Synthesis, Spectroscopic Studies of Fluorinated Pyrimido-1,2,4-Triazines: Protective Effect Against Some Plant Pathogenic Fungi

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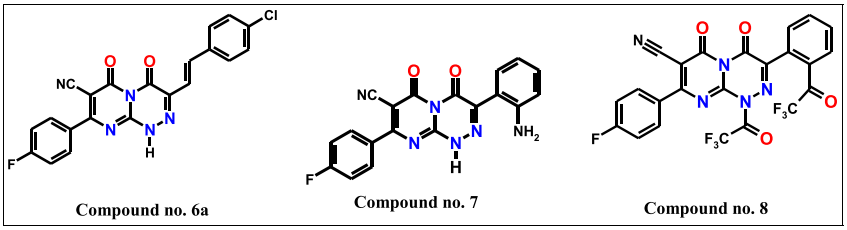


In search for new biologically active compounds, several new fluorine-substituted pyrimido [2,3-c][1,2,4] triazino (**3–16**) have been synthesized via the nucleophilic attack of 2-hydrazinopyrimidinone (**2**) toward more positive carbons under different conditions. Structure of the newly synthesized compounds was deduced from elemental analyses as well as their spectral data (UV, IR, NMR, and M/S). The antifungal activity of the target ring systems has been evaluated both *in vitro* and *in vivo* against some phytopathogenic fungi associated with wheat grains. Result showed that compounds **6**, **7,** and **8** have high protection of wheat grains against the fungal infection.

INTRODUCTION

The treatment of infectious diseases remains an important and challenging problem because of a combination of factors including the discovery of new infectious microorganisms and the increasing number of multi-drug resistant microbial pathogens. Therefore, there is real perceived need for the discovery of new compounds endowed with biocidal and/or enzymatic activity. Pyrimidines continue to receive much attention in recent years due to their unique physical, chemical, and pharmacological applications as potential microbial growth inhibitors [1–3], photochemical probe agents [4], and biocidal agents [5]. A variety of nitrogen-bridged pyrimidines was reported to be used as multi-targeted small molecule inhibitors and resistance-modifying agents [6–11]. On the other hand, 1,2,4-triazines and their condensed hetrobicyclic ring systems have attracted considerable interest in recent years as anti-HIV [12], antifungal agents [13,14], molluscicidal agents [15], and antitumor agents [16]. Furthermore, several triazine derivatives have been known to display a wide spectrum of medical and pharmacological activities [17,18]. In addition, some 1,2,4-triazine derivatives have shown an interesting activity as plant protection agents [19]. On the other hand, introduction of fluorine atoms into bioactive molecules often improves and enhances their medicinal, pharmacological, and biological activities, mainly because of the enhanced hydrophobicity, increase in membrane permeability, and increased stability against metabolic transformation. Moreover, the very high electronegativity of fluorine atom can modify the electronic distribution in the molecule, affecting its absorption, distribution, and metabolism [20,21].

In view of these observations, this work is devoted to the synthesis of fluorinated hybrid molecules containing both the pyrimidine and 1,2,4-triazine nuclei in one structure entity, namely, pyrimido[2,3-c][1,2,4]triazino hoping to get potential biologically active compounds as plant protecting agents against fungal infections.

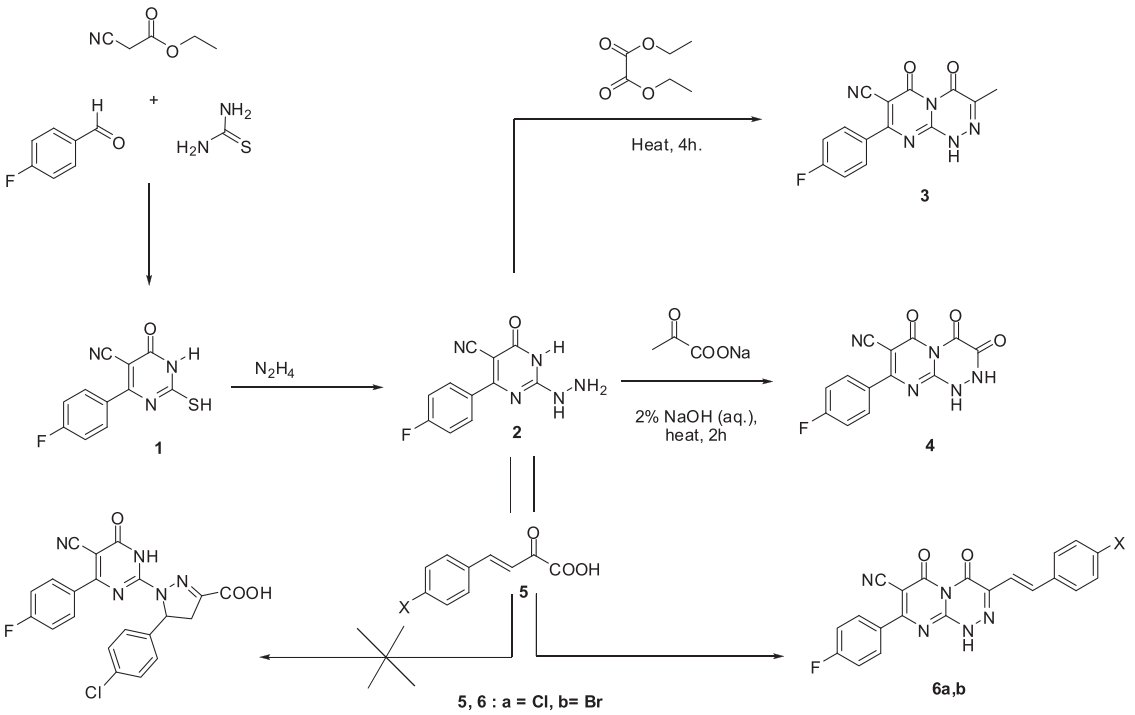
RESULTS AND DISCUSSION

Chemistry. The starting material 4-(4-fluorophenyl)-2-hydrazinyl-6-oxo-1,6-dihydropyrimidine-5-carbonitrile **2** was obtained from refluxing the corresponding 2-mercapto derivatives **1** with hydrazine hydrate in ethanol (Scheme 1).

