Asian Journal of Chemistry; Vol. 24, No. 6 (2012), 2516-2518

ASIAN JOURNAL OF CHEMISTRY

www.asianjournalofchemistry.co.in

Optimization and Antifungal Activity of Chalcone Analogues

XIAORONG TANG*, SHAOLING CHEN and LING WANG

School of Physics and Chemistry, Xihua University, Chengdu 610039, P.R. China

*Corresponding author: Tel: +86 28 87720208; E-mail: tangxiaorong0901@163.com

(*Received*: 29 April 2011; *Accepted*: 11 January 2012) AJC-10934

Using chalcone as a leading compound, a series of its analogues (compounds **1-16**) were designed and synthesized. Their activity of antipathogenic fungi of plants has been evaluated in the laboratory. The results showed that these compounds had certain antifungal activity against *Sclerotinia sclerotiorum* and *Botrytis cinerea*. Among them, the inhibition of growth for 2-chloro-N-phenylbenzamide (compound **2**) reached 92.6 and 90.1 % at a concentration of 100 mg L^{-1}, respectively.

Key Words: Antifungal activity, Chalcone analogues, Synthesis.

INTRODUCTION

In agriculture, various diseases of plants caused by pathogenic fungi are one of major economic damage in the world[1,2]. Many kinds of fungicides have been used to control them[3]. However, their over use has brought about side effects, such as the contamination of the environment and occurrence of resistance[4]. Therefore, novel fungicides which are both active against these diseases and safe to human being and the environment are necessary.

Botanical fungicides play an important role in controlling pathogenic fungi of plants[5-7]. Nevertheless, they have some drawbacks such as low yield, instability, complex components and limited antifungal spectrum. Thus, natural compounds with antifungal activity from plants are often used as a lead structure to synthesize new fungicides[8].

Chalcone (Fig. 1) is a natural compound existing in many plants and has many bioactivities[9-17]. Its structure is simple and its chemical synthesis is easy. Therefore, in the present study, a series of compounds have been designed and synthesized based on it. Meanwhile, their antifungal activity has been determined in the laboratory.

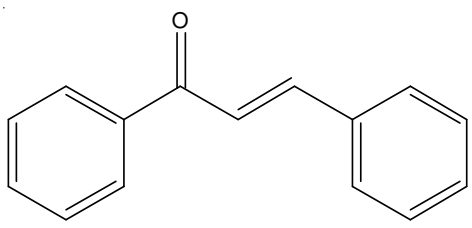

Fig. 1. Chemical structure of chalcone

EXPERIMENTAL

Sclerotinia sclerotiorum and *Botrytis cinerea* were obtained from the Chinese Academy of Agricultural Sciences. They were preserved at 4 ºC. All chemicals and solvents were purchased from commercial sources unless specified otherwise. IR spectra were recorded on a Thermofisher Nicolet-6700 spectrophotometer. ^{1}H NMR spectra were taken on a Varian Unity Inova-400 instrument using deuteron-chloroform as the solvent.

Synthesis of chalcone analogues: The chalcone analogues were synthesized according to the route shown in Fig. 2 and their yield was not optimized. Benzenamine or its derivatives (0.02 mol) and pyridine (0.02 mol) were dissolved in CH_2Cl_2 (25 mL). The mixture was stirred and heated to 35-45 ºC. Benzoyl chloride or its substituent (0.02 mol) was slowly to the mixture under stirring until the reaction was complete. The precipitate was filtered and washed with distilled water. The pure compounds **1-16** were obtained by re-crystallization in anhydrous ethanol. The details of the compounds (**1-16**) are shown in Table-1.

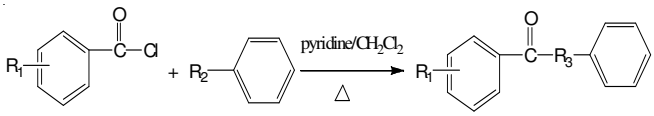

Fig. 2. Synthesis of chalcone analogues **1-16**

Compound 1: White powder, yield: 83.4 %, m.p. 155-157 ºC; IR (KBr, ν_{max}, cm^{-1}): 3344, 3025, 1655, 1600, 1578, 1535, 1492, 1439, 1301, 750, 716, 691, 616, 584; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.88 (s, 1H), 7.86 (d, J = 7.6 Hz,

2H), 7.65 (d, *J* = 7.6 Hz, 2H), 7.56 (t, *J* = 7.4 Hz, 1H), 7.49 (t, *J* = 7.4 Hz, 2H), 7.38 (t, *J* = 7.8 Hz, 2H), 7.16 (t, *J* = 7.4 Hz, 1H).

