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Synthesis of 2-Phenylchromen-4-one derivatives by conventional and microwave: Assisted techniques, and their antimicrobial evaluation

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Abstract
Two derivatives of 2-phenylchromen-4-one (flavone ring) with their precursors 1-(2-hydroxyphenyl)-3-(phenyl)-prop-2-en-1-one (chalcone ring) have been synthesized by conventional and microwave-assisted heating techniques. A comparative study was carried out between the two methods. The structures of the synthesized compounds were determined by IR, ^{1}H-NMR and ^{13}C-NMR spectral data. The antibacterial and antifungal screens were performed against six human pathogenic bacteria viz. *Bacillus cereus* (G+), *Staphylococcus aureus* (G+), *Escherichia coli* (G-), *Vibrio choloriae* (G-), *Pseudomonas aeruginosa* (G-), and *Salmonella typhi* (G-) by the filter paper disc diffusion method and four plant as well as mold fungi. viz. *Aspergillus flavus, Aspergillus ochraceus, Aspergillus niger and Rhizopus spp.* by the poisoned food technique, respectively. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of these synthesized compounds in comparison to ampicillin were also determined by broth micro-dilution method. Compounds 2-(2,4-dimethoxyphenyl)-chromen-4-one (**6)** and 2-(4-ethoxyphenyl)-chromen-4-one (**7)** were found to possess significant antibacterial and antifungal activities.

Keywords: Antimicrobial activity, Chalcone, Flavone, MBC, MIC, Microwave irradiation

1. Introduction
2-Phenylchromen-4-one derivatives are large group of natural products having wide range of biological as well as antimicrobial activities [1]. Survey of the literature informed that flavonoids containing various groups are widely occured in medicinal plants [2]. These flavonoid products have demonstrated many pharmacological activities e.g. biocidal [3], pharmaceutical [4], anti-inflammatory [5], antioxidant [6], antimicrobial [7] and inhibitory activities [8]. Various synthetic methods have been reported for synthesizing flavones e.g. oxidative cyclization [9, 10], cyclodehydration [11], and microwave irradiation [12, 13] from 2'-hydroxychalcones *via* intermolecular Wittig reaction [14]. Among these methods, microwave irradiation has gained popularity for its advantages, such as shorter reaction time, higher yields and better selectivity.
Encouraged by the exceptional biological importance of flavonoids and in continuation of search for antimicrobial active molecules [15-19], we synthesized 1-(2-hydroxyphenyl)-3-(phenyl)-prop-2-en-1-one derivatives, 4-5 and 2-phenylchromen-4-one derivatives, **6-7** by conventional and microwave irradiation technique under green protocol in order to evaluate their antimicrobial activities and a comparative study to assess reaction time, amount of solvent and percentage of yield.

2. Experimental
2.1. Materials and Methods
The IR spectra were recorded by FT-IR spectrophotometer (Model-8900, Shimadzu, Japan) by KBr matrix. ^{1}H-NMR (400 MHz) and ^{13}C-NMR (100 MHz) spectra were recorded on JEOL GS×400, GEOL JNM-AL 400 (400 MHz) and JEOL GS×400, GEOL JNMAL 400 (100 MHz) spectrometer in $CDCl_3$ as a solvent. Chemical shifts are expressed in δ units (ppm) with reference to TMS as an internal standard and *J* values are given in Hz. Melting points were determined on electro thermal melting point apparatus and were uncorrected. TLC was carried out on Kieselgel GF_{254} and checked by iodine vapour or UV light. LG microwave oven (MB-3947C) with a maximum power output of 800 W was used to carry out all reactions.

2.2. Synthesis of 1-(2-hydroxyphenyl)-3-(phenyl)-prop-2-en-1-one derivatives (4-5)

2.2.1. Conventional method

An equivalent mixture of 2-hydroxyacetophenone (1 mmol) and aromatic aldehydes (1 mmol) dissolved in ethanolic solution of KOH (10%, 15 mL) was placed in a conical flask and kept at room temperature for approximately 75 h [18]. Ice-cold water was mixed with the reaction mixture for dilution and acidified with dil. HCl. The solid was separated by filtration, washed with water and extracted with ether. After washing with water, the ether layer was dried by anhydrous Na_2SO_4 and purified by column chromatography over silica gel using ether-acetone (8:1). The product was recrystallized from absolute ethanol.

