Dariusz Wawro, Włodzimierz Stęplewski, Kinga Brzoza-Malczewska, *Wojciech Święszkowski

Institute of Biopolymers and Chemical Fibres,
ul. M. Skłodowskiej-Curie 19.27 Łódź, Poland
E-mail: dariusz.wawro@ibwch.lodz.pl

***Division of Materials Design,**
Faculty of Materials Science and Engineering,
Warsaw University of Technology,
Pl. Politechniki 1 00-661 Warsaw, Poland

Collagen-Modified Chitosan Fibres Intended for Scaffolds

Abstract
A method is presented for the preparation of collagen-modified chitosan fibres (Chit/Col) from chitosan-collagen solutions. The fibres may be used in the construction of scaffolds. Chitosan solutions with a concentration of 5.21% in an aqueous 3.0% acetic acid solution with the addition of glycerol were mixed with aqueous solutions of collagen. The applied chitosan was derived from the northern shrimps (Pandalus borealis) and the collagen from calf hide. The usability of chitosan and collagen was determined for the preparation of spinning solutions and Chit/Col fibres. Rheological properties are discussed for the chitosan-collagen solutions used in the spinning of Chit/Col fibres with linear density in the range of 3.30 – 8.36 dtex. The fibres are featured by increased nitrogen content and tenacity when compared to pure chitosan fibres. The performance of the spinning process was good.

Key words: *chitosan, collagen, wet-spinning, fibre, mechanical properties, morphological properties.*

Introduction

Chitosan fibres are used on the market as a raw material in the manufacture of textile, hygiene and medical products. The use of these fibres is beneficial thanks to such properties as biocompatibility, non-toxicity and biodegradability. The manufacture of chitosan fibres does not involve either expensive special equipment or toxic solvents, and the production process is simple and brings no harm to the environment [1]. The applied wet spinning process offers the chance of admixing additives to the aqueous chitosan solution, and thus widely modifying the fibre properties. Other polysaccharides, such as starch or alginates [2 - 4], carbon nanotubes, hydroxyapatites, and nano-silver [5-9], introduced to the spinning solution, count as suitable additives. Proteins such as fibroin, keratin, collagen or polypeptides with varying composition may find use in the production of chitosan fibre with tailored properties [2, 10 - 12]. Surface modification of ready-made textile products with chitosan or nano-silver is also applied by employing various cross-linking techniques. Chitosan fibres are a promising raw material in the manufacture of a variety of textiles [14 - 16]. Collagen is one of the main construction materials of living organisms. It is, as a result of a number of beneficial biological properties, widely used in medicine and in the pharmacological, cosmetic and alimentary industry [2, 17 - 18]. The family of collagens embraces a number of proteins, while the configuration of the polypeptide chains determines the structure of collagen. Elevated temperatures, changes of pH, and irradiation may destroy the natural structure of collagen and, hence, lead to its denaturation. This may occur as a change of second or third order of the structure or change of the super-molecular configuration, while the protein's first order structure remains unchanged. The collagen denaturation temperature depends upon its water content and degree of cross-linking.

Collagen fibres with diameters in the 100 – 1000 nm range made by electrospinning from volatile fluoroalcohols (HFP) like trifluoroethanol (TFE) [18 - 20] can be used in the engineering of cell scaffolds. During the electrospinning from HFP, collagen undergoes a denaturation which changes its biological properties [21].

Fibres with a diameter in the 100 to 150 nm range can be prepared by electrospinning from a solution of type I collagen and poly(ethylene oxide) (PEO with 900 kDa) in the proportion of 1:1 with the addition of sodium chloride [22].

Elastic collagen fibres with a diameter of 220 - 600 nm made by electrospinning from a solution of hydrochloric acid were used in the preparation of cell scaffolds [23].

A chitosan-collagen fibre with a new structure was used in the reconstruction of bones [24]. The physical and biological properties of chitosan may be modified by cross-linking with tri-calcium phosphate and collagen in the form of a medium for cell growth [25 - 26]. A material containing chitosan, hydroxyapatite and collagen may also be used in the regeneration of bones [27].

Chitosan/collagen and chitosan/tropocollagen fibres were prepared in the process of wet-spinning from a solution containing adequate compositions of the polymers [28]. Tropo-collagen made up to 50% of the blend with chitosan, while the collagen portion was up to 6%. Aqueous ammonium with a content of 40 - 43% ammonium sulphate constituted the coagulation bath. The fibre was spun at a speed of 8 m/min, and then drawn by 20-30% in a solution of ethylene glycol with a 2.0% content of NaOH. The resulting chitosan/tropo-collagen fibre revealed a tenacity of 9.50 - 14.52 cN/tex and elongation of 11 - 43%.

A collagen fibre with rather low mechanical parameters was prepared by wet spinning from a collagen solution and by melt spinning from a thermoplastic collagen [29]. The fibres both cross-linked with glutaric aldehyde and not cross-linked showed no cytotoxicity against mouse fibroblasts.

