Bull. Chem. Soc. Ethiop. **2024**, 38(4), 1077-1090.
DOI: https://dx.doi.org/10.4314/bcse.v38i4.21

ISSN 1011-3924
Printed in Ethiopia
Online ISSN 1726-801X

GREEN CHEMISTRY APPROACH FOR RAPID SYNTHESIS OF INDOL-3-YL-4H-PYRAN DERIVATIVES, BIOLOGICAL ASSESSMENTS, AND TOXICOLOGICAL ACTIVITIES AGAINST COWPEA APHID (APHIS CRACCIVORA)

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(Received January 12, 2024; Revised February 8, 2024; Accepted February 13, 2024)

ABSTRACT. From diverse or indole-3-carbaldehyde, certain unique indol-3-yl-4H-pyran derivatives were generated by condensing with different nucleophiles. IR, ^{1}HNMR, and elemental studies have all confirmed their chemical structures. The green catalyst for the formation of indol-3-yl-4H-pyran derivatives is a zinc-linked amino acid complex [Zn(L-proline)$_2$]. Furthermore, the environmental friendliness of this synthetic technique was investigated by assessing the reusability of the Zn(L-proline)$_2$ complex over five consecutive cycles with no significant loss of catalytic activity. This novel process has showed substantial advantages in terms of safety, simplicity, stability, mild conditions, a short reaction time, excellent yields, and good purity without the use of organic solvents. The antibacterial properties of the compounds produced were investigated and discovered to be promising. To determine whether compounds are appropriate as possible insecticidal agents, the toxicological activity of the synthesized compounds against *Cowpea aphid*, *Aphis craccivora*, was tested using leaf dip bioassay technique toxicity studies performed in the laboratory.

KEY WORDS: One pot synthesis, Zn(L-proline)$_2$, Indole-3-carbaldehyde, Aqueous media, Microwave irradiation, *Cowpea aphid,* toxicological activity

INTRODUCTION

L-Proline has been widely employed as a catalyst in enantioselective processes like aldol condensation [1]. Proline's catalytic action was linked to the production of an enamine with the nucleophile, which is similar to class I aldolases [2]. Darbre [3-5] reported complexes of several amino acids with Zn(II), which were also effectively employed to act as catalysts for the aldol condensation in water with a very high level of enantioselectivity. Because of the position of the secondary amine and carboxylate functional groups, proline is considered to be the most prominent amino acid for soft Lewis acid coordination with Zn. The Zn(L-proline)$_2$ complex may be easily made by combining zinc acetate Zn(AcO)$_2$ with the amino acid in methanol (MeOH) via triethylamine (Et_3N) as a basic catalyst [3, 4]. The water solubility of Zn(L-proline)$_2$ and other amino acid complexes is critical to their performance as a homogeneous catalyst in combined organic solvents and water, as it guarantees adequate solubility for both reagents involved in the reaction while also acting as a catalyst. Indole is a heterocyclic aromatic chemical molecule. It has a bicyclic structure with a six-membered benzene ring joined to a nitrogen-containing pyrrole ring. Indole is a common scent ingredient as well as a precursor to numerous medications. Indoles are compounds that include an indole ring. Notably, tryptophan, an indolic amino acid, is a

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precursor of the neurotransmitter serotonin. Furthermore, the indole ring is found in a variety of marine and terrestrial natural substances with beneficial biological effects. Indole, its bioisosters, and derivatives have anti-microbial properties toward gram-negative and gram-positive bacteria, as well as against the yeast *Candida albicans*, as well as antimicrobial agent activity toward *Enterobacter*, *Pseudomonas aeruginosa*, *E. coli*, and *Staphylococcus epidermidis*. Anti-inflammatory and analgesic [6-9], antifungal [10-12], antimicrobial [8, 9], anticancer [16, 17], anti HIV [18-20], antioxidant [21, 22], antiviral [23, 24], antitubercular [25, 26], antihypertensive [27-29], and antihistaminic [30]. All of these biological activities for indole and its derivatives, particularly insecticidal activity against some pests [31], lead to the selection of indole and its derivatives for further toxicological investigations to demonstrate their activity toward some dangerous pests such as *Cowpea aphid*. Aphids affect numerous agricultural crops in Egypt and across the world, inflicting significant economic loss because they take nutrients from diseased plants and spread viruses to them [31-33]. For many years, different insecticides have been used to control aphid pests, and some of them have a high insecticidal efficacy, such as neonicotinoids [34, 35], but the need for discovering new insecticides to control aphids and other pests is still present and growing, due to pest resistance and the search for new insecticides that are less harmful to humans.

