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Oxidation Assisted Fabrication of Sterculia Gum/Protein Hybrid Hydrogel Networks through Schiff Base Formation: Characterization and Swelling Kinetic Studies

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Polysaccharide/protein hybrid conjugate system becomes an emerging system with integrated characteristics of protein as well as polysaccharide to be exploited for recent advancements in biomedical sectors. Present study is an attempt to fabricate a sterculia gum/gelatin hybrid hydrophilic network system *via* Schiff base formation using oxidative route at ambient conditions. This route transforms hydrogel synthesis through green mode without employing any crosslinking systems so as to minimize the toxicity issues associated. Fabricated Schiff base based gel systems have been characterized by FT-IR, powdered XRD, FESEM and EDX to confirm the inclusion of new characteristics, morphological changes and functionalization. Oxidation of natural gum drastically alter the physico-chemical behaviour of the gum as confirmed by powdered XRD by incorporating crystalline nature of oxidized sterculia gum as compared to the native sterculia gum and the Schiff base formed. The changes observed are due to the chemical modification of sterculia gum during Schiff base formation. Further the swelling capacity of oxidized sterculia gum and crosslinked network formed is also modulated and was less as compared to native sterculia gum and gelatin. This article also elaborates the mechanistic changes which take place during oxidative route of hydrogel formation in various segments of sterculia gum during oxidation and Schiff base formation.

Keywords: Sterculia gum, Functionalization, Oxidation, Schiff's base, Swelling kinetics.

INTRODUCTION

Sterculia gum a heteropolysaccharide exudate gum derived from the bark of *Sterculia urens* is partially acetylated polysaccharide with rhamnose, galactose, galacturonic acid, glucuronic acid, present in different proportions [1-4]. Sterculia gum in its native form has food and medicinal applications [5-9]. Poor solubility and high swelling capacity of sterculia gum restricts its direct applicability, which could be removed or modulated by chemical functionalization. One major concern, which must be considered while functionalization is that the bio-compatibility and non-toxicity of native gum must be either improved or not compromised at all. The hydroxyl, carboxyl and acetyl groups present on the backbone are responsible for its characteristic properties as well as act as active centre's for its modification. From time to time, various strategies adopted for functionalization includes crosslinking, grafting, esterification, etherification and sequential mixing of any two or more functionalized components. Modification/functionalization depends upon reaction parameters and method used. Sterculia gum crosslinked with PVA-PVP (polyvinyl alcohol and polyvinyl pyrrolidine) improve thermal stability and modulate swelling behaviour and is influenced by PVA and PVP concentrations [10]. Barium ions cross-linked sterculia gum and alginate beads have different swelling behaviour as compared to the parent gum [11]. Esterification of sterculia gum with thioglycolic acid leads to improve mucoadhesiveness, increased crystallinity and stability of functionalized system [12]. Esterification of sterculia gum with dodecenylsuccinic anhydride incorporates antibacterial properties and improves porosity in the gel network structures [13]. Various pH sensitive sterculia gum based hydrogels networks have been synthesized through graft copolymerization using ammonium persulphate as an initiator and N′N′-methylenebisacrylamide as cross-linking agent with vinylic monomers [14-18]. Graft copolymerization although is one of the most adopted and system

friendly strategy but sometime is associated with some toxic crosslinking and initiating systems. Thus, there is a need to integrate different functionalization strategies while minimizing such drawbacks.

Oxidation is one of the modification strategy to incorporate functionalities on natural gums for further modification. There are various oxidizing reagents like hydrogen peroxide, sodium hypochlorite, chlorine dioxide, ozone, and sodium periodate used in literature for oxidation of polysaccharides [19-23]. The oxidation reaction is influenced by various factors such as pH, temperature, type, and oxidant concentration of the oxidant. Sodium periodate is the most useful and attractive reagent due to its highly selective nature, as it brings about selective oxidation of *vicinal* hydroxyl groups to the carbonyl groups. *vic*-Groups on the polysaccharide backbone are most exploited centers for suitable functionalization by many researchers. In literature, various types of polysaccharides have been functionalized through this route [24]. This route is used to study structural composition, and substitution pattern, physico-chemical properties, swelling behavior as well as mean for the synthesis of nanoparticles [25-30]. Oxidized pectin acts as a reducing system for the formation of silver nanoparticles, which attributes to aldehyde groups present in the oxidized pectin [30].

In general, a single chemical modification pathway is incapable of introducing all the desired changes into the polymeric network. Such changes can be introduced by adopting sequential strategies for the introduction of different functionalities on the polysaccharide backbone. The modification of polysaccharide through oxidation and subsequent crosslinking with suitable functionalities mainly with free amino groups containing moieties such as chitosan [31,32], carboxymethyl chitosan [33], urea [34], sulpha drugs [35], hydrazine [36], collagen [37] and gelatin through Schiff base formation can result in the formation of hydrophilic networks. Such stepwise fabrication either uses minimum or no chemical crosslinkers and gel formation took place at room temperature.

Functionalization of natural gums with proteins/amine-containing drugs paves a way toward biocompatibility with ease as they are free from any toxicity induced by some crosslinkers. Such hydrogel formed has physical behaviour similar to that of some biological tissues and also increased its stability in water, which made them useful for biomedical and environmental applications [37,38]. Oxidized dextran act as a crosslinker during the fabrication of chitosan mesoporous scaffold [31,32]. A wound dressing hydrogel was prepared by crosslinking periodate oxidized pectin with carboxymethyl chitosan [33]. The periodate oxidized starch crosslinked with urea result in a gel network used for the adsorption of heavy metal ions [34].

Gelatin obtained by the hydrolysis of collagen has food applications where it is used as a thickening and gelling agent [39]. Due to the presence of free -NH_2 groups, gelatin could be used as a crosslinking center for the synthesis of polysaccharide gel networks. A self healing hydrogel was prepared in the presence of borax using periodate oxidized alginate and gelatin [40]. A biodegradable extracellular matrix formed by borate assisted Schiff base formation between oxidized carboxymethyl cellulose and gelatin [41]. A nanogel of gelatin and oxidized gum arabic prepared through inverse miniemulsion technique has been used in drug delivery [42]. Oxidized corn starch/gelatin led to the formation of biodegradable active films [43]. Hydrogels have also been formed by Schiff base reaction between periodate oxidized pectin and gelatin [44,45]. Although there are many literature reports for functionalization of sterculia gum by various methods earlier, the oxidative route of modification is least explored. The present study is an attempt to functionalize sterculia gum through oxidative route and subsequent gelation with gelatin through Schiff base reaction. Chemical functionalities introduced in the system have been characterized through FTIR, XRD, FESEM, EDX, and swelling studies. The absence of chemical crosslinker and the aqueous environment make this method economically and ecologically attractive. The present article also discussed the mechanistic details of periodate oxidation of sterculia gum and its Schiff base formation with gelatin. Swelling studies have been carried out as a function of reaction parameters as well as function of pH. Swelling kinetics has been evaluated to interpret various swelling parameters of hydrophilic systems such as diffusion exponent, gel characteristic constant, and diffusion coefficients.

EXPERIMENTAL

Sterculia gum was purchased from the local herbal store. Ethylene glycol was obtained from CDH Pvt. Ltd., New Delhi, India. Sodium periodate and gelatin were obtained from Nice Chemicals Pvt. Ltd., Kochi, India. Ethanol was obtained from Merck Chemicals and Labware Pvt. Ltd., India. Double distilled water was used in the preparation of all solutions. Potassium chloride, sodium hydroxide pellets, methyl orange, and hydroxylamine hydrochloride were purchased from Merck specialties Pvt. Ltd. Buffer solutions of pH (1.2, 1.4, 2.2, 6.8. 7.2, and 7.4) were prepared as reported in the Pharmacopoeia of India [46]. All the chemicals were of analytical grade and used as such without further purification.

Synthesis of oxidized sterculia gum (OSG): Sterculia gum has been oxidized using sodium periodate by following a standard procedure [47]. Sterculia gum in variable proportions (2 g and 3 g) has been suspended in the double-distilled water (100 mL) with continuous stirring at room temperature for 30 min to obtain 2% and 3% concentration (w/v) suspensions. To each suspension, 25 mL of $NaIO_4$ (0.052 M) and (0.078 M) was added separately with continuous stirring at room temperature under dark conditions for 20 h. After 20 h, the oxidation process was seized by the addition of ethylene glycol (5 mL). The oxidized product was precipitated out by adding an excess of ethanol (approx. 500 mL). The oxidized product was filtered and washed with double distilled water three to four times to remove any unreacted sodium periodate. Oxidized product was dried to constant weight at room temperature. Samples were preserved in dry conditions in vacuum desiccators for further use.

Synthesis of sterculia gum/protein hybrid hydrogel networks through Schiff base formation: Sterculia gum/protein hybrid gel networks through Schiff base formation

between oxidized sterculia gum and gelatin have been synthesized using the method prescribed in literature with minor modifications [44]. Oxidized sterculia gum (0.5 g) was suspended in 25 mL of double distilled water and the reaction mixture was stirred on a magnetic stirrer at room temperature for 30 min under dark conditions to obtain a 2% (w/v) concentrated solution of OSG. A 10% (w/v) clear solution of gelatin was obtained by dissolving 2.5 g of gelatin in 25 mL of double distilled water and then stirred it on a magnetic stirrer at 55 ºC for 30 min. The equal amount of the two solutions (2% OSG and 10% gelatin) were mixed and then stirred on the magnetic stirrer at 45 ºC for 6 h to homogenize the two solutions. The homogenized mixture solution was then transferred to the petri dish, where gelation took place on cooling within 5 min. The yield and dimensions of the polymerized network were recorded. The synthesized gel networks were kept in a zero humidity environment for further use.

