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## Polyvinylalcohol/Polyethyleneimine Hydrogels: Evaluation of Swelling, Dehydration and Antibacterial Activity

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### Abstract

In the present study, Polyvinylalcohol/polyethyleneimine (PVA/PEI) hydrogels designed with different Glutaraldehyde (Glu) amount, PEI amount and PVA concentration were synthesized via solvent casting technique. The fabricated PVA/PEI hydrogels with different compositions were evaluated for swelling rate, dehydration properties and antibacterial activity to determine superior combination. The swelling tests were performed for 0-300 min at pH 7.4 and 37 °C. All of the prepared PVA/PEI hydrogels swelling ratio were observed in the range of 221-321%. The highest dehydration rate was found for the PVA/PEI hydrogel with the lowest PVA concentration and the lowest dehydration rate was found for the PVA/PEI hydrogel with the highest Glu amount. All the PVA/PEI hydrogels demonstrated high antibacterial activity against *E.coli*. PP5 (prepared with 5% PVA, 0.5 mL PEI and 30 µL Glu) was determined as the selected hydrogels with optimized characteristics in respect to swelling, dehydration and antibacterial activity data. This study highlights potential usage of the resultant PVA/PEI hydrogels as antibacterial wound dressings in wound care applications.

**Keywords:** Polyvinylalcohol, polyethyleneimine, swelling rate, dehydration properties, antibacterial activity

### INTRODUCTION

The hydrogels are crosslinked three-dimensional highly hydrophilic network polymers that the origin of the term hydrogel goes back to 1894 [1]. There has been received special attention because of their acidic or basic pendant groups such as carboxylic acid (-COOH) [2] sulfonic acid (-SO<sub>3</sub>H) [3], primary amine (CONH<sub>2</sub>) [4] and quaternized ammonium (NR<sub>4</sub><sup>+</sup>) [5]. Hence, these polymeric matrices able to hold a huge amount of water and biological fluids due to this groups.

In general, hydrogels swelling behavior takes places as a result of ionization. Thanks to the ionization property, it is possible to absorb the water formed in the hydrophilic functional groups connected to the polymer chains. Moreover, swelling behavior of the hydrogels depends on the swelling properties of hydrogels, polymer structure (hydrophilicity or hydrophobicity etc), polymer-solvent interactions (valence of counter ion of external swelling medium), and cross-linking density [6]. The swelling and deswelling behavior of hydrogel are important to improve the understanding of mechanism due to usage in for biomedical and industrial applications including agrochemistry [7], wastewater treatment [4], drug delivery systems [8], wound dressing [9], tissue engineering [10].

Polyethyleneimine (PEI) has received increased attention due to its cationic hydrophilic nature with the presence of amine groups (primary, secondary and tertiary amino) [11]. The hydrophilic character provides good water swelling capacity. Furthermore, high positive charges of the PEI responsive for antibacterial activity [12]. Poly (vinyl alcohol) (PVA) is biodegradable, biocompatible, non-toxic and eco-friendly which can be potentially used for promising biomaterials [13-15]. PVA exhibits water solubility properties thanks to its hydroxyl groups (-OH) [16]. Due to its hydrophilic character, the addition of a polymeric system can increase the swelling capacity. Glutaraldehyde (Glu) is a common chemical crosslinking agent for PVA polymer systems due to its high crosslinking rate [17, 18]. Glu has two aldehyde groups one of

may react with the hydroxyl group of polyvinyl alcohol [19]. There are very few studies in the literature addressing possible potential applications of copolymeric PEI and PVA hydrogels. In one of these studies, Wang et al., successfully prepared stretchable elastomer composite hydrogel based on PEI and PVA. Prepared hydrogels demonstrated excellent mechanical properties and high biocompatibility. With obtained results PVA and PEI hydrogels can be used different application areas such as healthcare monitoring, human-machine interfaces and soft robots [20]. In a following study, the biosensor application is entrapment of thermolysin enzyme into a PVA/PEI matrix containing gold nanoparticles and cross-linked using Glu vapors were shown to produce successfully [21]. Also, it has been well supported that heavy metal Cr (VI) was highly effective removal from aqueous solutions and exhibits a good reusability [22]. In the present study, we successfully fabricated PVA and PEI based hydrogels using Glu as crosslinker via solvent casting method. PVA/PEI hydrogels were fabricated with different amounts of Glu, PEI and PVA. The PVA/PEI hydrogels were investigated for swelling and dehydration properties as well as antibacterial performances against *E. coli*.

