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Research Article

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A study on the synthesis and biocidal efficacy of certain random copolyesters containing chalcone moiety in the main chain

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ABSTRACT

A series of four random copolyesters were synthesized by the polycondensation of a chalcone diol with two diacid chlorides namely terepththaloyl chloride and glutaryl chloride. The four chalcone diols used are (2E)-1,3-bis-(4-hydroxyphenyl)-prop-2-en-1-one, (2E)-1-(4-hydroxyphenyl)-3-(4-hydroxy-3-methoxyphenyl)-prop-2-en-1-one, (2E)-1-(4-hydroxy-3-methoxy phenyl)-3-(4-hydroxy phenyl)-prop-2-en-1-one and (2E)-1,3-bis-(4-hydroxy-3-methoxy phenyl)-prop-2-en-1-one were synthesized by acid catalyzed Claisen-Schmidt reaction. The random copolyesters were synthesized by solution polycondensation technique and they were characterized by qualitative solubility tests and viscosity measurements. The microstructure of the repeating units available in the copolyester backbone was established by FT-IR, ^{1}H-NMR and ^{13}C-NMR spectroscopic techniques. The inhibitory activity of the random copolyesters against certain microbes is well documented. Hence these random copolyesters may emerge as antimicrobial agents.

Key words: Chalcone, Random Copolyesters, Polycondensation, Bactericidal, Fungicidal

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INTRODUCTION

Chalcones are 1,3-diphenyl-2-propene-1-one derivatives in which two aromatic rings are linked by a three carbon α,β-unsaturated carbonyl system. They possess conjugated double bonds and a completely delocalized π-electron system on both benzene rings. Chalcone moieties are available in natural as well as synthetic compounds and are found to display a variety of pharmacological activity such as antibacterial [1], anti-tumour [2], anticancer [3], anti-tubercular, anti-inflammatory [4], antioxidant [5], anti-malarial [6], anti-ulcerative [7], etc. The presence of reactive methoxy and hydroxyl group in the chalcone backbone is responsible for their biological activity [8].

The objective of the present investigation is to generate four copolyesters by incorporating the chalcone moieties in the main chain by polycondensation, then characterizing them and finally studying their anti-microbial activities. Copolyesters [9] are a class of macromolecules which contain ester linkages and are synthesized by the copolymerization of diol, diacid chloride-I and diacid chloride-II in the ratio of 2:1:1.

EXPERIMENTAL SECTION

Aldrich samples of 4-hydroxybenzaldehyde, vanillin, 4-hydroxyacetophenone and 4-hydroxy-3-methoxyacetophenone were used as received. Ethanol (Merck) was used as a non-solvent for the precipitation of copolyesters and as a solvent for the preparation of the monomer diols. Aldrich samples of Terephthaloyl chloride and Glutaryl chloride were purchased and used for the copolymerization process. SD Fine AR sample of Dimethyl

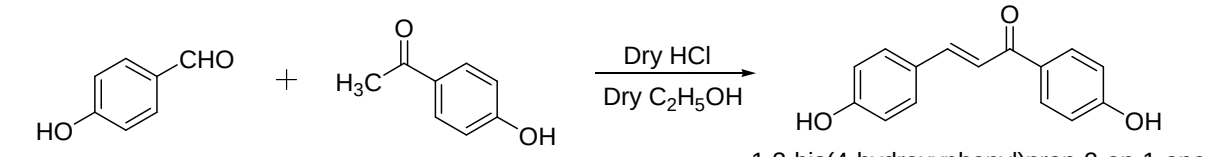
acetamide (DMAc) was used as such as solvent for finding out the inherent viscosity of the copolyester in solution. Spectral grade DMSO-d6 (Aldrich) containing as internal standard was used for recording NMR Spectra.