Considering the earlier described chitosan and collagen properties, it was intended to elaborate the conditions for the forming of chitosan/collagen (Chit/Col)

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fibres in a wide range of linear density to be used in the building of scaffolds. The spinning of the Chit/Col fibres for scaffolds at high speed like in unmodified chitosan fibres is a new concept.

The research was aimed at the preparation of collagen-modified chitosan fibres with widely varying linear density as a material for cell engineering to be used in the regeneration of cartilage tissue. The possible use of collagen type 2 and 3 derived from calf hide in the preparation of Chit/Col fibres was examined. The comparison of mechanical and morphological properties between Chit/Col fibres and regular chitosan fibres was another goal of the work.

Materials

Chitosan

Two forms of chitosan ChitoClear® derived from the shrimp *Pandalus borealis* provided by Primex Co were used in the research (see ***Table 1***).

Collagen

Two types of collagen with varying properties were used in the research. The material was provided in the form of an aqueous suspension by PROTEINA Co, Poland, (Producer of Natural Proteins) (see ***Table 2***).

The bioactive solution of collagen finds a wide use in cosmetic and dermatologic preparations.

In the preparation of solutions, coagulation baths and in analytical work, reagents provided by POCh. S.A., Gliwice, Poland were used: sodium hydroxide, acetic acid and glycerol.

Preparation of the chitosan-collagen spinning solution

Based on earlier experience, the chitosan solution was prepared with concentrations in the range of 5.14 - 5.21% in an aqueous 3.0% solution of acetic acid. Dissolving was performed over 75 minutes at 50 °C. After filtration and deaeration, the chitosan solution was used for the preparation of a mixture with a collagen suspension. The collagen suspension was poured into the chitosan solution with agitation at 25 °C to produce the chitosan-collagen solution ready for spinning of the Chit/Col fibres.

Wet spinning of collagen-modified chitosan fibres from the solution

Based on earlier experience, a platinum-rhodium spinneret was applied with 300 holes, 80 μm each in dia. A coagulation bath was used at 30 °C with the following composition: 7 wt. parts of a 3.0% aqueous sodium hydroxide and 3 wt. parts of 96% ethanol. The fibres were spun at a spun-draw-ratio of 0.4 - 1.1, speed of 17.5 m/min and then drawn in water at 30 °C with a total drawing rate of 34%. The fibre band was rinsed in water at 30 °C for 24 hours and then immersed for 10 minutes in aqueous ethanol (60%). The excess of the solvent was removed and the fibre was dried at 30 °C. The Chit/Col fibres were cut into staples with lengths of 38 mm.

Analytical methods

Dynamic viscosity of the aqueous chitosan solution was estimated by the use of a rotation viscometer (Brookfield LVT). The viscometer Brookfield, model RV DV-II+, with the program Rheocalc V3.1-1 was employed in rheological examinations at temperatures of 20, 25, 30, 35 and 40 °C.

Photo images of chitosan and chitosan-collagen solutions were made with the Biolar PZO polarising microscopes equipped with a Nikon camera and computer image MultiScan analyser.

Cross-sections and surfaces of the fibres were inspected by means of the scanning-electron microscope (SEM/ESEM), Quanta 200 (W), FEI Co., USA.

Photometric infrared spectra were prepared on the FTIR Unicam apparatus equipped with the steering program Winfirst ATI Mattson. The samples were tested in the form of moulded pieces in potassium bromide of Aldrich Co.

Fibre mechanical properties were tested according to PN-ISO 1973:1997 and PN-EN ISO 5079:1999 standards in an air–conditioned room at a RH of 65 ± 4% and a temperature of 20 ± 2 °C.

The Kjeldahl method was applied for analysing the nitrogen content in the collagen-modified chitosan fibres.

Gel chromatography GPC/SEC was employed in the examination of the molecular properties of chitosan notably: function of the distribution of molecular mass (*MMD*), average values of molecular mass ($\overline{M}_n$, $\overline{M}_w$) and polydispersity ($\overline{M}_w/\overline{M}_n$). The GPC/SEC analysis was done in a buffer solution containing 0.2 M of sodium acetate and 0.3 M of acetic acid at a flow speed of 0.8 ml/min. The GPC/SEC system with the isocratic pump HP 1050 (Hewlett Packard) was equipped with the refractometric detector HP 1047 (Hewlett Packard). The injection

Table 1. *Chitosan properties.*

Parameter	ChitoClear® fg 90	ChitoClear® fg 95
Dry mater content, %	92.4	97.5
Ash, %	0.5	0.2
Degree of Deacetylation, %	93.0	95.0
Viscosity (1% chitosan), mPas	92	99
Solubility, %	99.5	99.8
Feature	100% through 100 mesh, 0.5 g/cc	100% through 100 mesh, 0.55 g/cc
Appearance	white powder	white powder
Taste and smell	no taste or smell	no taste or smell
Caliform bakterii	absent	absent
Salmonella sp	absent	absent
Toxic heavy metals	none detected	none detected

