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Starch Based Sustainable Tough Hyperbranched Epoxy Thermoset

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With the growing concern to long term environmental and waste management problems, the recent trend in polymer industry is aimed to utilize environmentally benign substrates for development of sustainable polymers with required properties for their potential applications as substitutes for petrochemical derivatives. In this arena, the authors aspired to use starch, a natural renewable polysaccharide obtained from a wide variety of crops, as one of the reactants for a one-pot synthesis of a bio-based sustainable hyperbranched epoxy resin. Nuclear magnetic resonance (^{1}H NMR and ^{13}C NMR), Fourier transformed Infrared spectroscopy (FTIR) along with different analytical techniques confirmed the chemical structure of the resin. The poly(amido amine) cured epoxy thermoset exhibited acceptable biodegradation along with desirable properties. It exhibits excellent impact resistance (>100 cm), outstanding scratch hardness (>10 kg), exceptionally high tensile adhesive strength (up to 2906 MPa for aluminum), moderate tensile strength (up to 29 MPa), good elongation at break (up to 38 %), high toughness (up to 8.40 MJ m^{-3}) and very good chemical resistance against a number of chemical environments. Moreover, the thermoset displayed potent biomedical attributes by exhibiting cytocompatibility with the erythrocytes as obtained through hemolytic assay. Thus, the synthesized eco-friendly and sustainable hyperbranched epoxy thermoset with good toughness and exceptional adhesive strength can be worthy aspirants for replacing petroleum-based epoxy thermosets as an advanced engineering material.

Introduction

Rapid industrialization through scientific intervention has revolutionized the concept of sustainable development among the global communities. Genuine efforts are being made to generate naturally renewable alternatives with sustainable attribute with the depletion of the basic raw materials at an alarming rate, especially petroleum resources,.[1-3] In this milieu, the advancement of bio-based polymers from such renewable resources contributes a noteworthy transformation from conventional petroleum based ones. Further, they minimize the risk of adverse effect of non-biodegradability of discarded matter to health and environment, reduce scarcity of raw material and so on.[4-5] Starch, is a versatile and cheap polysaccharide with reactive primary hydroxyl groups. It is used as a thickener, water soluble binder, emulsifier, gel forming agent etc. Further, starch are used in many non-food items such as paper making, cardboard, textile sizing and adhesives.[6] However, the finished products from native starch suffers from demerits such as, poor process ability, low dimensional stability and mechanical properties. Therefore, various modifications such as blending, derivation and graft copolymerization have been attempted, to develop starch based sustainable polymers with enhanced properties.[7] One of such attempts is the development of environmental friendly biodegradable starch modified plastics.[8-10] Starch based biodegradable blends of poly(β-hydroxyalkanoates) (PHA), polylactide (PLA) and poly(ε-caprolactone) (PCL) have been reported to reduce total raw material cost and enhance their degradability. However, to attain the desired mechanical properties of such blends some additional agents like glycerol, sorbitol, urea and formamide have to be incorporated.[7] It is therefore difficult to achieve the expected level of properties of starch modified polymers to address the demands of many advanced applications. On the contrary, the use of starch as one of the reactants to synthesize industrially important polymer is rarely found in literature, in spite of its many advantages.

Again, amongst different important industrial polymers, epoxy is a unique class of high-performance thermoset because of its outstanding attributes like high mechanical strength, thermostability, chemical resistance, adhesive strength etc.[11] Thus it has garnered widespread attention to the industrial applications such as coatings, adhesives, binder in composites and so on. However, the commercial epoxy thermosets have limited advanced applications, as they possess disadvantages such as brittleness, low toughness, non-biodegradability etc.[12-14] Hence, in the recent past, a few attempts have been made to obtain bio-based epoxy with improved bio-degradability. Efforts have also been made to develop special architectural hyperbranched epoxy to address their other shortcomings. Such special structural polymers possess high reactivity, single step synthetic protocol, low viscosity and presence of large number of end functionalities along with internal cavities or free volume.[15-17] In almost all

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such cases, researchers have used vegetable oil-derivatives, which required additional energy and process step, to achieve biodegradability of the resultant thermosets. However, biodegradability of epoxy thermosets may also be achieved by the incorporation of hydrolysable bio-based component like starch, which is biodegradable in a wide variety of environments[18] to develop epoxy thermosets with sustainable attribute. Further, starch derivatives like isosorbide, (double dehydration product of sorbitol), have been reported to synthesize epoxy resin by etherification with epichlorohydrin in the presence of aqueous NaOH. Łukaszczyk et al. also showed the possibility of curing isosorbide based epoxy resins using typical aliphatic amine based hardeners and acid anhydrides. However, due to absence of aromatic moiety and improper crosslinking in the structure, high curing temperature with longer period of curing time was required. Also, the properties obtained were found to be inadequate.[19] East et al. attempted to cure isosorbide based epoxy resins but the study for both resins and thermosets were insufficient to judge the possibility to replace bisphenol A based epoxy resins.[20] Therefore, the design of a hyperbranched epoxy using starch as one of the reactants is an apt option. Additionally, such hyperbranched epoxy with combination of aromatic and aliphatic moieties may lead to excellent performance and hence may address all the above drawbacks of commercial epoxy thermosets.[21] This type of epoxy may also provide biocompatibility, which is required for different bio-medical applications.

Authors, therefore, wish to report here the synthesis, characterization and properties evaluation of starch based hyperbranched epoxy (HBSE) thermosets to address the shortcomings of commercial epoxy thermosets. The biodegradability and biocompatibility of the thermosets are also delved into. The performance of hyperbranched thermosets is also compared with diglycidyl ether epoxy of bisphenol A (DGEBA) thermoset without using starch to prove the superiority of the former.

Experimental

Materials

Soluble starch with molecular weight 342.30 g/mol was supplied by Sigma Aldrich, Germany and was vacuum dried for overnight, prior to use. Bisphenol-A (BPA, Sisco Research Laboratories Pvt. Ltd., India) was used after recrystallization from toluene. Tetrahydrofuran (THF, SD fine Chem., India) was used after distillation. Epichlorohydrin, (Sisco Research Laboratories Pvt. Ltd., India), sodium hydroxide (NaOH, Rankem, India), sodium sulfate anhydrous, (Merck, Mumbai, India), hydrochloric acid (Merck, Mumbai, India) and ethanol (Merck, Germany) were used as received. Poly(amido amine) hardener (HY 840, Ciba Geigy, Mumbai, India), with an amine value of 6.6–7.5 equiv/kg, was used as supplied. All other reagents used in this investigation were of reagent-grade.