Synthesis of Chalcone Diols

The monomers *(2E)*-1,3-bis-(4-hydroxyphenyl)-prop-2-en-1-one **(**BHPP), *(2E)*-1-(4-hydroxyphenyl)-3-(4-hydroxy-3-methoxyphenyl)-prop-2-en-1-one (HMPP), *(2E)*-1,3-bis-(4-hydroxy-3-methoxyphenyl)-prop-2-en-1-one **(**BHMP) and *(2E)*-1-(4-hydroxy-3-methoxyphenyl)-3-(4-hydroxyphenyl)-prop-2-en-1-one (HMHP) were synthesized by the process reported by Chitra and coworkers [10].

Preparation of BHPP

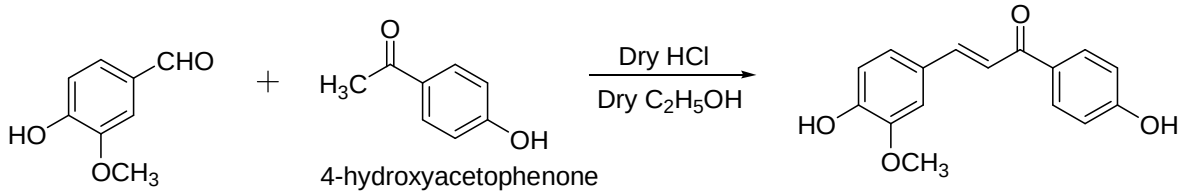
Dry HCl gas was passed through a well-cooled and stirred solution of 4-hydroxybenzaldehyde (50mmol) and 4-hydroxyacetophenone (50mmol) in 125mL of dry ethanol taken in a 250mL round-bottomed flask. The yellow coloured crystals of BHPP which got separated was washed with double distilled water and re-crystallized from hot methanol. Yield: 90% m.p.:197.2°C; IR(KBr) 3301 (b, O–H), 1648(s, C=O) cm^{-1}; ^{1}H NMR(DMSO-d6) δ9.9(s, 2H, –OH), δ7.4–8.4(m, 8H, aromatic), δ6.7–6.9(dd, 2H, –CH=CH–) and MS (EI) m/z 240 [M]+.



4-hydroxybenzaldehyde 4-hydroxyacetophenone 1,3-bis(4-hydroxyphenyl)prop-2-en-1-one

Preparation of HMPP

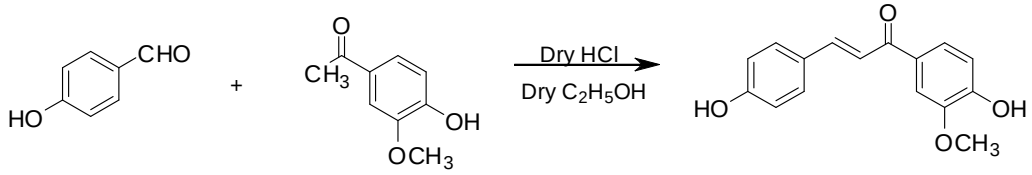
Dry HCl gas was passed through a well-cooled and stirred solution of vanillin (50mmol) and 4-hydroxyacetophenone (50mmol) in 125mL of dry ethanol taken in a 250mL round-bottomed flask. The yellow coloured crystals of HMPP which got separated was washed with double distilled water and re-crystallized from hot methanol. Yield: 85% m.p.:181°C; IR(KBr) 3329(b, O–H), 1663(s, C=O) cm^{-1}; ^{1}H NMR (DMSO-d6) δ9.8(s, 2H, –OH), δ7.2–8.3(m, 7H, aromatic), δ6.7–6.9(dd, 2H, –CH=CH–)δ3.4(s, 3H, –OCH_3) and; MS (EI) m/z 270 [M]+.



vanillin 4-hydroxyacetophenone 3-(4-hydroxy-3-methoxyphenyl)-1-(4-hydroxyphenyl)prop-2-en-1-one

Preparation of HMHP

Dry HCl gas was passed through a well-cooled and stirred solution of 4-hydroxybenzaldehyde (50mmol) and 4-hydroxy-3-methoxyacetophenone (50mmol) in 125mL of dry ethanol taken in a 250mL round-bottomed flask. The yellow coloured crystals of HMPP which got separated was washed with double distilled water and re-crystallized from hot methanol. Yield: 86% m.p.:178°C; IR(KBr) 3320 (b, O–H), 1655(s, C=O) cm^{-1}; ^{1}H NMR(DMSO-d6) δ 9.9(s, 2H, –OH), δ7.2–8.3(m, 7H, aromatic), δ6.7–6.9(dd, 2H, –CH=CH–)δ3.4 (s, 3H, –OCH_3) and; MS (EI) m/z 270 [M]+.