Structures of **1** and **2** were deduced from their IR and ^{1}HNMR spectra. IR spectra of **1** showed the presence of NH, CN, C=O, and S–H functional groups at λ 3437, 2232, 1702, and 1161 cm^{-1}, whereas that of **2** recorded a lack of SH functional groups, with present of NH_2, NH at λ 3400–3199 cm^{-1}. ^{1}HNMR spectra of **1** showed a signal at δ 4.95–4.93 and 8.0 ppm for SH and NH protons (Fig. 1).

Cyclocondensation of 2-hydraziano pyrimidinone **2** with a 1,2-bicarbonyl compounds as sodium pyruvate (in aqueous sodium hydroxide) or diethyl oxalate (in THF) [22] afforded 8-(4′-fluorophenyl)-7-cyano-3-methylpyrimido[3,4-c][1,2,4]triazine-4,6-dione **3** and/or 8-(4′-fluorophenyl)-7-cyano-1,2,3,4-

Scheme 1. Synthesis of compounds **1–6.**



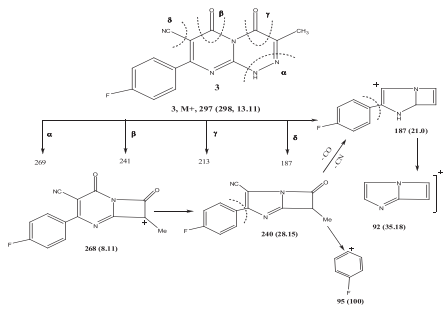


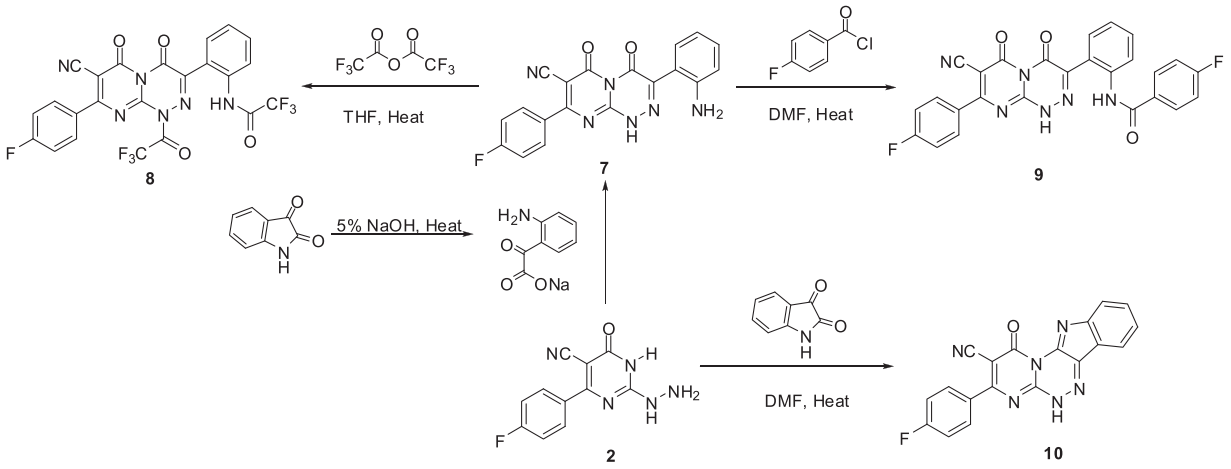
Figure 1. Mass fragmentation pattern of compound.

tetrahydropyrimido[3,2–c][1, 2, 4]triazine-3,4,6-trione **4**, respectively (Scheme 2). The IR of **3** showed λ at 1735 and 1678 cm^{-1} attributed to two carbonyl groups, whereas compound **4** recorded λ at 1670 and 1610 cm^{-1} for carbonyl and amide functions, respectively. The ^{13}C-NMR of compound **3** exhibited signals at δ 169.82 and 165.24 ppm for two C=O, whereas its MS recorded the molecular ion peak at *m/z* 298 with a base peak at *m/z* 95. An interesting result was obtained by the cycloaddition of 2-hydrazinopyrimidinone **2** with (*E*) 4-aryl-2-oxo-but-3-enoic acid **5** in refluxing sodium hydroxide solution [23], where 8-(4′-fluorophenyl-7-cyano-3-styryl-1*H*-pyrimido [3,2–c][1,2,4]triazine-4,6-diones **6a,b** were produced instead of the 5-(4-substitutedphenyl)-1-(5-cyano-4-(4-fluorophenyl)-6-oxo-1,6-dihydropyrimidin-2-yl)-4,5-dihydro-1*H*-pyrazole-3-carboxylic acids (Scheme 1). Compounds **6a,b** did not give acidity test with bicarbonate solution, instead, they showed various isomeric structural formulae. ^{1}H-NMR of compounds **6a,b** showed δ 8.16–7.97 ppm for 2H, coupling of aryl protons with the styryl moiety. In addition, the IR spectra recorded the presence of C–Cl, C–Br, and C–F at λ 756, 779, and 1250 cm^{-1}, whereas the MS of **6a** showed *m/z* at 420 with a base peak at 95 (Fig. 2).

In Scheme 2, compound **2** reacted with isatin under different reaction conditions (either in NaOH or DMF) [24], where 8-(4′-fluorophenyl-7-cyano-3-(2′-aminophenyl)-1*H*-pyrimido[3,2–c][1,2,4]triazine-4,6-dione **7** and 11-(4′-fluorophenyl-10-cyano-1H-pyrimido[3,2–c][1,2,4,]triazine[6,5-b] indole **10** were obtained, respectively. Chemical reactivity of compound **7** was evaluated by fluoroacetylation of the NH_2 group with trifluoroacetic anhydride in boiling THF, where the 1-(2′-trifluoroacetyl)-3-(2′-trifluoroacetyl)-3-(2′-trifluoroacetyl-aminophenyl)pyrimido[3,2-c][1,2,4] triazine-4,6-dione **8** was produced. Analogously, treatment of **7** with 4-fluorobenzoyl chloride in warm DMF resulted in the formation of the corresponding 4-fluoroanilido derivative **9**. The IR of **8** showed the presence of absorption bands of both NH_2 and two carbonyl groups at λ 3200, 1700, 1626 cm^{-1}, respectively. The mass spectrum of compound **10** exhibited the molecular ion peak at *m/z* 356 with a base peak at *m/z* 95 (C_6H_4F) (Fig. 3).

