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SYNTHESIS AND ANTIMICROBIAL EVALUATIONS OF NOVEL SPIRO CYCLIC 2-OXINDOLE DERIVATIVES OF *N*-(1*H*-PYRAZOL-5-YL)HEXAHYDROQUINOLINE DERIVATIVES

Said Ahmed Soliman Ghozlan, Muhammed Ali Ramadan, Amr Mohamed Abdelmoniem,* and Ismail Abdelshafy Abdelhamid*

Department of Chemistry, Faculty of Science, Cairo University, 12613 Giza, A. R. Egypt

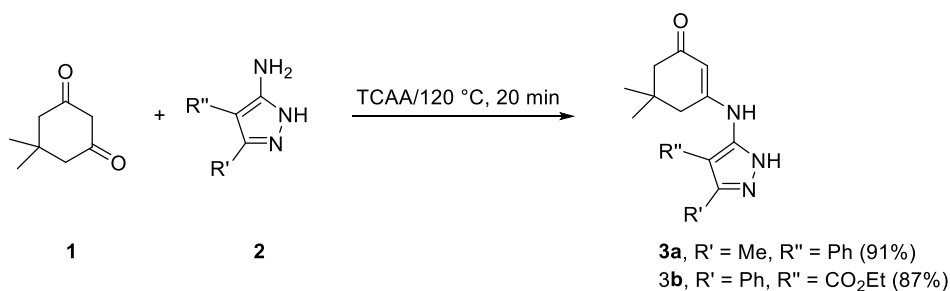
Abstract – A novel series of interesting spiro cyclic 2-oxindole derivatives of *N*-(1*H*-pyrazol-5-yl)hexahydroquinoline derivatives were prepared *via* the versatile readily accessible cyclic β -enaminones incorporating pyrazole. Antimicrobial evaluations were performed on the prepared compounds. Most of these compounds exhibited high to moderate antimicrobial activity.

INTRODUCTION

The Michael addition reaction has been widely used in organic synthesis for its C–C bond forming ability. Useful application of a tandem Michael addition is seen in the total synthesis of the antimicrobial compound Claenone.¹ In this respect, Yamada *et al.*¹ managed to construct a norbornane ring using two sequential Michael additions. The Michael reaction is also used in other reactions. The best known example is the Robinson annulation, where the Michael addition occurs as the first step.² This sequence of Michael addition followed by intramolecular Aldol has proved to be one of the most important chemical reactions introduced into in the area of steroid chemistry as shown in Woodward's synthesis of cortisone.² In addition, the spiro-oxindole ring system is one of the most distinguished heterocyclic ring systems, which constitutes the core structural element of many biologically active molecules that received an extensive synthetic interest.³⁻¹⁷ Moreover, quinoline and its derivatives exist in a variety of biologically significant compounds possessing anticancer,¹⁸ antioxidants,¹⁹ anti-inflammatory²⁰ and antimicrobial activities.²¹⁻²³ Considering the versatile bioactivities of the two structures of spiro-oxindole and 2-amino-tetrahydroquinolin-5-one, we expect that the integration of the two scaffolds into a spiro-oxindole incorporating 2-amino-tetrahydroquinolin-5-one can result in the discovery of new active drugs.