Table 2. *Properties of collagen.*

Type of collagen	Dissolvable collagen Type 2	Dissolvable collagen Type 3
Concentration of collagen, %	3.12	2.40
pH	3.2 - 4.2	3.5 - 5.0
Type of acid	citric	boric
source	protein of calf hide	protein of calf hide
appearance	opaque colorless fluid	opaque colorless fluid
Average molecular mass, Daltons	340.800	340.800
temperature of denaturation, °C	34	34
content of hydroxyproline, μg/ml	700	1500

volume of the solution of the tested sample was 100 µl at a concentration of approximately 0.05%. The separation of the macromolecules proceeded in the 2×PL aquagel OH Mixed, 300 mm long column system (Polymer Laboratories Ltd.) at a temperature of 30 °C. The calibration of the columns was made with the use of standards PEO/PEG from Varian Co in the range of 1970 D do 1,345,000 D. The results of the GPC/SEC analysis in the form of the distribution function of the molecular mass (*MMD*), average mass value $\overline{M}_n$, $\overline{M}_w$) and polydispersity ($\overline{M}_w/\overline{M}_n$) were calculated by applying the universal calibration method where the *a* and *k* parameters in the Mark-Houwink equation amount to: $a = 0.625$ and $k = 62\times10^{-5}$ ml/g for PEO/PEG and $a = 0.76$ and $k = 74\times10^{-5}$ ml/g [30] for chitosan.

Results and discussion

The distribution of molecular mass (*MMD*), average value of molecular mass (M_n, M_w) and polydispersity (M_w/M_n) were estimated for the chitosan grades ChitoClear®fg 90 and ChitoClear®fg 95 used in the preparation of fibres (see ***Figure 1*** and ***Table 3***).

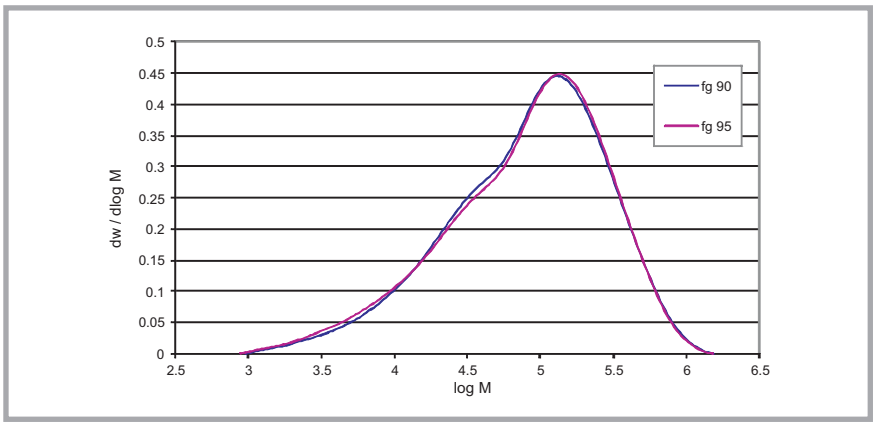

Figure 1. *Molecular weight distribution of ChitoClear chitosan used in the manufacture of fibres Chit/Col.*

Table 3. *Compilation of molecular mass and polydisperity values for chitosan.*

Parameter	Mn	Mw	Mw/Mn	Percentage of fractions, %						
	g/mol	g/mol		<5	5-50	50-100	100-200	200-400	400-800	>800
ChitoClear fg 90	31434	148908	4.7	3	29	20	23	17	7	1
ChitoClear fg 95	29570	148712	5.0	3	29	19	24	18	6	1

Table 4. *Some properties of chitosan spinning solutions.*

Symbol of solution	Chitosan grade	Glycerol concentration, %	Chitosan concentration, %	Dynamic viscosity / temp. Pas / °C
S/Ch - 2	ChitoClear fg 95	10	5.19	17.5 / 50
S/Ch - 3	ChitoClear fg 90	10	5.21	22.7 / 50
S/Ch - 4		10	5.14	19.0 / 50

Table 5. *Properties of chitosan-collagen spinning solutions (collagen type 3).*

Symbol of solution	Percentage of collagen in the polymer mix, %	Dynamic viscosity at 35 °C, Pas
S/Ch - 2	-	34.5
S/Ch - 2/Col 3/A	3.42	14.0
S/Ch - 2/Col 3/B	5.18	13.7
S/Ch - 2/Col 3/C	6.97	13.5

Table 6. *Properties of chitosan-collagen spinning solutions (collagen type 2).*

Symbol of solution	Percentage of collagen in the polymer mix, %	Dynamic viscosity at 35 °C, Pas
S/Ch - 4	-	36.0
S/Ch - 4/Col 2/A	4.94	9.0
S/Ch - 4/Col 2/B	8.55	8.5