Synthesis of hyperbranched epoxy resin

Starch, BPA and epichlorohydrin (1:3 mol ratio with respect to total hydroxyl groups of starch and BPA) were taken in a two necked round bottomed flask equipped with a condenser, a constant pressure dropping funnel, a magnetic stirrer and a thermometer dipped in an oil bath. The reaction was carried out at 100 °C for 5 h under constant magnetic stirring. At 60 °C, 5 N aqueous solution of NaOH (equivalent to the hydroxyl groups) was added drop wise to the reaction mixture through a dropping funnel, with continuous heating and reached the desired temperature within 10 min. About 1.5-2 h was required to complete the addition of NaOH. The reaction was continued for another 3 h. Then heating was stopped and allowed to cool down with continuous stirring. The white, viscous mass was poured into a separating funnel and washed with brine solution followed by distilled water, three times, to remove the water soluble impurities and unreacted reagents. Finally, the resin was dissolved in THF and vacuum dried at 70 °C to remove excess epichlorohydrin and entrapped water. Using the same technique, three different compositions of epoxy resins with 5, 10 and 20 wt % of starch were synthesized and coded as HBSE5, HBSE10, HBSE20, respectively. In addition to that, DGEBA was also prepared under the same reaction conditions for comparison purpose.

Curing study

For fabrication of HBSE thermosets, proper curing of resin is conducted using poly(amido amine), a resinous hardener system. It is a polycondensation product of diethylene triamine with dimmer acid of oleic acid.[13] An amount of 50 phr with respect to resin of poly(amido amine) hardener was mixed with epoxy resins, separately in a glass beaker to obtain a homogenous mixture of them by hand stirring. A little amount of THF was used to facilitate the mixing and stirred vigorously for about 10 min. The homogenous mixture was then coated uniformly on glass plates (75 mm × 25 mm × 1.3 mm) and commercially available mild steel plates (150 mm × 50 mm ×1.6 mm) for measurement of scratch hardness and impact strength, respectively. The coated plates were then degassed in vacuum desiccators to remove entrapped solvent and kept under room temperature to determine the touch-free time over a period of 24 h. Further, the films were kept under vacuum for another 24 h to keep them moisture free. They were cured at 100 °C followed by post curing at 120 °C for the required period of time as obtained from their respective swelling values, in a hot oven.

Characterization

FTIR spectra of the epoxy resins were recorded by a Nicolet FTIR spectrophotometer (Impact-410, Madison, WI) using KBr pellet. ^{1}H NMR and ^{13}C NMR spectra of the resins were recorded by a 500 MHz AV500 AVANCE-III FT-NMR spectrometer (BROKER, Switzerland), using TMS as the internal standard and d_6-DMSO and $CDCl_3$ as the solvents for ^{1}H NMR and ^{13}C NMR, respectively. The shear viscosities of epoxy

resins with 10 wt.% of starch at 4 different reaction times were determined by using CVO100 Rheometer (Malvern, U.K) at room temperature. The physical properties such as epoxy equivalent, hydroxyl value, solubility, and swelling value of the resins were measured by the standard test methods, as reported in literature.[22-23] Scratch hardness test on the cured films was carried out by a scratch hardness tester (Sheen instrument Ltd., U.K.), with stylus accessory and a travel speed of 30–40 mm/s. Swelling test on the thermosets was done to confirm the proper curing of the films, by immersing weighed amount of the cured films in THF. After 48 h, the final weight W_f of the swollen film was taken. The swelling value (%) was determined by the difference in weight between the dried film and the swollen film, as follows:

Swelling % = $[(W_f - W_i) / W_i] \times 100$ ---------- (1)

Where W_i and W_f are the initial and final weights of the film, respectively. Mechanical properties of the cured films were measured by a Universal Testing Machine (UTM, Zwick Z010, Germany) equipped with a 500 N load cell operated at a crosshead speed of 20 mm/min for tensile strength (ASTM D 638) and elongation at break. The lap shear adhesive strength (ASTM D 897) of the epoxy resins cured with poly(amido amine) were taken for the overlapping interfaces of the adherents (aluminum and plywood) and measured by UTM equipped with a 10kN load cell at a crosshead speed of 50 mm/min.[15] Impact strength of the thermosets was tested by an impact tester (S. C. Dey Co., Kolkata) as per the standard falling weight (ball) method (ASTM D 1709) where a weight of 850 g was allowed to fall on the film coated on a mild steel plate from minimum to maximum falling heights. The impact resistance is the maximum height up to which the film remained undamaged. The gloss characteristic of the cured films was evaluated by using a gloss meter (Minigloss meter, Sheen, UK), at an angle of incidence, 60°. Thermal properties of the epoxy thermosets were evaluated by thermogravimetric analysis (TGA) and differential scanning calorimetry (DSC). Thermogravimetric study was carried out by using a PerkinElmer 4000 thermal instrument in the temperature range of 30–700 °C, at a scanning rate of 10 °C/min, maintaining an inert atmosphere of nitrogen at a gas flow rate of 30 mL/min. The differential scanning calorimetric study was done by a PerkinElmer DSC 6000, USA instrument in the temperature range −30 to +150 °C (starting temperature was 0 °C) following a cycle of heating−cooling−heating under an atmosphere of nitrogen and at a scanning rate of 5 °C/min.

The chemical resistance test was determined in different chemical environments, such as aqueous NaOH (5%), aqueous HCl (10%), aqueous NaCl (15%), aqueous ethanol (20%) and water to study the effect of those chemicals on the thermosets. The small pieces of cured films were kept in 100 mL amber glass bottles containing the aforesaid media at 30 °C. The percent of weight loss was measured after 30 days of test. UV-visible spectra were analyzed by a Hitachi (U-2001, Tokyo, Japan) UV spectrophotometer. The surface morphologies of the biodegraded polymer films and control films (without bacterial strains) were obtained by scanning electron microscopy (SEM, model JSM-6390LV (JEOL)), after platinum coating on the surface.

