

Preparation of chitosan powder and investigation of its properties

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Abstract. For the preparation of chitosan powder, the thermocatalytic destruction method developed in our earlier works was applied. Chitosan samples obtained from exoskeletons of crustaceans with a molecular weight of 500 kDalton, deacetylation degree 80% and with a molecular weight of 900 kDalton, deacetylation degree 70% were investigated. For these samples, optimal thermocatalytic treatment conditions for partial destruction of chitosan, which was controlled from the change in intrinsic viscosity, to the levelling-off degree of polymerization (LODP) were developed. In our case, LODP was ~140 units. The destructed chitosan was dissolved in water and precipitated with adding a definite amount of NaOH. The precipitated chitosan was washed off, dried at 60 °C, and ground in a ball mill. As a result, a cream-like powder was obtained, whose major part of particles had sizes of 1–15 µm. The properties of the obtained powder were investigated by the microscopic, X-ray diffraction, IR spectroscopic, and physico-chemical methods.

Key words: chitosan, deacetylation degree, intrinsic viscosity, levelling-off degree of polymerization, thermocatalytic destruction.

INTRODUCTION

Chitosan, which is prepared from one of the most widespread natural biopolymers, chitin, is increasingly widely used in many fields owing to its valuable properties. In medicine and pharmacy, chitosan is used in anticholesterol, immune-stimulating, and anticancer preparations, for the production of microcapsules, wound healing dressings, prolonged-release drugs, as well as stitch materials and analysis membranes. In the food industry, it is used as a conservant, emulsifier, viscosity regulator, as well as a biologically active additive. In the paper industry, it is used for the production of high-quality paper that is

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stable for the external action and moisture. Besides, chitosan is applied in cosmetics, agriculture, and for wastewater purification from heavy metals and radionuclides. It is used in chromatography, in the textile industry, and in other fields. Chitosan is used in gel or powder form.

The aim of the present work was to obtain finely dispersed chitosan powder by the thermocatalytic destruction method, which had been developed in our earlier works on obtaining microcrystalline cellulose (MCC) [1].

EXPERIMENTAL

Chitosan samples obtained from exoskeletons of crustaceans, supplied by the Joint Stock Company Vostok-Bor (Vladivostok), with a molecular weight of 500 kDalton and “Hydagen CMFP” with a molecular weight of 900 kDalton were investigated.

As the thermal destruction catalyst, hydrochloric acid solution was chosen. Chitosan samples were impregnated with hydrochloric solutions of different concentration and then thermally treated at different temperatures until a dry state was reached. As a result of the thermocatalytic treatment, a partial destruction of chitosan occurred, which was controlled from the changes in intrinsic viscosity measured with a capillary viscosimeter using 0.5 M acetic acid – 0.2 M Na acetate as a solvent [2].

The intrinsic viscosity was determined from the Huggins equation:

$$\frac{\eta_{sp}}{c} = [\eta] + k[\eta]^2 c, \quad (1)$$

where η_{sp} is specific viscosity calculated from the solution and solvent flow time ratio; $[\eta]$ is intrinsic viscosity; c is chitosan concentration in the solution; k is a constant that varies from 0.3 to 0.7, but in our case 0.5 was accepted.

From the intrinsic viscosity, the molecular weight was determined employing the Mark-Houwink equation:

$$[\eta] = K M^a, \quad (2)$$

where M is viscosity average molecular weight; K and a are constants, whose values depend on the polymer type and the chosen solvent. As was shown in [3], for chitosan and the solvent 0.5 M AcOH – 0.2 M NaOAc, these constants are 3.5×10^{-4} and 0.76, respectively, and they do not depend on the deacetylation degree. By dividing the molecular weight of the chitosan by the molecular weight of the glucosamine link, the degree of polymerization (DP) was determined.

The chitosan destructed by the thermocatalytic treatment was dissolved in water, and, stirring in a mixer at a rotational speed of 150 rpm, a NaOH solution was added until pH = 8 was achieved. During this treatment time, aggregation and precipitation of chitosan molecules occurred. The precipitated chitosan was

washed off to pH = 6.5, dried at reduced pressure at 60 °C, and ground in a ball mill. The obtained powder samples were investigated by the microscopic, X-ray diffraction, IR spectroscopic, and physico-chemical methods.

The form and size of the particles of the powder samples, and the particle size distribution were investigated with an optical microscope BIOLAM at ×660 magnification.

The degree of crystallinity of the investigated samples was determined by the X-ray diffraction method. A diffractometer Dron-2 with CuK_α radiation, emitted by a nickel filter, was used. X-ray diffractograms were taken, and the degree of crystallinity was determined from the ratio of the total surface of crystalline peaks and the total surface of the diffractogram [4].