RESULTS AND DISCUSSION

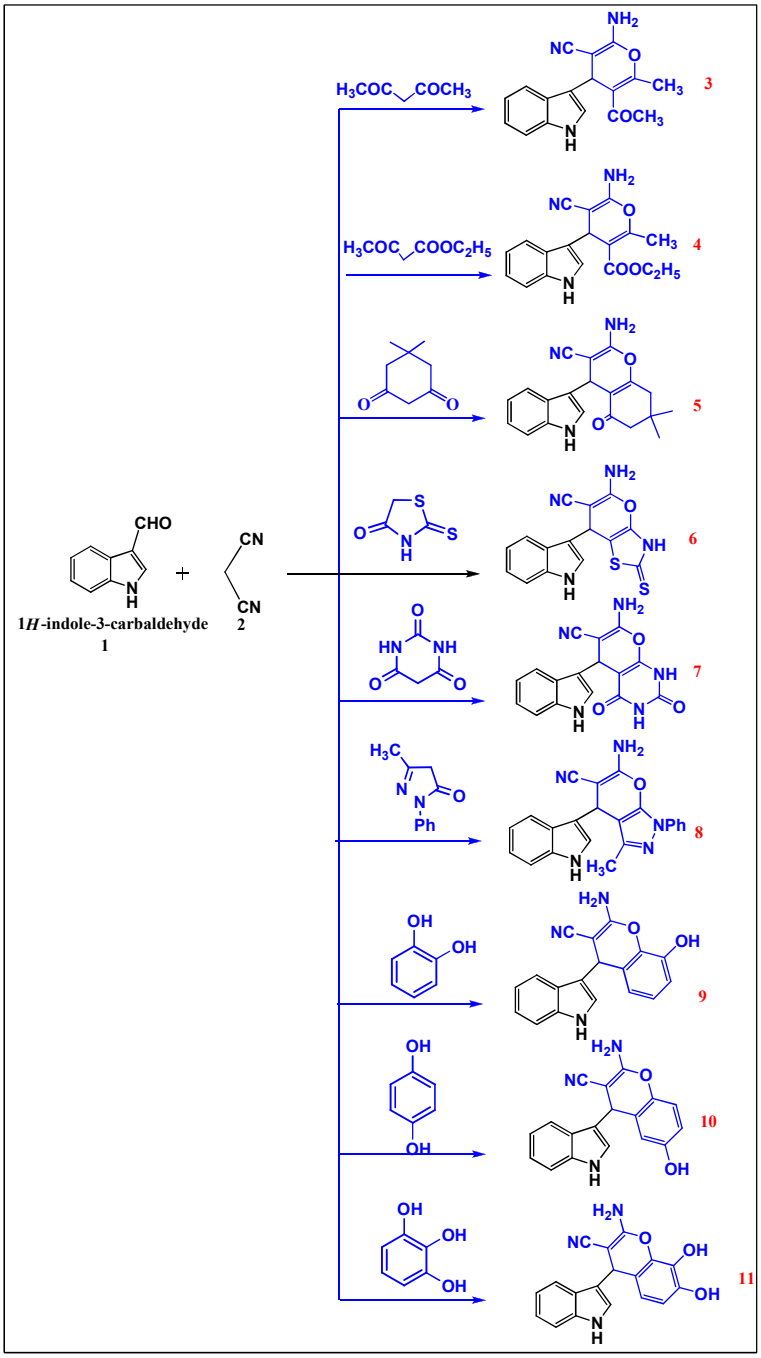
Chemistry

Green chemistry refers to the development of chemical products and processes that reduce or eliminate the use or production of hazardous substances. Green chemistry is concerned with reducing pollution and the amount of energy consumed in processes, thus in our study, we employed water as a green solvent for the reaction and use microwave radiation for minutes rather than usual heating for hours.

The importance of indole and indolyl compounds in multidisciplinary disciplines is widely acknowledged [35, 36]. This bicyclic arene has had a profound impact on pharmaceuticals, agrochemicals, and, more recently, organic electronics, and it continues to inspire enormous advances in organic synthesis [37-39]. The novel indol-3-yl-4H-pyran derivatives were synthesized using a catalytic quantity of [Zn(L-proline)$_2$] with MW irradiation (Scheme 1). In this study, these heterocyclic compounds were synthesized in a one-pot technique in a short amount of time with no intermediate isolation, and the reaction yields were enhanced. Spiro compounds were refined by filtering and recrystallization of crude products from a 70/30 ethanol/water solution (Scheme 1). Compound structures were determined using elemental analysis, IR, and ^{1}H NMR. For CHN analysis, all substances demonstrated good agreement between calculated and experimentally acquired results. Compound **3**'s 1H NMR spectrum, for example, showed a wide singlet at 10.39 ppm that was easily identified as NH-indole with multiplets (7.25-9.95) for the aromatic protons and NH_2 proton.

Bioassay results

Table 1 shows the toxicological activity of the produced chemicals. LC_{50} is the concentration that kills 50% of the insects in the experiment; it is a more valuable point statistically, for comparative purposes, and LC_{50} expressed on insecticidal efficacy, for calculating LC_{50} values using mortality data of aphids studied by probity analysis using some statistics (LDP-line) package.



Scheme 1. The synthesis of compounds **3-11**.

Table 1. Insecticidal effectiveness of compounds **3-11** towards cowpea aphid insects (nymphs and adults) after 24 hours of treatment.

