Biological and Physicochemical Study of the Implantation of a Modified Polyester Vascular Prosthesis

Abstract

Results are presented of an investigation into the susceptibility to hydrolytic and enzymatic degradation of poly(DL-lactide-co-glycolide) copolymers of varied composition. Assessment of the degradation was based on two factors: measurement of the mass loss and estimation of the molecular mass by the GPC method. The examination made it possible to select copolymers for the temporary sealing of polyester vessel prostheses. The sealed vessel prostheses manifested full biocompatibility in the range of irritant action, systemic toxicity, haemolytic action, cytotoxicity and genotoxicity. The usefulness of the polymeric material applied was also confirmed by examination of the local reaction of implants in test animals. The impact of accelerated ageing and radiation sterilisation upon the physical-mechanical properties of the vessel prostheses was also studied. The studies enabled to prepare tentative technology guidelines for the sealing of artificial blood vessels with a biocompatible and resorbable synthetic copolymer – lactide/glycolide.

Key words: vessel prosthesis, poly(DL-lactide-co-glycolide), degradation, sealing, biocompatibility, implantation.

Materials

Resomer RG 504H, copolymer of DL-lactide/glycolide (51% residues of DL-lactic acid, 49% residues of glycolic acid), inherent viscosity – 0.68, supplied by Boehringer Ingelheim;

Resomer RG 755S – copolymer of DL-lactide/glycolide (75% residues of DL-lactic acid, 25% residues of glycolic acid), inherent viscosity – 0.52, supplied by Boehringer Ingelheim;

Polyester double – side veloured prosthesis DALLON H of 8 mm diameter, supplied by Tricomed Co.

Methodology

Water permeability of the vessel prostheses

The water permeability of the modified prostheses was tested according to Standard ISO 7198 “Cardiovascular implants – Tubular vascular prostheses”. A device used to measure the volume in millimeters of water permeating through a surface unit (1 cm²) of the prosthesis wall during 1 minute at a pressure of 164 hPa (120 mm Hg).

Susceptibility to degradation

Testing of the susceptibility to hydrolytic and enzymatic degradation was carried out on films prepared from solutions with the same Resomers as those used for the sealing of prostheses. A 3% solution of the copolymers in 1.4-dioxane with a plasticiser (5% polyethylene glycol 600) was cast on a Teflon matrix to form the films. The films were next dried in the same way as with the modified prostheses.

The degradation proceeded for 90 days at 3°C in the following forms: hydrolytically in demineralised water at pH = 7.45, and enzymatically in a citric-phosphate buffer at pH 7.2-7.3 with a lysozyme concentration of 10-200 mg/cm³ and bath module of 1:250 w/w. The film samples were taken out of the bath at fixed time intervals, rinsed with demineralised water at ambient temperature, and dried at 25°C to constant weight. In the enzymatic degradation, the films were additionally submerged for 10 min in 70% ethanol to deactivate the enzymes. The proceeding degradation was assessed by the change in pH and mass loss as well as by GPC analysis.

Examination of the accelerated ageing

The dependence of useful properties of the prostheses on the shelf time was tested according to Standard ATM F 1980 Accelerated Ageing Testing, based on the Van’t Hoff theory, concerning the accelerated ageing of products at an elevated temperature.

Analytical methods

Samples (ca. 15 mg) of the material tested and 7 ml of chloroform HPLC (or 1.4-dioxane) were placed in a 10 ml measuring flask. On dissolving the sample, the flask was filled up to 10 ml with the solvent used, and the solution was filtered and injected into a chromatography column to carry out an analysis. The parameters of the chromatography analysis were as below:

- solvent (mobile phase): chloroform
- column: Plgel Mixed C, 300 mm, 5μm (Polymer Laboratories Ltd.)
- temperature of the column: 35°C,
- flow speed: 0.7 ml/min,
- injection volume: 100 ml,
- calibration standards: polystyrenes with a molecular weight in the range

Vessel prostheses, particularly those prepared by knitting, reveal an insufficient tightness which can be remedied by surface sealing to provide a temporary tightness. In earlier works [1] the family of poly(DL-lactide-co-glycolides) of the Resomer type was selected for this purpose. A sealing method was also prepared on a large laboratory scale, including the composition of the coating bath and coating conditions. The modification of the vessel prostheses provided the surgical tightness required (water permeability below 30 ml/cm²/min) and adequate physical-mechanical features including an elasticity sufficient for specific medical devices.

