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Antifungal Properties of a New Series of Terpolymer Resins Derived from Benzyl-2,4-dihydroxyphenyl Ketone, Formaldehyde/Furfural and 4-Oxoacids

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ABSTRACT

Antifungal properties of a new series of terpolymer resins, prepared by the polycondensation reaction of benzyl-2,4-dihydroxyphenyl keton (BPK), formaldehyde/furfural and 4-oxoacids using acid catalyst, was studied against two pathogenic fungi such as Fusarium oxysporum and Dreschlera halodes. All the newly prepared resins were characterised by spectral data, physicochemical and thermal analyses. The copolymer resins (BPK-formaldehyde and BPK-furfural) were devoid of antifungal activity, while all the terpolymer resins were toxic at one or other concentrations. Within the terpolymer resins, furfural based resins were highly toxic against both the fungi and caused total germination inhibition at 640 ppm concentration. These terpolymer resins were equipotent with that of standard Clotrimazole. Both the pathogens were equivally sensitive to the terpolymer resins.

Keywords: Terpolymer resins; Polycondensation; 4-oxoacids; Spectral analysis; Antifungal activity

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INTRODUCTION

The application of polymers in all spheres of life has been abundantly increased in recent years. On the other hand, invasion of polymers by microorganisms such as bacteria, fungi etc., is manifested by the loss of mechanical properties, surface degradation, discolouration, staining and other deteriorations leading to loss of their appearance and strength [1,2]. Generally addition of a biocide during polymer compounding is the most usual way to prevent the deterioration of polymers by microorganisms [3]. However, the biocide released from the polymer during its usage may be a hazard to the environment and also contribute to the growth of resistant microbial strains; hence protection is limited in the time scale. Fixation of a biocide on the polymer backbone by hydrolysable bond results only in a better control of time during which the polymer is protected, but this does not solve the problem of toxicity due to progressive loss of biocide with time [4,5]. A different and the best approach are to bind a biocidal group(s) to the polymer through covalent bond that will not be cleaved by microorganisms. The chemical group(s) present at the surface of modified polymer is active by contact with microorganism cells, a permanent activity may be

expected without liberation of toxic products to the environment. In-fact the use of polymers as antimicrobial agents presents several advantages, since these products usually exhibit long-term activity, limited residual toxicity, chemically stable, non-volatile, non-leaching and do not permeate through the skin.

Although various workers have prepared several biocidal polymers [6-12], but there is a noteworthy demand to synthesise polymers with permanent biocidal activity without liberation of toxic products to the environment. In this aspect, terpolymer resins are good candidates for this purpose. Therefore, in search of non-leaching eco-friendly biocides, it was considered worthwhile to screen some of the newly synthesised terpolymer resins, from benzyl-2,4-dihydroxyphenyl ketone (BPK), formaldehyde/furfural and 4-oxoacids, for their antifungal activity. This paper presents the synthesis, characterisation and antifungal properties of the newly synthesised terpolymer resins, hitherto unreported.

EXPERIMENTAL SECTION

All the chemicals used were of Analar or chemically pure grade and used after further purification. Formaldehyde (BDH) and furfural (S.D fine) were used as received. The monomer,benzyl-2,4-dihydroxyphenyl ketone (BPK) was prepared by following the reported procedure [13] and its purity was checked with its melting point (115°C) and spectral data. 4-Oxoacids (4-oxo-4-arylbutanoic acids) were prepared according to the reported procedures [14,15] and their chemical purity was checked by spectral analysis. Infrared spectra of the resins were recorded with the Perkin Elmer model BX-I spectrometer. ^{1}H–NMR spectra were recorded on Bruker BVT 3300 spectrometer, usind DMSO-d_6 solvent.

Resin Synthesis

A mixture of monomer BPK (0.01 mol), comonomer 4-oxoacid (0.01 mol) and condensing reagent formaldehyde/furfural (0.05 mol) was refluxed, in presence of 1:1 (v/v) hydrochloric acid and glacial acetic acid, in an oil bath for about six hours with intermittent shaking. After completion of reaction, the contents were poured in ice-cold water with stirring. The separated product was filtered off, washed repeatedly with hot distilled water and finally with methanol to remove unreacted reactants. The resin thus obtained was reprecipitated using dimethyl sulphoxide (DMSO) solvent and methanol non-solvent, vacuum dried and used for spectral characterisation and for the antifungal assay.