	Nymphs			Adults		
Comp.	LC_{50} (ppm)	Slope	Toxic ratio	LC_{50} (ppm)	slope	Toxic ratio
3	0.84	0.7206 ± 0.3475	0.21	2.60	0.8447 ± 0.3243	0.20
4	0.98	0.5724 ± 0.3232	0.18	2.41	0.8276 ± 0.3271	0.21
5	0.81	0.7456 ± 0.3531	0.22	4.03	0.9036 ± 0.3201	0.13
6	0.38	0.5244 ± 0.3364	0.47	0.85	0.4337 ± 0.3119	0.61
7	0.44	0.4261 ± 0.4567	0.40	1.83	0.2584 ± 0.4224	0.28
8	0.18	0.5234 ± 0.3629	1	0.52	0.2616 ± 0.3026	1
9	0.79	0.7391 ± 0.3527	0.22	1.64	0.6524 ± 0.3209	0.31
10	0.76	0.8329 ± 0.3674	0.23	1.11	0.7024 ± 0.3356	0.46
11	0.63	0.5622 ± 0.3310	0.28	1.00	0.5450 ± 0.3200	0.52

Toxicological activity towards nymphs of Cowpea aphid after one day of treatment

Table and figure 1 results demonstrate that all nine tested compounds exhibit moderate or high toxicological action against *Cowpea aphids* (nymphs), with LC_{50} values ranging from 0.18 to 0.98 ppm after 24 hours of treatment. Specifically, the LC 50 values of compounds **8, 6, 7, 11, 9, 10, 5, 3**, and **4** are 0.18, 0.38, 0.44, 0.63, 0.76, 0.79, 0.84, 0.81, and 0.98 ppm, respectively. Among all tested compounds, indoles derivatives **8**, **6**, and **7** have the highest insecticidal activity, with LC_{50} values of 0.18, 0.38, and 0.44 ppm, respectively.

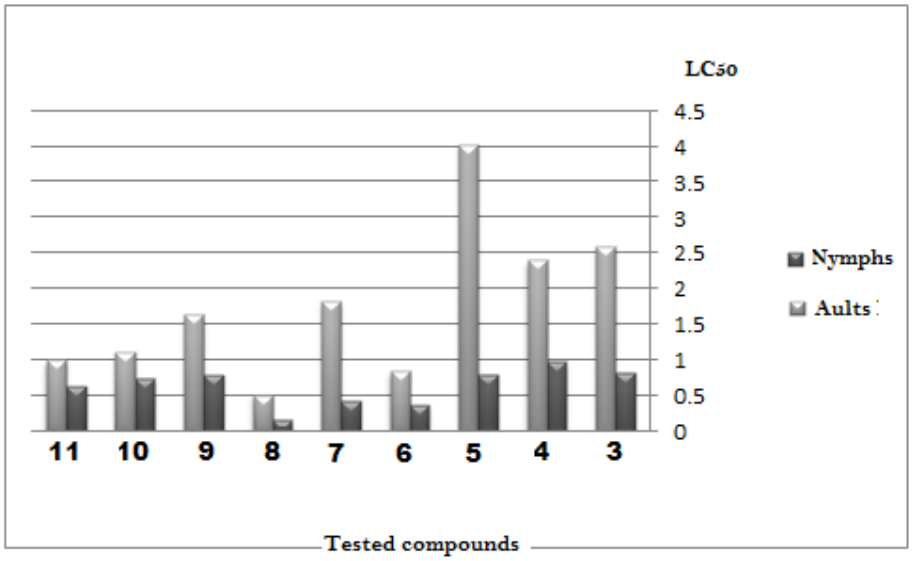


Figure 1. Insecticidal activity of selective indole derivatives **3-11** against the nymphs and adults of *Cowpea aphid.*

Toxicological activity towards adults of Cowpea aphid after 1 day of treatment

Table 1 and Figure 1 results demonstrate that all nine tested compounds exhibit moderate or high toxicological action against Cowpea aphids (adults), with LC_{50} values ranging from 0.18 to 0.98 ppm after 24 hours of treatment. Specifically, the LC_{50} values of compounds **8, 6, 7, 11, 9, 10, 5, 3** and **4** are 0.52, 0.85, 1.00, 1.11, 1.64, 1.83, 2.41, 2.60, 0.81, and 4.03 ppm, respectively. Among all tested compounds, indoles derivatives **8, 6, and 7** have the highest insecticidal activity with LC_{50} values of 0.52, 0.85, and 1.00 ppm, respectively.

EXPERIMENTAL

Materials and method

All the used chemical materials are from Aldrich, and they were used as received without any further purification. By using thin layer chromatography, all reactions were monitored. All melting points were recorded using the Kofler melting point device. IR spectroscopy was measured using a Bruker Alpha platinum-attenuated total reflection spectrophotometer. ^{1}H NMR and ^{13}C NMR data were detected using the Bruker BioSpin AG spectrometer at 400 MHz.

