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Synthesis, Characterisation and Comparative Study of Hydrogel and Nanogels of Psyllium

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ABSTRACT

Natural polysaccharides are being explored as the matrices for attaining speciality materials for pharmaceutical, medicinal and environmental applications via chemical modification such as grafting. Psyllium polysaccharide-based hydrogels and nanogels have potential biomedical and water purification applications due to their advantageous properties such as stimulus responsiveness, biocompatibility, target drug delivery and stability. The present study aims to synthesise hydrogel and nanogels of psyllium and attain comparative data for the two to undermine their potential applications. Psyllium-cl–*N,N*–MBAm–poly(AAm) hydrogel (Psy-MBAm-AAm–hg) and Psyllium–cl–*N,N*–MBAm–poly(AAm) nanogel (Psy-MBAm-AAm–ng) were synthesised by grafting acrylamide (AAm) onto psyllium using ammonium persulphate (APS) as a free radical initiator in a redox system where N, N-methylene bisacrylamide acted as a crosslinker. The comparative study of synthesised gels was carried out by studying swelling characteristics at acidic and basic pH (7 and 4) and at varied temperatures for both matrices. The synthesised hydrogel and nanogels were subjected to characterisation by Fourier transform infrared spectroscopy (FTIR), Field Emission Scanning Electron Microscopy (FESEM), and Zeta Potential Analysis to get evidence for successful synthesis and nanogel formation.

INTRODUCTION

Psyllium also known as Ispaghula and Isabgol is a natural polysaccharide obtained from Plantago ovate plant commonly known as desert Indian wheat (Mishra, S., Sinha, S., Dey, K. P., & Sen, G., 2014). More than 200 species of the Plantago genus (usually called psyllium) have been reported all over the world (Thakur, V. K., & Thakur, M. K. , 2014) with an average height of 10 – 15 cm. The Plantago psyllium husk is the raw material for psyllium mucilage with arabinose (~22%), xylose (~75%), galactose (~1.5%), glucose (~0.7%), rhamnose and uranic acid as the main constituents (Cui, S. W., Wu, Y., & Ding, H. 2013). Psyllium husk is the main source of arabinoxylan (Li, Q., Wang, S., Jin, X., Huang, C., & Xiang, Z., 2020) and is of laxative nature so used for the treatment of constipation and diarrhoea due to its water-solubility and gel-forming capacity. Psyllium-based polymeric materials and hydrogels have been reported for various therapeutic and pharmaceutical applications like cholesterol reduction (Uehleke, B., Ortiz, M., & Stange, R., 2008), wound dressing (Ahmad, N., *et al* , 2021), drug delivery (Singh, B., Sharma, *et al*, 2022, Kotta, S., *et al*,2022) tissue regeneration (Poddar, S., *et al*, 2021), cancer treatment (Kumar, D., Gautam, A., & Kundu, P. P., 2022, Masood, R., & Miraftab, M., 2010) lower normal LDL level (Jovanovski, E., *et al*, 2018, Everson, G. T., *et al*, 1992) and Industrial applications like treating of coloured effluents from water (Druzian, S. P., 2021), wastewater treatment (Das, N., Ojha, N., & Mandal, S. K., 2021, Singh, J., Kumar, S., & Sharma, S. 2022) such as removal of Hg(II) from the solution (Kumar, D., Pandey, J., Khan, N., Kumar, P., & Kundu, P. P. 2019) to mention a few.

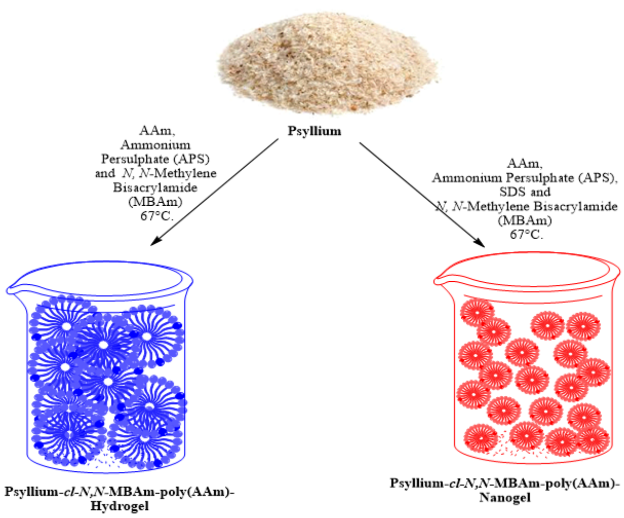

Figure 1: Graphical abstraction of Psy-MBAm-AAm – hg and Psy-MBAm-AAm – ng from Psyllium

