ANTIMICROBIAL EFFICACY OF EPOXY-MIXTURES OF ZEOLITE DERIVATIVES MATRIX COATING ON MS-STEEL PANELS

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Abstract
Mixtures of various Zeolite derivatives Viz; Propolis-Zeolite (PZ), Silver-Zeolite (SZ), and Silver-Sulfadiazine- Zeolite (ASZ) were applied as Matrix into commercial epoxy resin. The obtained coating materials were coated on MS steel panels. The uncoated materials were also taken in powder form. The antimicrobial activity of coated MS steel panels and of powder were evaluated by Agar cup method. The common bacteria *Staphylococcus aereus, Streptococcus pyrogenaes, Escherichia Coli., Pseudomonas aeruginos* and fungi *Aspergillus Niger, Aspergillus Flavus, Penicillium expansum, Candida albicans, Rhizopus nigrieuns, Botrydepladia theobromine* were used. The result show that the coating materials have excellent antimicrobial activity. The results indicate that such coating material can be used as food packaging materials.

Keywords: Epoxy Resin, Hardener, Zeolite derivatives, MS steel panel, Silver propolis, Sulfadiazine, antibacterial activity, fungi, antifungal activity.

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Introduction

There is increasing interest towards the development of materials which possess anti-microbial properties. Biomaterials impregnated with different types of organic antimicrobial material and used in medical field since many years [1-6]. The inorganic compounds such as silver and other heavy metals are also used as antimicrobial agents[7-9].These antimicrobial agents have also been impregnated into others materials like carpets, adhesives, road paving, food packaging, cleaners etc [10-14].

The Silver and silver-organo compounds are well known antimicrobial agents[15].Further zeolites and their derivatives also exhibit antimicrobial properties[16-20]. The recent review indicated that the epoxy embedded zeolite coating inhibit the growth of bacteria [21-22]. Recently, present authors reported the antimicrobial activity of mixture of zeolite derivatives [23]. In continuation of this work the present paper describes the antimicrobial efficacy of epoxy resin formulation having mixture of various zeolite derivatives viz; Propolis-Zeolite (PZ), Silver-Zeolite (SZ), and Silver-Sulfadiazine- Zeolite (ASZ). The above formulation was coated on MS steel panels and antimicrobial activity measurement was carried out on MS steel panel and non-coated material.

Experimental

A: Antimicrobial Zeolite preparation

The following zeolite derivatives were prepared using the methods reported [24-26] (Table – 1).

(i) Propolis-Zeolite (PZ)
(ii) Silver-Zeolite (SZ)
(iii) Silver-Sulfadiazine Zeolite (ASZ)

Table 1: Mixture of Zeolite derivatives

Mixture No.	Mixture of Zeolite derivatives (wt. in g)		
	PZ	ASZ	SZ
1	5	50	45
2	10	50	40
3	20	50	30
4	30	50	20
5	40	50	10
6	45	50	5

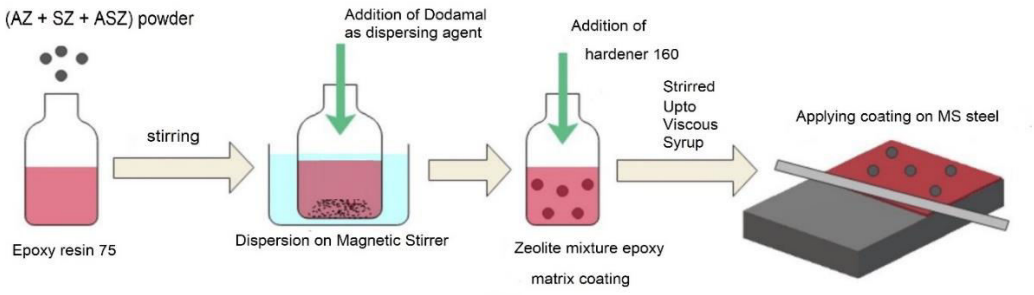
The so-called Zeolite derivatives were grounded to fine particles upto 300 mesh size and dried in hot oven upto 100°C.