Compounds synthesis

Preparation of the Zn linked amino acid complex catalyst

In an around-bottomed flask, an amino acid (20 mmol), triethylamine (20 mmol), and ethanol (50 mL) mixture was agitated for 20 min at room temperature. Then, in a tiny amount of second water, $Zn(AcO)_2.6H_2O$ (10 mmol) was added dropwise. For 5-7 hours, the mixture was continually mixed at room temperature. The resultant white Zn-linked amino acid complex was filtrated and vacuum-dried for 4 hours at 70 °C. IR and ^{1}H NMR were used to characterize the complexes [4, 6]. Using open glass capillaries on a gallenkamp apparatus, the melting point was measured uncorrected. The IR spectra were obtained using potassium bromide pellets and a Shimaduz 408 instrument. The ^{1}H NMR (400 MHz) spectra in DMSO-d_6 were measured with a Burker AM 400 with TMS as an internal standard. The microwave irradiation was carried out in domestic microwaves at 900 W, 50 Hz, and 220-240 V.

Experimental

In a beaker, a mixture consisting of a solution of indole-3-carbaldehyde (1 mmol), malononitrile **2** (1 mmol), nucleophile (1 mmol), and $Zn(L\text{-proline})_2$ catalyst (0.10 mmol) was combined with H_2O (10 mL) and EtOH (5 mL) and M.W at 900 W for 5 min. Following the conclusion of the reaction, the dry product was filtered and cleaned by washing it several times with aqueous ethanol.

*5-Acetyl-2-amino-4-(1H-indol-3-yl)-6-methyl-4H-pyran-3-carbonitrile (**3**)*. Powder of light yellow, yield 80%, m.p.: 274 ºC; IR (KBr, cm^{-1}): 3442, 3232 and 3151 (NH_2, NH), 2210 (CN), 1706, 1688 (CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 2.26(s, 3H, CH_3-pyran), 2.30 (s, 3H, CH_3- acetyl), 6.13 (s, 1H, CH), 7.27 (t, 2H, CH-Ar),7.43 (d, 1H, CH-Ar), 8.12 (d, 1H, CH-Ar), 8.20 (s, 2H, NH2), 9.88 (s, 1H, CH-Ar), 12.06 (s, 1H, NH).

*Ethyl-6-amino-5-cyano-4-(1H-indol-3-yl)-2-methyl-4H-pyran-3-carboxylate (**4**)*. Powder of light yellow, **y**ield 80%, m.p.: 269 ºC; IR (KBr, cm^{-1}): 3445, 3323 and 3276 (NH_2, NH), 2192 (CN),

1714, 1675 (CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 1.23 (t, 3H, CH_3-ester), 1.34 (s, 3H, CH_3-pyran), 4.17 (q, 2H, CH_2-ester), 5.81 (s, 1H, CH), 7.39 (t, 2H, CH-Ar), 7.51(d, 1H, CH-Ar), 7.93 (d, 1H, CH-Ar), 8.40 (s, 2H, NH_2), 8.61 (s, 1H, CH-Ar), 12.46 (s, 1H, NH).

2-Amino-4-(1H-indol-3-yl)-7,7-dimethyl-5-oxo-5,6,7,8-tetrahydro-4H-chromene-3-carbonitrile ***(5)***. Powder of light yellow, yield 80%, m.p.: 295 ºC; IR (KBr, cm^{-1}): 3361, 3265 and 3158 (NH_2, NH), 2207 (CN), 1712, 1678 (CO). ^{1}H NMR (400 MHz, , ppm, DMSO, d_6): δ 0.93 (s, 3H, CH_3), 1.08 (s, 3H, CH_3), 2.13 (s, 1H, CH_2), 2.21 (s, 1H, CH_2), 3.13 (s, 2H, CH_2), 4.38 (s, 1H, CH), 6.92-7.37 (m, 7H, CH-Ar, NH_2), 10.90 (s, 1H, NH).

5-Amino-7-(1H-indol-3-yl)-2-thioxo-3,7-dihydro-2H-pyrano[2,3-d]thiazole-6-carbonitrile ***(6)***. Powdered orange, yield 80%; m.p.: 285 ºC; IR (KBr, cm^{-1}): 3343, 3252 and 3136 (NH_2, NH), 2215 (CN), 1674 (CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 7.25 (t, 3H, CH-Ar), 7.55 (d, 1H, CH-Ar), 7.85 (s, 1H, CH-Ar), 8.07 (d, 1H, CH-Ar), 8.54 (s, 1H, NH_2), 8.64 (s, 1H, NH_2), 12.37 (s, 1H, NH), 12.72 (s, 1H, NH).

7-Amino-5-(1H-indol-3-yl)-2,4-dioxo-2,3,4,5-tetrahydro-1H-pyrano[2,3-d]pyrimidine-6-carbonitrile ***(7)***. Powdered white, yield 75%, m.p.: > 300 ºC; IR (KBr, cm^{-1}): 3373, 3282 and 3151 (NH_2, NH), 2190 (CN), 1677 (CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 6.83 (d, 1H, CH-Ar), 6.99 (t, 1H, CH-Ar), 7.17 (m, 3H, CH-Ar), 7.37 (s, 3H, CH-Ar, NH_2), 10.40 (s, 1H, NH), 11.04 (s, 1H, NH), 12.22 (s, 1H, NH).