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The hydrophilic nature of the psyllium husk imparting its excellent swell ability is one of the defining properties for its varied applications (Singh, B., Chauhan, G. S., Bhatt, S. S., & Kumar, K. (2006), Singh, B., Sharma, N., & Chauhan, N. (2007), Singh, B., & Chauhan, N., 2009). The psyllium hydrogel is explored for wide applications in various fields and little work is done with the psyllium-based nanogels. Hence, in order to get a better picture of the broad application area of Psyllium-based materials we synthesized hydrogels and nanogels of the same and tried to understand what effect the particle size has on the characteristic properties of the gels. Psyllium– cl –*N, N*–MBAm–poly (AAm) hydrogel (Psy-MBAm-AAm – hg) and Psyllium– cl –*N, N*–MBAm–poly(AAm) Nanogel (Psy-MBAm-AAm – ng) were synthesized by crosslinking Acrylamide (AAm) onto psyllium using ammonium persulphate (APS) as a free radical initiator. The synthesis of hydrogel and nanogel was characterized by swelling studies at different pH, and temperature and by subjecting them to FT-IR, FESEM, and Zeta Potential to get evidence for the successful synthesis of hydrogels and nanogels.

MATERIALS AND METHODS

Psyllium, *N, N*-Methylene Bis-acrylamide (Loba Chemie Pvt. Ltd.), Ammonium Persulphate (Loba Chemie Pvt. Ltd.), Sodium Lauryl Sulphate (Loba Chemie Pvt. Ltd.) and Acrylamide (Loba Chemie Pvt. Ltd.), Acetone (Qualikems Fine Chem Pvt. Ltd.), were used as received. Electronic Balance BL-220H of reliability 0.001 g, water bath with thermostat, all other reagents were analytically pure and all solutions were prepared with distilled water.

Synthesis of Psyllium–cl–N, N–MBAm–poly (AAm) hydrogel (Psy-MBAm-AAm – hg)

Psyllium commonly known as Ishab Ghol obtained in the form of small needles. The raw Psyllium was warmed at 35˚C to remove all the moisture content present in it and the dried Psyllium needles were crushed to a fine powder. 0.500g of AAm, 0.015g of Ammonium Persulphate (APS) and (0.015g) of *N, N*-Methylene Bisacrylamide (MBAm) were dissolved in 10 ml distilled water. To the homogeneous mixture 1.0g, powdered Psyllium was added slowly with continuous stirring on a magnetic stiller. The homogeneous mixture thus obtained was sonicated for 60 min at 67˚C. The Pool thus obtained was cooled to room temperature (25˚C) and the hydrogel formed was extracted with acetone. The homopolymer thus formed was extracted in a 60:40 ratio solution of acetone and ethyl alcohol. The extracted hydrogel was left undisturbed at 40˚C in a hot air oven for 24 h. The dried hydrogel was crushed to fine powdered with pestle and mortar and labelled as Psyllium– cl –*N, N*–MBAm–poly(AAm) hydrogel (Psy-MBAm-AAm – hg) shown in Figure 2

Figure 2: Psy-MBAm-AAm – hg hydrogel

Synthesis of Psyllium–cl–*N, N*–MBAm–poly(AAm) nano gel

For the synthesis of Psyllium–cl–N, N–MBAm–poly (AAm) nano gel (Psy-MBAm-AAm – ng), 0.500g of AAm, 0.015g of ammonium persulphate (APS), 0.015g of *N, N*-methylene bisacrylamide (MBAm) and 0.01g of sodium dodecyl sulphate (SDS) were mixed in 10 ml of distilled water. To the homogeneous mixture 1.0g, powdered Psyllium was added slowly with continuous stirring on the magnetic stirrer. The homogeneous mixture thus obtained was sonicated for 60 min at 67˚C. The Pool thus obtained was cooled to room temperature (25˚C) for 2h and the gel formed was extracted with acetone. Extract with acetone was again homogenized in a sonicator at 30˚C for 20 min. The homopolymer formed was removed by extracting the pool in a solution of 60:40 ratio of acetone and ethyl alcohol. The extracted nanogel was left undisturbed for cooling at 40˚C in a hot air oven for 24h. The dried gel was labelled as Psyllium– cl –*N, N*–MBAm–poly(AAm) Nanogel (Psy-MBAm-AAm – ng) shown in figure 3

