

Adsorption and Antibacterial Activity of Soluble and Precipitated Chitosan on Cellulose Viscose Fibers

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ABSTRACT

The aim and novelty of this work was to compare the adsorption of totally-soluble chitosan (acidic solution) against the adsorption of precipitated chitosan, onto cellulose fibers. The influences of both these chitosan-adsorption procedures on a final amino group's content in functionalized cellulose fibers were studied, using potentiometric titration and the conventional spectrophotometric C.I. Acid Orange 7 method. Surface modification and adsorption of chitosan were, in addition, monitored by determining XPS spectra. The antimicrobial activities of both chitosan- functionalised cellulose fibers were examined, in regard to pathogen bacteria and fungus.

Keywords: Functionalized viscose; chitosan solution; precipitates; potentiometric titration; spectrophotometry; antimicrobial activity

INTRODUCTION

Nowadays the development of high-quality hygienic and medical textiles is an important topic. Medical textiles are a major growth area within the technical textile industry and the range of applications continues to grow and diversify with every new development. Although the type of fiber used and the fabric structure varies according to the specific end-use, all medical fibers must be non-toxic, non-carcinogenic, non-allergic, and capable of being sterilized without suffering chemical or physical damage. In addition, absorbency is essential for many applications, when favouring the use of cotton or viscose cellulose materials. Cellulose fibers have a large active surface area and, due to their molecular structure, may be considered as an ideal matrix for the design of bioactive, biocompatible, and intelligent materials [1-3]. In regard to increasing humans' awareness of their health and well-being, there is a significant interest in cellulose materials' functionalization using natural biodegradable and non-toxic reagents – as, for example, less employed polysaccharides and their derivatives possessing antimicrobial and antifungal properties. Among the

various polysaccharide products used as antimicrobial substances, amino-functional polysaccharides are the most promising for many applications [4], including medical textiles. The amino groups interact with the cell surfaces of pathogen microorganisms and, in this way, destroy them by several possible mechanisms [5]. One of the most popular amino polysaccharides is chitosan, as obtained by the alkaline deacetylation of chitin. Chitosan's positive charge, the degree of N-deacetylation, the mean polymerization degree, and the nature of chemical modifications are those properties which strongly influence its antimicrobial effectiveness [5-11]. Many techniques are available for introducing chitosan onto cellulose fibers. These methods have been mainly limited to the treatment of cellulose fibers with cross-linking agents [12], the soaking of cotton fibers into chitosan solutions, or impregnation by chitosan solutions, etc. [13-14]. The adsorption of chitosan and its interactions with cellulose material are not only affected by the surface physicochemical characteristics of the cellulosic substrates but may also be strongly influenced by the chitosan aggregate's nature. The introduced amounts of amino groups onto cellulose fibers and, consequently the fiber antimicrobial activity, may increase with the attachment of polysaccharides in colloidal form, in comparison with attaching a totally-soluble form of chitosan, such as chitosan acidic solution.

The solubility of chitosan depends on the protonation of the amino groups. Hence, any method which removes or masks the charge of a sufficient number of free amino groups may be used to precipitate chitosan particles. In this work, they were simply precipitated by neutralization with sodium hydroxide.

The aim and novelty of this work was to compare the adsorption of fully soluble chitosan (acidic solution) with the adsorption of precipitated chitosan particles, onto cellulose viscose fibers. The content of amino groups in the functionalized cellulose fibers was

studied using potentiometric titration, and a conventional spectrophotometric method C.I. Acid Orange 7. The antimicrobial activities were examined in regard to the pathogen bacteria and fungus of the functionalised cellulose fibers.

EXPERIMENTAL

Materials

Viscose Fibers

Regenerated viscose cellulose fibers were obtained from Lenzing AG, Austria. The linear densities of all fibers were 1.3 dtex and the fiber lengths were 39 mm.

