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ORIGINAL ARTICLE

# Cerium oxide-catalyzed multicomponent condensation approach to spirooxindoles in water

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**Abstract** An efficient and facile green synthesis of spirooxindole derivatives bearing pyrano[2,3-*c*]pyrazole moiety has been achieved via a $CeO_2$-NPs catalyzed four-component reaction in water. The protocol offers an environmentally benign and effective approach to highly functionalized and biologically interesting spiro[indoline-3,4′-pyrano[2,3-*c*]pyrazole] derivatives. The synthesized compounds exhibit potent antioxidant and antibacterial activities.

**Keywords** Cerium oxide nanoparticles · Spirooxindoles · Antioxidant · Antibacterial · MCRs

## Introduction

The spirooxindole framework is an important structural motif found in many natural products and bioactive compounds [1–5]. These spirooxindole-based molecules have shown to possess varieties of important biological activities, such as antimicrobial [6–8], anti-inflammatory [9], antimalarial [10], antimycobacterial [11], antitubercular [12], antitumor and anticancer [13,14], and MDM2 inhibitor activity [15–17]. In addition, they are widely used as building blocks for the synthesis of bioactive natural products [18–20].

Due to their prominent biological and pharmacological activities, we became interested in the synthesis of a variety of spirooxindole derivatives using multicomponent reactions. Recently, we have developed simple and facile synthetic methods for the preparation of spirooxindole derivatives bearing hexahydroquinolines [21], dihydroquinazolinones [22], and 4-chromenes [23]. As a part of ongoing study on other spirooxindole derivatives, herein we examined four-component reactions of $\beta$-ketoesters, isatins, phenylhydrazines, and malononitrile to afford spirooxindole derivatives bearing pyrano[2,3-*c*]pyrazoles. It should be noted that a number of synthetic approaches using the **three**-component reaction of $\beta$-ketoesters with 3-methyl-2-pyrazolin-5-ones and malononitrile has been reported using $InCl_3$ [24], ZnS [25], $[Ch\text{-}OSO_3H]_3W_{12}PO_{40}$ [26], CAN/sonication [13], NaCl/sonication [27], $I_2$ [28], *L*-proline [29–35], 4-DMAP [36], and electrolysis [37] (Scheme 1). Also, several synthetic approaches to spirooxindole derivatives bearing pyrano[2,3-*c*]pyrazoles based on **four**-component reactions of $\beta$-ketoesters, isatins, phenylhydrazines, and malononitrile have been reported using $\beta$-cyclodextrin [38], chitosan/ionic liquid [39], piperidine [40,41], *L*-proline [42], and $ZrO_2$ [43] conditions. Still, there is a demand for more efficient and environmentally benign synthetic approaches to spirooxindole derivatives bearing pyrano[2,3-*c*]pyrazoles.

Green and sustainable chemical processes with reduction or even elimination of the use and production of hazardous materials are in high demand. Consequently, the use of non-toxic catalysts and pollution abatement solvents has become a prime choice for the researchers in both academia and industry. Recently, cerium oxide nanoparticles have emerged as environmentally benign and economical heterogeneous catalysts [44–49]. They have exhibited various advantages, such as sustainability in water, low corrosiveness and toxicity, high catalytic reactivity, recoverability and reusability, and ease of

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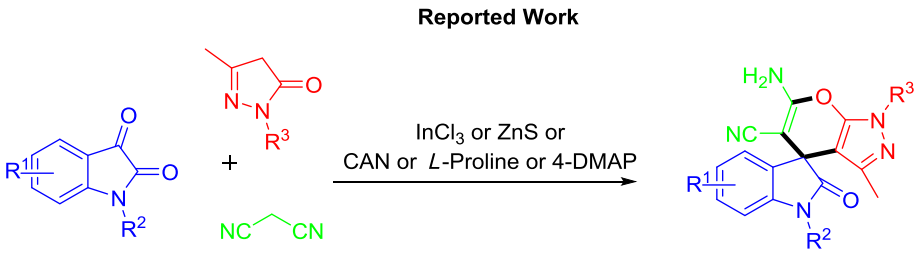
**Scheme 1** Reported three-component synthesis of spirooxindole derivative

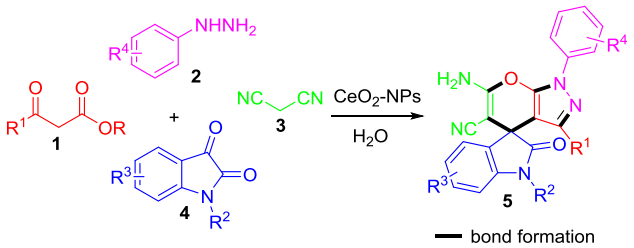



handling [50,51]. Because of these advantages, $CeO_2$-NPs, including core-metal/shell $CeO_2$ nanoparticles, have been extensively used as efficient and useful catalysts in various organic transformations [52–57]. Moreover, commercially available $CeO_2$-NPs are also being used in fluorescent applications [58], fuel cells [59], sunscreens [60], as antioxidants in cell model culture [61] and as gas sensors [62]. To the best of our knowledge, $CeO_2$ nanoparticle-catalyzed reactions of $\beta$-ketoesters with phenylhydrazines, malononitrile, and isatins for the construction of spirooxindoles have not been reported so far.

Herein, we describe a one-pot synthesis of biologically interesting spiro[indoline-3,4′-pyrano[2,3-*c*]pyrazole] derivatives using $CeO_2$ nanoparticle-catalyzed four-component reaction of $\beta$-ketoesters, phenylhydrazines, malononitrile, and isatins in water (Scheme 2). In addition, we report on the antibacterial and antioxidant activities of the synthesized spirooxindole derivatives.

## Results and discussion

The four-component reaction of methyl acetoacetate (**1a**), phenylhydrazine (**2a**), malononitrile (**3**), and isatin (**4a**) was first examined in the presence of several catalysts and solvents (Table 1). Initially, reaction in the absence of catalyst in water at 90 °C afforded the product **5a** in only 19 % yield (entry 1). Adding 30 mol% of ceric ammonium nitrate (CAN) and $CeCl_3$ at 90 °C allowed to increase the yield of **5a** to 47 and 35 %, respectively (entries 2 and 3). Further reactions were attempted with $CeO_2$-NPs in several solvents. The best yield (93 %) was obtained in the presence of 30 mol% of $CeO_2$-NPs in water at 90 °C (entry 5). Moreover, in polar



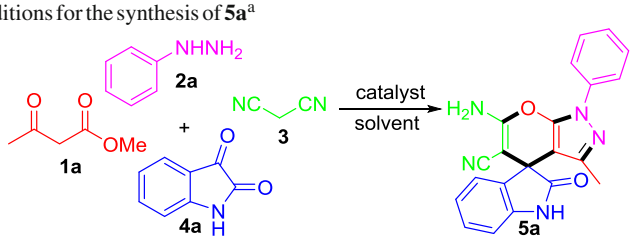

**Scheme 2** $CeO_2$ nanoparticle-catalyzed four-component reactions for the synthesis of **5**

solvents, such as ethanol and acetonitrile, **5a** was produced in a 65 and 81 % yield, respectively (entries 6 and 7) and in a non-polar solvent toluene, **5a** was obtained only in trace amounts (entry 8). The decrease or increase in loading of the catalyst ($CeO_2$-NPs) did not improve the yield of **5a** (entries 9, 10, and 11). Using 20 mol % of Lewis acids such as $FeCl_3$, $In(OTf)_3$ and $Cu(OTf)_2$ also gave the desired product in diminished yields (entries 12, 13, and 14). The identity of **5a** was confirmed by analysis of its spectroscopic data in comparison to reported values [25]. The $^1$H NMR of **5a** shows a methyl peak ($\delta$ = 1.54 ppm, singlet) and an amide proton ($\delta$ = 10.74 ppm, singlet). The $^{13}$C NMR exhibits a characteristic quaternary carbon peak at 47.7 ppm and an amide carbon peak at 177.4 ppm.