Figure: 3 Psy-MBAm-AAm – ng Nano gel

Separation of Hydrogels and Nanogels

The hydrogel and Nanogel synthesized with a cross-linker were treated repeatedly with ethyl alcohol to remove the homopolymer formed during the reaction. The unreacted and insoluble product was removed by simple filtration, without drying and treated with ethyl alcohol and water to get the constant weight. The dried gel was again treated with acetone.

Swelling Study

Psy-MBAm-AAm–hg hydrogel was dried at room temperature till constant weight is obtained and 0.050 g was measured with the help of electronic balance and immersed in a solution of pH 4.0 at room temperature (25°C). The swelling study was carried out using the following formula (Singh, B., Chauhan, G. S., Kumar, S., & Chauhan, N., 2007):

Percent Swelling = (Weight of the Hydrogel-Weight of the Xerogel)/Weight of the Xerogel X 100

The pH 4.0 solution was prepared in distilled water with the help of a buffer tablet and was tested with a Digital pH meter. The palate of Psy-MBAm-AAm–hg was immersed in 10ml pH 4.0 solution and taken out after 5 min for weight measurement. The experiment was performed in duplicate and repeated for 10, 20, 40, 80, 160 and 320 minutes time intervals. The swelling study was also carried out in a solution of pH 7.4 at the same time interval. The Swelling behaviour of Psy-MBAm-AAm – hg was also studied on the same pH and after the same time interval at a temperature of 37°C.

For the swelling study of nanogel, Psy-MBAm-AAm–ng nanogel was dried at room temperature to a constant weight and grinded to a fine powder. 0.010g of Nano gel fine power is taken in a centrifuge tube filled with 10 ml solution of pH 4 and 7.4 at room temperature (25°C).

The tube was left undisturbed for 4 minutes and centrifuged for 1 minute at 2000 rpm in a centrifuging machine. The water content was decanted off and blotted dry then the weight of swelled nanogel was calculated. Each experiment was performed in duplicate at the different time spans of 10, 20, 40, 80, 160 and 320 min at the same temperature. The Swelling study of nanogel was also carried out under the same conditions in a solution of pH 4.0, 7.4 at 37°C.

RESULTS AND DISCUSSION

Psyllium cross-linked hydrogel and nanogel were characterized by FTIR, FESEM, Zeta potential and Swelling Study

FTIR

FT-IR spectra of Psy-MBAm-AAm – hg and Psy-MBAm-AAm – ng were recorded using F.T. Infra-Red Spectrophotometer Model RZX(Perkin Elmer) 8010 between 400 to 4000 cm-1 to study the modification. The FT-IR of hydrogel and nanogels of psyllium is presented in Figure 3 and Figure 4 respectively. The broad absorption band due to -OH starching is observed at 3415.30 cm-1 for the hydrogel and nano gel. At 2925.52 cm-1 the band is observed due to asymmetric starching vibration – OH and – CH. The band at 1466.27 cm-1 witnessed the C-H deformation vibration.

The characteristic peaks are observed at 811.63 cm-1, 620.65 cm^{-1} for the grafted Psyllium hydrogel backbone. The absorption band at 1618.39 cm^{-1} and 1616.96 cm^{-1} in the FT-IR of Psyllium hydrogel and nanogels respectively are witnessed due to the stretching of the –C=O of the –COOH group and C-O-C starching vibrations. The out of plane bending of -NH and -CN groups in gel results in a peak at1382.68 cm^{-1}. -NH and -CH bending, generate a band at 897.22 cm^{-1}, while the band at 1034.14 cm^{-1} is due to C – O starching. In the case of Psy-MBAm-AAm – ng an additional peak at 479.29 Cm^{-1} is noticed due to C–C ring deformation in psyllium and $-CH_2$ wagging.

