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Synthesis and Herbicidal Activity of 4-Benzylidene-2-phenyl oxazol-5(4*H*)-one Derivatives using L-Proline as Catalyst

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A simple, efficient and environmentally benign method for the synthesis of 4-benzylidene-2-phenyl oxazol-5(4*H*)-one derivatives by the reaction of aromatic aldehydes and hippuric acid using acetic anhydride as dehydrating agent and L-proline as catalyst has been developed. L-proline acts as a highly efficient organocatalyst .It is thermally stable and can withstand harsh reaction conditions besides being easily available and environmentally non-hazardous. The structures of synthesized compounds were evaluated by FT-IR and ^{1}H-NMR spectroscopy. The present method is superior to the existing methods as it takes less reaction time, involves easy work up and affords products in excellent yield. The preliminary bioassay of synthesized compounds indicated that most of the compounds exhibited good seed germination inhibition activity against radish seed (*Raphanus sativus*).

Keywords: L-Proline, Organocatalyst, Oxazolone, Green method.

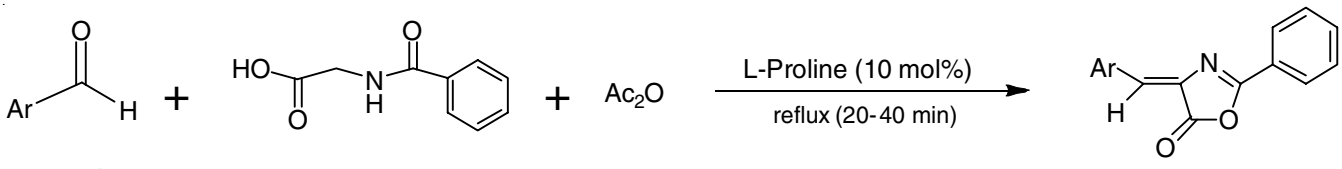
INTRODUCTION

During recent years, diversity oriented synthesis of small molecule libraries has become increasingly important to the development of new pharmaceuticals [1]. The first report on the synthesis of oxazolones using condensation of benzaldehyde with N-acetylglycine in the presence of sodium acetate as a basic catalyst and acetic anhydride as dehydrating agent was reported by Erlenmeyer [2]. Oxazolones are attractive building blocks in organic synthesis [3]. These are very promising intermediates for the preparation of amino acids [4], heterocyclic compounds [5], biosensors or coupling and photosensitive devices [6]. These compounds are biologically important as antimicrobials [7,8], antitumor [9], antioxidants [10], anti-inflammatory [11], anti-HIV [12,13] and tyrosinase inhibiting [14], antidiabetic [15], *etc*.

Recently, several methods for oxazolones synthesis such as cyclization [16], transition metal catalyzed synthesis [17-20], photocyclization [21], solid-phase synthesis [22] and cross-coupling of bromoalkyne with secondary *tert*-butyloxycarbamate [3] have been reported. Most of the methods used metal catalysts, in which separation is crucial because of residual toxicity of metals in the target compound. In past few years, promising methods for oxazolone synthesis were developed by Hashima *et al*. [23] and Gagosz *et al*. [3] wherein they reported the 3,5- disubtituted oxazolone *via* Au-catalyzed transformation of N-alkynyl ter-butyloxycarbamate. Yet, still now synthesis of 3,4,5-trisubtituted oxazolone remains challenging to the chemists. For the synthesis of oxazolone by the condensation of aldehyde, hippuric acid and dehydrating agent acetic anhydride various catalysts such as include Na_2CO_3 [24], $POCl_3$ [25], carbodiimides [26], polyphosphoric acid [27], Al_2O_3 [28], $Bi(OAc)_3$ [29], silica-supported heteropolyacids [30,31], $Yb(OTf)_3$ [32], $Ca(OAc)_2$ [33], supported KF [34], Fe_2O_3 [35], ZnO [36] and Al_2O_3-H_3BO_3 [37] have been used. Although these methods have some merits, but most of them still require high temperature, expensive and toxic reagents, hazardous transition metal catalysts.