Regarding Scheme 3, regioselective heterocyclization of he starting 2-hydrazino-pyrimidinone **2** as a nucleophile with different α-active electrophilic reagents as monochloroacetic acid in refluxing aqueous NaOH produced 8-(4′-fluorophenyl-7-cyano-1,2,3,4–tetrahydropyrimido

Scheme 2. Synthesis of compounds **7–10**.



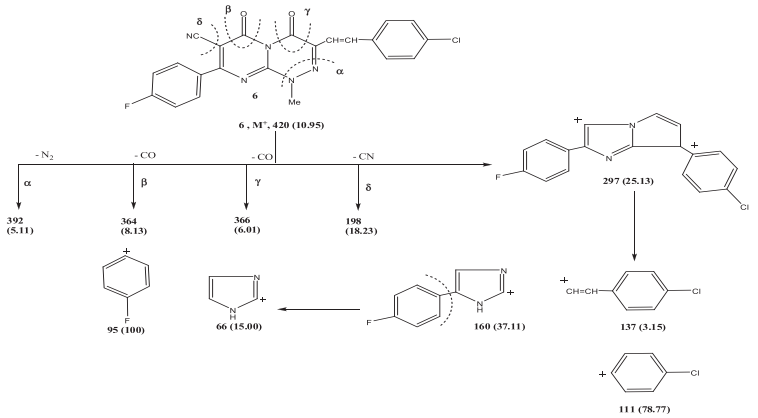


Figure 2. Mass fragmentation pattern of compound **6a**.

[3,2-c][1,2,4] triazine-4,6-dione **11**, whereas treatment of **2** with chloroacetyl chloride in warm DMF (accelerated SN^2 reaction) [20] yielded the isomeric structure **12**. Structures of **11** and **12** were confirmed from their UV, IR, and NMR spectra. The IR of compounds **11** and **12** recorded the presence of NH–NH and CH_2 as well as two carbonyl groups at λ 3366, 3152, 2890, 1668, and 1639 cm^{-1}, respectively. The UV of **10** recorded a λ_{max} at 373 nm attributed to n–π^* and π–π^* electronic transition. ^{13}C-NMR of **11** recorded δ at 170, 161 ppm for C=O and CONH carbons, whereas ^{1}H-NMR of **12** showed signals at δ ppm 12.32, 11.43 of two NH and 3.68 for CH_2 protons.

In their turn, compounds **11** and **12** were subjected to Kneogeval condensation with 4-chlorobenzaldehyde in refluxing ethanol using piperidine as a catalyst to produce the arylidene derivatives **13** and **14**, respectively (Scheme 3). The UV absorption of these compounds gave a good indication about their skeletons, where λ_{max} of **13** is higher than **14**, which is attributed to the aromatic stabilization of the heterocyclic system. On the other hand, oxidation of the pyrimidothiazinones **11** and **12** using $Fe_2(SO_4)$ in warm methanol yielded the corresponding oxidized products **15** and **16**, respectively. The prevalence of the aromatic phenolic form was chemically deduced by giving a positive test with neutral $FeCl_3$. The UV absorption spectrum of **15** recorded λ_{max} at 378 nm, whereas the IR spectra for both **15** and **16** exhibited λ at 2220, 1280, and 1250 cm^{-1} attributed to CN, C=O, and C–F functions, respectively, in addition to the absorption bands at λ 3450 cm^{-1} for OH in **15** and at 3470 cm^{-1} for **16**.

Biological activity. ***Plant protection and antifungal activity.*** It has been reported that several pyrimidinones are inhibitors of Zeta-carotene desaturase [25] and possess interesting herbicidal activity [26]. Recently, Abdel-Rahman et al. reported that some pyrimidopyrimidines acted as multi-targeted small molecule inhibitors and resistance-modifying agents. On the other hand, the NCNN group

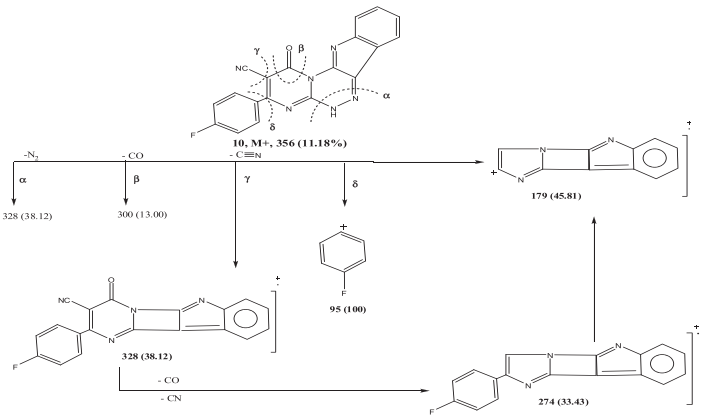
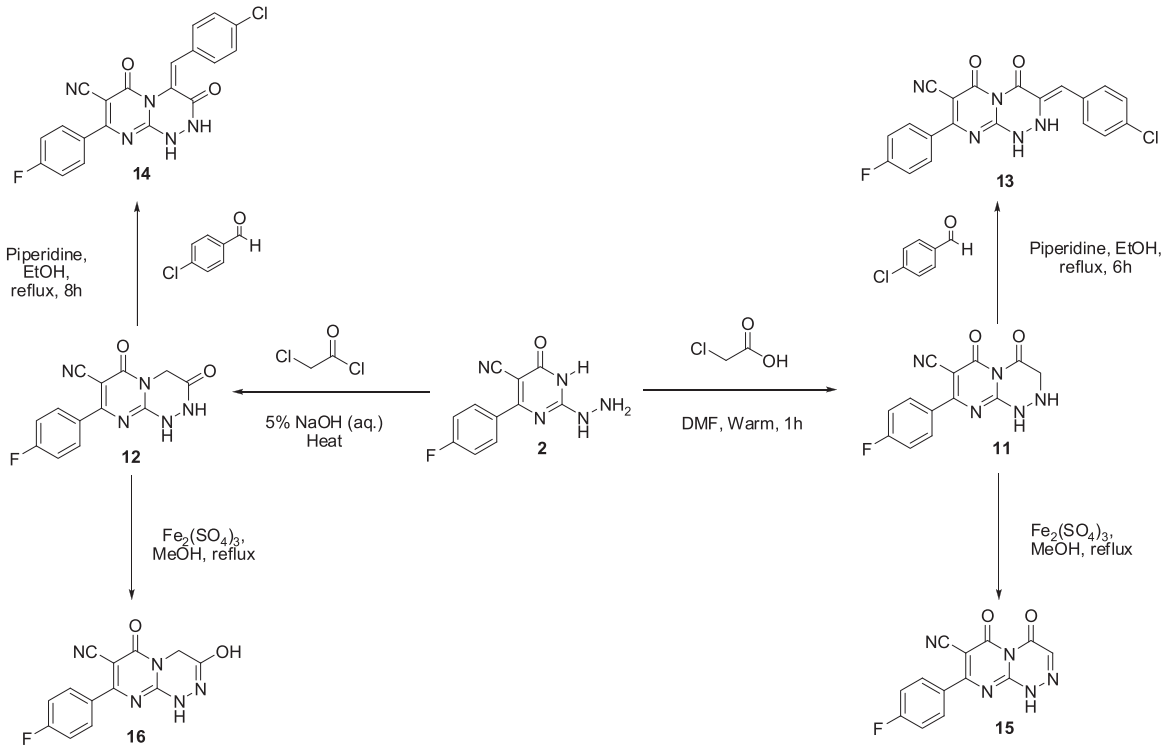