### MATERIALS AND METHODS

#### Chemicals and Materials

PVA (Mw 146000-186000; 99% hydrolyzed), PEI (average Mn ~1,800 by GPC, 50 wt. % in H<sub>2</sub>O) and Glu obtained from Sigma Aldrich. Luria Bertani (LB) agar and LB broth were obtained from Merck. All the other chemicals used were of analytical grade etc.

#### Preparation of PVA/PEI Hydrogels

PVA/PEI hydrogels with different combination of PVA, PEI and Glu were prepared via solvent casting method. Firstly, PVA was dissolved in distilled water at 90°C under magnetic stirring for 2 h. Then, PEI was added drop by drop to the PVA solution with vigorous mixing. PVA and PEI was interacted for 30 min to form a homogeneous solution. Glu as a crosslinker was added to the solution dropwise in the ice bath under

magnetic stirring. The mixture was poured onto 24-well plate and solvent was evaporated slowly at room temperature. Lastly, the hydrogels were dried under vacuum at 40°C for 4 h. The amounts of the components used for the preparation were listed in Table 1.

**Table 1** The compositions of PVA/PEI hydrogels

Codes	PVA (% w/v)	PEI (mL)	Glu (μL)
P30	10	1	30
P60	10	1	60
P120	10	1	120
P30-0.5	10	0.5	30
P30-0.1	10	0.1	30
P30-0.05	10	0.05	30
PP5	5	0.5	30
PP2.5	2.5	0.5	30
PP1	1	0.5	30

### Characterization of PVA/PEI Hydrogels

With using FTIR-ATR measurements the chemical structure of the PVA/PEI hydrogels was determined. The FTIR-ATR spectrum was obtained in the range of 600-4000 cm<sup>-1</sup>. The morphological properties of the selected hydrogels were performed with FESEM (Hitachi Regulus 8230 FE-SEM). The samples were coated with gold prior to SEM measurements.

#### Swelling ratio tests

Swelling measurements were carried out with using a thermostated water bath at 37 °C, at pH 7.4 and deionized water were used. Hydrogel samples were dried and measured ( $M_{dried}$ ) after ( $M_{dried}$ ) hydrogel samples were placed into buffer solution and the swollen hydrogels were measured after the excess water was removed from the surfaces of the samples ( $M_{swollen}$ ). Hydrogels swelling ratio were calculated with the given formula (1):

$$SR(\%) = \frac{M_{swollen} - M_{dried}}{M_{dried}} \times 100 \quad (1)$$

All of the swelling ratio experiments were performed in triplicate.

#### Dehydration tests

For the investigation of the hydrogels water loss percentage, dehydration tests were performed. Firstly, hydrogels were swollen in distilled water and keep under the room temperature. After at specific time intervals the wet hydrogels were weighed and the percentage of the water loss was calculated as given formula below (2). All of the dehydration experiments were performed in triplicate.

$$\text{Dehydration rate } (\%) = \frac{m_i - m_t}{m} \times 100 \quad (2)$$

At the formula  $m_i$  is represented wet hydrogels initial weight,  $m_t$  represented the measured weight of the wet hydrogel at

predetermined time  $t$  and  $m$  is represented the hydrogels water content. All of the dehydration experiments were performed in triplicate.

