

The influence of Ag⁺, Zn²⁺ and Cu²⁺ exchanged zeolite on antimicrobial and long term in vitro stability of medical grade polyether polyurethane

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Abstract. This study aims to investigate the limitations and applicability of different ion exchanged zeolites as antimicrobial additive in thermoplastic polyether type polyurethanes. These composites were designed to improve the health quality of hospitalized patients by expressing both biocompatibility and relevant antimicrobial activity. The zeolites were exchanged with silver, copper and zinc ions and single, binary and ternary ion-exchanged zeolite-polyurethane composites were prepared. The antimicrobial activity and the resistance of the composites against the human environment play vital role in the applicability of the materials as a medical device therefore these properties were investigated. The antimicrobial test were performed on Methicillin-resistant *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Candida tropicalis*. The tests showed that the efficiency of the silver ions is superior to the other single ionic systems. Besides, the binary and ternary ion-exchanged samples had similar antimicrobial efficiency regardless the type of the ions in the zeolite. The biocompatibility tests were carried out in-vitro in artificial body fluids for a period of 12 weeks. As a result of the in-vitro test, degradation of the composites were observed and the structural changes of the materials were detected and described by Scanning Electron Microscopy, Contact Angle measurements and Attenuated Total Reflection Fourier Transform Infrared Spectroscopy.

Keywords: biocomposites, body fluid, ion-exchanged zeolite, antibacterial, in-vitro test

1. Introduction

Polyether polyurethane (PU) is a biocompatible material that is preferably used in a variety of medical applications [1–3], e.g. as raw material for central venous catheters tracheostomy tubes and other medical devices. Although these polymers are biocompatible materials, device associated side effect

often occurs during the clinical use of the materials that can be inflammation, infection, biofilm formation and growth. This initiates the degradation of the material, e.g. previous studies have proven that the degradation of PU catheters is caused by either oxidation or hydrolysis [4–6]. The bacterial activity is responsible for the biofilm formation and growth

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and subsequent degradation of the polymeric material inside the human body. Once the biofilm has formed on the surface of the material it is difficult, or sometimes impossible, to remove it either by washing or antibiotic treatment [7, 8]. There are numerous solutions that have been presented in the literature in order to enhance the antimicrobial properties of polymeric materials e.g. antibiotics or silver containing coatings and composites [9–13]. Compared to some antibiotics, nano-silver coatings have shown superior antimicrobial properties. The antimicrobial activity of silver is based on the release of silver ions which is an ion-exchange process [14, 15]. Some studies have confirmed that the main drawback of silver is that during long term use if the released ions reach a certain concentration, silver could cause toxicity in both human and animal cells [16, 17]. Impregnation of silver ions into the surface is a safer way to ensure antibacterial properties of the material since the silver concentration is lower in this case, and the release is governed by the diffusion of the ion from the top surface layers of the material. For this application silver-sulfadiazine or other silver compounds are usually used [16–18]. Clinical studies have, however, proved that the antibacterial activity of silver-sulfadiazine impregnated catheters decreased rapidly. In addition, impregnated central venous catheters did not show a significant decrease in the number of infections compared to un-treated ones [18–20]. The aim of the antibacterial materials in clinical use is to decrease the number of infections, and thereby to improve the health of the patients, consequently reducing the healthcare costs. In addition, the production cost of these materials is almost twice that of the un-coated materials and the efficiency is questionable. Hence, clinical papers recommend the use of these materials not instead of, but in complement to un-coated materials [18–20]. It has been reported that silver ion exchanged zeolites have good antibacterial activity and therefore have a potential in the medical field to enhance antimicrobial properties of polymers. [21–23]. Since zeolite is a natural mineral, non-toxic, non-carcinogenic and has a high importance as food supplement and medical treatment agent for both humans and animals, it is completely safe to be used in medical devices [24, 25].

The mechanism of silver loaded zeolite's antimicrobial effect is similar to silver coatings and based on an ion exchange reaction. If moisture or liquid film interacts with the surface of the material, ion-exchange initiates where silver ions are exchanged to Na or other cations that can be found in the environment. The released silver ions attach to the bacteria by forming chelate complexes with the deoxyribonucleic acid (DNA) and block the transport processes in the cell [14, 15]. The applications of zeolite as filler in polymeric materials were previously reported in literature and proved that the antibacterial activity of these polymers was enhanced [26, 27]. Furthermore the effect of zeolite content on physical and thermal properties of the polymer was also examined [26, 27]. By increasing the silver-zeolite content in the polymer an increased antimicrobial activity was observed (due to the higher silver ion concentration) [22, 28] but, depending on the application the zeolite content may influence physical, thermal and/or chemical properties. There are two main reasons to exchange silver as filler in any kind of antimicrobial agent or composite. The first is the current European Union (EU) regulations that aim to ban silver products from the medical industry due to the increased risk to toxicity, while the second is based on economical issues due to the high price of silver products. The current study aims to investigate the potential of other metallic ions containing zeolites as antimicrobial agents that may replace the silver. This paper presents the preparation and manufacture of PU filled with silver and alternatively copper, zinc and multi-ion exchanged zeolite in order to determine whether the combination of these ions can lead to the elimination of silver. The influence of the presence of different ions in the zeolite on the antimicrobial activity of polyurethane composites was investigated. Furthermore, as a key requirement of the invasive medical applications, *in-vitro* tests were performed in two artificial body fluids in order to monitor the long-term stability of the materials, and to determine how the degradation was influenced by the different ions and the zeolite content. Contact angle measurements, Fourier-transform infrared spectroscopy (FTIR) and scanning electron microscopy (SEM) were used to monitor the changes in surface and chemical properties. In

addition, swelling and ion release measurements were performed to determine diffusion coefficients and the mechanism of zeolite diffusion from the bulk to the surface.