The aim of this work was to finally select the ultimate copolymer for the sealing of vessel prostheses and to confirm the usefulness of the modified prostheses as implants. The investigations were focused on the assessment of the susceptibility to hydrolytic and enzymatic degradation of films prepared from a selected group of copolymers. The biocompatibility of experimental vessel prostheses sealed with a selected copolymer was tested. Introductory implantation trials were also made.

Materials

- Resomer RG 504H, copolymer of DL-lactide/glycolide (51% residues of DL-lactic acid, 49% residues of glycolic acid), inherent viscosity – 0.52, supplied by Boehringer Ingelheim;
- Resomer RG 755S – copolymer of DL-lactide/glycolide (75% residues of DL-lactic acid, 25% residues of glycolic acid), inherent viscosity – 0.68, supplied by Boehringer Ingelheim;
- Polyester double – side veloured prosthesis DALLON H of 8 mm diameter, supplied by Tricomed Co.

Methodology

Water permeability of the vessel prostheses

The water permeability of the modified prostheses was tested according to Standard ISO 7198 “Cardiovascular implants – Tubular vascular prostheses”. A device used to measure the volume in millimeters of water permeating through a surface unit (1 cm²) of the prosthesis wall during 1 minute at a pressure of 164 hPa (120 mm Hg).

Susceptibility to degradation

Testing of the susceptibility to hydrolytic and enzymatic degradation was carried out on films prepared from solutions with the same Resomers as those used for the sealing of prostheses. A 3% solution of the copolymers in 1.4-dioxane with a plasticiser (5% polyethylene glycol 600) was cast on a Teflon matrix to form the films. The films were next dried in the same way as with the modified prostheses.

The degradation proceeded for 90 days at 3°C in the following forms: hydrolytically in demineralised water at pH = 7.45, and enzymatically in a citric-phosphate buffer at pH 7.2-7.3 with a lysozyme concentration of 10-200 mg/cm³ and bath module of 1:250 w/w. The film samples were taken out of the bath at fixed time intervals, rinsed with demineralised water at ambient temperature, and dried at 25°C to constant weight. In the enzymatic degradation, the films were additionally submerged for 10 min in 70% ethanol to deactivate the enzymes. The proceeding degradation was assessed by the change in pH and mass loss as well as by GPC analysis.

Examination of the accelerated ageing

The dependence of useful properties of the prostheses on the shelf time was tested according to Standard ATM F 1980 Accelerated Ageing Testing, based on the Van’t Hoff theory, concerning the accelerated ageing of products at an elevated temperature.

Analytical methods

Samples (ca. 15 mg) of the material tested and 7 ml of chloroform HPLC (or 1.4-dioxane) were placed in a 10 ml measuring flask. On dissolving the sample, the flask was filled up to 10 ml with the solvent used, and the solution was filtered and injected into a chromatography column to carry out an analysis. The parameters of the chromatography analysis were as below:

- solvent (mobile phase): chloroform
- column: Plgel Mixed C, 300 mm, 5μm (Polymer Laboratories Ltd.)
- temperature of the column: 35°C,
- flow speed: 0.7 ml/min,
- injection volume: 100 ml,
- calibration standards: polystyrenes with a molecular weight in the range
of 580 to 340,000 and polydispersity – from 1.04 to 1.14 (Polymer Laboratories).

Testing of mechanical properties


 susceptibilities to degradation

Susceptibility to degradation of poly(DL-lactide-co-glycolide) copolymers selected for the sealing of prostheses.

To better follow changes that occur in the course of the degradation of the polymeric materials chosen as sealants of prostheses, films with a constant thickness equivalent to the sealing coating on the vessel prostheses were examined. Films cast onto a Teflon matrix from 3% solutions of Resomer RG 755S and Resomer RG 504H in 1.4-dioxane with a plasticiser (5% polyethylene glycol 600) were used in the testing. Photos were taken of all preparations. The conditions at which the degradation tests were carried out are shown in Table 1.

Results of the degradation testing are shown in the two tables below: Table 2 – hydrolytic degradation, Table 3 – enzymatic degradation.