Nowadays, strategy towards for developing the heterocyclic compound is favoured which is particularly important for the drug promoted molecules for biological screeing. Recently, organocatalysis emerged as an area of research for efficient and environmentally benign synthesis. In view of the above perception, the development of environmentally benign and green method for high selectivity and high yield is desirable. Herein, we report a concise and efficient method for the synthesis of oxazolone derivatives using L-proline as organocatalyst *via* aldol condensation reaction (**Scheme-I**). L-proline is easily available and inexpensive catalyst [38].

Herein, we report the protocol which offers several merits over other existing synthetic methods in many ways such as use of thermally stable and inexpensive catalyst, high yield (85-95%), short reaction times and easy workup. To the best of our knowledge, L-proline is being used as catalyst first time for the synthesis of oxazolones.



1a-i **2** Yield= 85-95 %

3 a-i

1a: Ar=H
1b: Ar= 3-OMe-C_6H_4
1c: Ar= 2,3-Cl-C_6H_3
1d: Ar= 3-Br-C_6H_4
1e: Ar= 4-Me-C_6H_4
1f: Ar= Furfural
1g: Ar= 3-Cl-C_6H_4
1h: Ar= 4-N$(CH_3)_2C_6H_4$
1i: Ar= 4-OH-3,4-(MeO)-C_6H_4

Scheme-I: Synthesis of oxazolone derivatives

EXPERIMENTAL

All chemicals were obtained from Hi-Media, SDS and used as received. FT-IR spectra were recorded on Bruker model-alpha spectrophotometer using KBr disc technique. The ^{1}H NMR spectra were recorded on Spectrometer JNM-ECS400 MHz instruments. Chemical data for protons are reported in parts per million (ppm) downfield from tetramethylsilane as reference to residual proton in NMR spectrophotometer using $CDCl_3$ (7.26 ppm) as solvent.

Hippuric acid (2 mmol, 0.358 g), benzaldehyde (2 mmol, 0.212 g), acetic anhydride (0.6 mL) and L-proline (10 mol %, 0.023 g) as catalyst were taken in a round bottom flask and stirred at 80 ºC. The completion of reaction was monitored by TLC (5 % ethyl acetate:hexane). After completion of reaction, the mixture was cooled to room temperature. Solid mass obtained was filtered and washed with water. The crude product was recrystallized from ethanol to afford the pure product. The identity and purity of the products are confirmed by IR, ^{1}H NMR spectroscopic analysis and melting point.

4-Benzylidene-2-phenyloxazol-5(4*H*)-one (3a): Light yellow crystalline solid; Yield: 92 %; m.p.: 165-167 ºC, lit. m.p.:167-168 ºC [28]; Anal calcd. (%) for $C_{16}H_{11}NO_2$: C, 77.10; H, 4.45; N, 5.62. Found: C, 77.20; H, 4.48; N, 5.30. FTIR (ν_{max}, cm^{-1}): 1790 (C=O), 1650 (C=N), 1550 (C=C), 1150 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.29 (1H, =CH), 8.25-8.22 (m, 4H, ArH), 8.21-7.57 (m, 3H, ArH), 7.52-7.50 (m, 3H, ArH).

4-(3-Methoxybenzylidene)-2-phenyloxazol-5(4*H*)-one (3b): Pale yellow solid; Yield: 91 %; m.p.: 101-103 ºC, lit. m.p.: 99-102 ºC [28]; Anal calcd. (%) for $C_{17}H_{13}NO_3$: C, 73.11; H, 4.69; N, 5.02. Found: C, 72.89; H, 4.73; N, 5.10. FTIR (ν_{max}, cm^{-1}): 1775 (C=O), 1660 (C=N), 1590 (C=C), 1225 (C-N), 1155 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 3.95 (3H, s, -OCH_3), 7.25 (1H, s, =CH), 7.29-7.59 (m, 5H, ArH), 7.65 (t, 1H, ArH), 8.18 (d, 1H, ArH), 8.20 (d, 1H, ArH), 8.22 (s, 1H, ArH).