Pre-Treatment of Viscose Fibers

Efficient cleaning of the fibers is very important for clarifying the origins of any positively- charged groups present in functionalized fibers. Conventional cleaning procedures commonly used as pre-treatment processes in textile praxis were used in this research: i) alkaline washing, ii) bleaching and iii) demineralisation. Alkaline washing is aimed to remove oils, antistatic agents, etc. Bleaching decomposes substances such as colours, pigments, etc. which are hard to remove by alkaline treatment. The demineralization process is required in order to remove metal ions and to convert fibers into their H-form. These procedures are listed in *Table I*. Following this treatment, the samples were rinsed using distilled water until the conductivity of the rinsing water was less than 3 $\mu\text{S}/\text{cm}$. The processed material was air-dried and air-conditioned ($T = 20\text{ }^\circ\text{C} \pm 2\text{ }^\circ\text{C}$ and relative humidity = 65 % $\pm 2\text{ }%$).

TABLE I. Treatment conditions for viscose cellulose fibers' pre-treatment procedures.

Alkaline washing	Bleaching	Demineralisation
1 g/L Na_2CO_3	6 mL/L H_2O_2	0,01 mol/L HCl
1 g/ L Sandoclean PC (wetting agent, anionic)	2 mL/L Tanatex Geo (mineral stabiliser for H_2O_2 stabilisation)	
BR = 1 : 20	BR* = 1 : 20	BR = 1 : 100
pH 10.9	pH 10.7	pH 2
t = 30 min	t = 30 min	t = 30 min
T = 60 $^\circ\text{C}$	T = 98 $^\circ\text{C}$	T = 22 $^\circ\text{C}$

*BR = bath ratio

Chitosan

The chitosan used for fiber impregnation was chemically-graded, low-viscous chitosan (M= 200000; 90–95% deacetylated) from Gillet Chitosan, France. It was used without further purification.

Other Materials

All the chemicals were purchased by Sigma Aldrich (HCl, NaOH, and Na_2CO_3), Kemika Zagreb (KCl), Baker Dilut-it (KOH); Sandoz (Sandoclean PC), Tanatex (Tanatex Geo) and Belinka (H_2O_2) and were commercially-graded as well.

Functionalization of Viscose Fibers

Impregnation of Fibers Using Chitosan Acidic Solution

Chitosan was adsorbed onto viscose fibers by soaking the fibers (5 g) in a 0.5 % of 100 ml of acidic chitosan solution (pH adjusted to 3.6 by adding 1 M hydrochloric acid) for 10 min as described by YS Chung et al. (1998) [15]. The fiber impregnation was performed by passing through a foulard impregnation press (W. Mathis) at a pressure of 1.6 bars. After this treatment the fibers were dried at $T = 40\text{ }^\circ\text{C}$ and $t = 60\text{ min}$, and further air-conditioned for 48 h. Finally, the functionalized fibers were washed with distilled water until the conductivity of the water was less than 3 $\mu\text{S}/\text{cm}$. The processed material was air-dried and air-conditioned ($T = 20\text{ }^\circ\text{C} \pm 2\text{ }^\circ\text{C}$ and relative humidity = 65 % $\pm 2\text{ }%$).

Impregnation of Fibers using Chitosan Precipitate

The chitosan was precipitated onto the fibers by suspending viscose fibers (5 g) in rapidly stirred acidic solutions of chitosan (0.5 %, 100 ml), followed by the addition of sodium hydroxide (0.1 M NaOH). Precipitation of the fibers was followed by a turbidimetric titration experiment using a Mettler Toledo DL 53 titrator, and a Mettler Toledo Phototrode DP660 (wavelength of 660 nm).

The chitosan precipitated around pH = 6.8–7.0. The fibers were left in the chitosan precipitate suspension for 30 min and then passed through an impregnation-wringing machine (Foulard W. Mathis) at pressure 1.6 bars. After this treatment, the fibers were dried for 60 min at $T = 40\text{ }^\circ\text{C}$ and further air-conditioned for 48 h. Finally, the functionalized fibers were washed until the constant water conductivity was less than 3 $\mu\text{S}/\text{cm}$, demineralized, air dried, and stored under standard atmospheric conditions ($T = 20\text{ }^\circ\text{C} \pm 2\text{ }^\circ\text{C}$ and relative humidity RH = 65 % $\pm 2\text{ }%$). The samples' notations are given in *Table II*.

TABLE II. Sample notation.