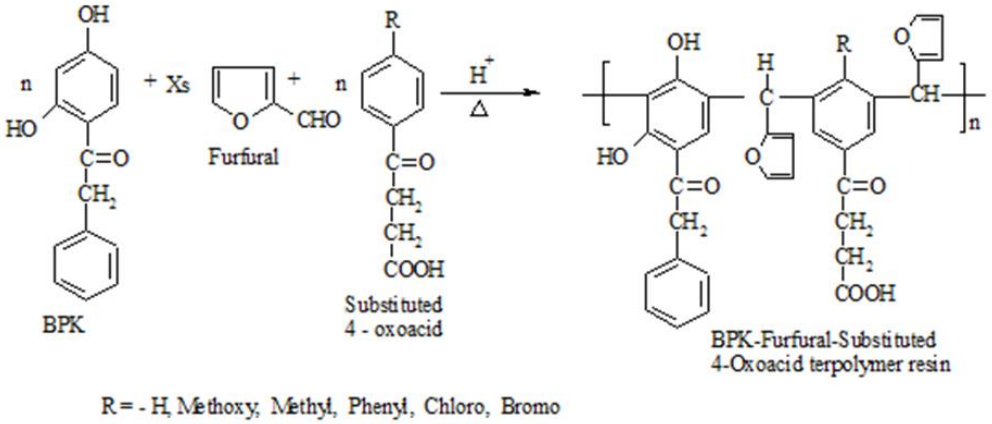
Antifungal Assay

Monosporic cultures of *Fusarium oxysporum* and *Dreschlera halodes* isolated from diseased fruits of tomato (*lycopersicon esculentum)* and maintained on Asthana and Hawker's medium-A (glucose 5g, potassium nitrate 3.5g, potassium dihydrogen phosphate 1.7g, magnesium sulphate 0.75g, agar-agar 15g and distilled water 1 liter) was employed for these studies. The antifungal activity of the resins was assayed by glass slide humid chamber technique as described by Horsfall [16], using varying concentrations of DMSO extract of the resin. The spore suspension of the fungi was prepared in resin solution of different concentrations so as to appear 30 – 40 spores in high power microscopic filed. A drop of such solution was placed on the sterilised glass slide and 100 % relative humidity was maintained by placing moistened sterilised blotter at the bottom of a petridish and incubacted at 28 ± 2 °C for 24 hours. At the end of incubation period, spores germinated and non-germinated were scored in ten

randomly selected microscopic fields so as to cover 350 – 400 spores. The percentage of spore germination inhibition was calculated as: % of spore germination inhibition = percentage of spore germination in the treated x 100/percentage of spore germination in control. Distilled water in place of resin solution was served as control. Average of three replicates was used to calculate the percentage of spore germination inhibition.

RESULTS AND DISCUSSION

The polycondensation of benzyl-2,4-dihydroxyphenyl ketone (BPK) with furfural and substituted 4-oxoacid may be represented as in Scheme 1.



Scheme 1. Synthesis of terpolymer resin

A large number of resins were prepared using different combinations of monomer (BPK), condensing reagents (formaldehyde/furfural) and comonomers (4-oxoacid and substituted 4-oxoacids) in the presence of acid catalyst. All these newly prepared resins were characterised by their spectral data, physicochemical studies and thermal analyses. The spectral data and possible assignments for a few resins are presented in Tables 1 and 2. X-ray diffraction studies revealed that the copolymer resins and phenyl substituted terpolymer resins were amorphous whereas other terpolymer resins were crystalline (Table 3). Formaldehyde based resins exhibited different colours (depending on the chemical nature of the comonomer), while furfural based resins were exclusively black or dark brown. Solubility behaviour of these resins was determined by using solvents of varying solubility parameter. All these resins were insoluble in water, methanol and toluene; sparingly soluble in acetone and chloroform; soluble in dimethyl formamide (DMF) and dimethyl sulphoxide (DMSO). Number–average molecular weight (Mn) of the resins, determined by non-aqueous conductometric titration [17,18] revealed that the Mn lies between 1920 and 3475. Thermogravimetric analysis indicated that the resins were thermally stable and the 50% decomposition/weight loss of resins occurred above 500°C temperature.