Figure 3. Mass fragmentation pattern of compound **10**.

Scheme 3. Synthesis of compounds **11–16**.



proved to be an essential part of various heterocycles bearing high biological activities [27,28]. In addition, 1,2,4-triazines and their fused derivatives have found applications as herbicides [29], amylolytic agent for fungi [30], and plant protection agents [31]. Therefore, it was of interest to evaluate the antifungal activity of the target compounds both *in vitro* and *in vivo* against some phytopathogenic fungi associated with wheat grains.

***In vitro* antifungal activity.** The *in vitro* antifungal activity of the new compounds was evaluated by inhibition of fungal mycelial growth of *Alternaria alterata*, *Helimenthosporium sativum,* and *Fusarium moniliforme*. DMF was used to make the desired concentrations, and sterile potato dextrose agar cultures were used. The culture plates were inoculated with a 4-mm diameter disc of inoculum of each fungus removed from a 7-day-old culture. Fungitoxicity was expressed as toxicity index depending on the ED_{50} values [32,33] (Table 1).

***In vivo* fungal activity.** The *in vivo* fungal toxicity activity of the compounds was tested on *A. alterata* and *F. moniliforme* fungi. Discs of orange rinds (3×3 cm)

Table 1

Antifungal activity toxicity index.

Comp. no.	*Alternaria alterata*	*Helimenthosporium sativum*	*Fusarium moniliforme*
1	4	3	2
3	5	2	0
4	2	0	4
6a	5	5	5
6b	4	5	5
7	5	0	0
8	1	0	0
11	3	3	0
12	2	1	1
13	2	0	1

Estimated ED_{50} for inhibition of mycelia growth.

were removed from sound novel orange fruits. The discs were surface sterilized by immersing 70% ethanol, the rinds were treated with the tested compounds by dipping. The treated discs were allowed to dry and were artificially inoculated with spores of tested fungi, commercial thiobendazol-2-(4-thiazolyl)benzimidazol was used as control. All treatments were replicated three times, and all the discs were stored in petri dishes containing a wet cotton plug to ensure a high relative humidity. After 1 week, the percentage of rotted discs was evaluated (Table 2).

From the results obtained in (Table 2), *F. moniliforme* is more sensitive to the tested compounds followed by *H. sativum* and *A. alterata.* Compounds **6a, 6b,** and **8** exhibited a high fungal toxicity activity. On the other hand, compound **7** is more toxic than the other tested compounds. It is interesting that compounds **11–13** showed a total loss of activity.

In the prevention of blue mold development [34], the action of the tested compounds on the decay control on rind discs is presented in Table 3. The results indicate that only compounds **3** and **6b** gave a good control at a concentration of 500 $\mu g/mL^{-1}$ against *A. alterata*, whereas compounds **1, 6b, 11,** and **12** gave a good control at a concentration of 1000 $\mu g/mL^{-1}$ against *F. moniliforme.* Finally, the best germination (80–90%) was achieved by treating the seeds with a solution containing 1000 µg/mL of compound **6a** followed by compound **8** under the same concentration (59–70% germination) (Table 3).

Table 2

ED_{50} (µg/mL) values of some fluorinated pyrimido-1,2,4-triazines.

Comp. no.	*Alternaria alterata*	*Helimenthosporium sativum*	*Fusarium moniliforme*
1	80.8	110.3	310.0
3	250.5	280.3	320.0
4	166.2	200.0	390.5
6a	7.2	8.3	11.2
6b	29.5	18.5	13.5
7	169.0	18.105	220.5
8	150.5	140.5	220.05
11	1000.0	1000.0	1000.0
12	1000.0	1000.0	1000.0
13	1000.0	1000.0	1000.0

Table 3

Germination percent of wheat grains treated the tested compounds and planted in soil infested with some fungi.

		% Germination in soil infested with	
Comp. no.	Concentration µg/mL	*Alternaria alterata*	*Fusarium moniliforme*
1	500	50.00	28.00
	1000	53.15	31.26
3	500	43.16	46.00
	1000	47.25	58.25
4	500	27.25	26.25
	1000	38.50	30.00
6a	500	63.50	79.5
	1000	80.15	89.5
6b	500	26.55	28.50
	1000	38.13	31.00
7	500	57.00	52.51
	1000	68.00	54.61
8	500	55.52	56.00
	1000	70.00	58.66
11	500	42.50	28.00
	1000	40.11	30.00
12	500	50.00	28.00
	1000	51.85	30.00
13	500	40.85	63.55
	1000	41.25	75.75
*TBZ	500	28.75	30.00

*Commercial thiobendazol-2-(4-thiazolyl)benzimidazole (TBZ) was used as reference standard.

CONCLUSION

In conclusion, compounds **6a, 6b, 7,** and **8** can be used as plant protective against fungal toxicities *A. alterata* and *F. moniliforme* by inhibition of mycelia growth compared with thiobendazol-2-(4-thiazolyl)benzimidazol as control. Also, compounds **6a** and **8** exhibited a higher germination percentage, whereas compounds **1, 6a, 11,** and **12** gave a good control at higher concentration. Thus, these compounds can be used as plant growth regulators.