Sample	Treatment of fibers
CH	Chitosan
CV	Pre-treated viscose fibers: alkaline washed, bleached, demineralized
CV – CH _{sol}	Viscose fibers treated by 0.5 % chitosan acid solution
CV – CH _{prec}	Viscose fibers treated by 0.5 % chitosan precipitates

Analytical Methods for Characterization of Chitosan-Functionalized Cellulose Fibers

Potentiometric Titration

A twin burette instrument (Mettler T-70) was used, equipped with a combined glass electrode (Mettler T DG 117). The burettes were filled with 0.1M HCl (Merck, Titrisol) and 0.1 M KOH (Baker, Dilut-it). All the solutions were prepared with deionized water with a very low carbonate content ($< 10^{-6}$ M), which was achieved through boiling, and subsequent cooling, within a nitrogen atmosphere.

The pure chitosan solution, pre-treated viscose fibers, and fibers functionalized in chitosan solution with chitosan precipitates (0.5 g of oven dry fibers), were all titrated in forward and back runs between pH = 3 and pH = 10. In order to avoid any sticking to the electrode and jamming with the stirrer, the fibers were kept in a stainless steel tea container. Prior to the titrations, the ionic strength was set at 0.1 M, by adding pure solid KCl (Kemika, Zagreb). The ionic strength therefore remained within 2 % of the initial value upon the additions of HCl and KOH. The blank HCl-KOH titrations were performed under the same conditions as above.

The equilibrium criteria for the timed addition was set at $dE/dt = 0.1$ mV/min. The total amount of weak acidic groups was calculated from the difference (ΔV) in the added KOH volume between the fiber sample (V_i) and the blank ($V_{i,Blank}$), and any given pH.

The molar concentration Q related to the overall charge of the weak ions was calculated from the charge balance equation:

$$Q = \sum_i c_i z_i = [OH^-] - [H^+] + [Cl^-] - [K^+] \quad (1)$$

where square brackets and c_i denote molar concentrations of ionic species, and z_i is the charge number of the species i . For a detailed description of the charge calculation, see Čakara *et al.* [16]. All

reported values are the mean values of triplicate determinations.

Spectrophotometric C.I. Acid Orange 7 Method

The adsorption of dye C.I. Acid Orange 7 (purified by recrystallization) into the reference and functionalized viscose fibers coated with chitosan acidic solution or chitosan precipitates (colloidal particles), respectively, was evaluated by determining the dye concentration in the solutions (pH 3.6 ± 0.5) in contact with the fibers, using on-line optical absorbance measurement, at a wavelength of maximum absorbance (484 nm). Equilibrium was reached in 6 h. The solutions were kept in a thermostat at (25 ± 0.5) °C and stirred with a magnetic stirrer. A Cary 50 Conc computer controlled UV spectrometer from Varian was used for this experiment. For this adsorption kinetics experiment, 0.25 g of viscose fiber was stirred with an acidic dye solution of 4×10^{-4} M. This procedure has been found to give a 100 % stoichiometric reaction at dye anionic sites with protonated amino groups along the chitosan polymer chains [17]. Therefore, the results are presented as the amount of amino groups per kg of fiber.

XPS Analysis

XPS spectra were recorded using the PHI model TFA XPS spectrometer at the Laboratory of Surface and Thin Film Analysis at the Jozef Stefan Institute, Ljubljana, Slovenia. The atomic composition was measured after chitosan treatment, and then compared to the elemental chemical composition of the surface of the non-treated viscose material. The base pressure in the XPS analysis chamber was about 6×10^{-10} mbar and the samples were excited with X-rays over a specific 400- μ m area using monochromatic Al $K\alpha_{1,2}$ radiations at 1486.6 eV. The photoelectrons were detected by a hemispherical analyzer, positioned at an angle of 45° with respect to the sample's surface normal. Energy resolution was about 0.6 eV. Survey-scan spectra were created at pass energy 187.85 eV. The spectra were recorded from at least three locations on each sample, using an analysis area of 1 x 1 mm. Surface elemental concentrations and O/C ratios were calculated from the survey -scan spectra. An additional electron gun for surface neutralization was used during the measurements, in order to compensate for the charging of the non-conducting samples. Three repetitions of the measurement were done for each sample.