The chitosan used in the investigations was varied in molecular mass and polydispersity. ChitoClear fg 95 shows a lower value of Mn and a higher viscosity of its 1% solution (***Table 1***) given by the manufacturer. Insignificant differences in the percentage of the individual fractions suggest high similarity between the lots of chitosan, which is confirmed in ***Figure 1***. The presented results and properties reported by the manufacturer present the full characteristics of the polymer. Considering also the properties of the fibres made, one can assess the suitability of a given chitosan to fibre spinning. Lower ash content, higher deacetylation degree and a higher viscosity (fg 95) promise a better suitability to the forming of fibre.

Aqueous solutions of chitosan in acetic acid (***Table 4***) were prepared for the forming of fibres. Chitosan was used with properties as shown in ***Table 1***.

Chitosan solutions with concentrations of 5.14 - 5.21% are characterised by dynamic viscosity in the 17.0 - 22.7 Pas range at a temperature of 50 °C. Higher viscosity values were found in the chitosan solution prepared from the chitosan with slightly lower molecular mass (ChitoClear fg 95) and higher concentration (5.19%). Filtration and de-aeration of the solution carried out at 50 °C did not present any problems. Microscopic inspection of the unfiltered solution revealed the presence of very few mechanical impurities.

Properties of chitosan-collagen solutions

When mixing the transparent light yellow chitosan solution (ChitoClear fg 95) with the opaque colourless colloid the al collagen solution, the dissolution of collagen could be observed. Both solutions mixed readily and the mixture turned to a transparent homogeneous chitosan-collagen solution. The properties of the blended solution are presented in ***Table 5***.

Chitosan-collagen solutions with collagen content in the range of 3.42 - 6.97% showed a dynamic viscosity of 13.5 - 14.0 Pas. Dynamic viscosity was decreased by 2.5-fold.

Chitosan grade ChitoClear fg 90 and colloidal solution of collagen type 2 were used in the consecutive trials. As in pre-

