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Tracking of the physical ageing of amorphous pharmaceutical polymeric excipients by positron annihilation spectroscopy

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Abstract

The structural changes observed in several amorphous polymers, commonly applied in the pharmaceutical technology to ensure conventional or controlled drug release, during relatively short storage periods are illustrated. The results suggest that the apparent structure formation of polymer molecules with water and, possibly, with other additives plays a significant role in the formation of such important physical and chemical parameters of tablets as drug release rate and solubility. Positron annihilation lifetime spectroscopy (PALS) was used to detect the changes of the free volume in the studied polymers under different storage conditions. Positrons react to the structural changes of amorphous polymers very sensitively, so the method can be recommended as useful means for stability tests during the development phase of dosage forms containing such excipients.
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1. Introduction

Knowledge of the release mechanisms and the properties of excipients is the key to developing different types of oral slow and extended release products, even with pulsatile release. When choosing a controlled release system, the important aspects include the ease of manufacturing, the reliability (i.e., the stability against the effects of motility, rate of stirring, pH of the stomach/intestines, and food) and the stability of the release rate during long storage periods. Among the excipients of these systems, the amorphous polymers are frequently used in different dosage forms. Most of these applications require long-term stability but, as amorphous polymers are not in equilibrium below their T_g, these polymers usually undergo spontaneous, however slow, transformations towards low-energy equilibrium states [1]. This so-called physical ageing is usually manifested also in volume and enthalpy relaxation indicating serious structural changes in the material. It has been repeatedly proven that the ageing of amorphous polymers is controlled by the type and the rate of their characteristic molecular motion [2,3]. The enhanced molecular mobility, caused by the plasticization effect of absorbed water, has been proposed to be the major underlying factor in chemical and physical instability of amorphous pharmaceutical materials [4–6].

Positron annihilation lifetime spectroscopy (PALS) is a unique method since it is exceptionally sensitive to the free volume. It is frequently used to determine the size distribution of free volume holes in polymers [7,8]. All of these measurements are based on the interaction of the free volume holes and the so-called *ortho*-positronium atom. When a positron meets with its particle counterpart, they annihilate

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and provide information on the surroundings of the annihilating pair. As the probability of such a meeting depends on the electron density in materials, positrons are exceptionally sensitive to free volumes, i.e., to the variation of electron density. In polymers, a large fraction of the injected positron forms a bound state with electrons before their annihilation. One of the bound states, the *ortho*-positronium atom or *o*-Ps, has a "long" lifetime: it lives for 1–10 ns in polymers. This lifetime is long enough for positronium atoms to scan their surroundings and, fortunately, it is long enough to be observed easily. Moreover, according to a simple model, the lifetime of an *o*-Ps atoms depends on the size of the free volume in which it is located:

$$\tau = \frac{1}{2}\left[1 - \frac{R}{R+\Delta R} + \frac{1}{2\pi}\sin\left(\frac{2\pi R}{R+\Delta R}\right)\right]^{-1} \quad (1)$$

Here τ is the lifetime of *o*-Ps; R the radius of the, expectedly, spherical free volume holes; and ΔR is a constant. The equation provides that the lifetime of *o*-Ps atoms increases with the size of free volume holes [9–12].

The purpose of the present study was to illustrate the plasticization effects of water on two amorphous polymers applied for conventional and controlled release coated tablets during a 30-day long storage period. Besides conventional physico-chemical methods, we have applied positron annihilation lifetime spectroscopy to follow the changes of the free volume of the polymeric films.

2. Materials and methods

2.1. Materials

Polyvinylpyrrolidone (PVP K25, Kollidon K25, BASF, Ludwigshafen, Germany) was selected as an amorphous binder for tablet preparation. Eudragit NE 30 D ("Polyacrylate dispersion 30%", Ph.Eur.4) containing 1.5% (w/w) nonoxynol 100 emulsifier, as additive, was applied for preparation of free films and the consequent film coating of tablets.

2.2. Preparation of free films of Eudragit NE

Approximately 10 g Eudragit NE 30D dispersions were poured on a Teflon plate (diameter = 10 cm) and dried in a sealed container above copper sulphate and stored at room temperature for 1 week. The dried films were cut into small pieces and blister-packed.