Antimicrobial Test

The antimicrobial properties of functionalized fibers were evaluated by ASTM E2149-01 (Standard Test

Method for Determining the Antimicrobial Activity of Antimicrobial Agents under Dynamic Contact Conditions), which is a quantitative antimicrobial test method performed under dynamic contact conditions. Gram-positive and Gram negative bacteria, as well as a fungi, were used as test organisms. An incubated test culture in a nutrient broth was diluted using a sterilised 0.3 mM phosphate buffer (KH_2PO_4 ; pH = 6.8) in order to give a final concentration of $1.5\text{--}3.0 \times 10^5$ colony forming units (CFU)/mL. This solution was used as a working bacterial dilution. Each sample (0.5 to 2 g) was cut into small pieces (1×1 cm) and transferred to a 250 mL Erlenmeyer flask containing 50 mL of the working bacterial dilution. All flasks were loosely capped, placed in the incubator, and shaken for 1 h at 37 °C and 120 rpm using a Wrist Action incubator shaker. After a series of dilutions using the buffer solutions, 1 mL of the diluted solution was plated in nutrient agar. The inoculated plates were incubated at 37 °C for 24 h and the surviving cells counted. The average values of the duplicates were converted to CFU/mL in the flasks, by multiplying with the dilution factor. The antimicrobial activity was expressed as $R = \%$ reduction of the organism after contact with the test specimen, compared to the number of bacterial cells surviving after contact with the control [14].

RESULTS

Potentiometric Titration

Reference Fibers

Figure 1 contains data from the titration of pure viscose fibers, alkaline washed fibers and viscose fibers that were alkaline washed and further bleached and demineralized. The charging isotherms are normalized with the mass of the fabric m_f , in order to facilitate comparison of data from the three experiments.

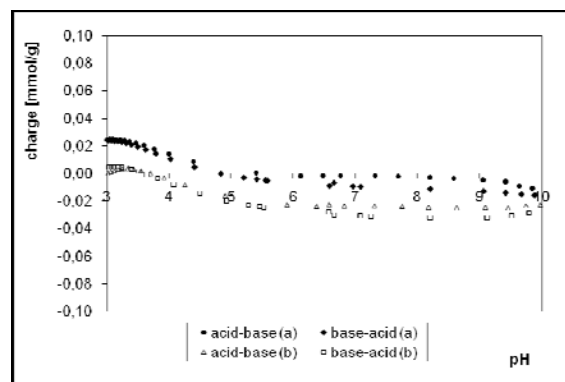


FIGURE 1. Potentiometric titration of alkaline washed viscose fibers (a), potentiometric titration of viscose fibers alkaline washed, bleached, and demineralised (b).

In order to obtain reliable analysis for the effect of chitosan impregnation, it is necessary to remove all impurities present on the referenced fiber's surface. The isotherms of alkaline washed fibers exhibit, in addition to negatively-charged groups, presumably carboxyl groups, a positive charge with a plateau at ca. 0.024 mmol/ g below pH ≈ 4 . Cellulose does not contain such groups; therefore, it is probable that the positive charge (cationic groups) originates from different fiber treatments and impurities associated with viscose fiber production. After bleaching and demineralization of the washed fibers the titration indicates the presence of anionic groups only, giving a negative charge with a plateau at ≈ 24 mmol/kg at pH > 6 . It can be assumed that the negative charge on the fibers is due to deprotonated carboxyl groups that may originate from terminal groups in cellulose molecules or from the oxidation of the cellulose (bleaching procedure) [4]. The amount of carboxyl groups determined by potentiometric titration is given in Table III. The pK value was calculated as described by Ćakara et al. [16]. pK = 3.8, which is typical for the glucuronic acid groups in cellulose.

Viscose Fibers Functionalized with Chitosan

Figures 2 and 3 show charging isotherms vs. pH for pure chitosan and viscose fibers functionalized by chitosan acidic solution and chitosan precipitates, respectively. Figure 2 shows the expected curve for a pure chitosan with a point of zero charge (PZC) at pH = 6.8.