2. Experimental

2.1. Materials

BASF Elastollan 1164D50 polyether type thermoplastic polyurethane (PU) was supplied by TRACOE medical GmbH (Frankfurt, Germany). A type zeolite (ZEOMIC AJ10D) was purchased from Sinanen Zeomic Co., Ltd. (Tokyo, Japan). The particles size was typically 2.5 μm . Silver-nitrate (purum p.a., crystallized, $\geq 99.0\%$), zinc nitrate hexahydrate (purum p.a., crystallized, $\geq 99.0\%$), copper nitrate trihydrate (purum p.a., 98.0–103%) sodium sulphate (purum, anhydrous, $\geq 99\%$), magnesium chloride (purum, anhydrous, $\geq 98.0\%$), sodium chloride (purum p.a., $\geq 99.5\%$), sodium hydroxide (purum p.a., $\geq 97.0\%$), sodium phosphate dibasic (purum p.a., anhydrous, $\geq 98.0\%$), potassium chloride (puriss. p.a., $\geq 99.5\%$) and calcium chloride dihydrate (purum p.a., 99–103%) were purchased from Fluka (Stockholm, Sweden). Sodium acetate (puriss. $\geq 99.0\%$, anhydrous), sodium bicarbonate (powder, $\geq 99.5\%$), citric acid (anhydrous, $\geq 99.5\%$), sodium citrate dihydrate (puriss. $\geq 99\%$), glycine (puriss. $\geq 99\%$), sodium L-tartrate dibasic dihydrate (ACS reagent, $\geq 99\%$) sodium L-lactate ($\sim 98\%$), and sodium pyruvate (SigmaUltra, $\geq 99\%$) were purchased from Sigma Aldrich (Stockholm, Sweden). Methicillin-resistant *Staphylococcus aureus* (MRSA) ATCC 43300, *Pseudomonas aeruginosa* ATCC 27853, *Candida tropicalis* ATCC 90874 were supplied by the Hungarian National Public Health and Medical Officer Service, Miskolc, Hungary.

2.2. Sample preparation

Single, binary and ternary ionic zeolites were prepared with silver, copper, zinc and the variation of these ions. In order to make the antimicrobial effectiveness of the different ions comparable, similar ion concentrations needed to be obtained in the zeolite structure. The ion absorption reactions were performed according to the following protocol: 1 g zeolite powder was added to 40 ml metallic nitrate solution. The reactions took place at room temperature for 24 hours under constant stirring then the samples were washed 5 times with distilled water to

Table 1. Ion content of the different zeolite samples

Sample/Ion	Ag	Cu	Zn
	[wt%]		
Ag	13.21 \pm 2.13		
Cu		9.25 \pm 0.32	
Zn			18.38 \pm 2.26
AgCu	12.60 \pm 1.30	10.29 \pm 0.69	
AgZn	15.54 \pm 1.51		13.37 \pm 0.88
CuZn		8.43 \pm 0.22	7.39 \pm 0.21
AgCuZn	11.94 \pm 0.38	9.30 \pm 0.28	5.23 \pm 0.11

remove the excess nitrate. This was followed by drying at 105°C. In case of single ionic systems 0.025 M silver-, 0.05 M zinc- and 0.1 M copper-nitrate was used in order to achieve fairly equal concentrations of each ion in the zeolite structure. For binary and ternary systems the variations of the different metallic solutions were mixed, however the concentration of each ion in the solution was kept at the same value as it was for the single ionic systems. The final ion concentration in the zeolite samples is presented in Table 1. The EDX results confirmed that similar ion concentration was reached in the binary and ternary ion exchanged zeolites by the applied solution concentrations. Altogether seven different samples were prepared: zeolites filled with Ag, Cu, Zn ions and the combination of these ions (Ag-Zn, Ag-Cu, Zn-Cu, Ag-Zn-Cu).

The second step was the incorporation of the zeolite samples into the PU. 1% of zeolite was mixed with PU pellets in a Haake PolyLab System equipped with a Rheomix 600 mixer (Thermo Fisher Scientific Inc., Karlsruhe, Germany). The mixing temperature was 210°C at 100 rpm and the duration of constant mixing was 10 minutes. Besides ion filled zeolite, 1–5% unfilled zeolite was also mixed in PU in order to monitor the influence of the zeolite content on the polymer's stability. After the composites were prepared, zeolite-containing PUs were ground into pellets and injection moulded into flat 100 \times 100 \times 1.5 mm plate with a Krauss Maffei 50-390/C1 type injection molding machine (KraussMaffei Technologie GmbH, München, Germany). Pure PU samples were also prepared and used as a reference sample for the experiments.

2.3. Antimicrobial test

The antimicrobial effect of zeolite-PU composites was determined for two bacterial strains (Methicillin-resistant *Staphylococcus aureus* (MRSA)

ATCC 43300, *Pseudomonas aeruginosa* ATCC 27853) and one mold (*Candida tropicalis* ATCC 90874) according to the ISO 22196 standard. Cell suspensions were prepared from each strain with a cell concentration of $5 \cdot 10^5$ CFU/mL. Then the suspension was inoculated onto the surface of the composite samples and the cells were allowed to grow on the surface for 24 h at 37°C and 98% relative humidity. The dimension of the test pieces was 50×50 mm. After the exposure the suspension was washed from the surface and mixed with agar that was deposited on petri dishes. This mixture was let to get solidified and were placed into growing ovens at 37°C. After 24–48 hours the number of the grown colonies in the agar plate was counted.

2.4. In-vitro degradation test

Two synthetic body fluids were chosen to simulate different in vivo conditions. Both fluids have been identified to cause degradation of plastic implant materials [29, 30]. Artificial Lysosomal Fluid (ALF) simulates the immunological reaction of the body while the Gamble's solution is similar to the interstitial fluid of the deep lung. The chemical composition of the two test media was previously described [30].

The pure PU and zeolite-PU composites were placed into acid-cleaned (1% HNO₃ for 24 h) glass vessels which were filled with 10 ml of either ALF or Gamble's solution, and thoroughly sealed with Parafilm. Subsequently, the samples were placed in an incubator and stored under static and dark conditions at 37°C during 84 days. Samples were withdrawn from the solution on a weekly basis, and analyzed by scanning electron microscopy (SEM), contact angle (CA) measurements and FTIR spectroscopy. Since the different ions in the zeolite structure may be reduced to oxides (especially silver) on contact with water and cause discoloration of the samples, the colour change was also monitored throughout the whole experiment.