2.3. Tablet preparation for release studies

Twelve batches of 300 tablets were produced using a wet granulation procedure beginning with the mixing of 100 g anhydrous theophylline (Ph.Eur.) and 10 ml of a 15% (w/v) PVP K25 aqueous solution. Next the wet granulated mass has been dried in a hot air drier at 60 °C for 24 h (Labor-Innova, Hungary). The dried granule mass was fractionated and the fractions of 0.250–0.630 mm particle size range were used for the compression. Tablets of 12 mm in diameter were compressed from the granules using a force of 1000 N with a single-punch tableting machine.

2.4. Storage conditions

The dried tablets used in release studies were divided into batches and transferred into four desiccators kept at 45, 55, 65 and 75% RH at 25 ± 1 °C for 30 days.

PVPK25 powder samples for solubility and PALS measurements were transferred into sealed containers kept at relative humidities of 25, 35, 45, 55, 65, and 75% RH and stored at 25 ± 1 °C for 1 day, 1 week and 1 month. The Eudragit NE films were stored at 75% RH.

2.5. Dissolution measurements

Dissolution measurements were performed by the USP paddle method at a stirring rate of 100 rpm in 900 ml of pH 1.2 HCl solution. The quantity of the dissolved theophylline was determined by UV–vis spectrophotometry (Unicam, ATI Unicam, Cambridge, UK) measuring the absorbance at 272 nm. Three tablets were tested at each sampling points.

2.6. Water content determination

After each storage period at a specific relative humidity, the water content of samples ($n = 4$) was determined by Karl–Fischer titration (Mettler Toledo DL 35, Lot, Belgium) using dry methanol and Hydranal® Composite 5 (Riedel-De-Haën, Seelze, Germany).

2.7. Positron lifetime measurements

The positron source applied for the measurements was made of carrier free ^{22}NaCl of the activity of 4×10^5 Bq. The active sodium chloride was sealed between two very thin (5 μm) titanium foils. The source was then placed between two pieces of Kollidon 25 treated identically before. Positron lifetime spectra were recorded by a conventional fast–fast coincidence system [13]. The system was constructed from standard ORTEC electronic units and the detectors from BaF_2 scintillator crystals and XP2020Q photomultipliers. The time resolution of the system was about 200 ps.

The spectra were first evaluated by the RESOLUTION computer code [14]. Three lifetime components were found in each case, from which only the medium long (τ_2) is used below to characterize the electron density of the material. After that a variation of the MELT code [15] was used to extract lifetime distributions from the spectra. These latter evaluations were used to characterize the size distribution of free volume holes in the samples throughout *o*-Ps lifetime.

3. Results and discussion

3.1. The release of theophylline from tablets containing PVP

The change of dissolution profiles of theophylline correlates with the well-known plasticization effect of water on PVP (Fig. 1). A low humidity of the air does not modify the structure of PVP, not even after a long time. Consequently, any change of dissolution profiles can only be observed on tablets stored at RH = 65%. At this point, a water-induced glassy-to-rubbery transition of PVP K25 occurs, which modifies the pore structure of the polymer [10,11]. This transition produces a densification and a reduction of porosity of the binder. This change of the physical state of the amorphous polymer might well lead to a change observed in the theophylline dissolution profile. The samples stored at RH = 75% were, due to the absorbed water, well above the glass transition temperature of PVP K25. In this state, the polymeric chains of the binder have a greater mobility and, consequently, the tablet loses its original pore structure [16]. The loss of pores results in a lower dissolution rate due to the hindered diffusion of water and theophylline molecules in the tablet.

Although this explanation for the change of dissolution profiles is reasonable, the nature and the dynamics of the observed structural change was not clear at this point. So, additional measurements were performed on poly(vinylpyrrolidone).

3.2. Solubility coefficient of poly(vinylpyrrolidone)

In general, the solubility coefficient, defined as the ratio of the concentration of water in the polymer and its relative pressure above it (S = cw/relative pressure), did not really change in samples stored under low relative humidity conditions. Although the water content of these samples varied between 0.14 and 0.32% (w/w), the solubility coefficient remained 0.56. Only a very small increase occurred in samples stored at RH = 45 and 55% after 30 days of storage.