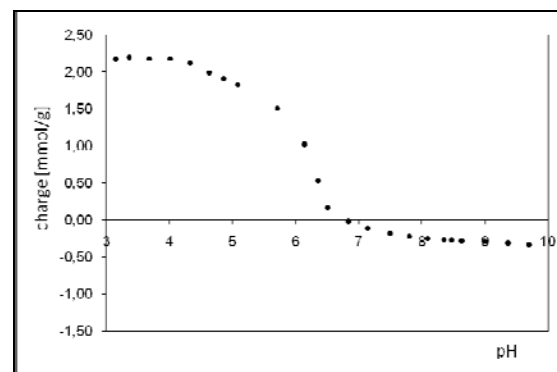


FIGURE 2. Potentiometric titration of chitosan solution, $c = 0.50$ % (w/v).

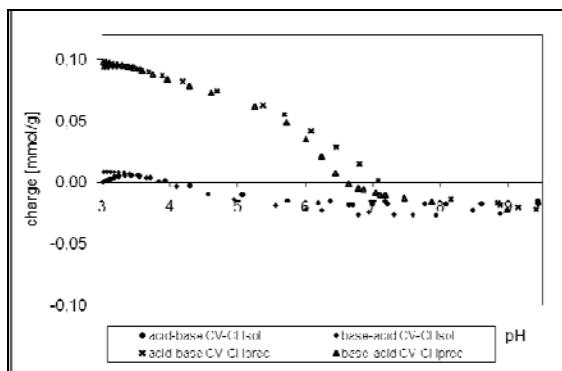


FIGURE 3. Potentiometric titration of viscose fibers, impregnated (CV-CH_{sol}) and precipitated (CV-CH_{prec}) with chitosan.

In *Figure 3* it is obvious that, in the case of fiber functionalization by the adsorption of dissolved chitosan from acid solution (CV-CH_{sol}), only a small amount of chitosan was bound to the fibers. Charge reversal (PZC) occurs in this case, at pH \approx 4, which indicates that its charge reversal is mainly due to the dissociation of carboxyl groups in the fibers. The PZC of fibers functionalized by precipitated chitosan (CV-CH_{prec}) is higher and occurs at pH \approx 6.6, i.e. almost identically as for free chitosan (*Figure 2*). The amount of amino groups in those fibers functionalised by chitosan precipitates was 97 mmol/kg, while in fibers impregnated by chitosan solution it was only 7 mmol/kg (*Figure 3*). While amino groups can be introduced onto the fibers by both methods, the use of chitosan precipitates is obviously better in the sense that it gives a much higher amount of amino groups responsible for the antimicrobial effect of chitosan [15].

Chitosan molecules have different conformational changes as a function of pH. At higher pH polymer chains become less extended with a smaller radius of gyration. This may be a reason for higher adsorbed amount of chitosan onto fibers in the case of precipitation procedure, and consequently higher fiber charge density (amino group content).

Moreover, it is likely that chitosan precipitates provide a larger surface area, which gives higher accessibility for the detection of total amino groups. Fiber negative charge as a result of carboxyl groups' presence was also determined from the charging isotherms (*Table III*). The effect of carboxyl groups from the impregnation of chitosan acidic solution is too small to be detected, and the somewhat lower amount of -COOH detected with precipitated chitosan (40 % less carboxyl groups than in fibers impregnated by acidic chitosan solution) is due to ionic bonds between the carboxyl and amino groups

in the chitosan. The charge calculated from charging isotherms is listed in *Table III*.

TABLE III. Positively and negatively-charged functional groups of chitosan (mmol/g of powder), reference fibers, viscose fibers impregnated by chitosan solution, and viscose fiber functionalised by chitosan precipitates (mmol/kg of fibers).

Sample	Positive charge	Negative charge
	[mmol/g/kg]	[mmol/g/kg]
CH	2.2	0.2
CV	1.4	24.0
CV - CH _{sol}	7.1	21.7
CV - CH _{prec}	96.7	13.6

The variation coefficient for the potentiometric results is within the region of 2 % - 5 %.