B: Epoxy resin and Hardener:

The epoxy resin and hardener ready for coating were purchased form local market. Their specification are as follow:

Epoxy resin 75: Solid content 75%
Epoxy Eq. Wt.: 2.1
Solvent: Xylene
Viscosity at 25°C: 12000 mpas
Epoxy Hardener 160: Polyamide 160
Amine Value: 350-400 mg KOH/gm
Viscosity at 25°C: 400-900 mpas

C: Incorporation of Antimicrobial Zeolite into Epoxy resin:



Antimicrobial Zeolite sample mixtures (10% by wt.) (Table -1) of a particle size 300 mesh (50 µm) were added to epoxy resin using a high-speed dispersion device. In order to provide homogeneous dispersion of antimicrobial zeolite particles pinch of a dispersant agent i.e., Dodamal was added. Then the hardener 160 (one fourth in volume) was added and dispersed for an hour of high-speed ensuring laminar flow. Six epoxy coatings were prepared by various antimicrobial zeolite derivatives mixture.

The coatings were applied by hand lay out process (ASTM D4147-93) on MS Steel panels (3 × 1 cm size). Prior to coaling the panels were degreased with solvent. An excess of coating sample was placed at the end of the MS steel panel and by taking K-Barrod (No. 5) applicator drawn across the substrate panel with uniform pressure. The excess coating material dushed through edge of panel. The panel was allowed curing at room temperature for at least 7 days. The excess cured coats outside the panel also collected and grounded to powder form.

D: Antimicrobial activity measurement of coatings

The antimicrobial and antifungal activity of all six coatings samples in form of MS steel panels and powder form were carried out:

The following pathogenic strains were used for testing antibacterial and antifungal activities of Zeolite derivatives containing epoxy coating samples.

Bacteria

A. Gram Positive
1. Staphylococcus aereus
2. Streptococcus pyrogenaes
B. Gram Negative
1. Escherichia Coli.
2. Pseudomonas aeruginosa

Fungal Strain:
1. Aspergillus Niger
2. Aspergillus Flavus
3. Penicillium expansum
4. Candida albicans
5. Rhizopus nigrieuns
6. Botrydepladia thiobromine

The antimicrobial activity was investigated using agar disk diffusion method[27-29]. The activity of Zeolite derivatives mixtures containing coatings (1 to 6) was studied against above mentioned bacteria. MS steel panel and reference drug were slurried separately in DMSO solvent. Bacteria from a 24 hr culture containing about 100-105 CFU/ml were spread on nutrient agar (prepared from 1% tryptone, 1% agar, 0.5% NaCl, 0.5 % yeast extract in 1 litre water at pH = 7) which was autoclaved at 120°C for half an hour. Then these disks were loaded with the sample slurried in DMSO. Ciprofloxacin (1mg/ml) was used as a reference. The disks with MS steel panel and coating powder were kept in an incubator at 37°C for 2 days and examined for inhibition zone of various zeolite derivatives mixtures. The experiment performed in duplicate. The inhibition zone was measured by calliper to receive mean value in mm.

All the coatings (1 to 6) were screened separately for their antifungal activity. The antifungal activity was performed by agar cup method.

The coated MS steel panels and coating powder 50mg each were separately placed in sterile Petry dish. A potato dextrose agar (PDA) (40:10:15) thick syrup was prepared and sterilised by autoclaving at 120 °C under pressure for half an hour. The sterilised syrup (20ml) poured into each sterilised Petry dish and kept aside in an incubator till it got solidified. Fungal strain (listed above) about 100-105 (FU/ml) was spread out uniformly on solidified perty dish by sterilised folded glass road. Dishes were left for 10 minutes till the culture is properly absorbed on the surface of PDA. Fluconazole (1mg/ml) was used as a reference. The petry dishes were kept for incubation at 30°C for 5 days. Then inhibition zone was measured in percentage.