Determination of aldehyde content: Aldehyde content in oxidized sterculia gum was determined by the titrimetric method [48]. Oxidized sterculia gum (0.1 g) was dissolved in 25 mL of 0.25 M hydroxylamine hydrochloride. The reaction mixture was covered with aluminium foil and stirred at room temperature for 1 h. The reaction of aldehyde groups present in the oxidized sterculia gum with hydroxylamine hydrochloride released hydrochloric acid. The released hydrochloride was titrated against 0.1 M, NaOH using methyl orange as an indicator. Light pink colour changed to pale yellow at the endpoint of the titration. The number of mole of aldehyde present in the oxidized sterculia gum is equivalent to the number of moles of NaOH consumed during the titration.

Characterization: Sterculia gum (SG), gelatin (G), oxidized Sterculia gum (OSG) and Schiff base sterculia gum-gelatin network (Schiff OSG-*cl*-G) were characterized by FT-IR powdered X-ray diffraction method, FESEM, and EDX. FTIR spectra were recorded on Perkin Elmer spectrum RX-IFTIR spectrophotometer using KBr pellets. Panalytical's X'Pert Pro X-ray diffractometer (powder method) was used to study the X-ray diffraction pattern. FESEM and EDX were recorded using SU8010 FESEM (Hitachi).

Swelling study: Swelling studies of native Sterculia gum (SG), gelatin (G), oxidized Sterculia gum (OSG), and Schiff base sterculia gum-gelatin network (Schiff OSG-*cl*-G) have been carried in distilled water in triplicate by gravimetric method [49]. The polymers of known weight were taken and filled in tea bags and then immersed in an excess amount of solvent (distilled water and buffer solutions of different pH (1.2, 1.4, 2.2, 6.8, 7.2 and 7.4) for different time intervals at 37 ºC. Samples were taken out at regular intervals of time and weighed after removing the extra surface water by drying using laboratory tissue paper. Swelling is noted for upto 24 h to get equilibrium swelling. The degree of swelling or percentage swelling (%) was evaluated by using the following formula:

$$\text{Swelling ratio, Q (water uptake per g of gel)} = \frac{W_t - W_o}{W_o} \text{ g/L} \quad (1)$$

where, W_t and W_o are the weights of swollen and dry hydrogels expressed in grams, respectively.

$$\text{Swelling (\%)} = \frac{W_t - W_o}{W_o} \quad (2)$$

Swelling study was carried out as a function of monomer and oxidant concentration during the hydrogel synthesis. Swelling kinetics has been evaluated using eqn. 3 [50]:

$$F = \frac{W_t}{W_\infty} = kt^n \quad (3)$$

where, W_∞ is the weight of the hydrogel at equilibrium swelling, k is the diffusion constant and n is the diffusion exponent. By taking natural log of eqn. 3 we get:

$$\ln (F) = \ln (k) + n \ln(t) \quad (4)$$

Plot of ln (F) against ln (t) gives value of n and k from the slope and intercept. Time was expressed in minutes. The initial diffusion coefficient (D_i), the average diffusion coefficient (D_A) and late diffusion coefficient has been calculated using eqns. 5, 6 and 7 [50].

$$\frac{W_t}{W_\infty} = 4\left(\frac{D_i t}{\pi l^2}\right)^{0.5} \quad (5)$$

$$D_A = \frac{0.49 l^2}{t^{1/2}} \quad (6)$$

$$\frac{W_t}{W_\infty} = 1 - \left(\frac{8}{\pi^2}\right)\exp\left[\frac{(-t\pi^2 D_L}{l^2}\right] \quad (7)$$

RESULTS AND DISCUSSION

Fabrication of sterculia gum/protein hybrid hydrogel networks through Schiff base formation: Sodium periodate is a very selective oxidizing agent. This selective behaviour is beneficial for the oxidation of polyhydroxyl systems (polysaccharides). $NaIO_4$ selectively brings about oxidation of *vic*-hydroxyl groups to the carbonyl groups. Singh & Sharma [51] have elaborated structural fragments of sterculia gum as composed of three segments. The segment (A) contains β-D-galactose and α-L rhamnose residue linked by (1,4β)glycosidic bond. Seg-ment (B) contain α-D-galacturonic acid and β-D-glucoronic residue having (1,3β)-glycosidic linkage and the segment (C) consists of (1,2α) linked α-D-galacturonic acid and α-L rhamnose residue units. The presence of hydroxyl and carboxylic groups on these residues acted as active centers for functionalization. Hydroxyl groups are present at position 2-, 3- (*vic*-) and 6-C in β-D-galactose unit and at position number 2-, 3- and 4-C in β-D-glucoronic unit of which (2-, 3-) and (3-, 4-) are *vic*- hydroxyl groups. In α-L rhamnose, the two hydroxyl groups present at 3- and 4-C were in the *vicinal* position, which present only one free hydroxyl group in α-D-galacturonic acid. The sodium periodate brings about cleavage of these *vicinal* hydroxyl groups present on the backbone of the sterculia gum. Cleavage of C2-C3 bond of β-D-galactose unit, C2- C3 and C3-C4β-D-glucoronic unit and C3-C4 of α-L rhamnose units of sterculia gum took place in the presence of sodium periodate and the carbonyl groups are generated at these positions (Fig. 1a-b). In second step, gelatin reacts with OSG resulting into a

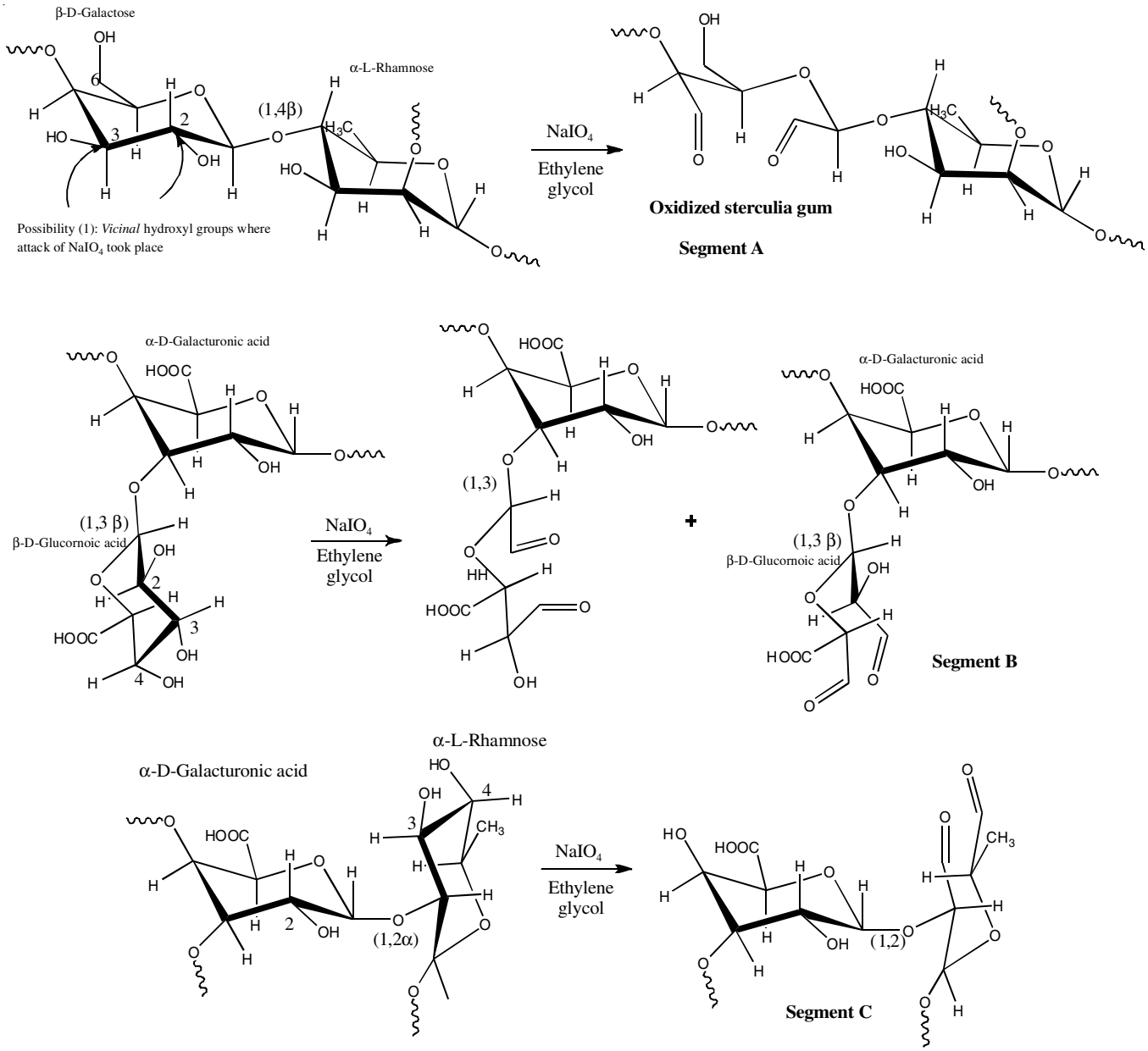

Fig. 1. (a,b) Schematic representation of oxidation of various segments of sterculia gum with $NaIO_4$

three dimensional network structure *via* formation of imine –CH=N- linkage through Schiff base reaction between the carbonyl groups present on the various residues of oxidized sterculia gum and the free amino groups present on the backbone of the gelatin (Fig. 2a-b). This reaction is fast and takes place at ambient temperature.

The aldehyde content in oxidized sterculia gum (2% and 3% sterculia gum oxidized using 0.052 M, $NaIO_4$ and 0.078 M, $NaIO_4$) is 2.5×10^{-2} mol/g, 1.7×10^{-2} mol/g, 1.45×10^{-2} mol/g and 2.0×10^{-2} mol/g, respectively. The maximum aldehyde content for oxidized sterculia gum was observed when 2% sterculia gum was oxidized by 0.052 M $NaIO_4$ and minimum was observed when 3% sterculia gum was oxidized by 0.052 M $NaIO_4$.