### Antibacterial Assay

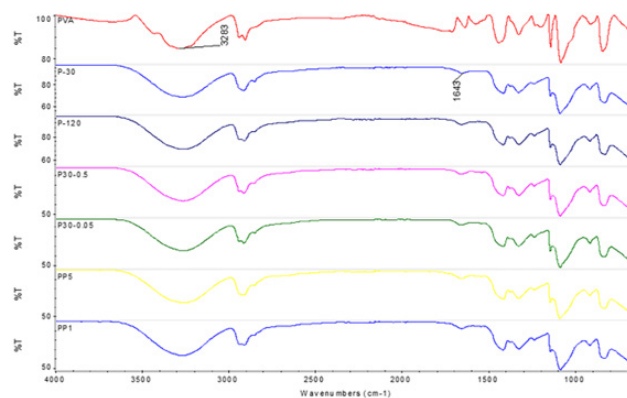
*Escherichia coli* (*E. coli*) was used for the antimicrobial studies of PVA-PEI hydrogels. *E. coli* was inoculated in LB medium and incubated for 24 h, at 37°C. After incubation bacterial solution diluted to be 10<sup>-3</sup> times and 100 μL of diluted bacterial suspensions were added to eppendorf tubes with 900 μL LB medium inside. Hydrogels were cut as 9 mm diameter, washed with deionized water and sterilized under ultraviolet (UV) light for 10 min for each side. Each hydrogel was added into eppendorf tubes containing bacterial suspensions. One eppendorf tube without added PVA/PEI hydrogel was considered as a control group. PVA/PEI hydrogels were incubated at 37°C for 24h inside of incubator set up for 150 rpm. After the incubation, PVA/PEI hydrogel containing bacterial solutions were diluted to be 10<sup>-6</sup> and control group diluted to be 10<sup>-7</sup>. 100 μL of diluted solutions were inoculated onto the LB agar plates and spread to the surface. Agar plates were incubated at 37°C for 24 h, after the number of colonies were counted and colony forming units (CFUs) was calculated. All of the experiments were performed three times and average colony numbers were taken into account. [23, 24].

## RESULTS AND DISCUSSION

### Characterization of PVA/PEI Hydrogels

#### FTIR-ATR measurements of PVA and PVA/PEI hydrogels

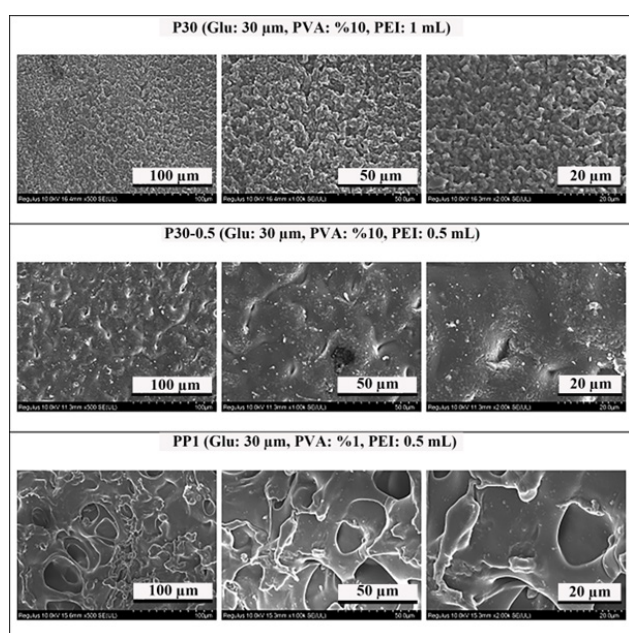
FTIR-ATR spectra of PVA/PEI hydrogels were shown in Fig. 1. It was clearly seen that they present very similar profiles. The band appeared at 1643 cm<sup>-1</sup> is assigned to the formation of C=N bonds supporting the crosslinking reactions between primary amine groups of PEI and glutaraldehyde [25]. The characteristic band of pure PVA at around 3283 cm<sup>-1</sup> corresponds to the stretching vibration of hydroxyl groups of PVA. The intensity of this band was decreased significantly after the synthesis of PVA/PEI hydrogel indicating the reduction of available hydroxyl groups which attributes to the formation of hydrogen bonding of PVA with PEI [26]. In addition, with the increased amount of Glu, the color of the hydrogels was observed to change from white to slight yellow indicating the cross-linking reaction took place.



**Figure 1** FTIR-ATR spectra of PVA and PVA/PEI hydrogels.

### SEM measurements of the P30, P30-0.5 and PP1 hydrogels

The SEM images of P30, P30-0.5 and PP1 were demonstrated in Figure 2 to explain the morphological characteristics of the hydrogels. All the hydrogels possessed porous structures with different porosity and homogeneity. The average pore sizes of the hydrogels were determined with ImageJ software using 15 individual measurements for each sample and found as  $3.5 \pm 0.9$ ,  $13.4 \pm 4.9$  and  $37.7 \pm 11.2$   $\mu\text{m}$  for P-30, P30-0.5 and PP1 respectively. P-30 had interconnected pores with high porosity. The pores of PVA/PEI hydrogel increased in size with the decrease in PEI amount. With the reduction of PVA ratio, the pores became larger which supports the higher swelling capacity enabling the penetration of water molecules. Morphological features were found to be consistent with the swelling results.