Spectrophotometric C.I. Acid Orange 7 Method

Figure 4 illustrates the results for amino groups' amounts of non-functionalized and functionalized fibers acquired by the C.I. Acid Orange 7 method. The obtained results are in accordance with potentiometric titration results (*Figure 4*) and further demonstrate that the adsorption of chitosan-precipitates onto fibers is more efficient than the adsorption of fibers by chitosan acidic solution. The amino groups' amount for viscose fibers functionalised by chitosan precipitates was 68.2 mmol/kg, whilst for fibers functionalised by chitosan solution only 9.1 mmol/kg. The referenced viscose fibers (pre-treated) had 1.5 mmol/kg of positive charge, which was caused by an insignificant quantity of C.I. Acid Orange 7 dye attached to the fibers due to possible hydrogen bonds or Van der Waals forces, at the beginning of experiment.

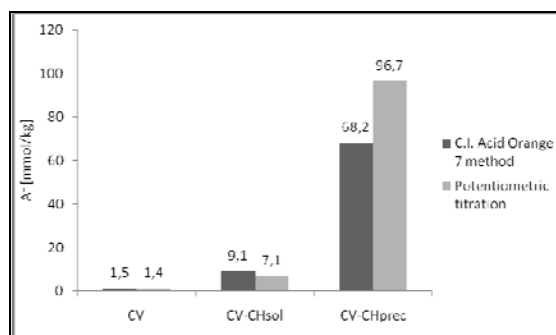


FIGURE 4. The amount of amino groups obtained by using C.I. Acid Orange 7 method.

The variation coefficient for results obtained by using C.I. Acid Orange 7 method is within the region of 8 % - 10 %.

XPS

The elemental surface compositions of non-treated, and both chitosan -treated viscose samples, are shown in *Table IV*. As the analysis depth (escape depth/attenuation length of electrons) in XPS is less than 10 nm [18-20], the results describe the composition of a limited number of molecular layers on the outermost fiber surfaces. Note that XPS does not detect hydrogen atoms. Analysis of the non-treated (reference) sample indicated that the surface consisted of about 43.3 atom% of oxygen and 58.1 atom% of carbon. For pure cellulose one would expect 45.5 atom% of oxygen and 54.5 atom % of carbon [18-21]. The O/C ratio of the non-treated sample is near to the theoretical cellulose value (0.83), indicating that by using all the above mentioned pre-treatment processes, resulted in effective cleaning of the viscose fibers surface, with negligible amounts of impurities.

TABLE IV. The elemental surface composition of non-treated and both chitosan treated viscose fibers.

Sample treatment	Surface composition (at. %)		
	C	O	N
CV	58.1	43.3	0.3
CV – CH _{sol}	62.4	36.1	1.1
CV – CH _{prec}	62.4	33.3	1.8

Significant amounts of nitrogen were detected only in those samples treated with chitosan (CV – CH_{sol} and CV – CH_{prec}). Thus, the nitrogen must be due to those amino groups introduced onto the viscose fibers by the adsorption of chitosan, i.e. XPS confirms that chitosan was adsorbed on all fiber surfaces treated with chitosan solutions and chitosan precipitates. The results listed in *Table IV* also clearly show that the treatment of fibers by chitosan precipitates increases the ability of fibers to adsorb chitosan and, hence, are in agreement with potentiometric titration and C.I. Acid Orange 7 method's results. The increase in the amount of N (atomic percentage) in samples treated by chitosan precipitates (CV – CH_{prec}) compared to samples treated by chitosan solution (CV – CH_{sol}) was about 63 %.

Comparison of Methods

Both wet analytical methods used, the potentiometric titration and the spectrophotometric C.I. Acid Orange 7 method, gave comparable results for the amino groups' amounts present in functionalized viscose fibers (see *Figure 4*). There is a satisfactory correlation between both techniques. For sample CV – CH_{prec}, however, there is an exception where the determined amounts of amino groups within the chosen methods varied by 28.5 mmol/kg, which might be explained by the fact that method C.I. Acid Orange 7 is an indirect method, whilst titration is a direct method, for the determination of dissociable functional groups. Obviously C.I. Acid Orange 7 indirect method does not satisfy the criteria of 100% stoichiometric reaction. This may be explained by accessibility of determined fiber amino groups. Potentiometric titration is based on titration of weak acid groups, such as amino groups, with small OH ions, while the spectrophotometric method is based on adsorption of a larger dye molecule (molecular weight of 350.3 g/mol) which cannot access all of the amino groups distributed in the fibers.