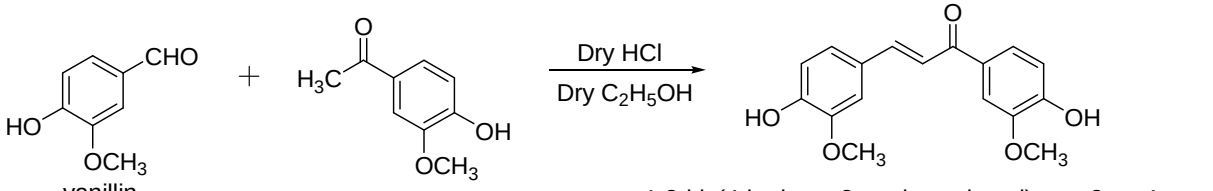


4-hydroxybenzaldehyde 4-hydroxy-3-methoxyyacetophenone

1-(4-hydroxy-3-methoxyphenyl)-3-(4-hydroxyphenyl)prop-2-en-1-one

Preparation of BHMP

Dry HCl gas was passed through a well-cooled and stirred solution of vanillin (50mmol) and 4-hydroxy-3-methoxyacetophenone (50mmol) in 125mL of dry ethanol taken in a 250mL round-bottomed flask. The yellow coloured crystals of HMPP which got separated was washed with double distilled water and re-crystallized from hot methanol. Yield: 83% m.p.:110°C; IR(KBr) 3322(b, O–H), 1658(s, C=O) cm^{-1}; ^1H NMR(DMSO-d6) δ9.9(s, 2H, –OH), δ7.2–8.3(m, 6H, aromatic), δ6.7–6.9(dd, 2H, –CH=CH–),δ3.4(s, 3H, $–OCH_3$), δ3.3(s, 3H, $–OCH_3$) and; MS (EI) m/z 300 [M]+.



Synthesis of Copolyesters

The procedure [11] involved in the synthesis of a typical aliphatic diacid-based random copolyester is represented here.

The monomer BHPP (1mmol.) was dissolved in 15ml of DMF in a 100mL round-bottomed flask. After 5 minutes 1mL of triethylamine was added and stirred. The mixture was allowed to stir at room temperature for 15 minutes in inert atmosphere. Then, the diacid chlorides terephthaloyl chloride (0.5mmol.) and glutaryl chloride (0.5mmol.) were added with constant stirring. Then the temperature was raised to 100°C and maintained at this temperature with continuous stirring for a span of 3 hours. At last the reaction mixture was poured into 100ml of methanol when the copolyester was precipitated. It was filtered, washed with dry methanol and dried in vacuum.

The copolyester PBTG, PCTG and PDTG were prepared by a similar method using the diols HMPP, HMHP and BHMP.

Table 1: Monomers used, copolyester code of the four copolyesters with their respective percentage yield and inherent viscosities

Diol	Diacid Chloride - I	Diacid Chloride - I	Copolyester Code	Yield (%)	η_{inh}, (dL/g)
BHPP	Terephthaloyl chloride	Glutaryl chloride	PATG	76	0.73
HMPP	Terephthaloyl chloride	Glutaryl chloride	PBTG	73	0.76
HMHP	Terephthaloyl chloride	Glutaryl chloride	PCTG	77	0.78
BHMP	Terephthaloyl chloride	Glutaryl chloride	PDTG	75	1.10

Antimicrobial Activity (Agar Disc Diffusion Method)

Preparation of Inoculums

Stock cultures were maintained at 4°C on slant of nutrient agar. Active cultures for experiments were prepared by transferring a loop full of cells from the stock cultures to test tubes of nutrient broth for bacteria that were incubated at 24 hours at 37°C. The Assay was performed by agar disc diffusion method.