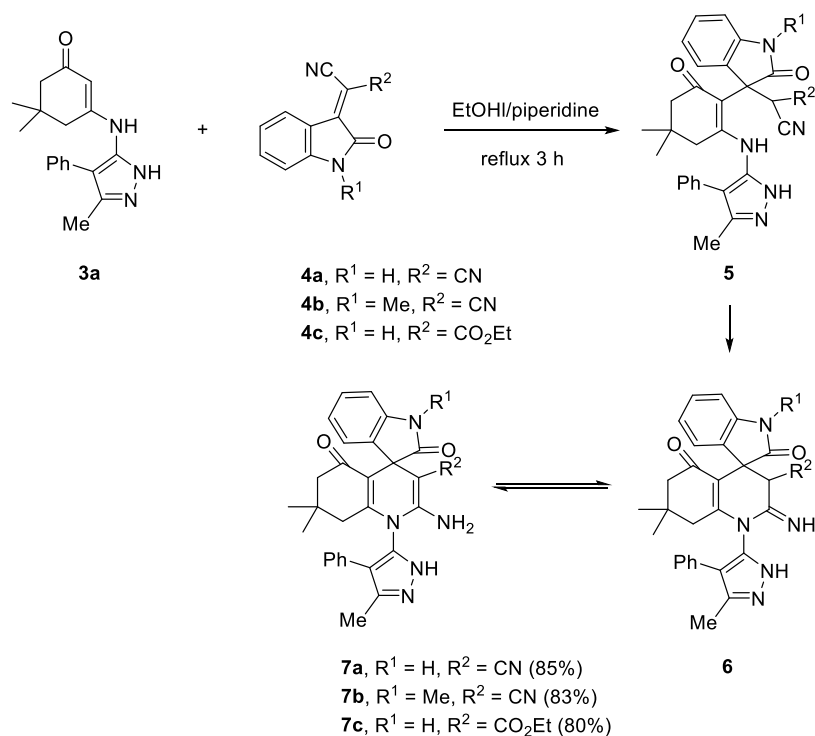
RESULTS AND DISCUSSION

In continuation to our interest on the chemistry of enamines²⁴⁻²⁷ as well as Michael addition reactions,^{10,28-32} we report herein a new series of novel spirocyclic 2-oxindole derivatives of *N*-(1*H*-pyrazol-5-yl)hexahydroquinoline derivatives *via* the cyclocondensation reaction of 3-cyanomethylidene-2-oxindoles and 3-(substituted pyrazol-5-yl)-5,5-dimethylcyclohex-2-enone. The required cyclic β -enaminoketones incorporating pyrazole moiety **3**, were prepared *via* the reaction of the dimedone **1** with the aminopyrazole derivatives **2** using a catalytic amount of trichloroacetic acid as a catalyst under solvent-free conditions.



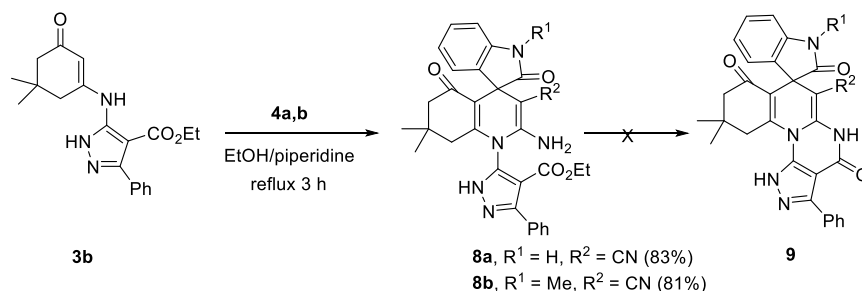
Scheme 1. Synthesis of enamines **3a,b**

The possibility of [3+3] atom combination of the prepared β -enaminones **3a,b** with 3-cyanomethylidene-2-oxindoles **4a-c** through Michael addition reaction was studied. Thus, the reaction of compound **3a** with compounds **4a-c** in the presence of piperidine resulted in the formation of spiroquinolines **7a-c**. Compounds **7** were formed due to the initial addition of the β -CH of **3a** to the activated double bond in **4**, to yield Michael adducts **5** that readily cyclized into **6** that tautomerized into **7**. (cf. Scheme 2). The structures of the novel hexahydroquinolines **7** were assigned based on the spectroscopic methods. Thus, IR data of **7c** indicated the absence of CN group. It also revealed two absorption bands at ν 3401 and 3246 cm^{-1} due to NH₂ and two NH. The bands at ν 1667 and 1635 cm^{-1} are assigned for the two carbonyl groups. ¹H NMR spectrum of **7c** revealed three singlet signals at δ 0.4, 0.82 and 2.41 ppm for three methyl groups. The ester group appears as triplet at δ 0.85 ppm and quartet at δ 3.68 ppm with $J = 7.2$ Hz. In addition, it displayed a prominent signal at δ 9.89 for NH group. The NH₂ and aromatic protons appear as multiplets in the area δ 6.62-7.53 ppm. All other signals appear at their expected position. Furthermore, ¹³C NMR was found to be in agreement with the proposed structure. The key signal at δ 32.3 ppm is assigned for the spiro carbon. The signals at δ 161.9 and 196.9 ppm are assigned to the two carbonyl groups. All other carbon signals appeared at their expected positions.