2.2.2. Microwave method

An equivalent mixture of 2-hydroxyacetophenone and aromatic aldehydes dissolved in minimum amount of ethanolic KOH (10%, 3 mL) was placed in an Erlenmeyer flask [15]. The flask was covered with a glass and then it was taken in a domestic microwave oven. The reaction mixture was irradiated under 320-watt microwave irradiation for 2-3 min. The progress of the reaction was monitored by TLC (*n*-hexane: ethyl acetate, 6:1). The products were purified by the same process as mentioned in conventional method.

2.2.3. Spectral data

(E)-1-(2-Hydroxyphenyl)-3-(2,4-dimethoxyphenyl)-prop-2-en-1-one (4): Orange solid crystals (yield: conventional method, 75%, microwave irradiation 98%), m.p. 84-85°C; IR (KBr) υ_{max}(cm^{-1}): 3471(-OH), 3055, 2951, 1635 (>C=O), 1558 (C=C), 1504 (C=C, Ar), 1307 (C-O), 1026, 817, 748, ^{1}H-NMR (400 MHz, $CDCl_3$) δ_H (ppm): 3.90 (s, 3H, C_2-OCH_3), 3.96 (s, 3H, C_4-OCH_3), 6.52 (s, 1H, C_3-H), 6.58 (d, 1H, *J*=8 Hz, C_5-H), 6.93 (t, 1H, *J*=8 Hz, $C_{5'}$-H), 7.03 (d, 1H, *J*=8 Hz, $C_{3'}$-H), 7.50 (dd, 1H, *J*=4 Hz and *J*=8 Hz, $C_{4'}$-H), 7.61 (d, 1H, *J*=8 Hz, C_6-H), 7.73 (d, 1H, *J*=15.2 Hz, C_α-H), 7.94 (d, 1H, *J*=8 Hz, $C_{6'}$-H), 8.19 (d, 1H, *J*=15.2 Hz, C_β-H), 12.95 (s, 1H, $C_{2'}$-OH); ^{13}C-NMR (100 MHz, $CDCl_3$) δ_C (ppm): 56.3 (C_4-OCH_3), 56.5 (C_2-OCH_3), 102.1 (C-3), 107.2 (C-5), 113.1 (C-1), 117.3 (C-3'), 121.9 (C-5'), 123.6 (C-α), 124.2 (C-1'), 128.3 (C-6), 130.6 (C-6'), 137.3 (C-4'), 142.6 (C-β), 158.3 (C-2'), 160.8 (C-2), 162.4 (C-4), 186.5 (>C=O).

(E)-1-(2-Hydroxyphenyl)-3-(4-ethoxyphenyl)-prop-2-en-1-one (5): Yellow crystals (yield: conventional method 72%, microwave irradiation 91%), m.p.111-113° C; IR (KBr) υ_{max} (cm^{-1}): 3464 (-OH), 2981, 1635 (>C=O), 1604, 1573 (C=C), 1512, 1423 (C=C, Ar), 1269 (C-O), 1111, 1029, 821; ^{1}H-NMR (400 MHz, $CDCl_3$) δ_H (ppm): 1.47 (t, 3H, *J*=7.2 Hz, C_4-OCH_2CH_3), 4.11 (q, 2H, *J*=7.2 Hz, C_4-OCH_2CH_3), 6.95 (d, 1H, *J*=8 Hz, $C_{3'}$-H), 6.96 (d, 2H, *J*=8 Hz, C_3-H and C_5-H), 7.05 (d, 1H, *J*=8 Hz, $C_{6'}$-H), 7.51 (m, 1H, $C_{5'}$-H), 7.57 (d, 1H, *J*=15.6 Hz, C_α-H), 7.65 (d, 2H, *J*=8 Hz, C_2-H and C_6-H), 7.92 (d, 1H, *J*=15.6 Hz, C_β-H), 7.95 (dd, 1H, *J*=8 Hz and *J*=4 Hz, $C_{4'}$-H), 12.96 (s, 1H, $C_{2'}$-OH); ^{13}C-NMR (100 MHz, $CDCl_3$) δ_C (ppm): 16.2 (C_4-OCH_2CH_3), 56.3 (C_4-OCH_2CH_3), 115.1 (C-3 & C-5), 116.6 (C-3'), 121.3 (C-5'), 124.1 (C-α), 124.6 (C-1'), 126.1 (C-1) 127.2 (C-2 & C-6), 132.2 (C-6'), 137.3 (C-4'), 143.4 (C-β), 157.2 (C-4), 159.1 (C-2'), 186.3 (>C=O).