EXPERIMENTAL

Melting points were determined with an electro thermal bib by Stuart Scientific Melting Point SMPI (UK). The IR spectra recorded for KBr discs were recorded on Perkins Lemer Spectrum RXI-FT-IR system 55529 (USA). ^{1}H-NMR spectra were determined for solution in deuterated (DMSO) with a Brucker NMR Advance DPX 400 MH (Germany) using TMS as an internal standard. Mass spectra were measured on a GCMS-Q 1000-Ex.

Spectrometer (Germany). Electronic absorption spectra were recorded on Shimadzu UV and visible 3101 PC spectrophotometer (Kyoto, Japan). Elemental analysis determination was performed in Microanalytical Center Cairo University-Egypt. ^{19}F-NMR spectra were recorded by using hexafluorobenzene, and the aromatic C–F exhibited at −122 to −124 ppm. Compound **5** was prepared according the reported method [35].

4-(4′-Fluorophenyl)-5-cyano-2-mercapto-3H-pyrionidin-6-one (1). Equimolar mixture of 4-Fluorobenzaldehyde and ethyl 2-cyanoethanoate was refluxed with thiourea (1:1 mmole) in sodium ethoxide, then stirred for 2 h and left over night to give the targets product 1 [22] by a high yield of 85% and crystallized from ethanol to give **1** as orange crystals. mp 209–210°C; IR (λ cm^{-1}) 3437 (NH), 3081 (Ar–CH), 2232 (C≡N), 1702 (C=O), 1552 (C=N), 386 (CN–C), 1250 (C–F), 1161 (C=S), 842 (P-substituted phenyl), 674 (C–F); ^{1}H-NMR (δ ppm): 8.00 (τ, 1H, NH), 7.75–7.73, 7.45–7.35, 7.24–7.21, 7.145–7.104 (each s, 4H, aromatic CH), 4.95–4.93, (s, 1H, SH); ^{13}C-NMR (δ ppm): 179.74 (C=S), 176.50 (C=O), 159.63–158.92 (C–F), 131.31–131.25, 130.38–130.75, 129.23–129.17, 128.95–128.89, 125.09–125.07 (aromatic carbons), 116.33–116.95 (C N), 115.59, 114.15 (C=N), 113.35 (CN), 90.62 (NCN), 77.67, 77.46, 77.25 (5,6–C=C of pyrimidine); *Anal.* Calcd. C, 53.44; H, 2.42; N, 17.00; S, 12.95; F, 7.96% for $C_{11}H_6N_3FSO$ (247). Found: C, 53.00; H, 2.30; N, 16.63; S, 12.69; F, 7.33%.

1H-2-hydrazino-4-aryl-5-cyano pyrimidin-6-one (2). Equimolar mixture of compound **1** (0.01 mol) and hydrazine hydrate (0.01 mol) was refluxed in ethanol (20 mL) for 6 h then cooled. The resulted solid was filtered and crystallized from EtOH to give **2** as deep yellow crystals. UV (λ $_{max}$ nm) 358.4; IR (λ cm^{-1}): 3400–3100 (b, NH, NH_2), 3067 (aromatic CH), 2228 (C≡N), 1667 (C=O), 1601 (deformation NH_2), 1551 (C=N), 1295 (C–N), 1245 (C–F), 830 (P-Substituted ring), 776 (C–F); ^{1}H-NMR (δ ppm): 8.62 (s, 1H, NH), 7.98–7.85, 7.75–7.65, 7.26–7.16, 7.16–7.022, (2H, 2H of aromatic protons), 3.61 (s, 2H, NH_2); ^{13}C-NMR (δ ppm): 176.43 (C=O), 160–158 (C–F), 131.32–131.26, 130.48–130.42, 130.23–130.21, 125.10 (six carbons of aryl pyridine), 115.93–115.79 (C≡N), 114.18 (C=N), 90.52 (N–C–N), 78.07, 77.85, 77.64 (4,5–CH=CH of pyrimidine); M^+/S (Int.%): 245 (246, M + 1, 13%), 214 (8.15), 148 (3.00), 95 (100), 52 (75); *Anal.* Calcd. C, 53.87; H, 3.26; N, 28.57; F, 7.75% for $C_{11}H_8FN_5O$ (245). Found: C, 53.51; H, 3.00; N, 28.33; F, 7.45%.

8-(4′-Fluorophenyl)-7-cyano-3-methyl-pyrimido[3,2-c][1,2,4-]triazin-4,6-dione (3). Equal amounts of **2** and diethyl oxalate (1:1 mmole) were refluxed with NaOH (2%, 100 mL) for 4 h then cooled and poured into ice HCl. The yielded solid was filtered and crystallized from EtOH to give **3** as yellowish crystals, mp 244–246°C, yield 68%; IR (υ_{cm}^{-1}) = 3128 (NH), 3080 (aromatic CH), 2937 (aliphatic CH), 2229 (C≡N) ,1735 (C=O), 1678 (C=O), 1587 (C=N), 1485 (deformation, CH_3), 1372 (cyclic NCN), 1252 (C–F), 847 (aromatic C=C), 664 (C–F); ^{1}H-NMR (δ ppm) = 12.87 (s, 1H, NH), 8.008–8.006, 7.993–7.981, 7.473–7.41, 7.193–7.167 (each s, 4H, aromatic protons), 2.258 (s, 3H, CH_3);

^{13}C-NMR (δ ppm) = 169.82 (C=O), 165.24–164.60 (C=O), 153 (C–F), 143.79, 132.01, 131.02, 130.96, 116.17,115.48 (aromatic carbons), 115.33 (C≡N), 89.71 (NCN), 77.69–77.67,77.46–77.24 (C=C of pyrimidine), 12.453 (CH_3); M/S (Int%) = 297 (298, M^{+1}, 13.11%), 269 (8.11), 241 (28.15), 187 (21.0), 95 (100), 92 (35.18); *Anal.* Calcd. C, 56.56; H, 2.69; N, 23.56; F, 6.39% for $C_{14}H_8N_5FO_2$ (297). Found: C, 56.33; H, 2.39; N, 23.11; F, 6.21%).