4-(2,3-Dichlorobenzylidene)-2-phenyloxazol-5(4*H*)-one (3c): Light yellow solid; Yield: 86 %; m.p.: 170-173 ºC; Anal calcd. (%) for $C_{16}H_9NO_2Cl_2$: C, 60.40; H, 2.85; N, 4.40. Found: C, 61.10; H, 2.40; N, 4.80. FTIR (ν_{max}, cm^{-1}): 1790 (C=O), 1654 (C=N), 1554 (C=C), 1450, 1320, 1160 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.29 (1H, s, =CH), 7.38-7.67 (m, 5H, ArH), 8.20 (1H, dd, ArH), 8.22 (1H, d, ArH), 8.86 (1H, d, ArH).

4-(3-Bromobenzylidene)-2-phenyloxazol-5(4*H*)-one (3d): Light yellow solid; Yield: 89 %; m. p.: 146-148 ºC, lit. m.p.: 148-149 ºC [20]; Anal calcd. (%) for $C_{16}H_{10}NO_2Br$: C, 58.56; H, 3.07; N, 4.27. Found: C, 59.03; H, 3.02; N, 4.16. FT-IR (ν_{max}, cm^{-1}): 1790 (C=O), 1660 (C=N), 1595 (C=C), 1224 (C-O) lactone , 980, 870, 775, 672; ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.35 (s, 3H, -CH_3), 7.10 (s, 1H, =CH), 7.50-8.01 (m, 5H, ArH), 8.13-8.15 (dd, 2H, ArH), 8.38-8.44 (dd, 2H, ArH).

4-(4-Methylbenzylidene)-2-phenyloxazol-5(4*H*)-one (3e): Yellow solid; Yield: 94 %; m.p.: 144-146 ºC, lit. m.p.: 145-146 ºC [33]; Anal calcd. (%) for $C_{17}H_{13}NO_2$: C, 77.55; H, 4.98; N, 5.32. Found: C, 76.93; H, 5.23; N, 4.87. FTIR (ν_{max}, cm^{-1}): 1800 (C=O), 1660 (C=N), 1590 (C=C), 1220 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.47 (s, 3H, -CH_3), 7.31 (s, 1H, =CH), 7.56-7.58 (m, 2H, ArH), 7.33 (d, 2H, ArH), 8.13 (dd, 1H, ArH), 8.22 (d, 2H, ArH), 8.24 (dd, 2H, ArH).

4-[(Furan-2-yl)methylene]-2-phenyloxazol-5(4*H*)-one (3f): Light yellw solid; Yield: 88 %; m.p.: 171-173 ºC, lit. m.p.: 171 ºC [39]; Anal calcd. (%) for $C_{14}H_9NO_3$: C, 70.29; H, 3.79; N, 5.86. Found: C, 71.02; H, 3.42; N, 5.20. FTIR (ν_{max}, cm^{-1}): 1790 (C=O), 1660 (C=N), 1640 (C=N), 1520 (C=C), 1210 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.69 (1H, s, =CH), 7.21-7.60 (m, 5H, ArH), 7.71 (dd, 1H, ArH), 8.18 (d, 1H, ArH), 8.20 (d, 1H, ArH).

4-(3-Chlorobenzylidene)-2-phenyloxazol-5(4*H*)-one (3g): Yellow crystalline solid; Yield: 90 %; m.p.: 176-178 ºC; Anal calcd. (%) for $C_{16}H_{10}NO_2Cl$: C, 67.74; H, 3.55; N, 4.94. Found: C, 68.04; H, 3.17; N, 4.23. FTIR (ν_{max}, cm^{-1}): 1787 (C=O), 1651 (C=N), 1554 (C=C), 1158 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.22 (1H, =CH), 7.28-7.31 (m, 3H, ArH), 7.54 (t, 2H, ArH), 7.59-7.63 (m, 1H, ArH), 8.11 (d, 2H, ArH), 8.18-8.20 (d, 2H, ArH).