Under the optimized reaction condition, the generality of this multicomponent reaction was further explored by employing various $\beta$-ketoesters **1a–1d**, phenylhydrazines **2a–2e**, and isatins **4a–4k** (Table 2). Reactions of methyl 3-oxobutanoate (**1a**) with **2a**, **3**, and isatin **4b** or **4c** bearing electron-donating groups provided products **5b** and **5c** in an 84 and 86 % yield, respectively, whereas those with isatin **4d**, **4e**, or **4f** bearing electron-withdrawing groups afforded the desired products **5d–5f**, in a 91, 90, and an 87 % yield, respectively. Reactions of methyl and acetyl substituted isatins **4g** and **4h** provided products **5g–5h** in an 80 and 89 % yield, respectively. With other $\beta$-ketoesters of methyl 3-oxohexanoate (**1b**) or methyl 4-methyl-3-oxopentanoate (**1c**), the desired products **5i–5q** were produced in 86–94 % yields. Next, treatment of methyl 3-oxo-3-phenylpropanoate (**1d**) with phenylhydrazine (**2a**), malononitrile (**3**), and isatins **4a**, **4g**, or **4h** afforded the desired products **5r–5t** in 75–84 % yields.

In addition, further reactions of substituted phenylhydrazines **2b–2e** bearing electron-donating or -withdrawing substituents on various positions of benzene ring were successful. For example, treatment of **1a** with 4-methyl phenylhydrazine (**2b**), **3**, and **4a** provided the desired product **5u** in an 84 % yield, whereas that of **1c** with 2-ethyl phenylhydrazine (**2c**), **3**, and **4h** afforded the product **5v** in a 79 % yield. Moreover, treatment of **1c** or **1a** with 4-chloro phenylhydrazine (**2d**) or 2-chloro phenylhydrazine (**2e**), **3**, and **4a** gave products **5w** and **5x** in 73 and 74 % yields. However, when malononitrile was replaced by cyanoesters like methyl cyanoacetate or ethyl cyanoacetate, no desired

**Table 1** Optimization of reaction conditions for the synthesis of **5a**[a]




| Entry | Catalyst | Solvent | Temp. (°C) | Time (h) | Yield (%)[b] |
|---|---|---|---|---|---|
| 1 | – | Water | 90 | 24 | 19 |
| 2 | CAN (30 mol%) | Water | 90 | 12 | 47 |
| 3 | $CeCl_3$ (30 mol%) | Water | 90 | 12 | 35 |
| 4 | $CeO_2$ (30 mol%) | Water | rt | 12 | 77 |
| 5 | $CeO_2$ (30 mol%) | Water | 90 | 5 | 93 |
| 6 | $CeO_2$ (30 mol%) | Ethanol | 90 | 7 | 65 |
| 7 | $CeO_2$ (30 mol%) | Acetonitrile | 90 | 7 | 81 |
| 8 | $CeO_2$ (30 mol%) | Toluene | 90 | 7 | Trace |
| 9 | $CeO_2$ (20 mol%) | Water | 90 | 5 | 88 |
| 10 | $CeO_2$ (40 mol%) | Water | 90 | 5 | 90 |
| 11 | $CeO_2$ (5 mol%) | Water | 90 | 24 | 72 |
| 12 | $FeCl_3$ (20 mol%) | Water | 90 | 5 | 67 |
| 13 | $In(OTf)_3$ (20 mol%) | Water | 90 | 7 | 54 |
| 14 | $Cu(OTf)_2$ (20 mol%) | Water | 90 | 7 | 43 |

[a] Reaction conditions: methyl acetoacetate (**1a**, 1 mmol), isatin (**2a**, 1 mmol), phenylhydrazine (**3a**, 1 mmol), and malononitrile (**4**, 1 mmol).
[b] Isolated yield after column chromatography

products were obtained, instead intractable mixtures were produced. Our procedure provides a rapid synthetic route to a variety of highly functionalized spiro[indoline-3,4′-pyrano[2,3-*c*]pyrazole] derivatives in good yield. Moreover, most of the synthesized compounds of **5f**, **5i**, **5k–5q**, and **5t–5w** are novel and reported for the first time.

The formation of **5a** can be explained by the mechanism as shown in Scheme 3 [40,41]. In the presence of $CeO_2$-NPs, the intermediate **7** is first formed by condensation of methyl acetoacetate (**1a**) with phenylhydrazine (**2a**). The Knoevenagel condensation of **3a**, derived from **3** and **4a**, provides the intermediate **8**, which is reacted with **7a** to furnish **9** through Michael reaction. Tautomerism of **9** followed by intramolecular cyclization gives intermediate **10**, which undergoes further isomerization to furnish the final product **5a**.

## In vitro antioxidant activity

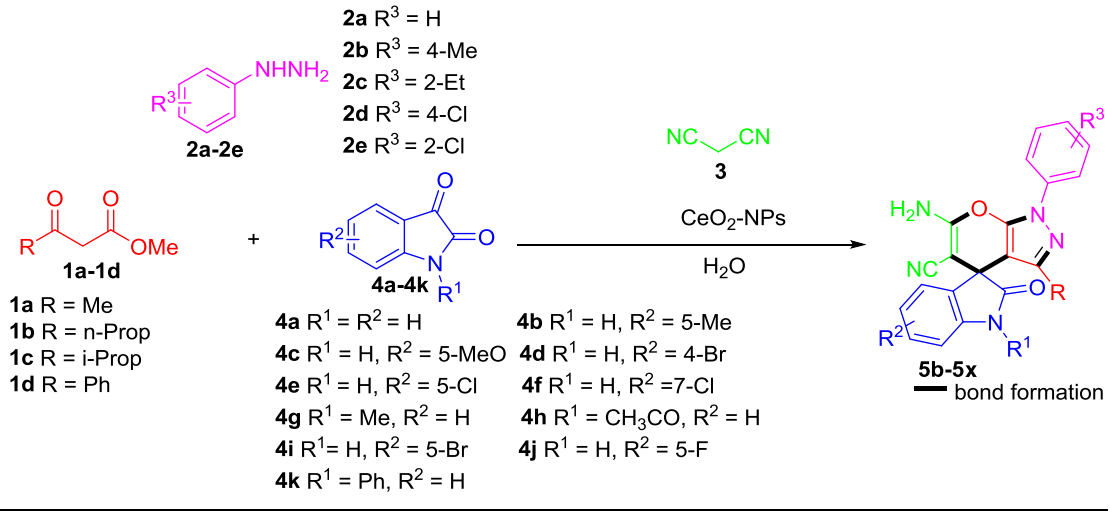
Furthermore, the synthesized spirooxindoles were screened for their antioxidant activity by using ferric reducing/antioxidant tests [63]. The FRAP assay measures the ability of a compound to reduce the ferric 2,4,6-tripyridyl-*s*-triazine complex to the colored ferrous complex with development of intense blue color at the maximum wavelength of 593 nm. FRAP values were obtained by comparing the absorbance change in test reaction mixtures with those containing ferrous ions of known concentration. The results of the antioxidant test are expressed as Trolox equivalent antioxidant capacity (TEAC) values as shown in Fig. 1. A higher value of TEAC suggests a higher antioxidant capacity. The majority of the tested compounds in the series revealed moderate interactions with the FRAP reagent. Compounds **5f**, **5h**, **5k**, and **5t** exhibited superior activity as compared to other synthesized compounds.