FTIR studies: To ascertain the chemical composition & presence of different functionalities on native (SG), gelatin (G), oxidized sterculia gum (OSG) and Schiff base sterculia gum-gelatin network (Schiff OSG-*cl*-G), FT-IR have been observed in the range of 4000-400 cm^{-1} (Fig. 3). In the spectrum of sterculia gum the broad and strong peak around 3500-3200 cm^{-1} due to -OH stretching vibration. This peak was also present in the spectrum of oxidized sterculia gum indicating that free primary hydroxyl group at 6-C of β-D-galactose residue remain unchanged during the oxidation process. The carboxylic groups in the uronic acid residues show peaks in the range of 1730-1600 cm^{-1} in the IR spectra of sterculia gum and oxidized sterculia gum. The C-O stretching give rise to a strong and sharp peak at around 1050 cm^{-1}. We expect a characteristic peak around 1730 cm^{-1} due to the C=O stretching on the oxidized Sterculia gum, but this peak is overlapped by C=O stretching bands of carboxylic and acetyl groups of uronic acid residues. The peak around 819 cm^{-1} in the oxidized sterculia gum is due

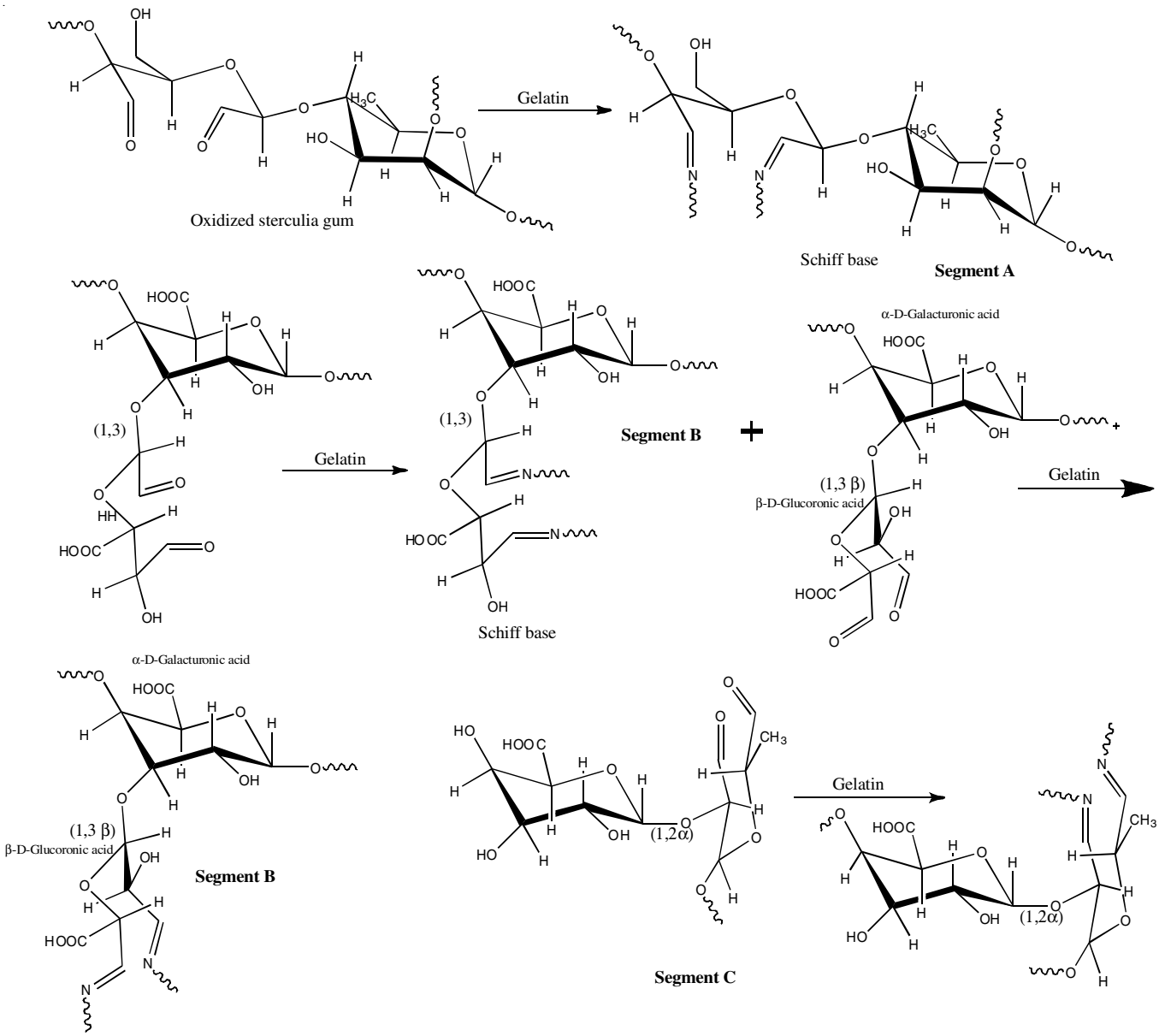

Fig. 2. (a,b) Schematic representation of Schiff base formation as a result of reaction between oxidized sterculia gum and gelatin

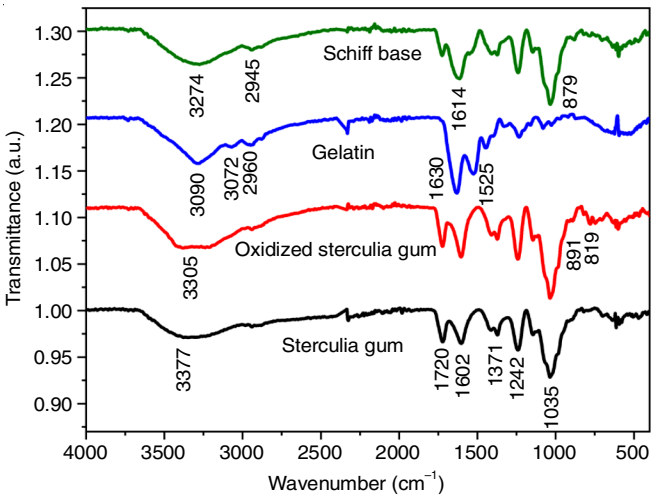

Fig. 3. FTIR of sterculia gum, oxidized sterculia gum, gelatin and Schiff base OSG-*cl*-G

to hemiacetal group because in molecular structure most of the carbonyl groups are in hemiacetal form [25,52,53] and this indicated that sodium periodate oxidized *vicinal* hydroxyl groups to carbonyl groups in the sterculia gum. The FTIR spectra of Schiff base show bands due to both oxidized sterculia gum and gelatin. The absence of strong band around 1720 cm^{-1} confirmed the crosslinking between oxidized sterculia gum and gelatin. The peak due to the –C=N band was expected around 1600-1640 cm^{-1} but this peak was overlapped by the peak due to amide group in gelatin [42,54].

XRD studies: The powdered XRD spectra of sterculia gum, oxidized sterculia gum, gelatin and Schiff base are shown in Fig. 4. The presence of broad peak in the powdered XRD of sterculia gum, gelatin and Schiff base is due to their amorphous nature. The powdered XRD of oxidized sterculia gum was different from that of sterculia gum. The two sharp peaks were present at $2\theta = 21.0297°$ and $2\theta = 27.6449°$ indicates the

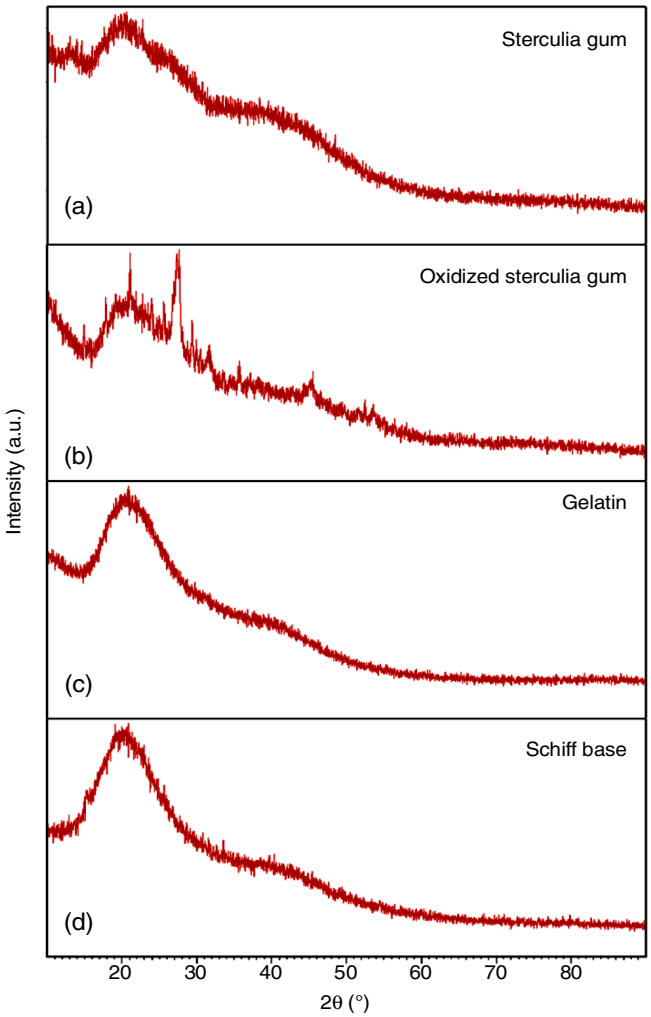

Fig. 4. XRD of (a) sterculia gum, (b) oxidized sterculia gum (OSG), (c) gelatin and (d) Schiff base OSG-*cl*-G

crystalline nature of oxidized sterculia gum. Crystallinity is also confirmed through swelling behaviour since OSG become more soluble as compared to native gum.