Compound 2: Grey crystal, yield: 86.3 %, m.p. 114-116 ºC; IR (KBr, ν_{max}, cm^{-1}): 3239, 3042, 1641, 1600, 1548, 1489, 1444, 1330, 1050, 779, 761, 717, 692, 652, 588; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.87 (s, 1H), 7.77 (dd, *J* = 1.6 Hz, 2.0 Hz, 1H), 7.65 (d, *J* = 8.0 Hz, 1H), 7.47 (d, *J* = 1.6 Hz, 1H), 7.45 (s, 1H), 7.44 (d, *J* = 1.6 Hz, 1H), 7.42 (d, *J* = 2 Hz, 1H), 7.40 (d, *J* = 8.0 Hz, 1H), 7.38 (d, *J* = 7.2 Hz, 1H), 7.18 (t, *J* = 7.2 Hz, 1H).

Compound 3: White crystal, yield: 81.2 %, m.p. 195-196 ºC; IR (KBr, ν_{max}, cm^{-1}): 3351, 3057, 1654, 1599, 1571, 1530, 1487, 1440, 1326, 1093, 848, 758, 729, 690, 624; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.82 (d, *J* = 8.4 Hz, 2H), 7.76 (s, 1H), 7.63 (d, *J* = 7.6 Hz, 2H), 7.47 (d, *J* = 8.4 Hz, 2H), 7.39 (t, *J* = 7.8 Hz, 2H), 7.17 (t, *J* = 7.4 Hz, 1H).

Compound 4: Grey powder, yield: 78.5 %, m.p. 210-212 ºC; IR (KBr, ν_{max}, cm^{-1}): 3322, 1652, 1598, 1533, 1518, 1494, 1441, 1348, 1326, 761, 744, 724, 694, 666; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 8.36 (d, *J* = 8.4 Hz, 2H), 8.05 (d, *J* = 8.4 Hz, 2H), 7.81 (s, 1H), 7.65 (d, *J* = 7.6 Hz, 2H), 7.43 (t, *J* = 7.6 Hz, 2H), 7.22 (t, *J* = 7.6 Hz, 1H).

Compound 5: Colourless crystal, yield: 60.6 %, m.p. 110-112 ºC; IR (KBr, ν_{max}, cm^{-1}): 3325, 3060, 2928, 1643, 1602, 1577, 1548, 1497, 1452, 1316, 727, 694, 669, 601; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.79 (d, *J* = 7.6 Hz, 2H), 7.50 (t, *J* = 7.5 Hz, 1H), 7.43 (t, *J* = 7.8 Hz, 2H), 7.26-7.37 (m, 5H), 6.41 (s, 1H), 4.66 (d, *J* = 6 Hz, 2H).

Compound 6: White crystal, yield: 78.6 %, m.p. 161-163 ºC; IR (KBr, ν_{max}, cm^{-1}): 3313, 3031, 1639, 1595, 1488, 1451, 1322, 1093, 762, 737, 712, 695, 671; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.73 (d, *J* = 8.4 Hz, 2H), 7.40 (d, *J* = 8.4 Hz, 2H), 7.31-7.37 (m, 5H), 6.39 (s, 1H), 4.64 (d, *J* = 5.6 Hz, 2H).

Compound 7: White crystal, yield: 75.9 %, m.p. 163-165 ºC; IR (KBr, ν_{max}, cm^{-1}): 3313, 3031, 1639, 1594, 1555, 1488, 1451, 1322, 1093, 850, 762, 737, 712, 694; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.73 (d, *J* = 8.8 Hz, 2H), 7.40 (d, *J* = 8.8 Hz, 2H), 7.29-7.36 (m, 4H), 6.44 (s, 1H), 4.63 (d, *J* = 5.6 Hz, 2H).