Table 1. FTIR spectral analysis of terpolymer resins

Frequency ($\bar{\upsilon}$, cm^{-1})	Assignment of absorption bands
BPK – Formaldehyde-Paramethoxy-4-Oxoacid terpolymer resin	
3423	O – H Stretching of monomer moiety
2984	C – H Stretching of aromatic rings

2927	C – H Stretching of methylene groups
1682	C = O Stretching of comonomer (P-OCH_3OXA) moiety
1620	C = O Stretching of monomer moiety
1593	C = C Stretching of aromatic rings
1449	– CH_2 – Bending of methylene bridge
1374	O – H In-plane bending of monomer moiety
1325	C – O Stretching of comonomer (P-OCH_3OXA) moiety
1232	C – O Stretching of monomer moiety
840	Stretching vibration of tetra substituted phenyl ring
BPK – Furfural-Parachloro-4-Oxoacid terpolymer resin	
3445	O – H Stretching of monomer moiety
3000	C – H Stretching of aromatic (benzene & furan) rings
2928	C – H Stretching of methylene groups
1686	C = O Stretching of comonomer (P-ClOXA) moiety
1650	C = O Stretching of monomer moiety
1600	C = C Stretching of aromatic rings
1470	– CH – Bending vibration of methine bridge
1320	O – H In-plane bending of monomer moiety
1270	C – O Stretching of comonomer (P-ClOXA) moiety
960, 915, 853	Furanyl ring vibrations of furfural moiety
832	Stretching vibration of tetra substituted phenyl ring
635	C –Cl stretching vibration of comonomer (P-ClOXA) moiety

Table 2. ^{1}H–NMR spectral analysis of terpolymer resins

δ (ppm)	Assignments
BPK – Formaldehyde - Parabromo-4-Oxoacid terpolymer resin	
3.9 (2H)s	– CH_2 – Protons of terminal methylol group
4.0 (2H)s	Bridged – CH_2 – group protons
4.1 – 4.6 (6H)m	- CH_2 – group protons of monomer and comonomer (P-BrOXA) moieties
6.2 (1H)s	Para – OH group proton of the monomer moiety
7.1 (5H)s	Phenyl group protons of the monomer moiety
7.3 (1H)s	Meta hydrogen of the terminal monomer moiety
7.4 (1H)s	Ortho hydrogen of the monomer moiety
7.8 (2H)s	Ortho hydrogens of the comonomer (P-BrOXA) moiety
12.4 (1H)s	Ortho – OH group proton (H-bonded) of the monomer moiety
14.2 (1H)s	Carboxylic group proton of comonomer (P-BrOXA) moiety
BPK – Furfural-Parachloro-4-Oxoacid terpolymer resin	
4.1 – 4.3 (6H)m	– CH_2 – group protons of monomer and comonomer moieties

6.3 (1H)s	Para – OH group proton of the monomer moiety.
6.35 (1H)s	Triaryl – CH – group proton (bridged)
7.2 (5H)s	Phenyl group protons of the monomer moiety
6.4 – 7.9 (3H)m	Furanyl ring portions of furfural moiety
7.39 (1H)s	Ortho hydrogen of the monomer moiety
7.40 (2H)s	Ortho hydrogens of the comonomer (P-ClOXA) moiety
12.5 (1H)s	Ortho – OH group proton (H-bonded) of the monomer moiety
14.2 (1H)s	Carboxylic group proton of comonomer (P-BrOXA) moiety