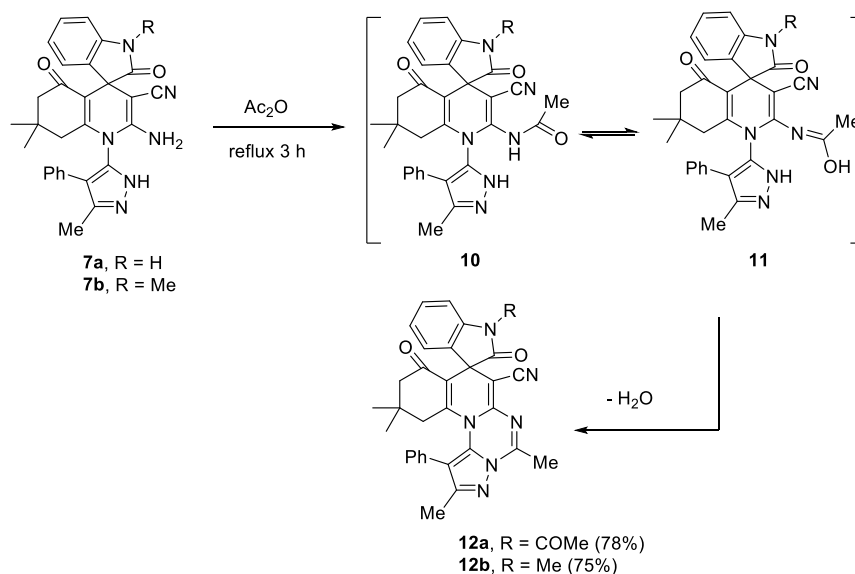
Scheme 2. Synthesis of spiro[indoline-3,4'-quinoline] derivatives **7a-c**

Conducting the above-mentioned reaction under the same conditions on the cyclic enamines **3b**, carrying ester group on the pyrazole ring, leads to the formation of **8a,b**. Trials to affect a further cyclization to prepare compounds **9** did not succeed.



Scheme 3. Synthesis of spiro[indoline-3,4'-quinoline] derivatives **8a,b**

Quinolines **7a,b** were used as precursors for the synthesis of the spiro-hexacyclic products. Thus, boiling compounds **7a,b** in acetic anhydride for a long period results in the formation of **10** that readily tautomerize into **11** followed by water removal to give the spiro[indoline-3,8'-pyrazolo[1',5':5,6][1,3,5]triazino[1,2-*a*]quinoline] derivatives **12a,b**, respectively. The structures of compounds **12** were confirmed based on their spectral data. The analyses clearly indicate the absence of the NH and NH₂ groups. It also revealed the presence of additional methyl group.



Scheme 4. Synthesis of spiro[indoline-3,8'-pyrazolo[1',5':5,6][1,3,5]triazino[1,2-*a*]quinoline] derivatives **12a,b**

ANTIMICROBIAL ACTIVITY

The antibacterial activity of the synthesized compounds was screened against the Gram-positive bacteria: *Streptococcus pneumoniae* and *Bacillus subtilis*, and the Gram-negative bacteria: *Pseudomonas aeruginosa* and *Escherichia coli* using diffusion agar medium. The antifungal activity of the compounds was tested against *Candida albicans* and *Aspergillus fumigatus* using diffusion agar medium. The minimum inhibitory concentration (MIC) was carried out using microdilution susceptibility method.³³ Ampicillin and Gentamycin were used as standard antibacterial drugs. Amphotericin B was used as a standard antifungal drug. The observed data of the antimicrobial activity of compounds and control drugs are given in Table 1. It is clear that, however compound **12b** showed the lowest activities, most of the other screened samples, showed significant antibacterial and antifungal activities (Table 1). The MIC values were determined as the lowest concentration that completely inhibited visible growth of the microorganisms. The investigation of antibacterial screening (Table 2) reveals that most of the compounds showed excellent antibacterial activity at MIC 0.49-3.9 $\mu\text{g/mL}$ in DMSO. Amongst all the synthesized quinoline derivatives, compounds **7a**, **12a** and **8b** exhibited good activities against *Bacillus subtilis* (MIC 0.24, 0.49, 0.98 $\mu\text{g/mL}$) and *Streptococcus pneumoniae* (MIC 0.49, 0.98 1.95 $\mu\text{g/mL}$) and *Escherichia coli* (MIC: 0.49, 0.98, 1.95 $\mu\text{g/mL}$). On the other hand, they showed moderate activities against *Pseudomonas aeruginosa* (MIC: 1.95, 3.9, 3.9 $\mu\text{g/mL}$). Compound **7c** displayed moderate activities towards *Bacillus subtilis*, *pseudomonas aeruginosa* and *Escherichia coli* (MIC 3.9 $\mu\text{g/mL}$). Compounds **8a** and **7b** revealed moderate activity towards *Streptococcus pneumoniae*, *Bacillus subtilis* and *Escherichia coli*. The antifungal screening study also revealed that the newly synthesized compounds

showed moderate-to-good inhibition against *Aspergillus fumigatus*. However, all the tested compounds showed no bioactivities towards *Candida albicans*.