Biodegradation study

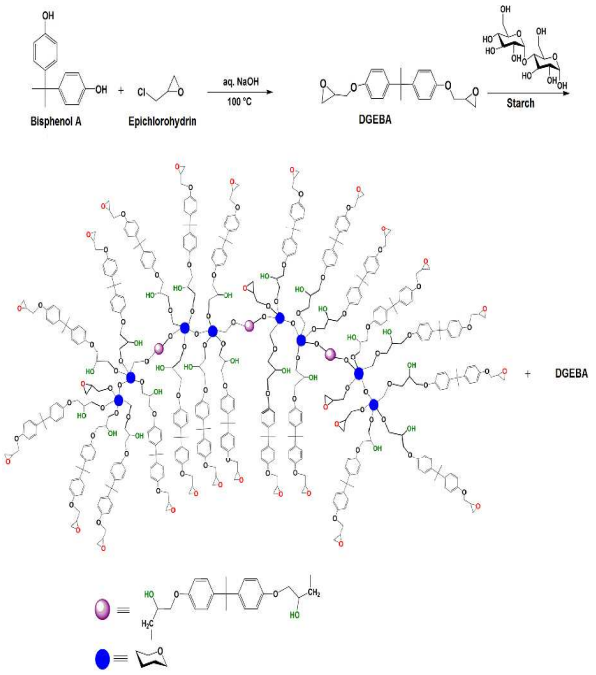
Biodegradation study on the epoxy thermosets were done by McFarland turbidity method using *P. aeruginosa* and *B. subtilus* as the bacterial strains.[23-25] A nutrient salt media was arranged for bacterial growth culture, comprising of 2.0 g $(NH_4)_2SO_4$, 3.61 g KH_2PO_4, 1.75 g $MgSO_4{\cdot}7H_2O$, 0.2 g $CaCl_2{\cdot}2H_2O$, 2.0 g Na_2HPO_4, 50 mg $FeSO_4{\cdot}7H_2O$, 1 mg $CuSO_4{\cdot}7H_2O$, 50µg $MnSO_4{\cdot}5H_2O$, 70 µg $ZnSO_4{\cdot}7H_2O$, 10 µg $H_3BO_3{\cdot}5H_2O$ and 10 µg MoO_3 in 1.0 L in distilled water. An amount of 10 mL of the above prepared liquid culture media was poured into 100 mL conical flasks and was sterilized using autoclave machine at 121 °C and 15 lb pressure for 15 min and allowed to cool down to room temperature. *P. aeruginosa* and *B. subtilus* bacterial strains were cultured in the medium inside an incubator shaker at 37 °C for 48 h. As calculated by the McFarland turbidity method, 100 µL (10^8 microbes/mL) of the cultured medium was taken in a conical flask containing 10 mL of the prepared salt medium and was sterilized by exposure to UV light of wavelength 254 nm. The sterilized films were incubated inside the medium under sterile condition at 37 °C. The flask containing mineral salt medium but without any bacterial strain was used as the control. The extent of biodegradation was studied from the measurement of weight loss (%) of the degraded thermoset films, measured after 6 weeks of exposure to the bacterial strains, based on the following equation.

Weight loss, $W = (W_0 - W_t)/ W_t \times 100$ ---------- (2)

Where W_0 and W_t are the weights of sample before and after degradation, respectively, at the time of interest, t. The bacterial growth is indicated by increase in turbidity of the medium with time. The optical density (OD) of the microorganism was monitored by measuring the absorbance of the medium at 600 nm with respect to the control. The experiment was performed in triplicate. SEM images of the degraded films were taken to study their surface morphology after 6 weeks of bacterial degradation.

Hemolytic assay

To have a preliminary understanding about the cytocompatibility of both HBSE and DGEBA resins and thermosets, a hemolytic assay was performed to investigate the lysis of red blood cell (RBC) membrane by them. Goat's blood was collected from a slaughter house in a heparinized tube containing 4% sodium citrate. It was centrifuged (MPW) at 3000 rpm for 20 min at 4 °C to separate the plasma from erythrocytes and further washed thrice with phosphate buffer saline (PBS). A 5% haematocrit was obtained by re-suspending the packed erythrocytes in PBS (10 mM at pH = 7.4). Different concentrations of samples (0.5, 1, 2, 4, and 6 mg mL^{-1}) were prepared. Finally, 1900 µL of haematocrit along with 100 µL of the test samples were added in each microfuge tube (Eppendorf) and incubated at 37 °C for 30 min.



Scheme 1 Synthesis of HBSE20 resin.

Post-incubation, the cells were kept in an ice bath for 60 s to halt the incubation of cells, followed by centrifugation for 5 min at 3000 rpm at 4 °C. The haemoglobin concentration was determined by taking UV-absorbance of the supernatants at 540 nm, as a measure of haemolysis.[2, 26] The experiment was performed in triplicate.

Results and discussion

Synthesis and characterization

HBSE resins were synthesized by a polycondensation reaction using bisphenol A, epichlorohydrin and starch, keeping the hydroxyl to epichlorohydrin ratio constant, in presence of alkali as given in Table 1. At first, DGEBA was obtained as the reactivity of hydroxyl groups of bisphenol A towards epichlorohydrin is greater than that of starch.[21] Again, the reactivity of primary hydroxyl groups of starch is higher than that of secondary hydroxyl groups. The desired hyperbranched epoxy resin was then obtained by the reaction between the DGEBA and starch. The unreacted hydroxyl groups of starch were reacted with epichlorohydrin, in due course of reaction time to obtain the final hyperbranched epoxy.

Table 1 Composition (mol) of HBSE and DGEBA resins

Reactant	HBSE5	HBSE10	HBSE20	DGEBA
Starch	0.000730	0.00146	0.00292	0
Bisphenol A	0.0219	0.0219	0.0219	0.0219
Epichlorohydrin	0.14965	0.1679	0.2044	0.1314

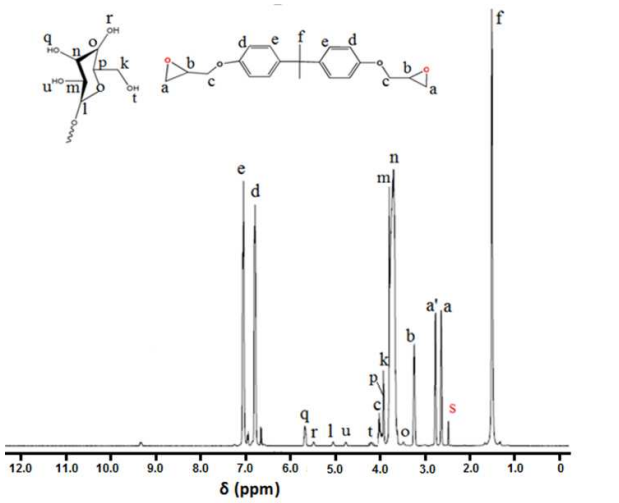


Fig. 1 ^{1}H NMR spectrum of epoxy resin with 10 wt.% just after complete addition of alkali solution.

The viscosity of the resin increases with the reaction time and the reactions were carried out for 5 h at 100 °C so that the hydroxyl groups of starch underwent reaction. This increase in viscosity was confirmed by determining the variation of shear viscosity of the epoxy resin with 10 wt.% of starch at 100 °C for 2 h, 3 h, 4 h and 5 h of reaction time. Under a constant shear stress of 100 Pas, the viscosity increases with reaction time from 0.8 to 3.2 Pas, with a value of 2.4 Pas at 4 h. The low final viscosity is due to unique hyperbranched structure with globular like shape of the epoxy resin.