Films of Resomers RG 755S and RG 504H degrade rather differently, marked by a loss of mass and pH change in the bath. More prone to degradation is Resomer RG 504H, having an equimolar content of glycolide and lactide residues. A complete disintegration of the film followed after 30 days (Figure 1), and the mass loss exceeded 70%; at the same time the pH of the bath dropped to about 3, caused by the migration of hydrolysis products such as glycolic – and lactic acids to the solution [2, 3]. The lowered pH added to the acceleration of the hydrolysis rate. It must be mentioned that Resomers marked with index “H” are additionally processed by the manufacturer to increase the susceptibility of the material to degradation. The other Resomer marked RG 755S, with a higher content of lactide residues, is less susceptible to hydrolysis. A decrease in its mass of about 44% could be observed only after 90 days. After 7 days of incubation the films became opaque, and their appearance remained unchanged until the 30th day (Figure 2). Only after 60 days did they become brittle and undergo disintegration, the pH of which was on the decrease, reaching 3.9 after 90 days.

As for hydrolytic degradation, films of Resomer RG 504H degrade faster than those of Resomer RG 755S, also in the presence of lysozyme. In Resomer RG 504H, an extensive mass loss, C, in the range of 88-89% for all lysozyme concentrations could be seen as soon as after 60 days, which is slightly lower than in the hydrolytic mode. The Resomer RG 504H films had almost entirely degraded after 90 days; however, the change in pH in the buffer was not as distinct as in the bath of the hydrolytic process. A higher pH in the enzymatic process explains the lower degradation rate compared to that using the hydrolysis route. Figure 3 presents images of Resomer RG 504H films during degradation up to 30 days.

In Resomer 755S, the mass loss after 90 days, depending on the lysozyme concentration, was distinctly lower, amounting to about 39-57% (Figure 4).

### Table 1. Conditions of the hydrolytic and enzymatic degradation of films of Resomers RG 755S and RG 504H.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Hydrolytic degradation</th>
<th>Enzymatic degradation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time, days</td>
<td>0, 7, 14, 30, 60, 90</td>
<td>0, 7, 14, 30, 60, 90</td>
</tr>
<tr>
<td>Temperature</td>
<td>37°C</td>
<td>37°C</td>
</tr>
<tr>
<td>Medium</td>
<td>water pH = 7.45</td>
<td>Phosphate-citric buffer pH = 7.24 and 7.30</td>
</tr>
<tr>
<td>Enzyme</td>
<td>-</td>
<td>lysozyme (concentr. 10, 100, 200 μg/cm²)</td>
</tr>
<tr>
<td>Bath module</td>
<td>250:1 w/w</td>
<td>250:1 w/w</td>
</tr>
</tbody>
</table>

### Table 2. Impact of the incubation time of films of Resomer RG 755S and Resomer RG 504H in water at 37°C upon the change in mass and pH of the bath.

<table>
<thead>
<tr>
<th>Degradation time, days</th>
<th>Resomer RG 755S</th>
<th>Resomer RG 504H</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH</td>
<td>Mass loss, %</td>
</tr>
<tr>
<td>0</td>
<td>7.42</td>
<td>–</td>
</tr>
<tr>
<td>7</td>
<td>7.33</td>
<td>4.68</td>
</tr>
<tr>
<td>14</td>
<td>7.22</td>
<td>5.10</td>
</tr>
<tr>
<td>30</td>
<td>7.15</td>
<td>5.45</td>
</tr>
<tr>
<td>60</td>
<td>4.68</td>
<td>11.30</td>
</tr>
<tr>
<td>90</td>
<td>3.91</td>
<td>43.73</td>
</tr>
</tbody>
</table>

### Table 3. Impact of the degradation time and enzyme concentration time upon the change in mass, and pH of the bath of Resomer RG 755S and Resomer RG 504H.