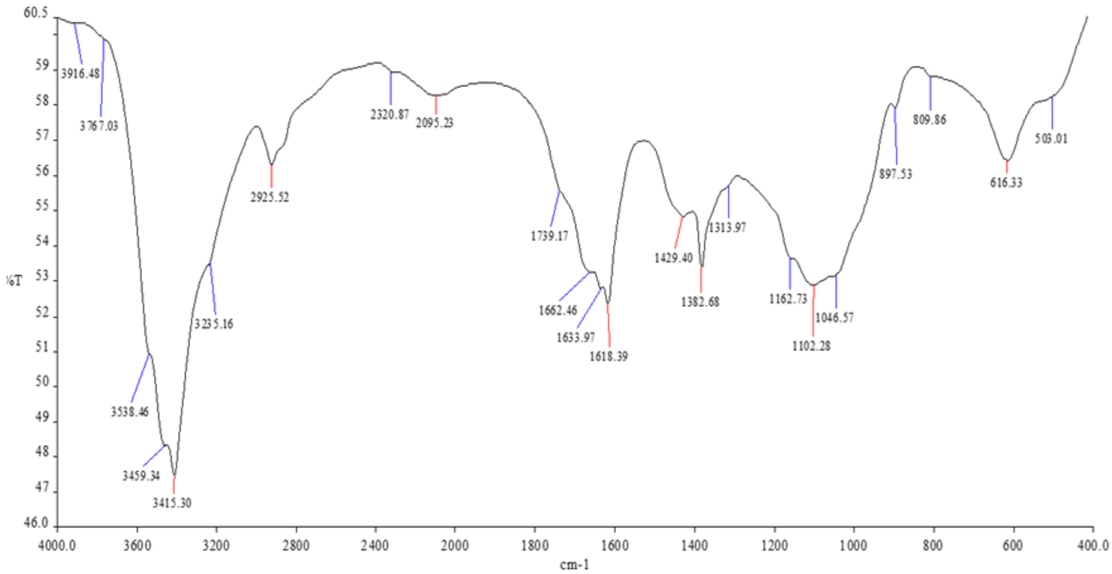

Figure 4.1: FT-IR of Psy-MBAm-AAm – hg hydrogel

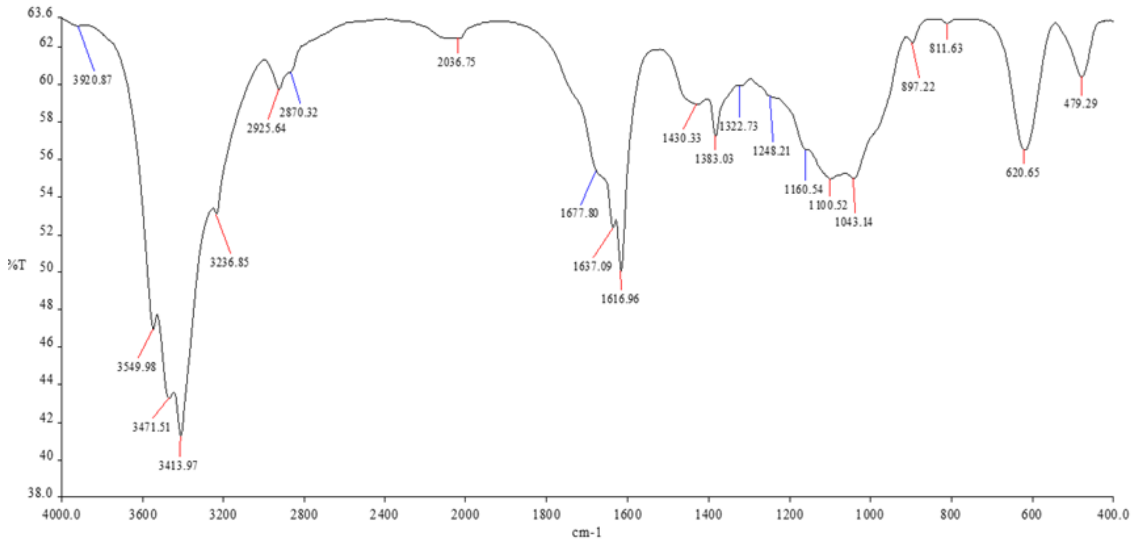

Figure 4.2: FT-IR of Psy-MBA-AAm – ng Nanogel

FESEM

The surface morphology of Psy-MBAm-AAm – hg hydrogel and Psy-MBAm-AAm – ng nano gel was studied using Hitachi SU 8010 Series and images at different resolutions are presented in Figure 5 (1 and 2). FESEM data indicated the somewhat rough surface of the hydrogels with small grooves and ridges. FESEM images of nanogel indicate structural heterogeneity in its surface, where nanoparticles of size 37.4 – 97.2 nm in diameter get accumulated together to form an aggregate molecule of diameter 500 nm. The agglomeration rate is observed to increase over time due to the hydrophilic and H-bonding nature of the nanogels.