SEM-EDX studies: The surface morphology and three dimensional network structure of native (SG), oxidized sterculia gum (OSG) and Schiff base sterculia gum-gelatin network (Schiff OSG-*cl*-G) have been studied through SEM (Fig. 5). The oxidized sterculia gum has layered and smooth structure with smaller pore size as compared to the parent sterculia gum. The rough and denser network structure of the Schiff base indicated the crosslinking between the oxidized sterculia gum and gelatin. The increase in the nitrogen content and decrease in the oxygen content in EDX (Fig. 6) of OSG-*cl*-G as compared to OSG further confirmed the formation of Schiff base involving carbonyl group of OSG and NH_2 groups of gelatin.

Swelling behaviour: Swelling studies is one very crucial properties of hydrophilic networks correlated to their crosslinked structure, which decide the fate/applicability of the gel networks in different fields. Swelling characteristic of the hydrogel is the step to understand the network structure of the gel and its capacity to function as drug delivery carrier.

Swelling of OSG and OSG-*cl*-G as a function pH of the swelling medium: The hydrogels formed under different conditions showed dynamic swelling behaviour. The pH sensitivity of the prepared OSG and OSG-*cl*-G was confirmed by their swelling behaviour toward buffers of different pH (1.2, 1.4, 2.2, 6.8, 7.0, 7.2 and 7.4) values. The equilibrium swelling capacity of hydrogels after 24 h at different pHs is shown in Fig. 7. The swelling capacity of hydrogels depends upon their porous nature. The parent sterculia gum showed maximum swelling under alkaline medium (pH = 6.8, 7.2 and 7.4) with maximum swelling of 608.79% in the distilled water. Under alkaline conditions the acidic groups present on the sterculia gum get deprotonated to O^- and COO^- and leads to electrostatic repulsion between these negatively charged groups, which increased swelling. The crystalline nature of OSG as confirmed by powdered XRD was the reason for the decreased swelling nature of OSG as compared to the parent gum. The free NH_2 groups present in the natural protein, gelatin get protonated to NH_3^+ under acidic conditions and the electrostatic repulsion

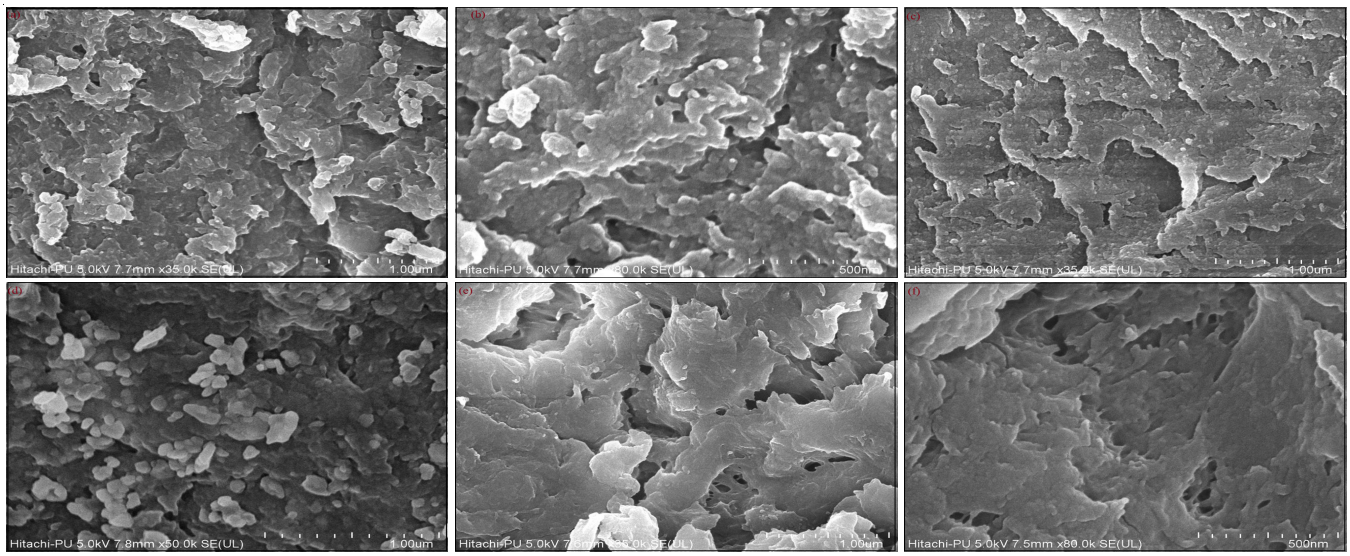

Fig. 5. FESEM of sterculia gum (a,b), OSG (c,d) and OSG-*cl*-G (e,f)

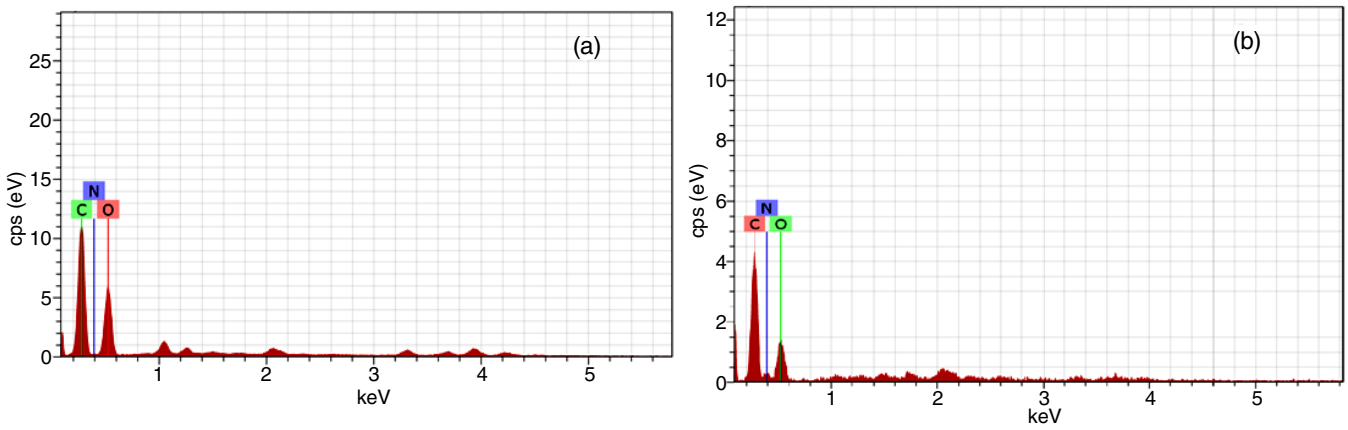

Fig. 6. EDX of (a) oxidized sterculia gum (OSG) and (b) OSG-*cl*-G Schiff base

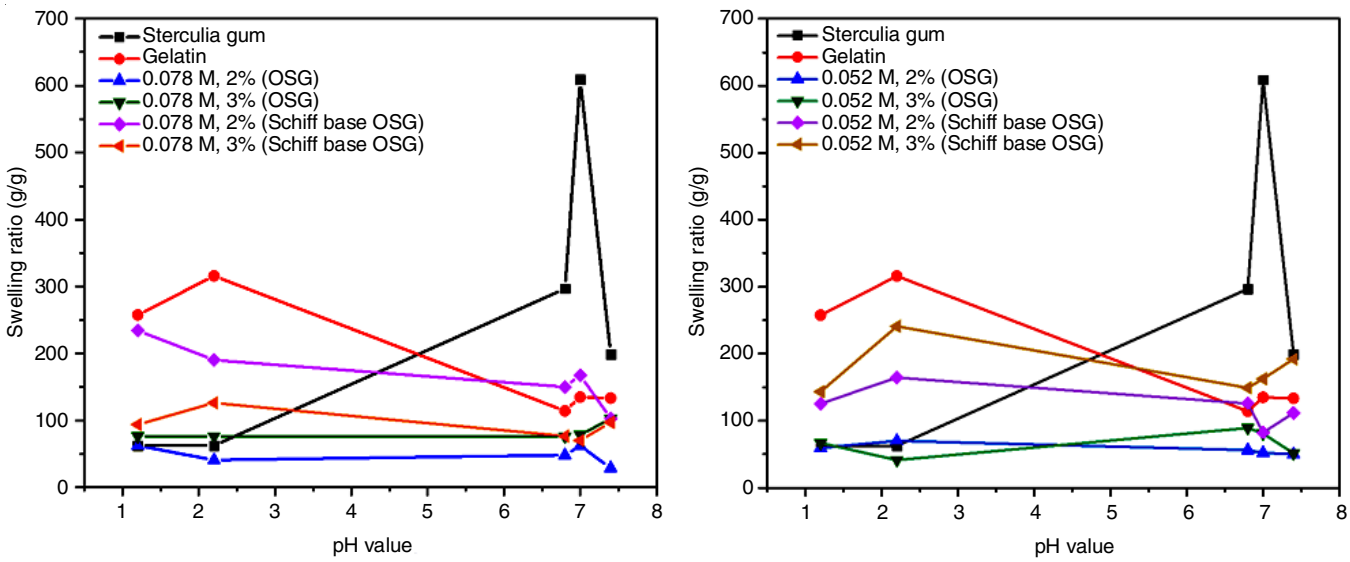

Fig. 7. Effect of pH on the swelling ratio of OSG and OSG-*cl*-G Schiff base

between these positively charged groups increased their swelling under acidic conditions with maximum swelling of 377.78% at pH = 1.4. The crosslinked network formed between the OSG and the natural protein has decreased swelling ratio as compared to the parent protein because crosslinking increased density and decreased porosity. It has maximum swelling of 164.5% at pH = 2.2. The crosslinking between carbonyl groups of OSG and amino groups of natural protein decreased the number of free NH_2 groups in the crosslinked network formed which decreased its swelling ratio as compared to the gelatin under acidic conditions.