Compound 8: White crystal, yield: 39.5 %, m.p. 139-140 ºC; IR (KBr, ν_{max}, cm^{-1}): 3279, 3035, 2935, 2883, 1630, 1597, 1536, 1514, 1454, 1346, 1318, 872, 754, 726, 698, 623, 584; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 8.28 (d, *J* = 9.2 Hz, 2H), 7.95 (d, *J* = 8.8 Hz, 2H), 7.33-7.40 (m, 5H), 6.50 (s, 1H), 4.67 (d, *J* = 5.2 Hz, 2H).

Compound 9: White crystal, yield: 84.2 %, m.p. 113-114 ºC; IR (KBr, ν_{max}, cm^{-1}): 3344, 3026, 1640, 1602, 1579, 1545, 1496, 1455, 1314, 749, 722, 695, 667, 598; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.70 (d, *J* = 7.3 Hz, 2H), 7.49 (t, *J* = 7.4 Hz, 1H), 7.42 (t, *J* = 7.7 Hz, 2H), 7.35 (t, *J* = 7.6 Hz, 2H), 7.26-7.29 (m, 3H), 6.14 (s, 1H), 3.74 (q, 2H), 2.95 (t, *J* = 6.9 Hz, 2H).

Compound 10: Yellow crystal, yield: 86.3 %, m.p. 107-109 ºC; IR (KBr, ν_{max}, cm^{-1}): 3281, 3024, 1662, 1593, 1556, 1494, 1453, 1307, 1062, 786, 753, 726, 696, 646; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.60 (d, *J* = 7.2 Hz, 2H), 7.24-7.38 (m, 8H), 6.22 (s, 1H), 3.75 (q, 3H).

Compound 11: Yellow crystal, yield: 80.0 %, m.p. 127-129 ºC; IR (KBr, ν_{max}, cm^{-1}): 3346, 3028, 1638, 1596, 1570, 1541, 1487, 1455, 1316, 1094, 845, 762, 752, 722, 699, 625, 599; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 8.08 (d, *J* = 8.4 Hz, 2H), 7.62 (d, *J* = 8.4 Hz, 2H), 7.52 (d, *J* = 8.8 Hz, 1H), 7.32-7.39 (m, 2H), 7.25 (q, 2H), 6.08 (s, 1H), 3.72 (q, 2H), 2.93 (t, *J* = 6.8 Hz, 2H).

Compound 12: Yellow powder, yield: 78.8 %, m.p. 145-146 ºC; IR (KBr, ν_{max}, cm^{-1}): 3256, 3031, 1636, 1597, 1557, 1515, 1493, 1454, 1348, 1303, 827, 752, 704, 623, 578; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 8.26 (d, *J* = 8.8 Hz, 2H), 7.83 (d, *J* = 8.8 Hz, 2H), 7.23-7.36 (m, 5H), 6.20 (s, 1H), 3.75 (q, 2H), 2.96 (t, *J* = 6.8 Hz, 2H).

Compound 13: White crystal, yield: 88.0 %, m.p. 179-180 ºC; IR (KBr, ν_{max}, cm^{-1}): 3065, 3026, 1654, 1590, 1489, 1447, 786, 769, 755, 725, 703, 691; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 7.46 (d, *J* = 7.2 Hz, 1H), 7.45 (d, *J* = 1.2 Hz, 1H), 7.29 (t, *J* = 7.6 Hz, 4H), 7.27 (d, *J* = 3.6 Hz, 2H), 7.22 (d, *J* = 7.6 Hz, 1H), 7.20 (d, *J* = 1.2 Hz, 1H), 7.19 (d, *J* = 4.0 Hz, 1H), 7.15 (d, *J* = 7.6 Hz, 4H).