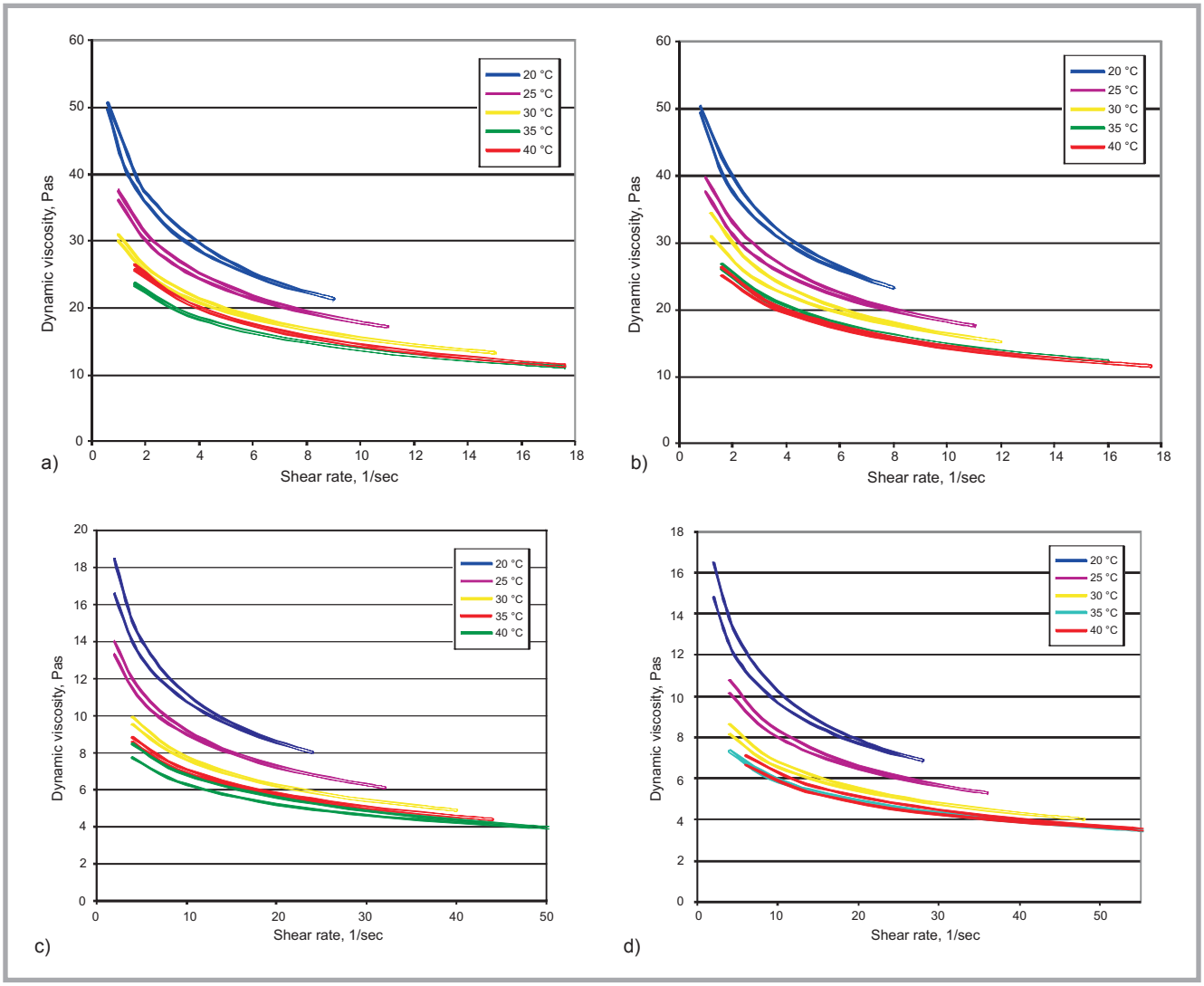

Figure 2. *Correlation between apparent dynamic viscosity, shear rate and temperature; for chitosan solution with symbol S/Ch – 3 (a) & after 24 h of storage at 20 °C (b), for chitosan-collagen S/Ch - 4/Col 2/A (c) & S/Ch - 4/Col 2/B (d) solution.*

ceding trials, the solutions mixed well in various proportions producing homogeneous chitosan-collagen blends (see ***Table 6***).

Microscopic inspection of the chitosan-collagen blend revealed the presence of a few objects with diameters in the range of few micrometres, which were probably mechanical impurities and were removed from solutions by filtration.

The solutions with the composition and properties shown in ***Tables 5*** and ***6*** were used in the spinning of Chit/Col fibres.

Investigation of the rheology of chitosan-collagen solutions

The rheology of the chitosan-collagen solutions at 20, 25, 30, 35 and 40 °C was examined to establish optimal conditions for the formation of fibres. The impact of shear rate and temperature upon apparent dynamic viscosity of the chitosan- and chitosan-collagen solutions is presented in ***Figure 2***. The flow curves were drawn at increasing and decreasing shear rates.

The chitosan solutions (***Figure 2.a***) reveal the character of non-Newtonian fluids in which the viscosity of the solution at 20 and 25 °C decreases with increasing shear rate. The flow curves coincide for both decreasing and increasing shear rates; solutions in that case show the character of a pseudo-plastic fluid which undergoes thinning by shearing, in a phenomenon that is typical to polymer solutions. The increase of temperature to 25 °C has a distinct impact upon the decrease of the apparent dynamic viscosity. The solutions tested at 35 and 40 °C also show a remarkable decrease of viscosity at increasing shear rate.

A chitosan solution left for 24 hours at 20 °C does not in fact change its rheological properties and behaves as before at increased temperatures and shear rate; an indication that the solution is still suitable for spinning after storage (see ***Figure 2.b***).

When collagen was introduced, the apparent dynamic viscosity dropped distinctly depending upon the amount of collagen added, due to the smaller viscosity of collagen related to the viscosity of chitosan solution. Increases of temperature to 30 °C and above cause a distinct decrease of the apparent dynamic viscosity (see ***Figure 2.c***).

Increase of the collagen in the collagen-chitosan solution (***Table 6***) has a rather insignificant influence on the decrease of viscosity. An elevation of the temperature of the collagen-chitosan solution

Table 7. Mechanical properties of chitosan and chitosan/collagen fibres.

Symbol of fibres		Linear density, dtex	Tenacity, conditioned, cN/tex	Elongation at break, conditioned, %
FS/Ch-2		7.82	5.88	18.0
		5.06	6.07	29.0
F S/Ch - 2/Col 3	A	4.14	7.21	25.0
		7.96	7.75	33.0
	B	3.35	10.0	33.0
		8.36	8.21	26.0
	C	3.78	9.40	29.0
		6.96	9.55	32.0
F S/Ch - 4/Col 2	A	3.30	6.85	29.0
		5.28	5.53	25.0
	B	3.35	6.66	26.0
		5.41	5.77	25.0