Table 1. Antimicrobial activity of a novel series of spirocyclic 2-oxindole derivatives of 2-amino-tetrahydroquinolin-5-one

Compounds	Inhibition Diameter Zone (mm)					
	Fungi		Gram Positive Bacteria		Gram Negative Bacteria	
	<i>Aspergillus fumigatus</i>	<i>Candida albicans</i>	<i>Streptococcus pneumoniae</i>	<i>Bacillus subtilis</i>	<i>Pseudomonas aeruginosa</i>	<i>Escherichia coli</i>
St.	Amphotericin B		Ampicillin		Gentamicin	
	23.7	25.4	23.8	32.4	17.3	19.9
7a	24.3	0	23.5	28.3	21.3	25.2
7b	20.3	0	19.3	21.6	17.3	19.4
7c	19.3	0	18.3	20.4	19.3	20.1
8a	19.3	0	20.6	22.6	14.3	18.6
8b	21.3	0	20.8	23.4	18.9	21.3
12a	22.6	0	21.9	25.7	20.4	23.6
12b	12.6	0	14.6	16.2	11.2	13.7

Table 2. The minimum inhibitory concentration ($\mu\text{g/mL}$)

Compounds	The minimum inhibitory concentration (MIC) ($\mu\text{g/mL}$)					
	Fungi		Gram Positive Bacteria		Gram Negative Bacteria	
	<i>Aspergillus fumigatus</i>	<i>Candida albicans</i>	<i>Streptococcus pneumoniae</i>	<i>Bacillus subtilis</i>	<i>Pseudomonas aeruginosa</i>	<i>Escherichia coli</i>
St.	Amphotericin B		Ampicillin		Gentamicin	
	0.98	0.49	0.98	0.24	15.63	3.9
7a	0.49	>100	0.49	0.24	1.95	0.49
7b	3.9	>100	3.9	0.98	15.63	3.9
7c	3.9	>100	7.81	3.9	3.9	3.9
8a	3.9	>100	1.95	0.98	62.5	3.9
8b	1.95	>100	1.95	0.98	3.9	1.95
12a	0.98	>100	0.98	0.49	3.9	0.98

In conclusion, the Michael reaction of β -enaminones **3a,b** with 3-cyanomethylidene-2-oxindoles **4a-c** represents a versatile tool for the synthesis of various spirocyclic structures combining both oxindole and hexahydroquinoline fragments. The formation of spiro-polycondensed derivatives **12a,b** was also achievable by the action of acetic anhydride. Preliminary antimicrobial evaluation tests indicate that the majority of the synthesized compounds showed promising antimicrobial activities.

EXPERIMENTAL

Melting points were measured with a Stuart melting point apparatus and are uncorrected. The IR spectra were recorded using a FTIR Bruker–vector 22 spectrophotometer as KBr pellets. The ^1H and ^{13}C NMR

spectra were recorded in DMSO- d_6 and $CDCl_3$ as solvent on Varian Gemini NMR spectrometer at 400 MHz and 100 MHz, respectively, using TMS as internal standard. Chemical shifts are reported as δ values in ppm. Mass spectra were recorded with a Shimadzu GCMS-QP-1000 EX mass spectrometer in EI (70 eV) model. The elemental analyses were performed at the Micro analytical center, Cairo University.

Synthesis of enamines (3a,b). A mixture of dimedone **1** (1 g, 7.14 mmol) and 3-methyl-4-phenyl-1*H*-pyrazol-5-amine **2a** (1.24 g, 7.17 mmol) or ethyl 5-amino-3-phenyl-1*H*-pyrazole-4-carboxylate **2b** (1.65 g, 7.14 mmol) was heated in an oil bath at 120 °C in presence of trichloroacetic acid (0.2 g, 1.23 mmol) for 20 min. The oily residue was extracted with $CHCl_3$ (25 mL). The solvent was removed at reduced pressure and the crude solid was crystallized from EtOH.