<table>
<thead>
<tr>
<th>Degradation time, days</th>
<th>Enzyme concentration, μg/cm²</th>
<th>Resomer RG 755S</th>
<th>Resomer RG 504H</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH</td>
<td>Mass loss, %</td>
<td>pH</td>
</tr>
<tr>
<td>0</td>
<td>–</td>
<td>7.30</td>
<td>–</td>
</tr>
<tr>
<td>7</td>
<td>10</td>
<td>7.09</td>
<td>4.84</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>7.08</td>
<td>3.62</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>7.07</td>
<td>4.28</td>
</tr>
<tr>
<td>14</td>
<td>10</td>
<td>7.07</td>
<td>4.76</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>7.05</td>
<td>3.44</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>7.03</td>
<td>5.08</td>
</tr>
<tr>
<td>30</td>
<td>10</td>
<td>7.05</td>
<td>6.40</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>7.03</td>
<td>5.68</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>7.01</td>
<td>7.32</td>
</tr>
<tr>
<td>60</td>
<td>10</td>
<td>7.03</td>
<td>9.21</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>7.01</td>
<td>9.88</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>7.00</td>
<td>11.46</td>
</tr>
<tr>
<td>90</td>
<td>10</td>
<td>6.99</td>
<td>38.91</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>6.98</td>
<td>51.63</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>6.97</td>
<td>57.71</td>
</tr>
</tbody>
</table>
The proceeding degradation, both hydrolytic and enzymatic, of the Resomer films was also assessed by the function of the molecular mass distribution (GPC analysis). Examples of molecular mass distribution curves for Resomer RG 755S are shown in Figure 5 (hydrolytic degradation) and in Figure 6 (enzymatic degradation, lysozyme concentration: 200 µg/cm³), starting with a curve presenting the molecular mass distribution of the polymer after sterilisation.

When the films were kept in the buffer, hydrolytic degradation proceeded very slowly, reflected by only an insignificant shift of the distribution curves towards decreasing values. Only after 60 days were the changes more pronounced, coinciding with the measurements of mass loss.

Similarly, an inconsiderable change in molecular mass occurred up to the 14th day of enzymatic degradation with a lysozyme concentration of 200 µg/cm³. The changes are more distinct after 30 days, and even more so after 60 days, showing a more intensive degradation than in the hydrolytic route; however, the mass loss is comparable for both processes (11.5%).

Considering the excessive susceptibility of Resomer RG 504H to degradation, Dallon H prostheses with Resomer RG 755S were chosen for the assessment of biocompatibility.

Impact of sterilisation and ageing upon the properties of the modified vessel prostheses

Modified Dallon H prostheses were radiation-sterilised (fast electrons) and then subjected to accelerated ageing to define the impact of the treatment upon the physical-mechanical properties of the prostheses.

The prostheses were prepared according to methodology described elsewhere [1], wrapped in a blister pack, and radiation-sterilised. Sterilisation with fast electrons at a dose of 25 kGy was done at the Institute of Nuclear Chemistry and Technique, Warsaw, by a high-energy electron bundle (10 MeV) generated in an Elektrona 10/0 accelerator.

The impact of the radiation upon the structure of the seal coating of the prostheses was assessed by means of SEM inspection (Figure 7) as well as by testing of their physical-mechanical properties (Table 4).
The examination proved that radiation at the dose applied does not significantly change the structure of the polymeric coating.

The ageing examination was aimed at defining the time during which the modified prosthesis preserves its properties and remains fit for its destination. It would take quite long to establish the time in real conditions, hence the method of accelerated ageing was applied. Such products, according to the method, are stored at an elevated temperature for a given time defined by the Arrhenius equation.

The storage time at an elevated temperature is calculated from the equation:

\[ C_{T_1} = C_{RT} / AF \]

where:
- \( C_{T_1} \) – shelf time of the product at the ageing temperature
- \( C_{RT} \) – storage period to be simulated
- \( AF \) – ageing coefficient \( AF = Q_{10}^{(T_{11}-T_{RT})/10} \)
- \( Q_{10} \) – constant of reaction rate; assumed as 2
- \( T_{T1} \) – temperature at which the accelerated ageing process is to be conducted
- \( T_{RT} \) – ambient temperature

Accelerated ageing was accomplished at the R&D Laboratory of Tricomed Co according to guidelines contained in ASTM F 1980. To examine the influence of accelerated ageing upon the properties of the prostheses, the quality parameters of Dallon H were analysed before and after ageing.

The examination was carried out in an ageing chamber at +55°C for 32.3 days, equivalent to 1 year of ageing at ambient temperature (20°C).

In Table 4 are compiled the basic properties of the modified prostheses before and after sterilisation, and after accelerated ageing.

The testing of the physical-mechanical properties after radiation sterilisation revealed that undesired changes only occur in the bursting strength, which dropped by 17%. The 10% increase in the displacement at burst evidences an increase in the elasticity of the prostheses. The suture retention strength remained unchanged. It is quite important that the water permeability remained unchanged after the action of the 25 kGy sterilisation dose.