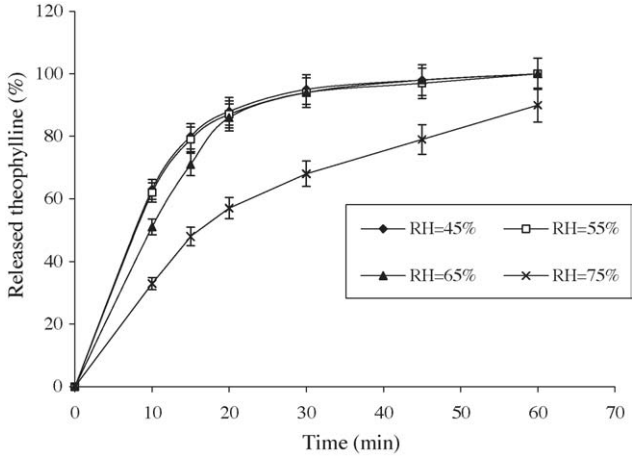

Fig. 1. Theophylline release from PVP-containing tablets stored under different relative humidity conditions for 30 days.

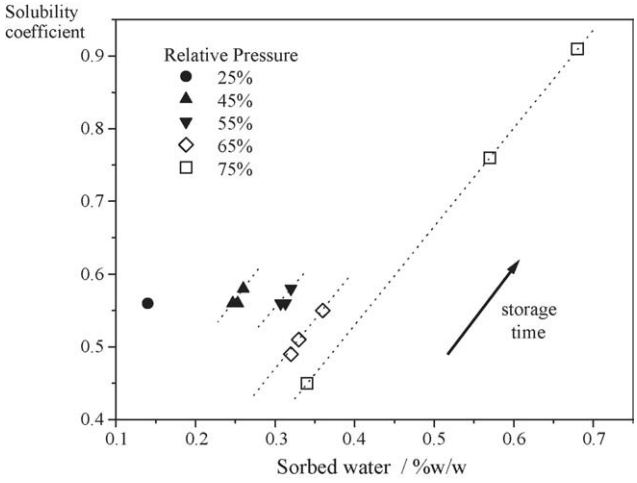

Fig. 2. The solubility coefficient of poly(vinylpyrrolidone) as a function of water content, the storage time and the relative pressure of water during the storage. Open symbols indicate samples in which a water-induced glassy-to-rubbery phase transition takes place. The dashed lines are only shown to guide the eye.

In samples stored at a relative pressure of water of 0.65 (RH = 65%) and above, this trend was abruptly broken (Fig. 2). For these samples, not only their actual water content but also their storage history determines the solubility coefficient. After a day of storage the solubility coefficient was decreased considerably but increased later on. During the 30-day storage period both the water content of the samples and the solubility coefficient increased considerably. However, the increase is obvious only if we separate the samples according to storage conditions (dashed lines in Fig. 2). Otherwise the picture is quite confuse and the water content and the solubility of a sample do not seem to correlate. The analysis of Fig. 2 suggests that, at and above RH = 65%, the structure of PVP is modified very rapidly and the structural change is determined by the relative pressure of water above the polymer. During a longer storage period the newly formed structure absorbs additional water and determines the properties (e.g., the solubility coefficient) of the polymer [17].

Our results are in agreement with those of Fitzpatrick et al., who reported [16] that the glassy to rubbery conversion of PVP produced a densification and a reduction of porosity of PVP. This change in the physical state of amorphous PVP led to a change in the dissolution rate of tablets in their study.

According to the Flory–Huggins theory of polymer solutions, if the mixing process were driven by an entropic gradient (nonpolar solvent), the solubility coefficient would increase as a function of the solvent concentration. In the case of an exothermic interaction between the solvent and the polymer (i.e. $\chi > 0$), the solubility would be reduced. To apply this theory to PVP, we should separate the sets of sam-

ples according to storage conditions. The separated sets fulfil the Flory–Huggins theory totally, although the water uptake is almost negligible at low relative pressures of water (Fig. 2). After the separation, we obtain sets of points in which the increasing water content results in an increasing solubility coefficient. Thus, we should expect that, in every separate sets, the absorption of water is a process driven by an entropic gradient.

On the other hand, if we consider the initial states of samples (i.e., the state reached after the first day of storage), the variation of parameter *S* indicates an exothermic interaction between water molecules and the polymer. The enthalpy of mixing is reduced in this initial step providing a solubility coefficient decreasing with the increasing water concentration.

This suggests that the process of mixing between water and the polymer occurs by two distinct mechanisms. The first step of the process, at any relative pressure of water, is mainly driven by enthalpic gradients, whereas entropic gradients become dominant later on under every relative pressure of water. The enthalpic driving force might be due to specific interactions between water and the oxygen of vinylpyrrolidone, i.e., to a formation of hydrogen bonded "clusters" of water and PVP. This specific interaction is manifested most obviously in the anomalous behaviour [10,11] of PVP under the relative pressure of water of 0.65. Consequently, the overall sorption process can be envisaged as occurring by two distinct mechanisms: (a) absorption of water molecules on hydrophilic groups and (b) water dissolution in the polymer matrix [18].