5,5-Dimethyl-3-((3-methyl-4-phenyl-1*H*-pyrazol-5-yl)amino)cyclohex-2-enone (3a): Yellow crystals (1.92 g, 91%), Mp 248-250 °C, IR (KBr): ν 3437, 3226 (br, 2NH), 1712 (CO_2Et), 1535 (CO) cm^{-1} , 1H NMR (400 MHz, DMSO- d_6): δ 0.94 (s, 6H, 2 CH_3), 1.94 (s, 2H, CH_2), 2.26 (s, 2H, CH_2), 2.30 (s, 3H, pyrazole CH_3), 5.01 (s, 1H, =CH), 7.21-7.39 (m, 5H, ArH), 8.49 (br s, 1H, enamine NH), 12.58 (br s, 1H, pyrazole NH) ppm, ^{13}C NMR (100 MHz, $CDCl_3$): δ 10.3 (CH_3), 27.9 (2 CH_3), 32.3 (C), 41.8 (CH_2), 50.0 (CH_2), 99.2 (C), 112.0 (C), 125.9 (C), 128.1 (CH), 128.2 (CH), 132.0 (C), 138.2 (C), 152.9 (C), 161.9 (C), 196.9 (C) ppm, MS (EI, 70 eV): m/z (%) 295 ($[M^+]$, 78), 280 (99), 267 (22), 252 (25), 239 (59), 211 (100), 194 (32), 115 (32), 77 (20), Anal. Calcd for $C_{18}H_{21}N_3O$: C, 73.19; H, 7.17; N, 14.23. Found: C, 73.06; H, 7.12; N, 14.11.

Ethyl 5-((5,5-dimethyl-3-oxocyclohex-1-en-1-yl)amino)-3-phenyl-1*H*-pyrazole-4-carboxylate (3b): Yellow crystals (2.14 g, 85%), Mp 222-224 °C, IR (KBr): ν 3429 (br, 2NH), 1675 (CO_2Et), 1546 (CO) cm^{-1} , 1H NMR (400 MHz, DMSO- d_6): δ 0.99 (s, 6H, 2 CH_3), 1.05 (t, 3H, $J = 7.2$ Hz, CH_3CH_2), 2.10 (s, 2H, CH_2), 2.46 (s, 2H, CH_2), 4.12 (q, 2H, $J = 7.2$ Hz, CH_2CH_3), 6.51 (s, 1H, =CH), 7.48-7.62 (m, 5H, ArH), 8.53 (br s, 1H, enamine NH), 13.30 (br s, 1H, pyrazole NH) ppm, ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.9 (CH_3), 28.3 (CH_3), 32.9 (C), 44.2 (CH_2), 50.2 (CH_2), 60.3 (CH_2), 97.5 (C), 104.3 (CH), 127.8 (CH), 128.7 (C), 129.5 (CH), 129.8 (CH), 145.9 (C), 152.3 (C), 156.3 (C), 165.5 (C), 200.3 (C) ppm, MS (EI, 70 eV): m/z (%) 353 ($[M^+]$, 21), 338 (20), 308 (20), 238 (21), 231 (36), 217 (20), 185 (100), 181 (33), 128 (35), 96 (23), 77 (37), Anal. Calcd for $C_{20}H_{23}N_3O_3$: C, 67.97; H, 6.56; N, 11.89. Found: C, 67.88; H, 6.47; N, 11.82.

General method for synthesis of compounds (7a-c). A mixture of enamine **3a** (0.30 g, 1 mmol) and 3-cyanomethylidene-2-oxindole derivatives **4a-c** (1 mmol) was heated at reflux in absolute EtOH (15 mL) in presence of piperidine (0.2 mL) for 3 h. The crude solid was crystallized from EtOH-dioxane (3:1, v/v).

2'-Amino-7',7'-dimethyl-1'-(3-methyl-4-phenyl-1*H*-pyrazol-5-yl)-2,5'-dioxo-5',6',7',8'-tetrahydro-1'*H*-spiro[indoline-3,4'-quinoline]-3'-carbonitrile (7a): Yellow crystals (0.42 g, 85%), Mp 248-250 °C, IR (KBr): ν 3448, 3398, 3311, 3204 (2NH and NH₂), 2187 (CN), 1706, 1638 (2CO) cm⁻¹, ¹H NMR (400 MHz, DMSO-*d*₆): δ 0.44 (s, 3H, CH₃), 0.82 (s, 3H, CH₃), 1.71 (m, 2H, CH₂), 2.08 (m, 2H, CH₂), 2.44 (s, 3H, pyrazole CH₃), 5.62 (br s, 2H, NH₂), 6.70-7.57 (m, 9H, ArH), 10.19 (br s, 1H, oxindole NH), 13.29 (br s, 1H, pyrazole NH) ppm; ¹³C NMR (100 MHz, CDCl₃): δ 10.3 (CH₃), 25.3 (CH₃), 28.8 (CH₃), 31.4 (C), 48.2 (CH₂), 49.3 (C), 56.3 (CH₂), 61.2 (C), 108.8 (C), 110.1 (C), 116.1 (CH), 118.4 (C), 121.1 (CH), 122.5 (CH), 126.9 (CH), 127.3 (C), 128.0 (CH), 128.5 (CH), 130.4 (CH), 135.9 (C), 138.1 (C), 140.9 (C), 150.9 (C), 151.5 (C), 179.6 (C), 193.5 (C) ppm; MS (EI, 70 eV): *m/z* (%) 490 ([M⁺], 39), 482 (29), 466 (41), 394 (46), 295 (37), 143 (100), 128 (48), 75 (54), Anal. Calcd for C₂₉H₂₆N₆O₂: C, 71.00; H, 5.34; N, 17.13. Found: C, 71.13; H, 5.29; N, 17.08.