Results and Discussion

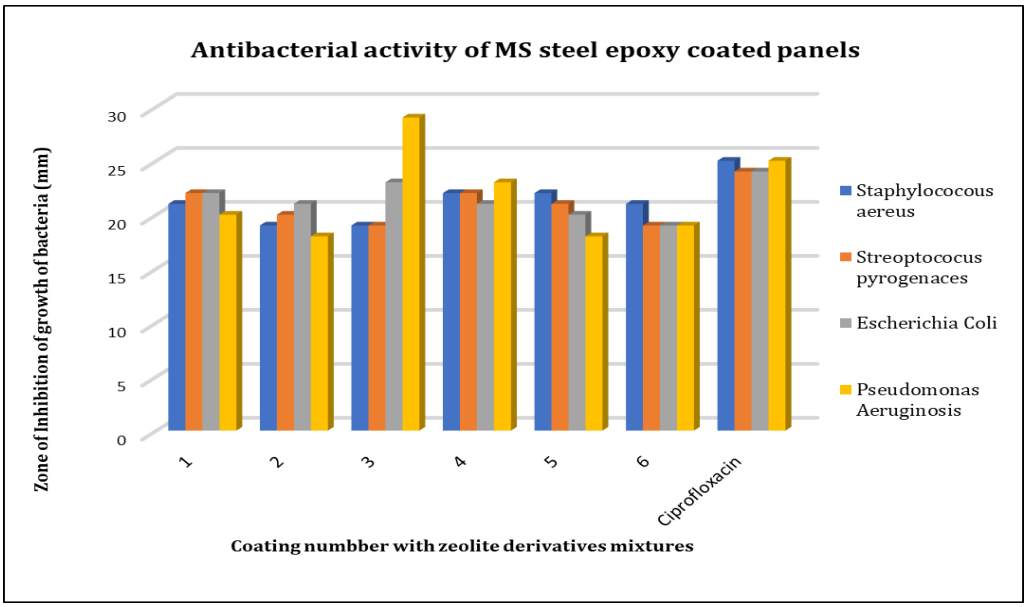
The present study focuses on the effect of mixtures of antimicrobial zeolite derivatives reported earlier [23] on epoxy resin coating against various bacterial and fungal strains. Tables 2 and 3 show the activity of both the epoxy coatings, on MS steel coating panels and coating powders.

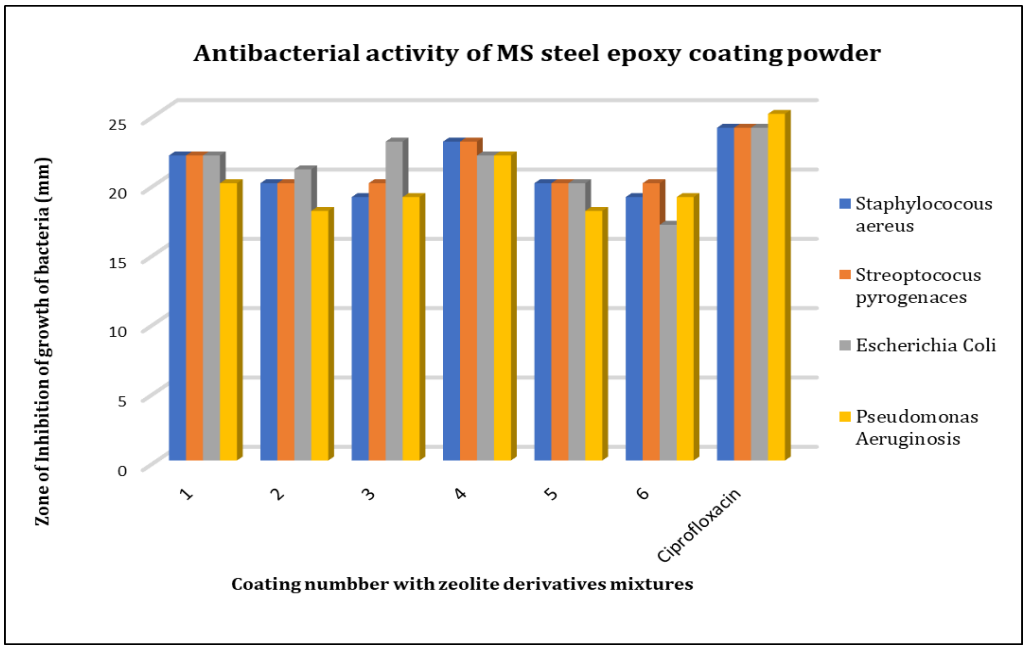
Table – 2: Antibacterial activity of MS steel epoxy coated panels and epoxy coating powder