4-[4-(Dimethylamino)benzylidene]-2-phenyloxazol-5(4*H*)-one (3h): Red needle crystal; Yield: 96 %; m.p.: 213-215 ºC, lit. m.p.: 215 ºC [40]; Anal calcd. (%) for $C_{18}H_{16}N_2O_2$: C, 73.95; H, 5.52; N, 9.58. Found: C, 72.15; H, 4.96; N, 10.03. FTIR (ν_{max}, cm^{-1}): > 3612, 2875, 1827, 1643, 1523, 1368, 1157, 809, 687, 1755 (C=O), 1643 (C=N), 1597 (C=C), 1191 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 3.10 (s, 6H, $(CH_3)_2$), 6.72-6.75 (m, 2H, ArH), 7.21 (s, 1H, =CH), 7.48-7.55 (m,3H, ArH), 8.03-8.23 (m, 4H, ArH).

4-(4-Hydroxy-3,5-dimethoxybenzylidene)-2-phenyloxazol-5(4*H*)-one (3i): Yellow solid; Yield: 94 %; m.p.: 216-218 ºC, lit. m.p.: 217-219 ºC [20]; Anal calcd. (%) for $C_{18}H_{15}NO_5$: C, 66.46; H, 4.65; N, 4.31. Found: C, 67.03; H, 4.88; N, 4.93. FTIR (ν_{max}, cm^{-1}): 3565 (O-H), 1767 (C=O), 1651 (C=N), 1594 (C=C), 1222 (C-O) lactone; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.18 (s, 1H, =CH), 6.72 (s, 1H, O-H), 7.44-7.64 (m, 5H, ArH), 7.80-7.88 (m, 1H, ArH), 8.13 (d, 1H, ArH).

RESULTS AND DISCUSSION

For the optimization of amount of catalyst, the model reaction using reactants benzaldehyde (**1a**), hippuric acid (**2**) and acetic anhydride was carried out at 80 ºC using different amounts of catalyst (Table-1, entries 1-3). In starting 15 mol% catalyst was loaded, it took 20 min to complete the reaction and afforded 92 % yield. The reaction was carried out with 10 mol % amount of catalyst, took 20 min and afforded 92 % yield. Further decreasing the amount of catalyst to 5 mol% decreased the yield to 65% by taking a time of 50 min. Thus,10 mol % amount of catalyst was used for carrying out the reaction for other substrates.

TABLE-1
EFFECT OF CATALYST LOADING ON THE MODEL REACTION

Entry	Catalyst	Time (min)	Yield[a] (%)
1	15	20	92
2	10	20	92
3	5	50	65

[a]Isolated yield of products

In order to optimize the temperature, the model reaction was carried out at different temperatures (Table-2, entries 1-4). Initially, the reaction was carried out at ambient temperature it took 5 h to complete the reaction affording 70 % yield. By carrying out reaction at 80 ºC, there was significant decrease in time. The reaction completed within 20 min affording 92 % yield. Further increase in temperature to 90 ºC did neither improved yield nor decreased the time. Thus, we selected 80 ºC for carrying out the reaction.