**Figure 2** SEM images of the PVA/PEI hydrogels (P30, P30-0.5 and PP1).

### Swelling ratio of the PVA/PEI hydrogels

The effect of Glu amount on swelling kinetics was examined and the obtained results were demonstrated in Figure 3a. The mass ratios of Glu were utilized as 0.5, 1 and 2 %w/v. PVA/PEI hydrogels follow the similar trend; firstly, they swelled rapidly at the initial periods after immersion, then the swelling ratio decreases and finally reached an equilibrium value. The swelling amount of PVA/PEI fabricated with 30  $\mu\text{L}$  Glu was higher than that of PVA/PEI fabricated with 60 and 120  $\mu\text{L}$  Glu. PVA/PEI hydrogels swelling ratio was decreased when the amount of Glu was increased due to formation of more crosslinks between PEI and Glu. The increase in Glu amount results in more rigid polymeric structure since the mobility of the polymer chains is hindered [27]. Thus, with the higher crosslinking available free area for the diffusion of water molecules is diminished and the swelling capability is lowered. 30  $\mu\text{L}$  is selected and used for the further experiments.

Figure 3b shows the effect of PEI content on swelling ratio of

PVA/PEI hydrogels when PEI amount was changed as 1, 0.5, 0.1 and 0.05 mL. The highest SR was observed for PVA/PEI hydrogel synthesized using 0.5 mL PEI. The increased swelling capacity caused by incorporation of PEI chains providing high amount of amine groups available for hydrogen bonding with water molecules. PEI with branched structure having high amount of amine groups enables interaction with more water molecules [28]. However, SR was reduced when PEI amount was 1 mL indicating more than adequate amount of PEI blocks the diffusion of water molecules throughout the polymeric matrix. The obtained SR results were compatible with the results reported by Mohan and Geckeler [29].

The effect of PVA content on water holding capacity of PVA/PEI hydrogels was evaluated in Figure 3c. The highest swelling capacity was found with PVA/PEI hydrogels prepared with 1% PVA content and the lowest swelling amount was obtained from the hydrogels having PVA (10 %). The swelling capability of PVA/PEI hydrogels decreased due to the increase in PVA content. Higher PVA amount in the polymeric structure may cause stronger hydrogen bonding leading less interaction with water molecules. Chaturvedi et al. and Shim et al were reported similar results [14, 30]. In addition, increased amount of PVA content provides the integration of PEI chains more effectively leading the polymeric structure more hydrophilic.

In order to reach the swelling equilibrium ( $t_{eq}$ ), evaluations were carried out at different time periods. Required time periods were follows as: 90, 120 and 150 min for P30, P60 and P120; approximately 60 min for P30-0.5, P30-0.1, P30-0.05; approximately 45 min for PP5, PP2.5 and PP1. The increased amount of Glu caused the increment in  $t_{eq}$ . The hydrogel becomes denser with the increase of cross-linker amount making the hydrogels swollen in a slower manner. For the other PVA/PEI hydrogels,  $t_{eq}$  remained almost stable due to the constant cross-linker amount.

### Statistical Analysis

In this study, Design Expert 7.0 trial version software package program was used to construct design by using historical data. ANOVA Table comprises the predicted the effective parameters on responses.

### Swelling kinetic models

The swelling kinetics of PVA/PEI hydrogels in distilled water were examined with first order kinetic model and second order kinetic model which are defined as the linearized forms:

$$\log\left(1 - \frac{M}{M_e}\right) = K_f t \quad (3)$$

$$\frac{t}{M} = A + Bt \quad (4)$$

$$A = \frac{1}{K_s M_e^2} \quad (5)$$

$$B = \frac{1}{M_{e,theo}} \quad (6)$$



where,  $M$  and  $M_{e,theo}$  are water swelling of the hydrogel at time  $t$  (min) and at equilibrium. The slope of  $\log(1 - M/M_e)$  versus  $t$  gives  $K_1$  that is the first order rate constant. The slope of  $t/M$  versus  $t$  yields  $K_2$  which is the second order rate constant. The kinetic parameters were listed in Table 2. The swelling data fits more to second order kinetic model rather than first order kinetic model according to the regression coefficients. Also, the theoretical swelling ratio is close to the experimental swelling ratios. As seen in Table 2, the  $K_2$  values of PVA/PEI hydrogels decreased with the increasing crosslinker amount. There existed no significant difference when PEI amount was changed. The value of  $K_2$  was enlarged by lowering PVA content. The results were all comparable with the swelling data since  $K_2$  depends on many parameters like crosslinking degree, hydrophilicity of the polymeric structure [31].