2.3. Synthesis of 2-phenyl-chromen-4-one derivatives (6-7)

2.3.1. Conventional method

1-(2-Hydroxyphenyl)-3-(phenyl)-prop-2-en-1-one derivatives (1 mmol) were suspended in DMSO (20 mL) and a crystal of iodine (0.02 mmol) was added [9]. The reaction mixture was refluxed for 30-40 minutes in an oil bath. The progress of the reaction was monitored by TLC (*n*-hexane: ethyl acetate, 4:1). The reaction mixture was diluted with excess water, and extracted with diethyl ether (2 × 15 mL). The ether layer was washed with aqueous 20% sodium thiosulphate and water, and dried over anhydrous Na_2SO_4. This crude solid was purified by column chromatography using *n*-hexane: ethyl acetate (4:1) as an eluent. The product was obtained as solid crystals. The solid products were recrystallized using ethyl alcohol and it gave blue fluorescence in UV light.

2.3.2. Microwave method

1-(2-Hydroxyphenyl)-3-(phenyl)-prop-2-en-1-one derivatives (1 mmol) were suspended in DMSO (2 mL) and iodine (0.02 mmol) was added. The mixture was irradiated under 320-watt microwave irradiation for 4-5 minutes. The progress of the reaction was monitored by TLC (*n*-hexane: ethyl acetate, 4:1). The product was obtained by the same process as in conventional method.

2.3.3. Spectral data

2-(2,4-Dimethoxyphenyl)-chromen-4-one (6): Colorless needles (yield: conventional method 67%, microwave irradiation 95%), m.p.155-156°C; IR (KBr) υ_{max} (cm^{-1}): 3143, 2947, 2927, 1639 (>C=O), 1608, 1558 (C=C), 1465 (C=C, Ph), 1377, 1257 (C-O), 1130, 1014, 829, 756; ^{1}H-NMR (400 MHz, $CDCl_3$) δ_H(ppm): 3.92 (s, 3H, -OCH_3), 3.96 (s, 3H, -OCH_3), 6.59 (s, 1H, C_3-H), 6.67 (dd, 1H, *J*=8.8 Hz and 2.0 Hz, $C_{5'}$-H), 7.29 (d, 1H, *J*=5.2 Hz, $C_{3'}$-H), 7.44 (t, 1H, *J*=12.4 Hz, C_8-H), 7.55 (d, 1H, *J*=8.4 Hz, $C_{6'}$-H), 7.70 (dt, 1H, J=8.4 Hz & 1.6 Hz, C_6-H), 7.94 (d, 1H, *J*=8.8 Hz, C_7-H), 8.25 (dd, 1H, J= 8 Hz & 1.6 Hz, C_5-H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ_C (ppm): 55.56 (C4' -OCH_3), 55.69 (C2' -OCH_3), 98.96 (C-3), 105.34 (C-3'), 111.32 (C-5'), 113.64 (C-1'), 117.89 (C-8), 123.79 (C-6), 124.76 (C-4a), 125.59 (C-6'), 130.45 (C-4), 133.35 (C-7), 156.41 (C-8a), 159.71 (C-2'), 160.94 (C-4'), 163.30 (C-2), 178.95 (>C=O).

2-(4-Ethoxyphenyl)-chromen-4-one (7): Pale yellow crystals (yield: conventional method 66%, microwave irradiation 93%), m.p.124-125°C; IR (KBr) υ_{max} (cm^{-1}): 2981.95, 2927, 1651 (>C=O), 1604, 1512 (C=C), 1465 (C=C, Ph), 1377, 1265 (C-O), 1184, 1037, 817, 771; ^{1}H-NMR (400 MHz, $CDCl_3$) δ_H (ppm): 1.49 (t, 3H, *J*=6.8 Hz, C_4-$OCH_2C\underline{H}_3$), 4.15 (q, 2H, *J*= 6.8 Hz, C_4'-$OC\underline{H}_2CH_3$), 6.78 (s, 1H, C_3-H), 7.03 (d, 2H, *J*=8.8 Hz, $C_{3'}$-H and C_5'-H), 7.44 (t, 1H, *J*=7.2 Hz, C_8-H), 7.57 (d, 1H, *J*=8 Hz, C_6-H), 7.71 (m, 1H, C_7-H), 7.91 (d, 2H, *J*=8 Hz, $C_{2'}$-H and $C_{6'}$-H), 8.25 (dd, 1H, *J*=6.4 Hz & 1.6 Hz, C_5-H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ_C (ppm): 15.6 (C4'-OCH_2CH_3), 66.7 (C4' -OCH_2CH_3), 97.3 (C-3), 115.8 (C-3' & C-5'), 118.5 (C-8) 124.3 (C-6), 126.1 (C-4a), 127.2 (C-1'), 127.5 (C-2' & C6'), 131.7 (C-5), 136.4 (C-7), 156.9 (C-8a), 158.2 (C-4'), 168.3 (C-2), 178.5 (>C=O).