Swelling of OSG and OSG-*cl*-G as a function of amount of backbone and concentration of the oxidizing medium: The swelling behaviour of functionalized sterculia gum OSG was affected by the amount of backbone used and by the concentration of the oxidizing medium (Fig. 8a). The concentration of the oxidizing medium strongly affected the extent of oxidation of sterculia gum, which further is directly correlated to swelling behaviour of the oxidized sterculia gum as well as crosslinked network. Swelling capacity of oxidized product declines as compared to native gum. This is attributed to increase in crystalline behaviour of the oxidized sterculia gum, this make it more soluble which leads to decrease in the swelling behaviour of the oxidized sterculia gum. The swelling capacity of the oxidized sterculia gum increased with increase in the amount of sterculia gum used. The increase in the swelling ratio was more under basic conditions due to deprotonation of more number of hydroxyl and carboxylic groups present on the backbone, resulting in more electrostatic repulsion between O^- and COO^-, which increased the swelling. The maximum swelling at pH 7.4 for oxidized sterculia gum was observed when 3% sterculia gum was oxidized by 0.052 M $NaIO_4$ and minimum is observed when 2% sterculia gum was oxidized by 0.078 M $NaIO_4$, due to the less crystalline nature of former than the latter due to the different degree of oxidation by the different concentration of the oxidizing agent.

The crosslinked network formed between the oxidized sterculia gum and natural protein gelatin OSG-*cl*-G has less swelling capacity as compared to the natural protein, gelatin and the sterculia gum (Fig. 8b). The Schiff base formed between free carbonyl centers on oxidized backbone and protein show more swelling under acidic conditions than basic conditions, as under acidic conditions free NH_2 groups present protonated to NH_3^+ and the electrostatic repulsion between

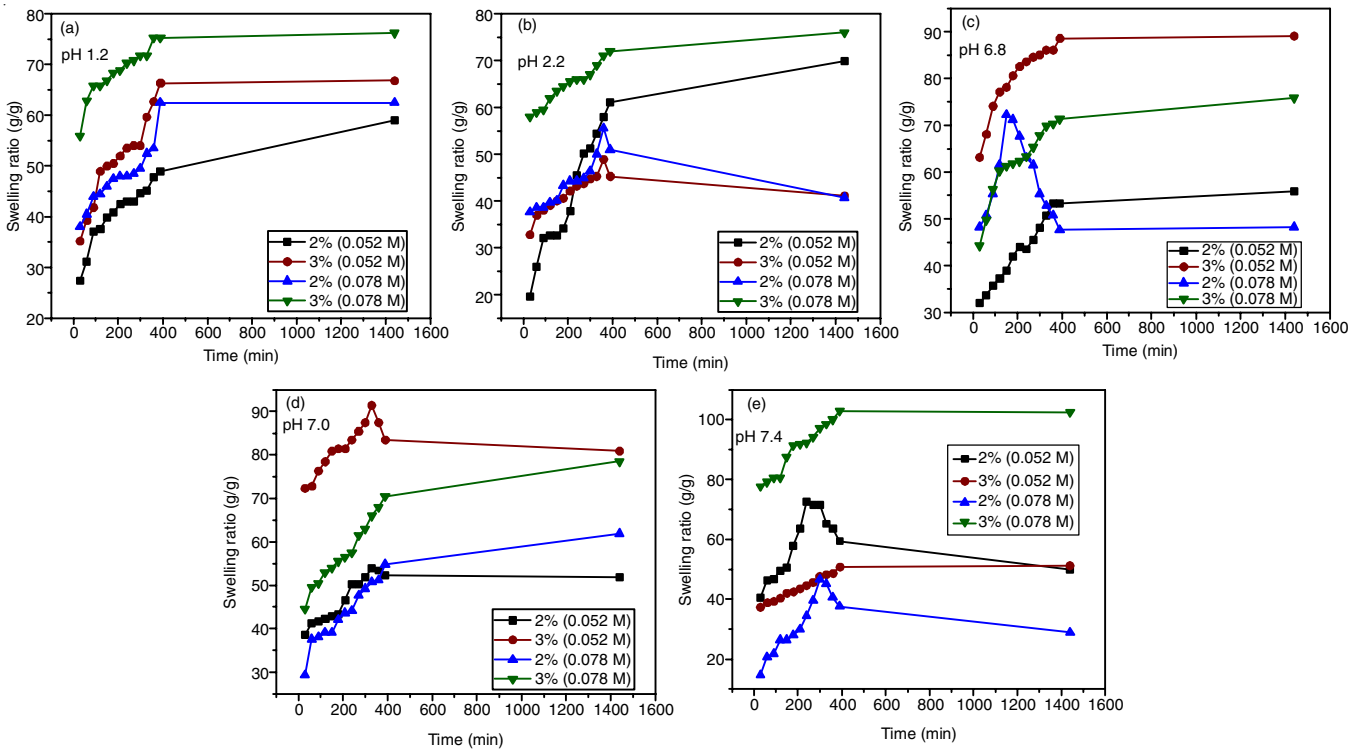

Fig. 8a. Variation of swelling ratio of oxidized sterculia gum (OSG) with the amount of backbone and the concentration of the oxidizing medium

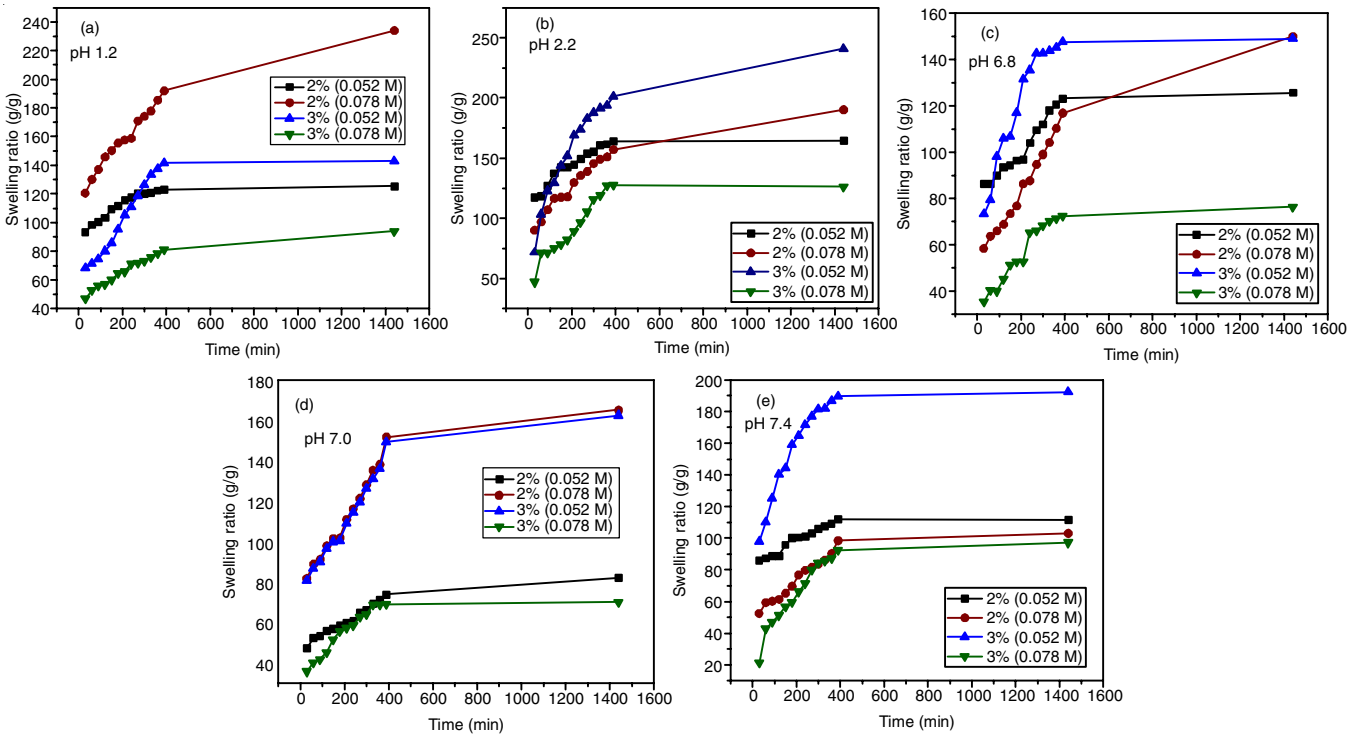

Fig. 8b. Variation of swelling ratio of OSG-*cl*-G Schiff base with the amount of backbone and the concentration of the oxidizing medium

these positively charged groups increased their swelling under acidic conditions with maximum swelling of 164.5% at pH = 2.2. The decrease in the swelling ability of Schiff base as compared to sterculia gum and natural protein gelatin is due to the decrease in the number of free amino groups as a result of crosslinking with the oxidized sterculia gum. The constant variation was observed in the swelling behaviour of OSG-*cl*-G with the amount of sterculia gum and the concentration of the oxidizing medium (Fig. 8b). It has been observed that the swelling capacity of Schiff base get decreased with the increase in the concentration of the oxidizing medium. At pH 2.2, where equilibrium swelling capacity of the Schiff base is maximum

and the minimum was observed when gelatin was crosslinked with 3% sterculia gum oxidized by 0.078 M $NaIO_4$. The minimum was observed due to the fact that the high concentration of oxidizing agent brings about maximum functionalization of sterculia gum, which ultimately leads to maximum crosslinking with the natural protein. The increase in the crosslinking decreased the porosity of hydrogel, which leads to decrease in the water absorption. The higher swelling was observed when the oxidizing medium used has low concentration (0.052 M) and the sterculia gum has high percentage (3%), where the more number of free amino groups protonated to NH_3^+ and the electrostatic repulsion between these positively

