

PHYSICOCHEMICAL PROPERTIES OF CHITIN ISOLATED FROM SHELL
OF INDUSTRIAL CRABS OF VARIOUS SPECIES

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Abstract

Differential thermal analysis was carried out over the range of $-190 \div 400$ °C on six samples of chitin from shells of industrial crabs of various species. All samples of the chitin have several relaxation transitions of endothermal character (g_1 -, g_2 -, β - and γ -).

Key words: *chitin, physicochemical properties, shell, industrial crabs of various species.*

1. Introduction

Recycling of waste products from crab processing plants has an important ecological impact. Chitin obtained from the shells requires additional processing to reduce its molecular weight. This reduction can be achieved by using microorganisms. It is important to investigate the physicochemical characteristics, particularly the temperatures of physical transitions of chitin and its derivative chitosan, and their differences between various species of crabs in order to understand mechanisms of chitin/chitosan microbial destruction. Therefore differential thermal analysis (DTA) was carried out over the range of $-190 \div 400$ °C on six samples of chitin produced in Russian Federal Research Institute of Fisheries and Oceanography (Moscow, RUSSIA) from shells of industrial crabs of various species.

2. Materials and methods

Six samples of chitin that were isolated from various species of crabs such as golden king crab (*Lithodes aequispinus*) (No 1), snow crab (*Chionoecetes opilio*) (No 2), Korean hair crab (*Erimacrus isenbeckii*) (No 3), blue king crab (*Paralithodes platypus*) (No 4), red king crab (*Paralithodes camtchaticus*) (No 5), tanner crab (*Chionoecetes bairdi*) (No 6) were studied. These crabs were caught at the intermolting stage in the Sea of Okhotsk during one season. The samples were demineralized. The separation of a protein component of the shell (deproteination) was carried out with 5% solution of NaOH at 20 - 22 °C during 24 h. Then the chitin was washed with distilled water till neutral pH of washing waters. Pigments – carotenoids (astoxanthine, xanthoxanthine – remained in the samples.

Our experiments showed that air-dried samples of the chitin contained from 8.3 to 11.8 mass.% water that evaporated from them at 120 - 127 °C as shown by the data from DTA (**Table 1**, **Figure 1**).

As a method of research works, the differential thermal analysis was used [1 - 3]. Runs over the range of -190 to 400 °C were conducted in a helium atmosphere using a device developed in the Laboratory of Thermochemistry at the Research Institute of Chemistry Nizhny Novgorod State University [4]. Quartz served as standard. The mass of a sample

Table 1. Physicochemical properties and mean temperatures of physical transitions in chitin isolated from shell of crab of various species.

Sample	No 1	No 2	No 3	No 4	No 5	No 6
Sample mass, g	0.1870	0.2054	0.1861	0.2330	0.2580	0.2558
Content of H ₂ O, mass%	9.0	9.0	11.8	8.5	10.4	8.3
t _{vap} (H ₂ O), °C	120.5	124	122.5	127	123	127
t _v , °C	-----	-----	0.5	-----	-22	-33
t _β , °C	22	19	30	-----	33	63
t _{g1} , °C	103	102.5	107	106.5	85	124
t _{g2} , °C	212	213	209	-----	262	206
t _{dest1} , °C	294	287	300.5	283.5	299.5	262
t _{dest2} , °C	326	324	325.5	326	325	326