2.5. Diffusion and swelling

During the in-vitro ageing of the materials in ALF and Gambles solution, the liquid uptake of the samples was determined on short and long term. Sampling was performed after 1, 2, 3, 4, 5, 6, 8, 12, 24, 32, 48, 60, 72, 96, 120 and 144 hours in the short term test, while weekly sampling was done during

the long term test that lasted 12 weeks. The liquid uptake was determined by weight measurement and calculated by Equation (1).

$$WA [\%] = \frac{m_v - m_k}{m_k} \cdot 100 \quad (1)$$

where WA [%] is the absorbed liquid uptake in weight percentage, m_k is the initial weight and m_v is the weight after sampling of the samples.

The diffusion coefficients were calculated from the fluid absorption curves. These curve had two sections that can be clearly distinguished (Figure 1). The first section follows a Fickian characteristic ($\sqrt{h} \leq 7$), where the amount of the absorbed liquid increases linearly as a function of exposure time. The second section is parallel to the X axis, which means that the amount of the liquid remains constant in the composite and indicates the saturation of the material.

This second section is the saturation level, which is marked by M_∞ in the literature. The time that is necessary for the saturation is t_s . This was determined at 95% saturation and the diffusion coefficients of the liquid into the polymer phase was calculated by Fick's second law (Equation (2)) from the linear section of the normalized absorption curves (Figure 1):

$$\frac{M_t}{M_\infty} = \frac{4}{L} \cdot \left(\frac{D}{\pi}\right)^{0.5} \cdot t^{0.5} \quad (2)$$

where M_t is the absorbed liquid amount at a certain time, M_∞ is the maximum absorbable liquid amount, L is the sample thickness and t is the time.

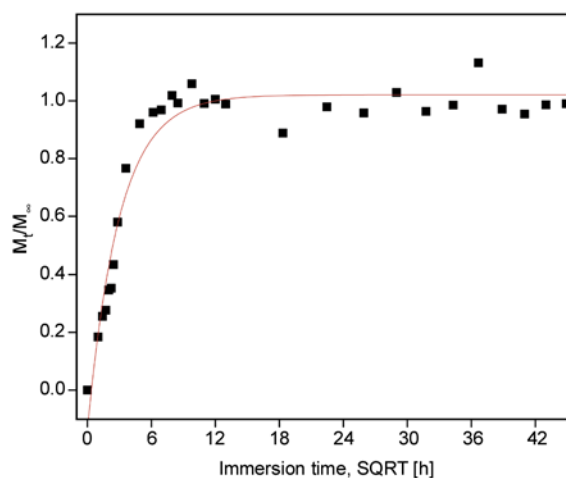


Figure 1. The method to determine the diffusion coefficient from the absorption plot

2.6. Contact angle measurements

The contact angle measurements were performed on a CAM 200 instrument from KSV Instruments Ltd. (Helsinki, Finland). MilliQ grade water from a Synergy 185UV Ultrapure water system (18.2 M Ω -cm, Millipore AB, Solna, Sweden) was used. The contact angle was measured on a reference and all exposed samples, the average value of three pieces cut from the same sample was calculated. Five 3 μ l droplets were analyzed on each piece, i.e. a total number of 15 droplets, and ten images were taken for each droplet at 100 frames/s.

2.7. Attenuated Total Reflection Fourier Transform Infrared Spectroscopy (ATR-FTIR)

The sample surfaces were analysed using a Spectrum 2000 FTIR spectrometer from Perkin Elmer (Wellesley, MA, USA) equipped with a Golden Gate single-reflection accessory for Attenuated Total Reflection (ATR). Triplets of the samples were analysed, 16 scans per piece between 4000 and 600 cm^{-1} at intervals of 1 cm^{-1} with a resolution of 4 cm^{-1} . All the spectra were normalized to 1.5 cm^{-1} and average results are presented.

2.8. Scanning electron microscopy (SEM)

SEM micrographs from each sampling area at the different exposure times were taken with a Hitachi S-4800 Ultra-High Resolution Field Emission Scanning Electron Microscope (FE-SEM) (Hitachi High Technologies Europe GmbH, Krefeld, Germany) at various magnifications. The samples were coated with gold/palladium.

3. Results and discussion

3.1. Antimicrobial activity

The results of the antimicrobial test are presented in Figure 2. The reference sample was the pure PU that did not contain any zeolite particles. It was observed that this sample had no antimicrobial effect, all of the microorganisms started to grow intensively on the surface. After 24 hours exposure the number of viable cells increased approximately one order of magnitude for MRSA and *Candida tropicalis* while for *Pseudomonas aeruginosa* this value was two times higher than the initial cell concentration ($5 \cdot 10^5$ CFU/ml). In addition, the number of viable MRSA cells also increased slightly on the

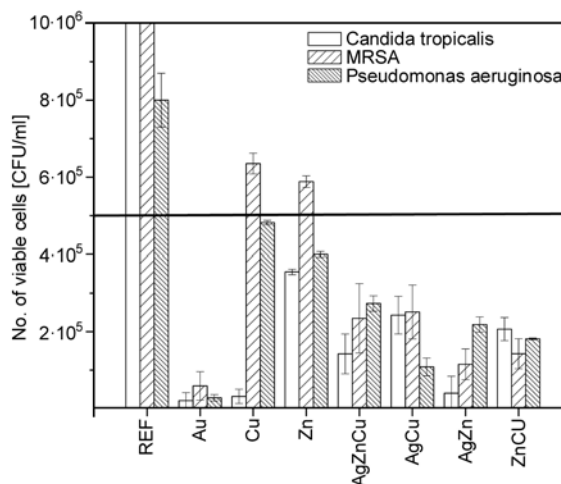


Figure 2. The number of viable cells on the surface of the different ion-filled samples after 24 hours exposure

surface of samples that contained only copper or zinc zeolite. In contrast these samples showed antimicrobial or growth inhibition effect against *Ps. aeruginosa* and *C. trop.* Both copper and zinc containing samples decreased the number of viable cells for *Ps. aeruginosa* slightly, while copper decreased the number of *Candida tropicalis* cells significantly. Microorganisms have different tolerance against antibiotics and antimicrobial material. This good activity of the copper could be explained by the low resistance of *C. tropicalis*. Binary and ternary ionic zeolite samples showed similar antimicrobial activity against all three microorganisms. This is due to the ion distribution of in the zeolite. It was determined during previous experiments that in binary and ternary zeolite systems the different ions may block each others' way and therefore highly influence each others' release. Since silver was superior to all of the other samples. It can be assumed that in silver containing multi-ionic systems the silver ion could be the responsible for the increased antimicrobial activity compared to single ionic copper and zinc zeolite. Although, these two ions did not express significant antimicrobial activity, the binary zinc-copper system decreased the number of viable cells with the same magnitude as the silver containing multi-ionic systems. This increased efficiency could be due to joint activity of copper and zinc. It can be assumed that each of these ions initiate different toxic mechanism within the cell, therefore the duplex system can be more harmful for the cells than single ones.