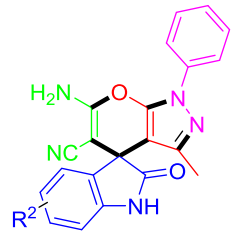
The antioxidant activity of organic compounds seems to be related to the presence of hydroxyl groups, double bond conjugation, and resonance effects [64]. The synthesized compounds containing both electron-withdrawing and -donating groups showed moderate activity. Compounds containing electron-withdrawing or -donating group on ortho or para positions to the spirooxindolic NH or acetyl group on spirooxindolic nitrogen atom showed higher activity than other compounds. In FRAP tests, the antioxidant activities of the compounds are well correlated with their $EC_{50}$ (half maximal effective concentration) values, as shown in Fig. 2.

## Antibacterial activity

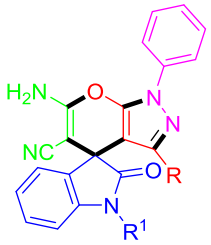
The antibacterial activity of the synthesized spirooxindoles was tested against two gram-negative *Escherichia coli*

**Table 2** Additional reactions for the synthesis of a variety of spirooxindole derivatives bearing pyrano[2,3 *c*]pyrazoles

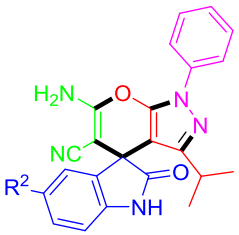


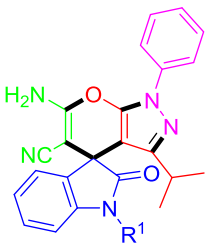
**5b**: $R^2$ = 5-Me, 6 h (84%)
**5c**: $R^2$ = 5-OMe, 6 h (86%)
**5d**: $R^2$ = 4-Br, 6 h (91%)
**5e**: $R^2$ = 5-Cl, 5 h (90%)
**5f**: $R^2$ = 7-Cl, 7 h (87%)



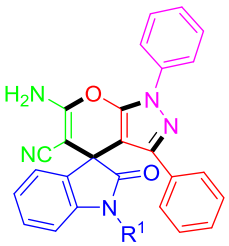
**5g**: $R^1$ = Me, R = Me, 7 h (80%)
**5h**: $R^1$ = $CH_3CO$, R = Me, 6 h (89%)
**5i**: $R^1$ = $CH_3CO$, R = n-Prop, 7 h (90%)



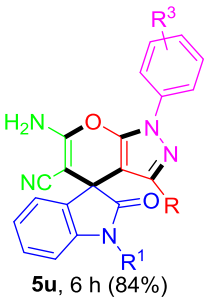
**5j**: $R^2$ = H, 6 h (94%)
**5k**: $R^2$ = OMe, 6 h (91%)
**5l**: $R^2$ = Br, 5 h (92%)
**5m**: $R^2$ = Cl, 5 h (92%)
**5n**: $R^2$ = F, 6 h (93%)



**5o**: $R^1$ = Me, 6 h (86%)
**5p**: $R^1$ = $CH_3CO$, 6 h (90%)
**5q**: $R^1$ = Ph, 7 h (92%)



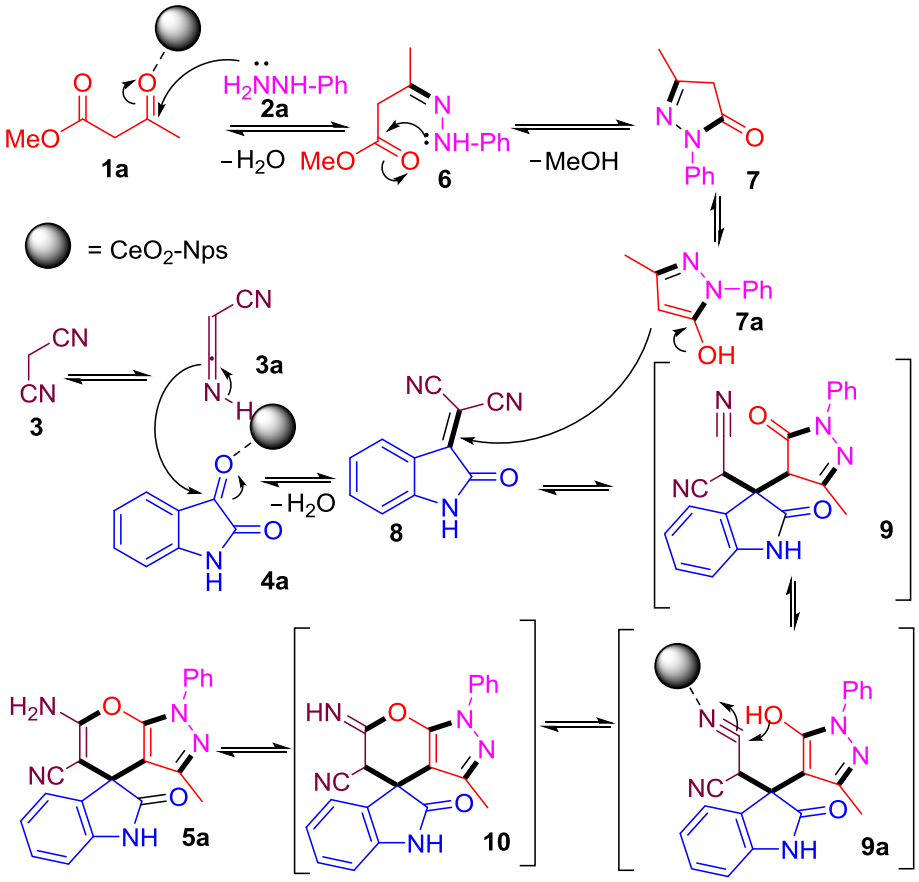
**5r**: $R^1$ = H, 8 h (83%)
**5s**: $R^1$ = Me, 8 h (75%)
**5t**: $R^1$ = $CH_3CO$, 7 h (84%)