In order to determine the swelling mechanism of PVA/PEI hydrogels, the following equation was used:

$$F = \frac{M_t}{M_e} = kt^n \quad (7)$$

$M_t$ : the mass of the water diffused into the hydrogel at regular time interval

$M_e$ : the mass of the water diffused into the hydrogel at equilibrium state

$k$ : the characteristic constant of network structure

$n$ : the swelling exponent which indicates the transport mode of diffusion.

Table 2 shows swelling kinetic parameters of swelling exponent ( $n$ ) and characteristic constant ( $k$ ). The  $n$  value defines the diffusion mechanism through the polymeric matrix, such as Fickian diffusion where  $n \leq 0.5$  and Non-Fickian diffusion where  $0.5 < n < 1$ . For Fickian diffusion, the relaxation rate of the polymer chains is greater than the diffusion rate of solvent molecules through the matrix [6]. The effect of Glu concentration on diffusion was investigated. Due to the increased amount of cross-linker, the  $n$  value was increased from 0.35 to 0.53. The Fickian diffusion was occurred when 30  $\mu$ L of Glu was utilized while non-Fickian diffusion was appeared with the increased amount of Glu. The reason may be the formation of more rigid structure with more cross-linker. There exist no significant effect of the amount of PEI on the diffusion type. Fickian diffusion was also observed for PVA/PEI hydrogels with varying PVA content (5%-1%). PVA/PEI hydrogels presented important potential as biomaterials since almost all of the hydrogels demonstrated a Fickian diffusion [32].

The diffusion coefficients ( $D$ ) were also calculated via the short time approximation method using the first 60% of the swelling data with the following equation:

$$\frac{M_t}{M_e} = 4 \left[ \frac{Dt}{\pi r^2} \right]^2 \quad (8)$$

where  $r$  is the radius of the hydrogels (cm). The diffusion coefficient was calculated using the plot of  $M_t/M_e$  versus  $t^{0.5}$ . It was clearly observed that the diffusion coefficient decreased

with increasing Glu amount due to the fabrication of more compact structure with the crosslinking reactions [33]. There existed no significant effect of PEI amount on  $D$  values. The decrease of the diffusion coefficients with decreasing content of PVA may be caused by the hydrolytic degradation of the hydrogels namely, PP2.5 and PP1.

Table 3. ANOVA table for  $n$  value of swelling of the hydrogels. In Table 3, it can be said that model is significant owing to the lower  $p$ -value (0.0022). The most effective parameters was found as PVA since it showed higher  $F$ -value than the other selected two parameters. The insignificant factor was found to be glutaraldehyde because of the higher  $p$ -value (0.1214). Besides, statistical indicators such as  $R^2$  and Adj- $R^2$  were calculated by design expert 7.0. The determination coefficient ( $r^2$ ) was calculated as 0.9347. This implies that model data is fitted well to experimental data. Furthermore, Adjustment  $R$ -squared was calculated as 0.8956.

**Table 2** The kinetic parameters for swelling of PVA/PEI hydrogels.

Codes	n	k	R <sup>2</sup>	D x 10 <sup>2</sup> (cm <sup>2</sup> /min)
P30	0.35	0.21	0.98	0.28
P60	0.52	0.15	0.90	0.29
P120	0.53	0.14	0.93	0.17
P30-0.5	0.50	0.25	0.97	0.29
P30-0.1	0.50	0.13	0.91	0.31
P30-0.05	0.51	0.12	0.93	0.33
PP5	0.23	0.39	0.97	0.34
PP2.5	0.18	0.47	0.95	0.31
PP1	0.19	0.50	0.97	0.23

**Table 3** ANOVA table for  $n$  value of swelling of the hydrogels.

p-value Source Prob>F	Sum of Squares	df	Mean Square	F value
Model 0.0022	0.16 significant	3	0.053	23.87
A-PEI 0.0232	0.023	1	0.023	10.43
B-Glu 0.1214	7.696E-003	1	7.696E-003	3.47
C-PVA 0.0008	0.12	1	0.12	52.29
Residual	0.011	5	2.216E-003	
Cor Total	0.17	8		

The linear equation for  $n$ -value in terms of coded factors was given in Eq. (9)