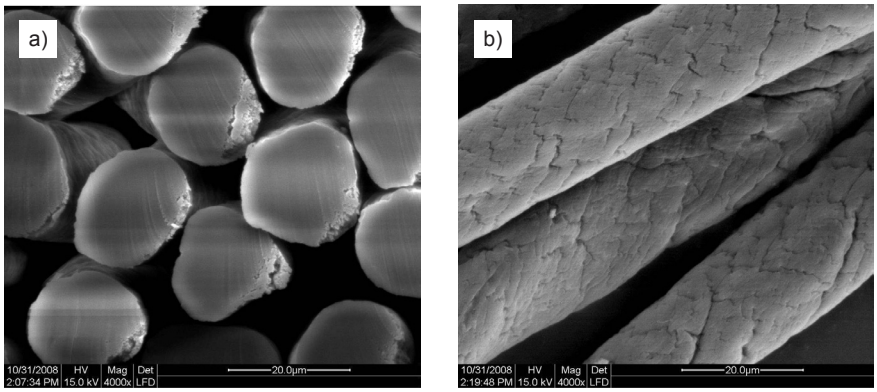

Figure 3. *SEM pictures of chitosan fibres; F S/Ch - 2.*

above the denaturation point of collagen did not cause any changes in its properties, such as gelatinising ability and viscosity. Structural changes of the collagen, like decay of bioactivity, could have happened as a result of the temperature. The result was a decrease of viscosity of the solution, which is an indication of suitability for the formation of Chit/Col fibres. Based on the rheology investigation, the temperature of 25 °C was adopted as adequate for the spinning of the Chit/Col fibres. Spinning performance could be better at higher temperatures, which, however, is not advisable considering the need to preserve the original properties of collagen.

Investigation in the spinning of collagen-modified chitosan fibres

Admixing of the collagen to the chitosan solution influences the spinning process, which is reflected in a lower tenacity of the fibres under wet conditions. The addition of ethanol to the alkaline coagulation bath resulted in a distinct improvement of spinning stability and wet tenacity of the fibres. A 34.0%, the draw ratio was applied during the spinning. The formation of the structure comes to an end in the course of the drying process which also confers the ultimate tenacity upon the fibre. Bearing in mind the potential use of the fibres in scaffolds, it is essential to provide fine fibres. The authors elaborated the process parameters for the spinning of Chit/Col fibres in the range of 3.30 – 8.36 dtex of the linear density, meaning that the minimal diameter of the fibres in the conditioned state decreases to 20 micrometres. A fibre with 3.30 dtex was prepared.

Mechanical properties of the collagen-modified chitosan fibres

Mechanical properties of chitosan fibres were tested in an accredited laboratory (see ***Table 7***) according to the standards in force.

The decline of linear density resulted in a slight increase of tenacity and elongation of the FS/Ch 2 fibres. Elongation was maintained in the range of 18 - 29%.

No problems arose during the spinning of the Chit/Col fibres; the process proceeded smoothly. Mechanical properties of the Chit/Col fibres are shown in ***Table 7***.

An increase in tenacity and elongation was found in the Chit/Col when compared to regular chitosan fibres (***Table 7***). The presence of collagen type 3 exerted a positive influence on the mechanical properties, regardless of linear density. The highest increase of tenacity was found for the fibre with a 5.18% content of collagen**.**

Employing chitosan ChitoClear fg 90 and collagen type 2 caused a drop of tenacity to the value of 6 cN/tex; tenacity was further lowered as a result of the decrease in linear density to 5.41 dtex, while elongation remained high, at above 25%.

Fibres of chitosan ChitoClear fg 95 and collagen type 3 are characterised by higher tenacity and elongation compared to those made of ChitoClear fg 90 and collagen type 2. This may be a result of higher collagen content in the spinning solution (see ***Tables 5*** and ***6***).

Examination of fibre morphology

In ***Figure 3***, SEM pictures of chitosan fibres are shown.

The cross-section of chitosan fibres is presented in ***Figure 3.a***. The fibres are characterised by a regular, close to circular shape of the cross-section. ***Figure 3.b*** depicts the longitudinal view of the fibre, which reveals the characteristic regular cavities and furrows on the fibre's surface.

In ***Figure 4*** presented are SEM pictures of Chit/Col fibres.

Chit/Col fibres are characterised by an oval, less regular shape of the cross-section. The surface structure is clearly marked with shallower furrows and cavities. Also seen are regular cracks on the surface. With a higher amount of collagen in the fibre the cracks become deeper, and the amount of fibres sticking to each other increases.

The investigation confirmed the impact of chitosan concentration in the spinning solution (presence of collagen) on the morphological structures of Chit/Col fibres. The lowest diameter of the Chit/Col fibres that could be achieved was 20 μm. The morphology of the fibres may display an impact on their application in the construction of scaffolds.