2.4. Antimicrobial activities

All the synthesized compounds 4-7 were evaluated for their *in vitro* antibacterial and antifungal activity. The antibacterial activity was studied by the filter paper disc diffusion method [20,21] against six human pathogenic bacteria viz. *Bacillus cereus* (G+), *Staphylococcus aureus* (G+), *Escherichia coli* (G-), *Vibrio choloriae* (G-), *Pseudomonas aeruginosa* (G-) and *Salmonella typhi* (G-). The antifungal activity was also assessed by poisoned-food technique [22] towards four plant mold fungi, viz. *Aspergillus flavus, Aspergillus ochraceus, Aspergillus niger and Rhizopus spp.*. All the compounds were dissolved in DMSO to get a solution and discs with only

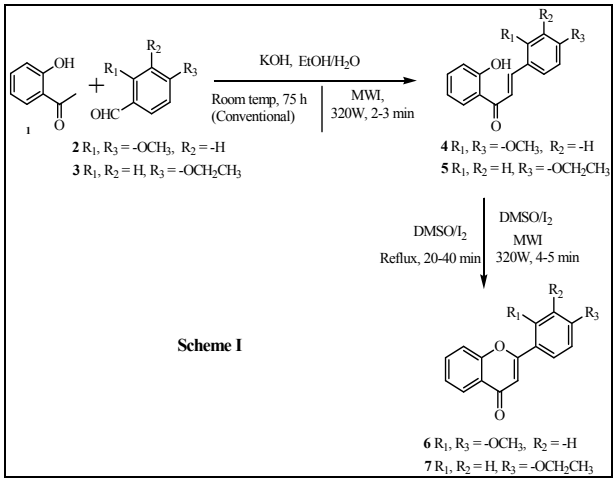
DMSO were used as a control. Commercial Ampicillin and Nystatin were used as a standard antibiotic and fungicide, respectively under similar conditions. Nutrient agar (NA) and potato dextrose agar (PDA) were used as a basal medium for bacteria and fungi, respectively. Inhibitory activity was measured in millimeters (mm) from the observed inhibition zones. The MIC and MBC values in comparison to Ampicillin were also determined against those selected bacteria by broth micro-dilution method [23]. The media used in this respect were nutrient broth. Dilution series were set up with 2, 4, 8, 16, 32, 64, 128, 256, 512 and 1024 µg/mL of nutrient broth medium. To each well, 100 µL of standardized suspension of the testing bacteria (10^7cell/mL) were added and incubated at 30 °C for 24 hr.

3. Results and discussion

3.1. Chemistry

The main aim of the present work involves the synthesis and the antimicrobial activity of two derivatives of 2-(phenyl)-chromen-4-one viz., 2-(2,4-dimethoxyphenyl)-chromen-4-one (6) and 2-(4-ethoxyphenyl)-chromen-4-one (7) as well as their precursors (E)-1-(2-Hydroxyphenyl)-3-(2,4-dimethoxyphenyl)-prop-2-en-1-one (4) and (E)-1-(2-Hydroxyphenyl)-3-(4-ethoxyphenyl)-prop-2-en-1-one (5). The synthesis of the derivatives has been accomplished in conventional method and under microwave irradiation, as shown in Scheme 1. A comparison has been shown in Table 1 among reaction times, solvent and percentage of yields. The formation of the compounds 4-7 has been confirmed by IR, ^{1}H-NMR and ^{13}C-NMR spectral data.