8-(4′-Fluorophenyl)-7-cyano-1,2-dihydro-pyrimido[3,2-c][1,2,4]triazin-3,4,6-trione (4). A mixture of **2** (0.01 mol) and sodium pyruvate (0.01 mol) in THF (50 mL) was refluxed for 4 h and then cooled. The solid product was filtered and crystallized from dioxan to give **4** as yellowish crystals, mp 278–280°C, yield 65%; IR (υ_{cm}^{-1}): 3250–3143 (b, NH–NH), 2210 (C≡N), 1670, 1610 (C=O,–CONH), 1588 (C=N), 1392 (NCN), 1239 (C–F), 837,811 (P-substituted ring), 659 (C–F); ^{1}H-NMR (δ ppm): 12.38, 12.14 (each s, 2H, NH, NH of 1,2,3-triazine), 8.17, 8.6, 8.01, 7.96, (each s, 4H of aryl, 7.19, 7.11 (coupling, doubled, doubled of F on CH protons); ^{13}C-NMR (δ ppm) = 169.61, 164.52, 163.11, 162.82, 161.80, 153.39, 146.20, 132.48, 130.72, 116.87–115.06, 86.37, 78.56–78.12. *Anal.* Calcd. C, 52.17; H, 2.06; N, 23.41; F, 6.35% for $C_{13}H_6N_5FO_3$ (299). Found: C, 51.89; H, 1.88; N, 23.11; F, 6.20%.

8-(4′-Fluorophenyl)-7-cyano-3-styryl-pyrimido[3,2-c][1,2,4]triazin-4,6-diones (6a,b). A mixture of **2** (0.01 mol) and compound **5a** and/or **5b** [35] (0.01 mol) in NaOH (5%, 100 mL) was refluxed for 2 h then cooled and poured into ice HCl. The yielded solid was filtered and crystallized from THF to give **6a** and/or **6b**, respectively, as yellow crystals, **6a** yield 70%; mp 170–172°C; **6a**: UV (λ$_{max}$ nm) = 375; IR (υ_{cm}^{-1}): 3180 (NH), 3080 (aromatic CH), 2923 (aliphatic CH), 2217 (C≡N), 1680, 1660 (2C=O), 1615 (C=C), 1586 (C=N), 1485 (deformation, CH=CH), 1384 (NCN), 1252 (C–F), 827, 815 (P-substituted ring), 756 (C–Cl), 680 (C–F); ^{1}H-NMR (δ ppm): 8.62–8.55 (d, 1H, NH), 8.16–7.97 (each s, 2H, coupling of aryl protons with –CH=CH– protons); 7.95–7.90, 7.89–7.74, 7.73–7.68, 7.60–7.58, 7.57 7.53, 7.18–7.16, 7.12–7.09, 6.73–6.67 (each s, 8H, aromatic protons); ^{13}C-NMR (δ ppm) = 167.42 (C=O), 153.29 (C–F), 146.53 (C–C),131.94, 131.92, 131.69, 131.41, 131.32, 130.83, 130.78, 130.56, 130.49, 130.42, 129.96, 129.88 (12 aromatic carbons), 127.36 (CN) (115.98–115.95, 115.83–115.81 (C=N), 115.59, 115.38, 115.19 (C–N), 111.53, 111.46 (CH=CH), 77.82–77.39 (NCN); M/S (Int%) = 420 (10.95), 392 (5.11), 364 (8.13), 336 (6.01), 198 (18.23), 297 (25.13), 160 (37.11), 137 (3.15), 95 (100), 111 (78.77), 66 (15.00); *Anal.* Calcd. C, 60.14; H, 2.62; N, 16.70; F, 4.53, Cl, 8.35% for $C_{21}H_{11}N_5FClO_2$ (419). Found: C, 60.01; H, 2.55; N, 16.49; F, 4.29; Cl, 8.21%.

6b, yield 65%; mp 128–130°C; IR (υ_{cm}^{-1}) = 3190 (NH), 3080 (aromatic CH), 2923, 2880 (aliphatic CH), 2099 (CN), 1680, 1676 (2C=O), 1584 (C=N), 1484, 1424 (deformation CH=CH), 1399 (NCN), 1259 (C–F), 928, 884, 808 (P-substituted ring), 756, 679 (C–Br, C–F); ^{1}H-NMR (δ ppm) = 9.973 (s, 1H, NH), 8.62–8.59 (each m, 2H, coupling CH=CH– with aryl protons), 7.96–7.95, 7.86–7.853, 7.851–7.842, 7.80–7.78, 7.44–7.43, 7.16–7.14, 6.73–6.69 (each s, 8H, aromatic protons), 3.69–3.68 (s, 1H, ,B CH=) 3.69–3.68 (s, 1H, α, CH =), 3.09–3.05 (s, 1H, β–CH=); ^{13}C-NMR (δ ppm) = 160.93 (C=O), 160.71 (C=O), 137.04 (C–F), 132.48–132.41 (C–Br), 130.91–130.86, 130.60–130.54, 130.48, 130.25–130.23, 130.95–130.174, 129.739–129.687, 129.734, 129.06–129.05, 128.93–128.59 (12 aromatic carbons), 116.04, 116.02 (CN), 115.90–115.88 (α CH=), 115.44–115.30 (β–CH=), 111.59 (CN); *Anal.* Calcd. C, 54.31; H, 2.37; N, 16.50; F, 4.09, Br, 17.24% for $C_{21}H_{11}N_5FBrO_2$ (464). Found: C, 54.02; H, 2.15; N, 16.35; F, 3.85; Br, 17.01%.

8-(4′-Fluorophenyl)-7-cyano-1H-3-(2′-aminophenyl)pyrimido[3,2-c][1,2,4]triazin-4,6-dione (7). A mixture of **2** (0.01 mol) and isatin (0.01 mol) in NaOH (5%, 100 mL) was refluxed for 2 h then cooled and poured into ice HCl. The yielded solid was filtered and crystallized from EtOH to give **7** as yellowish crystals, mp 277–279°C, yield 75%.