Antibacterial Activity

The disc diffusion method [12] was employed to establish the antibacterial activity. Antibacterial activity of the copolyester sample was determined by disc diffusion method on Muller Hinton agar (MHA) medium. The Muller Hinton Agar was weighed as 3.8gms and dissolved in 100ml of distilled water after which 1gm of agar was added. Then the medium was kept for sterilization. After sterilization the media was poured into sterile Petri plates and were permitted to solidify. After the medium was solidified, the inoculums were spread on the solid plates with sterile swab moistened with the bacterial suspension. The discs were placed in MHA plate and 20µl of sample [concentration: 500µg, 250µg, 125µg, 62.5µg] were added. The plates were incubated for 24 hours, at 37°C. Then the microbial growth was determined by measuring the diameter of zone of inhibition.

Antifungal Activity Assay

Antifungal activity of copolyester sample was determined by antifungal susceptibility test. Prepare PDA Broth and inoculate the culture. Then it is kept in shaker for a day. The potato dextrose agar was weighed as 3.9gms and

dissolved in 100ml of distilled water to which 1gm of agar was added. Then the medium was kept for sterilization. After sterilization the media was poured in to sterile Petri plates and were allowed to solidify for twenty minutes. After solidification, the inoculums were spread on the solid plates with sterile swab moistened with the fungal suspension. The discs were placed in PDA plate and 20µl of sample [concentration: 500µg, 250µg, 125µg,62.5µg] were added. The plates were kept at room temperature. Then the microbial growth was determined by measuring the diameter of zone of inhibition.

RESULTS AND DISCUSSION

Solubility of all the polyesters was determined in various solvents qualitatively. The inherent viscosity (η_{inh}) of the polyesters was determined in DMAc solution at a concentration 0.1 gdL^{-1} using Ubbelohde viscometer in which the pure solvent had a flow rate of 104 seconds at 30°C. FT-IR spectra of the entire random copolyesters were recorded using Shimadzu FT-IR instrument. The ^{1}H and ^{13}C-NMR spectra were recorded with BRUKER AV III 500 MHz NMR instrument in DMSO-d6 solvent.

Solubility

The copolyesters reported here are found to be soluble in highly polar solvents such as DMAc and dimethyl formamide, partially soluble in moderately polar solvents like tetrahydrofuran and acetone but thoroughly insoluble in least polar solvents like benzene and hexane. Copolyesters with methoxy substituent in the benzene ring of the chalcone moiety had better solubility which may be to their competence to disrupt the macromolecular chain. Similar explanation was offered by Sidharthan and coworkers [13] in a series of copolyester.

Viscosity Measurements

The η_{inh} value of all the four copolyesters was determined in DMAc solution at 30°C using Ubbelohde viscometer. In each case 25mg of pure dry copolyester sample was dissolved in 25ml of DMAc, kept aside for some time with occasional shaking. The η_{inh} was calculated from the flow time measurements. The inherent viscosity values were found to be in the range of 0.73–1.10dL/g and are presented in table 1. The data shows that these copolyesters are reasonably of high molecular weight.

Spectral Studies

FT-IR spectrum of the four copolyesters was recorded using Shimadzu FT-IR instrument. The FT-IR spectrum of all the four copolyesters showed characteristic absorption in the range of 1742–1764cm^{-1} due to ester C=O stretching frequency. Similar observations were made by Samuel and coworkers [14] in a series of copolyesters.

The NMR spectra were recorded with BRUKER AV III 500 MHz NMR instrument in DMSO-d6 solvent to identify the structural units present in the copolyester chain. The aromatic protons are observed in the range of 7.5–8.2ppm. The vinyllic protons attached to the carbonyl carbon are observed in the range of 6.7–7.0ppm. The methoxy protons in the chalcone moiety are represented in the range of 3.3–3.5ppm. The methylene protons are observed in the range of 2.0–3.0ppm. Similar remarks were made Chitra and coworkers [15] in a series of copolyesters derived from bischalcones.