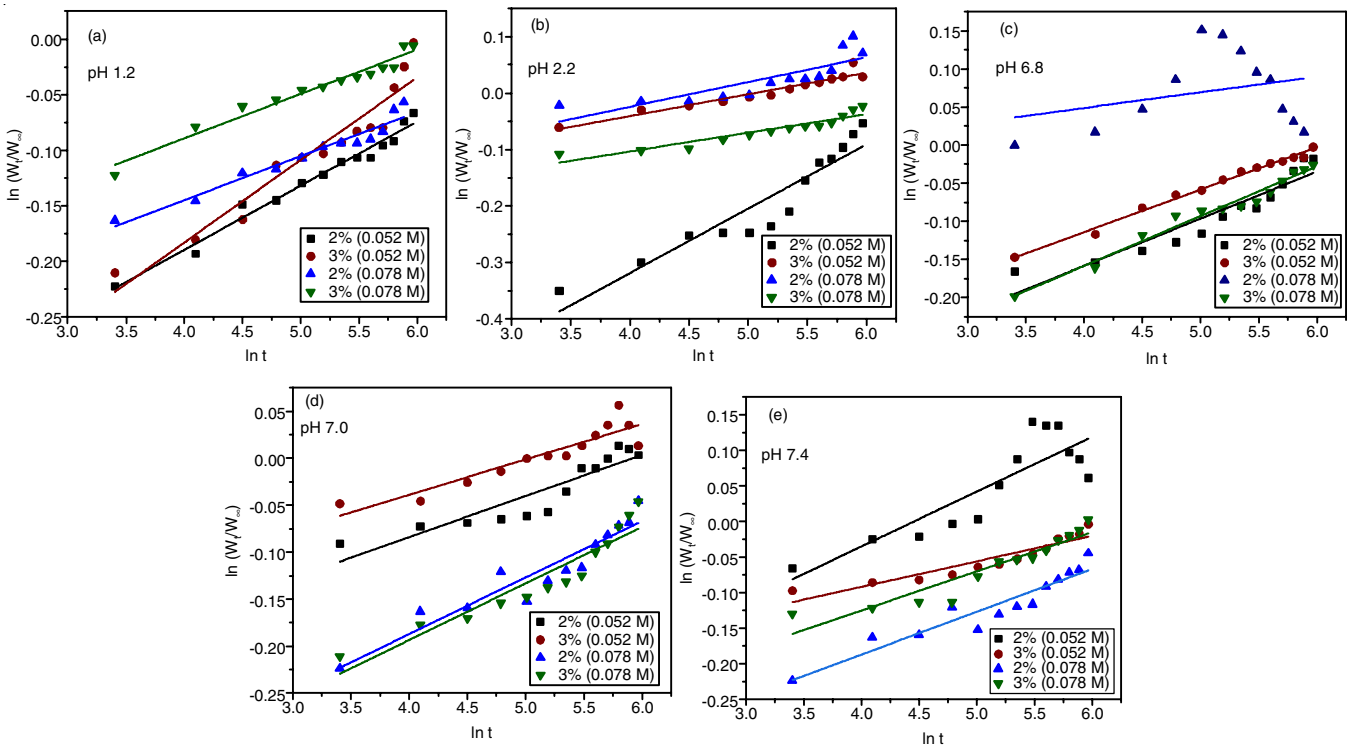

Fig. 9a. Plot of ln (W_t/W_∞) with ln (t) for OSG at different concentration of amount of backbone and the concentration of the oxidizing medium

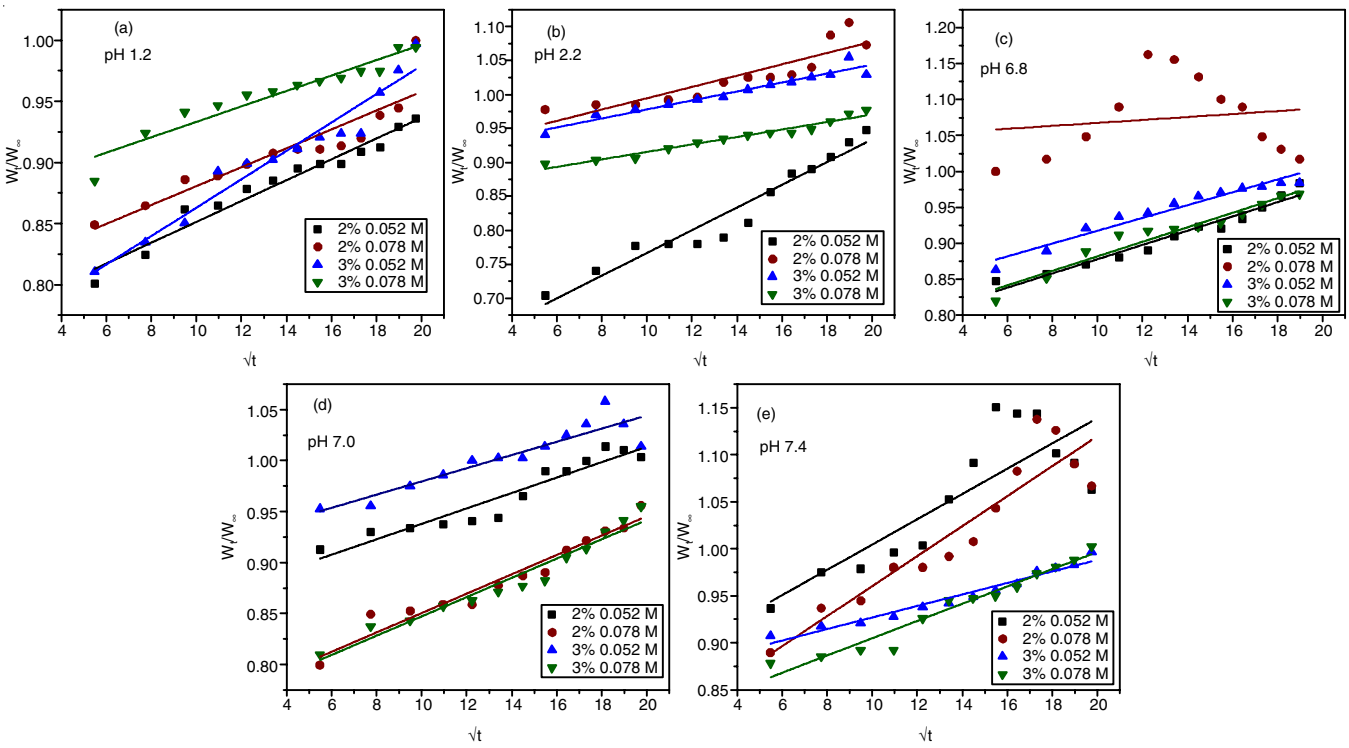

Fig. 9b. Plot of W_t/W_∞ and $\sqrt{t}$ for OSG at different concentration of amount of backbone and the concentration of the oxidizing medium

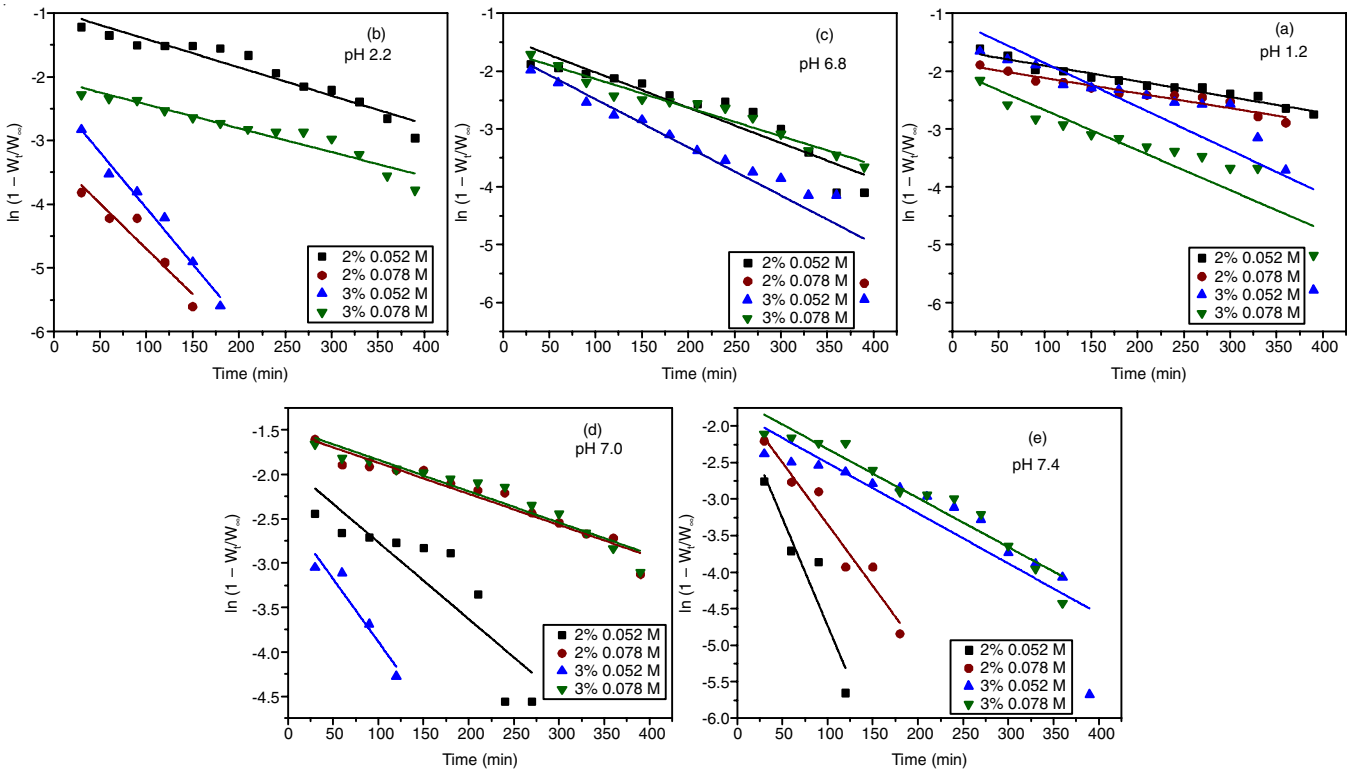

Fig. 9c. Plot of ln $(1 - W_t/W_\infty)$ with t for OSG at different concentration of amount of backbone and the concentration of the oxidizing medium