6-Amino-4-(1H-indol-3-yl)-3-methyl-1-phenyl-1,4-dihydropyrano[2,3-c]pyrazole-5-carbonitrile ***(8)***. Powdered orange, yield 80%, m.p.: 251 ºC; IR (KBr, cm^{-1}): 3358, 3264 and 3179 (NH_2, NH), 2211 (CN), 1683(CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 2.40 (s, 3H, CH_3), 7.22 (t, 1H, CH-Ar), 7.37 (d, 2H, CH-Ar), 7.50 (t, 2H, CH-Ar), 7.64 (d, 1H, CH-Ar), 8.09 (s, 2H, NH_2), 8.17 (s, 1H, CH-Ar), 8.23 (s, 1H, CH-Ar), 9.80 (s, 1H, CH-Ar), 12.47 (s, 1H, NH).

2-Amino-8-hydroxy-4-(1H-indol-3-yl)-4H-chromene-3-carbonitrile ***(9)***. Powdered beige, yield 80%, m.p.: 167 ºC; IR (KBr, cm^{-1}): 3430 (OH), 3373, 3309 and 3263 (NH_2, NH), 2203 (CN), 1684 (CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 7.23 (m, 3H, CH-Ar), 7.55 (d,2H, CH-Ar), 8.10 (d, 2H, CH-Ar), 8.33 (s, 1H, CH-Ar), 8.54 (s, 1H, NH_2), 8.71 (s, 1H, NH_2), 9.90 (s, 1H, CH-Ar), 12.08 (s, 1H, NH), 12.70 (s, 1H, OH).

2-*Amino-6-hydroxy-4-(1H-indol-3-yl)-4H-chromene-3-carbonitrile* ***(10)***. Powdered beige, yield 80%, m.p.: 183 ºC; IR (KBr, cm^{-1}): 3452(OH), 3373, 3263 and 3166 (NH_2, NH), 2211 (CN), 1687 (CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 7.25 (m, 3H, CH-Ar), 7.52 (d, 2H, CH-Ar), 8.10 (d, 2H, CH-Ar), 8.34 (s, 1H, CH-Ar), 8.50 (s, 1H, NH_2), 8.72 (s, 1H, NH_2), 9.87 (s, 1H, CH-Ar), 12.16 (s, 1H, NH), 12.71 (s, 1H, OH).

2-Amino-7,8-dihydroxy-4-(1H-indol-3-yl)-4H-chromene-3-carbonitrile ***(11)***. Powdered light green, yield 75%, m.p.: 173 ºC; IR (KBr, cm^{-1}): 3452 (OH), 3366, 3249 and 3179 (NH_2, NH), 2201 (CN), 1687(CO). ^{1}H NMR (400 MHz, ppm, DMSO, d_6): δ 7.21 (m, 3H, CH-Ar), 7.50 (d, 2H, CH-Ar), 8.09 (d, 2H, CH-Ar), 8.35 (s, 1H, NH_2), 8.53(s, 1H, NH_2), 8.69 (s, 1H, CH-Ar), 9.88 (s, 1H, NH), 12.16 (s, 2H, OH).

Insect collection and rearing

Cowpea aphid is found all over the world; in Egypt, it causes significant harm to commercial crops; until now, chemical pesticides have been the most effective

approach to protect crops. We investigated various novel indole compounds as possible insecticides against Cowpea aphid insects that were gathered, and all toxicity investigations were carried out in the insect laboratory at the Plant Protection Research Institute, Agricultural Research Center.

Laboratory bioassay

Evaluated the toxicity of the mentioned indole derivatives by leaf dip bioassay method under the same reported laboratory conditions [31-33]. The laboratory experiment recorded the concentrations of the synthesized compounds required to kill 50% (LC_{50}) of Cowpea aphids. LC_{50} used as expression for the toxicological activity, compound with low LC_{50} is the most effective compound. Solution of Each compound from Nine synthesized compounds was created by dissolving 0.1 g. in 10 mL acetone as a solvent (different concentration was prepared by diluting with distilled water) and 0.1% Tween-80 as a surfactant. While, acetone, distilled water and 0.1% Tween-80 used as control, 20 nymphs and 20 adults of similar size Cowpea aphids, immersed for ten seconds into each concentration of the compound solution (repeated 3 times). After treatment, the treated leaved to air dry for about 1/2 h. at room temperature. Once the treated aphids had dried, they moved to Petri dishes with a diameter of 9 centimeters and left for 24 hours, a 12-hour light/dark photoperiod, with a temperature of 22 ± 2 °C and relative humidity of 60 ± 5%. After about 24 hours, the aphid mortality rate counted by using a binocular microscope. Aphids that were cannot move forward in a coordinated manner considered dead. The resulting data corrected using Abbott's formula [34]. Used probit analysis program to calculate the median lethal concentrations (LC_{50}) and slope values for each one of the synthesized compounds [35].

