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# **Obtaining Chitin and Chitosan from the Subspecies of Apis Mellifera Bees**

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**ABSTRACT:** The article presents the synthesis of chitin and chitosan obtained from the subspecies of *Apis mellifera* bees. The optimal conditions for obtaining chitin and chitosan from the bee subsurface were investigated. The composition and molecular structures of chitin and chitosan were studied using IR spectrophotometry and elemental analysis of chitosan, and microscopic images were taken using a scanning microscope. The resulting chitosan is polydisperse by molecular weight of D-glucosamine, containing 5-15 % of acetamide groups, as well as up to 1 % of groups connected to amino acids and peptides. Biopolymers chitin and chitosan were obtained and characterized based on a new promising source – the dry subspecies of *Apis mellifera* bees. Taking into account the natural origin of the bee subsurface and its degree of renewability, we can conclude that this raw material is of great importance as a source material for obtaining polyelectrolytes with a set of unique properties.

**KEYWORDS:** synthesis, research, chitin, chitosan, amino acid, polysaccharide, IR spectroscopy, scanning microscope.

## **I. INTRODUCTION**

In recent years, renewable natural resources have become increasingly popular, among which chitin, the second most common polymer in nature after cellulose, and its derivative chitosan occupy a special place. The substance chitosan is a part of many supplements, with a positive effect on lowering cholesterol and strengthening the immune system. The natural polysaccharide chitosan and its derivatives, due to its wide range of useful properties, are also increasingly used in various fields, such as: the textile industry - for dressing and anti - shrink or water - repellent treatment of fabrics; medicine-as suture materials, early-and burn-healing dressings, as part of ointments and various medicinal preparations, such as enterosorbent; in perfumes and cosmetics, it is part of moisturizing creams, lotions, gels, hair sprays, etc.shampoos; when cleaning water, it serves as a sorbent and flocculant. The indisputable advantages of chitosan include its perfect safety for humans and the environment: it is environmentally friendly and completely decomposes in natural conditions [1].

If data on the structure and properties of cellulose (the most important representative of the class of polysaccharides, which is the main structural material of plants) are presented in the available literature, information about chitin is significantly less. At the same time, chitin is the basis of the skeletal system that supports the cellular structure of tissues in the shells of crustaceans, the cuticle of insects, the cell wall of fungi and bacteria [2].

## **II. THE MAIN FINDINGS AND RESULTS**

The most accessible for industrial development of chitin production in the Republic of Uzbekistan is the silkworm and honeybees. The raw material for obtaining chitin from bees can serve as a subspecies of bees [3].

A special type of chitosan – PHALLOSAN the more powerful the biologically active substance than crustacean chitosan. A significant reserve of raw materials for chitosan production is represented by subspecies of *Apis mellifera* bees (Fig.1).



**Figure 1. Honeybees *Apis mellifera* (honey bearer) and bee Podmore**

In this regard, it is quite appropriate to obtain reproducible biopolymers of chitin and chitosan from the bee subspecies *Apis mellifera*. Podmore – bees that died, mainly during the wintering period and fell to the bottom of the hive. In summer, the death of bees is much more significant than in winter, but less noticeable, since they usually die outside the hive. Apizán or, as it is called in scientific terms, a low-molecular chitosan-melanin complex is obtained from bee subsurface [4].

In summer, during the period of active honey collection and in spring after wintering, the bee family is updated by almost 60-80%. The strength of the bee family (the mass of worker bees in the bee family, measured in kg) is, on average, 7.5-8 kg. This makes it possible to consider the bee subspecies as a new promising source of chitin and chitosan along with traditional raw materials [5].

We used a dry Podmore of bees collected during the spring renewal of the bee family and containing a significant amount of chitin. The raw material is a black-brown mass with a specific smell. When viewed in detail, you can see whole undisturbed bees and various parts of bees (head, chest, legs, abdomen, wings, etc.). Bee Podmore contains a minimal amount of minerals, since the cuticle of insects is practically not mineralized. The Podmore mass was dried at a temperature of about 35°C, laid out in a thin layer. The dried raw material mass of 30 g was crushed and demineralized (DM), then deproteinated (DP) according to the following scheme (Fig.2).



**Figure 2. General scheme of chitin production**

Demineralization (DM) was performed according to the naskmap Method by treating a subpopulation of bees with 2 M hydrochloric acid for 5 hours at room temperature. Deproteination was performed by treating the crushed raw materials with 1 N sodium hydroxide solution for 1 hour at 80°C. The mass was filtered and dried at room temperature. Each process was accompanied by washing the raw material to a neutral reaction of the washing water (pH=7).