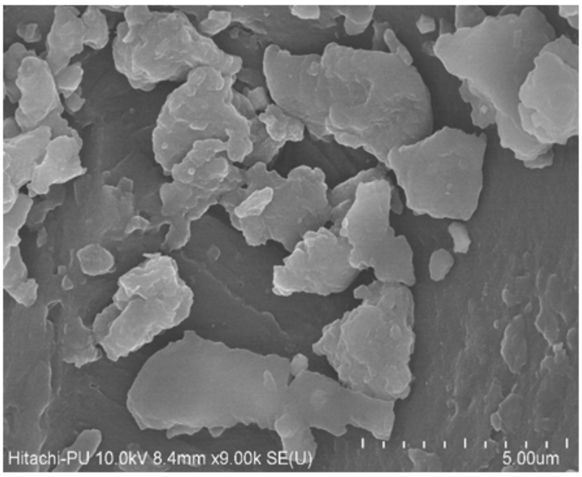

Figure 5.1: FESEM of Psy-MBAm-AAm – hg

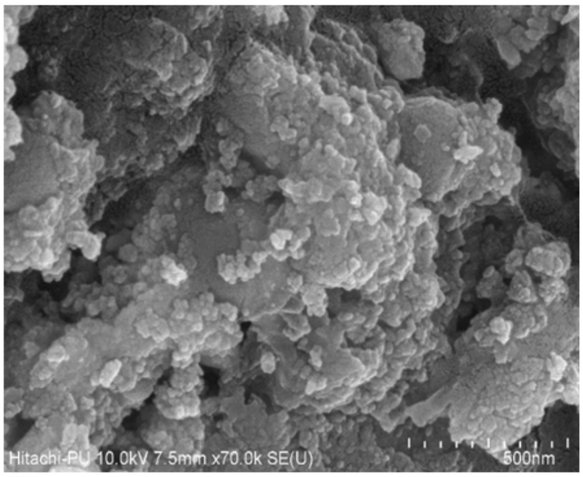

Figure 5.2: FESEM of Psy-MBAm-AAm – ng

FESEM indicated successful synthesis of nanogels of Psyllium acrylamide nanogels crosslinked with MBAm.

Particle Size Analysis

The successful synthesis of Psy-MBAm-AAm–ng nanogel is also verified by Zeta Size particle analysis. 1g/L of aqueous solution at 25°C is used to study the size distribution of nanogel using Malvern Zetasizer Nano ZS. Zeta potential is the electrokinetic potential of the colloidal system and is the measure of phase difference in the boundaries of Solid and liquid phases in a colloidal solution.

The Polydispersity Index (PDI) value of Zeta Sizer (Figure 6.1) is 0.558, verifying that nanoparticles are of varied size with little variation at 25ºC. The average radius Z (Z- average) of synthesized nanoparticles in an aqueous dispersion medium was 278.7 nm at 100% intensity.

Since the Zeta potential is the measure of the surface charge of the nanoparticles in the solution and hence is used to measure the stability of nanoparticles in the dispersion medium. The higher the positive or negative value of the Zeta potential (ꞩPotential) more will be the stability of nanoparticles in the dispersion medium.