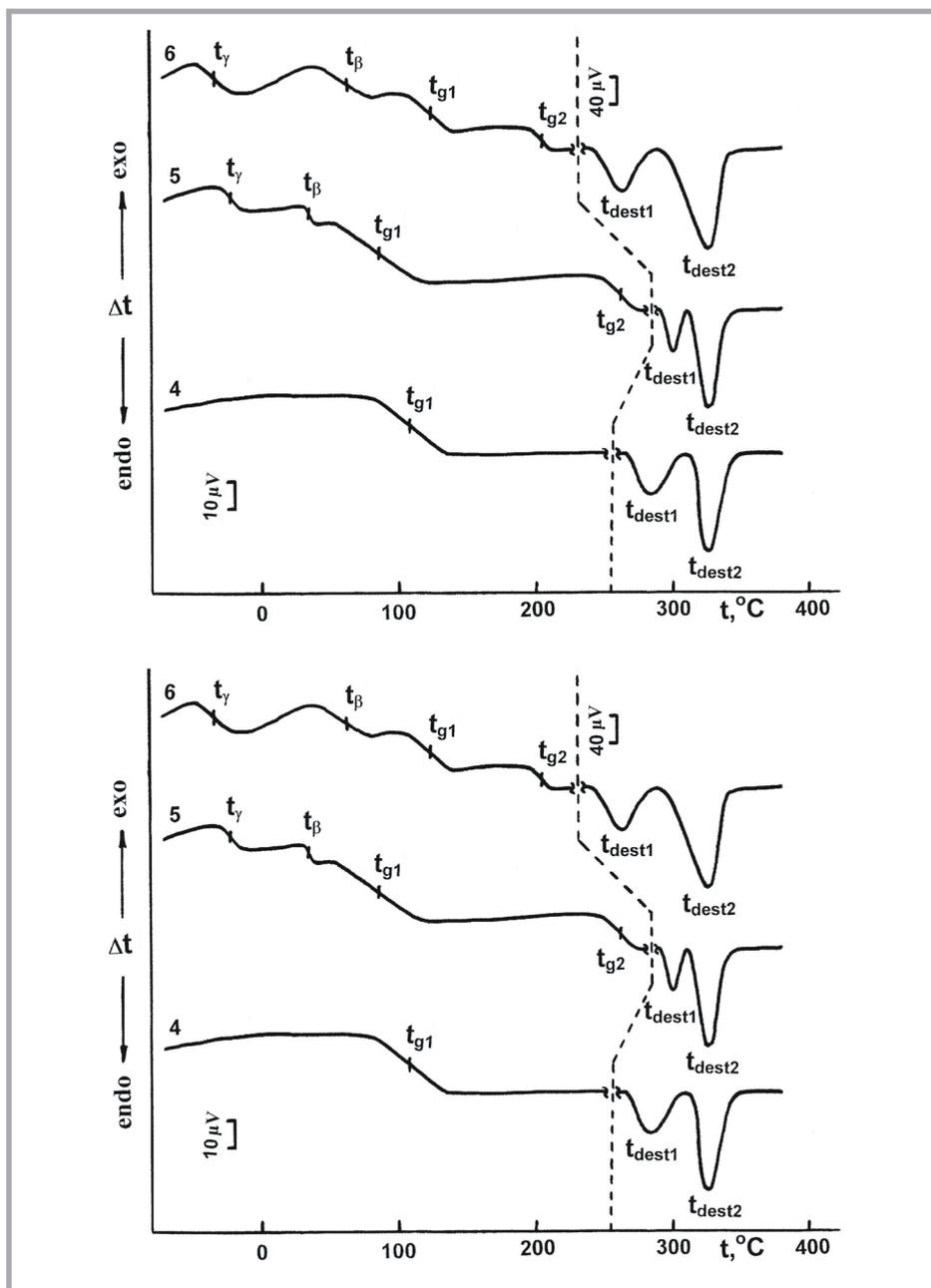


Figure 1. Thermograms of dried chitin that were isolated from various species of crabs: 1 – golden king crab (*Lithodes aequispinus*); 2 – snow crab (*Chionoecetes opilio*); 3 – Korean hair crab (*Erimacrus isenbeckii*), 4 – blue king crab (*Paralithodes platypus*); 5 – red king crab (*Paralithodes camchaticus*); 6 – tanner crab (*Chionoecetes bairdi*).

and of the standard was $\sim 0.25 - 0.35$ g. The temperature of the sample and the difference in temperatures between the sample and the standard were measured with a chromel-copel thermocouple to within $\pm 0.5^\circ$. The rate of heating in experiments was $5^\circ/\text{min}$ and the deviation from linearity did not exceed 1%. To check the DTA device operation the melting temperature (t_m) of standard n-heptane and the glass transition temperature (t_g) of purified glycerin were determined. The results obtained by us coincided with the corresponding reliable literature data for n-heptane [5] with an uncertainty of 0.2°C and glycerin 1°C [6].