The IR absorption band at 3364 cm^{-1} indicated the presence of hydroxyl group in the compound 4 and a positive ferric chloride test also supported a free hydroxyl group. Moreover, a characteristic singlet at δ_H 12.65 proved the presence of a chelated phenolic proton at $C_{2'}$-OH integrating for one proton. The presence of olefinic protons of an α, β-unsaturated ketone in compound 4 which was clearly observed at δ 7.73 (*J*=15.2 Hz) and δ 8.19 (*J*=15.2 Hz) corresponding to C_α-H and C_β-H, respectively. The higher coupling values show that the olefinic protons are in a *trans*-relationship. The absence of a hydroxyl group in IR spectra band confirmed the oxidation of chalcone 4 into chromenone (flavone) 6 and signal of -OH was also not observed in the ^{1}H-NMR spectrum. This spectrum of 6 also displayed a singlet at δ_H 6.94 for one proton corresponding to C_3-H and showed that the C_β-H of the corresponding chalcone 4 involved in cyclization of chalcone to form corresponding flavone. The entire ^{13}C- NMR spectral data were in accordance with the structure of compounds 4-7.



Scheme I

Table 1: Comparison between conventional and microwave technique.

Compound	Conventional Method			Microwave Irradiation		
	Time	Solvent (mL)	Yield %	Time	Solvent (mL)	Yield %
4	75 h	15	75	2 min	3	98
5	72 h	15	72	2.5 min	3	91
6	35 min	20	67	4.5 min	2	95
7	40 min	25	66	5.5 min	2	93

3.2. Antimicrobial activities

The result (Table 2) of antibacterial activity indicates that compounds 6 and 7 showed higher activity and compound 4 showed low antibacterial activity while compound 5 was unable to show any inhibition. The overall results of antifungal (Table 2) investigation showed that 2-(phenyl)-chromen-4-one derivatives 6 and 7 are somewhat higher activity than their corresponding 1-(2-hydroxyphenyl)-3-(phenyl)-prop-2-en-1-one derivatives 4 and 5 towards the selected organisms (fungi). After analyzing the MIC and MBC values (Table 3) of all the compounds 4-7, compounds 6 and 7 showed lower MIC values against both gram-positive and gram-negative bacteria strains. Meanwhile, compounds 6 and 7 showed lower MBC values and compounds 5 showed higher MBC values for all bacteria strains.

Table 2: Antibacterial and antifungal activities of the synthesized compounds 4-7.

Compound	Diameter of zone of inhibition in mm (100 µg (dw)/ disc)						% Inhibition of mycelial growth (100 µg(dw)/ml PDA)			
	B. Cereus	*S. Aureus*	*E. Coli*	*V. Choloriae*	*P. Aeruginosa*	*S. Typhi*	*A. flavus*	*A. niger*	*A. ochraceus*	*Rhizopus spp.*
4	08	09	09	*10	…	08	68	72	77	74
5	…	…	…	07	…	08	41	50	51	63
6	*10	*11	*12	*11	08	*10	41	72	63	66
7	*11	09	*11	*11	09	*13	41	47	54	53
AMP	12	13	12	12	11	14	53	61	66	68

'(…)' Means no inhibition, '*' Means good inhibition, dw.= dry weight.

Table 3: MIC and MBC level of the synthesized compounds 4-7.

Test organism	MIC (μg mL^{-1})					MBC (μg mL^{-1})				
	4	5	6	7	AMP	4	5	6	7	AMP
B. cereus	64	128	64	32	4	64	256	64	32	8
S. aureus	64	128	64	32	4	64	256	64	32	8
E. coli	32	128	32	16	4	64	256	32	16	8
V. cholerae	128	128	64	32	4	128	256	64	32	8
P. aeruginosa	64	256	32	16	8	128	>256	32	16	8
S. typhi	64	128	32	16	4	64	256	64	32	8

4. Conclusion

In this work, we have demonstrated the synthesis of 2-(phenyl)-chromen-4-one derivatives using conventional method and microwave irradiation technique. The main advantages of microwave method are relatively shorter time, higher yields, a simple experiment and isolation procedure. Moreover, this method reduces sharply the volume of DMSO required. From the result of antibacterial and antifungal activities, it can be concluded that the 2-(phenyl)-chromen-4-one ring system and presence of alkoxy group are mainly responsible for the antimicrobial effects. The antimicrobial activity data obtained during this study will be certainly useful for further research for drug designing and synthesizing new flavone derivatives.