The proposed structure of HBSE20 resin is shown in Scheme 1. The hydroxyl value, epoxy equivalent and weight percent of starch are calculated theoretically from the proposed structure. This structure contains 28 hydroxyl groups so the calculated hydroxyl value would be 152.55 mg KOH/g whereas the experimental hydroxyl value is 149.93 mg KOH/g. This indicates the presence of 26.6 hydroxyl groups which is close to the theoretical value. Similarly, the weight percent of starch in the given structure is 22 wt.% with respect to bisphenol A. This is close to the experimentally used amount of starch (20 wt.%). Further, the ratio of substituted hydroxyl group/ unsubstitued hydroxyl group is 0.78. This is again close to experimental result (0.72, obtained from ^{1}H NMR spectrum, as discussed later). However, there are 26 epoxy groups in the structure which indicates epoxy equivalent of 386.5 g/equiv. The value deviates from the experimentally observed value of 253.35 g/equiv (39 epoxy groups) which may be due to the presence of DGEBA moiety with epoxy equivalent of 170 g/equiv (theoretical). Thus, this result indirectly supports the formation of hyperbranched structure of the starch based epoxy, though there is certain amount of DGEBA.

Further, for better understanding of the reaction mechanism for the formation of hyperbranched structure, a ^{1}H NMR spectrum was taken just after complete addition of alkali solution. This is just to prove the formation of DGEBA at the initial stage without formation of any starch based epoxy. This is indeed found in the ^{1}H NMR spectrum. The ^{1}H NMR (Fig. 1)

spectrum shows δH, ppm (500 MHz, d_6-DMSO (S), Me_4Si): 3.3 (1H, CH of oxirane ring), 2.6 and 2.8 (2H, CH_2 of oxirane ring), 3.9 (2H, CH_2 next to oxirane ring), 6.8 (4H, aromatic protons of bisphenol A), 7.1 (4H, aromatic protons of bisphenol A), 1.6 (3H, CH_3 of bisphenol A), 3.87 (1H, proton of starch attached to C_5), 3.83 (1H, proton attached to C_6 of starch) 5.01 (1H, proton attached to C_1 of starch), 3.73 (1H, proton attached to C_2 of starch), 3.64 (1H, proton attached to C_3 of starch), 3.45 (1H, proton attached to C_4 of starch), 5.48 (1H, OH attached to C_4 of starch), 5.66 (1H, OH attached to C_3 of starch), 4.21 (1H, OH attached to C_6 of starch), 4.73 (1H, OH attached to C_2 of starch). (C_1, C_2, C_3, C_4, C_5 and C_6 of starch are numbered according to the rules of IUPAC and designated as l, m, n, o, p and k, respectively in Fig. 1). Thus, the peaks in ^{1}H NMR spectrum clearly indicate the presence of DGEBA moiety with unreacted starch moiety. No substitution of hydroxyl group of starch and opening of epoxide group of DGEBA was observed. Therefore at the initial stage, only DGEBA is formed which is transformed to hyperbranched structure with the reaction of hydroxyl groups of starch. However, it is difficult to rule out the presence of some amount of DGEBA moiety in the hyperbranched resins. Thus, the final structure may be a mixture of starch based hyperbranched epoxy with DGEBA.

FTIR spectral analysis

To confirm the different chemical functionalities present in the structure of the epoxy resin, FTIR spectroscopy was used, as shown in Fig. 2. Compared to the IR spectrum of starch, the IR spectra of DGEBA and HBSE5 showed a sharp band for asymmetric vibration of oxirane ring at 912-825 cm^{-1}.[11,12,13,21] The bands at 1604 cm^{-1} and 3059 cm^{-1} are due to aromatic C=C stretching vibrations and aromatic C-H vibrations of bisphenol A moiety in the hyperbranched epoxy HBSE5. The sharp absorption band at 1036 cm^{-1} confirmed the presence of alkyl–aryl ether groups in the epoxy resin.

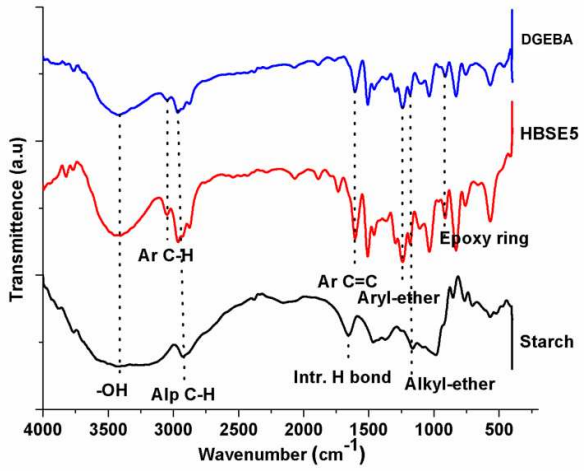


Fig. 2 FT-IR spectra of HBSE5, starch and DGEBA resin

The presence of the band observed at 1247 cm^{-1} is due to the aryl ether linkage in the structure of the resin. The broad and strong absorption band in the region of 3252–3547 cm^{-1} is attributed to the stretching vibration of OH.[27] The bands at 2965-2871 cm^{-1} and 1161 cm^{-1} are assigned to the vibrational absorptions of C-H bond and C-O bond of the starch moiety in the epoxy structure.[27]