The infrared spectroscopic method with a FTIR spectrometer “Spectrum One” used was applied to determine the deacetylation degree, hydrogen bond energy, and crystallinity index.

The deacetylation degree and hydrogen bond energy were calculated from the following relationships [5]:

$$DD = 100 - \left[\frac{(A_{1660 \text{ cm}^{-1}} / A_{3450 \text{ cm}^{-1}}) \times 100}{1.33} \right] (\%), \quad (3)$$

where DD is deacetylation degree; $A_{1660 \text{ cm}^{-1}}$ and $A_{3450 \text{ cm}^{-1}}$ are absolute heights of absorption bands of amide and hydroxyl groups; and

$$E_H = \frac{1}{k} \frac{v_0 - v}{v_0} \quad (\text{kcal}), \quad (4)$$

where E_H is hydrogen bond energy; v_0 is free OH groups vibration frequency (3650 cm^{-1}); v is bound OH groups vibration frequency; and k is a constant equal to $1.6 \times 10^{-2} \text{ kcal}^{-1}$.

Crystallinity index was determined by infrared spectroscopy from the absorbances ratio $A_{1423 \text{ cm}^{-1}} / A_{897 \text{ cm}^{-1}}$, where $A_{1423 \text{ cm}^{-1}}$ and $A_{897 \text{ cm}^{-1}}$ correspond to the crystalline and amorphous parts, respectively [6].

The water retention value was determined with the Jayme method [7]. Chitosan samples were immersed in water and allowed to stand for 20 h. Then the surplus water was poured off, and the samples were centrifuged for 25 min at 5500 rpm. The water retention value was determined from the relationship:

$$WRV = \frac{m_1 - m_0}{m_0} \times 100 \quad (\%), \quad (5)$$

where WRV is the water retention value; m_1 and m_0 are the sample weights after centrifuging and after drying at 105 °C, respectively.

The physico-chemical and quality parameters such as solubility, water extract pH, mass loss upon drying, content of water-soluble substances, etc. were determined by the methods specified by pharmacopoeias (for example [8]).

RESULTS

Figure 1 shows dependences of DP on the hydrochloric solution concentration at a constant treatment temperature (120°C) for Vostok-Bor and “Hydagen CMFP” chitosan samples. A dramatic decrease in DP can be observed in a definite solution concentration range. At a further increase in the solution concentration, DP changes little. For untreated “Hydagen CMFP” chitosan samples, whose deacetylation degree (70% from IR spectra) is lower than in the case of Vostok-Bor chitosan samples (80% from IR spectra), DP is 1.9 times higher, while the solution concentration range in which a dramatic decrease in viscosity occurs is shifted towards low concentrations. Figure 2 shows dependences of DP on the thermal treatment temperature at the catalyst solution concentration that corresponds to reaching a constant DP (see Fig. 1). With increasing the thermal treatment temperature up to 80°C for both the Vostok-Bor and “Hydagen CMFP” chitosan samples, DP falls, and then becomes practically constant. However, when the chitosan samples impregnated with catalyst solution are dried at 20°C, their DP is approximately 2.5 times lower than in the case the chitosan samples unimpregnated with catalyst solution are dried at 120°C.

Taking into account the obtained results, the following thermocatalytic treatment conditions were set to reach LODP: thermal treatment temperature 80–120°C, impregnation time with catalyst solution 2 h, hydrochloric acid solution concentrations 1.5–3.0% and 0.7–1.0% for chitosan samples with deacetylation degrees 80% and 70%, respectively.

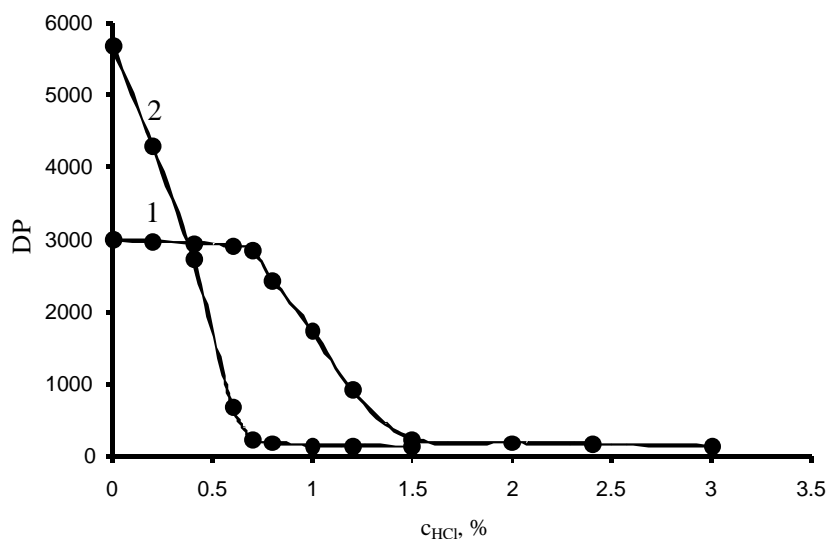


Fig. 1. Degree of polymerization (DP) versus hydrochloric acid solution concentration at the thermal treatment temperature 120°C for Vostok-Bor (1) and “Hydagen CMFP” (2) chitosan samples.