Compound 14: Yellow powder, yield: 85.4 %, m.p. 137-139 ºC; IR (KBr, ν_{max}, cm^{-1}): 3064, 3024, 1656, 1590, 1491, 1452, 1051, 769, 758, 748, 718, 699, 693; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 8.04 (d, *J* = 8.0 Hz, 1H), 7.53 (d, *J* = 3.6 Hz, 1H), 7.39 (d, *J* = 3.6 Hz, 1H), 7.37 (t, *J* = 2.8 Hz, 1H), 7.35 (d, *J* = 3.2 Hz, 1H), 7.34 (d, *J* = 3.6 Hz, 1H), 7.21 (d, *J* = 2.8 Hz, 1H), 7.20 (d, *J* = 2.8 Hz, 1H), 7.19 (d, *J* = 3.2 Hz, 1H), 7.18 (t, *J* = 2.4 Hz, 1H), 7.16 (d, *J* = 5.2 Hz, 1H), 7.18 (d, *J* = 2.4 Hz, 1H), 7.14 (t, *J* = 2.4 Hz, 1H), 7.13 (d, *J* = 5.2 Hz, 1H).

Compound 15: White powder, yield: 77.0 %, m.p. 130-132 ºC; IR (KBr, ν_{max}, cm^{-1}): 3096, 3039, 1652, 1591, 1489, 1448, 1088, 836, 760, 749, 699, 691, 681; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 8.08 (d, *J* = 8.4 Hz, 1H), 7.52 (d, *J* = 8.8 Hz, 1H), 7.39 (d, *J* = 8.8 Hz, 2H), 7.31 (d, *J* = 7.2 Hz, 1H), 7.30 (d, *J* = 8.0 Hz, 2H), 7.21 (d, *J* = 7.6 Hz, 3H), 7.19 (d, *J* = 8.4 Hz, 2H), 7.14 (d, *J* = 8.0 Hz, 2H).

Compound 16: Yellow powder, yield: 32.2 %, m.p. 150-152 ºC; IR (KBr, ν_{max}, cm^{-1}): 3068, 3040, 1667, 1603, 1593, 1521, 1490, 1450, 1337, 839, 771, 759, 714, 700; ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 8.07 (d, *J* = 8.8 Hz, 2H), 7.60 (d, *J* = 8.4 Hz, 2H), 7.31 (d, *J* = 6.8 Hz, 6H), 7.22-7.26 (m, 4H).

Assay of antifungal activity: All the synthesized compounds were screened for their activity against *Sclerotinia sclerotiorum* and *Botrytis cinerea* using the plate growth rate method[18].

Each tested compound and carbendazim (purity 90 %) were dissolved in dimethyl sulfoxide (DMSO), respectively. They were added to the sterile culture medium (potato-dextrose agar) at 45 ºC, mixed to homogeneity and transferred to sterile Petri dishes to solidify. For primary screening, each compound was used at a concentration of 100 mg L^{-1}. A mycelium agar disc (5 mm in diameter) of the target fungi was placed in the center of potato-dextrose agar plates. They were incubated at 28 ºC in the dark until the target fungi used as the controls covered the surface of these plates. Control groups were treated with the corresponding solutions without the tested compound or carbendazim. The diameter of the fungi in the cultures was measured and the inhibition of growth was calculated according to the formula of Abbott. Each compound was replicated three times.

TABLE-1
ANTIFUNGAL ACTIVITY AGAINST *Sclerotinia sclerotiorum* AND *Botrytis cinerea* OF COMPOUNDS **1-16** AT 100 mg L^{-1}

Compound	R_1	R_2	R_3	Inhibition of growth (%)*	
				Sclerotinia sclerotiorum	*Botrytis cinerea*
1	H	NH_2	NH	11.8	24.4
2	2-Cl	NH_2	NH	92.6	90.1
3	4-Cl	NH_2	NH	0.1	10.1
4	4-NO_2	NH_2	NH	16.3	8.0
5	H	NH_2 CH_2	$NHCH_2$	24.1	47.7
6	2-Cl	NH_2 CH_2	$NHCH_2$	14.0	16.0
7	4-Cl	NH_2 CH_2	$NHCH_2$	13.1	9.8
8	4-NO_2	NH_2 CH_2	$NHCH_2$	13.6	25.5
9	H	$NH_2CH_2CH_2$	$NHCH_2CH_2$	45.7	36.1
10	2-Cl	$NH_2CH_2CH_2$	$NHCH_2CH_2$	40.7	33.9
11	4-Cl	$NH_2CH_2CH_2$	$NHCH_2CH_2$	23.8	35.6
12	4-NO_2	$NH_2CH_2CH_2$	$NHCH_2CH_2$	9.7	13.0
13	H	NHPh	NPh	2.3	19.0
14	2-Cl	NHPh	NPh	9.7	20.9
15	4-Cl	NHPh	NPh	14.0	20.4
16	4-NO_2	NHPh	NPh	8.9	16.2
Carbendazim				99.8	91.3

*Based on the mean of triplicates.