**5u**, 6 h (84%)
**5u**: R = Me, $R^1$ = H, $R^3$ = 4-Me, 6 h (84%)
**5v**: R = i-Prop, $R^1$ = $CH_3CO$, $R^3$ = 2-Et, 8 h (79%)
**5w**: R = i-Prop, $R^1$ = H, $R^3$ = 4-Cl, 7 h (73%)
**5x**: R = Me, $R^1$ = H, $R^3$ = 2-Cl, 6 h (74%)

(KCTC-1924) and *Pseudomonas aeruginosa* (KCTC-2004) and two gram-positive *Staphylococcus aureus* (KCTC-1916) and *Bacillus cereus* (KCTC-1012) bacteria, respectively, by using a modified Kirby-Bauer disk diffusion method [65]. The inhibition zone against the growth of the verified bacteria for the compounds is reported in Table 3. Aliquots of bacterial suspension (100 μL) were spread on Difco™ nutrient broth containing the test microorganism with an optical density of 0.7 at 595 nm. From the results, synthesized compounds showed antibacterial activity toward the investigated bacte-

**Scheme 3** Proposed mechanism for the formation of **5a**



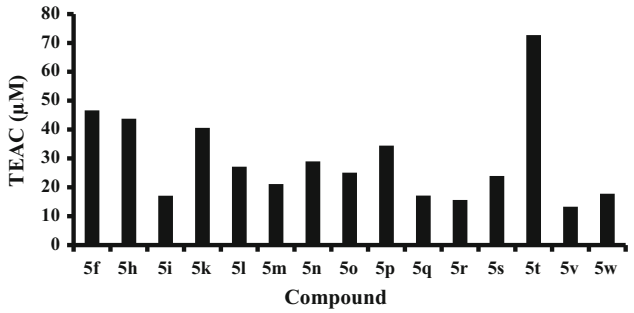




**Fig. 1** Antioxidant activity of the synthesized spirooxindoles expressed in TEAC values

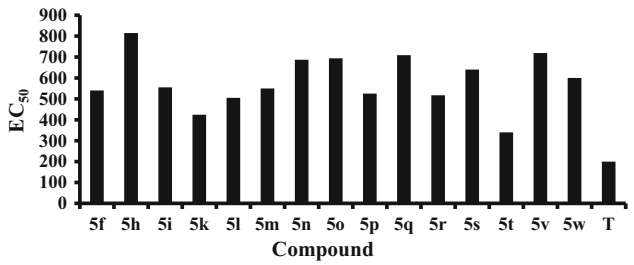



**Fig. 2** $EC_{50}$ (μg/mL) of the synthesized spirooxindoles. Abbreviation: *T* Trolox, *EC* effective concentration

**Table 3** Antimicrobial activity of synthesized spirooxindoles against several standard strains

| Compound | Diameter of growth inhibition zone (mm) | | | |
|---|---|---|---|---|
| | Gram-negative | | Gram-positive | |
| | *E. coli* | *P. aeruginosa* | *S. aureus* | *B. cereus* |
| **5f** | 10 | 11 | – | – |
| **5k** | 12 | 11 | – | – |
| **5m** | 22 | 21 | 15 | 16 |
| **5n** | 9 | 11 | – | – |
| **5q** | 10 | 11 | 11 | 10 |
| **5s** | 12 | 12 | 11 | 10 |
| **5t** | 21 | 20 | 15 | 16 |
| **5v** | 18 | 17 | 14 | 15 |
| **Ciprofloxacin** | 20 | 19 | 18 | 18 |

rial strains. In particular, compounds **5m** and **5t** exhibited excellent activity toward the gram-negative bacteria compared to standard ciprofloxacin. Compounds **5m**, **5t**, and **5v** displayed moderate levels of antimicrobial activity toward the gram- positive bacteria. Compounds containing electron-withdrawing group on spirooxindolic NH or its para position showed higher activity than other compounds.

## Conclusions

A green and efficient protocol for the construction of spirooxindole derivatives bearing pyrano[2,3-*c*]pyrazole] by a multicomponent coupling of $\beta$-ketoesters, phenylhydrazines, malononitrile, and isatins was developed. This method offers several advantages such as mild reaction conditions, ease of handling, high yields, and the use of an effective green catalyst. The synthesized spirooxindole derivatives show potent antioxidant and antibacterial activities.

## Experimental

All experiments were conducted under a nitrogen atmosphere. Merck precoated silica gel plates (Art. 5554) with a fluorescent indicator were used for analytical TLC. Flash column chromatography was performed using silica gel 9385 (Merck). The melting points were determined using micro-cover glasses on a Fisher-Johns apparatus and were uncorrected. The $^1$H NMR spectra were recorded on a Varian-VNS (300 MHz), DPX (300 MHz), and VNS (600 MHz) spectrometer in DMSO-$d_6$ setting the solvent chemical shift at 2.50 ppm. The $^{13}$C NMR spectra were recorded on a Varian-VNS (75 MHz), DPX (75 MHz), and VNS (150 MHz) spectrometer in DMSO-$d_6$ setting the solvent chemical shift at 39.5 ppm. Chemical shifts ($\delta$) are expressed in units of ppm and $J$ values are given in Hz. Multiplicities are abbreviated as follows: s = singlet, d = doublet, t = triplet, q = quartet, br s = broad singlet, dd = doublet of doublets, tt = triplet of triplet, and m = multiplet. The IR spectra were recorded on PerkinElmer FT-IR spectrometer Spectrum Two$^{TM}$. High-resolution mass spectrometry (HRMS) was obtained with a JEOL JMS-700 spectrometer (EI) at the Korea Basic Science Institute.

### General procedure for the synthesis of spirooxindole derivatives (5a–5x)

A mixture of $\beta$-ketoester **1** (1 mmol), phenylhydrazine **2** (1 mmol), malononitrile **3** (1 mmol), isatin **4** (1 mmol), and $CeO_2$-NPs (30 mol%) in water (5 mL) was stirred at 90 °C for the time mentioned, until the completion of reaction as indicated by TLC. After completion of the reaction, the reaction mixture was cooled to room temperature. The precipitated product was then filtered and dissolved in EtOAc. The solution was then dried over $MgSO_4$ and filtered. After evaporating solvent, the residue was recrystallized from EtOAc to provide pure product.

*6′-Amino-3′-methyl-2-oxo-1′-phenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5a)**

Compound **5a** (342 mg, 93 %) was obtained as a white solid: mp 237–239 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.74 (1H, s), 7.78 (2H, d, $J$ = 7.8 Hz), 7.56–7.49 (4H, m), 7.37–7.26 (2H, m), 7.17 (1H, d, $J$ = 7.2 Hz), 7.02 (1H, t, $J$ = 7.5 Hz), 6.95 (1H, d, $J$ = 7.5 Hz), 1.54 (3H, s); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.4, 161.0, 144.9, 143.8, 141.6, 137.2, 132.1, 129.4, 129.2, 126.5, 124.8, 122.7, 120.1, 117.8, 109.8, 96.3, 56.2, 47.7, 11.6; IR (ATR) 3451, 3254, 3080, 2247, 1891, 1739, 1562, 1428, 1348, 1236, 1099, 988, 931 cm$^{-1}$; HRMS $m/z$ (M$^+$) calcd for $C_{21}H_{15}N_5O_2$: 369.1226. Found: 369.1229.