Coating number with zeolite derivatives mixtures	Zone of inhibition of growth of bacteria (mm)							
	Gram +Ve bacteria				Gram – Ve bacteria			
	Staphylococous aereus		*Streoptococus pyrogenaces*		*Escherichia Coli*		*Pseudomonas Aeruginosis*	
	Mild steel panel	Coating powder	Mild steel panel	Coating powder	Mild steel panel	Coating powder	Mild steel panel	Coating powder
1	21	22	22	22	22	22	20	20
2	19	20	20	20	21	21	18	18
3	19	19	19	20	23	23	29	19
4	22	23	22	23	21	22	23	22
5	22	20	21	20	20	20	18	18
6	21	19	19	20	19	17	19	19
Ciprofloxacin	25	24	24	24	24	24	25	25

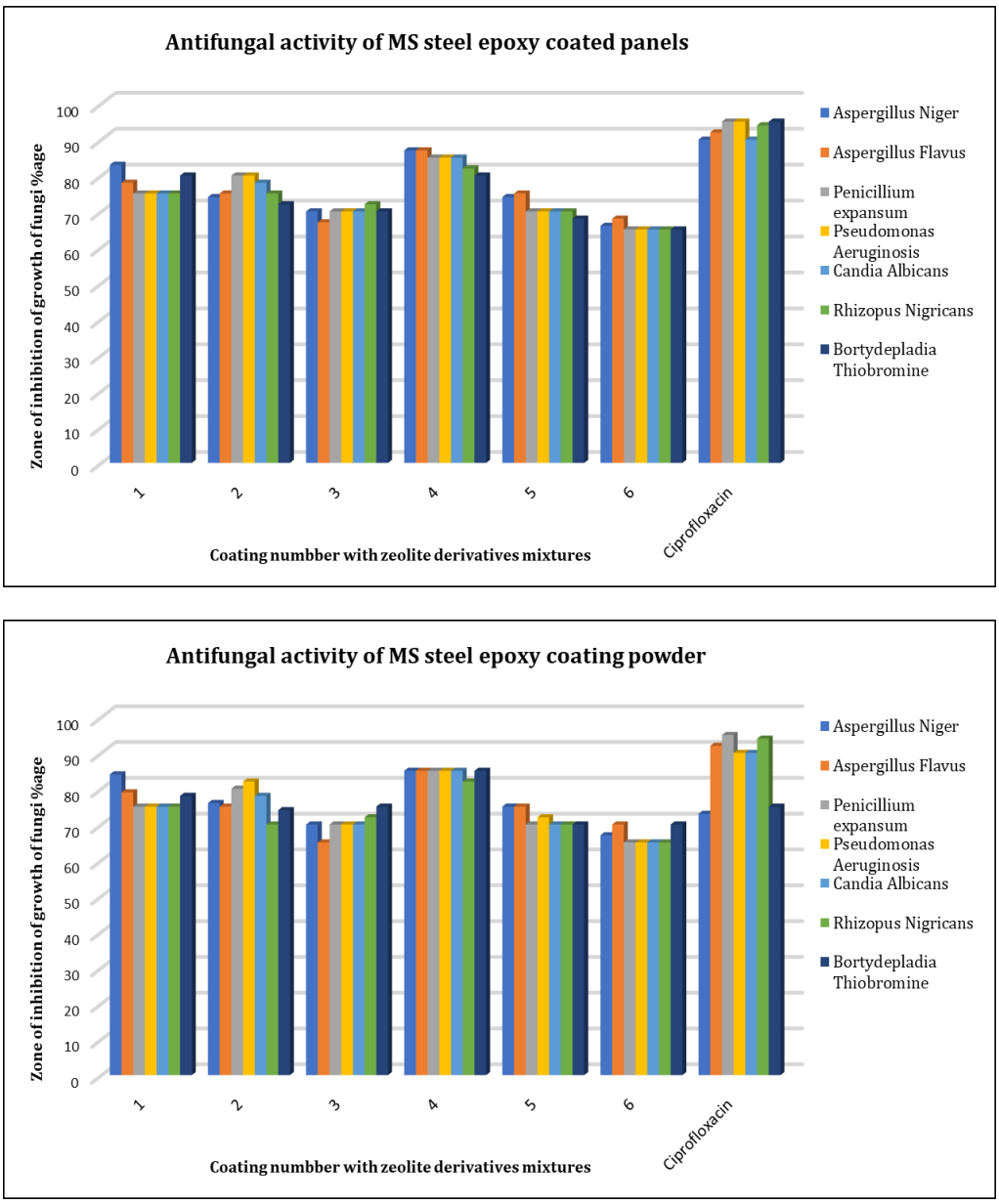
Table – 3: Antifungal activity of MS steel epoxy coated panels and epoxy coating powder