UV (λ_{max} nm) = 370; IR (υ_{cm}^{-1}) = 3199 (NH), 2224 (C≡N), 1680, 1660 (2C=O), 1621 (deformation NH_2), 1607, 1541 (C=N), 1384 (NCN), 1237 (C–F), 893, 843, 815, 785 (O,P-substituted ring), 656 (C–F); ^{1}H-NMR (δ ppm) = 10.70 (s, 1H, NH), 7.55, 7.67–7.61, 7.34–7.31, 7.21–7.208, 7.202–7.197, 7.095–7.077, 6.967–6.90 (each m, 7H, aromatic), 6.89–6.88 (s, 1H, NH of NH_2), 3.677–3.651 (s, 2H, NH_2); ^{13}C-NMR (δ ppm) = 164.23 (C=O), 145.16–145.14 (C–F), 133 (C–NH_2), 128.76–122.5 (aromatic carbons), 116 (NCN), 111–113 (CN), 78.09–77.666 (carbons of 1,2, 4-triazine), 66 (C=N); *Anal.* Calcd. C, 60.96; H, 2.94; N, 22.24; F, 5.08% for $C_{19}H_{11}N_6FO_2$ (374). Found: C, 60.59; H, 2.88; N, 22.05; F, 4.95%.

Trifluoroacetyl-3-(trifluoroacetyl amino phenyl-8-4-fluoro-phenyl-7-cyano-pyrimido-[3,2-c][1,2,4] triazin-4,6-dione (8). A mixture of compound **7** (0.285 g and trifluoroacetic anhydride (2 mL) in THF (20 mL) was reflux for 24 h and cooled. The resulted solid filtered off and crystallized from dioxan to give **8** as yellowish crystals, yield 70%; mp 239–240°C; IR (υ_{cm}^{-1}): 3200 (NH_2), 2215 (C≡N), 1700, 1626 (C=O), 1598 (CONH), 1585 (C=N), 1316 (NCN), 1295, 1224 (C–F), 805, 825 (O,P-substituted ring), 707 (C–F); ^{1}H-NMR (δ ppm): 12.8, 10.8 (each s, NH↔OH), 7.88–6.77 (m, 8H, aryl protons); ^{13}C-NMR (δ ppm) = 184.50, 159.40, 145.08, 138.33, 128.58, 128.32, 124.79, 122.87, 120.71, 115.95–111.06, 78.51; *Anal.* Calcd. C, 49.64; H, 1.61; N, 23.92; F, 15.10 for $C_{23}H_9N_6F_7O_4$ (556). Found: C, 49.39; H, 1.51; N, 23.55; F, 14.88%.

8-(4′-Fluorophenyl)-3′-[2-(4′-fluorobenzoyl)aminophenyl]-7-cyano–pyrimido[3,2-c] [1,2,4] triazin-4,6-dione (9). Equimolar mixture of compound **7** and 4-fluorobenzoylchloride in DMF (10 mL) was warmed for 30 min, cooled then poured into ice. The solid thus obtained filtered off and crystallized from dioxan to give **9** as reddish crystals, yield 60%; mp 221–223°C; IR (υ_{cm}^{-1}): 3220–3190 (b, NH), 3020 (Ar CH), 2220 (C≡N) 1660, 1590 (C=O, CONH), 1250 (C–F),900,800–820 (aryl CH), 660 (C–F); ^{1}H-NMR (δ ppm) 10.8 (s, 1H, OH), 8.18–6.89 (m, 8H, aryl protons); ^{13}C-NMR (δ ppm) = 163.0 162.12, 145.16, 133.94, 130.76, 130.71, 129.54, 128.74,125.17, 122.57, 121.43, 116.04,110.37, 78.14–77.55, 66.79; *Anal.* Calcd. C, 62.90; H, 2.82; N, 16.93; F, 7.66%. For $C_{26}H_{14}N_6F_2O_3$ (496). Found: C, 62.49; H, 2.58; N 16.61; F 7.35%.

11-(4′-Fluorophenyl)-10-cyano-1H-pyrimido[3,2-c][1,2,4]triazino [6,5-b]indole (10). A mixture of **2** (0.01 mol) and isatin (0.01 mol) in DMF was refluxed for 2 h then cooled. The yielded solid was filtered and crystallized from dioxan give **10** as brown crystals, yield 65%; mp 304–305°C; UV (λ_{max} nm): 373; IR (υ_{cm}^{-1}) = 3201 (NH), 2225 (C≡N), 1666 (C=O), 1608, 1541 (C=N), 1384 (cyclic NCN), 1238 (C–F), 894, 843, 815, 785 (O,P-substituted ring), 656 (C–F); MS (Int%) = 356 (11.18), 328 (38.12), 300 (13.00), 274 (33.43), 179 (45.81), 95 (100); *Anal.* Calcd. C, 64.04; H, 2.52; N, 23.59; F, 5.33%. For $C_{19}H_9N_6FO$ (356). Found: C, 63.74; H, 2.35; N, 23.29; F, 5.21%.

8-(4′-Fluorophenyl)-7-cyano-1,2,3,4-tetrahydro-pyrimido[3,2-c][1,2,4]triazin-4,6-dione (11). Equimolar mixture of **2** and chloroacetic acid in NaOH (5% 1:1, 100 mL) was refluxed for 4 h then cooled and poured into ice HCl. The produced solid was filtered and crystallized from THF to give **11** as yellowish crystal, yield 66%; mp 167–168°C.

IR (υ_{cm}^{-1}) = 3366 (NH), 3152 (NH), 3090 (aromatic CH), 2890 (aliphatic CH), 2217 (C≡N), 1668 (C=O), 1639 (CONH), 1612 (C=N), 1556 (C=N), 1489 (deformation CH), 1395 (cyclic NCN), 1238 (C–F), 842, 811 (P-substituted ring), 779 (C–F); ^{1}H-NMR (δ ppm): 12.21–12.06 (s, 1H, NH), 11.70 (s, 1H, NH), 7.98–7.95, 7.88–7.87, 7.77–7.45, 7.24–7.10 (each s, 4H, aromatic protons), 5.41 (s, 1H, OH of –$COCH_2$), 3.065–2.97 (s, 2H, NH); ^{13}C-NMR (δ ppm): 170.01 (C=O), 161.76 (C=O), 153.23 (C–F), 132.15, 130.86–130.81, 130.04–129.71, 129.23, 129.15–129.07, 129–016–129.91 (6C aromatic), 116 (C≡N), 115.78, 115.23 (C=N), 87.24 (C–N), 77.83, 77.62, 77.40 (carbons of 1,2,4-triazine), 40.14–39.58 (aliphatic carbons); *Anal.* Calcd. C, 54.73; H, 2.2.80; N, 24.56; F, 6.69%. For $C_{13}H_8N_5FO_2$ (285). Found: C, 54.55; H, 2.39; N, 24.28; F 6.51%.