*6′-Amino-3′,5-dimethyl-2-oxo-1′-phenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5b)**

Compound **5b** (320 mg, 84 %) was obtained as a white solid: mp 288–289 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.63 (1H, s), 7.79 (2H, d, $J$ = 8.1 Hz), 7.52 (4H, t, $J$ = 8.1 Hz), 7.35 (1H, t, $J$ = 7.5 Hz), 7.08 (1H, d, $J$ = 7.5 Hz), 7.0 (1H, s), 6.83 (1H, d, $J$ = 7.8 Hz), 2.23 (3H, s), 1.56 (3H, s); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.4, 161.0, 144.9, 144.0, 139.1, 137.3, 132.3, 131.6, 129.5, 129.4, 126.5, 125.3, 120.1, 118.0, 109.6, 96.5, 56.3, 47.8, 20.6, 11.7; IR (ATR) 3420, 3254, 3089, 2246, 1875, 1732, 1673, 1554, 1427, 1347, 1237, 1158, 989 cm$^{-1}$; HRMS $m/z$ (M$^+$) calcd for $C_{22}H_{17}N_5O_2$: 383.1382. Found: 383.1384.

*6′-Amino-5-methoxy-3′-methyl-2-oxo-1′-phenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5c)**

Compound **5c** (342 mg, 86 %) was obtained as a white solid: mp 213–215 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.54 (1H, s), 7.79 (2H, d, $J$ = 8.1 Hz), 7.54–7.49 (4H, m), 7.34 (1H, t, $J$ = 7.8 Hz), 6.8 (3H, d, $J$ = 8.1 Hz), 3.68 (3H, s), 1.56 (3H, s); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.5, 161.0, 155.7, 145.0, 144.0, 137.3, 134.8, 133.4, 129.5, 126.5, 120.1, 118.0, 114.4, 111.3, 110.4, 96.4, 56.3, 55.5, 48.3, 11.7; IR (ATR) 3318, 3184, 3062, 2203, 1696, 1486, 1392, 1293, 1199, 1029, 962 cm$^{-1}$; HRMS $m/z$ (M$^+$) calcd for $C_{22}H_{17}N_5O_3$: 399.1331. Found: 399.1328.

*6′-Amino-4-bromo-3′-methyl-2-oxo-1′-phenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5d)**

Compound **5d** (405 mg, 91 %) was obtained as a white solid: mp 268–270 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 11.02 (1H, s), 7.78 (2H, d, $J$ = 9.0 Hz), 7.65 (2H, s), 7.51 (2H, t, $J$ = 7.8 Hz), 7.35 (1H, t, $J$ = 7.2 Hz), 7.25 (1H, t, $J$ = 7.8 Hz), 7.18 (1H, d, $J$ = 7.8 Hz), 6.98 (1H, d, $J$ = 7.8 Hz), 1.60 (3H, s); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 176.7, 161.7,

145.5, 143.7, 143.6, 137.2, 131.3, 129.5, 128.6, 126.7, 126.1, 120.1, 119.5, 117.7, 109.5, 94.1, 53.9, 49.4, 11.7 $cm^{-1}$; IR (ATR) 3320, 3193, 2198, 1723, 1655, 1583, 1527, 1447, 1394, 1221, 1127, 1037 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{21}H_{14}BrN_5O_2$: 447.0331. Found: 447.0328.

*6′-Amino-5-chloro-3′-methyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5e)**

Compound **5e** (362 mg, 90 %) was obtained as a white solid: mp 229–231 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.89 (1H, s), 7.79 (2H, d, $J$ = 7.2 Hz), 7.63 (2H, s), 7.53–7.49 (2H, m), 7.35–7.32 (3H, m), 6.97 (1H, d, $J$ = 7.5 Hz), 1.59 (3H, s); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.3, 161.1, 145.0, 143.8, 140.4, 137.2, 134.3, 129.4, 129.2, 126.7, 126.6, 125.2, 120.3, 117.9, 111.3, 95.7, 55.6, 48.0, 11.7; IR (ATR) 3305, 3181, 3015, 2194, 1842, 1725, 1646, 1451, 1382, 1216, 1123, 1068 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{21}H_{14}ClN_5O_2$: 403.0836. Found: 403.0835.

*6′-Amino-7-chloro-3′-methyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5f)**

Compound **5f** (350 mg, 87 %) was obtained as a white solid: mp 235–237 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 11.19 (1H, s), 7.78 (2H, d, $J$ = 7.5 Hz), 7.63 (2H, s), 7.52 (2H, t, $J$ = 7.5 Hz), 7.35 (2H, t, $J$ = 7.5 Hz), 7.18 (1H, d, $J$ = 7.2 Hz), 7.06 (1H, t, $J$ = 7.8 Hz), 1.57 (3H, s); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.5, 161.0, 144.9, 143.7, 139.3, 137.1, 133.9, 129.4, 129.2, 126.6, 123.9, 123.6, 120.2, 117.7, 114.1, 95.8, 55.7, 48.6, 11.7; IR (ATR) 3439, 3253, 3093, 2248, 1969, 1737, 1600, 1430, 1352, 1236, 1160, 992 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{21}H_{14}ClN_5O_2$: 403.0836. Found: 403.0837.

*6′-Amino-1,3′-dimethyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5g)**

Compound **5g** (306 mg, 80 %) was obtained as a white solid: mp 212–214 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 7.78 (2H, d, $J$ = 8.4 Hz), 7.60 (2H, s), 7.51 (2H, t, $J$ = 8.4 Hz), 7.39 (1H, t, $J$ = 7.2 Hz), 7.35 (1H, t, $J$ = 7.2 Hz), 7.23 (1H, d, $J$ = 7.8 Hz), 7.15 (1H, d, $J$ = 7.8 Hz), 7.11 (1H, t, $J$ = 7.2 Hz), 3.24 (3H, s), 1.45 (3H, s); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 175.8, 161.2, 144.9, 143.8, 143.0, 137.2, 131.4, 129.5, 126.6, 124.6, 123.4, 120.2, 117.8, 108.9, 96.2, 55.8, 47.4, 26.5, 11.7; IR (ATR) 3445, 3236, 3095, 2229, 1780, 1541, 1419, 1331, 1214, 952 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{22}H_{17}N_5O_2$: 383.1382. Found: 383.1381.

*1-Acetyl-6′-amino-3′-methyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5h)**

Compound **5h** (364 mg, 89 %) was obtained as a white solid: mp 224–226 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 8.20 (1H, d, $J$ = 8.1 Hz), 7.8 (4H, d, $J$ = 8.1 Hz), 7.55–7.43 (3H, m), 7.39–7.29 (3H, m), 2.64 (3H, s), 1.51 (3H, s); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.2, 170.6, 161.2, 144.9, 144.0, 139.3, 137.1, 130.4, 129.7, 129.5, 126.8, 126.2, 125.1, 120.4, 117.6, 115.8, 95.9, 56.0, 48.5, 26.3, 11.9; IR (ATR) 3379, 3319, 3192, 2924, 2202, 1723, 1651, 1517, 1393, 1257, 1162, 1031, 908 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{23}H_{17}N_5O_3$: 411.1331. Found: 411.1335.