H and ^{13}C NMR spectral analysis

The ^{1}H NMR spectrum confirmed the simple structure of the epoxy resin HBSE20. ^{1}H NMR (Fig. 3a) δH, ppm (500 MHz, d_6-DMSO (S), Me_4Si): 3.3 (1H, CH of oxirane ring), 2.6 and 2.8 (2H, CH_2 of oxirane ring), 3.9 (2H, CH_2 next to oxirane ring), 6.8 (4H, aromatic protons of bisphenol A), 7.1 (4H, aromatic protons of bisphenol A), 1.6 (3H, CH_3 of bisphenol A), 3.7 (2H, CH_2 next to bisphenol A), 4.1 (1H, CH attached with OH), 2.02 (1H, OH), 3.64 (1H, proton attached to C_5 of starch), 3.74 (1H, proton attached to C_6 of starch) 4.95 (1H, proton attached to C_1 of starch), 3.43 (1H, proton attached to C_2 of starch), 3.39 (1H, proton attached to C_3 of starch), 3.35 (1H, proton attached to C_4 of starch), 5.38 (1H, OH attached to C_4 of starch), 5.58 (1H, OH attached to C_3 of starch)[28-29,11,21] (C_1, C_2, C_3, C_4, C_5 and C_6 of starch are numbered according to the rules of IUPAC and designated as l, m, n, o, p and k, respectively in Fig. 3a.) The degree of branching of the hyperbranched starch based epoxy was calculated by determining the intensity of the peaks of the substituted and unsubstituted hydroxyl groups from the ^{1}H NMR spectrum of hyperbranched epoxy (Table 2). The considered unsubstituted hydroxyl groups are generated by ring opening reaction of oxirane ring of DGEBA. In the same manner the carbons present in different chemical environments in the structure of hyperbranched epoxy HBSE20 were confirmed by ^{13}C NMR spectrum (Fig. 3b), δC, ppm ($CDCl_3$, S): 31.04 (CH_3, bisphenol A), 42.5 and 50 (oxirane ring carbons), 31.04, 114, 128, 143.5, and 156 (bisphenol A carbons), 68–70 (carbon attached with –OH and ether linkages),[21] 105.5 (C_1 of starch), 79.5 (C_2 of starch), 72.2 (C_3 of starch), 71.1 (C_4 of starch), 85.8 (C_5 of starch), 67.4 (C_6 of starch),[28] (where the carbon atoms C_1, C_2, C_3, C_4, C_5 and C_6 of the starch moiety are numbered according to the rules of IUPAC and are designated as o, p, q, r, n and m, respectively, in Fig. 3b).

Physical properties of the resins

Different compositions of the synthesized starch based hyperbranched epoxy resins appeared to be odorless and colorless viscous mass. The physical properties like hydroxyl value, epoxy equivalent etc. of the resins were determined as shown in Table 2. It is observed that all three compositions of epoxy resins possess low epoxy equivalent, which may be due to the presence of 3 moles of epichlorohydrin with respect to 1 mole of total hydroxyl groups, present in starch and BPA. However, the resin HBE10 possesses the lowest epoxy equivalent, and therefore, the highest number of epoxy groups present in its structure.

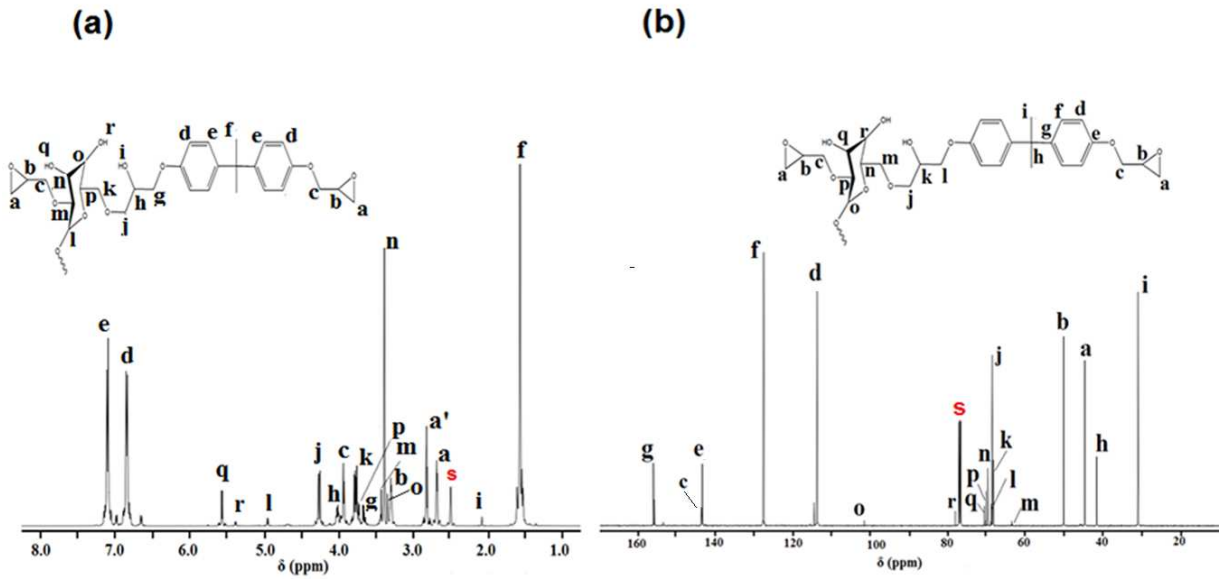


Fig. 3 (a) ^{1}H NMR spectrum of HBSE20 and (b) ^{13}C NMR spectrum of HBSE20.

This may be due to the favorable composition of reactants at 10 wt% of starch, which helps to form the hyperbranched structure with the highest degree of branching (Table 2). Further, from the solubility studies, it has been observed that the resins were soluble in most of the common organic solvents such as dichloromethane, chloroform, acetone, ethanol, DMAc, DMF, and DMSO etc., and partially soluble in methanol, xylene and toluene, whereas insoluble in water. This high solubility of the resins is mainly due to the presence of large numbers of polar functionalities along with flexible ether linkage and globular in structure.[13] However, compared to hyperbranched structure, DGEBA resin exhibited less solubility in different solvents. The gloss of the thermosets was found to be high owing to its highly smooth surface texture and good dimensional stability.

Table 2 Physical properties of HBSE and DGEBA resins and thermosets

Physical property	HBSE5	HBSE10	HBSE20	DGEBA
Hydroxyl value (mg KOH/g)	178.46	125.48	149.93	89.37
Epoxy equivalent (g/equiv)	281.66	206.4	253.35	190.73
Yield (%)	96.32	96.6	95.25	97.12
Gloss (60^0)	104 ± 4	101 ± 4	74 ± 3	71 ± 3
Specific gravity of resins	0.9 ± 0.01	1 ± 0.01	1 ± 0.02	1.05 ± 0.01
Specific gravity of thermosets	1.06 ± 0.01	1.1 ± 0.01	1.3 ± 0.01	1.03 ± 0.01
Degree of branching	0.63	0.80	0.72	-

The specific gravity of the resins varies from 1 to 1.3 with the increase of starch content in the resins which may be due to increase of compactness in the structure, as the number of polar groups increases with the same. However, the values are within the normal range of such resins and thermosets (Table 2).