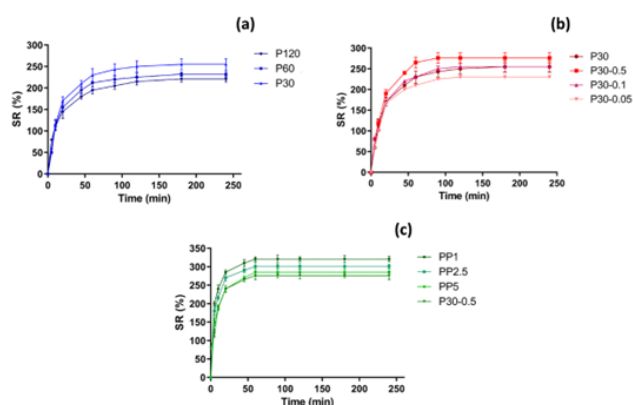
$$n = 0.35 - 0.086 * A + 0.059 * B + 0.15 * C \quad (9)$$

where  $A$ ,  $B$  and  $C$  represents PEI, Glu, PVA, respectively. The optimal conditions were found 0.05 x mL of PEI, 119.99  $\mu$ L of Glu and 10.00 of % PVA

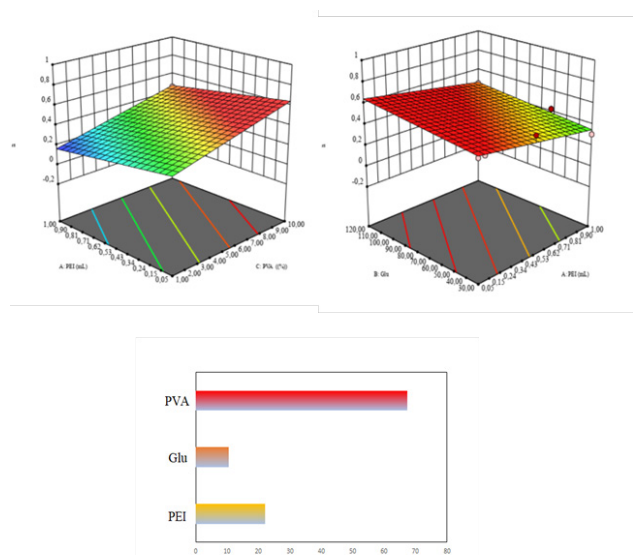
Figure 4.a depicted the binary interaction of PEI and Glu on n-value at optimal conditions. When n-value increased with increasing the amount of Glu. If the amount of PEI was increased from 0.05 to 1.0, n-value was decreased. The binary interaction effect of PEI and PVA on n-value at optimal conditions were shown in Figure 4.b. n-value increased with increasing the amount of PVA in 3-D graph. Also, Increasing PEI has reverse effect on n-value. The Pareto chart was constructed were given in Eq. (10) [34].

$$P_i = \left( \frac{\beta_i^2}{\sum \beta_i^2} \right) \times 100 (i \neq 0) \quad (10)$$

In Eq (10),  $\beta_i$  represents the coefficient of variables,  $P_i$  is the percentage of each variable, respectively. Thus, the variables affecting on the n-value (%) were depicted in Figure 4.c. The most important variable was the amount of PVA (C, 67.41%). The other variables are listed as follows: PEI (A, 22.16 %) and Glu (B, 10.43%). The least effective variable was found to be Glu (B) under  $\alpha=0.05$  significance.

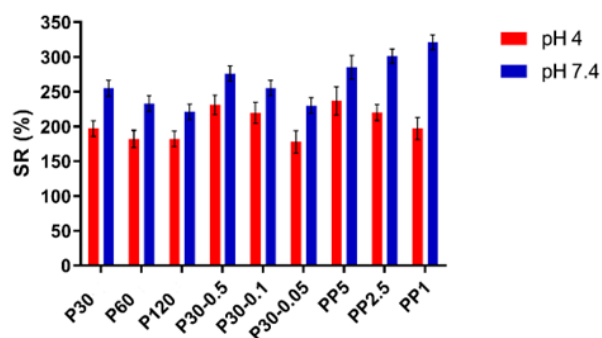


**Figure 3** The effect of Glu amount (PVA: 10%, PEI: 1 mL) (a), PEI amount (PVA: 10%, Glu: 30 µL) (b) and PVA content (Glu: 30 µL, PEI: 0.1 mL) (c) on the swelling degree of PVA/PEI hydrogels.