The exact place of the absorption–dissolution transition point as a function of the relative pressure and storage time might have many practical consequences concerning the stability of certain dosage forms containing PVP. Moreover, the knowledge of free volume changes connected with the transition point might help in the design of dosage forms.

3.3. o-Ps lifetime distributions around the absorption–dissolution transition point

Previous studies [19] confirmed that, when positron lifetime distributions are measured in PVP, the only parameter showing significant variation with water content is the peak position (centroid) of the *o*-Ps lifetime distribution, i.e., the average size of free volumes around the annihilating *o*-Ps atoms. The samples stored at 65% relative humidity are located just at the transition range. Instead of large free volume holes, the sample stored at 65% relative humidity for a week contains smaller ones. Ab initio calculations revealed that a networked structure could be formed under this condition, where "crosslinks" are formed by water molecules bound to separate polymeric chains by hydrogen bonds. The energy gain supported by the formation of this complex can be sufficient to cause the creation of such "crosslinks" at a certain water concentration. As this process requires a certain position of the neighbouring polymeric chains, separate hydrogen bonds are preferred below this water concentration. Similarly, in the presence of excess water, the chains depart from each other and the possibility of "crosslink" formation vanishes [10].

Other methods revealed very similar results to those of positron lifetime measurements. A water molecule is much smaller than the distance between the oxygen atoms of neighbouring PVP carbonyls in a glassy state polymer (0.417 nm). On the other hand, the pyrrolidone rings of the PVP macromolecule are able to move closer to each other in hydrated solid state PVP above the T_g or in a solution, allowing the water molecule to use both its proton-donating vacancies to form two hydrogen bonds simultaneously with neighbouring PVP repeat unit [20]. Lebedeva et al. reported [21] similar results that the formation of such intermolecular hydrogen bonds becomes only possible at relatively large degrees of hydration. The significance of 65% RH is also emphasized by the results of former physical hardness measurements [22].

A close examination of positron lifetime distribution curves reveals the free volume changes caused by the water-induced structural changes of PVP (Fig. 3). In samples stored under low humidity conditions (RH = 55%), nothing serious happens with the free volume. Exactly as it is expected from theophylline release and solubility measurements. The *o*-Ps lifetime distribution is broad and almost undisturbed during

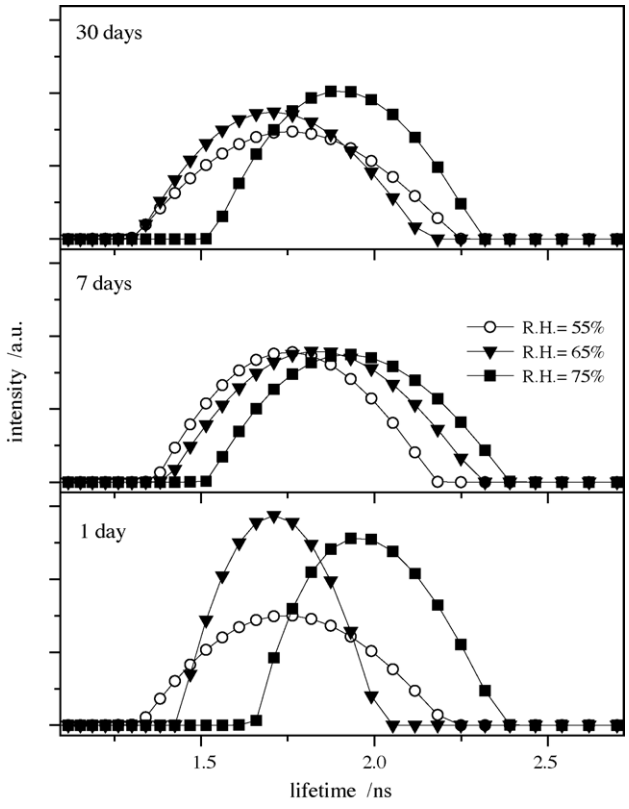

Fig. 3. The effect of storage conditions on the positron lifetime distribution in PVP. Note that the lifetime distribution is connected with the size distribution of free volume holes.

the whole storage period. The very few water molecules, absorbed by the polymer, are not able to modify the initial structure of PVP. Note that the polymer is in its glassy state.