Table 3. Physicochemical properties of the newly synthesised terpolymer resins

R.No.	Terpolymer resin	Yield %	Colour	Nature	MP/Dec. temp. (°C)	Density (gm/cm^3)	$\overline{Mn}$
R-01	BPK-FM (copolymer)	80	Pale yellow	Amorphous	282	1.0611	1920
R-02	BPK-FM-OXA	67	Yellow	Crystalline	273	1.1721	3010
R-03	BPK-FM-P-OCH_3OXA	65	Yellow	Crystalline	278	1.1919	2963
R-04	BPK-FM-P-CH_3OXA	72	Orange	Crystalline	269	1.1894	2986
R-05	BPK-FM-P-C_6H_5OXA	68	Light Brown	Amorphous	280	1.0996	3036
R-06	BPK-FM-P-ClOXA	70	Yellow	Crystalline	305	1.1792	3189
R-07	BPK-FM-P-BrOXA	72	Brown	Crystalline	312	1.2026	3256
R-08	BPK-FF (copolymer)	76	Black	Amorphous	>360	1.1812	2217
R-09	BPK-FF-OXA	72	Black	Crystalline	>360	1.2712	3106
R-10	BPK-FF-P-OCH_3OXA	73	Black	Crystalline	>360	1.2189	3014
R-11	BPK-FF-P-CH_3OXA	70	Dark brown	Crystalline	>360	1.2175	3217
R-12	BPK-FF-P-C_6H_5OXA	68	Black	Amorphous	>360	1.2366	3244
R-13	BPK-FF-P-ClOXA	69	Dark brown	Crystalline	>360	1.2738	3212
R-14	BPK-FF-P-BrOXA	67	Black	Crystalline	>360	1.2322	3475

BPK: Benzyl-2,4-dihydroxyphenyl ketone; FM: Formaldehyde; FF: Furfural; OXA: 4-Oxoacid (4-Oxo-4-phenylbutanoic acid).

Table 4. Antifungal activity of the newly synthesised terpolymer resins against Fusarium oxysporum and Dreschlera halodes*

R.No.	Terpolymer resin	Percentage of spore germination inhibition			
		160	320	480	640 (ppm)
R-01	BPK-Formaldehyde (Copolymer)	NA (NA)*	NA (NA)	NA (NA)	NA (NA)
R-02	BPK-Formaldehyde-4-Oxoacid	9.26 (8.76)	18.00 (17.94)	25.73 (23.00)	44.15 (43.16)
R-03	BPK-Formaldehyde-*P*-Methoxy-4-oxoacid	17.63 (9.26)	33.42 (25.77)	44.61 (34.72)	80.00 (60.88)
R-04	BPK-Formaldehyde-*P*-Methyl-4-oxoacid	24.38 (18.61)	53.53 (33.52)	71.74 (68.78)	100.00 (100.00)
R-05	BPK-Formaldehyde-*P*-Phenyl-4-oxoacid	20.66 (17.02)	37.40 (34.13)	75.01 (59.15)	93.76 (87.47)
R-06	BPK-Formaldehyde-*P*-Chloro-4-oxoacid	23.63 (20.74)	63.63 (57.79)	76.21 (75.72)	100.00 (100.00)
R-07	BPK-Formaldehyde-*P*-Bromo-4-oxoacid	22.42 (18.64)	62.17 (60.10)	70.84 (71.05)	100.00 (100.00)
R-08	BPK-Furfural (copolymer)	NA (NA)	NA (NA)	NA (NA)	NA (NA)
R-09	BPK-Formaldehyde-4-Oxoacid	15.78 (9.55)	22.64 (20.77)	34.33 (32.44)	48.74 (46.71)
R-10	BPK- Furfural-*P*-Methoxy-4-oxoacid	18.29 (15.33)	22.84 (20.96)	51.82 (49.25)	74.76 (67.84)
R-11	BPK- Furfural-*P*-Methyl-4-oxoacid	20.66 (17.08)	38.00 (34.36)	74.71 (68.15)	100.00 (100.00)
R-12	BPK- Furfural-*P*-Phenyl-4-oxoacid	18.17 (18.01)	43.12 (41.62)	67.45 (61.01)	100.00 (100.00)
R-13	BPK- Furfural-*P*-Chloro-4-oxoacid	24.55 (23.32)	58.02 (60.15)	87.25 (86.43)	100.00 (100.00)
R-14	BPK- Furfural-*P*-Bromo-4-oxoacid	22.47 (20.97)	53.85 (50.00)	77.92 (76.65)	100.00 (100.00)
15	Clotrimazole (Reference)	-	-	100.00 (100.00)	-

*Values in parentheses are the results obtained with *D.halodes*; NA: Not active.

The antifungal activity of resins, screened against the pathogenic fungi such as *Fusarium oxysporum* and *Dreschlera halodes*, revealed that the toxicity of resins varied significantly both with the chemical nature of resins and the fungi employed. Copolymer resins such as BPK formaldehyde (R-01) and BPK – furfural (R-08) were devoid of toxicity as they failed to inhibit the spore germination of both the fungi. On the other hand, the terpoloymer resins exhibited good to excellent antifungal activity against the fungi under investigation (Table 4).