**Figure 4** The binary interaction effect of (a) PEI and Glu on n-value at optimal conditions and (b) PEI and PVA on n-value at optimal conditions and (c) Pareto graph for the influences of factors affecting on the n-value.

One of the major parameters of the swelling process is the pH of the biological medium. The pH of the healthy skin is in the range of 4.0-6.0 while it becomes around pH 7.4 when exposed to blood after injury [35]. pH-responsive polymers contain ionizable functional groups and thus their swelling degree is mainly based on the protonation state of these groups due to pKa values. The charged polymeric matrix facilitates the interaction of hydrogen bonding with the hydrated deposit. The prepared PVA/PEI hydrogels showed the similar trend since they swell more at pH 7.4 in comparison with pH 4 (Figure 5). PEI as a cationic polymer (pKa ≈ 7.4) bears high amount of amine groups having higher amount of protonated amine groups at acidic pH. However great number of protonated amine groups led to a significant reduction of swelling capacity by disarranging the hydrated layer. Avais and Chattopadhyay prepared PEI hydrogels via chemical cross-linking using azetidinium based crosslinker and reported pH-responsivity of the prepared PEI hydrogels with maximum swelling capacity at pH 7.4 [36]. The removal of exudate from the wound site is a crucial step for effective wound healing. The prepared PVA/PEI hydrogels swell more at pH of wound area enabling the removal of exudate from the wound site efficiently.



**Figure 5** The swelling ratio of PVA/PEI hydrogels with pH 4 and 7.4.

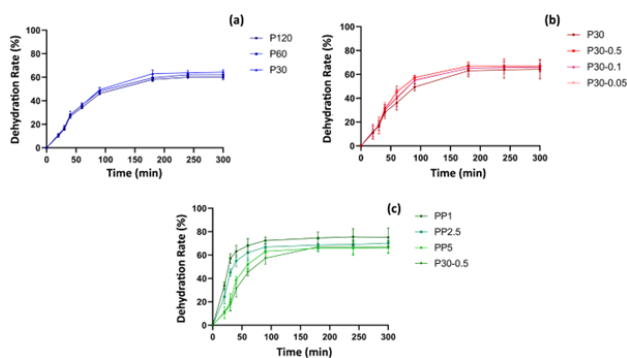
### Dehydration tests

The dehydration kinetic curves were shown in Figure 6. Firstly, water loss from the hydrogels occurred fast, then reduced slowly and finally reached an equilibrium value. The hydrogel prepared with the largest Glu amount had the lowest dehydration rate since more rigid polymeric structure inhibit the water loss (Fig 6(a)). The hydrogel synthesized with the lowest PVA content showed the biggest dehydration rate while having large pores would favor the dehydration of the hydrogel (Fig 6(c)). The bulk water molecules diffuse easily from the large pores however they can be retained as bound water via hydrogen bonding in the pores [37]. PVA has the dominant effect on the dehydration process.

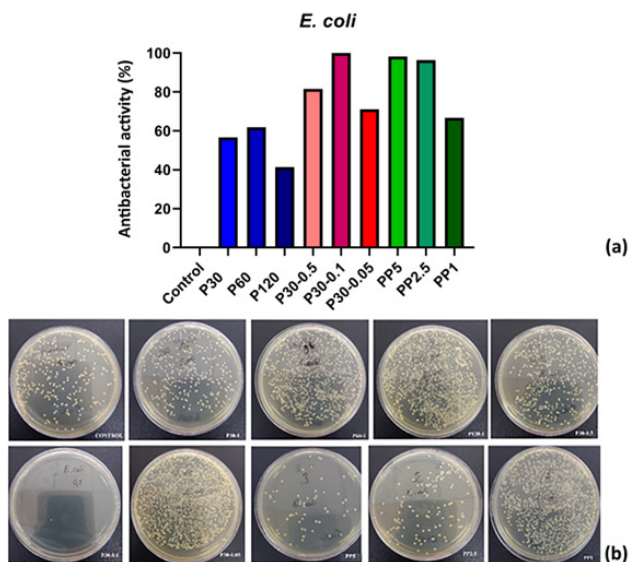
### Antibacterial activity assay

The antibacterial properties of the hydrogels were examined by colony formation counting assay and percentage antibacterial activity of PVA-PEI hydrogels against *E. coli* were given in Figure 7. PVA ratio increases in PP1, PP2.5 and PP5 hydrogels, respectively. As gelation of these hydrogels increased, PEI were expected to effectively incorporate into