In the case of Psy-MBAm-AAm – ng nanogel the Zeta potential value of 93.3 % particles is -3.52mV, indicating that these nanoparticles were negatively charged and have a rapid coagulation tendency in aqueous solution. The 6.5 % of nanoparticles have a higher positive zeta potential of 39.3 and are moderately stable with a positive charge, while only 0.1 % of nanoparticles with -119 mV zeta potential show excellent stability with Negative charge in aqueous

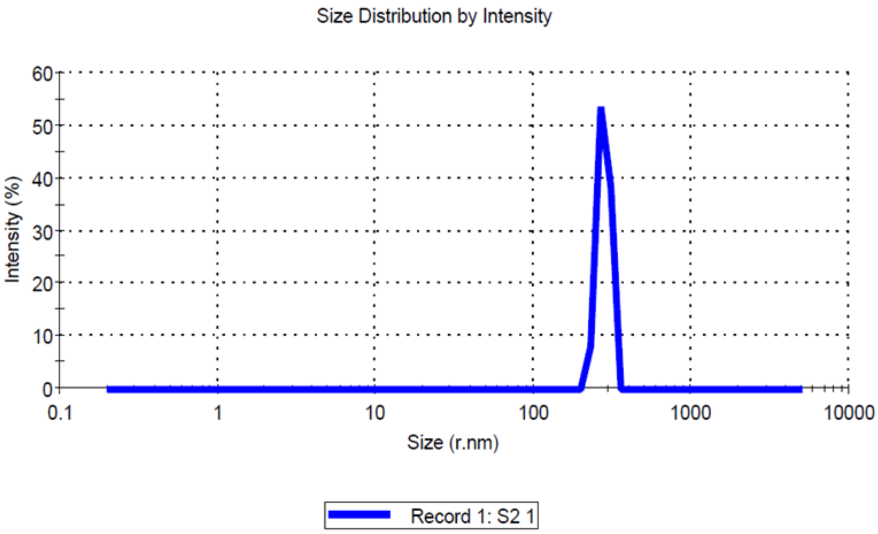

Figure 6.1: Zeta Sizer Analysis of Psy-MBAm-AAm – ng Nanogel

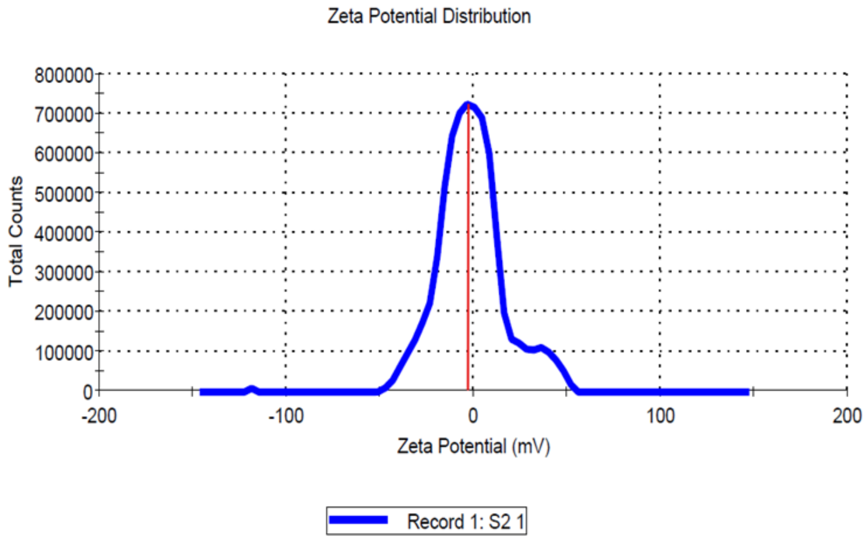

Figure 6.2: Zeta Potential Analysis of Psy-MBAm-AAm – ng Nanogel

solution. In nutshell, average particles are negatively charged with a zeta potential of -1.37 mV and get rapidly coagulated. Due to hydrophobic interactions, these nanoparticles have a higher affinity for water molecules in an aqueous solution and are hence responsible for swelling behaviour. The ζ Potential curve for Psy-MBAm-AAm – ng nanogel is shown in Figure 6.2

Swelling Behaviour

The swelling characteristics for the synthesized hydrogel and nanogel observed at 4.0 and 7.0 pH (25°C and 37° C) are presented in Table 1 and Figure 1.8. Figure 1.9 shows the swelling behaviour of Psy-MBAm-AAm – ng nanogel at different reaction conditions. Table 2 gives the details of the variables used.

Swelling Behaviour of Psyllium–cl–poly (AAm) Hydrogel and Nanogel

An aqueous medium was used to carry out the swelling study of the polymeric network of psyllium hydrogel and nanogel. The swelling was carried out till equilibrium swelling is obtained at a fixed pH maintained at a particular temperature.