2'-Amino-1,7',7'-trimethyl-1'-(3-methyl-4-phenyl-1*H*-pyrazol-5-yl)-2,5'-dioxo-5',6',7',8'-tetrahydro-1'*H*-spiro[indoline-3,4'-quinoline]-3'-carbonitrile (7b): Yellow crystals (0.42 g, 83%), Mp 248-250 °C, IR (KBr): ν 4377, 3404, 3237 (br, 2NH and NH₂), 2187 (CN), 1720, 1632 (2CO) cm⁻¹, ¹H NMR (400 MHz, DMSO-*d*₆): δ 0.43 (s, 3H, CH₃), 0.85 (s, 3H, CH₃), 1.71 (m, 2H, CH₂), 2.06 (m, 2H, CH₂), 2.39 (s, 3H, pyrazole CH₃), 3.11 (s, 3H, NCH₃), 5.73 (br s, 2H, NH₂), 6.74-7.57 (m, 9H, ArH), 13.25 (br s, 1H, pyrazole NH) ppm; ¹³C NMR (100 MHz, CDCl₃): δ 9.8 (CH₃), 12.1 (CH₃), 24.6 (CH₃), 28.2 (CH₃), 30.3 (C), 47.9 (CH₂), 48.7 (C), 49.5 (CH₂), 57.8 (CH₂), 78.4 (C), 106.9 (C), 111.9 (C), 115.5 (CH), 119.2 (CH), 121.5 (CH), 125.7 (CH), 126.2 (C), 127.2 (CH), 127.3 (C), 127.8 (CH), 129.9 (CH), 136.4 (C), 142.4 (C), 149.5 (C), 151.8 (C), 167.8 (C), 181.3 (C), 192.6 (C) ppm; MS (EI, 70 eV): *m/z* (%) 504 ([M⁺], 22), 499 (15), 384 (38), 356 (47), 305 (62), 117 (100), Anal. Calcd for C₃₀H₂₈N₆O₂: C, 71.41; H, 5.59; N, 16.66. Found: C, 71.33; H, 5.48; N, 16.58.

Ethyl 2'-amino-7',7'-dimethyl-1'-(3-methyl-4-phenyl-1*H*-pyrazol-5-yl)-2,5'-dioxo-5',6',7',8'-tetrahydro-1'*H*-spiro[indoline-3,4'-quinoline]-3'-carboxylate (7c): Yellow crystals (0.43 g, 80%), Mp 248-250 °C, IR (KBr): ν 3401, 3246 (br, 2NH and NH₂), 1721 (CO₂CH₂CH₃), 1667, 1635 (2CO) cm⁻¹, ¹H NMR (400 MHz, DMSO-*d*₆): δ 0.40 (s, 3H, CH₃), 0.82 (s, 3H, CH₃), 0.85 (t, 3H, *J* = 7.2 Hz, CO₂CH₂CH₃), 1.68 (m, 2H, CH₂), 2.05 (m, 2H, CH₂), 2.41 (s, 3H, pyrazole CH₃), 3.68 (q, 3H, *J* = 7.2 Hz, CO₂CH₂CH₃), 6.62-7.53 (m, 11H, ArH and NH₂), 9.89 (br s, 1H, oxindole NH), 13.26 (br s, 1H, pyrazole NH) ppm; ¹³C NMR (100 MHz, CDCl₃): δ 9.6 (CH₃), 25.2 (CH₃), 28.0 (CH₃), 30.7 (C), 35.8 (CH₃), 47.2 (CH₂), 48.5 (C), 55.8 (CH₂), 60.4 (C), 106.5 (C), 109.5 (C), 115.6 (CH), 120.5 (C), 121.1 (CH), 121.7 (CH), 126.3 (CH), 126.9 (C), 127.3 (CH), 127.8 (CH), 128.1 (CH), 129.6 (C), 134.2 (C), 137.6 (C), 141.7 (C), 150.4 (C), 151.0 (C), 177.3 (C), 193.2 (C) ppm; MS (EI, 70 eV): *m/z* (%) 537 ([M⁺], 2), 464 (100), 128 (62), 115 (48), 83 (57), Anal. Calcd for C₃₁H₃₁N₅O₄: C, 69.26; H, 5.81; N, 13.03. Found: C, 69.18; H, 5.77; N, 13.12.