3.2. Diffusion and swelling

The mechanism of the antimicrobial effect of zeolite-PU composites is based on the diffusion of the ions from the bulk to the surface where they interact with microbial cells. Water uptake is a key factor since it can influence the diffusion of the zeolite and ions in a positive way. Besides, the type of the ion may also influence the diffusion rate of zeolite, which is based on the ion activity. Since zeolites are known to be hygroscopic materials, the zeolite content in the composites may result in an increased water uptake. Figure 3 presents the water uptake of zeolite-PU composites with increasing zeolite content as a function of exposure time in ALF and Gamble's solution.

It can be determined from the water uptake plots that the increasing zeolite content has influence on the rate of the water absorption and slightly increased the amount of total absorbed solution. During the short-term experiment all the composites became saturated with the aging solutions. The rate of absorption of ALF and Gamble solution did not increase at 1 to 3% zeolite content, while at 4 to 5% the rate increased for both ageing solutions.

The calculated diffusion coefficients and maximum absorption values are presented in Table 2. The absorption plots and the measured values show that with the increasing zeolite content the absorption slightly increased in ALF-solution while it did not change in Gamble's solution. Based on this it can be assumed that besides temperature, molecular structure and sample thickness, the diffusing liquid also influences the absorption. The diffusion coefficients show increasing tendency as a function of zeolite content. This is also obvious from Figure 2

Table 2. The maximum solution absorption capacity and diffusion coefficients of the PU-zeolite samples in ALF and Gamble's solution

Zeolite content [wt%]	ALF		Gamble	
	WA [%]	D [m^2/s]	WA [%]	D [m^2/s]
0	2.21	$4.01 \cdot 10^{12}$	2.23	$4.31 \cdot 10^{12}$
1	2.28	$3.71 \cdot 10^{12}$	2.21	$4.10 \cdot 10^{12}$
2	2.36	$3.61 \cdot 10^{12}$	2.54	$3.92 \cdot 10^{12}$
3	2.33	$3.51 \cdot 10^{12}$	2.5	$2.82 \cdot 10^{12}$
4	2.41	$3.46 \cdot 10^{12}$	2.53	$2.72 \cdot 10^{12}$
5	2.45	$1.25 \cdot 10^{11}$	2.53	$2.26 \cdot 10^{11}$

since the time that is needed for the saturation decreased as a function of zeolite content. The diffusion coefficients were compared with the literature data and showed a good match.

3.3. SEM study

Different surface changes were observed during the in vitro degradation of PU composite samples in artificial ageing solutions. Pure PU samples showed surface changes as a function of time in both ALF and Gamble solution (Figure 4). As a result, the surface roughness increased and smaller degradation signs (cracks, holes) appeared on the surface. Some of these changes can originate from manufacturing and preparation of the samples. In addition, high amount of salt residues were also found on the surface of Gamble aged samples.

On the surface of zeolite-PU composites obvious degradation signs were identified (Figure 5). An increasing amount of small holes were observed as a function of time and zeolite content on the surface of ALF exposed samples. These holes started to appear after 1 month and their formation increased intensively during the whole exposure. Since the

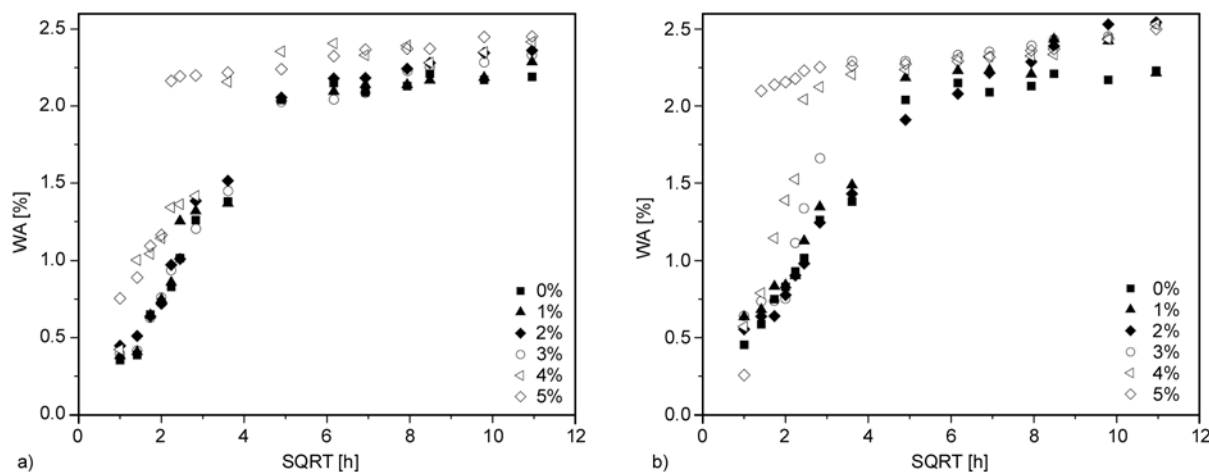


Figure 3. The water uptake of PU-zeolite samples as a function of immersion time in ALF (a) and Gamble's (b) solution

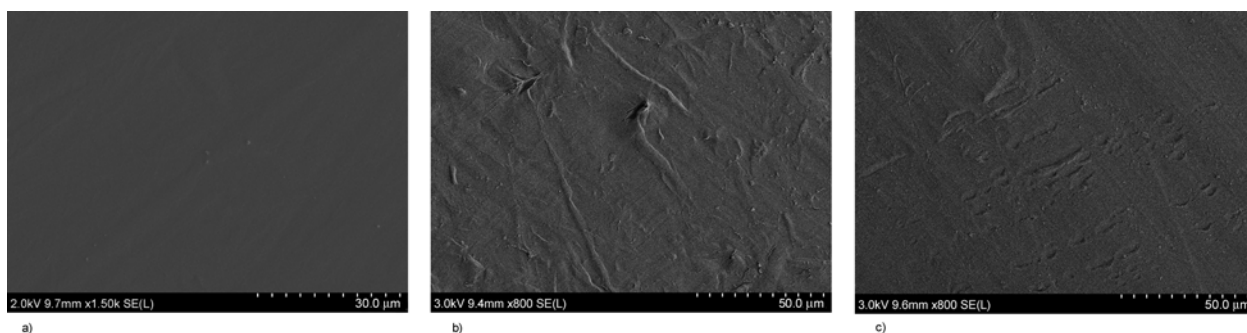


Figure 4. SEM micrographs of the surface of unaged pure PU (a) and aged in ALF (b) and Gamble's (c) solution pure PU samples for 3 months