Effect of solvent and catalyst

For the optimization of the reaction, we looked at a model reaction including indole-3-carbaldehyde (1 mmol), malononitrile **2** (1 mmol), and nucleophile (1 mmol) with varying concentrations (mol %) of the Zn(L-proline)$_2$ catalyst. The catalyst concentration has a major impact on product yields. It was discovered that increasing the catalyst loading from 7 to 10% resulted in a 94% yield. Adding extra catalyst does not enhance the process and does not speed up the reaction (Table 2).

Table 2. The amount of Zn(L-proline)$_2$ catalyst for the synthesis of 1H-indol-3-ylderivatives.

Entry	Cat. mol%	Yield %[a]	Entry	Cat. mol%	Yield %[a]
1	1	10	5	7	65
2	2	25	6	9	80
3	4	45	7	10	94
4	5	50	8	11	90

[a]Isolated yields based on **3**, [b]Reaction conditions: indole-3-carbaldehyde (1 mmol), malononitrile 2 (1 mmol), nucleophile (1 mmol) and Zn(L-proline)$_2$ catalyst (0.1 mmol) in mixture of water and ethanol was heated in microwave 5 min.

In this part, we looked at the role of solvents in this chemical process, investigating several solvents using a simulated reaction. Following a solvent screening, it was revealed that polar protic solvents (MeOH, EtOH, AcOH, and H_2O) produced typically high yields. The aprotic solvents (DCM, DMF, THF, CH_3CN, and $CHCl_3$) produced the lowest yields. We noticed, in particular, that a mixed solvent [H_2O and EtOH (v/v) (2/3)] improved the synthesis of the indole-3-carbaldehyde derivative 3 (Table 3).

Table 3. Effect of solvent for the synthesis of pyrimidine derivative 3[b].

Solvent	Time (min)	Yield (%)[a]
DCM	5	30
DMF	5	35
THF	5	45
CH_3CN	5	55
$CHCl_3$	5	65
MeOH	5	70
ACOH	6	67
EtOH	5	90
H_2O	5	80
H_2O/ EtOH	3	95

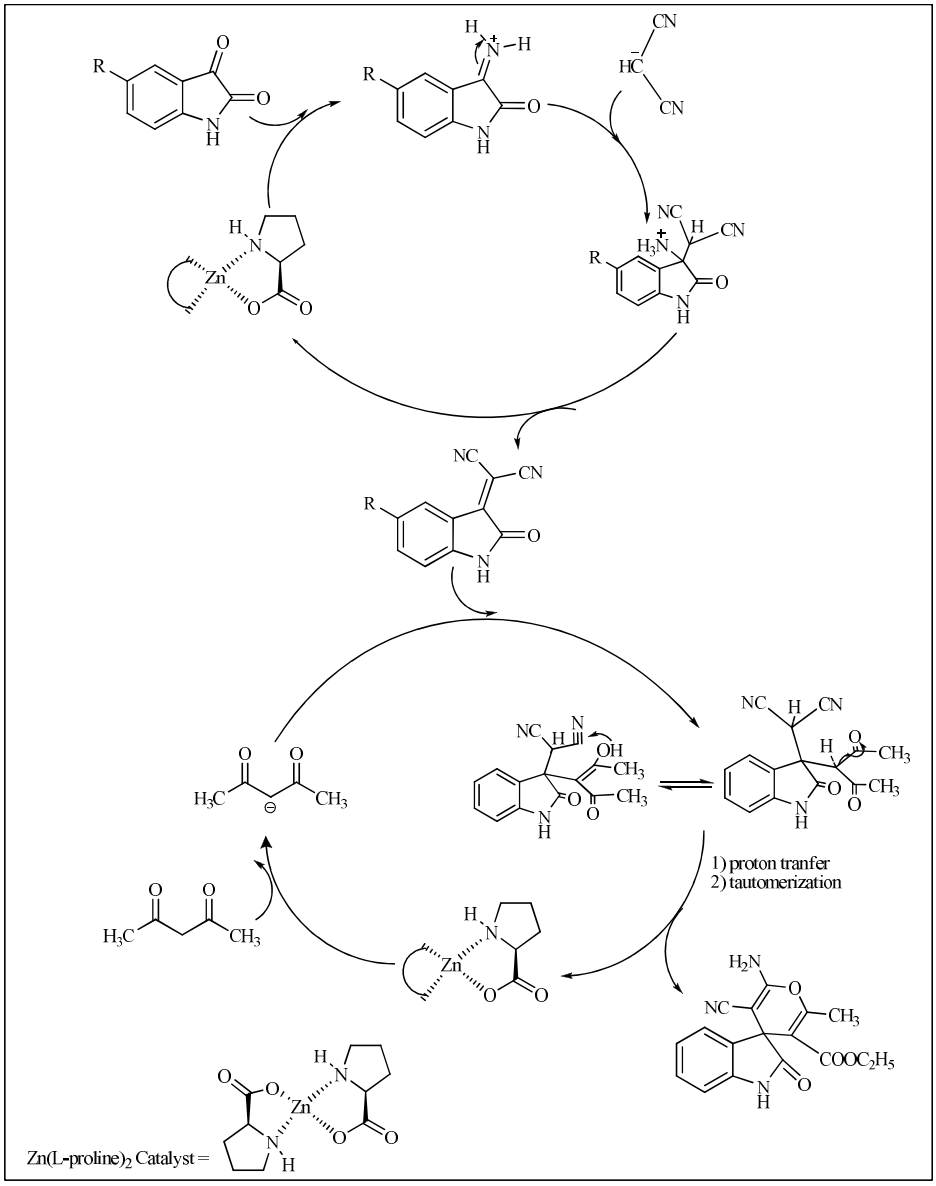
[a]Isolated yields based on **3**, [b]Reaction conditions: indole-3-carbaldehyde (1 mmol), malononitrile **2** (1 mmol), nucleophile (1 mmol) and Zn(L-proline)$_2$ catalyst (0.1mmol) in mixture of water and ethanol (2:3 ratio) was refluxed 3 h.