8-(4′-Fluorophenyl)-7-cyano-1,2,3,4-tetrahydro-pyrimido[3,2-c][1,2,4]triazin-3,6-dione (12). A mixture of **2** (0.01 mol) and chloroacetyl chloride (0.01 mol) in DMF (100 mL) was warmed for 1 h then cooled. The obtained solid was filtered and crystallized from dioxan to give **12** as faint yellow crystals, yield 60%; mp 254–255°C; UV (λ_{max} nm): 351; IR (υ_{cm}^{-1}): 3400–3200 (b, OH, NH–NH), 2210 (C≡N), 1668 (C=O), 1591 (C=N), 1494, 1410 (deformation CH_2), 1392 (NCN), 1254 (C–F), 857, 815, 786 (P-substituted ring), 658 (C–F); ^{1}H-NMR (δ ppm) = 12.32 (s, 1H, NH, 1,2,4-triazine), 11.43 (s, 1H, NH, 1,2,4-triazine), 8.61–8.55, 7.96–7.90, 7.85–7.51, 7.48–7.37, 7.18–7.098 (each s, aromatic protons), 3.68–3.66 (m, 2H, CH_2), 2.900–2.58 (each s, 2H, N–CH_2–COH); *Anal.* Calcd. C, 54.73; H, 2.80; N, 24.56; F, 6.69% for $C_{13}H_8N_5FO_2$ (285). Found: C, 54.33; H, 2.51; N, 24.13; F, 6.48%.

8-(4′-Fluorophenyl)-3-arylidene-7-cyano-1,2-dihydro-pyrimido [3,2-c][1,2,4]triazin-4,6-dione (13). Equimolar mixture of **11** and p-chlorobenzaldehyde in EtOH (50 mL)-piperidine drops was refluxed for 2 h then cooled and poured into ice. The produced solid was filtered and crystallized from dioxan to give **13** as yellowish crystals, yield 65%; mp 256–258°C; UV (λ_{max} nm):360; IR (υ_{cm}^{-1}): 3300,3143 (NH, NH), 2209 (C≡N), 1663 (C=O), 1640 (C=O), 1586 (C=N), 1487, 1458 (deformation CH=), 1391 (cyclic NCN), 1256 (C–F), 862, 826, 812, 780 (P-substituted ring), 680 (C–Cl), 656 (C–F); ^{1}H-NMR (δ ppm): 8.61, 8.59 (each s, 1H, 1H, NH, NH), 8.12–8.10, 8.097–8.08 (m, 2H of coupling aryl with α and β carbons of styryl), 7.999, 7.95–7.92, 7.89–7.82, 7.74–7.65, 7.60–7.55, 7.248–7.229, 7.214–7.200, 7.176–7.143 (each s, 8H, aromatic protons), 3.82–3.45, 3.229–3.024 (each m, 2H, CH=CH); ^{13}C-NMR (δ ppm): 160.81 (C=O), 160.75 (C=O), 160.50 (C–F), 132.82 (C–Cl), 132.03, 131.94–131.92, 131.16–131.10, 130.56–130.50, 130.48–130.43, 130.25, 129.896, 129.83, 128.96, 128.51, 125.52 (12 carbons of aromatic rings), 115.98–115.955, 115.83–115.81 (C=C of pyrimidine), 115.63 (6C of 1,2,4-triazine), 115.49 (C≡N), 77.96–77.53 (NCN of 1,2,4-triazine), 56.54 (C=N), 36.29, 33.54 (two aliphatic carbons (=CH–Ar); *Anal.* Calcd. C, 58.82; H, 2.69; N, 17.15; F, 4.65; Cl, 8.82% for $C_{20}H_{11}N_5FClO_2$ (408). Found: C, 58.52; H, 2.34; N, 16.88; F, 4.35; Cl, 8.59%.

8-(4′-Fluorophenyl)-4-aryldene-7-cyano-1,2-dihydro-pyrimido [3,2-c][1,2,4]triazin-3,6-dione (14). Equimolar mixture of **12** and p-chlorobenzaldehyde in EtOH (100 mL)-piperidine drops was refluxed for 2 h then cooled and poured into ice. The produced solid was filtered and crystallized from ethanol to give **14** as faint yellow crystals, yield 58%; mp 231–232°C.; IR (υ_{cm}^{-1}): 3500–3400 (**b**, NH, OH), 3020 (Ar–CH), 2910 (R–CH), 2180 (C≡N), 1708 (C=O), 1590, 1580 (C=C, C=N), 1380 (NCN), 1240 (C–F), 840, 810 (Ar–CH), 710, 660 (C–Cl and C–F); *Anal.* Calcd. C, 58.82; H, 2.69; N, 17.15; F, 4.65; Cl, 8.82% for $C_{20}H_{11}N_5FClO_2$ (408). Found: C, 58.55; H, 2.39; N, 16.89; F, 4.41; Cl, 8.55%.

8-(4′-Fluorophenyl)-7-cyano-4-hydroxy-pyrimido[3,2-c][1,2,4] triazin-6-one (15) and 8-(4′-fluorophenyl)-7-cyano-3-hydroxy-pyrimido[3,2-c][1,2,4]triazin-6-one (16). A mixture of compounds **11** and/or **12** (0.2 g) and $FeCl_3$ (0.2 g) in MeOH (20 mL) was refluxed for 3 h then filtered while hot. The solid thus obtained from filtrates filtered off and crystallized from dioxan to give **15** and **16** as deep brown crystals.

15, yield 68%; mp 154–155°C; UV (λ_{max} nm): 375 nm; IR (υ_{cm}^{-1}): 3450 (OH), 2220 (C≡N), 1680 (C=O), 1250 (C–F)**;** *Anal.* Calcd. C, 55.12; H, 2.12; N, 24.73; F, 6.71% for $C_{13}H_6N_5FO_2$ (283). Found: C, 54.89; H, 1.98; N, 24.35; F, 6.0% soluble in NaOH solution in cold water.

16, yield 66%; mp 228–230°C; UV (λ_{max} nm): 378 nm; IR (υ_{cm}^{-1}): 3470 (OH), 2195 (C≡N), 1670 (C=O), 1230 (C–F)**;** *Anal.* Calcd. C, 55.12; H, 2.12; N, 24.73; F, 6.71% for $C_{13}H_6N_5FO_2$ (283). Found: C, 54.69; H, 1.88; N, 24.49; F, 6.55% soluble in NaOH solution in cold water.

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