Curing study

For the evaluation of various performance characteristics of the synthesized HBSE and DGEBA resins, curing was conducted with an appropriate hardener, poly(amido amine). During the curing process, the viscous liquid resins are transformed to solid form by a chemical crosslinking reaction with the hardener, where high strain oxirane rings of the epoxy resin readily open up in the presence of a labile proton of an amine of the hardener. This hardener contains labile amino protons that readily react with epoxy groups at high temperature to form crosslinked three- dimensional network structures, which was monitored by the decrease in the FTIR bands in the region 914–832 cm^{-1}.[30]

Table 3 Curing parameters for HBSE and DGEBA resins

Curing parameter	HBSE5	HBSE10	HBSE20	DGEBA
Touch free time (at room temperature) (h)	4	3	3.5	6
Curing time at 100 °C (min)	85	75	80	85
Post curing time at 120 °C (min)	20	10	15	25
Swelling value at 25 °C	28	22	25	27

The curing time of epoxy resin with the poly(amido amine) hardener depends on the epoxy equivalent of the resin and the time decreases with the decrease in epoxy equivalent, which directly indicates the increase in number of epoxy groups in the structure of resin.[21] Among all three compositions of epoxy resins, HBSE10 took the lowest time to cure at the given temperature, owing to its lowest epoxy equivalent. The state of crosslinking or curing was monitored by measuring the swelling value (Table 3). The curing time is the time required to attain the swelling value of the epoxy thermoset in the range of 20–30% (Table 3). Due to the presence of sufficient crosslinking and its hyperbranched structure, HBSE took a lesser curing time than DGEBA (Table 3).

Mechanical properties

The mechanical properties of the three HBSE thermosets and their linear analog were determined (Table 4). The tensile strength of the thermoset increases with the increase in aromatic moiety and number of epoxy groups which provides optimum level of crosslinking in the structure. HBSE10 thermoset has the highest tensile strength as it has the lowest epoxy equivalent leading to sufficient crosslinking as evident from its lowest swelling value (Table 3) and highest degree of branching (Table 2). Although, DGEBA thermoset exhibited sufficient tensile strength, but possessed low elongation at break due to its brittle nature, as it contains only an aromatic rigid moiety as a major constituent in its structure.[21] Elongation at break of the thermosets decreased as the percentages of aromatic moiety and starch content (which contains amylose and amylopectin moieties) were increased.

Table 4 Mechanical properties of thermosets

Property	Thermoset			
	HBSE5	HBSE10	HBSE20	DGEBA
Tensile strength (MPa)	24 ± 0.5	29 ± 0.4	26 ± 0.5	62 ± 0.4
Elongation at break (%)	28 ± 1.0	36 ± 2.0	38 ± 1.0	7.5 ± 0.6
Toughness[a] (MJ m^{-3})	5.60 ± 10	8.40 ± 12	8.28 ± 15	2.30 ± 8
Impact resistance (m)[b]	>1	>1	>1	6.5 ± 5
Scratch resistance (kg)[c]	>10	>10	>10	7.0 ± 0.4
Adhesive strength (MPa) (ply-wood)[d]	1984 ± 5	1837 ± 4	1337 ± 5	863 ± 5
Adhesive strength (MPa) (aluminium)	1086 ± 6	2906 ± 4	1337 ± 4	598 ± 5

[a]Calculated by integrating the area under stress–strain curves, [b]Limit of the instrument for impact strength was 100 cm (highest), [c]Limit of the instrument for scratch hardness was 10.0 kg (highest), [d]In all the cases the used plywood substrate failed.

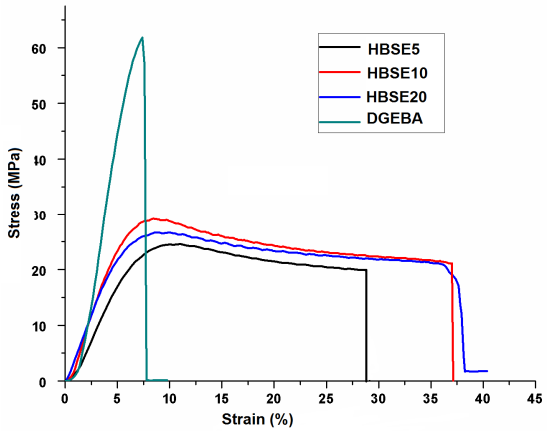


Fig. 4 Toughness of thermosets (obtained by integrating stress-strain curves).

For the same reason, HBSE5 exhibited the lowest elongation whereas HBSE20, the highest elongation at break. It is noteworthy to mention that HBSE10 thermoset showed the highest toughness as measured by integrating the area under the stress-strain curve of it (Fig. 4 and Table 4). This may be due to the combined effect of high strength and elongation of the thermoset. The impact resistance was found to be high in all the hyperbranched epoxy thermosets (impact resistance was above 1 m as that is the maximum limit of the instrument). However, DGEBA thermosets exhibited comparatively low scratch hardness and impact resistance because of its brittleness character. The adhesive strength of the epoxy was evaluated for two different substrates viz. ply-wood and aluminum sheets. However, in all three cases of HBSE5, HBSE10 and HBSE20, there was substrate failure of the cellusoic wood substrates, which indicates that their adhesive strengths were higher than the tensile strength of the wood. This is attributed to the strong interactions of the polar hydroxyl, ether and epoxy groups along with the branched structure with the terminal epoxide groups and the polar linkages of the poly(amido amine) hardener, with the cellusoic wood substrates.[22] Further, the resins also showed high adhesive strength with the metal substrates because of their hyperbranched structures and low viscosity which facilitate the physical interlocking with the metal substrates.[13] In case of DGEBA, the adhesive strength for both the substrates was less than the hyperbranched resins. This is attributed to the lesser number of polar functionalities like hydroxyl groups and ether groups in its linear structure.

Thermal properties

The thermograms and derivatives of the thermograms of the hyperbranched and DGEBA thermosets are shown in Fig. 5a and Fig. 5b. The initial degradation temperature, peak temperatures for first and second stages of degradation and char residue at 700 °C are given in Table 5. In all the cases it was observed that the epoxy thermosets exhibited two step degradation patterns, which can be attributed to the presence of both aliphatic and aromatic moieties in the structure of the thermosets.

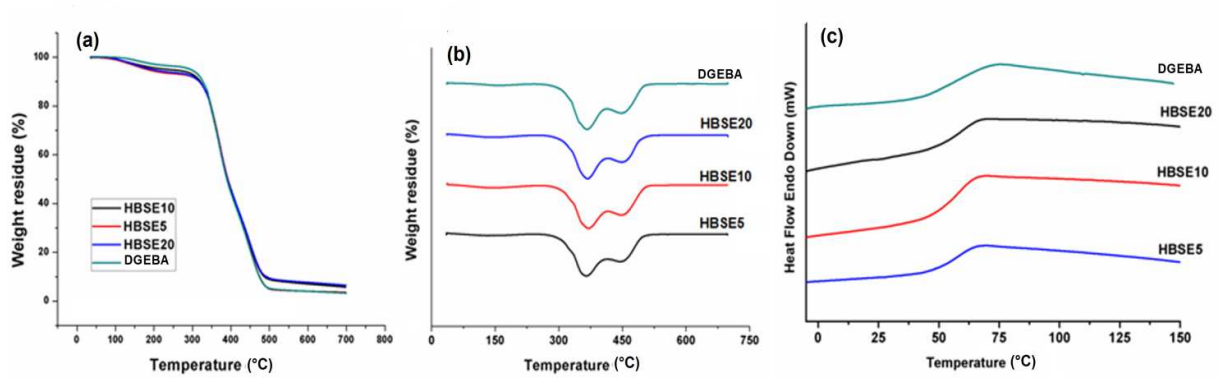


Fig. 5(a) TGA thermograms of thermosets (b) DTG curves of thermosets (c) Glass transition temperatures of thermosets.

