13.08 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3164 (ν_{N-H}); 3090 ($\nu_{Ar\,C-H}$); 2850 (ν_{CH2} C-H); 1734, 1697, 1662 ($\nu_{C=O}$); 1590 ($\nu_{C=N}$); 1561 ($\nu_{C=C}$); 1313 (ν_{C-N}); 1247 (ν_{C-F}); 1080 (ν_{C-O}); 684 (ν_{C-S}); Anal. calcd. (%) for $C_{20}H_{14}FN_5O_4S$: C, 54.67; H, 3.21; N, 15.94; S, 7.30. Found (%): C, 54.78; H, 3.34; N, 16.12; S, 7.86.

IVn: m.p. 261–263°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 0.83 (t, J = 7.5 Hz, 3H, CH_3), 1.19–1.22 (m, 20H, $10CH_2$), 2.56 (t, J = 10.3 Hz, 2H, CH_2), 4.67 (s, 2H, CH_2CO), 7.32–7.84 (m, 4H, Ar-H), 8.09 (d, J = 11.0 Hz, 1H, FC=CH), 11.98 (s, 1H, CONH), 12.98 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3200 (ν_{N-H}); 3030 ($\nu_{Ar\,C-H}$); 2930, 2860 (ν_{CH3} C-H, ν_{CH2} C-H); 1734, 1691, 1662 ($\nu_{C=O}$); 1586 ($\nu_{C=N}$); 1540($\nu_{C=C}$); 1344 (ν_{C-N}); 1226 (ν_{C-F}); 690 (ν_{C-S}); Anal. calcd. (%) for $C_{26}H_{34}FN_5O_3S$: C, 60.56; H, 6.65; N, 13.58; S, 6.22. Found (%): C, 60.60; H, 6.72; N, 13.28; S, 6.55.

IVo: m.p. 252–254°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 0.83 (t, J = 7.1 Hz, 3H, CH_3), 1.23–1.59 (m, 12H, $6CH_2$), 2.58 (t, J = 10.7 Hz, 2H, CH_2), 4.67 (s, 2H, CH_2CO), 7.32–7.83 (m, 4H, Ar-H), 8.09 (d, J = 10.0 Hz, 1H, FC=CH), 11.97 (s, 1H, CONH), 13.02 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3195 (ν_{N-H}); 3045 ($\nu_{Ar\,C-H}$); 2923, 2862 (ν_{CH3} C-H, ν_{CH2} C-H); 1735, 1687, 1660($\nu_{C=O}$); 1580 ($\nu_{C=N}$); 1556($\nu_{C=C}$); 1360 (ν_{C-N}); 1225 (ν_{C-F}); 693 (ν_{C-S}); Anal. calcd. (%) for $C_{22}H_{26}FN_5O_3S$: C, 57.50; H, 5.70; N, 15.24; S, 6.98. Found (%): C, 57.56; H, 5.80; N, 15.33; S, 6.78.

IVp: m.p. 226–228°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 4.69 (s, 2H, CH_2CO), 7.41–7.54 (m, 5H, Ar-H), 7.78 (d, J = 12.5 Hz, 1H, FC=CH), 13.22 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3198 (ν_{N-H}); 3083 ($\nu_{Ar\,C-H}$); 2860 (ν_{CH2} C-H); 1724, 1670, 1631 ($\nu_{C=O}$); 1570 ($\nu_{C=N}$); 1543 ($\nu_{C=C}$); 1312 (ν_{C-N}); 1242 (ν_{C-F}); 692 (ν_{C-S}); Anal. calcd. (%) for $C_{14}H_{10}FN_5O_3S$: C, 48.41; H, 2.90; N, 20.16; S, 9.23. Found (%): C, 48.45; H, 2.87; N, 20.13; S, 9.11.

IVq: m.p. 249–251°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 4.69 (s, 2H, CH_2CO), 7.55–7.59 (m, 1H, Ar-H), 8.10 (d, J = 11.5 Hz, 1H, FC=CH), 8.31–9.12 (m, 3H, Ar-H), 11.99 (s, 1H, CONH), 13.17 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3225 (ν_{N-H}); 3012 ($\nu_{Ar\,C-H}$); 2986 (ν_{CH2} C-H); 1733, 1700, 1657 ($\nu_{C=O}$); 1593 ($\nu_{C=N}$); 1550 ($\nu_{C=C}$); 1316 (ν_{C-N}); 1251 (ν_{C-F}); 702 (ν_{C-S}); Anal. calcd. (%) for $C_{13}H_9FN_6O_3S$: C, 44.83; H, 2.60; N, 24.13; S, 9.21. Found (%): C, 44.80; H, 2.60; N, 24.11; S, 9.46.