The proposed mechanism begins with a Zn(L-proline)$_2$ catalyst catalyzing the formation of an imine between indol-3-yl-4H-pyran and the catalyst's proline moiety, activating the carbonyl group, and follows by the nucleophilic addition of the carbanion of malononitrile **2** at the imine intermediate. Following the addition of active methylene, cyclization and tautomerization occur, culminating in the production of the product of interest (Scheme 2).

Antimicrobial activities

The biological experiment's antibacterial activity was diverse. Of the substances tested, compounds **6, 7,** and **8** were the most efficient against both gram-positive and gram-negative bacteria. It exhibited the best effectiveness at 20 mg/mL when compared with *ampicillin* and *Gentamicin* as a reference standard, with a percentage of 89%, 93%, and 91.6% inhibition zone on gram-positive bacteria and 98%, 94.2%, and 98.8% inhibition zone on gram-negative bacteria. The remaining compounds synthesized showed moderate to weak action, as stated in Table 4.

Compound **8**, on the other hand, has shown significant action against several fungal species at doses of 5, 10, and 20 mg/mL. Compound **7** exhibited the highest impact against *Cryptococcus neoformans* at a dosage of 20 mg/mL, with a 96.81% inhibition zone. Compound **8** had the strongest impact on *Candida albicans*, with an inhibition zone of 87.9%. Compounds **6, 7**, and **8** had a high impact on *Syncephalastrum racemosum,* with 90%, 95%, and 78.7%, respectively. At varying dosages, the other drugs displayed moderate to modest activity against fungal species as cleaned (Table 5).



Scheme 2. Proposed mechanism for synthesis of novel indol-3-yl-4H-pyran derivatives by Zn(L-proline)$_2$ catalyst.

Table 4. Antibacterial activity of the synthesized compounds (zone of inhibition in diameter in mm and (%) value).

Compound	Conc. (mg/mL)	Gram-positive bacteria inhibition zone diameter in mm and (%) value		Gram-negative bacteria inhibition zone diameter in mm and (%) value	
		Streptococcus pneumoniae	*Bacillus cereus*	*Pseudomonas aeruginosa*	*Escherichia coli*
3	20	21.4 ± 0.5 89.9%	19.2 ± 0.5 59.2%	14.1 ± 0.3 81.5%	13.6 ± 0.5 68.3%
4	20	18.4 ± 0.2 77.3%	18.8 ± 0.1 58.0%	15.1 ± 0.2 87.3%	14.8 ± 0.3 74.3%
5	20	17.2 ± 0.8 89.1%	18.8 ± 0.1 55.8%	13.9 ± 0.5 80.3%	16.8 ± 0.5 84.4%
6	20	21.2 ± 0.5 93.2%	26.1 ± 0.5 80.5%	17.0 ± 0.8 98.2%	18.7 ± 0.1 93.9%
7	20	22.2 ± 0.4 91.5%	24.7 ± 0.1 76.2%	16.3 ± 0.1 94.2%	16.2 ± 0.1 81.4%
8	20	21.8 ± 0.5 86.5%	25.2 ± 0.3 77.7%	17.1 ± 0.7 98.8%	17.9 ± 0.1 89.9
9	20	20.6 ± 0.5 86.5%	21.2 ± 0.7 65.4%	15.1 ± 0.2 84.3%	14.1 ± 0.5 70.8%
10	20	19.7 ± 0.1 82.75	19.2 ± 0.7 59.2%	12.3 ± 0.1 71.1%	12.1 ± 0.3 60.8%
11	20	21.4 ± 0.1 88.6%	20.2 ± 0.3 62.3%	14.7 ± 0.3 84.9%	13.0 ± 0.1 65.3%
Ampicillin	20	23.8 ± 0.2	32.4 ± 0.3	-	-
Gentamicin	20	-	-	17.3 ± 0.1	19.9 ± 0.3

Table 5. Antifungal activity of the synthesized compounds (zone of inhibition in diameter in mm and (%) value).