A 3-5 wt% of initial loss is due to the loss of volatiles, including water, present in the thermosets as they contain many polar groups. The actual initial degradation temperature varies from 262-275 °C as observed from DTG for the thermosets (Table 5). The variations of degradation temperature for the thermosets are according to their epoxy equivalent values (Table 1), which also indicated the adequate degree of crosslink of the thermosets. The peak temperature for second step of degradation ranges from 448 to 454 °C due to the degradation of the thermally stable aromatic moiety and amide linkages in the epoxy thermoset.[11]

Table 5 Thermal degradation parameters and glass transition temperatures of thermosets

Parameter	HBSE5	HBSE10	HBSE20	DGEBA
Onset temperature (°C)	262	275	266	287
Peak temperature for 1st stage degradation (°C)	363	373	367	374
Peak temperature for 2nd stage degradation (°C)	448	451	454	445
Char residue (%)	5.6	3.59	6.39	3.35
Glass transition temperature (°C)	64.54	65.15	66.75	71.69

However, an overlap of degradation steps was observed which may be due to the fact that during the degradation of aliphatic moieties, especially starch based ones, are transformed to thermostable aromatic moieties through decarboxylation followed by aromatization. This is indirectly supported by the observed higher thermal stability of the thermosets with higher amount of starch, even though it has lower amount of aromatic moiety based bisphenol-A. The thermosets also exhibited char residue of 3.35 - 6.39% due to the formation of carbonaceous products. The glass transition temperatures (T_g) of the thermosets were measured with the help of DSC given in Table 5, which indicate that the HBSE thermosets possess good dimensional stability. The DSC curves indicate that the T_g of HBSE thermosets are lower than that of DGEBA thermoset (Fig. 5c). This can be attributed to the nature of hyperbranched structure in HBSE thermosets and the brittleness character of DGEBA thermosets.[11] Further, the T_g value increases with an increase in wt.% of starch in the HBSE thermoset. This is due to the fact that starch exhibits hydrophilic properties which in turn lead to a large number of intramolecular interactions and strong intermolecular associations via hydrogen bonding formed by the hydroxyl groups in the structure.[7]

Table 6 Change in weight (%) of the thermosets in different chemical media

Chemical Medium	HBSE5	HBSE10	HBSE20	DGEBA
10% aq HCl	0.0015	0.0013	0.0017	0.0025
5% aq NaOH	0.0013	0.0011	0.0014	0.0031
15% aq NaCl	0.0012	0.0015	0.0011	0.0024
20% aq EtOH	0.0031	0.0025	0.0038	0.0041
Water	0.0011	0.0013	0.0012	0.0026

Chemical resistance

Chemical resistance is an essential attribute of epoxy thermosets. The starch based hyperbranched epoxy as well as the DGEBA thermosets exhibited very good chemical resistance in almost all the chemical environments as given in Table 6. The presence of aromatic moiety and adequate crosslinking density favor the exceptional chemical resistance of the epoxy thermoset films.[11] However, relatively better chemical resistance was observed for the hyperbranched thermosets compared to their linear analog which may be due to the presence of strong intra- and inter-molecular secondary interactions such as polar-polar and H-bonding, and the fact that the former has better crosslinking and unique structural architecture than the latter.[31]

Hemolytic assay

Some of the major requirements of sustainable material are protection of human health and prevention of permanent damage to the environment, during its life cycle.

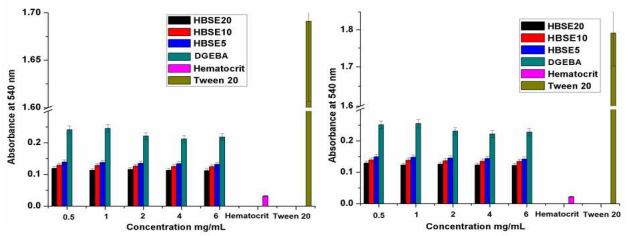


Fig. 6 (a) Anti- haemolytic activity of HBSE and DGEBA thermosets (b) Anti- haemolytic activity of HBSE and DGEBA resins.

Thus, red blood cell (RBC) hemolysis protection assay was performed as a biocompatibility representative tool on the synthesized HBSE and DGEBA, to explore material safety. The hemocompatibility of both the resins and thermosets of HBSE and DGEBA were evaluated by detecting the hemolyzation of erythrocytes by their direct interaction with the erythrocyte membrane. The toxicity of HBSE and DGEBA thermosets and resins was evaluated by extent of disruption of the erythrocyte membrane.[2] The RBC protection assay showed that HBSE5, HBSE10 and HBSE20 resins and their thermosets possessed good compatibility with the erythrocytes. However, lysis of the RBC membrane was observed to be the highest in case of DGEBA, and the lowest in case of HBSE20. Again for both resin and thermoset of HBSE20, the difference is very minute with the hematocrit as the percentage of biocompatible component, starch is high. Tween 20, the negative control displayed drastic destruction of the RBC membrane against the positive control haematocrit, as evident from the comparatively large value of hemoglobin absorbance. Hematocrit exhibited negligible absorbance value. Thus, in-vitro hemolytic assay confirmed the cytocompatibility of the tested epoxy with mammalian RBC (Fig. 6).

Biodegradation study

From the biodegradation study of HBSE and DGEBA thermoset films, it was established that they were gradually degraded on exposure time by *P. aeruginosa* and *B. subtilus* bacterial strains (Fig. 7). The differences in the cell wall structure of the gram negative bacteria, *P. aeruginosa* and gram positive bacteria *B. subtilus* act as an active barrier which is supposed to control the degradation of the polymer substrate.[32] It was found that the epoxy thermosets were degraded to a considerable amount after 6 weeks of inoculations, by the bacterial strains (Fig 8). This signifies that the polymer (carbon source) acted as catabolite to the bacteria. The growth of both the bacterial strains was found to be the highest for HBSE20, as it contains the highest amount of starch in its structure, and thus provides a better susceptible surface for bacterial growth, as evident from the OD curves (Fig. 7a and Fig. 7c). The weight percentages of weight loss of the epoxy thermosets were found after 6 weeks of exposure to the bacterial strains which showed reduction in weight of the thermosets.