IVr: m.p. 245–247°C; ^{1}H NMR (DMSO-d_6) δ(ppm): 4.70 (s, 2H, CH_2CO), 7.89–7.92 (m, 2H, Ar-H), 8.09 (d, J = 11.5 Hz, 1H, FC=CH), 8.72–8.74 (m, 2H, Ar-H), 11.99 (s, 1H, CONH), 13.26 (s, 1H, $(CO)_2NH$); IR (cm^{-1}): 3260 (ν_{N-H}); 3072 ($\nu_{Ar\,C-H}$); 2980 (ν_{CH2} C-H); 1731, 1698, 1663 ($\nu_{C=}$); 1589 ($\nu_{C=N}$); 1562 ($\nu_{C=C}$); 1305 (ν_{C-N}); 1250 (ν_{C-F}); 704 (ν_{C-S}); Anal. calcd. (%)

for $C_{13}H_9FN_6O_3S$: C, 44.83; H, 2.60; N, 24.13; S, 9.21. Found (%): C, 44.89; H, 2.55; N, 24.09; S, 9.46.

Biological Assay

All bioassays were performed on representative test organisms reared in the laboratory. The bioassay was repeated at 25 ± 1°C, according to statistical requirements. All compounds were dissolved in acetone and diluted with water containing Tween80 (500 mg L^{-1}) to obtain a series of concentrations of 400.0, 100.0, 4.0 mg L^{-1}, and others for bioassays. Assessments were made on a dead/alive basis and the data were subjected to probit analysis. Evaluations were based on a percentage scale of 0 to 100 in which 0 = no activity and 100 = total kill.

Contact Toxicity Against *Tetranychus cinnabarinus* (Spider Mite)

The horsebean plant leaves with adult mites were dipped in diluted solutions of the chemicals containing Tween80 (500 mg L^{-1}) for 10 seconds. After that, the leaves were transferred to penicillin vials within clean water for cultivation. Water containing Tween80 (500 mg L^{-1}) was used for contrast. The number of live and dead insects were counted after 72 hours. Each treatment was performed three times. For comparative purposes, avermectins was tested under the same condition. The results were listed in Table 2.

Contact Toxicity Against *Aphis Craccivora* (Pea Aphids)

The horsebean seedlings with insects (third instar *Aphis craccivora*) were dipped in diluted solutions of the chemicals containing Tween80 (500 mg L^{-1}) for 10 seconds. After that, the seedlings were transferred to penicillin vials within clean water for cultivation. Water containing Tween80 (500 mg L^{-1}) was used for contrast. Mortality was assessed after 72 hours. Each treatment was performed three times. For comparative purposes, imidacloprid was tested under the same condition. The results of the contact toxicities of the title compounds **IVa–r** and imidacloprid against *Aphis craccivora* (pea aphids) were listed in Table 4 and Table 5. The data for the mortality-regression lines of the compounds were subjected to probit analysis. The LC_{50} values of the title compounds **IVe**, **IVg**, **IVm**, imidacloprid, and thiacloprid against *Aphis craccivora* are listed in Table 6.

Stomach Toxicity Against *Aphis craccivora* (Pea Aphids)

The horsebean seedlings with insects (third instar *Aphis craccivora*) were dipped in diluted solutions with different concentrations of the title compounds **IVa–r** for cultivation. Mortality was calculated 72 hours after

treatment, and LC_{50} values were established. The data for the mortality-regression lines of the compounds were subjected to probit analysis. Each treatment was performed three times. Water containing Tween80 (500 mg L^{-1}) served as a contrast, and imidacloprid and thiacloprid were used as positive control samples. The LC_{50} values of the title compound **IVe**, thiacloprid, and imidacloprid against *Aphis craccivora* are shown in Table 7.