TABLE-2
EFFECT OF TEMPERATURE ON THE MODEL REACTION

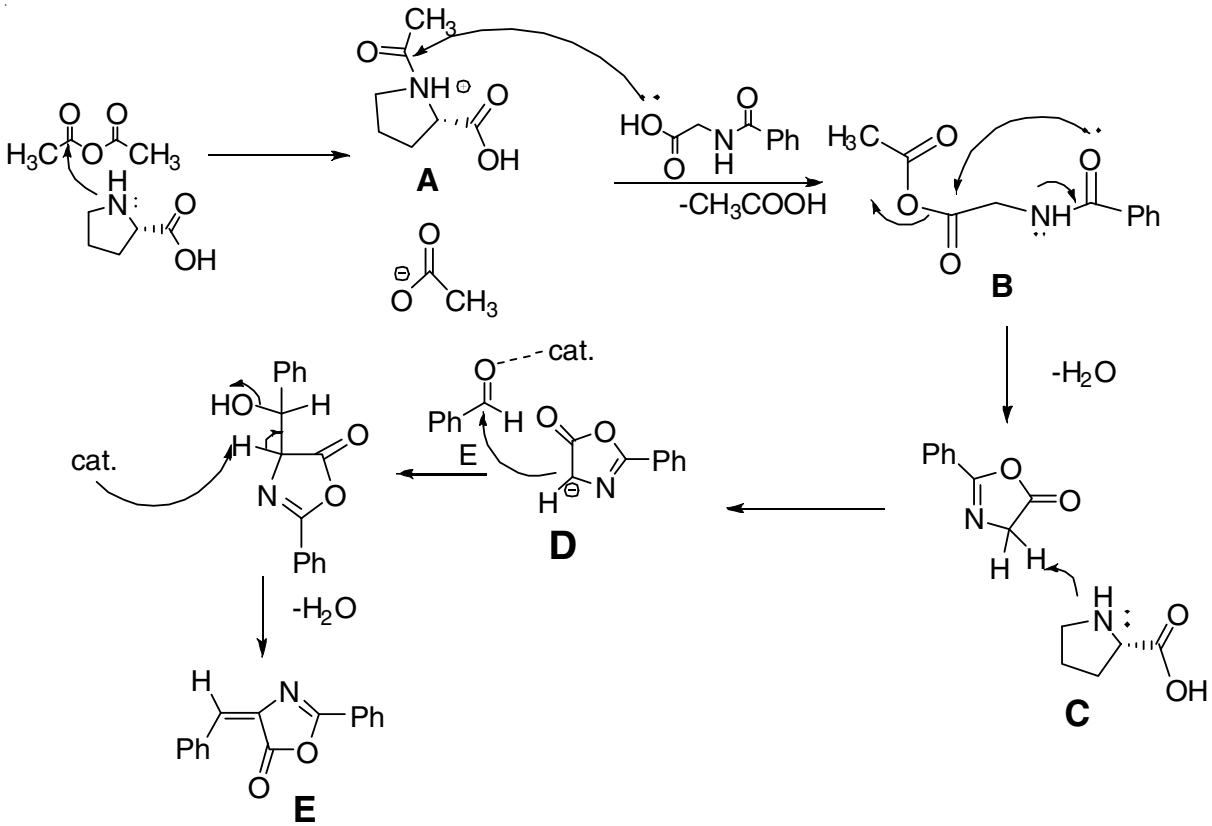
Entry	Temp. (°C)	Time (min)	Yield[a] (%)
1	Room temperature	300	70
2	60	80	80
3	80	20	92
4	90	20	92

[a]Isolated yield of products

In order to test the substrate scope of aldehydes, the reaction was carried out with different aromatic aldehydes having *o*-, *m*- and *p*-directing substituents on benzene ring. Aldehydes bearing various functional groups, such as -OMe, -F, -Br, -OH, -Cl, $N(CH_3)_2$, $-CH_3$, produced the corresponding oxazolones (**3a-i**). Aromatic aldehydes having *o*- and *p*-directing substituents gave slightly higher yield in comparison to the compounds having *m*-directing substituents (Table-3, entries 1-9). The substituents on the aromatic aldehydes were found to have a minimal electronic effects.

A plausible mechanism of reaction catalyzed by L-proline is shown in **Scheme-II**. The complex A formed by interaction of acetic anhydride and L-proline initially activates the carboxyl group of hippuric acid to give compex B which cyclizes to give intermediate (C). Formation of intermediate (C) has been supported by earlier workers [41,42]. L-proline abstracts hydrogen ion from intermediate (C) to convert it to nucleophile (D). Nucleophile (D) attacks on catalyst activated carbonyl carbon of aldehyde. Finally removal of water molecule gives the product (E).

Herbicidal activity of synthesized compounds: The pre-emergence herbicidal activity of synthesized oxazolone derivatives was evaluated against radish seeds (*Raphanus sativus*).