### Table 4. Physical-mechanical properties of the modified vessel prostheses before and after sterilisation, and after accelerated ageing.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Before sterilisation</th>
<th>After sterilisation</th>
<th>After ageing</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inner diameter, mm</td>
<td>8.0</td>
<td>7.97</td>
<td>7.97</td>
</tr>
<tr>
<td>Bursting strength, N</td>
<td>270</td>
<td>222</td>
<td>242</td>
</tr>
<tr>
<td>Displacement at burst, mm</td>
<td>6.9</td>
<td>7.6</td>
<td>6.1</td>
</tr>
<tr>
<td>Suture retention strength, N</td>
<td>17.1</td>
<td>17.1</td>
<td>17.2</td>
</tr>
<tr>
<td>Water permeability, ml/cm²/min</td>
<td>28.0</td>
<td>27.9</td>
<td>2100</td>
</tr>
</tbody>
</table>
Changes in the main mechanical parameters after accelerated ageing do not exceed the 20% limit assumed in comparison with the after-sterilisation values. The bursting strength increased by 9%, and the displacement at burst and suture retention strength dropped by 19.8% and 5%, respectively.

However, the most significant change occurred with respect to the water permeability, which, after accelerated ageing, decreased to about 2100 ml/cm²/min (3500 ml/cm²/min for the unsealed Dallon H prosthesis), indicating substantial degradation of the polymer at the temperature of accelerated ageing (55°C).

The inference is that Dallon H prostheses modified with Resomer RG 755S need to be, as with some pharmaceuticals, kept at a low temperature (fridge).

It may be expected that the resistance of the prostheses to ageing could be improved by the use of the earlier considered sterilisation with ethylene oxide, during which the degradation of seal coatings of poly(lactide-co-glycolide) [4] does not occur. Further investigation is needed in this direction.

**Figure 8.** Macroscopic image of the implant locality (a & b – sealed prostheses, c & d – unsealed prostheses): a) Broad band of fibroblast tissue in the proximity of the implant with a rich deposit from polymorphonuclear cells, lymphocytes and macrophages; b) Band of fibroblast tissue in the proximity of the implant with a cell deposit and visible proliferation of blood vessels; c) Site wall with infiltration of polymorphonuclear cells and lymphocytes with the dominance of eosinophilic granulocytes; d) Numerous giant cells of the round – foreign body type and numerous fresh blood vessels in the band of fibroblast tissue around the implant.

**Biocompatibility according to Standard PN-EN ISO 10993-1:2004**

A number of vessel prostheses were prepared to test their biocompatibility according to Standard PN-EN ISO 10993-1:2004. The examination was carried out in vitro and in vivo on animals at the specialised unit of the ‘National Institute of Drugs’, Warsaw:

- irritation action after an intradermal injection of the extract according to ISO 10993-10:2007
- sensibilisation action according to ISO 10993-10:2007
- systemic toxicity according to Standard ISO 10993-11:2008
- cytotoxicity according to Standard ISO 10993-5: 2001
- haemolytic action according to Standard 0993-4:2007
- genotoxicity according to Standards ISO 10993-3:2008, ISO 10993-12:2008 and BB/PM 5.4/002. The genotoxicity was examined by two methods : the Ames test and micronuclear test.

All these tests evidenced that Dallon H vessel prostheses modified with poly(D,L-lactide-co-glycolide) do not exert any cytotoxicity-, irritation-, sensibilisation- haemolytic- nor genotoxic action.

**In vivo examination with experimental animals according to Standard PN-EN 30993-6:2000.**

Testing was carried out on six rabbits of a New Zealand breed. Fragments of the prostheses sized 12 x 1.5 mm were implanted into subcutaneous tissue on both sides of the animal’s back: left- modified Dallon H, right- unmodified Dallon H as reference. The animals were killed after 30 days, and the locality of the implant was macroscopically assessed. Six implants along with the surrounding tissue were cut out and collected for histopathology inspection by means of confocal microscopy.

It was found that the modified vessel prostheses tolerated well, and after implantation into subcutaneous tissue do not differ from the unmodified reference Dallon H. The medium in the implant locality found by microscopic observation is a typical cell response to a foreign body commonly appearing with medical devices.

Implantation and post-implantation examinations were carried out at the National Institute of Drugs and Centre of Biostructure of the Medical University, Warsaw.