*1-Acetyl-6′-amino-2-oxo-1′-phenyl-3′-propyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5i)**

Compound **5i** (394 mg, 90 %) was obtained as a white solid: mp 224–226 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 8.20 (1H, d, $J$ = 8.1 Hz), 7.83–7.81 (4H, m), 7.56–7.44 (3H, m), 7.41–7.29 (3H, m), 2.64 (3H, s), 1.80 (2H, t, $J$ = 7.5 Hz), 1.21–1.02 (2H, m), 0.58 (3H, t, $J$ = 7.5 Hz); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.6, 170.4, 161.0, 147.8, 144.8, 139.1, 137.1, 130.8, 129.7, 129.4, 126.8, 126.2, 125.2, 120.4, 117.6, 115.8, 95.5, 56.2, 48.6, 28.5, 26.2, 20.8, 13.6; IR (ATR) 3313, 3198, 2947, 2204, 1757, 1648, 1458, 1394, 1262, 1153, 1071, 907 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{25}H_{21}N_5O_3$: 439.1644. Found: 439.1641.

*6′-Amino-3′-isopropyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5j)**

Compound **5j** (372 mg, 94 %) was obtained as a white solid: mp 217–219 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 10.75 (1H, s), 7.80 (2H, d, $J$ = 7.8 Hz), 7.53–7.51 (4H, m), 7.35 (1H, t, $J$ = 6.6 Hz), 7.27 (1H, t, $J$ = 7.2 Hz), 7.19 (1H, d, $J$ = 7.2 Hz), 7.02 (1H, t, $J$ = 7.2 Hz), 6.94 (1H, d, $J$ = 7.8 Hz), 2.10-2.05 (1H, m), 1.01 (3H, d, $J$ = 6.6 Hz), 0.69 (3H, d, $J$ = 6.6 Hz); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 178.0, 160.7, 153.1, 144.7, 141.5, 137.3, 132.8, 129.4, 129.3, 126.5, 125.0, 122.6, 120.2, 117.9, 109.9, 95.0, 56.7, 47.8, 26.4, 21.4, 21.0; IR (ATR) 3412, 3254, 3091, 2248, 1891, 1675, 1619, 1425, 1345, 1437, 1160, 1096, 989 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{23}H_{19}N_5O_2$: 397.1539. Found: 397.1540.

*6′-Amino-3′-isopropyl-5-methoxy-2-oxo-1′-phenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5k)**

Compound **5k** (388 mg, 91 %) was obtained as a white solid: mp 221–223 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.57 (1H, s), 7.81 (2H, d, $J$ = 8.1 Hz), 7.54–7.52 (4H, m), 7.34

(1H, t, $J$ = 7.2 Hz), 6.85 (3H, s), 3.67 (3H, s), 2.16–2.07 (1H, m), 1.01 (3H, d, $J$ = 6.9), 0.74 (3H, d, $J$ = 6.6); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.9, 160.7, 155.7, 153.2, 144.7, 137.4, 134.7, 134.1, 129.4, 126.5, 120.2, 118.0, 114.5, 111.4, 110.4, 95.1, 56.8, 55.6, 48.4, 26.4, 21.5, 21.1; IR (ATR) 3313, 3188, 2964, 2193, 1839, 1704, 1645, 1475, 1390, 1199, 1030, 760 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{24}H_{21}N_5O_3$: 427.1644. Found: 427.1642.

*6′-Amino-5-bromo-3′-isopropyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5l)**

Compound **5l** (436 mg, 92 %) was obtained as a white solid: mp 200–202 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.90 (1H, s), 7.80 (2H, d, $J$ = 7.5 Hz), 7.57–7.45 (6H, m), 7.36 (1H, t, $J$ = 7.5 Hz), 6.90 (1H, d, $J$ = 8.1 Hz), 2.13–2.06 (1H, m), 1.02 (3H, d, $J$ = 6.9 Hz), 0.74 (3H, d, $J$ = 6.9 Hz); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.6, 160.8, 152.9, 144.8, 140.7, 137.3, 135.4, 132.1, 129.4, 128.0, 126.6, 120.4, 117.8, 114.3, 111.9, 94.4, 56.0, 48.0, 26.5, 21.4, 21.1; IR (ATR) 3313, 3289, 2972, 2193, 1707, 1640, 1452, 1393, 1215, 1085 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{23}H_{18}BrN_5O_2$: 475.0644. Found: 475.0641.

*6′-Amino-5-chloro-3′-isopropyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5m)**

Compound **5m** (396 mg, 92 %) was obtained as a white solid: mp 203–205 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.90 (1H, s), 7.80 (2H, d, $J$ = 8.1 Hz), 7.58–7.50 (4H, m), 7.38–7.32 (3H, m), 6.96 (1H, d, $J$ = 8.1 Hz), 2.14–2.05 (1H, m), 1.02 (3H, d, $J$ = 6.9 Hz), 0.74 (3H, d, $J$ = 6.6 Hz); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.8, 160.8, 152.9, 144.8, 140.3, 137.3, 135.1, 129.4, 129.3, 126.7, 126.6, 125.3, 120.5, 117.9, 111.4, 94.4, 55.9, 48.1, 26.5, 21.5, 21.1; IR (ATR) 3345, 3225, 3016, 2248, 1751, 1680, 1561, 1423, 1351, 1220, 1095, 920 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{23}H_{18}ClN_5O_2$: 431.1149. Found: 431.1153.

*6′-Amino-5-fluoro-3′-isopropyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5n)**

Compound **5n** (385 mg, 93 %) was obtained as a white solid: mp 226–228 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.79 (1H, s), 7.80 (2H, d, $J$ = 7.8 Hz), 7.57–7.49 (4H, m), 7.35 (1H, t, $J$ = 7.2 Hz), 7.22–7.19 (1H, m), 7.14–7.07 (1H, m), 6.96–6.91 (1H, m), 2.14–2.07 (1H, m), 1.02 (3H, d, $J$ = 6.9 Hz), 0.74 (3H, d, $J$ = 6.9 Hz); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 178.0, 160.8, 158.7 (d, $J$ = 237.9 Hz), 153.0, 144.7, 137.5 (d, $J$ = 46.1 Hz), 134.7 (d, $J$ = 15.5 Hz), 129.4, 126.6, 120.3, 117.8, 115.7 (d, $J$ = 46.1 Hz), 112.8 (d, $J$ = 48.5 Hz), 110.9, 94.5, 56.2, 48.4, 26.4, 21.5, 21.0; IR (ATR) 3318, 3191, 2972, 2192, 1762, 1645, 1471, 1391, 1179, 1082, 757 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{23}H_{18}FN_5O_2$: 415.1445. Found: 415.1443.