Scheme-II: Plausible mechanism for synthesis of oxazolone derivatives using L-proline as catalyst

TABLE-3
SYNTHESIS OF 4-ARYLIDENE-2-PHENYL-5(4*H*)-OXAZOLONES DERIVATIVES[a] **3(a-i)**

Entry	Aldehyde	Products	Time[b] (min)	Yield[c] (%)
1	**1a**	**3a**	18	92
2	**1b**	**3b**	22	91
3	**1c**	**3c**	12	86
4	**1d**	**3d**	25	89
5	**1e**	**3e**	16	94
6	**1f**	**3f**	23	88
7	**1g**	**3g**	17	90
8	**1h**	**3h**	18	96
9	**1i**	**3i**	40	94

[a]Reaction conditions: aldehyde (**1a,** 2 mmol), hippuric acid (**2**, 2 mmol), acetic anhydride (0.6 mL) and L-proline (10 mol %), 80 °C.
[b]Reaction progress by TLC.
[c]Isolated yield of products.

The herbicide butachlor was taken as standard. Each of the compound was first dissolved in minimum amount of ethanol, two drops of 1 % Tween 20 were added to the solution and made up with distilled water to prepare a stock solution of concentration 500 mg/L. Then by serial dilution with distilled water, solutions of concentration 0.02, 0.01 and 0.005 M were prepared. Three replicates were used for each treatment. Prior to putting the seeds in Petri-dish for germination, the seed of radish (*Raphanus sativus*) were sterilized in 1% sodium hypochlorite solution for 30 min. Ten seeds of raddish were taken in each petri dish containing a piece of filter paper and 7 mL solution of each test solution. Distilled water was used as control. The petri dishes were closed by parafilm to prevent the loss of moisture. The petri dishes were kept in incubator at 25 ± 2 ºC for germination. The seed germination was assessed by taking observations after 120 h of incubation. The number of seeds germinated were counted. Shoot and rooth lengths were also measured.

The germination inhibition, root length inhibition and shoot length inhibition were subjected to analysis of variance (ANOVA) by STPR3 software and critical differences (CDs) at $p > 0.05$ were calculated.

The mean percent germination inhibition values of synthesized compounds and standard (butachlor) with their CD values are presented in Table-4, which clearly indicates that with increase in concentration from 0.005 M to 0.01 M for all the compounds including standard except compounds **3b** and **3i**, there is significant increase in activity while with increase in concentration from 0.01 M to 0.02 M there is no significant increase in activity for the compounds except compounds **3d**, **3e** and **3h**. Though for standard significant increase in activity is observed. So, concentration 0.01 M may be taken to get limiting activity for the compounds mentioned previously. Comparison of activity of compounds at 0.005 M concentration with standard revealed that only the compound **3h** exhibited activity (46.7 %) at par with standard (56.7 %). At concentration 0.01 M all the compounds exhibited significant activity at par with standard (66.7 %). At concentration 0.02 M also all the compounds except compound **3e** (56.7%), significant activity at par with standard (80 %). It may be inferred that all the compounds exhibit significant activity at par with standard at concentration 0.01 M and this concentration may be taken as basal concentration for achieving limiting activity.

TABLE-4
MEAN PERCENT GERMINATION INHIBITION VALUES OF SYNTHESIZED COMPOUNDS

Compd. code	0.005 M	0.01 M	0.02 M	CD at 5%
3a	40.0	70.0	86.7	21.0
3b	40.0	66.7	86.7	27.4
3c	30.0	63.3	80.0	17.6
3d	40.0	63.3	76.7	24.0
3e	36.7	46.7	56.7	NS
3f	40.0	70.0	83.3	17.6
3g	30.0	60.0	76.7	17.6
3h	46.7	56.7	70.0	17.6
3i	30.0	56.7	76.7	22.1
Stand. (Butachlor)	56.7	66.7	80.0	9.4
CD at 5%	15.2	20.4	15.6	–

CD = Critical difference

The mean per cent shoot length inhibition values of synthesized compounds along with standard are presented in Table-5. The results revealed that with increase in concentration from 0.005 M to 0.01 M and then to 0.02 M, there is significant increase in activity for all the compounds except compounds **3e** and **3h** for which the data are not statistically significant. Comparison of the shoot inhibition values of the compounds with standard at concentration 0.005 M indicates that all the compounds exhibit significantly lower activity in comparison to standard (75.2 %). At concentration 0.01 M, all the compounds exhibit significantly lower activity in comparison to standard (83.6 %) while at concentration 0.02 M for all compounds except compound **3e** (83.7 %) and compound **3h** (85.2%) again significant activity at par with standard (94.4%) is observed.