RESULTS AND DISCUSSION

All the synthesized analogues of chalcone were screened for their activity against *Sclerotinia sclerotiorum* and *Botrytis cinerea*. The results are presented in Table-1. Some of them were active against the two different pathogenic fungi at a concentration of 100 mg L^{-1}. Among them, the inhibition rate of compound **2** reached 92.6 and 90.1 %, respectively, which was close to carbendazim.

A series of analogues of chalcone have been successfully synthesized and tested for their antifungal activity against *Sclerotinia sclerotiorum* and *Botrytis cinerea*. The preliminary results suggested that the design and synthesis of these compounds may be conducive to the antifungal activity of analogues of chalcone. They are also promising and beneficial to further studies in developing new and more effective fungicides in the agricultural chemistry field. However, there is more work to be done. A number of analogues of chalcone should be further synthesized for screening and surveying quantitative structure-activity relationships so as to find novel fungicides with high effect and low toxicity. The structural modification and optimization of compound **2** need to be done. Meanwhile, its mode of action and its safety to humans and non-target organisms also need to be investigated.

ACKNOWLEDGEMENTS

This project was supported by the Scientific Research Fund of Sichuan Provincial Education Department (No. 09ZA113), the Key Scientific Research Fund of Xihua University (No. Z0913302) and the Organic Chemistry Key Discipline of Xihua University.

REFERENCES

1. M.J. Gilbert, C.R. Thornton, E. Gavin, G.E. Wakley and N.J. Talbot, *Nature*, **440**, 535 (2006).
2. L.P. Partida-Martinez and C. Hertweck, *Nature*, **437**, 884 (2005).
3. P.E. Russell, *J. Agric. Sci*., **143**, 11 (2005).
4. M.T. McGrath, *Plant Dis*., **85**, 236 (2001).
5. J.Y. Cho, H.Y. Kim, G.J. Choi, K.S. Jang, H.K. Lim, C.H. Lim, K.Y. Cho and J.C. Kim, *Pest Manage. Sci*., **62**, 414 (2006).
6. J.Y. Cho, G.J. Choi, S.W. Son, K.S. Jang, H.K. Lim, S.O. Lee, N.D. Sung, K.Y. Cho and J.C. Kim, *Pest Manage. Sci*., **63**, 935 (2007).
7. M.A. Abbassy, S.A.M. Abdelgaleil, A.S.H. Belal and M.A.A.A. Rasoul, *Indian Crop Prod*., **26**, 345 (2007).
8. G.D. Crouse, *Chemtech*, **28**, 36 (1998).
9. T.G. Liao, Q.A. Wang, W.Q. Fang and H.J. Zhu, *Chin. J. Org. Chem*., **26**, 685 (2006).
10. L.P. Guan, X.M. Yin and H.M. Quan, *Chin. J. Org. Chem*., **24**, 1247 (2004).
11. R. Laliberte, *Can. J. Pharm. Sci*., **2**, 37 (1967).
12. R. De Vincenzo, C. Ferlini and M. Distefano, *Cancer Chemother. Phamacol*., **46**, 305 (2000).
13. J.C. Le Bail, C. Pouget and C. Fagnere, *Life Sci*., **68**, 751 (2001).
14. S. Shibata, *Stem Cells*, **12**, 44 (1994).
15. A. Lopez, D.S. Ming and G.H.N. Towers, *J. Nat. Prod*., **65**, 62 (2002).
16. M.M. Salem and K.A. Werbovetz, *J. Nat. Prod*., **68**, 108 (2005).
17. S. Marakami, M. Muramatsu and H. Aihara, *Biochem. Pharmacol*., **42**, 1447 (1991).
18. W. Huang and G.F. Yang, *Bioorg. Med. Chem*., **14**, 8280 (2006).