General procedure for synthesis of compounds (8a,b). A mixture of enamine **3b** (0.30, 1 mmol) and 3-cyanomethylidene-2-oxindole derivatives **4a,b** (1 mmol) was heated at reflux in anhydrous pyridine (5 mL) for 3 h. The excess pyridine was evaporated at reduced pressure and the residue was then treated with dil. HCl (1 N, 10 mL). The collected crude solid was crystallized from EtOH-dioxane (3:1, v/v).

ethyl

5-(2'-amino-3'-cyano-7',7'-dimethyl-2,5'-dioxo-5',6',7',8'-tetrahydro-1'H-spiro[indoline-3,4'-quinolin]-1'-yl)-3-phenyl-1H-pyrazole-4-carboxylate (8a): Yellow crystals (0.46 g, 83%), Mp 248-250 °C, IR (KBr): ν 3466, 3370, 3313, 3143 (2NH and NH₂), 2188 (CN), 1705 (CO₂CH₂CH₃), 1643 (br, 2CO) cm⁻¹, ¹H NMR (400 MHz, DMSO-*d*₆): δ 0.87 (s, 3H, CH₃), 0.94 (s, 3H, CH₃), 1.11 (t, 3H, CO₂CH₂CH₃), 1.87-2.29 (m, 4H, 2CH₂), 4.20 (q, 2H, CO₂CH₂CH₃), 5.73 (br s, 2H, NH₂), 6.76-7.78 (m, 9H, ArH), 10.19 (br s, 1H, oxindole NH), 13.98 (br s, 1H, pyrazole NH) ppm; ¹³C NMR (100 MHz, CDCl₃): δ 12.8 (CH₃), 26.0 (CH₃), 27.8 (CH₃), 31.0 (C), 42.7 (CH₂), 47.7 (C), 48.8 (CH₂), 59.2 (CH₂), 60.8 (C), 94.0 (C), 108.0 (C), 117.8 (CH), 120.6 (C), 122.6 (CH), 124.4 (C), 126.6 (CH), 126.9 (CH), 127.0 (C), 128.3 (CH), 128.4 (CH), 128.6 (C), 134.4 (C), 140.3 (C), 148.4 (C), 151.0 (C), 160.3 (C), 171.7 (C), 179.1 (C), 193.1 (C) ppm; MS (EI, 70 eV): *m/z* (%) 548 ([M⁺], 5), 505 (33), 464 (44), 418 (100), 384 (53), 287 (49), 104 (66), 77 (83), Anal. Calcd for C₃₁H₂₈N₆O₄: C, 67.87; H, 5.14; N, 15.32. Found: C, 67.79; H, 5.08; N, 15.36.

ethyl

5-(2'-amino-3'-cyano-1,7',7'-trimethyl-2,5'-dioxo-5',6',7',8'-tetrahydro-1'H-spiro[indoline-3,4'-quinolin]-1'-yl)-3-phenyl-1H-pyrazole-4-carboxylate (8b): Yellow crystals (0.46 g, 81%), Mp 248-250 °C, IR (KBr): ν 3394, 3226 (br, 2NH and NH₂), 2184 (CN), 1701 (CO₂CH₂CH₃), 1650, 1611 (2CO) cm⁻¹, ¹H NMR (400 MHz, DMSO-*d*₆): δ 0.85 (s, 3H, CH₃), 0.93 (s, 3H, CH₃), 1.11 (t, 3H, CO₂CH₂CH₃), 1.88-2.30 (m, 4H, 2CH₂), 3.14 (s, 3H, NCH₃), 4.20 (q, 2H, CO₂CH₂CH₃), 5.79 (br s, 2H, NH₂), 6.94-7.77 (m, 9H, ArH), 13.99 (br s, 1H, pyrazole NH) ppm, ¹³C NMR (100 MHz, CDCl₃): δ 10.3 (CH₃), 27.9 (2CH₃), 32.3 (C), 41.8 (CH₂), 50.0 (CH₂), 99.2 (C), 112.0 (C), 125.9 (C), 128.1 (CH), 128.2 (CH), 132.0 (C), 138.2 (C), 152.9 (C), 161.9 (C), 196.9 (C) ppm, MS (EI, 70 eV): *m/z* (%) 562 ([M⁺], 22), 533 (13), 508 (25), 478 (85), 432 (100), 405 (32), 301 (21), 115 (33), 77 (57), Anal. Calcd for C₃₂H₃₀N₆O₄: C, 68.31; H, 5.37; N, 14.94. Found: C, 68.25; H, 5.33; N, 14.88.