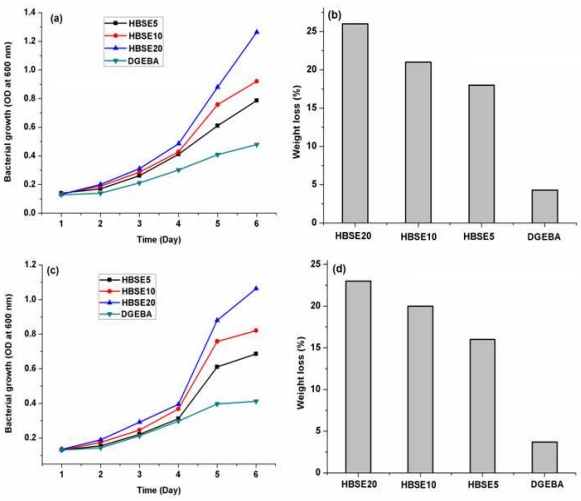


Fig. 7 (a) Variation of *P. aeruginosa* bacterial growth against exposure time for the thermosets (b) Weight loss of the thermosets after 60 days of exposure to the *P. aeruginosa* bacterial strain (c) Variation of *B. subtilus* bacterial growth against exposure time for the thermosets (d) Weight loss of the thermosets after 60 days of exposure to the *B. subtilus* bacterial strain.

This is due to dissolving or degradation of starch by microorganism attack.[8] However, *P. aeruginosa* bacterial strain exhibited comparatively higher degradation rate compared to the *B. subtilus* bacterial strain, owing to the fact that the former possesses higher biosurfactant activity and cell surface hydrophobicity (CSH) than the latter bacterial strain (Fig. 8).[33] Hence, a faster colonization and growth of the former bacterial strain on the polymeric surface was observed than the latter bacterial strain. This further contributes in biodegradation of the starch based hyperbranched epoxy thermosets. Also, as HBSE20 thermoset consisting of the highest amount of starch in its structure, the growth of the bacteria was found to be the highest in it. About 26 wt. % and 23 wt. % of weight losses were found after 6 weeks of their exposure to the *P. aeruginosa* and *B. subtilus* bacterial strains, respectively (Fig. 7b and Fig. 7d). On the other hand, DGEBA thermoset was found to be biodegradable to a very small extent, owing to the presence of the non-biodegradable DGEBA moiety as the major constituent in its structure. The minor amount of biodegradation of it is due to presence of bio-based poly(amido-amine) hardener in the thermoset.[21] The SEM images (Fig. 8a and Fig. 8b) of the biodegraded HBSE20 thermoset films revealed significant surface erosion and bacterial adherence as compared to the control (Fig. 8e) which further confirmed the bacterial biodegradation. The surface of DGEBA thermoset films also showed slight surface erosion (Fig. 8c and Fig. 8d) as compared to the control film of DGEBA (Fig. 8f). Thus from the biodegradation study it could be concluded that the rate of biodegradation of all three hyperbranched epoxy thermosets depends upon the starch content.

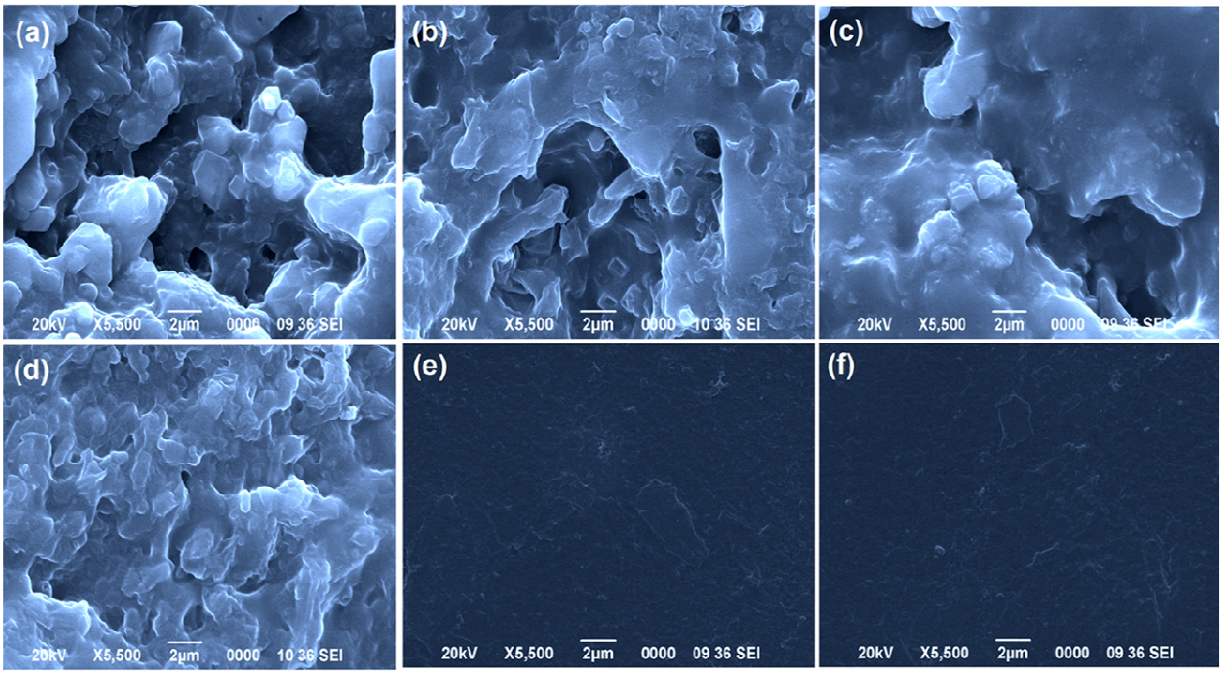


Fig. 8 SEM images of biodegraded (a) HBSE20 by *P. aeruginosa* (b) HBSE20 by *B. subtilus* (c) LE by *B. subtilus* (d) LE by *P. aeruginosa* and respective controls of (e) HBSE20, and (f) DGEBA thermoset.

Conclusion

A novel bio-based biodegradable epoxy thermoset was developed using starch as one of the prime reactants. Poly(amido amine) cured thermoset exhibited high toughness, high adhesive strength and high thermostability (up to 275 °C), which solved problems like brittleness and non biodegradability of conventional epoxy. This thermoset also showed excellent chemical resistance and cytocompatibility with erythrocytes. Thus, superior adhesive strength, toughness, flexibility, chemical resistance and biodegradability of the thermoset, over DGEBA may offer a suitable position to the former in advanced engineering and biomedical fields.

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