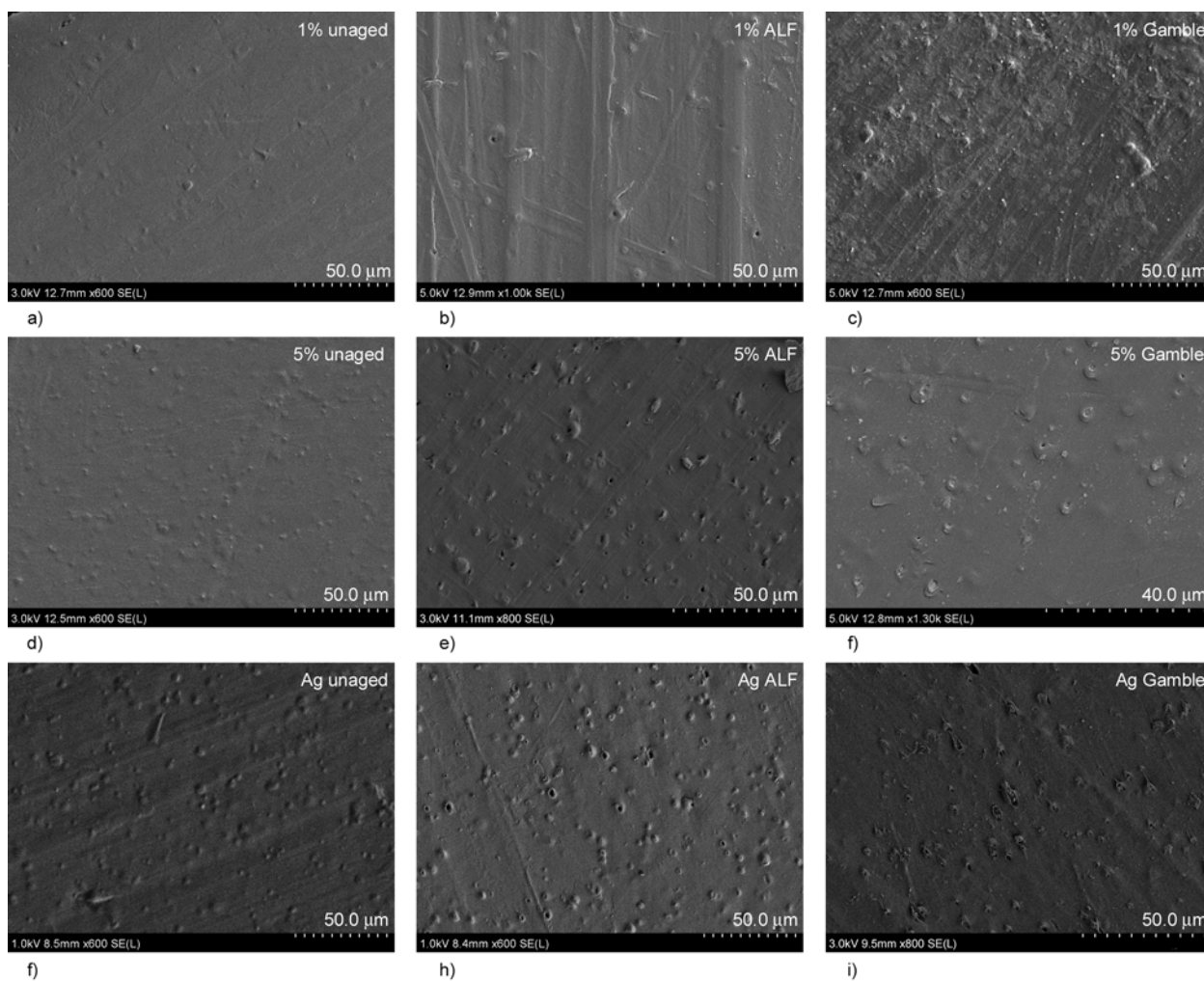


Figure 5. SEM micrographs of the surface of unaged, ALF and Gamble exposed 1% (a, b, c), 5% zeolite-PU (d, e, f) and silver filled zeolite-PU (g, h, i)

dimension of the holes is between 2–4 microns, it is probable that zeolite particles diffused out of the materials by leaving such a cavity behind. The highest amount of cavities was found on the surface of samples that contained Ag ions and 5% zeolite. This can be explained with the high ion-exchange activity of silver in zeolites since it is known that silver exchanges easily with other ions in the envi-

ronment. Therefore besides the concentration gradient, the high activity of silver may increased the diffusion of zeolite to the surface. This surface alteration was seen on non-silver containing composites and also on the surface of zeolite-PU composites that were not filled with any ions. This indicates that apart from the activity of the ion, the zeolite itself is capable to diffuse to the surface of the polymer and

migrate out in order to form equilibrium between the bulk and the environment.

In case of Gamble aged samples the previously mentioned cavities were found irregularly on the surface of different samples. Their shape differed from each other. In addition, high amount of salt remained on the surface after cleaning. The reason for this could be that the different salts from the Gamble solution interacted with the surface of the polymer and bonded to the surface structure or reacted with the ions of the zeolite on the surface and stuck there afterwards. This salt layer can highly influence the ion and zeolite release and the hydrophobicity of the composites.

3.4. *In-vitro* biocompatibility test

Contact angle measurements were performed on multi-ionic systems and on composites with increasing zeolite content. Figure 6 represents the contact angle change of the pure PU as the reference and ion-exchanged zeolite-PU samples as a function of exposure time in ALF (Figure 6a) and Gamble's solution (Figure 6b).