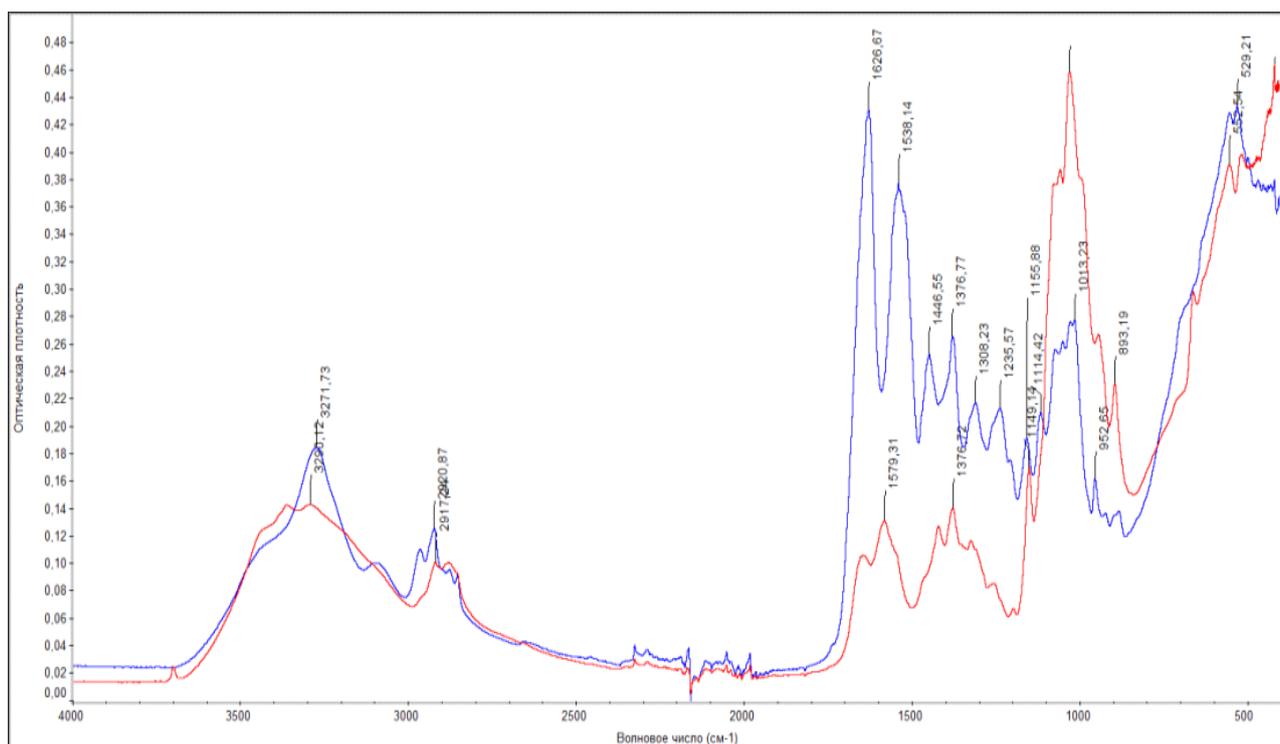
Chitosan is an aminopolysaccharide obtained by removing the acetyl group in chitin as a result of treating it under harsh conditions with an alkali solution, which allows replacing the acetyl groups of chitin with amino groups. Thus, the stage of deacetylation of chitin is always preceded by the process of its isolation from chitin-containing raw materials. Chitosan was obtained by deacetylating chitin with 35% aqueous NaOH solution for 4 hours at a temperature of 85°C and dried at 50-55°C.

The deacetylation reaction is ACCOMPANIED by simultaneous breaking of the glycoside bonds of the polymer. Thus, chitosan is a polydisperse D-glucosamine by molecular weight, containing 5-15 % of acetamide groups, as well as up to 1 % of groups connected to amino acids and peptides.

When dried at higher temperatures, chitosan compacts, darkens and loses solubility, which reduces the possibility of its use. Then the resulting mass was discolored with a 3% solution of hydrogen peroxide and washed with ethanol. The reaction product is a light beige mass with a specific smell.

Interpretation of the obtained biopolymers was carried out by taking IR spectra on a Nicolet S 50 (Thermo Fisher Scientific, USA) Fourier transform spectrometer, which are shown in Fig. 3.

As can be seen in the IR spectrum of chitin are observed the characteristic absorption bands in the regions  $3290\text{ cm}^{-1}$  is related to the fluctuation of N-H, and absorption bands  $1371\text{ cm}^{-1}$  which indicate the presence of  $-\text{CH}_3$  group, the absorption at  $1579\text{ cm}^{-1}$  is characteristic of C=O group. The IR spectrum of chitosan shows peaks in the region of  $3272\text{ cm}^{-1}$  and  $1377-1028\text{ cm}^{-1}$ , which indicate the presence of the  $\text{NH}_2$  group.