The signals in the range of 170–205ppm and 165–175ppm in the ^{13}C-NMR spectra of the copolyesters are owing to the carbonyl carbon of the α,β-unsaturated ketone and ester groups, respectively, which indicates the formation of copolyester.

Bactericidal Study

The antibacterial activity [16,17] of the four copolyesters PATG, PBTG, PCTG and PDTG were assayed against *Staphylococcus aureus*, *Enterococcus faecalis*, *Bacillus cereus*, and *Escherichia coli* by disc diffusion method [18,19].

Streptomycin inhibited the escalation of *Staphylococcus aureus* by 20mm, *Enterrococcus faecalis* by 19mm, *Bacillus cereus* by 26mm, and *Escherichia coli* by 17mm. From table 2 it is apparent that the four copolyesters were found to be bactericidal in nature. With increase of concentration of the copolyester material it was observed that the inhibition effect increased. Parallel observations were made by Kannappan and *et al*. [20] in a series of poly(ester-amides).

Table 2: Inhibition effects of the four copolyesters on the growth of *Staphylococcus aureus Enterococcus faecalis, Bacillus cereus* and *Escherichia coli*

Test Material	*Staphylococcus aureus* Zone of inhibition in mm				*Enterococcus faecalis* Zone of inhibition in mm			
	500 µg	250 µg	125 µg	62.5 µg	500 µg	250 µg	125 µg	62.5 µg
PATG	15	12	10	10	16	15	11	10
PBTG	18	16	16	16	15	13	11	11
PCTG	13	12	12	12	13	10	10	10
PDTG	12	12	11	11	10	9	8	7
Test Material	*Bacillus cereus* Zone of inhibition in mm				*Escherichia coli* Zone of inhibition in mm			
	500 µg	250 µg	125 µg	62.5 µg	500 µg	250 µg	125 µg	62.5 µg
PATG	19	18	15	12	12	12	12	12
PBTG	15	14	13	13	-	-	-	-
PCTG	15	14	14	16	11	11	11	11
PDTG	13	12	12	12	13	12	11	11

Fungicidal Study

The antifungal activity [21] of the synthesized copolyesters PATG, PBTG, PCTG and PDTG were assayed against *Candida albicans*, A*spergillus flavus*, *Penicillium*, *T. mentogrophyte* by disc diffusion method. Streptomycin subdued the growth of *Candida albicans* by 14mm, A*spergillus flavus* by 13mm, *Penicillium* by 10mm, and *T. mentogrophyte* by 10mm. From table 3 it is obvious that the four copolysters were found to be fungicidal in nature.

Table 3: Inhibition effects of the four copolyesters on the growth of *Candida albicans,* A*spergillus flavus, Penicillium* and *T. mentogrophyte*

Test Material	*Candida albicans* Zone of inhibition in mm				A*spergillus flavus* Zone of inhibition in mm			
	500 µg	250 µg	125 µg	62.5 µg	500 µg	250 µg	125 µg	62.5 µg
PATG	11	10	8	6	10	10	9	9
PBTG	12	10	10	10	9	9	-	-
PCTG	12	12	12	12	9	9	9	9
PDTG	10	10	10	10	10	10	9	9
Test Material	*Penicillium* Species Zone of inhibition in mm				*T. mentogrophyte* Zone of inhibition in mm			
	500 µg	250 µg	125 µg	62.5 µg	500 µg	250 µg	125 µg	62.5 µg
PATG	11	11	11	11	8	8	7	7
PBTG	9	9	9	-	7	7	-	-
PCTG	10	10	-	-	9	8	-	-
PDTG	10	10	10	10	-	-	-	-