The contact angle of pure-PU slightly increased in the beginning and after 2 weeks it remained constant and did not change during the aging process in any of the solutions. This phenomenon is believed to be caused due to the interaction between the polymer surface and the aging solution and as a result hydrophobic groups formed on the surface.

Initially the contact angle of these materials started to increase similarly to the pure PU and reached a maximum value after 4–6 weeks. Then it started to decrease moderately and after 12 weeks exposure

the contact angle of the samples aged in ALF solution decreased back to the initial contact angle or to a slightly lower value. This change in hydrophobicity was similar in case of the most ions; however the contact angle change of Cu containing sample showed similar characteristic to the pure PU. After it reached a maximum value, it remained constant through the whole duration of the experiment. In Gamble's solution similar change was observed, but after the contact angle reached the maximum value, a more intense decrease was seen and after 12 weeks the contact angle value of all samples became lower than the initial. By comparing the results of the different ions, the Zn and multi-ionic system Ag-Zn-Cu decreased the most, the contact angle of the surface became lower than 90° which is the value where a hydrophobic surface becomes hydrophilic. Based on the results it can be assumed that the change in contact angle as a function of exposure time was ion dependant.

In Figure 7, the contact angle change of samples with increasing zeolite content is presented as a function of exposure time. These samples didn't contain any ions. Few weeks after the immersion, the contact angle started to increase. This was seen before in case of multi-ionic samples. It reached a maximum value around 4–6 weeks and started to decrease intensively. Compared to the ion-filled zeolite-PU samples, the change of contact angle during the exposure was bigger in these samples that did not contain any ions. The different ions may influence the zeolites hygroscopic behaviour and water absorption as well. This contact angle decrease was slightly influenced by the zeolite content, and

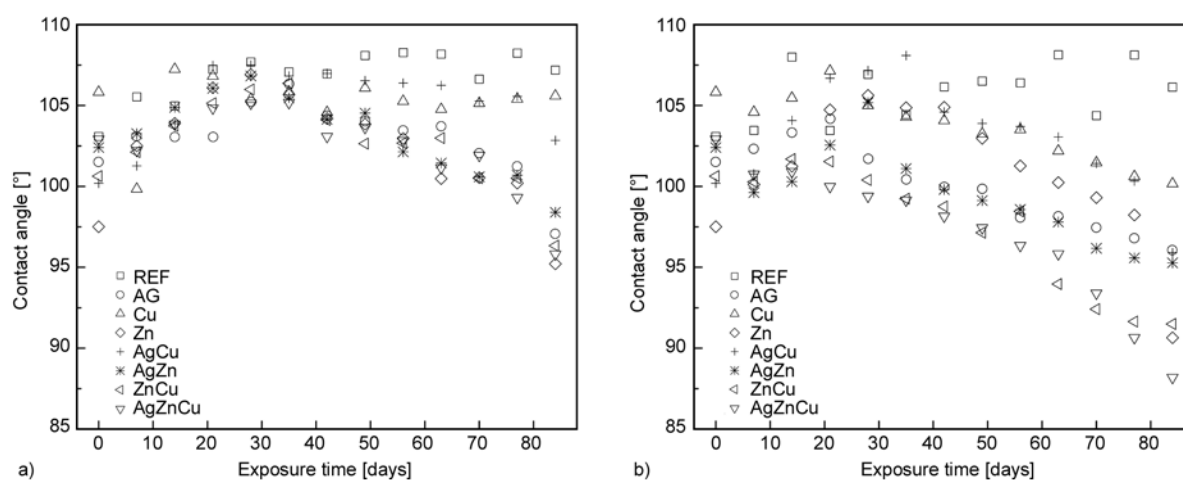


Figure 6. The contact angle change of different ionic PU-zeolite samples as a function of exposure time in ALF (a) and Gamble's solution (b)

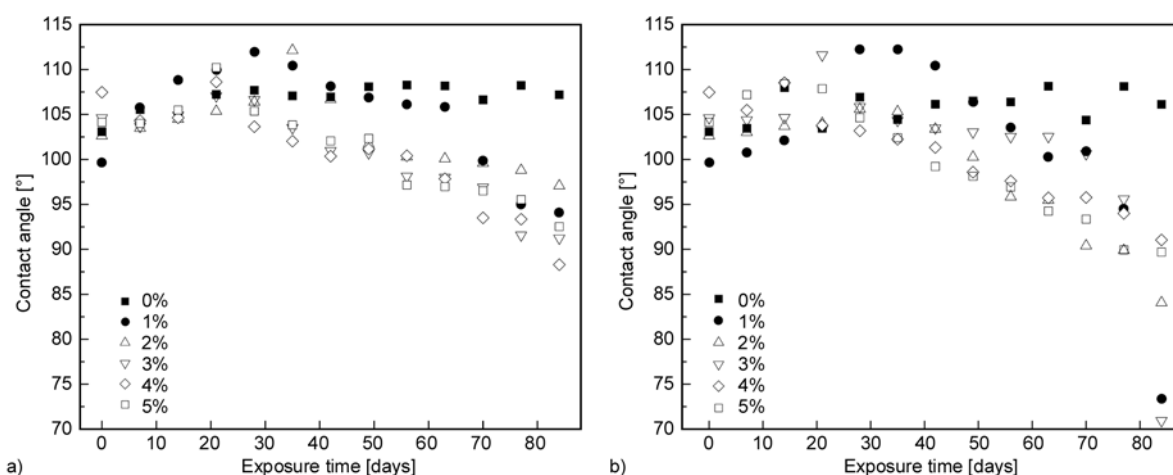


Figure 7. The contact angle change of PU-zeolite samples with increasing zeolite content as a function of exposure time in ALF (a) and Gamble's solution (b)

after 8 weeks exposure these values became lower than the initial and decreased significantly in the last period of the experiment. The contact angle of the samples decreased to slightly lower than 90° in ALF solution, which was observed in multi-ionic systems as well. In Gamble's solution the contact angle change was similar to ALF solution, however in the last weeks of the exposure the contact angle dropped significantly and decreased to $70\text{--}80$ degrees in some cases.