The thermal chamber with the sample and standard was cooled with liquid nitrogen to -190°C at a rate of $\sim 20^\circ/\text{min}$. Then it was heated at a rate of $5^\circ/\text{min}$ up to $\sim 140^\circ\text{C}$ (the first warming-up) to record the endothermal peak of evaporation of sorbed water (**Table 1**). The heating was ended, the sample was cooled at a rate of $5^\circ/\text{min}$ down to room temperature, and simultaneously the evaporated sorbed water was pumped out *in situ* in the thermal chamber. Then the crucible with the sample was taken out from the thermal chamber, weighed on an analytical balance and the quantity of water contained in an air-dried sample was determined. Afterwards the sample was placed in a desiccator over CaCl_2 . The next day the dried sample was placed in the thermal chamber and the experiment was repeated with heating the sample up to 380°C (**Figure 1**).

3. Results and discussion

Figure 1 is examples demonstrating typical thermograms for samples No 1 - 6 of dried chitin that were isolated from various species of crabs. The averaged results and some physicochemical characteristics of the samples tested are given in **Table 1**.

As seen from the thermograms, all the samples of chitin have several relaxation transitions of endothermal character (g_1 -, g_2 -, β - and γ -). Such processes were observed in earlier studies of samples of chitin isolated from various sources [7 - 10] as well as other polysaccharides [11 - 16].

According to the accepted classification [17 - 19] the γ -type transition appearing at a temperature below 0°C (**Table 1, Figure 1**), is related to the excitation of vibrations of side branches of a polymer chain. It is suggested that those are present in macromolecules of the tested chitin. A similar pattern was observed by us for branched amylopectin [9, 14] and samples of hydrolyzed crab chitin and chitosan [10]. The β -type transition is caused by the excitation of vibrations of polymer chain segments that are smaller than a kinetic segment [17 - 19]. In cellulose, for example, an analogous process is explained as the excitation of vibrations of pyranose rings around a glucoside bond [20]. Two temperature intervals of devitrification (t_{g1} and t_{g2}) point to the availability of microregions of different degree of ordering (highly- and poorly ordered) in the examined chitin. The temperature of the first glass transition (t_{g1}) concerns the process in amorphous microregions. The temperature of the second glass transition (t_{g2}) is attributed to highly ordered microregions of the polymer.

Chitin derived from blue king crab (*Paralithodes platypus*) shell (No 4) showed unusual properties and its behavior was different from the rest of the samples analyzed. It

exhibited a single devitrification temperature ($t_{g1} = 106.5$ °C) and did not exhibit a γ - or a β - transition (**Table 1, Figure 1**, curve 4).

The γ -transition was observed in three samples (No 3, 5 and 6) with the highest, $t_{\gamma}=0.5$ °C, in sample No 3 (Korean hair crab, *Erimacrus isenbeckii*), and the lowest, $t_{\gamma}=-33$ °C, in sample No 6 (tanner crab, *Chionoecetes bairdi*).

All samples except No 4 registered a β -transition. The highest, $t_{\beta}=63$ °C, was recorded for sample No 6 (tanner crab, *Chionoecetes bairdi*), and the lowest, $t_{\beta}= 19$ °C, for sample No 2 (snow crab, *Chionoecetes opilio*).

Vitrification temperatures were not significantly different between the analyzed chitin samples ($t_{g1} \approx 110$ °C, $t_{g2} \approx 210$ °C).

The destruction of the chitin proceeds in two stages ($t_{destr1} 290$ °C and $t_{destr2} \approx 325$ °C) with the absorption of energy in the form of heat (**Table 1, Figure 1**).