Compound	Conc. (mg/mL)	Fungi inhibition zone diameter in mm and (%) value			
		Aspergillus niger	*Syncephalastrum racemosum*	*Candida albicans*	*Cryptococcus neoformans*
3	20	16.3 ± 0.4 67.3%	14.9 ± 0.5 73.7%	13.1 ± 0.6 52.6%	17.2 ± 0.1 66.9%
4	20	16.9 ± 0.6 69.8%	13.8 ± 0.3 68.3	14.6 ± 0.7 58.6%	17.3 ± 0.4 67.3%
5	20	13.1 ± 0.3 54.1%	15.3 ± 0.3 75.7%	14.2 ± 0.7 57.0%	14.1 ± 0.4 54.8%
6	20	19.3 ± 0.1 79.75	18.2 ± 0.2 90.1%	19.0 ± 0.5 76.3%	22.9 ± 0.3 89.1%
7	20	21.9 ± 0.1 90.5%	19.2 ± 0.2 95.05	18.9 ± 0.5 75.9%	24.9 ± 0.3 96.8%
8	20	22.5 ± 0.5 92.9%	15.9 ± 0.4 78.7%	21.9 ± 0.6 87.9%	23.2 ± 0.1 90.2%
9	20	11.6 ± 0.5 47.9%	11.5 ± 0.3 56.9%	15.9 ± 0.3 63.8%	18.9 ± 0.6 73.5%
10	20	13.6 ± 0.5 56.1%	12.5 ± 0.3 61.8%	15.2 ± 0.3 61.0%	17.9 ± 0.6 69.6%
11	20	15.6 ± 0.5 64.4%	15.5 ± 0.3 76.7%	16.2 ± 0.3 65.0%	16.9 ± 0.6 65.7%
Amphotericin B	20	24.2 ± 0.1	20.2 ± 0.2	24.9 ± 0.1	25.7 ± 0.2

Structure-activity relationship

The goal of this work was to find novel compounds with insecticidal action against Cowpea aphid insects. Chemically, all derivatives consist of an indole ring linked to a pyran ring, and the only variation between the nine derivatives is the chemical groups attached to the pyran ring. As a result, the variation in toxicological activity may be attributable to differences in substituents connected to the pyran ring, as well as their location on the pyran ring. All Cowpea aphid chemical versions 6-Amino-4-(1H-indol-3-yl)-3-methyl-1-phenyl-1,4-dihydropyrano[2,3-c] are more efficient against nymphs than adults. The most powerful chemical is pyrazole-5-carbonitrile (**8**), which has LC_{50} values of 0.18 and 0.52 for nymphs and adults, respectively. This result might be attributed to the pyrazole ring, which is linked to the pyran ring. Numerous research investigations [c] have shown the insecticidal activity of the pyrazole ring. Another example is the compound 5-Amino-7-(1H-indol-3-yl)-2-thioxo-3,7-dihydro-2H-pyrano[2,3-d].7-Amino-5-(1H-indol-3-yl)-2,4-dioxo-2,3,4,5-tetrahydro-1H-pyrano[2,3-d] also demonstrated significant insecticidal action against nymphs and adults, with LC_{50} values of 0.38 and 0.85 ppm, respectively. Pyrimidine-6-carbonitrile (**7**), which has a pyrimidine ring linked to a pyran ring, displayed significant insecticidal activity against nymphs and adults with LC_{50} values of 0.44 and 1.00 ppm, respectively. Other chemicals' insecticidal effectiveness ranged from mild to strong. Other derivatives, such as those with the cyano group, amino, methyl, or ethyl esters, the chromene ring, and the hydroxide group, demonstrated mild to considerable insecticidal activity independent of the various substituents attached to the pyran ring. In order to increase insecticidal efficacy, nine derivatives have been ordered as follows: **8, 6, 7, 11, 10, 9, 5, 3,** and lastly, **4**.

CONCLUSIONS

Finally, the green bio-organic catalyst Zinc combined with the amino acid complex $Zn(L\text{-proline})_2$ has been demonstrated to be an efficient and environmentally friendly catalyst for the synthesis of novel indol-3-yl-4H-pyran derivatives using a one-pot multicomponent approach. This study has a number of significant benefits over other procedures, such as the prevention of hazardous organic solvents, low catalyst loading, harmless catalyst, short reaction time, high efficacy, mild reaction conditions, the use of an environmentally friendly solvent mixture of H_2O and C_2H_5OH, easy operability, a wide tolerance of starting materials, and great catalyst recoverability. According to the bioassay results, those compounds have potential for use as insecticides, and this work, together with previous investigations on the insecticidal activity of indole compounds, paves the way for future research. The antimicrobial activity of the produced compounds was examined and shown to be effective against bacteria and fungi. Comparing with the reference standards, compounds **6**, **7**, and **8** were the most promising, demonstrating potency against the tested microorganisms and meriting further preclinical research as an effective therapy for various microbial illnesses.

ACKNOWLEDGEMENT

This work was supported by the Deanship of Scientific Research, Vice Presidency for Graduate Studies and Scientific Research, King Faisal University, Saudi Arabia [GRANT5741].

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