The Swelling Percent (%Sw) of the gel was calculated as using the following formula (Singh, B., Chauhan, G. S., Kumar, S., & Chauhan, N., 2007):

% Sw = (W¬sw – Wd)/ Wd x 100

Where

Wsw = Weight of swollen Gel

Wd = Weights of swollen gel

The swelling behaviour of the hydrogel and nanogel networks was studied as a function of time, temperature and pH. The data thus obtained indicated that the increase in surface area on the psyllium matrix on conversion to nanogel resulted in a prominent increase in the percent swelling at all studied pH and temperatures.

Table 1: Swelling of Psy-MBAm-AAm – hg and Psy-MBAm-AAm – ng Nanogels

Psy-MBAm-AAm – hg					**Psy-MBAm-AAm – ng**			
Duration	T1 = 24°C		T2 = 37°C		T1 =24°C		T2 = 37°C	
	pH = 4	pH = 7	pH = 4	pH = 7	pH = 4	pH = 7	pH = 4	pH = 7
% Swelling	Sw1	Sw2	Sw3	Sw4	S'w1	S'w2	S'w3	S'w4
5 Min	176	200	228	276	1434	898	2012	1710
10 Min	294	314	300	372	1554	990	2178	2118
20 Min	398	420	386	460	1872	1016	2286	2204
40 Min	532	580	596	648	2068	1054	2746	2374
80 Min	666	762	750	888	2290	878	2620	2362
160 Min	866	1074	1006	1190	2556	778	2450	2352
320 Min	1232	1398	1256	1500	2804	776	2138	2288
640 Min	1624	2406	1288	1716	3212	908	2114	2266

Table 2: Variable used with their description

Sr. No.	**Variable**	**Description**
1	Psy-MBAm-AAm – hg	Psyllium– *cl* –N, N–MBAm–poly (AAm) hydrogel
2	Psy-MBAm-AAm – ng	Psyllium– *cl* –N, N–MBAm–poly (AAm) nanogel
3	AAm	Acrylamide
4	APS	Ammonium persulphate
5	SDS	Sodium dodecyl sulphate
6	MBAm	N, N-Methylene Bisacrylamide
7	% Sw	Percent Swelling

Effect of temperature and pH on the Swelling of Psy-MBAm-AAm – hg Hydrogel

The swelling studies carried out at the two temperatures indicated the percent swelling of Psy-MBAm-AAm – hg hydrogel increases initially at all pH and temperature due to the penetration of solvent molecules into the crosslinked matrix. A steady increase was noticed up to 320 min at all reaction conditions. However, at 37°C and pH 4, the % Swelling becomes constant. At pH 7 (24°C) a sharp increase in swelling behaviour was noticed and hydrogel behaves as a super absorbent. Whereas under other conditions the %swelling of hydrogel was observed to increase at a constant rate. The nanogel exhibited almost double %swelling than the corresponding hydrogel at pH 4 (24°C and 37°C).