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5. References

1. Berg L. G.; (1969) *Introduction to thermography*. Moscow.
2. Wendland W.; (1978) *Thermal methods of analysis*. Moscow.
3. *Newer methods of polymer characterization*; (1966) Bacon Ke (ed) Moscow.
4. Uryash V. F., Mochalov A. N., Pokrovsky V. A.; (1978) *Device for differential thermal analysis. Thermodynamics of Organic Compounds Interinstitution Bull Gorky, 7*, pp. 88-92.
5. Douglas T. B., Furukava G. T., McCoskey R. E., Ball A. L.; (1954) *Calorimetric properties of normal heptane from 0 to 520 K. J Res Natl Bur Standards 53*, pp. 39-43.
6. Ahlberg J. E., Blanchard E. R., Lundberg W. O.; (1937) *The heat capacities of benzene, methyl alcohol and glycerol at very low temperatures. J Chem Phys 5*, pp. 539-551.
7. Tsvetkova L. Ya., Novoselova N. V., Golitsin V. P., Ivanov A. V., Khlyustova T. B., Uryash V. F.; (1993) *Thermodynamic characteristics of chitin and chitosan. Zh Khim Termodinam i Termokhim Ivanovo, 2*, pp. 88-93.
8. Uryash V. F., Kokurina N. Yu., Maslova V. A., Larina V. N., Iosilevich I. N.; (1998) *Calorimetric study of chitin from mushrooms and its mixtures with water. Bulletin of Nizhny Novgorod State University, Ser Chem, Nizhny Novgorod, 1*, pp. 165-170.
9. Uryash V. F.; (2002) *Thermodynamics of chitin and chitosan. In: Skryabin K. G., Vikhoreva G. A., Varlamov V. P. (eds), Chitin and chitosan. Preparation, properties and application. Moscow, pp. 119-129.*
10. Uryash V. F., Kokurina N. Yu., Larina V. N., Varlamov V. P., Ilina A. V., Grishatova N. V., Gruzdeva A. E.; (2007) *Influence of acidic hydrolysis on heat capacity and physical transitions of chitin and chitosan. Bulletin of Nizhny Novgorod State University, Nizhny Novgorod 3, pp. 98-104.*
11. Uryash V. F., Maslova V. A. Chizhikova V. A.; (1994) *Thermal analysis of agar and agar-water mixtures as a polymeric carrier for drugs. Biomater-Liv Syst Inter 2*, pp. 71-77.

12. **Gruzdeva A. E., Uryash V. F., Karyakin N. V., Kokurina N. Yu., Grishatova N. V.;** (2000) Heat capacity and physicochemical analysis of agar and agarose. *Bulletin of Nizhny Novgorod State University. Ser. Chem., Nizhny Novgorod, 1*, pp. 139-145.
13. **Uryash V. F., Karyakin N. V., Gruzdeva A. E.;** (2001) Optimization of preparation process of biologically active substances on the base of calorimetric data. *Advancing materials, Moscow, 6*, pp. 61-69.
14. **Uryash V. F., Gruzdeva A. E., Kokurina N. Yu., Grishatova N. V., Larina V. N.;** (2004) Thermodynamic characteristics of amylose, amylopectin and starch in the range 6–320 K. *Zh Fiz Khimii Moscow 78*, pp. 796-804.
15. **Uryash V. F., Gruzdeva A. E., Kokurina N. Yu., Grishatova N. V., Larina V. N.;** (2005) Thermodynamic characteristics of pectin of different degree of esterification in the range 6-330 K. *Zh Fiz Khimii Moscow 79*, pp. 1383-1389.
16. **Uryash V. F., Gruzdeva A. E., Kokurina N. Yu., Grishatova N. V., Faminskaya L. A.;** (2007) Calorimetric determination of solubility of water in pectin and diagram of physical states of pectin-water system. *Vysokomol Soed Ser A Moscow 49*, pp. 1672-1678.
17. **Hoffman J. D., Williams G., Passaglia E.;** (1966) Analysis of α -, β - and γ -relaxation processes in polychlorotrifluoroethylene and polyethylene: dielectric and mechanical properties. In Boyer R. F. (ed), *Transition and relaxations in polymers*, New York, pp. 193-272.
18. **Bartenev G. M., Sanditov D. S.;** (1986) *Relaxation processes in glassy systems*, Moscow.
19. **Perez J.;** (1998) Study of polymer materials by method of mechanical spectrometry. *Vysokomol Soed, Ser B Moscow 40*, pp. 102-135.
20. **Uryash V. F., Rabinovich I. B., Mochalov A. N., Khlyustova T. B.;** (1985) Thermal and calorimetric analysis of cellulose, its derivatives and mixtures with plasticizers. *Thermochim. Acta 93*, pp. 409-412.