On the other hand, samples stored at RH = 65 and 75% show the discussed two-step swelling mechanism. In these cases, lifetime distributions change dramatically after a day of storage. They become narrow and shift toward longer lifetime with the increasing water content. As the *o*-Ps lifetime is connected with the size of free volume holes very strongly (Eq. (1)), we can conclude that the first exothermic step of mixing (or swelling) results in a narrower free volume size distribution. This is in a very good agreement with the results of former calculations [10,19]. As the chains of PVP becomes "networked" by water molecules, the structure becomes more ordered and free volume holes more uniform because the forming water bridges fix the distance between polymeric chains. The enthalpy driving the whole reorientation of molecules is provided by the binding energy of the forming hydrogen bonds.

A longer exposure of PVP to water leads to different structures in samples stored at RH = 65 and 75%. At RH = 65%, the entropy driven water uptake does not possess enough strength to overpower the binding force of hydrogen bonds. The result is an intermediate structure: the size distribution of free volume holes widens but does not shift towards larger holes (Fig. 3). The remaining crosslinks formed by H-bounded water molecules prevents the "streaming" of water into the bulk of the polymer. However, the widening of the distribution after 30 days of storage suggests that a large fraction of "crosslinks" was ruined by excess water molecules. This result agrees with those of theophylline release and solubility measurements.

On the other hand, the tremendous excess of water molecules leads to larger holes and more absorbed water in the case of RH = 75%. The lifetime distribution measured after a day of storage suggests that both steps of the "swelling" are fast. The lifetime (and hole size) distribution is narrow at this point but shifted towards longer lifetimes (larger holes) indicating a considerable amount of absorbed water. An even larger amount of water leads to a widening of the distribution peak showing the breaking of crosslinks and the forming of a less ordered structure. Note that the case observed at RH = 75% is the regular way of swelling of polar polymers in water [10,18].

As a conclusion drawn from *o*-Ps lifetime distributions, we can state that the initial exothermic step of the "mixing" of PVP with water leads to a structure which is more ordered than the initial dry glassy state. Free volume holes in this structure have more uniform sizes then in the dry polymer. However the size distribution of holes becomes wider, if the driving force of the excess water is enough break the new structure, and the exothermic absorption process becomes an entropy-driven dissolution process. The widening of the size distribution of free volume holes benefits solution of PVP.

3.4. Induced absorption–dissolution transition in Eudragit NE

The mentioned absorption–dissolution transition occurs in Eudragit NE samples in a very strange way. In pure Eudragit NE, the transition does not appears at all (Fig. 4). Not even a storage at RH = 75% for 30 days modifies the original size distribution of free volume holes. The *o*-Ps lifetime distribution is wide and unchanged during the whole storage period. On the other hand, the addition of nonoxynol to Eudragit NE starts processes very similar to those observed in PVP.

Fig. 4 illustrates the effect of nonoxynol on the *o*-Ps distribution of Eudragit NE samples. The characteristic peak becomes narrower in samples containing the additive and, therefore, the size distribution of free volume holes becomes more uniform. From the point of view of controlled release applications, this alteration could be useful because the uniform size distribution enables constant rate diffusion through the polymer film. The only problem is that the structural change caused by the additive is very similar to that observed in the case of PVP–water "mixing". The structural change benefits the "streaming" of water into the bulk polymer, as it is indicated by shifting and widening lifetime distributions. Thus nonoxynol, applied to form uniform free volume holes, destroys the long-term stability of the film.