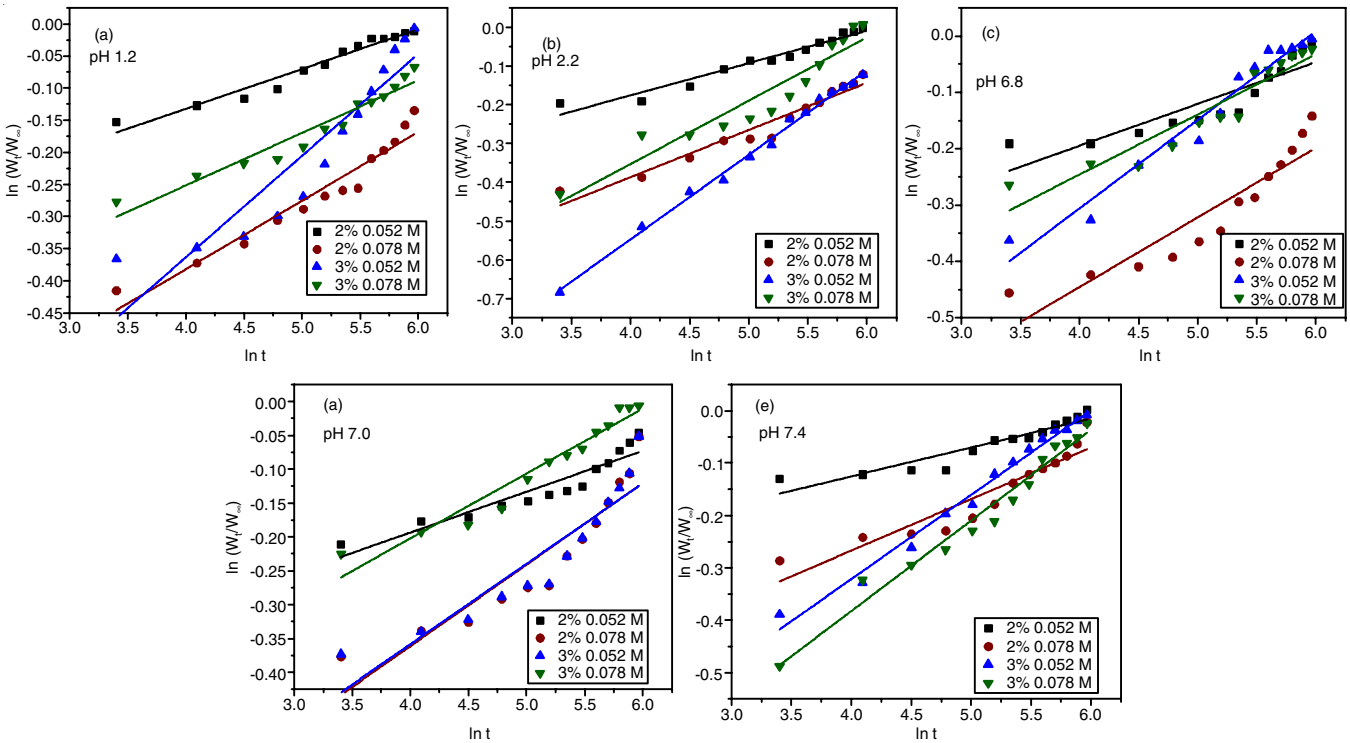

Fig. 10a. Plot of ln (W_t/W_∞) with ln (t) for OSG-*cl*-G Schiff base at different concentration of amount of backbone and the concentration of the oxidizing medium

charged groups increased their swelling under acidic conditions. The slope and intercept of the plot of ln M_t/M_∞ *vs.* ln t give the value of diffusion constant, n and gel characteristic constant, k at different pH values for oxidized sterculia gum and the Schiff base formed with the natural protein gelatin respectively at different concentration of sterculia gum and the different concentration of the oxidizing medium (Figs. 9a and 10a). Value of n and k for OSG and OSG-*cl*-G have been presented in Tables 1 and 2, respectively. The n values were of magnitude of order $n < 0.5$, which indicated that the pseudo-Fickian

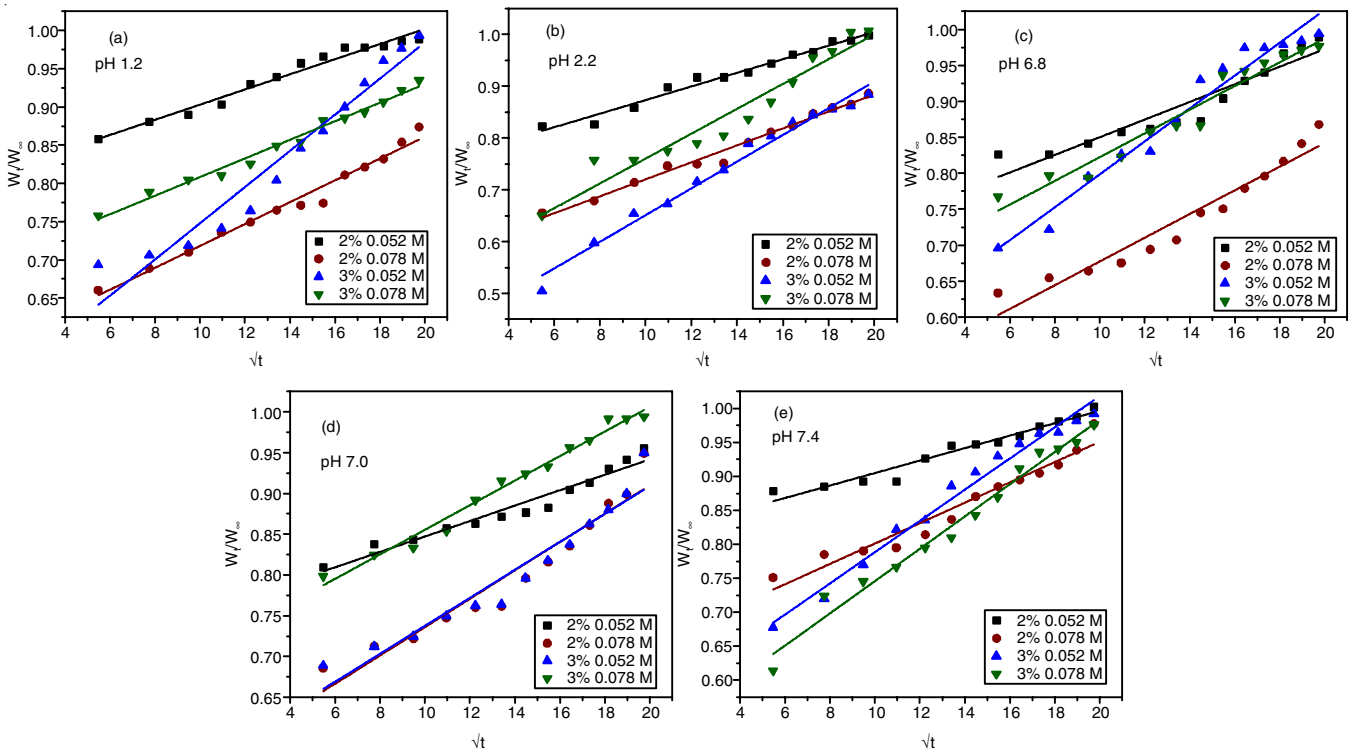

Fig. 10b. Plot of W_t/W_∞ and √t for OSG-*cl*-G Schiff base at different concentration of amount of backbone and concentration of oxidizing medium