Therefore, in the case of higher charge fiber density (amino group content) it is more difficult to access all of these groups with Acid Orange 7 dye. XPS analysis shows some additional details for the surface chemistry of functionalised fibers. It is obvious that the trend for both “wet” techniques results (for amino group content determination in the bulk) and XPS (for nitrogen content determination on limited fiber surface layer) is almost the same. The XPS method confirms that the surface concentration of nitrogen as a consequence of adsorbed chitosan is considerably lower in sample CV – CH_{sol} than in CV – CH_{prec}. The use of titration methods in combination with XPS analysis seems to be very promising for observing the influence of chemical functionalization on the fiber surfaces in order to develop bioactive properties.

Antimicrobial Test

The aim of antimicrobial testing was to examine the influence of amino group amounts on the antimicrobial properties of functionalized viscose fibers. Furthermore, it was in our interest to analyse the influence of chitosan aggregation nature (solid, liquid) as a fiber adsorbent, on the final antimicrobial properties. Both functionalised viscose fibers were tested by using the ASTM 2149-01 dynamic shake flask method. The results of *in vitro* growth inhibition for the various microorganisms are shown in *Table V*.

TABLE V. Reductions in pathogenic bacteria and fungi.

Sample	Reduction R [%]				
	Pathogenic bacteria			Pathogenic fungi	
	S. <i>aureus</i>	E. <i>coli</i>	S. <i>agalactiae</i>	C. <i>albicans</i>	C. <i>glabrata</i>
CV	0	0	80	20	30
CV-CH _{sol}	90	0	100	78	88
CV-CH _{prec}	100	0	100	86	78

The variation coefficient for all results is less than 5 %. Both chitosan-functionalized viscose fibers successfully reduced pathogenic bacteria, such as *S. aureus* and *S. agalactiae*, and fungi, *C. albicans* and *C. glabrata*, but were completely ineffective against the pathogenic *E. coli*. Similar results were obtained by Gao and Cranston (2008) [1]. There is almost no difference in antimicrobial activity between both samples, i.e. CV-CH_{sol} and CV-CH_{prec}. The adsorption of chitosan precipitates onto fibers slightly improved the reduction of pathogenic bacteria *S. aureus* and fungus *C. albicans* in comparison with the adsorption of chitosan from solution. The reduction of *C. glabrata* with sample CV-CH_{prec} was about 12 %, lower than with CV-CH_{sol}. It is interesting that the amino group amount did not proportionally influence the antimicrobial activity of the fibers. Obviously, the already small amount of amino groups introduced onto the fibers is sufficient for antimicrobial activity. Several high-quality investigations have examined the relationship between the amino group content of chitosan solution and its antibacterial activity. When the functionalized fiber (viscose-chitosan) system is examined from an antimicrobial point of view, several other parameters are obviously important besides the amino groups' amount; such as fiber structure (porosity), fiber acidity, its hydrophobicity/hydrophilicity, etc. [14]. Therefore, the microbiological results for chitosan adsorbed onto fibers are often widely conflicting [7], owing to inter- and intra-assay variation when susceptibility testing. Detailed future investigations are needed on this topic in order to understand the mechanisms regarding the antimicrobial activity (microorganisms' inhibition) of chitosan-functionalised fibers.

CONCLUSIONS

The most important conclusion of this study is that both fibers (functionalized by chitosan solution, and chitosan precipitates) show antimicrobial capacity (R > 75 %) against Gram positive bacteria, and may be applied in the future as new antimicrobial sanitary products or medical textiles. In addition, the following conclusions can be made:

- Determinations of the amount of amino groups on the functionalized fibers analysed by potentiometric titration and spectrophotometry, using C.I. Acid Orange 7, gives comparable results.
- XPS spectra yield a qualitative estimation of bonded chitosan onto viscose fibers and support the "wet" chemical analyses.
- The combination of the spectrophotometry method and potentiometric titration with XPS appears to be a very powerful approach when evaluating in detail, how chitosan is applied as a fiber coating influences a fiber amino group content. The functionalization of viscose fibers using precipitated chitosan is more efficient, in contrast to the functionalization of fibers with acidic chitosan solution in the sense that it contributes more amino groups but, surprisingly, this evidently does not influence the growth of pathogen microorganisms.

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