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6. References

1. Weidenbörner M, Jha HCC. Int. Symp. on Natural phenols in plant resistance, Int. Soc. Horticultural Sci. Acta Horticulture 2003; 381.
2. Tanaka T, Iinuma M, Yuki K, Fujii Y, Mizuno. Flavonoids in root bark of Pongamia pinnata. Phytochemistry 1992; 31(3):993-998.
3. Rao KV, Chattopadhyay SK, Reddy GC. Flavonoids with mosquito larval toxicity. J. Agric. Food. Chem. 1990; 38:1427-1430.
4. Wolfman C, Viola H, Marder M, Wasowski C, Ardenghi P, Izquierdo I, *et al.,* Anxio-selective properties of 6,3′-dinitroflavone, a high-affinity benzodiazepine receptor ligand. Eur. J. Pharmacol. 1996; 318:23-30.
5. Dao TT, Chi YS, Kim J, Kim HP, Kim S, Park H. Synthesis and inhibitory activity against COX-2 catalyzed prostaglandin production of chrysin derivatives. Bioorg. Med. Chem. Lett. 2004; 14:1165-1167.
6. Chan ECH, Patchareewan P, Owen LW. Relaxation of flavones and flavonoids in rat isolated thoracic aorta: mechanism of action and structure-activity relation-ships. J. Cardiovasc. Pharmacol. 2000; 35:326-333.
7. Mazimba O. Antimicrobial activities of heterocycles derived from thienylchalcones. J. King Saud Univ. Sci. 2015; 27:42–48.
8. Bors W, Heller W, Michel C, Saran M. In Methods in enzymology (eds) L. Packer, A. N. Glazer, Academic Press, vol. 18, New York, 1990, 6343-355.
9. Doshi AG, Soni PA, Ghiya BJ. Oxidation of 2'-hydroxychalocones. Indian J. Chem. 1986; 258:759-759.
10. Hoshino Y, Ohinata T, Taken N. The direct preparation of flavones from 2'- hydroxychalcones using disulphides. Bull. Chem. Soc. Jap. 1986; 59:2351-2352.
11. Kabalka WG, Mereddy A. Microwave-assisted synthesis of functionalized flavones and chromones. Tetrahedron Lett. 2005; 46(37):6315-6317.
12. Varma RS. Solvent-free organic syntheses using supported reagents and microwave irradiation. Green Chem. 1999; 1(1):43-55.
13. Hayes BL. Microwave synthesis: Chemistry at the speed of light. Matthews, NC, CEM Publishing, 2002.
14. Floc'h YL, Lefeuvre M. Synthese de dihydroxy phenacylidene. Tetrahedron Lett. 1986; 27(24):2751-2752.
15. Bhuiyan MMH, Hossain MI, Mahmud MM, Al-Amin M. Microwave-assisted efficient synthesis of chalcones as probes for antimicrobial activities. Chem Journal 2011; 1(1):21-28.
16. Bhuiyan MMH, Hamidunnessa, Mahmud MM, Multicomponent reactions: Microwave-assisted efficient synthesis of dihydropyrimidinones (thiones) and quinazolinones under green chemistry protocol as probes for antimicrobial activities, J. Sci. Res. 2012; 4(1):143-153.
17. Bhuiyan MMH, Rahman KMM, Alam MA, Mahmud MM. Microwave assisted Knoevenagel condensation: Synthesis and antimicrobial activities of Some -cyanoacrylates. Pak. J. Sci. Ind. Res. 2013; 56(3):131-137.
18. Rahim A, Ali R, Islam A. Studies of antimicrobial activity of 2′,4′,5′- and 2′,3′,4′-trimethoxy flavanones. J. Sci. Res. 2016; 8(2):209-216.
19. Ali R, Rahim A, Islam A. Synthesis and antimicrobial activity of 7-hydroxy-3□□□□-methylenedioxy- and 7-benzyloxy-3□□□□-methylenedioxy flavanones. J. Sci. Res. 2017; 9(3):297-306.
20. Arima H, Ashida H, Danno GI. Rutin-enhanched antibacterial activities of flavonoids against *Bacillus cereus* and *Salmonella entertidis*. Biosci. Biotechnol. Biochem. 2002; 66(5):1009-1014.
21. Jeongmok K, Maurice RM, Cheng IWJ. Antibacterial activity of some essential oil components against five foodborne pathogens. J. Agric. Food Chem. 1995; 43:2839-2845.
22. Grover RK, Moore JD, Toximetric studies of fungicides against the brown rot organisms, sclerotinia fructicola and S. laxa. Phytopathology, 1962; 52:876-880.
23. Amsterdam D. In: V. Loman, Antibiotics in laboratory medicine, 1996; 4:52-111.