Examination of the chemical structure

It may be assumed that, during the solidification of the streaming chitosan and

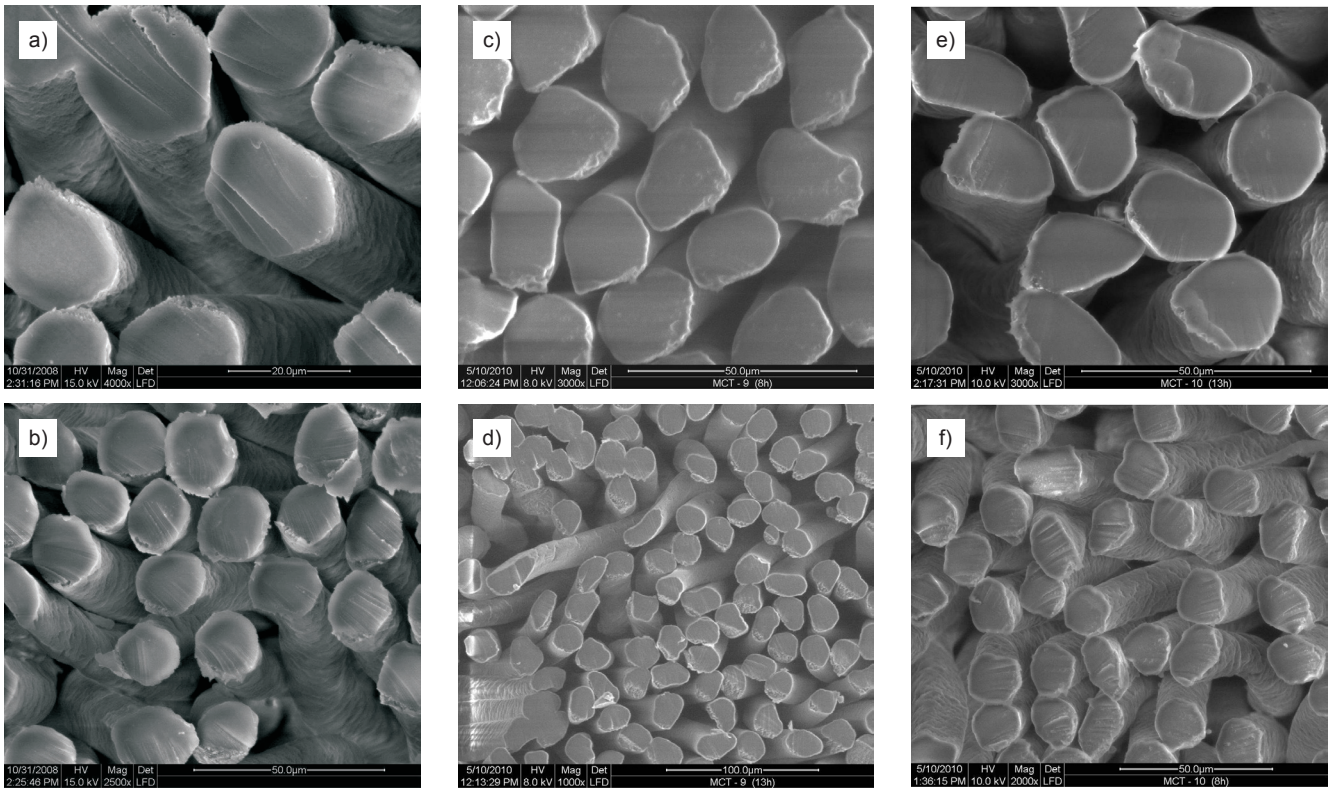

Figure 4. *SEM pictures of Chit/Col fibres with symbols: a, b - F S/Ch - 2/Col 3/C, c, d - F S/Ch - 4/Col 2/A: e, f - F S/Ch - 4/Col 2/B.*

collagen solutions, differences emerge in the diffusion rate of the solvent and coagulation bath molecules. The spun fibres appeared to be poorly solidified in the spinning trials, and they were prone to mechanical deformation in the wet state.

Testing of nitrogen content was performed for selected chitosan and Chit/Col fibres as shown in ***Table 8***.

Nitrogen content in chitosan- and Chit/Col fibres is in the range 6.92 - 7.44%. There are, in most cases, insignificant differences resulting from the kind of chitosan and inhomogeneity of the material lots. The influence of collagen content in the fibres on the nitrogen percentage is rather feeble. The examinations confirmed the presence of collagen in the Chit/Col fibres. The collagen content in the fibres has a minor influence on the nitrogen percentage; The FTIR examination described below confirmed the presence of collagen in the Chit/Col fibres. FTIR spectra of Chit/Col fibres are shown in ***Figure 5*** see page 38.

The presence of groups that are characteristic for chitosan is confirmed by the intensity of peaks at given wavelengths, notably: NH_2 (1506 cm^{-1}), amide I (1637 cm^{-1}), amide II (1559 and 1414 cm^{-1}), amide III (1319 cm^{-1}). Anti-symmetrical stretching vibrations are characteristic for C-O-C bonds (1160 cm^{-1}), stretching vibrations for N-H bonds (3298, 3500 cm^{-1}), and stretching vibrations for O-H bonds (3445 cm^{-1}). Highly intensive peaks were observed at a wavelength of 1160 cm^{-1} reflecting the anti-symmetrical stretching vibrations of C-O-C bonds. Wavelengths of 1032 and 1086 cm^{-1} show the presence of symmetrical stretching vibrations of C-O bonds, which are characteristic to sugars [32 - 34], while the peaks at 2600 and 3566 and 3700 cm^{-1} highlight the stretching vibrations of the – OH group in chitosan.