PU absorbs moisture and zeolite is highly hygroscopic, it was shown that the water absorption of the zeolite-PU composites is increasing with the increased zeolite content. This water uptake is a reasonable explanation for the significant decrease in the contact angle, since more aging solution can reach the bulk and initiate degradation processes. However, for the initial contact angle increase, a change in the chemical structure of the surface can be a good explanation. In order to describe this phenomenon, FTIR analysis was performed on the samples weekly during the aging experiment. In Figure 8 the region of N-H (3445 cm^{-1}) and CH₂ ($2854, 2933\text{ cm}^{-1}$) stretching is presented. Up to 4–6 weeks of exposure the area of CH₂ increased in case of most of the samples and decreased afterwards. The most significant increase of CH₂ was observed on the FTIR spectrum of pure-PU. In addition the area of CH₂ for this sample remained constant after the initial increase. This region of CH₂ represents the bonds of polyether and cyclohexane and the increase indicates the formation of these compounds. It is known that cyclic alkanes are hydrophobic. This may be the reason for the ini-

tial contact angle increase in the samples. The formation of these compounds started right after one-week exposure and reached a maximum level within a few weeks. This observation matches well with the contact angle results of zeolite-PU samples, where the maximum value was also reached in 4 weeks and then a decreasing tendency was observed.

In addition, the peak at 1360 cm^{-1} , which corresponds to the wagging of CH₂ increased until 8 weeks and then started to decrease (Figure 9). After 10 weeks exposure the characteristic of the peak changed and started to form two different peaks. Besides CH₂, the formation of –OH groups was also detected in the region $3400\text{--}3600\text{ cm}^{-1}$. In ALF solution this peak started to form after one week and increased slightly as a function time. The formation of –OH groups is a consequence of the water uptake and indicates oxidative degradation of the soft segment. The bond scission in the soft segment occurs on the methylene of the ether group by the formation of hydro-peroxide (3388 cm^{-1}) [3]. In Gamble solution apart from the pure PU the –OH peak broadened and formed an overlap with the N-H region after four weeks exposure. It is confirmed by the literature that the N-H region overlaps with the region of the –OH groups [2, 4, 31, 32]. This alteration of the FTIR spectrum indicates oxidative degradation and the formation of new hydrogen bonds [3, 5, 33]. During the oxidative degradation of the hard segment bond scission takes place on the urethane linkage, which indicates the broadening of the N-H region and is due to the formation of primary amines. In addition, since the carbonyl groups are attached to the N-H group by hydrogen bonds in the

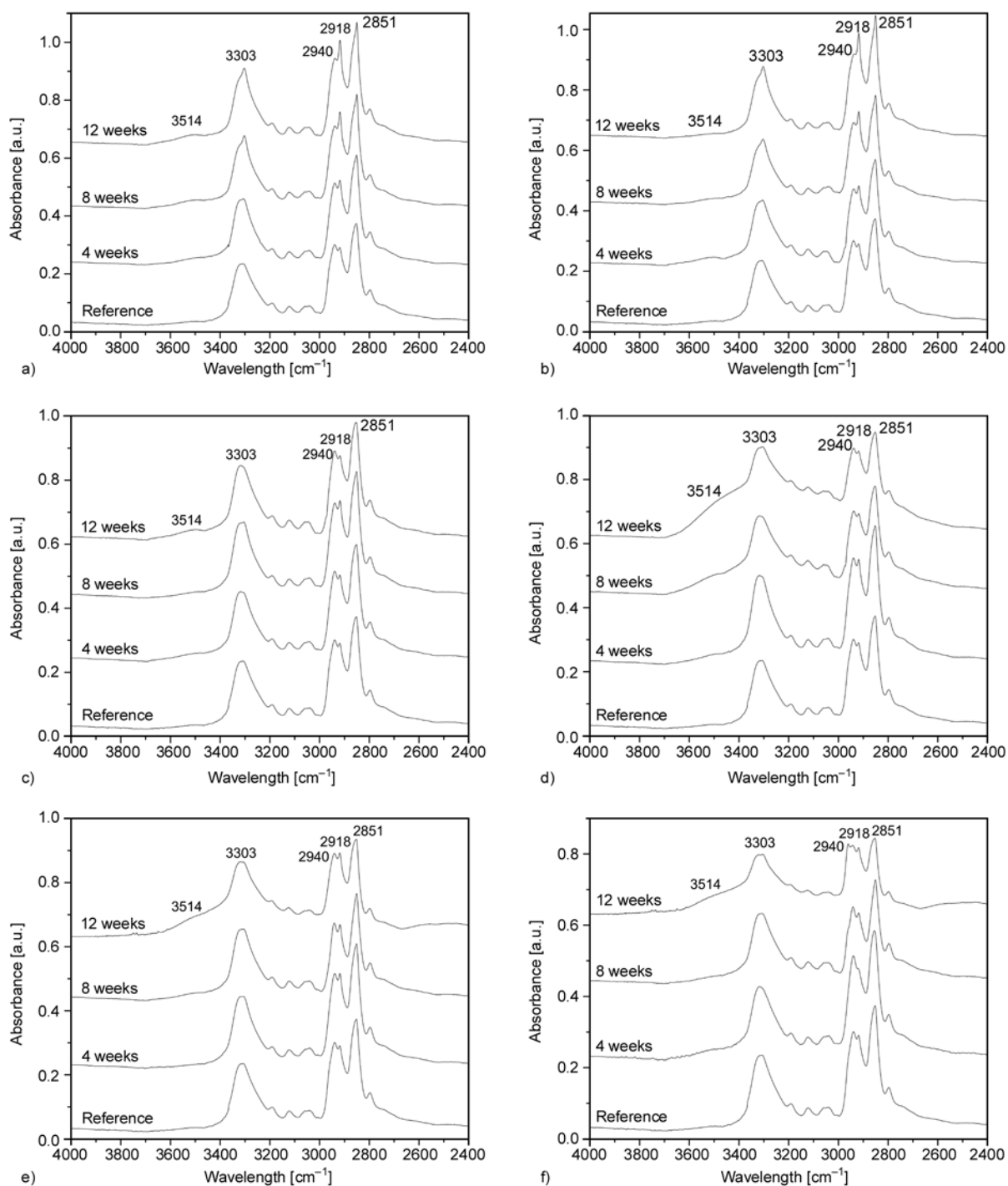


Figure 8. The FTIR spectrum of the N–H and CH₂ (4000–2400 cm⁻¹) region of pure PU (a–b), 1% (c–d) and 5% zeolite-PU (e–f) in ALF (a, c, e) and Gamble's solution (b, d, f)

polymers hard segment, the formed shoulder between the N–H and –OH group can also correspond to the increase of this bond. Since the N–H groups overlap with the –OH, the formation of hydrogen bonds can only be monitored by the carbonyl ratios.