General method for synthesis of compounds (12a,b). Compound **7a** or **7b** (1 mmol) was heated at reflux in acetic anhydride (5 mL) for 3 h. The excess solvent was removed at reduced pressure. The formed residue was extensively washed with aq. NaHCO₃ (1 N, 10 mL). The collected precipitate was air-dried and crystallized from EtOH-dioxane (3:1, v/v).

1-Acetyl-2',5',11',11'-tetramethyl-2,9'-dioxo-1'-phenyl-9',10',11',12'-tetrahydrospiro[indoline-3,8'-pyrazolo[1',5':5,6][1,3,5]triazino[1,2-*a*]quinoline]-7'-carbonitrile (12a): Yellow crystals (0.43 g, 78%), Mp 248-250 °C, IR (KBr): ν 2113 (CN), 1754, 1739, 1660 (3CO) cm⁻¹, ¹H NMR (400 MHz, DMSO-*d*₆):

δ 0.23 (s, 3H, CH_3), 0.84 (s, 3H, CH_3), 1.82 (m, 2H, CH_2), 2.08 (m, 2H, CH_2), 2.22 (s, 3H, $COCH_3$), 2.43 (s, 3H, pyrazole CH_3), 2.60 (s, 3H, triazine CH_3), 7.29-8.14 (m, 9H, ArH) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.9 (CH_3), 23.3 (CH_3), 24.8 (CH_3), 26.5 (CH_3), 28.4 (CH_3), 32.5 (C), 40.4 (CH_2), 49.5 (CH_2), 51.4 (CH_2), 94.0 (C), 112.4 (C), 113.5 (C), 116.8 (CH), 121.9 (C), 123.9 (CH), 125.8 (CH), 128.8 (CH), 129.1 (C), 129.5 (CH), 129.7 (CH), 130.2 (CH), 131.0 (C), 139.3 (C), 142.1 (C), 143.9 (C), 145.4 (C), 150.8 (C), 170.6 (C), 170.8 (C), 172.0 (C), 194.7 (C) ppm; MS (EI, 70 eV): m/z (%) 556 ($[M^+]$, 5), 513 (11), 446 (45), 129 (70), 83 (100), Anal. Calcd for $C_{33}H_{28}N_6O_3$: C, 71.21; H, 5.07; N, 15.10. Found: C, 71.17; H, 5.11; N, 15.21.

1,2',5',11',11'-Pentamethyl-2,9'-dioxo-1'-phenyl-9',10',11',12'-tetrahydrospiro[indoline-3,8'-pyrazolo[1',5':5,6][1,3,5]triazino[1,2-*a*]quinoline]-7'-carbonitrile (12b): Yellow crystals (0.40 g, 75%), Mp 248-250 °C, IR (KBr): ν 2121 (CN), 1767, 1725 (2CO) cm^{-1} , 1H NMR (400 MHz, $DMSO-d_6$): δ 0.22 (s, 3H, CH_3), 0.83 (s, 3H, CH_3), 1.77 (m, 2H, CH_2), 2.06 (m, 2H, CH_2), 2.40 (s, 3H, pyrazole CH_3), 2.60 (s, 3H, triazine CH_3), 3.18 (s, 3H, NCH_3), 7.08-7.67 (m, 9H, ArH) ppm, MS (EI, 70 eV): m/z (%) 528 ($[M^+]$, 54), 524 (87), 267 (32), 252 (22), 239 (47), 211 (100), 194 (34), 115 (30), 77 (15), Anal. Calcd for $C_{32}H_{28}N_6O_2$: C, 72.71; H, 5.34; N, 15.90. Found: C, 72.63; H, 5.28; N, 15.81.

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