The peaks of collagen confirm the presence of the functional groups C=O, amide I (1658 cm^{-1}), N-H amide II bending vibrations and C-N stretching vibrations (1554 cm^{-1}), C-N and N-H amide III (1240 cm^{-1}) [35, 36].

The FTIR analysis of Chit/Col indicates amide bonds I, and stretching vibrations indicate C=O (1600 - 1690 cm^{-1}) at a wavelength of 1652 cm^{-1}, which correlate with the nitrogen content in the fibres. The presence of amide bonds II, stretching vibrations for CN=O and bending vibrations for NH (1480 - 1575 cm^{-1}) are also observed at a wavelength of 1517 cm^{-1}, which correlates with nitrogen content in the fibres. Very distinct changes can be seen in the intensity of peaks at a wavelength of 1320 cm^{-1} which correspond with the amide bonds of chitosan. The intensity of the peak lowers with an increasing content of collagen. The high intensity of the peak at 1168 cm^{-1} for chitosan fibres decreases with the increasing content of collagen in the fibres.

The presence of amide bonds I and II in the Chit/Col fibre provides a convenient condition for the formation of hydrogen bonds. Change of absorbency values at wavelengths of 2900 and 3450 cm^{-1} witness the presence of both intra- and intermolecular hydrogen bonds. The intensity of the 3445 cm^{-1} peak lessens with increasing collagen content.

Table 8. *Nitrogen content in chitosan- and Chit/Col fibres.*

Symbol of fibre	Nitrogen content, %
F S/Ch - 2	7.03
F S/Ch - 2/Col 3/A	7.24
F S/Ch - 2/Col 3/B	7.35
F S/Ch - 2/Col 3/C	7.44
F S/Ch - 4	6.92
F S/Ch - 4/Col 2/A	7.08
F S/Ch - 4/Col 2/B	7.17

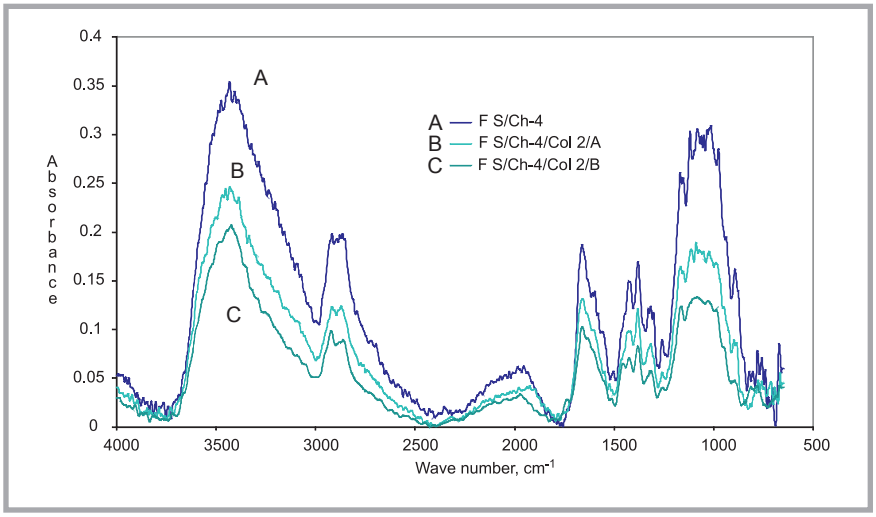

***Figure 5**. FTIR spectra of Chit/Col fibres; A) F S/Ch – 4, B) F S/Ch - 4/Col 2/A, C) F S/Ch - 4/Col 2/B.*

The presented results of the investigation gave evidence that the commercial ChitoClear chitosan is a suitable starting material for the preparation of chitosan fibres. The impact of the concentration of ChitoClear fg 95 in the solution upon the tenacity of the fibres was also confirmed. Collagen type 2 and 3 derived from calf hide buffered with citric or boric acid are the correct materials for the preparation of chitosan-collagen solutions suitable for the spinning of Chit/Col fibres. The portion of collagen up to 5.18% in the chitosan fibres positively influences the tenacity and elongation of the fibres.

Conclusions

1. The presented method permits the preparation of collagen-modified chitosan fibres with tenacity and elongation higher than in regular chitosan fibres.
2. Chitosan-collagen solutions are suitable for the formation of Chit/Col fibres.
3. A better suitability of ChitoClear fg 95 chitosan and collagen type 3 was demonstrated for the preparation of Chit/Col fibres.
4. The presence of collagen in the Chit/Col fibres was confirmed by an increased content of nitrogen and by the examination of the morphological structure.
5. Chit/Col fibres with the lowest diameter of 20 micrometres were prepared.

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