The degradation of the hard segment was confirmed by the formation of a new small peak at 1637 cm⁻¹ after 1 week exposure. This peak slightly increased

as a function of exposure time. The peak is assigned to the free aromatic amine and correlated to the degradation of the hard segment [34]. Besides, in case of zeolite content higher than 3% in Gamble's solution the C–O–C asymmetric stretching of polyether (1103 and 1070 cm⁻¹), urethane (1017 cm⁻¹) and the asymmetric ring stretching of the cyclohexane increased significantly [3]. This gives evi-

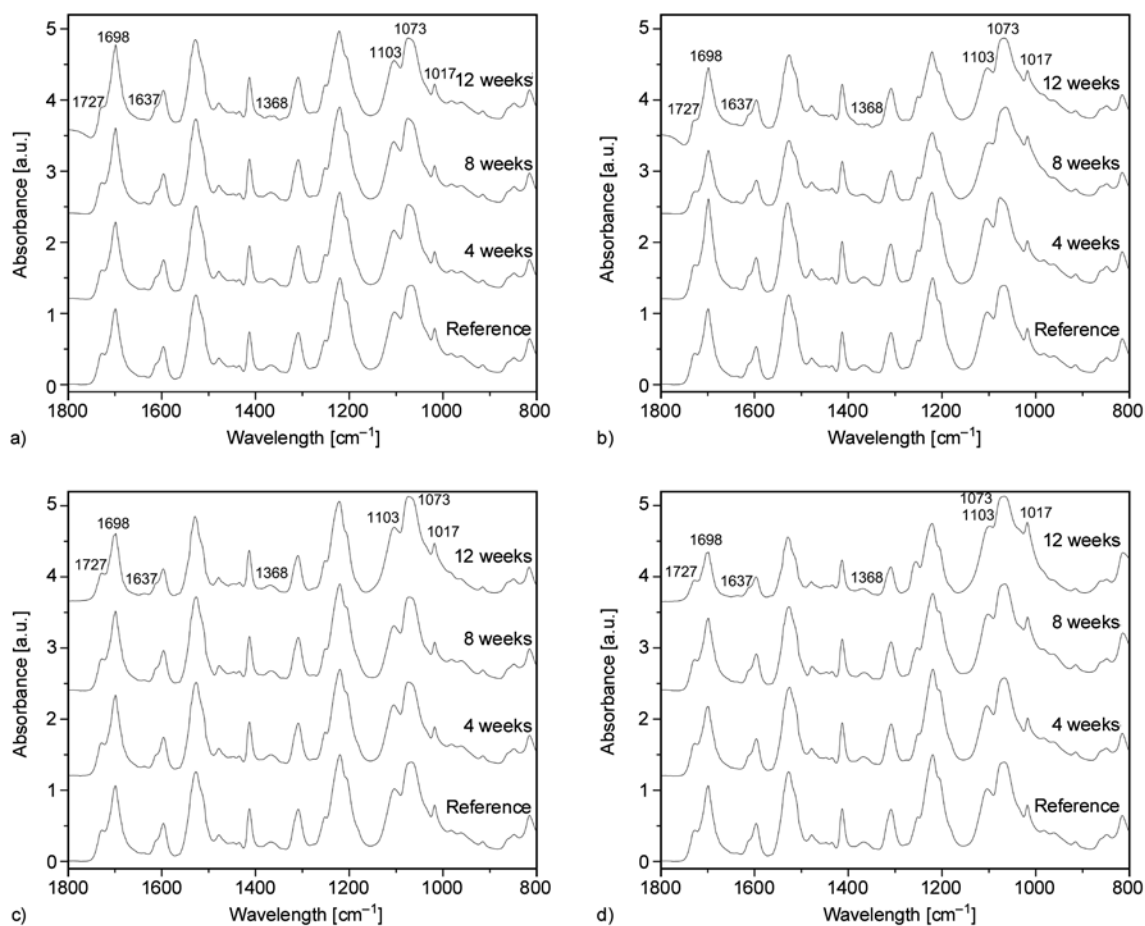


Figure 9. The FTIR spectrum of 1% (a–b) and 5% zeolite-PU (c–d) in ALF (a, c,) and Gamble's solution (b, d) in the region 1800–800 cm^{-1}

dence that oxidative degradation and bond scissions occurred in both the hard and soft segment with increased functionality of ether and aromatic groups. In general the intensity of this change in the chemical structure was the same for pure PU and zeolite-PU up to 3% zeolite. Above this amount more intense alteration of the FTIR spectrum was observed [4]. This is due to the increased solution uptake of the composites, which results an increased degradation rate. It was observed that the different ageing solutions caused the same type of degradation of the composites.

4. Conclusions

It can be concluded that the increasing zeolite content in the polymer resulted an increased water uptake and diffusion coefficient. This may influence the diffusion of the zeolite and the different ions within the polymer matrix. The antimicrobial tests showed that the efficiency of the silver ions is superior to the other single ionic systems. The

binary and ternary ion-exchanged samples had similar antimicrobial efficiency. This is due to the ion release and the ion distribution in zeolite. In these systems the different ions may block each others' way that influences the ion release and therefore the antimicrobial properties.

During the ageing of the polymer composites, significant surface changes were observed. The surface of the samples became rough and small holes were detected. These holes were formed due to the zeolite that diffused out of the matrix.

Besides topography, changes in the contact angle first showed increasing tendency up to a couple of weeks and then decreased. This change is thought to be due to the degradation and the complex chemical changes in the structure. The FTIR results confirmed that chemical changes occurred in the materials during exposure to both ALF and Gamble's solutions. It was confirmed that oxidative degradation of the hard and soft segment of PU occurred during the in vitro exposure. In addition the forma-

tion of hydrogen linkages and small amount of cross-linking was also observed as a result of the degradation. The zeolite content has shown to have an influence on the degradation in case of both aging solutions. This can be linked to the increased solution uptake of the composites.

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