*6′-Amino-3′-isopropyl-1-methyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5o)**

Compound **5o** (353 mg, 86 %) was obtained as a white solid: mp 221–223 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 7.80 (2H, d, $J$ = 7.8 Hz), 7.56 (2H, s), 7.52 (2H, t, $J$ = 7.8 Hz), 7.40–7.34 (2H, m), 7.26 (1H, d, $J$ = 7.8 Hz), 7.16 (1H, d, $J$ = 8.4 Hz), 7.11 (1H, t, $J$ = 7.8 Hz), 3.33 (3H, s), 1.92–1.87 (1H, m), 0.91 (3H, d, $J$ = 7.2 Hz), 0.69 (3H, d, $J$ = 6.6 Hz); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 176.2, 160.8, 152.9, 144.7, 142.9, 137.3, 132.0, 129.4, 124.7, 123.3, 120.4, 120.2, 117.7, 108.9, 108.8, 94.8, 56.2, 47.5, 26.4, 26.4, 21.4, 20.7; IR (ATR) 3306, 3181, 2965, 2201, 1703, 1654, 1457, 1397, 1083, 937 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{24}H_{21}N_5O_2$: 411.1695. Found: 411.1697.

*1-Acetyl-6′-amino-3′-isopropyl-2-oxo-1′-phenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5p)**

Compound **5p** (394 mg, 90 %) was obtained as a white solid: mp 221–223 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 7.80 (2H, d, $J$ = 7.8 Hz), 7.56 (2H, s), 7.52 (2H, t, $J$ = 7.8 Hz), 7.40–7.34 (2H, m), 7.26 (1H, d, $J$ = 7.8 Hz), 7.16 (1H, d, $J$ = 8.4 Hz), 7.11 (1H, t, $J$ = 7.8 Hz), 3.33 (3H, s), 1.92–1.87 (1H, m), 0.91 (3H, d, $J$ = 7.2 Hz), 0.69 (3H, d, $J$ = 6.6 Hz); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 177.7, 170.3, 160.7, 152.9, 144.7, 139.0, 137.2, 131.1, 129.7, 129.4, 126.7, 126.1, 125.2, 120.5, 117.4, 115.8, 94.5, 56.5, 48.6, 26.5, 26.1, 21.5, 20.7; IR (ATR) 3422, 3254, 3093, 2247, 1875, 1675, 1610, 1425, 1343, 1243, 1160, 1099, 989 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{25}H_{21}N_5O_3$: 439.1644. Found: 439.1641.

*6′-Amino-3′-isopropyl-2-oxo-1,1′-diphenyl-1′H-spiro [indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5q)**

Compound **5q** (435 mg, 92 %) was obtained as a white solid: mp 238–240 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 7.82 (2H, d, $J$ = 7.5 Hz), 7.66–7.62 (4H, m), 7.55–7.53 (3H, m), 7.40–7.31 (5H, m), 7.18–7.14 (1H, m), 6.8 (1H, d, $J$ = 7.8 Hz), 2.19-2.12 (1H, m), 1.02 (3H, d, $J$ = 6.6 Hz), 0.78 (3H, d, $J$ = 6.6 Hz); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 175.8, 160.6, 153.0, 144.8, 142.5, 137.3, 134.0, 131.8, 130.0, 129.5, 129.3, 128.5, 126.6, 126.3, 125.2, 124.0, 120.3, 117.6, 109.2, 94.5, 56.6, 47.7, 26.6, 21.6, 20.7; IR (ATR) 3456, 3278, 3160, 2962, 2191, 1704, 1651, 1595, 1489, 1384, 1211, 1084, 933 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{29}H_{23}N_5O_2$: 473.1852. Found: 473.1850.

*6′-Amino-2-oxo-1′,3′-diphenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5r)**

Compound **5r** (357 mg, 83 %) was obtained as a white solid: mp 208–210 °C[1]; H NMR (600 MHz, DMSO-$d_6$) $\delta$ 10.60 (1H, s), 7.90 (2H, d, $J$ = 7.8 Hz), 7.59–7.54 (4H, m), 7.42 (1H, t, $J$ = 7.2 Hz), 7.23–7.19 (3H, m), 7.13 (2H, t, $J$ = 7.2 Hz), 6.97–6.92 (3H, m), 6.79 (1H, d, $J$ = 7.8 Hz); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.6, 160.3, 147.4, 145.7, 141.7, 137.1, 133.5, 132.0, 129.5, 129.2, 128.3, 127.9, 127.3, 124.8, 122.5, 121.0, 120.8, 117.6, 109.8, 95.6, 57.5, 48.1; IR (ATR) 3408, 3254, 3095, 2246, 1891, 1816, 1730, 1671, 1549, 1422, 1345, 1158, 1099, 989 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{26}H_{17}N_5O_2$: 431.1382 . Found: 431.1379.

*6′-Amino-1-methyl-2-oxo-1′,3′-diphenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5s)**

Compound **5s** (333 mg, 75 %) was obtained as a white solid: mp 198–200 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 7.89 (2H, d, $J$ = 7.8 Hz), 7.61 (2H, s), 7.57 (2H, t, $J$ = 7.8 Hz), 7.43–7.41 (1H, m), 7.28 (1H, t, $J$ = 7.8 Hz), 7.23 (2H, t, $J$ = 7.8 Hz), 7.14 (2H, t, $J$ = 7.8 Hz), 7.02 (1H, t, $J$ = 7.8 Hz), 6.93 (1H, d, $J$ = 7.8 Hz), 6.80 (2H, d, $J$ = 7.2 Hz), 2.98 (3H, s); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 175.9, 160.5, 147.4, 145.4, 142.8, 137.1, 132.6, 131.8, 129.5, 129.3, 128.3, 127.9, 127.3, 127.2, 124.4, 123.2, 120.9, 117.6, 108.7, 95.8, 56.7, 47.7, 26.2; IR (ATR) 3364, 3185, 3068, 2199, 1863, 1660, 1515, 1382, 1249, 1079 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{27}H_{19}N_5O_2$: 445.1539. Found: 445.1542.

*1-Acetyl-6′-amino-2-oxo-1′,3′-diphenyl-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5t)**

Compound **5t** (397 mg, 84 %) was obtained as a white solid: mp 204–206 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 7.97–7.90 (3H, m), 7.81 (2H, s), 7.58 (2H, t, $J$ = 7.8 Hz), 7.46–7.34 (3H, m), 7.27-7.21 (2H, m), 7.11 (2H, t, $J$ = 7.8 Hz), 6.79 (2H, d, $J$ = 7.5 Hz), 2.39 (3H, s); $^{13}$C NMR (75 MHz, DMSO-$d_6$) $\delta$ 177.4, 169.9, 160.6, 147.5, 145.3, 139.0, 137.0, 131.8, 131.5, 129.5, 128.6, 128.0, 127.4, 127.3, 126.0, 125.0, 121.0, 117.4, 115.6, 96.0, 56.7, 48.8, 25.7; IR (ATR) 3317, 3197, 2925, 2200, 1717, 1645, 1454, 1385, 1682, 1153, 1027, 913 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{28}H_{19}N_5O_3$: 473.1488. Found: 473.1487.