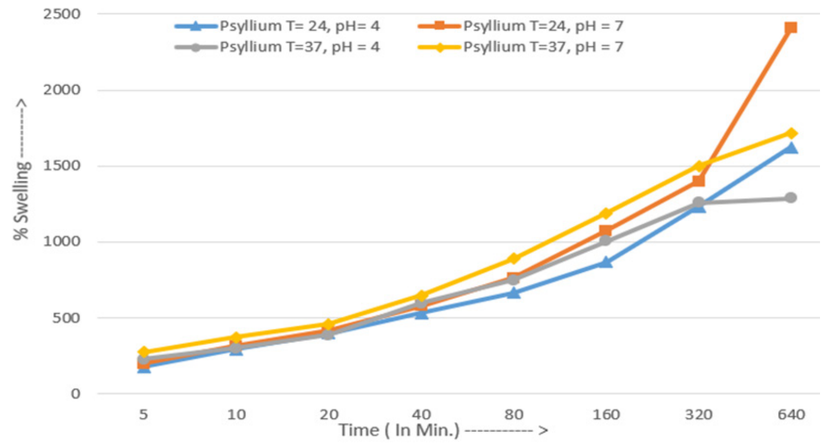

Figure 7: Swelling behaviour of (Psy-MBAm-AAm – hg) Hydrogel

Effect of temperature and pH on the Swelling of Psy-MBAm-AAm – ng) Nanogel

In Psy-MBAm-AAm – ng, the variation of % Swelling is not as regular as noticed in the case of a hydrogel. The significant effect of temperature was noticed at pH = 4, nanogels act as super observant at normal room temperature (24°C) but at 37°C the maximum swelling is noticed at 40 min and thereafter a steady decrease in swelling up to 320 min was observed till it becomes constant. This change in swelling behaviour is due to rupturing of the polymeric backbone at high temperatures. At high temperature, the binding forces are mainly hydrogen bonding which binds the different polymer chains together and creates a cavity for holding the solvent molecule becomes inoperative and release the excess water molecules back.

This results in the deswelling of the nanogels and retains only those solvent molecules which help in maintaining the equilibrium in different forces. The further increase in the time period results in the dissolution of nanogels. The Porosity and particle size of gels play a crucial role in maintaining the balance between different cohesive

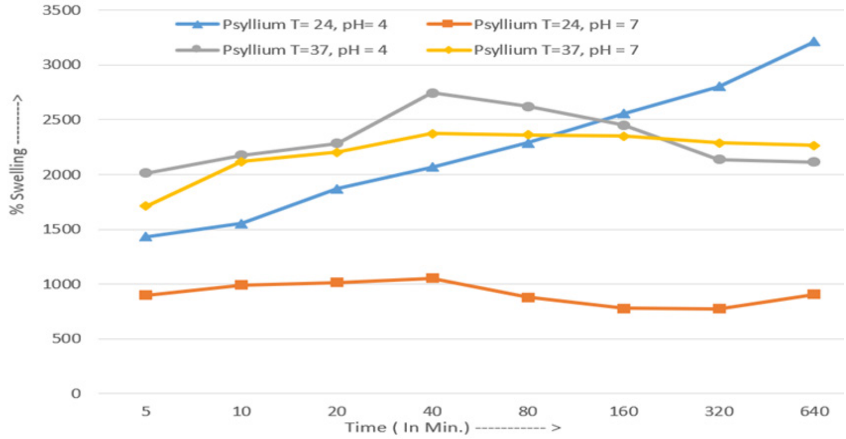

Figure: 8 Swelling behaviour of Psy-MBAm-AAm – ng Nanogel

forces. From Table 1, it is observed that at T_1 and pH = 4, initially, the rate of adsorption of water molecules by Psy-MBAm-AAm–hg hydrogel is eight folds than that of Psy-MBAm-AAm–ng and starts decreasing with the increase in time and finally reach to double. The high rate of adsorption by nanogel is attributed due to the high porosity and particle size and the decrease in adsorption is because of the occupancy of the empty void by solvent molecules in the polymeric chain.

The same adsorption behaviour with a higher adsorption rate is observed at pH=7 and the same temperature. The lower swelling rate at pH=4 is due to the breaking of the polymer network chain in an acidic medium. At high-temperature T_2, and low pH the Psy-MBAm-AAm–hg swelled continuously but for Psy-MBAm-AAm–ng, adsorption of the solvent molecule increases initially, and reaches the maximum at 40 min and then starts decreasing. The swelling of Psy-MBAm-AAm – ng nanogel at T_2 and pH=7 is much higher than that of Psy-MBAm-AAm– hg hydrogel.

CONCLUSION AND RECOMMENDATIONS

The paper aims to draw a comparative between the psyllium-based acrylamide grafted hydrogel and nanogels. Psy-MBAm-AAm–hg and ng were successfully synthesized and characterized. The FESEM and particle size analysis data indicated that the nanogels of size below 250nm that exhibited negative zeta potential value were obtained. The swelling characteristics studied at varied pH and temperatures indicated that the nanogels exhibited greater swelling than the corresponding hydrogels due to an increase in the surface area.

The results pointed towards the potential applications in site-specific drug loading and release studies as at body temperature high % swelling was observed at acidic pH. The nanogel exhibiting a continuous increase in water absorption at room temperature and in mildly acidic pH, opens the application of the same for sewage or industrial wastewater management. There are a number of applications already reported for hydrogel but the method reported in this paper is fast and simple and also suitable for the synthesis of Psyllium – Acrylamide-based nanogels.

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