**Figures 8a and 9b** present microscopic images of the implantation locality of the modified prostheses, and **Figures 8c and 8d** show microphotos of the implant locality of the reference prosthesis (magnification- x 100).

**Preparation of technical outlines for the manufacture of partly resorbable vessel prostheses.**

Based upon the R&D works presented, tentative technical outlines have been prepared for the manufacture of modified (sealed) vessel prostheses on a large laboratory scale. The manufacture starts from DALLON H made by Tricomed Co as a half product, which is a double-side coloured polyester prosthesis of 8 mm diameter. The resorbable copolymer of DL lactide and glycolide produced by Boehringer Ingelheim under the trade name of Resomer RG 755 S was chosen as the sealing substance. Spraying with Resomer RG 755 S in 1.4 dioxygen is recommended for the sealing. The prostheses, after drying, can be packed in blister packs and radiation-sterilised at a dose of 25 kGy.

**Conclusions**

1. Based on the examination of selected poly(DL-lactide-co-glycolide) copolymers of varied composition (RG
4. The sterilisation of modified DALLON H prostheses with fast electrons at a 25 kGy dose does not cause an increase in water permeability.

3. The modified prostheses do not exert any cytotoxicity-, irritation-, sensibilisation- haemolytic- nor genotoxic action.

4. The modified DALLON H prostheses tolerate well. When implanted into the subcutaneous tissue of rabbits, it does not differ from the unmodified reference. The medium in the implant locality found by microscopic observation is a typical cell response to a foreign body commonly appearing with medical devices.

5. The modified DALLON H prostheses revealed unsatisfactory durability after radiation sterilisation and accelerated ageing at 55°C.

6. A complementary investigation is recommended for the ageing of modified DALLON H prostheses after sterilisation with ethylene oxide. The susceptibility of the modified prostheses to ageing at low temperature is also to be assessed in further works.

Acknowledgements

The investigation was carried out within research project No 3 T08E 03727 sponsored by the Ministry of Science and Higher Education. We would also like to thank TRICOMED S.A. for their cooperation in this project.

References


Dr Maria Ratajska (1945–2010)

Maria Ratajska was born in 1945, in Łódź, Poland. Since 1963 she had been studying physics at the University of Łódź and graduated in 1968. Next she started her professional work at the Textile Research Institute in Łódź, where she specialised in research on the structure of polymers and fibres using X-ray methods. She obtained a Ph.D. degree in 1978 from the Textile Faculty of the Technical University of Łódź with a thesis entitled “The Influence of Organic Solvents on Changes in the Submicroscopic Structures of Wool and Selected Chemical Fibres”. In 1990 she accepted an offer from The Institute of Chemical Fibres to join their team at the laboratory of the physical chemistry of polymers, directed by the undersigned, and be active in the biodegradation of polymers and fibres. For over 10 years she directed the section concerning biodegradation, additionally working on IR spectrophotometry. We worked on the influence of microorganisms on the structure of polymers and fibres. The main subject of our research was natural polymers, especially cellulose and chitosan, and products of their chemical modification. In the 90s, we presented our research works at conferences and symposia, published numerous scientific articles together and finished some research projects.

When I left the Institute in 2001, Dr Maria Ratajska was in charge of the laboratory of physicochemistry, which lasted till 2008 when she retired.

Besides her scientific work she was also involved in the organisational activity of the Institute. She was also a member of the Scientific Committee of the Institute for certain periods as well as a member of the Editorial Committee and a careful reviewer of Fibres & Textiles in Eastern Europe, the journal of the Institute. Throughout her research work at the Institute of Biopolymers and Chemical Fibres she was the author or co-author of 13 articles published in international journals, such as Fibres & Textiles in Eastern Europe, Polimery and Polymer for Advanced Technology. She was also the author of 40 presentations at symposia and conferences as well as of numerous research projects (the COST 628 project, amongst others).

Her high scientific qualifications, open mind to new ideas and organisational abilities enabled her to manage the laboratory through the difficult period (2001–2003) of applying for accreditations of test and research methods. Among her co-workers she was known as a reliable creative and hard-working person, with great serenity and kindness up to the last days of her life. She was an honest and modest person and this is how she will be remembered.

Stefan Boryniec

Received 18.09.2009 Reviewed 8.11.2010