*6′-Amino-3′-methyl-2-oxo-1′-(p-tolyl)-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5u)**

Compound **5u** (321 mg, 84 %) was obtained as a white solid: mp 242–244 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 10.72 (1H, s), 7.65 (2H, d, $J$ = 8.4 Hz), 7.54 (2H, s), 7.31–7.26 (3H, m), 7.16 (1H, d, $J$ = 7.2 Hz), 7.02 (1H, t, $J$ = 7.2 Hz), 6.94 (1H, d, $J$ = 7.8 Hz), 2.35 (3H, s), 1.53 (3H, s); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 177.5, 161.0, 144.7, 143.6, 141.6, 136.0, 134.9, 132.2, 129.9, 129.7, 129.2, 124.8, 122.6, 120.2, 120.1, 118.0, 109.8, 96.1, 56.2, 47.8, 20.5, 11.6; IR (ATR) 3443, 3326, 3155, 2189, 1707, 1471, 1394, 1328, 1224, 1068, 1044, 933, 751, 615 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{22}H_{17}N_5O_2$: 383.1382. Found: 383.1385.

*1-Acetyl-6′-amino-1′-(2-ethylphenyl)-3′-isopropyl-2-oxo-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5v)**

Compound **5v** (368 mg, 79 %) was obtained as a white solid: mp 211–213 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 8.18 (1H, d, $J$ = 8.4 Hz), 7.59 (2H, s), 7.50–7.44 (4H, m), 7.41–7.38 (1H, m), 7.36–7.34 (2H, m), 2.63 (3H, s), 2.45–2.41 (2H, m), 1.97–1.92 (1H, m), 1.04 (3H, t, $J$ = 7.2 Hz), 0.91 (3H, d, $J$ = 6.6 Hz), 0.65 (3H, d, $J$ = 6.6 Hz); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 177.8, 170.4, 160.8, 152.4, 145.5, 141.3, 139.0, 134.8, 131.3, 129.8, 129.6, 127.6, 126.8, 126.2, 124.9, 117.5, 115.8, 92.7, 56.6, 48.8, 26.5, 26.1, 23.9, 21.6, 20.8, 14.4; IR (ATR) 3395, 3322, 2965, 2204, 1766, 1648, 1526, 1393, 1331, 1268, 1155, 1090, 1040 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{27}H_{25}N_5O_3$: 467.1957. Found: 467.1954.

*1-Acetyl-6′-amino-1′-(4-chlorophenyl)-3′-isopropyl-2-oxo-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5w)**

Compound **5w** (357 mg, 83 %) was obtained as a white solid: mp 250–252 °C; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ 10.75 (1H, s), 7.84 (2H, d, $J$ = 8.7 Hz), 7.58–7.54 (4H, m), 7.27 (1H, t, $J$ = 7.5 Hz), 7.19 (1H, d, $J$ = 7.2 Hz), 7.01 (1H, t, $J$ = 7.5 Hz), 6.90 (1H, d, $J$ = 7.8 Hz), 2.11–2.02 (1H, s), 0.99 (3H, d, $J$ = 6.9 Hz), 0.68 (3H, d, $J$ = 6.9 Hz); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 177.8, 160.6, 153.5, 144.7, 141.4, 136.2, 132.6, 130.6, 129.3, 125.0, 122.6, 121.6, 117.7, 109.9, 95.3, 56.7, 47.8, 26.3, 21.3, 20.9; IR (ATR) 3460, 3334, 3186, 2190, 1852, 1709, 1644, 1508, 1391, 1329, 1221, 1092, 1044, 931 cm$^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{23}H_{18}ClN_5O_2$: 431.1149. Found: 431.1146.

*6′-Amino-1′-(2-chlorophenyl)-3′-methyl-2-oxo-1′H-spiro[indoline-3,4′-pyrano[2,3-c]pyrazole]-5′-carbonitrile* **(5x)**

Compound **5x** (298 mg, 74 %) was obtained as a white solid: mp 250–252 °C; $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 10.72 (1H, s), 7.72 (1H, d, $J$ = 7.8 Hz), 7.64 (1H, d, $J$ = 6.6 Hz), 7.59–7.53 (2H, m), 7.40 (2H, s), 7.29 (1H, t, $J$ = 7.8 Hz), 7.11 (1H, d, $J$ = 7.2 Hz), 7.04 (1H, t, $J$ = 7.2 Hz), 6.94 (1H, d, $J$ = 7.2 Hz), 1.53 (3H, s); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 177.4, 161.0, 146.1, 144.2, 141.6, 133.9, 132.2, 131.3, 130.8, 130.4, 129.9, 129.3,

128.3, 124.7, 122.6, 118.0, 109.8, 95.0, 56.2, 48.0, 11.7; IR (ATR) 3315, 3160, 2922, 2198, 1711, 1531, 1473, 1381, 1128, 1042, 932, 750, 623 $cm^{-1}$; HRMS $m/z$ ($M^+$) calcd for $C_{21}H_{14}ClN_5O_2$: 403.0836. Found: 403.0833.

### Ferric reducing antioxidant power (FRAP) assay

The FRAP assay was carried out according to the procedure described by Benzie and Strain [63]. FRAP reagent was prepared freshly by adding 10 vol. of 300 mM acetate buffer, pH 3.6 (3.1 g sodium acetate and 16 mL glacial acetic acid) and 1 vol. of 10 mM 2,4,6-tripyridyl-s-triazine (TPTZ) prepared in 40 mM HCl and 1 vol. of 20 mM $FeCl_3$. For each assay, 3 mL of FRAP reagent was mixed with 0.1 mL diluted synthesized compound. The mixture was shaken and incubated at 37 °C for 30 mins. Absorbance of the reaction mixture was measured at 593 nm. The standard calibration curve was carried out using Trolox and values expressed in terms of TEAC (μM). For calculating $EC_{50}$ values, aliquots of the different concentrations of the synthesized compounds were prepared and incubated as described above.

### Antibacterial activity

The antibacterial activities of synthesized compounds were determined using a modified Kirby-Bauer disk diffusion method [65]. Briefly, the test bacteria were grown in 10 mL of fresh Difco™ nutrient broth for 24 h. Optical density of test bacteria was measured using an Optizer 3220 (Double beam) UV–Vis spectrophotometer and found to be 0.7 at 595 nm. Aliquots of above bacterial suspension (100 μL) were then spread on Difco™ nutrient broth agar, which corresponded to the broth in which they were maintained. Two gram-negative bacteria *Escherichia coli* (KCTC-1924) and *Pseudomonas aeruginosa* (KCTC-2004) and two gram-positive bacteria *Staphylococcus aureus* (KCTC-1916) and *Bacillus cereus* (KCTC-1012) were obtained from the Korean Collection for Type Cultures (KCTC). The bacteria were incubated at 37 °C for 20–36 h, and then the diameters of the inhibition zones were measured in millimeters. Two mg of each test compound was dissolved in DMSO and further diluted to 100 μg/mL. Standard disks of ciprofloxacin served as positive controls and filter disks impregnated with DMSO as negative controls. Further, the depth of the agar in the plate is a factor to be considered in the disk diffusion method. Blank paper disks of diameter of 8.0 mm were impregnated with 10 μL of above diluted sample solutions. When a filter disk impregnated with a tested chemical is placed on agar, the chemical diffuses from the disk into the agar. The solubility of the chemical and its molecular size determine the size of the area of infiltration. When an organism is placed on the agar, it will not grow in the area around the disk, if the chemical is active. This area of no growth around the disk is referred to as the zone of inhibition.

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