Coating number with zeolite derivatives mixtures	Zone of inhibition of growth of fungi % age											
	Aspergillus Niger		*Aspergillus Flavus*		*Penicillium expansum*		*Candia Albicans*		*Rhizopus Nigricans*		*Bortydepladia Thiobromine*	
	Mild steel Coating	Coating powder	Mild steel panel	Coating powder	Mild steel Coating	Coating powder	Mild steel panel	Coating powder	Mild steel panel	Coating powder	Mild steel panel	Coating powder
1	83	84	78	79	75	75	75	75	75	75	80	78
2	74	76	75	75	80	80	78	82	75	70	72	74
3	70	70	67	65	70	70	70	70	72	72	70	75
4	87	85	87	85	85	85	85	85	82	82	80	85
5	74	75	75	75	70	70	70	72	70	70	68	70
6	66	67	68	70	65	65	65	65	65	65	65	70
Ciprofloxacin	90	73	92	92	95	95	90	90	94	94	95	75





Histogram 1 A and B: Antibacterial activity of MS steel epoxy coated panels and epoxy coating powder



Histogram 2 A and B : Antifungal activity of MS steel epoxy coated panels and epoxy coating powder

The reported derivatives viz., PZ, SZ, and ASZ [24-26] are already good antimicrobial agents.

Recently the present authors evaluated the antimicrobial activity of the mixture of these derivatives. The results show good antimicrobial activity present study is the extension of that work which covers the novel coating materials prepared by mixtures of zeolite derivatives reported earlier [23]. The epoxy resin coating is chosen selected for the study.

The coating materials are generally manufactured using polymers and are used in food packaging material based on antimicrobial property. These materials can be prepared by incorporation of antimicrobial agents into coating formulation. These agents include many organic compounds, metal salts, plant extracts etc. [30-34]. Several zeolites have been used as antimicrobial agents [35-39].

As expected, all the epoxy coatings (Tables – 2,3) with six mixtures of antimicrobial zeolite derivatives shows the very good results. specifically the coating No. 4 is more antimicrobial. M comparison to others. This may be due to the presence of more content of silver and sulfadiazine drug. It was also observed that more quantity of mixture decolourises the coating film. Therefore up to 10% of mixture has been added. It was also observed that less mesh size of zeolite mixtures led to the inhomogeneous distribution in coating formulation and It also attached the coating thickness to be uneven.

Both the activities for epoxy coatings are presented in Histograms – 1 A, B and 2 A, B.

In this work the antimicrobial activity of epoxy coatings was evaluated using MS steel panel and coatings powder. The results of both studies indicates that in both system high antimicrobial efficiency is achieved against four bacteria and six fungi.

Conclusion

Various mixtures of antimicrobial Zeolite derivatives and epoxy results upto 10% by wt. were incorporated to epoxy resins. The coating formulations prepared by were applied on MS steel panels. The antimicrobial and antifungal activities of MS steel panels and uncoated coatings (i.e. powder form) drugs were measured by agar cup method. The result show similar behaviour for MS steel panel and coating powder. This study suggests that the coating of such mixtures of antimicrobial Zeolite derivatives on substrate like glass, MS steel, Aluminium foil can minimize the microbial contamination of food products surfaces during their storages, transportation, and handling. The coatings of epoxy resin with antimicrobial zeolite can be applied to food packages substrate like metal foils or plastic films or glass plates and thus can be used as an antimicrobial agent in food packaging.

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