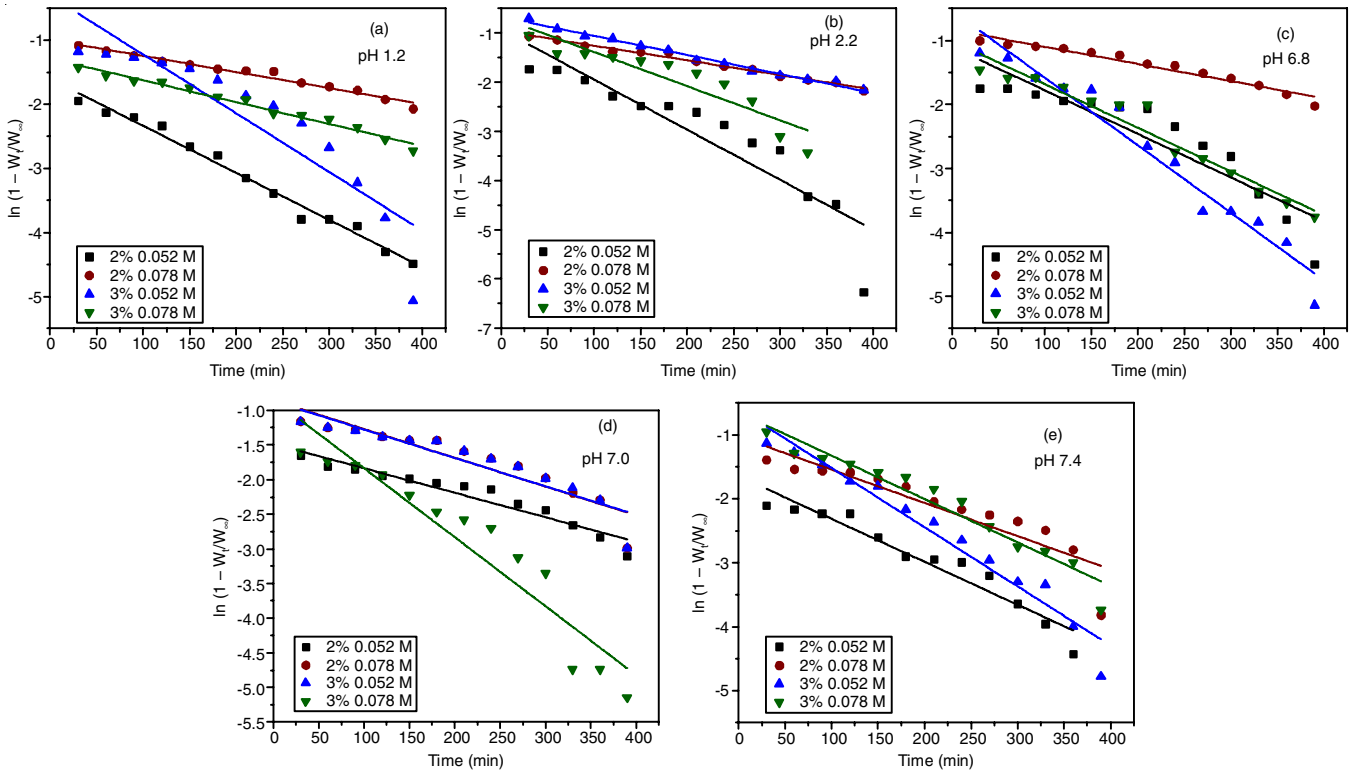

Fig. 10c. Plot of ln (1 – W_t/W_∞) with t for OSG-*cl*-G Schiff base at different concentration of amount of backbone and concentration of oxidizing medium

diffusion governed the absorption mechanism of the hydrogels. A pseudo-Fickian curve resembled Fickian curves, but in this case the approach to the equilibrium is at slower rate [55]. For OSG and OSG-*cl*-Gel, initial diffusion coefficient (D_i), average diffusion coefficient (D_A) have been evaluated from the plot of √t *vs.* W_t/W_∞ (Figs. 9b and 10b) whereas late diffusion coefficient D_L have been calculated from plot of ln (1-W_t/W_∞) with t (Figs. 9c and 10c).

TABLE-1
SLOPE AND INTERCEPT FOR OSG
(ln t *vs.* ln W_t/W_∞) AT DIFFERENT pH

S. No.	SG (%)	Conc. of $NaIO_4$ (M)	pH	Diffusion exponent (n)	Gel characteristic constant (k)
1	2	0.052	1.2	0.05777	-0.42076
2	2	0.078	1.2	0.03971	-0.30382
3	3	0.052	1.2	0.07434	-0.48042
4	3	0.078	1.2	0.03996	-0.24915
5	2	0.052	2.2	0.11404	-0.77485
6	2	0.078	2.2	0.04374	-0.19923
7	3	0.052	2.2	0.03845	-0.19449
8	3	0.078	2.2	0.03303	-0.23546
9	2	0.052	6.8	0.06174	-0.4053
10	2	0.078	6.8	0.02051	-0.03337
11	3	0.052	6.8	0.05574	-0.33713
12	3	0.078	6.8	0.06528	-0.41959
13	2	0.052	7.0	0.04377	-0.25895
14	2	0.078	7.0	0.06024	-0.18953
15	3	0.052	7.0	0.03769	-0.42809
16	3	0.078	7.0	0.06026	-0.43457
17	2	0.052	7.4	0.07712	-0.34363
18	2	0.078	7.4	0.06024	-0.42809
19	3	0.052	7.4	0.03592	-0.23571
20	3	0.078	7.4	0.05504	-0.34534

TABLE-2
SLOPE AND INTERCEPT FOR OSG-*cl*-G SCHIFF
BASE (ln t *vs.* ln W_t/W_∞) AT DIFFERENT pH

S. No.	SG (%)	Conc. of $NaIO_4$ (M)	pH	Diffusion exponent (n)	Gel characteristic constant (k)
1	2	0.052	1.2	0.06195	-0.37993
2	2	0.078	1.2	0.10681	-0.80932
3	3	0.052	1.2	0.15823	-0.99646
4	3	0.078	1.2	0.08185	-0.57899
5	2	0.052	2.2	0.08328	-0.50906
6	2	0.078	2.2	0.12185	-0.87456
7	3	0.052	2.2	0.21797	-1.41925
8	3	0.078	2.2	0.16340	-1.00675
9	2	0.052	6.8	0.07427	-0.49178
10	2	0.078	6.8	0.12350	-0.93937
11	3	0.052	6.8	0.15712	-0.93433
12	3	0.078	6.8	0.10657	-0.67209
13	2	0.052	7.0	0.06026	-0.43457
14	2	0.078	7.0	0.12033	-0.84240
15	3	0.052	7.0	0.11870	-0.83302
16	3	0.078	7.0	0.09575	-0.58518
17	2	0.052	7.4	0.05504	-0.34534
18	2	0.078	7.4	0.09827	-0.65996
19	3	0.052	7.4	0.16075	-0.96415
20	3	0.078	7.4	0.17272	-1.07304

In OSG, initial diffusion coefficient (D_i) has been found to be lower than the late diffusion coefficient (D_L) at variable pH (Table-3) indicating faster swelling in the later stage than the initial stage. In case of OSG-*cl*-G Schiff base except at pH 1.2 where the D_i is almost same as that of D_L, at all other pH values the late diffusion coefficient has lower value than the initial diffusion coefficient which clearly indicate the faster swelling initially than the later stage (Table-4).

TABLE-3
DIFFUSION COEFFICIENTS FOR
OSG ($cm^2\ min^{-1}$), D_i, D_L AND D_A

S. No.	SG (%)	Conc. of $NaIO_4$ (M)	$D_i \times 10^5$	$D_L \times 10^5$	$D_A \times 10^3$
1	2	0.052	1.42	3.44	1.289
2	2	0.078	1.17	3.28	1.992
3	3	0.052	2.65	9.55	1.725
4	3	0.078	0.781	8.76	2.398
5	2	0.052	5.48	5.64	0.263
6	2	0.078	1.36	18.1	3.019
7	3	0.052	0.857	22.2	2.606
8	3	0.078	0.594	4.75	2.500
9	2	0.052	1.92	7.72	3.079
10	2	0.078	0.0825	–	3.267
11	3	0.052	1.55	10.5	2.318
12	3	0.078	1.99	6.25	1.904
13	2	0.052	1.12	11	2.434
14	2	0.078	1.77	4.44	1.225
15	3	0.052	0.824	18	2.924
16	3	0.078	1.76	4.49	1.798
17	2	0.052	3.55	37.3	2.648
18	2	0.078	4.99	21.4	1.656
19	3	0.052	0.735	8.72	2.376
20	3	0.078	1.65	8.51	2.482

TABLE-4
DIFFUSION COEFFICIENTS FOR OSG-*cl*-G
SCHIFF'S BASE ($cm^2\ min^{-1}$), D_i, D_L AND D_A

S. No.	SG (%)	Conc. of $NaIO_4$ (M)	pH	$D_i \times 10^5$	$D_L \times 10^5$	$D_A \times 10^3$
1	2	0.052	1.2	1.93	9.31	2.434
2	2	0.078	1.2	4.01	3.18	1.682
3	3	0.052	1.2	11.0	11.6	0.817
4	3	0.078	1.2	46.4	4.32	1.634
5	2	0.052	2.2	54.0	128.0	2.333
6	2	0.078	2.2	84.1	3.78	0.942
7	3	0.052	2.2	208.0	4.91	0.576
8	3	0.078	2.2	180.0	8.8	0.961
9	2	0.052	6.8	47.2	8.63	2.248
10	2	0.078	6.8	85.3	3.4	0.289
11	3	0.052	6.8	164.1	13.4	1.361
12	3	0.078	6.8	85.3	8.63	1.061
13	2	0.052	7.0	28.1	4.49	1.887
14	2	0.078	7.0	95.0	5.25	1.581
15	3	0.052	7.0	92.5	5.2	1.571
16	3	0.078	7.0	71.2	12.6	1.639
17	2	0.052	7.4	26.5	8.51	2.510
18	2	0.078	7.4	70.6	65.9	1.665
19	3	0.052	7.4	165.0	11.7	1.665
20	3	0.078	7.4	177.1	8.6	0.490

Conclusion

In this work, it has been concluded that oxidation-assisted fabrication of functionalized sterculia gum/protein hybrid gel network is an efficient method since it replaces the use of initiating systems and toxic crosslinking agents. Sodium periodate-assisted reaction of sterculia gum brings about oxidation of *vicinal* hydroxyl groups to carbonyl groups. During oxidation process, cleavage of C2-C3 bond of β-D-galactose, C2-C3, and C3-C4 bond of β-D-glucuronic acid and C3-C4 bond of α-L rhamnose residue units of sterculia

gum took place. The Schiff base reaction between the carbonyl groups present in the oxidized sterculia gum and the amino group in gelatin brings about effective gelation at ambient conditions. Oxidized sterculia gum displays district behaviour in term of an increase in crystallinity and decreased swelling as compared to the native sterculia gum. Oxidized sterculia gum (OSG) and OSG-*cl*-G Schiff base show pH-responsive behaviour and porous structure as confirmed from SEM analysis. Newly synthesized gel networks follow pseudo-Fickian diffusion mechanism. These hydrogel networks could be exploited further as drug delivery systems.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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