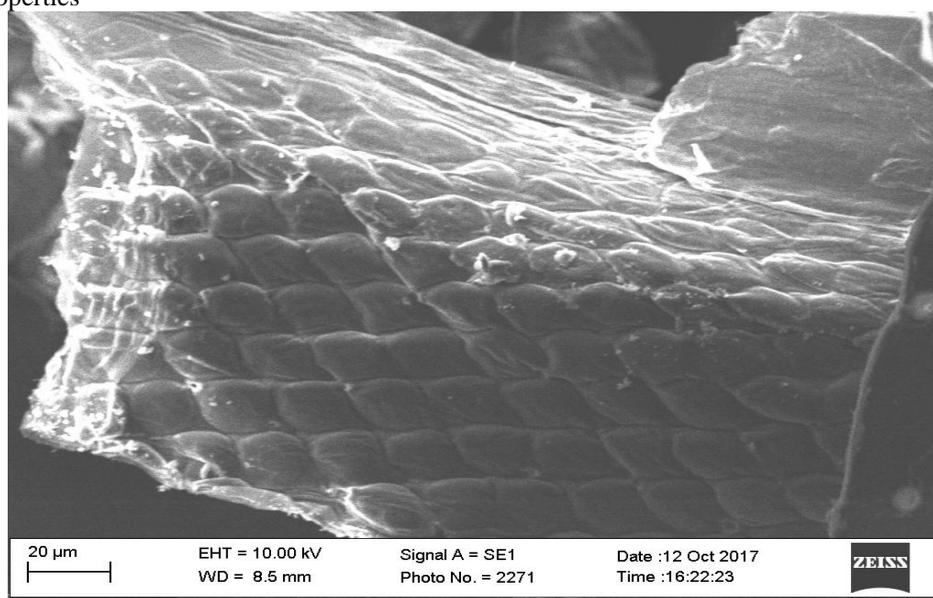
**Figure 3. Fourier - IR spectra of chitin and chitosan obtained from bee Podmore**

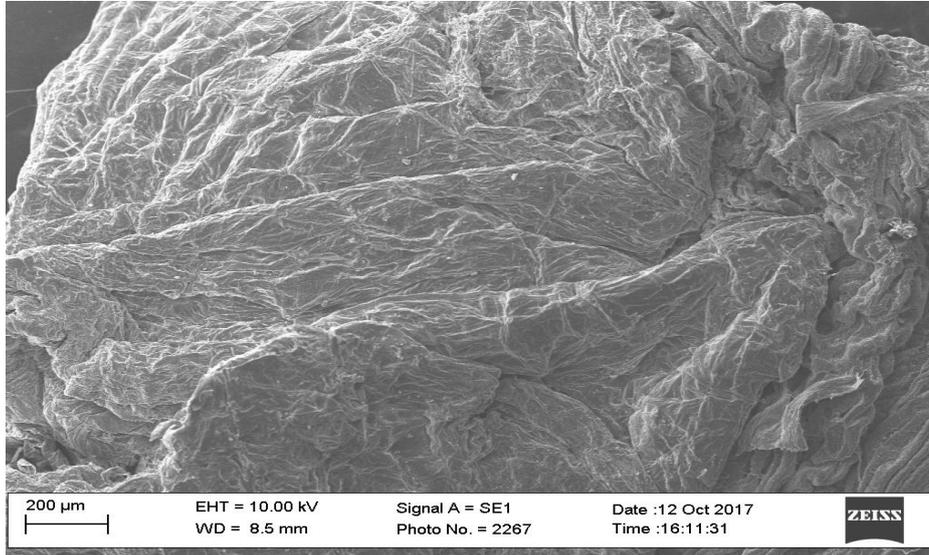
At this absorption in the region of  $1360-1000\text{ cm}^{-1}$ , all types of amines have absorption bands caused by the participation of the C–N bond in the skeletal vibrations of the molecule. In the sample of chitin and chitosan, bands with maxima at  $1446\text{ cm}^{-1}$  of the strain oscillation of the  $\text{CH}_2$  - and  $\text{CH}_3$ -groups and  $1373\text{ cm}^{-1}$  (inflection) of the strain oscillation of the OH-bond were also recorded. In the chitosan sample, a wide band of average intensity is observed in the region of  $1320-1387\text{ cm}^{-1}$ , corresponding to the oscillation of the OH bond.

An elemental analysis of chitosan obtained from a subspecies of bees was also performed, and microscopic images of the chitin and chitosan structure were taken using a scanning microscope (Fig.4).

These figures indicate that when processing chitin with an alkali solution, chitin molecules quickly pass into an amorphous state, and then into a crystal state, that is, amorphization occurs, the crystal lattice is gradually destroyed. From structural and molecular and IR spectrophotometric data, it can be concluded that during deacetylation, chitin is converted to the form of chitosan, that is, the molecular structures and crystals of chitosan differ from the molecular structures and crystals of chitin. These can be considered as copolymers of chitin and chitosan.

Thus, the biopolymers chitin and chitosan were obtained and characterized based on a new promising source – the dry subspecies of *Apis mellifera* bees. Taking into account the natural origin of bee Podmore and its degree of renewability, we can conclude that this raw material is of great importance as a source material for obtaining polyelectrolytes with a set of unique properties





**Figure 4. Electron micrographs of chitin and chitosan obtained from bee subpestilence**

### III. CONCLUSION

The presented material can be considered only as primary information about the structure of chitin, chitosan obtained from the subspecies of *apis millifera* bees. As already mentioned, chitin and chitosan are similar in structure to cellulose, one of the main fiber - forming natural polymers. It is natural, therefore, that, like cellulose, these polymers and their derivatives have fiber - and film-forming properties. However, what has already been said allows us to conclude that these materials may take a prominent place in various areas of our life in the near future.

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