hydrogel structure. The PP5 and PP2.5 hydrogels showed 98.2% and 96.3% antibacterial activities, respectively. However, antibacterial effect of PP1 was 66.7%. Antibacterial efficiency of P30, P60 and P120 hydrogels which have different Glu ratio were 66.7%, 61.8% and 41.3%, respectively. As we conclude from the percentage antibacterial activity results, antibacterial activities of PP2.5 and P30-0.1, PP5 hydrogels was over 96.4%. Thus, these three hydrogels showed excellent antibacterial activity for *E. coli*. P30-0.1 with 99.9% antibacterial activity was found as the most effective among the hydrogels containing different amount of PEI. The antibacterial performance of the antibacterial hydrogels depends on chemical and morphological characteristics of the polymeric matrix. PEI as a cationic polymer improves the antibacterial activity of the hydrogels due to the disruption of the bacterial cell membrane. The hydrogels with high porosity facilitate the interaction of the microorganisms with the polymeric structure [38].

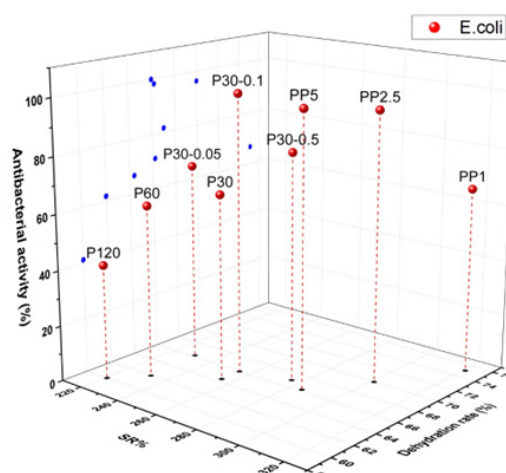


**Figure 6** The effect of Glu amount (PVA: 10%, PEI: 1 mL) (a), PEI amount (PVA: 10%, Glu: 30  $\mu$ L) (b) and PVA content (Glu: 30  $\mu$ L, PEI: 0.1 mL) (c) on the dehydration rate of PVA/PEI hydrogels.



**Figure 7** Colony formation counting assay results, (a) percentage antibacterial activities of PVA/PEI hydrogels against *E. coli*, (b) optic images of agar plates. Dilution factor: 10<sup>-7</sup> for control group and 10<sup>-6</sup> for hydrogels. Hydrogels treated *E. coli* was inoculated in agar plates and incubated for 24 h at 37 °C.

Consequently, the correlation between swelling amount, dehydration rate, and antibacterial activity was created in the 3D plot to evaluate the optimum PVA/PEI hydrogel for antibacterial wound dressing materials. It has been reported that PEI added to PVA/PEI hydrogels consists of high positive charges that are sensitive to antibacterial activity [12]. As can be seen from Figure 8, the PVA/PEI hydrogel (PP5) with the highest PEI (0.5 mL) and PVA (5%) content has significantly demonstrated the excellent antibacterial activities of the PEI in the PVA/PEI hydrogels. In addition, it is clear that the optimum PVA/PEI hydrogel as an excellent antibacterial wound dressing material is the PP5 hydrogel due to its high swelling amount, low dehydration rate and great antibacterial activity.



**Figure 8** 3D plot of PVA/PEI hydrogels against antibacterial activity, swelling amount and dehydration rate.

## CONCLUSION

The aim of this study was to investigate the swelling amount, dehydration rate and antibacterial activity performance that are crucial key parameters for wound healing applications. For this propose, PVA/PEI hydrogels were prepared via solvent casting technique with different amount Glu, PEI and PVA concentration. All of the hydrogels showed great swelling degree at pH 7.4 simulating human biological fluids and 37 °C. PVA concentration showed the dominant effect on the swelling degree as well as the dehydration results. All of the prepared PVA/PEI hydrogels demonstrated high antibacterial activity against *E. coli*. The obtained results were related with the incorporation of PEI into the polymeric structure and the morphological properties of the hydrogels. 3D plot containing swelling degree, dehydration percentage and antibacterial activity displayed the optimum hydrogel depending on these parameters. PP5 was determined as the best hydrogel with the optimized data.

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