Many of the newly synthesized terpolymer resins such as BPK-formaldehyde-*p*-methyl-4-oxoacid (R-04), BPK-formaldehyde-*p*-chloro-4-oxoacid (R-06), BPK-formaldehyde-*p*-bromo-4-oxoacid (R-07), BPK-furfural-*p*-methyl-4-oxoacid (R-11), BPK-furfural-*p*-phenyl-4-oxoacid (R-12), BPK-furfural-*p*-chloro-4-oxoacid (R-13) and BPK-furfural-*p*-bromo-4-oxoacid (R-14) were highly toxic as characterised by their complete (100%) spore germination inhibition at 640 ppm concentration with both the test organisms, and the activity is almost equal to the standard clotrimazole. Other terpolymer resins exhibited moderate to good fungicidal property against the tested fungi. In general, *F. oxysporum* is more sensitive than *D.halodes* and the terpolymer resins with methyl and halogen (Cl, Br) substituents are displaying significant antifungal activity than the resins with other substitutents. The antifungal activity of resins under study could be referred to a number of causes like injurious effect on the cell wall or cell division, effect on permeability of cell membrane and cell enzyme system, chalation and precipitation of chemicals. Infact, a synergistic structural effects are playing a role in fungicidal property of resins and no single factor may be responsible for such activity. Therefore, it is difficult at this stage to correlate the structure-activity relationship.

CONCLUSION

Most of the prepared tempolymer resins were found to have excellent antifungal activity against the chosen fungi, which may be exploited as water insoluble surface coatings. The use of these terpolymer resins result a permanent activity to the material without liberation of toxic products to the environment. Hence recommended for the preparation of surface coatings; which would serve as non-leaching, eco-friendly antifouling biocidal coatings with long-life.

REFERENCES

[1] CU Pittman; KS Ramachandran; KR Lawryer. *J Coating Technol.* **1982**, 54, 27-40.

[2] G Patrick; A Werner; B Hans Iris; D Peter; E Michael; W Thomas. *US Patent.* **2005**, 200543, 499.

[3] C Potin; A Pleurdeau; CM Bruneau. *Chim Peintures.* **1984**, 347, 15-33.

[4] DH Lewis.*Controlled Release of Bioactive Materials*, Plenum press, New York, **1981**, 161-170.

[5] J Hazziza-Laskar; N Nurdin; G Helary; G Sauvet. *J Appl Polymer Sci.* **1993**, 50, 801-806.

[6] C Soykan; I Erol, *J Polymer Sci A.* **2003**, 41(3), 1942-1951.

[7] SS Rahangdale; WB Gurnule. *Der Pharmachemica.* **2011**, 3 (4), 314-322.

[8] MA Riswan Ahmad; RS Azarudeen; M Karunakaran; AR Burkanudeen. *Iranian Polymer J.* **2010**, 19(8), 635-646.

[9] HJ Patel; MG Patel; AK Patel; KH Patel; RM Patel. *Express Polymer Letters.* **2008**, 2(10), 727-734.

[10] AR Burknudeen; MA Riswan Ahmad; RS Azarudeen; M Shabana Begum; WB Gurnule. *Arabian J Chem.* **2011**, 4, 33-43.

[11] AD Kushwaha; AB Kalambe; VV Hiwase; DN Urade. *J Chem Pharm Res.* **2012**, 4(2), 1111-1116.

[12] M Ravichander; G Anand Reddy; V Malathi; P Jalapathi; T Raja Komuraiah. *J Chem Pharm Res.* **2015**, 7(9), 868-871.

[13] P Price; SSJ Israelstan. *Organic Chem.* **1964**, 29, 2800-2803.

[14] LF Fieser; AM Saligman. *J Amer Chem Society.* **1938**, 60, 170-176.

[15] WG Douben; RE Adams. *J Amer Chem Society.* **1948**, 70, 1559-1566.

[16] JS Horsfall. *Principles of Fungicidal Action.* Waltham MA, **1956**,176-178.

[17] SK Chatterjee; UB Agarwal. *J Polymer Sci Polymer Chem.* **1971**, 3225-3230.

[18] SK Chatterjee; N Datta Gupta. *J Macromolecular Sci: Chem.* **1974**, A8 (2), 451- 460.