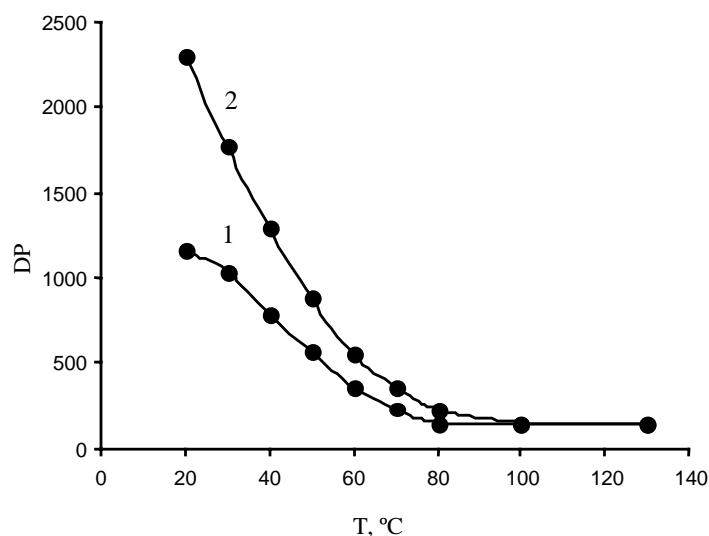


Fig. 2. Degree of polymerization (DP) versus thermal treatment temperature for Vostok-Bor (1) and “Hydagen CMFP” (2) chitosan samples at hydrochloric acid solution concentrations of 3% and 1%, respectively.

After the chitosan samples were treated under the above-mentioned thermo-catalytic treatment conditions, dissolved in water, precipitated, washed off from NaOH, and dried, an odourless cream-coloured powder was obtained.

The microscopic studies showed that the sizes of powder particles in the longitudinal and transversal directions differ little. Figure 3 shows percentage distribution graphs of the number of particles with respect to longitudinal and transversal sizes for the obtained chitosan powders. As can be seen, 84% of the

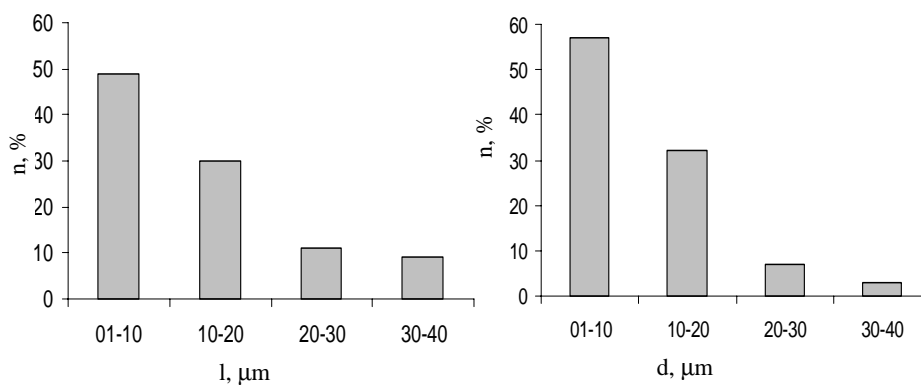


Fig. 3. Percentage distribution graphs in terms of longitudinal (l) and transversal (d) sizes of particles for Vostok-Bor chitosan powder.

chitosan powder particles have an average size up to 20 μm , and the maximum size does not exceed 40 μm . In comparison with the MCC powder obtained by the thermocatalytic destruction method from softwood bleached sulphate pulp [9], which has only 70% of particles with sizes up to 20 μm , but the maximum particle size reaches 100 μm , the chitosan powder has a lower particle size.

The deacetylation degree and hydrogen bond energy were determined for chitosan samples with the IR spectroscopic method. The deacetylation degree for Vostok-Bor chitosan samples was 80% and for “Hydagen CMFP” 70%. The deacetylation degree did not change after thermocatalytic treatment. The hydrogen bond energy for untreated and treated samples was ~ 3.67 and 3.76 kcal, respectively.

The X-ray diffraction and IR spectroscopic studies showed that the crystallinity for the obtained chitosan powder samples was by 30–50% higher than for the initial chitosan samples.