CONCLUSION

The four copolyesters are synthesized using a diacid chloride-I (Terephthaloyl chloride), diacid chloride-II (Glutaryl chloride) and a chalcone diol. The chalcone diols are varied. The chalcone diols used are (2E)-1,3-bis-(4-hydroxyphenyl)-prop-2-en-1-one **(**BHPP), (2E)-1-(4-hydroxyphenyl)-3-(4-hydroxy-3-methoxyphenyl)-prop-2-en-1-one (HMPP), (2E)-1,3-bis-(4-hydroxy-3-methoxyphenyl)-prop-2-en-1-one **(**BHMP) and (2E)-1-(4-hydroxy-3-methoxyphenyl)-3-(4-hydroxyphenyl)-prop-2-en-1-one (HMHP). These copolyesters are highly soluble in polar organic solvents. These random copolyesters are characterized by solubility studies, viscosity measurements and spectral data. These copolyesters exhibited significant bactericidal activity against pathogenic bacteria. Some of them are also found to be fungicidal in nature.

REFERENCES

[1] SS Mokle; MA Sayeed; Kothawar; Chopde, *Int. J. Chem. Sci.*, **2004**, 2(1), 96–100.
[2] PM Siva Kumar; SK Geetha Babu; D Mukesh, *Chem. Pharm. Bull.*, **2007**, 55(1), 44–49.
[3] E Francesco; G Salvatore; M Luigi; C Massimo, *Phytochem.*, **2007**, 68, 939–953.
[4] HK Hsieh; LT Tsao; JP Wang, *J. Pharm. Pharmacol.*, **2000**, 52, 163–171.
[5] CL Miranda; GLM Aponso; JF Stevens; ML Deinzer; DR Buhler, *J. Agric. Food Chem.*, **2000**, 48, 3876–3884.
[6] M Liu; P Wilairat; LM Go, *J. Med. Chem.*, **2001**, 44, 4443–4452.
[7] S Mukarami; M Muramatsu; H Aihara; S Otomo, *Biochem. Pharmacol.*, **1991**, 42, 1447– 1451.

[8] A Rajendra Prasad; A Lakshmana Rao; R Rambabu, *E- J. Chem.*, **2008**, 5(3), 461–466.
[9] J Arul Moli; S Vasanthi; N Prakash; D Roop Singh, *High Perform. Polym.*, **2012,** 24(6), 507–520.
[10] M Chitra; D Reuben Jonathan; YC Rajan; V Duraipandiyan, *Int. J. Chem. Appl.*, **2013**, 5(2), 241–250.
[11] E Arumugasamy; B Baskar; V Kannappan, *J. Polym. Mater.*, **2000**,17, 4–9.
[12] P Rajakumar; K Ganesan; S Jayavelu; K Murugesan, *Syn. Lett*, **2006**, 11, 1121–1124.
[13] J Sidharthan; D Reuben Jonathan; T Peter Amaladhas, *Int. J. Chem. Appl.*, **2012,** 4(3), 241–250.
[14] R Sugaraj Samuel; D Reuben Jonathan; Y Christurajan; S Jayakumar; R. Pichai, *Ind. J. Sci. Tech.*, **2010**, 3(6), 696–701.
[15] M Chitra; TV Rajendran; V Duraipandiyan; YC Rajan; D Reuben Jonathan, *Ind. J. Sci. Tech.*, **2010**, 3(8), 890–893.
[16] R Senthamizh Selvi; R Nanthini; G Sukanyaa, *J. Chem. Pharm. Res*., **2012**, 4(1): 393–397.
[17] KA Nandekar; JR Dontulwar; WB Gurnule, *J. Chem. Pharm. Res*., **2012**, 4(7), 3628–3636.
[18] N Malathi; D Roop Singh, *Ind. J. Sci. Tech.,* **2012**, 5, 2302–2306.
[19] YC Rajan; CC Kanakam; S Periyar Selvam; K Murugesan, *Tetrahedron Lett.,* **200**7, 48, 8562–8565.
[20] V Kannappan; D Reuben Jonathan, *J. Chem. Pharm. Res.*, **2013**, 5(4), 393–397.
[21] Sheenam Watts; Ramhari Meena; RV Singh, *J. Chem. Pharm. Res*., **2013**, 5(10), 260–265.