TABLE-5
MEAN PERCENT SHOOT LENGTH INHIBITION VALUES OF SYNTHESIZED COMPOUNDS

Compd. code	0.005 M	0.01 M	0.02 M	CD at 5%
3a	51.5	68.1	95.2	2.1
3b	57.0	70.3	95.1	2.7
3c	54.2	72.9	96.7	1.8
3d	60.5	74.5	100	2.4
3e	54.7	67.2	83.7	NS
3f	58.7	74.2	95.6	1.8
3g	57.0	70.1	93.9	1.8
3h	54.5	60.6	85.2	NS
3i	58.4	71.6	96.3	2.2
Stand. (Butachlor)	75.2	83.6	94.4	0.9
CD at 5 %	10.7	6.8	4.3	–

The anomaly of getting higher activity at lower concentration and *vice versa* may be explained by keeping in view the fact that some compounds may exhibit plant growth regulatory activity depending upon the dose [43].

The mean percent root length inhibition values of synthesized compounds including standard are presented in Table-6. The results indicates that with increase in concentration from 0.005 to 0.01 M and then to 0.02 M, there is significance increase in concentration for all the compounds including standard. Comparison of activity of synthesized compounds with standard at 0.005 M concentration revealed that all the compounds except compound **3d** (76.1%) exhibit significantly lower activity in comparison to standard (73.7 %). At concentration 0.01 M, the

TABLE-6
MEAN PERCENT ROOT LENGTH INHIBITION VALUES OF SYNTHESIZED COMPOUNDS

Compd. code	0.005 M	0.01 M	0.02 M	CD at 5%
3a	55.7	83.2	96.7	4.3
3b	42.7	68.6	97.0	4.9
3c	63.7	83.9	93.6	7.9
3d	76.1	85.3	94.9	1.3
3e	44.9	55.6	61.2	3.9
3f	46.9	67	86.1	6.6
3g	43	65.2	83.1	1.4
3h	48.3	68	93.6	3.0
3i	65.7	74.7	85.5	1.2
Stand. (Butachlor)	73.7	81.9	94.9	0.6
CD at 5 %	3.1	2.0	5.0	–

compounds **3a** (83.2 %), **3c** (83.9 %) and **3d** (85.3 %) exhibit activity at par with standard (81.9 %) while the remaining compounds exhibit significantly lower activity in comparison to the standard. At concentration 0.02 M, the compounds **3a** (96.7 %), **3b** (97.0 %), **3c** (93.6 %), **3d** (94.9 %) and **3h** (93.6 %) exhibit activity at par with standard (94.9 %) while compounds **3e** (61.2%), **3f** (86.1%), **3g** (83.1 %) and **3i** (85.5 %) exhibit activity significantly lower in comparison to the standard.

In conclusion, pre-emergence seed germination inhibition studies clearly indicate that most of the compounds are at par with standard butachlor in activity. So, these compounds may be developed as potential herbicides. Moreover, with variation in concentration, shoot growth is not inhibited with increase in concentration which indicates plant growth regulatory activitiy present in compounds. Though, it needs further research to ascertain plant growth regulatory activity of these compounds.

Conclusion

A novel and eco-freiendly approach for the synthesis of 4-benzylidene-2-phenyloxazol-5(4*H*)-one derivatives in excellent yield using L-proline as convenient and inexpensive organocatalyst is reported. The preliminary bioassay showed that most of the synthesized compounds had good to excellent seed germination inhibition activity against radish seed (*Raphanus sativus*).

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