Physico-chemical and quality indices of the obtained chitosan powder samples are presented in Table 1. For comparison, also the corresponding indices of the MCC powder are given. As can be seen, the obtained chitosan powders, just as MCC powder, are practically insoluble in water, many dilute acids, sodium hydroxide solutions, and organic solvents. Their mass losses upon drying and WRV are higher, but the chemical purity is lower than of MCC powders.

Table 1. Physico-chemical and quality parameters of the obtained chitosan and MCC powder samples

| Parameter | Vostok-Bor chitosan | “Hydagen CMFP” chitosan | MCC |
|--|---|---|---|
| Solubility | Practically insoluble in water, diluted sulphuric, hydrochloric, nitric, and formic acids, 8.5% sodium hydroxide, 96% ethanol, toluene, acetone. Soluble in diluted acetic acid and oxalic acid | Practically insoluble in water, diluted sulphuric, hydrochloric, nitric, and formic acids, 8.5% sodium hydroxide, 96% ethanol, toluene, acetone. Soluble in diluted acetic acid and oxalic acid | Practically insoluble in water, diluted acids, 8.5% sodium hydroxide, 96% ethanol, toluene, acetone |
| Water extract pH | 6.5–7.0 | 6.4–6.8 | 6.2–6.8 |
| Mass loss upon drying, % | 12.0–12.4 | 11.7–12.1 | 3.7–5.8 |
| Content of water-soluble substances, % | 0.45–0.67 | 0.35–0.50 | 0.12–0.16 |
| Sulphate ash, % | 0.92–1.14 | 0.80–0.90 | 0.19–0.30 |
| Starch and dextrine | Does not contain | Does not contain | Does not contain |
| WRV, % | 95–97 | 93–95 | 54–62 |

CONCLUSIONS

A thermocatalytic destruction method for the preparation of chitosan destructed to the levelling-off degree of polymerization was developed, and thermocatalytic treatment conditions were determined. After precipitation of the destructed chitosan, its washing-off from hydroxide, drying, and grinding in a ball mill, a finely dispersed chitosan powder was obtained.

The structural and physico-chemical properties of the obtained chitosan powder samples were investigated. The deacetylation degree of the prepared chitosan powder samples was equal to 70% and 80%. It was found that the size of the major part of particles is 1–15 µm. The degree of crystallinity of the obtained chitosan powders is higher in comparison with that of untreated chitosan. They are not soluble in water, many diluted acids, sodium hydroxide, and organic solvents, and they have a relatively high water retention value, namely 96%.

REFERENCES

1. Laka, M. & Chernyavskaya, S. Method for obtaining microcrystalline cellulose. Latvian Republic Patent 11184, 1996. *Patenti un Preču Zīmes*, 1996, **4**, 878.
2. Terbojevidh, M. & Cosani, A. Molecular weight determination of chitin and chitosan. In *Chitin Handbook* (Muzzarelli, R. A. A. & Peter, M. G., eds). European Chitin Society, 1997, 87–101.
3. Terbojevich, M., Cosani, A. & Muzzarelli, R. A. A. Molecular parameters of chitosans depolymerized with the aid of papain. *Carbohydr. Polym.*, 1996, **29**, 63–68.
4. Ioelovich, M. & Leykin, A. Nano-cellulose and its applications. *Sci. Israel – Technol. Advantages*, 2004, **6**, 17–24.
5. Struszczyk, H. Microcrystalline chitosan. I. Preparation and properties of microcrystalline chitosan. *J. Appl. Polymer Sci.*, 1987, **33**, 177–189.
6. Ferruz, R. & Pages, P. Water retention value and degree of crystallinity by infrared absorption spectroscopy in caustic-soda-treated cotton. *Cellulose Chem. Technol.*, 1977, **11**, 633–637.
7. Jayme, G. & Hahn, G. Vereinfachte Messung des Wasserrückhaltenvermögen von Zellstoffen. *Das Papier*, 1960, **14**, 138–139.
8. *British Pharmacopeia*, 1988, 1.
9. Laka, M., Chernyavskaya, S. & Maskavs, M. Cellulose-containing fillers for polymer composites. *Mech. Compos. Mater.*, 2003, **39**, 183–188.

Kitosaanpulbri saamine ja selle omaduste uurimine

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Kitosaanpulbri saamiseks on kasutatud termokatalüütilist osalise lagundamise meetodit. Optimaalsete lagundamistingimuste leidmiseks on kontrollitud kitosaani-proovide piirviskoossuse muutust kuni polümerisatsiooni tasakaaluni. Saadud pulbri omadusi on uuritud mikroskoopiliste, röntgenidifraktsiooni-, IP-spektroskoopia- ja füüsikalise-keemiliste meetoditega.