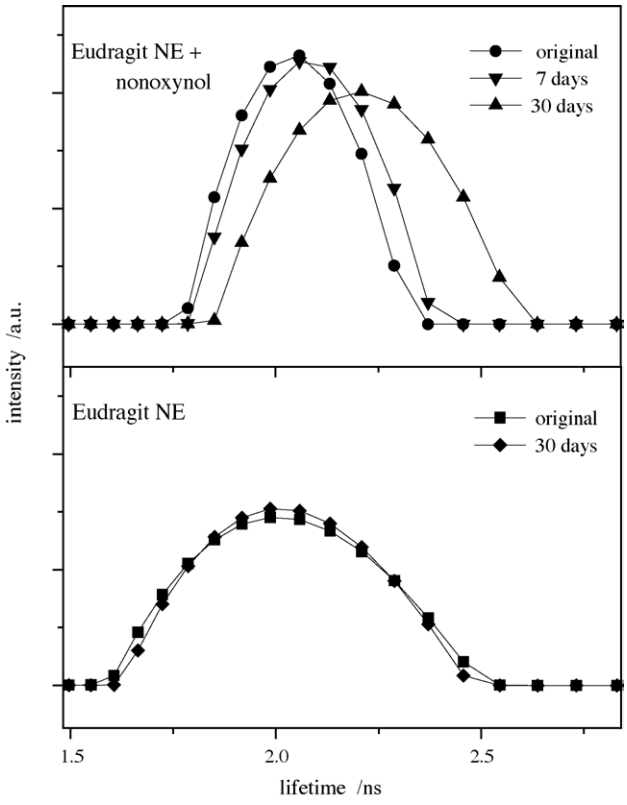

Fig. 4. The effect of storage time on Eudragit NE films with and without nonoxynol 100. The relative pressure of water was 0.75 (RH = 75%) in every case.

4. Conclusions

It was shown that the dissolution of poly(vinylpyrrolidone) is a two-step process. It starts with an exothermic absorption step and the mixing of polymeric chains and water molecules starts only after this initial structural rearrangement. The structure formed in the absorption step determines the kinetics and dynamics of the dissolution.

Positron lifetime distributions revealed the changes of the free volume during the absorption–dissolution transition. It was shown that the first absorption process results in a structure with free volume holes more uniform than those in the dry polymer. The dissolution starts with the breaking of this new structure and the "streaming" of water into free volume holes.

In the case of Eudragit NE, it was shown that the addition of nonoxynol initiates the absorption–dissolution transition in the inert polymer.

The above results emphasize that the positron annihilation lifetime method can be recommended as sensitive means for the stability tests of pharmaceutical dosage forms containing polymeric drugs and excipients.

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References

[1] J.-H. Guo, J. Pharm. Sci. 83 (1994) 447–449.
[2] B.C. Hancock, G. Zografi, J. Pharm. Sci. 86 (1997) 1–12.
[3] J.M.G. Cowie, R. Ferguson, Polymer 34 (1993) 2135–2141.
[4] C.A. Oksanen, G. Zografi, Pharm. Res. 7 (1990) 654–657.
[5] C.A. Oksanen, G. Zografi, Pharm. Res. 10 (1993) 791–799.
[6] B.C. Hancock, G. Zografi, Pharm. Res. 11 (1994) 1166–1173.
[7] A.H. Baugher, W.J. Kossler, K.G. Petzinger, R.H. Pater, Mater. Sci. Forum 57 (1997) 57–61.
[8] K. Süvegh, A. Domján, Gy. Vankó, B. Iván, A. Vértes, Macromolecules 31 (1998) 7770–7775.
[9] K. Süvegh, A. Vértes, T. Hyodo, Adv. Mol. Struct. Res. 5 (1999) 313–357.
[10] K. Süvegh, R. Zelkó, Macromolecules 35 (2002) 795–800.
[11] R. Zelkó, K. Süvegh, S. Marton, I. Rácz, Pharm. Res. 17 (2000) 1028–1030.
[12] Q. Deng, Y.C. Jean, Macromolecules 26 (1993) 30–34.
[13] I.K. MacKenzie, in: W. Brandt, A. Dupasquier (Eds.), Positron Solid-state Physics, North-Holland, Amsterdam, 1983, p. 196.
[14] P. Kirkegaard, M. Eldrup, O.E. Mogensen, N.J. Pedersen, Comput. Phys. Commun. 23 (1981) 307–338.
[15] A. Shukla, M. Peter, L. Hoffmann, Nucl. Instrum. Meth. A 335 (1993) 310–325.
[16] S. Fitzpatrick, J.F. McCabe, C.R. Petts, S.W. Booth, Int. J. Pharm. 246 (2002) 143–151.
[17] R. Zelkó, Polym. Degrad. Stabil. 87 (2005) 355–359.
[18] P.A. Netti, L. Ambrosio, M.A. Del Nobile, G. Mensitieri, L. Nicolais, J. Bioact. Compat. Polym. 11 (1996) 312–316.
[19] R. Zelkó, K. Süvegh, Eur. J. Pharm. Sci. 21 (2004) 519–523.
[20] M.M. Feldstein, G.A. Shandryuk, N.A. Platé, Polymer 42 (2001) 971–979.
[21] T.L. Lebedeva, M.M. Feldstein, S.A. Kuptsov, N.A. Platé, Polym. Sci. 42 (2000) 989–1005.
[22] F. Kiekens, R. Zelkó, J.P. Remon, Pharm. Res. 17 (2000) 490–494.