REFERENCES

1. Luo, Y.P.; Yang, G.F. Discovery of a new insecticide lead by optimizing a target-diverse scaffold: Tetrazolinone derivatives. *Bioorg. Med. Chem.* **2007**, 15, 1716–1724.
2. Hiroshi, K.; Rokuro, S.; Isao, H.; Takuo, O. Herbicidal activity of 1,3,4-thiadiazole derivatives. *J. Agric. Food Chem.* **1970**, 18, 60–65.
3. Ward, J.S. N-(1,3,4-thiadiazol-2-yl)benzamides. US Pat. No. 4141984, 1979.
4. Schaefer, C.H.; Miura, T.; Wilder, W.H. Biological activities of two new substituted benzamides against mosquitoes and nontarget organisms. *J. Econ. Entomol.* **1981**, 74, 658–661.
5. Gabrieli, S. N-(1,3,4-thiadiazol-2-yl)benzamides, their preparation and insecticidal compositions containing them. Israel Patent. IL 55355, 1982, 1–26.
6. Robertson, J.L. Toxicity of experimental molt inhibitors to western spruce budworm. *J. Ga. Entomol. Soc.* **1982**, 17, 413–416.
7. Liu, X.H.; Shi, Y.X.; Ma, Y.; Zhang, C.Y.; Dong, W.L.; Pan, L.; Wang, B.L.; Li, B.J.; Li, Z.M. Synthesis, antifungal activities and 3D-QSAR study of N-(5-substituted-1,3,4-thiadiazol-2-yl)cyclopropanecarboxamides. *Eur. J. Med. Chem.* **2009**, 44, 2782–2786.
8. Silvia, S.; Chiara, B.; Olga, B.; Francesco, B.; Angelo, R.; Walter, F.; Barbara, R.; Annalisa, C.; Giuseppe, F. New 1,3,4-thiadiazole derivatives endowed with analgesic and antiinflammatory activities. *Bioorg. Med. Chem.* **2006**, 14, 1698–1705.
9. Matysiak, J.; Opolski, A. Synthesis and antiproliferative activity of N-substituted 2-amino-5-(2,4-dihydroxyphenyl)-1,3,4-thiadiazoles. *Bioorg. Med. Chem.* **2006**, 14, 4483–4489.
10. Alireza, F.; Saeed, E.; Abdolreza, H.; Majid, R.; Kazem, S.; Mohammad, H.M.; Abbas, S. Synthesis and antibacterial activity of N-(5-benzylthio-1,3,4-thiadiazol-2-yl) and N-(5-benzylsulfonyl-1,3,4-thiadiazol-2-yl)piperazinyl quinolone derivatives. *Bioorg. Med. Chem. Lett.* **2005**, 15, 4488–4492.
11. Sara, T.; Tahmineh, A.; Mohammad, R.F.; Hossein, J.; Abbas, S. Synthesis and antibacterial activity of 1-[1,2,4-triazol-3-yl] and 1-[1,3,4-thiadiazol-2-yl]-3-methylthio-6,7-dihydrobenzo[c]thiophen-4(5H)ones. *Bioorg. Med. Chem. Lett.* **2005**, 15, 1023–1025.
12. Thomasco, L.M.; Gadwood, R.C.; Weaver, E.A.; Ochoada, J.M.; Ford, C.W.; Zurenko, G.E.; Hamel, J.C.; Stapert, D.; Moerman, J.K.; Schaadt, R.D.; Yagi, B.H. The synthesis and antibacterial activity of 1,3,4-Thiadiazole phenyl oxazolidinone analogues. *Bioorg. Med. Chem. Lett.* **2003**, 13, 4193–4196.
13. Longley, D.B.; Harkin, D.P.; Johnston, P.G. 5-Fluorouracil: Mechanisms of action and clinical strategies. *Nat. Rev. Cancer.* **2003**, 3, 330–338.
14. Cheng, C.Y.; Lin, Y.H.; Su, C.C. Anti-tumor activity of Sann-Joong-Kuey-Jian-Tang alone and in combination with 5-fluorouracil in a human colon cancer colo 205 cell xenograft model. *Mol. Med. Rep.* **2010**, 3, 227–231.
15. Lee, S.C.; Chan, J.Y.; Pervaiz, S. Spontaneous and 5-fluorouracil-induced centrosome amplification lowers the threshold to resveratrol-evoked apoptosis in colon cancer cells. *Cancer. Lett.* **2010**, 288, 36–41.
16. Won, H.J.; Ha, T.K.; Sung, C.H.; Hur, S.J.; Baik, H.H.; Suh, S.I.; Ha, E.; Kim, Y. Differential effects of 5-fluorouracil on glucose transport and expressions of glucose transporter proteins in gastric cancer cells. *Anti-Cancer Drugs.* **2010**, 21, 270–276.
17. Zemanova, M.; Petruzelka, L.; Pazdro, A.; Kralova, D.; Smejkal, M.; Pazdrova, G.; Honova, H. Prospective non-randomized study of preoperative concurrent platinum plus 5-fluorouracil-based chemoradiotherapy with or without paclitaxel in esophageal cancer patients: long-term follow-up. *Dis. Esophagus.* **2010**, 23, 160–167.